Basic Science for Improving Survival & Quality of Life

Sub Topics:
- Botany
- Environmental Science and Technology
- Instrumental and Measurement
The 7th Basic Science International Conference
Basics Science for Improving Survival and Quality of Life

7 – 8 March 2017
Ijen Suites Resorts & Convention
Malang, East Java
Indonesia

Proceedings Book

Sub Topics:
✓ Botany
✓ Environmental Science and Technology
✓ Instrumentation and Measurement
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# BASIC 2017 COMMITTEE

**Steering Committee**

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ABOUT BASIC

The Annual Basic Science International Conference is a scientific meeting aimed to promote mutual exchange between scientists and also experts, to discuss innovative ideas in scientific research, and to tackle contemporary problems through the application of knowledge that rise from sciences. The scope of this conference is fundamental and applied research in chemistry, biology, physics, and mathematics. The origin of this conference was initiated in year 2000, by the Faculty of Mathematics and Natural Sciences of Brawijaya University, under the name of Seminar Nasional Kemipaan (National Sciences Conference). Since then, the conference has been organized regularly on annual basis. In 2004, the conference changed its name to Basic Sciences Seminar (BSS) and started to invite international speakers and participants. The conference then expands its scope to international in 2011 and formally adopting the current name. The previous Basic Sciences International Conference was held at Atria Hotel Malang in 2016 with participants from many countries including Australia, Malaysia, Thailand, Japan, UK and Germany.
WELCOME MESSAGE

On behalf of the organizing committee, I would like to welcome you to the 7th Annual Basic Science International Conference.

Firstly, I would like to thank all participants who have spent their time to come and join us for the conference. I believe that we will not be able to hold this conference successfully without participation from all of you. Secondly, I would like to thank the dean of faculty of Mathematics and Natural Sciences, Brawijaya University, because the faculty has provided us supports and facilities. I am thankful to our great keynote and invited speakers for their willingness to join the conference and share their scientific knowledge to all of us. Thanks to our reviewers who have made assessments and suggestions related to the abstracts. I also want to thank the sponsors which have made their contributions to this conference. Finally, I want to thank all members of the committee for their hard work to make this conference successful.

The Basic Science International Conference is held every year since 2010, and always organized by the Faculty of Mathematics and Natural Sciences, Brawijaya University. This conference is a forum that enables us to share our ideas among us. The participants are expected also to take their time and opportunities to know each other during the conference, in order to strengthen their networks and collaborations. In this conference, we have more than 300 participants from counties such as Indonesia, Japan, Australia, Germany, Switzerland, and Thailand. In the conference, we have plenary lectures and sessions for parallel oral presentations as well as poster presentations.

We hope that all participants enjoy all activities during the conference and this proceedings book will be useful for all of us.

Thank you very much.

Best regards,

Hari Arief Dharmawan, Ph.D.

Chairman of BaSIC 2017
WELCOME MESSAGE

On behalf of the Dean of Faculty of Mathematics and Natural Sciences, Brawijaya University, I would like to extend my warmest welcome to all delegates from all over the world. Welcome to Malang, where Malang is one of the educational city in Indonesia. Malang, which is about more than 400 meters above sea level, has many tourist destinations. Malang is like a bowl, surrounded by some volcanoes in the east (Semeru and Bromo), west (Kawi and Kelud) and north (Arjuna and Welirang Complex), and in the south are coastal areas, where we have many beautiful new opening beaches.

We are very pleased to welcome you in the proceedings book of the seventh Annual Basic Science International Conference 2017. I would like to express my gratitude to all of the participants, keynote and invited speakers as well. Many thanks also go to the reviewers and the editorial team for their big effort in supporting this book of abstracts. Last but not least my big appreciation to the steering and organizing committees, in realizing this proceedings book.

Thank you.

Faculty of Mathematics and Natural Sciences,

Dean,

Adi Susilo, Ph.D.
CONFERENCE VENUE

The Room Map of The 7th Annual Basic Science International Conference 2017

- Parallel Session Rooms
- Ballroom

- Photobooth area
- Registration
- Poster area and booth
- Booth sponsor
# CONFERENCE PROGRAM

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CRISPR/Cas9: Basics and Applications in "gene surgery".  
Prof. Dr. Wolfgang Nellen, Institut fur biology, Germany |
| 09.45 – 10.00 | Coffee Break                                                         |
| 10.00 – 10.45 | **Plenary Lecture 2:**  
Use of Wavelet Analyses with Potential Field Data  
in Exploration and Monitoring Studies  
Dr. Guillaume Mauri, Neuchatel University, Switzerland |
| 10.50 – 11.35 | **Plenary Lecture 3:**  
Mathematics for Solving 5G Massive Wireless IoT Networks Problems  
Dr. Eng. Khoirul Anwar, S. T., M. Eng., Telkom University |
| 11.35 – 12.30 | Lunch                                                                |
| 12.30 – 15.00 | Parallel Session 1                                                   |
| 15.00 – 15.30 | Poster Session & Coffee Break                                        |
| 15.30 – 17.30 | Parallel Session 2                                                   |
| 17.30 – 19.00 | Breaks                                                               |
| 19.00 – 21.00 | Gala Dinner                                                          |

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The Roles of Metal Ions in Diabetes – Metal Drugs and Supplements  
Prof. Peter Andrew Lay, Sydney University, Australia |
| 09.00 – 09.45 | **Plenary Lecture 5:**  
Functionalization of Stainless Steels Via Low Temperature Plasma Nitriding  
Prof. Tatsuhiko Aizawa, Shibaura Institute of Technology (SIT), Japan |
| 09.45 – 10.00 | Coffee Break                                                         |
| 10.00 – 12.00 | Parallel Session 3                                                   |
| 12.00 – 13.00 | Lunch                                                                |
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Plenary Lectures
CRISPR/Cas9 – the new gene surgery

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Abstract – The new gene editing tool CRISPR/Cas9 allows for easy and efficient targeted changes in the genomes of microbes, plants and animals (including humans). CRISPR/Cas is a prokaryotic immune system that “memorizes” infections by phages and plasmids. The molecular biology and biochemistry of the machinery is very well understood and it has been engineered in different ways to serve specific needs in biotechnology. I will briefly present the origins and biochemistry of bacterial CRISPR/Cas systems, the technical use in gene technology and some applications that have already been achieved and others that are in the pipe-line.

1. SUMMARY

In 2012, a revolutionary paper appeared in the journal Science: Jennifer Doudna and Emmanuelle Charpentier and co-workers published the application of the bacterial CRISPR immune system to modify, delete and insert genes in essentially every living cell.

In bacteria and archaea, CRISPR represents an array of short DNA pieces that were captured from infecting phages or plasmids. Using enzymes encoded by the adjacent Cas locus, the microorganisms can fight subsequent infections. Transcripts of the short DNA pieces (crRNA) serve as guides to direct a nuclease encoded in the Cas locus to the invader and destroy it.

The Cas-nucleases can thus be programmed to cleave any specific sequence in any genome. Out of a plethora of different Cas-nucleases from different bacteria and archaea, the Cas9 enzyme from Streptococcus pyogenes proved to be the most convenient enzyme from the family for technical applications. In eukaryotes, the cut in the DNA is reversed by the cellular repair machinery. However, relegation of the cut ends usually results in sequence mistakes by short deletions or short insertions. Consequently, the gene targeted by a synthetic crRNA becomes functionless.

An additional piece of DNA with (partial) homology to the targeted sequence can be introduced to the cell. This will be used as a template and can replace the original sequence around the cut – including large insertions and deletions as well as single nucleotide changes.

CRISPR/Cas can thus be used in any organism to delete, insert or modify genes at a defined target site. The applications are endless and only a few examples will be presented here.

1. Compatibility in bone marrow transplantation: For BMT in leukaemia patients, compatible donors have to be found which have the same surface antigens as the recipient. With CRISPR/Cas9, genes for surface antigens of the donor can be adjusted to the recipient. If the genetic defect in the patient is exactly known, bone marrow cells can be removed, repaired in vitro and reintroduced into the patient.

2. Resistance of crop plants to fungal or viral diseases: In many cases natural resistances of plants against viral and fungal infections were accidentally lost during breeding and selection over the last few hundred years. Resistance genes are still present in some old variants or the wild forms of modern crops. Backcrossing these traits into the modern strains would take decades while introducing the resistance by CRISPR/Cas9 could be done in approximately one year.

3. Animal diseases: The same approach can be used to deal with diseases of farm animals like chicken flu or swine fever, where resistance genes exist in related species and can be transferred to livestock by CRISPR/Cas9.

4. Nutritional value: Malnutrition is a substantial problem, though sufficient calories are supplied, in many regions of the world insufficient amounts of vitamins and micro-nutrients are contained in the common staple crops. Multiple genes to construct biochemical pathways for the synthesis e.g. vitamin A can be introduced into plants easily by CRISPR/Cas9.

5. Gene drive: CRISPR/Cas9 can be used for a copying (recombination) mechanism that generates homozygous individuals from heterozygous ones. This is currently being used for spreading infertility genes in disease vectors like Anopheles and Aedes. A realistic aim is to reduce the population of mosquitos by 80 to 90% and thus reduce infection by Malaria, Dengue, Yellow Fever and others.
6. Antibiotic resistant bacteria: it may be feasible to target resistance genes in pathogenic bacteria by CRISPR/Cas9 and thus solve the increasing problem of multi-resistant infections.

The advantages of CRISPR/Cas9 are that it can be universally applied, that the method is quite easy to perform and that it is inexpensive. These advantages could lead to a “democratisation of gene technology”, meaning that the method is not only available to large, financially strong companies but also to small companies and even local breeders.

These advantages may also create the risk of intentional or unintentional misuse since any well trained molecular biologist may carry out CRISPR/Cas9 constructions with quite simple and inexpensive equipment.

There are also concerns on ethical issues, e.g. the interference with ecosystems or the genetic modification of human life. Though especially the latter issue requires serious and rational discussion, we are still very far from “designer babies” with specific (complex) traits and behaviour and it is questionable if this can be achieved at all. Rules and regulations on gene technology in general and CRISPR/Cas9 in particular have to be made wisely. In Europe, especially in Germany, legislation has strangled progress in genetic engineering, companies have moved out of the country and innovation is significantly slowing down. Even worse, western political parties and NGOs try to force their anti-science opinion onto developing countries, preventing progress that is so much needed. One should always consider that using a technology requires responsibility. However, prohibiting a technology also requires responsibility. Indonesia should find its own way to make best and responsible use of this new breakthrough technology. There are many challenges in agriculture, animal breeding, nutrition and human health that can be approached by CRISPR/Cas9. Unfortunately, after more than five years, no university or research institution has yet picked the technology to pursue serious projects.

For further details: there is a huge number of excellent review papers, cartoons, videos, graphs, podcasts and discussions in various languages available on the internet. A reference list would be endless or a very subjective, arbitrary choice. Readers interested in CRISPR/Cas will have no problem to find further material on every level of knowledge.
Use of Wavelet Analyses with Potential Field Data in Exploration and Monitoring

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1. INTRODUCTION

Developed in the 1980’s, first for seismic analyses [1,2,3,4,5], the wavelet method is used since the 1990’s to analyses potential field data [6,7,8,9]. The aim of this work is to present some of the benefits when using wavelet analyses on potential field data (e.g., gravity, self-potential) when conducting geothermal exploration and volcano monitoring.

2. METHOD

Wavelets are mathematical equations that allow for analyzing either time series or spatial data set, which are used in a wide variety of domains, which include but not limited to seismic/acoustics [1], image or signal processing [9,10], fluid mechanics (turbulence) [12], archeology [13], volcano monitoring [14], or biology [15].

The wavelets analyses are organized into different categories, such as orthogonal wavelet [16], discrete wavelet analyses [20] and continuous wavelet analyses [5,8,10]. Many wavelet exists and are organized into families [8,9,10,12]. Here, we present the use of the Poisson wavelet family [8,9,13]. Wavelet analysis is a multi-scale analyses method, which is used to determine power and frequency spectrum, and distribution (space, time) of a processed signal [1,9,13,16,17].

3. RESULTS AND DISCUSSION

In 2010, we developed the multi-scale wavelet tomography approach, which use multiple wavelets to better constrain depth of source generating observed potential field anomaly [18]. Later on, we published a wavelet code using Poisson family for potential field data, named MWTmat, based on matlab platform [19]. Since, we have applied MWTmat on several projects that cover volcano monitoring [20,21] and geophysical study for geothermal exploration [18,20,22].

Our results show the usefulness of the MWTmat method that brings information on depths, location and structural shape of the source. We present examples that include but not limited to Kawah Ijen aquifer monitoring (Indonesia) [21], geothermal fluids circulation on Waita volcano (Japan) [18]. Fig. 1 presents an example of MWT method applied on gravity data to better constrain the geological structures affected by increase of fracture density due to fault movement and rock dissolution through karstification processes (study case of Jura range, Switzerland) [22].

![Fig. 1. Example of MWT analyses using Poisson wavelet on gravity data to locate rock density decrease associated to fault and karstification on geothermal exploration [22].](image-url)
4. REFERENCES


Mathematics for Solving 5G Massive Wireless IoT Networks Problems

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1. INTRODUCTION

Requirements of the fifth telecommunication generation (5G) aspects expected in 2020, according to the definition of international telecommunications union (ITU), are: (1) data rate beyond 10 Gbps, (2) massive machine-type communications, and (3) latency below 1 milisecond. This talk considers solution to massive machine-type communications, which is expected to serve about 40-50 billion devices connected to the internet, called the internet of things (IoT). Based on the contention-based access mechanism, IoT technologies are divided into four categories: (i) pure ALOHA, (ii) slotted ALOHA, (iii) non-slotted carrier sense multiple access with collision avoidance (CSMA/CA), and (iv) slotted CSMA/CA.

In this talk, we propose new categories of IoT [1]-[6] to provide better probability of success in detection, where collision is even beneficial [1]-[2]. Using the basic concept of mathematics [3]-[4], especially on probability, binomial and exponential distribution (combined with some findings in information theory), we provide better IoT technologies [5]-[6] in terms of: (a) higher throughput (serve more devices), (b) lower packet loss rate , (c) optimizable networks (using extrinsic information (EXIT) chart analysis), and the most important finding, i.e., (d) the theoretical IoT limits given multiple user/devices detection capability per time slot.

2. METHOD, RESULTS AND DISCUSSION

We use EXIT chart analysis to design the rate of each IoT devices. To make it applicable in practice, the time-slot can not be set very large. As a consequence, the stopping set happens causing performance limitation. We derive stopping sets from multinomial distribution. We found 6 stopping sets shown in Table I [4] for the case of no multiuser detection. We found that the probability is accurate enough to predict the performance of massive IoT wireless networks.

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The roles of metal ions in diabetes – metal drugs and supplements

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1. INTRODUCTION
Metal ions have diverse roles in controlling diabetes, with Cu, Zn and Mn deficiencies known to contribute to the disease through loss of control of oxidative stress, and there is debate about whether vanadium may also be an essential trace element that is involved in both insulin mimetic and insulin enhancing roles [1]. Both vanadium and chromium supplements are consumed widely for glucose metabolism and control of diabetes, and there has been considerable interest in developing anti-diabetic drugs based on these metals and also Mo and W [1]. Recently, we have shown that it is important to consider speciation of such species in cell media and other biological fluids [2], and that supposedly safe Cr(III) supplements can be oxidized to carcinogenic Cr(VI) and that there are natural pathways to prevent Cr toxicity [3,4]. This and other evidence points to neither a natural role for Cr in diabetes nor safety in its long-term consumption [1-4]. Thus, most evidence is currently focused on vanadium and to a lesser extent Mo and W. These aspects will be discussed in this talk.

2. METHOD
The speciation of metal ions within cells and tissues were investigated with X-ray absorption spectroscopy, EPR spectroscopy and UV/Vis spectroscopy [2], whereas elemental distribution in cells and tissues were measured by X-ray fluorescence microscopy [3]. Various biochemical assays including vibrational spectroscopy and imaging of glucose metabolism in cells, capillary immunoassays of cell signaling and phosphatase inhibition have been studied.

3. RESULTS AND DISCUSSION
The results of the experiments described above throw considerable doubt on the efficacy and safety of widely consumed Cr dietary supplements for the control of glucose metabolism. On the other hand, evidence has been obtained for both a potential essential role for vanadium and insights into the reasons for its efficacy as a drug, which will be discussed.

4. REFERENCES
Functionalization of Stainless Steels via Low Temperature Plasma Nitriding

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Abstract – AISI420 type martensitic stainless steels were utilized to experimentally describe the low temperature plasma nitriding behavior with respect to the inner nitriding process in matrix, the lattice expansion and straining process, the strain-induced refinement process and the phase transformation from α’- to γ-phases. The physical modeling was built from these experimental results, which was difficult to explain by the classical nitriding models. This model took account of the inner nitriding process with consideration on the occupation of nitrogen solute with high contents into the vacancy sites in the α’-lattice. The strain induced phase transformation and microstructure refinement was investigated to describe the material model in Fe(Cr) – N system.

1. INTRODUCTION

The plasma-based nitrogen processes have been high-lighted; e.g. the plasma nitriding, the ion implantation, and the plasma immersion ion implantation [1]. In particular, the DC and DC-pulsed plasma nitriding processes have been utilized in industries with the name of ion-nitriding and radical-nitriding [2]. Those conventional plasma nitriding processes are characterized by higher holding temperature than 773 K and longer processing time than 20 ks. The hardening process is also driven by the precipitation strengthening where the synthesized chromium nitride (CrN) precipitates with nano-meter size and large volume fraction in the stainless steel substrates. The inner nitriding process is mainly governed by the nitrogen diffusion; e.g., the nitried layer thickness is proportional to the square root of nitriding time [2-4]. As pointed out by [5], the high temperature plasma nitriding process for most of stainless steels and tool steels abides by this diffusion-oriented mechanism; the nitrogen solute content makes exponential decrease from the maximum content at the surface to the depth. Besides for the bound nitrogen in content in the form of CrN or FeN, the surface maximum nitrogen solute content is limited by 0.1 mass %, which is equivalent to the maximum nitrogen solubility limit in the phase diagram. In the low temperature plasma nitriding below 673 K, the diffusing nitrogen interstitial atoms never react with the chromium and iron to form CrN or FeN but occupy the vacancy sites in the fcc- and bcc-structured supercells in the stainless steel [6]. Owing to this occupation of vacancies by nitrogen solute, the original fcc- and bcc-structured lattices expand in their c-axis and contact in their a- and b-axes, respectively [7, 8]. In those previous studies, the nitrogen solute content [N] was limited at most by 20 at%; e.g., [N] = 10 at% in case of the ion implantation [9], [N] = 12 at% in case of the glow discharge plasma nitriding [10], and, [N] = 18 at% when using the low temperature plasma nitriding [11].

Authors have developed the high density plasma nitriding process to be working in low temperature [12-15]. In this low temperature plasma nitriding of martensitic stainless steels type AISI420 and austenitic stainless steels type AISI304, the nitrogen interstitials were infiltrated into the stainless steel substrates with significantly high content. Under this condition, the nitrogen interstitials occupy the vacancy sites in the α’-phase bcc structured lattices of martensitic stainless steels, and in the γ-phase fcc structured lattices of austenitic stainless steels, respectively. Due to this occupation of nitrogen solutes in the lattices with high concentration, each constituent grain is refined in the nitried layer together with high straining by the lattice expansion with and without phase transformation. Hence, the nitrogen solute diffusion process still plays an important role in this low temperature nitriding; the occupation process must be taken into account for description of this process. In the present study, new physical modeling is proposed to describe the inner nitriding process without formation of nitrides as well as the strain-induced phase transformation from α’- to γ-phases. In addition, the strain-induced refinement in microstructure is also discussed to develop the heterogeneous structuring in stainless steels.

2. EXPERIMENTAL PROCEDURE

2.1 High density plasma nitriding

The high density nitriding system has no mechanical matching box with slow response time of 1 s to 10 s to adjust the applied power. Since both the input and output powers are automatically matched by frequency
adjustment around 2 MHz, the matching response time is only limited to 1 ms at most. This prompt power control provides to make full use of mesoscopic plasma pressure range over 50 Pa. Figure 1 a) illustrates the standard RF/DC plasma nitriding system. Different from the conventional processes, the vacuum chamber is electrically neutral so that RF-power and DC-bias should be controlled independently from each other. A dipole electrode is utilized to generate RF-plasma; DC bias is directly applied to the specimens. Heating unit is located under this DC-biased cathode plate. In the following nitriding experiments, the specimens are located in the inside of the hollow as shown in Fig. 1 b) before evacuation down to the base pressure of 0.1 Pa. This hollow cathode device is effective to intensify the density of activated nitrogen atoms and NH-radicals as well as electrons toward its outlet of hollow.

![Diagram](image)

**Figure 1** High density plasma nitriding set-up. a) Standard RF/DC plasma nitriding system, and, b) Hollow cathode device to intensify the plasma density.

### 2.2 Measurement and observation

The microstructure of the nitrided layer was analyzed by scanning electron microscope (SEM). Energy dispersive spectroscopy (EDS) and electron backscattering diffraction (EBSD; HITACHI SU-70) were employed to make a precise analysis. The phase analysis was also performed by the X-ray diffraction.

### 3. RESULTS AND DISCUSSION

Figure 2 depicts the SEM image and nitrogen mapping on the cross-section of the nitrided AISI420 substrate at 673 K for 14.4 ks by 70 Pa. The original coarse grains were seen below the nitriding front end in Fig. 2 a); little grain boundaries can be detected even with the trace level in the nitrided layer. This implies that grain size should be significantly reduced during this low temperature plasma nitriding. Highly concentrated nitrogen atoms are present from 31 at% to 10 at% at the vicinity of surface down to 20 μm in depth. The nitrogen contents detected are much higher than the solubility limit of 0.08 at%; no chromium nitride (CrN) precipitates are detected. The high nitrogen concentration at the surface is responsible for driving the nitrogen diffusion process down to the nitriding front end at 80 μm in depth.

![Image](image)

**Figure 2** Cross-sectional view of the plasma nitrided AISI420 specimen at 673 K for 14.4 ks. a) SEM image of cross-section, and, b) Nitrogen mapping by EDX.

Figure 3 depicts the nitrogen content depth profile from the surface to the nitriding front end in correspondence to Fig. 2 b). In the present plasma nitriding, the nitrogen content decreases form the surface to the nitriding front end; however, the plateau of nitrogen content is observed to have almost 9 at% from 20 μm to 80 μm in depth.
In the normal nitrogen diffusion process, the nitrogen content decreases from its maximum solubility limit of 0.1 at% to the nitriding front end exponentially. This essential difference in the nitrogen depth profiles is never explained by the normal diffusion model. The evolution of the nitrided layer thickness (E) is described by the classical Wagner’s law where \( E^2 \) is proportional to the nitriding duration time (\( \tau \)). This \( E^2 \) of the nitrided AISI420 specimen at the higher temperature than 723 K was proportional to \( \tau \); in case of this low temperature nitriding, no proportionality held on between \( E^2 \) and \( \tau \).

**Fig. 3 Comparison of the nitrogen diffusion process between the high temperature and low temperature plasma nitriding treatments.**

This nitrogen diffusion process in the low temperature nitriding accompanies with occupation process of nitrogen solute atoms into octahedral vacancy sites in the \( \alpha' \)-bcc martensitic lattice structure. This occupation process is detected as an \( \alpha' \)-lattice expansion by XRD analysis. As shown in Fig. 4, the original peak for \( \alpha' \) (110) shifted itself to \( \alpha'_N \) (100) in the lower 20 direction; this large peak shift reveals that \( \alpha' \)-lattice expands by occupation of nitrogen solute into lattice.

**Figure. 4 Comparison of the XRD diagrams before and after nitriding at 673 K for 14.4 ks.**

After [16], the original diffusion partial-differential equation is modified by considering this occupation process. The chemical reaction between the vacancies and the diffusing nitrogen atoms is taken into account in formulation; the occupation process is theoretically modeled by the single-step reaction kinetics. For simplicity, the nitrogen content is normalized to be represented by \( u(t, x) \) for \( 0 < u < 1 \). Then, this new diffusion model is expressed by

\[
\frac{\partial u}{\partial t} = \frac{\partial^2 u}{\partial x^2} + \left(au - bu^2\right), \quad a, b > 0 \tag{1}
\]

where the second term in its right hand side denotes for the occupation process, and, \( a \) and \( b \) are material constants. If \( a \) and \( b \) are assumed to be unity, Eq. (1) reduces to the Fisher – Kolmogorov equation. Hence, this solution of Eq. (1) predicts that the nitriding front end advances in the depth with a wave velocity.

Beside for the \( \alpha' \)-lattice expansion, the \( \gamma \)-phase peaks were seen in Fig. 4. The unknown peak around 20 = 38° was identified to correspond to the Fe – N bonding state; more precise analysis is necessary in future. The physical analysis by EBSD works to describe this phase transformation from \( \alpha' \)- to \( \gamma \)-phases during the plasma
nitriding. Figure 5 shows the relationship among the crystalline structure, the staining and the phase mapping. As shown in Fig. 5 b), the highly strained network is formed into the matrix by $\alpha'$-lattice expansion in each grain. Corresponding to this network, the phase transformation to $\gamma$-phase is induced in the network structure in Fig. 5 c). In parallel with this high straining, the grain size is significantly refined to be invisible by EPSD-analysis at the vicinity of the surface in Fig. 5 a).

4. CONCLUSIONS

The low temperature plasma nitriding provides a method to describe the structure of $\alpha'$- or $\gamma$-lattices in the stainless steels in the wider range of nitrogen contents up to 30 at%. Two phase structure such as ($\alpha'$, $\gamma$) is induced by high straining during this nitriding. Furthermore, the original coarse grain structure is refined by this high straining to have ultra-fine grain sizes. This low temperature nitriding also becomes a tool to make heterogeneous structuring for functionalization of stainless steels.

5. ACKNOWLEDGEMENTS

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6. REFERENCES


![Fig. 5 EBSD analysis. a) IPF-mapping, b) KAM distribution, and c) Phase-mapping.](image-url)
The 7th Basic Science International Conference

Basics Science for Improving Survival and Quality of Life

Invited Papers
Complexity and Nano Science Approach in Life Sciences: The way to overcome our partial understanding on living system

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Abstract – Universe is organizational system built from complex interactions and interconnections of components where biological system is complex structure with specific function dedicated to perform normal ordered organizational system. Developing Complexity Science and Nano Biological perspective giving the ideas of interfacing between modern physical and biological sciences into more comprehensive understanding of life system. The ideas initiated for more than 7 decades by physicist Erwin Schrodinger in a 1944 book What is Life? The argument that life feeds on negative entropy or negentropy was asserted and may initiate the way to more comprehensive and better understanding life. We can then develop understanding biological behavior on nano size biological materials and its higher order using modern physics. While other physicist namely Firtjof Capra give another stimulating definition of biological system that enable to apply thermodynamic law. He defined that life is the ceaseless flow of energy and material through complex chemical interaction namely metabolism to perform self organization, perpetuation, regeneration, reparation as well as movement. These ideas are interesting while most people are not even conscious that biologists when they coming into nano size molecular biological discussion, they remain using what is called Newtonian principles respecting the mechanical view of reality when they think and talk. This Newtonian based principles, in order to fulfil the scientific methodologies, many biological scientists including who works in applied sciences such as edical Sciences, they practiced reductionism, reducing things into their parts and examining the parts to understand what made them tick. They reduced life to cells, molecules while the concepts is remain within Newtonian. In the early 20th Century, the certainty of Newton’s mechanics was undermined by quantum mechanics and the Uncertainty Principle developed by Werner Heisenberg. This is the new challenges to the old Newtonian view of reality. So some biological scientists began abandoning the Newtonian worldview, while most of them and the ordinary people held on to it.
Surface Modification for Quartz Crystal Microbalance using Polystyrene as a Basis for Biosensor

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*Corresponding authors: [sakti@ub.ac.id]

Abstract – Surface modification of the Quartz Crystal Microbalance is one importance factor in the application of the sensor as a biosensor. The property of the surface physically must not affect the performance of the sensor. It is indicated by a minimal damping of the sensor, while biochemically the sensor surface must be able to bind or adsorb the biomolecules being immobilized as a sensitive layer of the biosensor. Polystyrene is one from many materials which can be used as a sensor coating for the basis of the biosensor as the polystyrene can be used as a matrix for immobilizing antigen or antibody. Polystyrene coating on top of the QCM sensor can be done by using spin coating, airbrush spray coating, and ultrasonic spray coating. Those different methods result in a different surface structure. Good coating using glassy material will not increase the electrical impedance of the sensor. Therefore it is suitable to be used as a mass sensitive biosensor. In addition, modification of the polystyrene surface can be done to alter the surface hydrophobicity. UV radiation and plasma treatment can effectively alter the polystyrene surface hydrophobicity without affecting the electrical impedance of the QCM sensor which can be influenced by the acoustic impedance characteristic of the coating layer. Surface hydrophobicity plays an important role in the used of the QCM biosensor which detects the viscosity and density of the target molecule in liquid. Polystyrene surface with hydrophilic surface decreases the resonance frequency of the QCM biosensor in contact with liquid more than the decreasing resonance frequency on a hydrophobic surface.

1. INTRODUCTION

Since the first Quartz Crystal Microbalance has been used, the application of the quartz crystal microbalance can be found in many areas. The interesting aspect of the sensor is its ability to works in gas and liquid. The sensor responds to physical, chemical and biological quantity results in a frequency change or impedance change. The wide use of the QCM sensor for chemical and biological sensors are mostly based on the sensor responds to mass change on its surface, and it responds to viscosity and or density of contacting material on the sensor surface 1. The first respond is described by Sauerbrey equation 2 and the second respond by Kanazawa and Gordon 3.

The electrical behavior of the QCM sensor and its coating and loading can be modeled as an electrical circuit model (RLC model) 4,5. The model is well known as a Butterworth van Dyke (BvD) model. The electrical model is presented in Figure 1a. The right part of the model is the electromechanical equivalent model of the piezoelectric property of the sensor, while the capacitance in the left is the capacitance of the sensor as a parallel plate capacitor with the quartz as the dielectric material.

![Figure 1. Electrical equivalent model of the QCM sensor without coating (a) and with coating (b)](image-url)
The coating layer and liquid behaviour is also modelled as a resistive and inductive elements in the extended BVD model. The model is depicted in Figure 1b. The load contribution of the coating layer can also be expressed in an RLC model. The resistive part of the model shows a dissipative behavior of the sensor and the coating material. A glassy coating material does not contribute to the resistance equivalence value, and those the value of the resistive element is close to zero. The coating material behaves only as a rigid mass on top of the sensor which can be expressed only by an inductor element. In the impedance spectrum of the sensor, the glassy coating material causes the series resonance frequency of the sensor goes down, and the minimum impedance of the sensor at the series resonance remains. In contrast, a rubbery coating material lowers the series resonance frequency of the sensor together with an effect of increasing minimum impedance of the series resonance sensor and decreasing maximum impedance at the parallel resonance. It was studied that the glassy and rubbery property of the coating material is determined by the shear moduli of the coating material. Also, the thickness of the coating material affects the property of the material. A very thin coating most likely behaves as a glassy material at high frequency, for example at 10MHz.

The rubbery coating material is not preferred to be used for QCM sensor because it contributes an additional damping to the sensor indicates by the increasing impedance of the coated sensor at series resonance. In the worst condition, the additional damping causes the sensor to stop oscillating. Therefore it is important to maintain the low damping caused by the coating materials. The low damping is indicated by a non-significant or zero increase of the sensor impedance at series resonance.

Polystyrene is one from many polymers which can be used as a coating material for QCM biosensor. The high shear moduli of the polystyrene make the polystyrene coating behaves as a glassy coating material. Those it is expected that a polystyrene coating on QCM sensor does not add damping to the sensor. The surface property of the polystyrene can be modified by physical and chemical modification. Method to deposit the polystyrene on top of the QCM sensor affects the surface roughness of the sensor. UV radiation to the polystyrene coating alters the hydrophobicity of the polystyrene surface. Plasma treatment can also change the hydrophobicity.

2. COATING FOR MASS SENSITIVE BIOSENSOR

Those indicated that the polystyrene coating is a good matrix layer to immobilized sensitive biomolecule for QCM biosensor. The high shear moduli of the polystyrene make the polystyrene coating behaves as a glassy coating material. Those is it expected that a polystyrene coating on QCM sensor does not add damping to the sensor. The sensor surface needs to be modified to make the sensor responds only to a specific quantity target. Surface modification of the QCM sensor using polystyrene can be done easily and targeted for many difference applications. The polystyrene coating on the sensor can be used as immobilization matrix for a biomolecule to be used as an immunosensor for various target molecules such as insulin, human serum albumin, a specific protein of cow milk, matrix metalloproteinase-3 (MMP-3) antibody and many others.

The simple method of coating development using polystyrene on top of the QCM sensor is done by spin coating, air pressure spray coating, and ultrasonic spray coating. Proper selection of the coating method results in a homogeneity coating thickness with no damping contribution to the QCM sensor. However, come condition of the inhomogeneous coating exists caused by preparations and coating process. Inhomogeneous coating thickness raises the minimum impedance at series resonance of the QCM sensor.

Spin coating is the simplest method to make a polystyrene coating on top of the QCM sensor with HC-49U form. Rotation speed, concentration, and solvent selection can be done to achieve the desired film thickness and surface roughness. The surface roughness of the polystyrene which was deposited using spin coating method is affected by the solvent. Polystyrene coating which was deposited using spray coating (airbrush) resulted in more roughness. However, optimization of some parameter of the coating such as solvent, pressure, and the distance between the nozzle and the sensor surface need to be optimized to get a homogeneous surface. The same action is also required when the deposition is done using an ultrasonic deposition method.

The surface roughness of the coating, however, does not contribute any additional damping to the sensor. Inhomogeneity of the coating deposition can add damping to the sensor. By measuring the electrical impedance of the sensor, one can identify whether the coating layer is electromechanically good or bad. A good coating layer will not add additional impedance in the measured impedance spectrum, while a bad coating results in an increasing minimum impedance of the sensor at series resonance.

Figure 2 shows impedance curves of a QCM sensor before and after coating. Both were coated using polystyrene. In the left figure, the minimum impedance of the sensor at series resonance after coating does not increase. Before and after coating the minimum impedance is around 10Ω. On the right side, the impedance of the sensor with the coating is around 38Ω. The frequency change of both sensors caused by the coating layer is around 27KHz. The increasing minimum impedance of the sensor in the right figure is caused by inhomogeneity of the coating.
Figure 2. Impedance spectrum of QCM sensor with coating with no additional damping (left) and with additional damping (right)

Immobilized The surface roughness of the coating, however, does not contribute any additional damping to the sensor. Inhomogeneity of the coating deposition can add damping to the sensor. By measuring the electrical impedance of the sensor, one can identify whether the coating layer is electromechanically good or bad. A good coating layer will not add additional impedance in the measured impedance spectrum, while a bad coating results in an increasing minimum impedance of the sensor at series resonance.

3. COATING FOR LIQUID VISCOSITY AND DENSITY CHANGE

Hydrophobicity of the polystyrene surface affects the loading properties of the liquid to the sensor response. This result in a different frequency change of the sensor at the same viscosity caused by different hydrophobicity. Biological sensor by detecting viscosity change of the sample liquid caused by interaction between the substance in the liquid and target molecule was demonstrated for endotoxin detection using QCM sensor 11. In those works, the change in the hydrophobicity of the surface resulted in a significant effect on the sensor response. A higher signal was obtained using a hydrophilic surface.

For the application where the detected target sample will result in viscosity and density change of the liquid on top of the QCM sensor, the surface property of the coating layer needs to be modified to effectively couples the liquid and the sensor. The good coupling is required to transfer the energy of the vibrating quartz to the liquid. The surface property of the sensor should be modified to minimize the slip effect of the liquid 12,13. The mechanical coupling between the surface and the liquid affects the frequency change of the sensor caused by the liquid viscosity and density change as described by Kanazawa and Gordon.

Different surface hydrophobicity can be achieved using many different approaches. One can use different materials or by modifying the surface property of the material by chemical or physical treatment. Polystyrene is one material where its surface hydrophobicity can be changed physically and chemically. The surface hydrophobicity of the polystyrene can be change to be more hydrophilic by UV radiation or by using plasma treatment. Both modifications do not change the bulk property of the polystyrene coating. Therefore the modulus elasticity of the polystyrene coating remains. Figure 3 shows an example of impedance curve of a QCM sensor with polystyrene coating before and after UV radiation. It can be seen that the minimum impedance of the sensor remains.

Figure 3. Impedance spectrum of QCM sensor with coating with no additional damping (left) and with additional damping (right) before and after UV radiation

A significant change of the impedance curve can be observed when the sensor surface is in contact with water. The minimum impedance of the sensor in contact with water increases significantly after the Polystyrene coating treated with UV radiation and become a hydrophilic surface (Figure 4). When the polystyrene surface become hydrophilic, the shift of the resonance frequency is higher, and the minimum impedance increases. Therefore for
a biosensor which uses a liquid viscosity and density change of the water-based solution needs a hydrophilic surface to get a higher signal change.

![Figure 4. Impedance spectrum of QCM sensor in contact with water](image)

### 4. CONCLUSIONS

Polystyrene can be used as a coating material for QCM sensor. The mechanical property of the polystyrene as a glassy material gives an advantage to the polystyrene coating which does not add additional damping to the sensor. Meanwhile, the surface property of the polystyrene can be modified to make the surface as a good matrix for biomolecule immobilization and to make a better coupling between the sensor and water solution containing target molecule.

### 5. REFERENCES

**Structure and Dynamics of Water: An Insight from Molecular Simulation**

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**Abstract** – While being termed as the "matrix of life", in which the majority of important biochemical reactions took place, water is the most puzzling substance and does not share many properties with other liquids [1]. Upon cooling and/or pressurized, water has at least 14 crystalline polymorphs in pure state [2]. This number increases with the presence of small hydrophobic molecules that can be encaged in a network of water molecules that are connected by hydrogen bonds [3,4]. A deep understanding on the structure and dynamics of water is of paramount importance in life science, cryobiology, climate prediction, and material design. Molecular simulation methods provide an access to experimentally difficult conditions such as extreme temperature and pressure or metastable state; thus provides possibilities to discover novel phenomena and peculiar character of a chemical system [5–7]. The methods enable a systematic investigation at nanoscale level with a great numerical detail. In the absence of technical difficulty in preparing certain material, molecular simulation methods facilitate creative minds to tweak the system with more freedom, and the results are useful to interpret the nature and to design innovative materials. Here we will introduce a number of our molecular simulation works in attempt to unravel the structures and dynamics of a water dominated system.

**REFERENCES**

Electrochemical Sensor for Industry and Medical

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Abstract – Electrochemical sensor had been developed for many purposes. This type sensor mostly can be easily adopted to solve the problem in industry and medical with a high degree of accuracy, precision, sensitivity and selectivity. This study we review a highlight recent in the using of electrochemical sensor for industry and medical. We developed a spicy sensor based gold nanoparticle for industry, which can be used for spicy analysis of any kind food. Both of anodic and cathodic can be used for spicy measurement with limit of detection of the sensor at about 0.500 μM. The sensitivity at anodic and cathodic is 0.102 and -0.268 μA.μM⁻¹.mm⁻² respectively. The spicy sensor has been applied to detect the quantity of capsaicin at the seed and fruit of red pepper. We also fabricate and utilize the extract of Maja (Aegle marmelos) leaves and pulp to modify silver electrode for medical biosensor. The maja leaf extract modified silver electrode gives a good performance as a urea biosensor at pH 13 with LOD and sensitivity are 2.299 μM and 19.781 μA.μM⁻¹.mm⁻² (for anodic) and 5.165 μM and 38.177 μA.μM⁻¹.mm⁻² (for cathodic) respectively. Whereas, the maja pulp extract modified silver electrode can be used as glucose biosensor. The biosensor has a good LOD and sensitivity for glucose detection, i.e., 1.920 μM and 1.222 μA.μM⁻¹.mm⁻² (for anodic) and 1.701 μM and 1.522 μA.μM⁻¹.mm⁻² (for cathodic) respectively. The urea and glucose biosensors were also proven have good selectivity.

1. INTRODUCTION

Jaroslav Heyrovsky, Czech scientist discovered voltammetry which is a form of electrochemistry in 1920s. This technique can analyze a samples by measuring current as a function of the applied potential [1]. In recent years, the sensor technology was developed using electrochemical technique. This sensor commonly used because it was relatively fast, simple and low-cost. Therefore, the electrochemical sensor have been widely applied in industry and medical [2–6].

Improvement of selectivity and sensitivity of the sensor can be conducted by modify the electrode surface using definite material. Most of the electrodes that modified are platinum, gold and silver. The modification on the electrode surface give general result in the:

1. Transfer of physicochemical properties of the modifier to the electrode
2. Enhanced electrocatalytic activity due to the use of materials with large surface area which in turn allows better sensitivity
3. Selectivity towards analyte due to immobilized functional groups and dopents
4. Fast diffusion kinetics in case of some materials
5. Extraction and accumulation of an analyte at the electrode surface [7].

Based on the above explanation, the scientist already modify many electrodes to increase the sensor performance [8–12]. In this article, we reviewed our electrode modification as a sensor for industry and medical purpose. Our sensor was proven have a good limit of detection (LOD), selectivity and sensitivity. For industry purpose, we develop a gold nanoparticles for spicy sensor. We also utilize the Indonesian original plant (i.e., Maja (Aegle marmelos)) to modify silver electrode for urea and glucose biosensor. The performance of our modified electrode will be compared with another works.

2. APPLICATION ELECTROCHEMICAL SENSOR FOR INDUSTRY

2.1 SPICY SENSOR

Chili is one of important commodity in the world, include Indonesia. In the period 2000-2014, Indonesia is able to export an average of 8.38% per year for fresh chili and 31.74% per year for processed chili such as sauce. Determination of the pungency level of chili is needed, especially in the export-import industry. The pungency level of chili is related to the presence of capsaicinoid compounds [10,13–16]. Monitoring of this compound is
important because excessive intake of capsaicinoid may be harmful to health [15,17]. In fact, the Council of Europe has recommended a certain limit to the total capsaicinoid content permitted in a few categories, for instance, 5 ppm as the general limit for food and beverages, 10 ppm for spicy food and beverages, 20 ppm for hot sauces, and 50 ppm for tobacco, harissa, pimento hot oil, and so forth [15].

Conventional methods used to determine the pungency level or capsaicin concentration are using tongue by a panel of tasters (Scoville Organoleptic test method). This test is leveled in Scoville, which is the spiciness measurement of 30 chilies or its derivatives. Pure capsaicin itself has 16 million Scoville. Currently, the industrial Scoville test measurement becomes the standard of spicy flavor, but it is less accurate due to its subjectivity [18]. Quantitative methods such as spectrophotometry and high performance liquid chromatography (HPLC) has been used for the detection of capsaicinoid. HPLC results are reported in spicy ASTA units (15 multiple converted into Scoville) and claimed more reproducible than the conventional method. Unfortunately, this technique requires high cost and complex preparation. Therefore, a simple and accurate technique is still needed to detect and determine capsaicinoid.

Electrochemical method have been reported for fast, simpler and accurate technique. The first claimed of this method for capsaicin analysis is reported on the patent number WO 2009/115840A1. The active material used is double walled carbon nanotubes. Fabrication double walled carbon nanotube is relatively difficult since it requires relatively expensive. We provides another alternative active material that work in the similar system but using simpler process, i.e., composite polyamide 11 and gold nanoparticles. Synthesis of gold nanoparticles was done using wet method, which is simpler than the making of double walled nanocarbon active materials.

![Fig 1. Polyamide composite electrode-gold nanoparticle.](image)

In general, the composition of our modified electrode can be seen at Fig. 1. The surface of gold electrodes was coated by gold nanoparticles (Fig. 1a). The edge of the gold electrodes was isolated (Fig. 1b) and make the bottoms were opened. Deposition of polyamide 11 on the gold electrode surface is carried out using LbL (Layer by Layer) technique. Solution of polyamide 11 appeared by dissolving 0.2 grams powdered polyamide 11 with 7 mL of concentrated H2SO4. Gold electrodes immersed for 5 seconds in a solution of polyamide 11 and was directly included in the distilled water to rinse. The electrode was dipped in the suspension of gold nanoparticles for 24 hours (Fig. 1c), then rinsed again with distilled water. The electrodes are ready to analyze, after dried. The characterization of electrode surface by microscope optic was presented at Fig. 2.

![Fig 2. Gold electrode surface before (a) and after deposited by polyamide 11 (b), and the surface of modified gold electrode with polyamide 11-gold nanoparticles.](image)

The combination of polyamide and gold nanoparticles are able to oxidize capsaicin, which cannot be done by gold electrodes. The peak of oxidation and reduction are observed at 0.439 V and 0 V, respectively. The detection limit (LOD) of electrode polyamide 11 gold nanoparticles at 0.439 V reaches up to 0.5 μM. Linear concentration range that can be used as a calibration curve is in the region of concentration 220 μM. In linear regression equation of the calibration curve obtained y = 6.75 + 0.08 x with a value of R of 0.99468, so the sensitivity of electrodes
According to Table 2, our Maja leaf extract appears glucose. This result proves that the el at Fig 5. It shows that the Maja leaf extract-modified silver electrode can detect urea without any interference from glucose. This result proves that the electrode have a good selectivity. The cathodic and anodic current signals are observed at 0.4 V and 0.625 V, respectively. The performance of the electrode also proven with LOD and sensitivity. According to Table 2, our Maja leaf extract-modified silver electrode is better than the other studies.

Comparison with the patent number WO 2009/115840A1 indicates that the polyamide11-gold nanoparticles modified electrode has nearly the same value of LOD (Table 1). Thus, the polyamide11-gold nanoparticles modified gold electrode is demonstrated as an alternative spicy sensor for industrial purposes.

![Fig 3. Calibration curve of anodic.](image)

![Fig 4. Calibration curve of cathodic.](image)

<table>
<thead>
<tr>
<th>Electrode</th>
<th>Linear range (µM)</th>
<th>LOD (µM)</th>
<th>Sensitivity (µA.µM⁻¹.mm²)</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Modified MWCNTs</td>
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<td>0.45</td>
<td>-</td>
<td>[19]</td>
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<tr>
<td>Modified gold</td>
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<td>0.500 (for anodic and cathodic)</td>
<td>0.102 (anodic and cathodic)</td>
<td>-0.268 (cathodic)</td>
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</table>

2.2 APPLICATION ELECTROCHEMICAL SENSOR FOR MEDICAL

Since medical analysis in laboratory are expensive and time-consuming process, more measurements of analytes are performed in various locations, including hospital point-of-care setting, by caregivers in non-hospital settings and by patients at home. Today one of main challenges is the development of methods to perform these rapid ‘in situ’ analysis. These methods must be sensitive and accurate, and able to determine various substances with different properties in ‘real-life’ samples. Electrochemical sensors for the measurement of analytes of interest in medical analysis are ideally suited for these new application, due to their high sensitivity and selectivity, portable field-based size, rapid response time and low-cost. We already develop and use the extract of leaves and pulp of Maja (Aegle marmelos), Indonesian original plant, to modify the silver electrode and used it as urea and glucose biosensors. Similar with spicy sensor, we also provide a simpler process to urea and glucose detection.

2.2.1 UREA SENSOR

Urea is widely distributed in nature and its analysis is of considerable interest in clinical and agricultural chemistry. It is known to be an important marker for evaluating uremic toxin levels. The normal level of urea in serum is from 15 to 40 mg/dl (2.5–7.5 mM/l). In patients suffering from renal insufficiency, urea concentrations in serum vary from 180 to 480 mg/dl and, at elevated levels above 180 mg/dl, hemodialysis is required [8,12]. Hence, urea detection is important. In our study, we presented another alternative method for urea detection, that is electrochemical non-enzymatic biosensor using the extract of Maja leaf [5].

The composition of the electrode was reported in our previous paper [5]. The response of electrode was presented at Fig 5. It shows that the Maja leaf extract-modified silver electrode can detect urea without any interference from glucose. This result proves that the electrode have a good selectivity. The cathodic and anodic current signals are appeared at 0.4 V and 0.625 V, respectively. The performance of the electrode also proven with LOD and sensitivity. According to Table 2, our Maja leaf extract-modified silver electrode is better than the other studies.
2.2.2 GLUCOSE SENSOR

Development of simple, reliable and fast methods for glucose detection is important, especially for medical diagnostics. The methods useful to determination of glucose in the blood for diabetes screening and treatment. Many researcher reported and develop the electrochemical glucose sensors and biosensors both in enzymatic or non-enzymatic form. Enzymatic sensors normally have a short life due to the properties of the enzyme, which is very easy to decompose. Therefore, we used Maja pulp extract to modify silver electrode for non-enzymatic glucose biosensor [4].

The composition of the electrode was reported in our previous paper [4]. The response of electrode was presented at Fig 6. It shows that the Maja pulp extract-modified silver electrode can detect glucose. The signals of cathodic and anodic current discovered at 0.38 V and -0.25 V, respectively. There is no interference signal from urea and ascorbic acid. The performance of our Maja pulp extract-modified silver electrode was compared with other studies, which summarized at Table 3. It justifies that our electrode show a better performance than other research.

![Graph showing response comparison between silica gel and silica gel/Maja leaf extract to glucose solution and urea at pH 13 condition.](image)

**Table 2. Comparison of the non-enzymatic urea biosensors.**

<table>
<thead>
<tr>
<th>Electrode</th>
<th>Linear range (µM)</th>
<th>LOD (µM)</th>
<th>Sensitivity (µA.µM⁻¹.mm²)</th>
<th>Reference</th>
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</tr>
<tr>
<td></td>
<td>2-7 (cathodic)</td>
<td>5.165</td>
<td>38.177 (cathodic)</td>
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</tr>
</tbody>
</table>
Fig 6. Response comparison between silica gel and silica gel/Maja pulp extract to glucose solution and urea at pH 13 condition.

Table 3. Performance comparison of the non-enzymatic glucose biosensors.

<table>
<thead>
<tr>
<th>Electrode</th>
<th>Linear range (µM)</th>
<th>LOD (µM)</th>
<th>Sensitivity (µA.µM⁻¹.mm⁻²)</th>
<th>Reference</th>
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<td>0.002</td>
<td>[21]</td>
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<tr>
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<td>820</td>
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<td>9</td>
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<tr>
<td></td>
<td>1.701 (cathodic)</td>
<td></td>
<td>1.522 (cathodic)</td>
<td></td>
</tr>
</tbody>
</table>

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Polaraniline-Modified Zeolite NaY: A New Sorbent for Dispersive Solid Phase Extraction of Multiclass Pesticides

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1. INTRODUCTION

Pesticide residues are highly concerned as one of contaminants in environment, foods and agricultural products since their applications are continually expanding and their consumptions are ever increasing. Trace analyses of these substances require analytical techniques for the detection of the greatest number of compounds possible, with the fewest number of extraction and clean-up steps. The analytical techniques for these trace residues usually consist of sample preparation and quantification using analytical instruments. Our work is intended to developed new materials for preconcentration of chemical residues as well as to propose a sample preparation procedure [1]. A novel polyaniline (PANI)-modified zeolite NaY was proposed as sorbent for the dispersive solid-phase extraction (DSPE) of diverse pesticide residues [2].

2. METHOD

The PANI-coated zeolite NaY sorbent was created via oxidative polymerization of aniline onto the surface of the zeolite. The sorbent was applied for DSPE of commonly used pesticides belonging to five different chemical groups, including carbamate, organophosphate, sulfonylurea, pyrethroid and neonicotinoid. To perform the DSPE, 150 mg sorbent was added to 125 mL of sample. A suspension was then shaken for 4 min to promote the sorption of the analytes onto the sorbent before transferring to the SPE eluting column. The column was washed with 10 mL water before eluting with an appropriate eluent. The final extract was analyzed by HPLC-PDA.

3. RESULTS AND DISCUSSION

The applicability of synthesized PANI-modified zeolite NaY as a sorbent for multiclass pesticides extraction was investigated. An average sorption capacity of 833 mg kg⁻¹ sorbent was reached. The experimental conditions of the DSPE procedure were optimized. The coupling of DSPE and HPLC resulted in an efficient method for multiresidue analysis of pesticides. The preconcentration factor of up to 42 was observed. The LODs and LOQs of the proposed method were found in the ranges of 0.001–1.00 mg L⁻¹ and 0.005–2.50 mg L⁻¹, respectively. The method was successfully applied to the determination of pesticide residues in environmental and food samples. The satisfactory recoveries were obtained.

4. REFERENCES

Mathematical Model of a Growing Tumor and Its Interaction with Immune System: The role of dendritic cell in controlling the immune system

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1. INTRODUCTION

Immune system such as natural killer (NK) cells and cytotoxic T cells are capable of recognizing and killing tumor cells. In recent years, it is found that dendritic cells (DCs) also have a potential role in controlling a tumor growth, besides their function as a regulator of immune system [1]. This talk relates to a mathematical model of a growing tumor and its interactions with the immune system that is emphasized in the role of dendritic cells in controlling immune system. First, we construct a mathematical model in the form of system of ordinary differential equation. Then, existence of the equilibrium points and their stability are analyzed. We also investigate which parameters that play important role in this model. Second, we develop a hybrid cellular automata model that can describe the system in more detail, including cell-cell interactions of every single cell in the system. Besides providing more detail of the immune system, this model also include the chemokine which function to activate dendritic cells. Furthermore, to include the effect of chemokine, we use a partial differential equation to describe the concentration of chemokine.

2. METHOD

First, we construct the model based on some assumptions stated in [2]. This model consists of tumor cells, NK cells, cytotoxic T cells, DCs and chemokine in the form of system of nonlinear ordinary differential equations. The stability analysis is done by linearizing the system near equilibrium points and Routh-Hurwitz criterion is then used. In the second model, we combine the numerical solution of partial differential equation model with the discrete cellular automata used to model individual cells [3]. Finally, some numerical simulations are conducted to illustrate the behavior of the solution of both models.

3. RESULTS AND DISCUSSION

Based on analysis result, the first model has two equilibrium points which exist and asymptotically stable under certain conditions. It is also obtained that the parameters such as tumor growth, NK cells tumor cell kill rate, death rate of NK cells and source term of NK cells play important role in the growth of tumor.

Hybrid cellular automata model can describe the evolution of the tumor cells both in space and in time. From simulation, increasing the concentration of DCs will decrease the concentration of tumor cells.

4. REFERENCES

Spatial Panel Dynamic Econometrics Model of Land Value, Land Use Externalities and Their Dynamic: Case Study of the Jakarta’s Fringe

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Abstract – Land value is the product of past decision of its use leading to its value. It is also affected by the local characteristic and the observed surrounded land use (externalities) from the previous period. The effect of each factor on land value has dynamic and spatial virtues. An empirical land value model with a spatial panel dynamic setting is appropriate to capture them. The model will be useful to estimate the extent of land use externalities on land value in the short run as well as in the long run. It serves as a basis to formulate an effective urban growth management’s policy. The objective of this paper is to derive the indirect effects – externalities on land value and their dynamic. The result will be applied to analyze the significance of land use externalities on land value in the fringe of Jakarta Metropolitan. There is some evidence about the significance of neighborhood urban activity – negative externalities, the previous land value and local accessibility on land value. The effects are accumulated dynamically over years, but they will fully affect the land value after 5 until 6 years.
How Data Science Shapes Personalized Medicine Revolution

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Abstract – Personalized Medicine coined firstly in 1969 is defined as tailoring of medical treatment to the individual characteristics. This field has made significant progress with key discoveries boosted by rapid development of new biotechnologies such as Microarray Chip and Next Generation Sequencing (NGS).

From the completion of Human Genome Project 2003 scientist in this field have produced billions of data points on properties, structure and functions of genes, proteins and other molecules are compiled in enormous databases and systematically studied. As the price is getting cheaper, the growth of the data on genomes, proteins and cells would be unstoppable by approximately 25 petabyte/year worldwide.

Dealing with this “big data” requires a different approach and poses challenges for data scientist in retrieving, managing, visualizing, analyzing, interpreting and presenting the data. Conventional statistical approaches can no longer be used. Numerous advanced statistical techniques have been proposed to understand and extract more insight and information of the biological data.

The objective of this article is to review and describe the role and increasing importance of data scientist in the research of personalized medicine, as well as discuss the challenges and some current topics.

Furthermore, this article would discuss the development of various advanced statistical techniques from simple to the latest implemented in Bioinformatics in shaping the revolution of personalized or precision medicine. Wide range approaches in classification and clustering techniques used in Microarray and NGS would be presented. Future development and research direction would be discussed as well.

In conclusion, data scientists not only have major role in all aspects of discovering precision medicine but also would be able to speed up the translation of all findings to the clinical practice.
The 7th Basic Science International Conference
Basics Science for Improving Survival and Quality of Life

Botany
Antibacterial Activity of Endophytic Fungi Isolated from *Talinum paniculatum* (Jag.) Gaertn.

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**Abstract** – Some endophytic fungi species could produce an antibacterial active compound, so that the fungi could potentially be used as natural antibiotics substance producer. Seven species of endophytic fungi have been isolated from *Talinum paniculatum* (Jag.) Gaertn. plant tissue, i.e: *Colletotrichum acutatum*, *Colletotrichum coccodes*, *Colletotrichum gloeosporioides*, *Xylohypha sp.*, *Fusarium semitectum*, *Fusarium lateritium*, and *Aspergillus candidus*. This research is done with purposes: 1) to examine the antibacterial activity of each endophytic fungi secondary metabolite towards *Escherichia coli* and *Bacillus subtilis*; 2) to determine the endophytic fungi species that have the highest antibacterial activity towards *Escherichia coli* and *Bacillus subtilis*; 3) to analyze the secondary metabolite content in each endophytic fungi liquid culture. Each endophytic fungi species was isolated and inoculated in Potato Dextrose Broth (PDB) medium shaken in the rate of 120 rpm during 7x24 hours, then filtered, and centrifugated in the rate of 300 rpm during 10 minutes. The antibacterial activity of each fungi metabolites towards *Escherichia coli* and *Bacillus subtilis* were examined using disc diffusion method. The research results show: 1) each endophytic fungi species have antibacterial activity towards *Escherichia coli* and *Bacillus subtilis*; 2) *Aspergillus candidus* has the highest antibacterial activity towards *Escherichia coli* and *Bacillus subtilis*; 3) Each endophytic fungi species could produce secondary metabolites: alkaloid, flavonoid, terpenoid, and tannin.

1. INTRODUCTION

The endophytic fungi live in the internal healthy plant tissues beneath the epidermal layers, without causing damage to the host plant[1]. The fungi have an ability to colonize within the intercellular space of the tissue. Some endophytic fungi have been identified. *Trichoderma viridae* and *Alternaria* sp. are endophytic fungi species that found in Zingiber nimmoni root[2]. There also found some endophytic fungi, i.e: *Aspergillus candidus*, *Aspergillus terreus*, *Aspergillus versicolor*, *Penicillium chrysogenum*, *Fusarium avenaceum*, *Alternaria humicola*, *Alternaria japonica*, and *Mycelia sterilia* in *Ocimum sanctum* leaf[3]. Some endophytic fungi also found in medicinal plant, Azadirachta indica, i.e: *Acremonium sp.*, *Cladosporium sp.*, *Curvularia lunata*, *Fusarium oxysporum*, *Fusarium solani*, *Nigrospora oryzae*, *Phoma euprena*, *Phyllosica sp.*, *Trichoderma sp.*, and *Verticillium alboatrum*[4].

Java ginseng (*Talinum paniculatum* (Jag.) Gaertn.) is a medicinal plant. This plant has some function, i.e: antiinflammation on skin, antifefection on gastrointestinal disease[5], and antioxidant[6]. Some endophytic fungi have been isolated in preliminary research. Seven endophytic fungi species isolated from *Talinum paniculatum* (Jag.) Gaertn. plant are: *Fusarium semitectum*, *Aspergillus candidus*, *Colletotrichum accutatum*, *Colletotrichum coccodes*, *Fusarium lateritium*, *Xylohypha sp.*, and *Colletotrichum gloeosporioides*.

Endophytic fungi produce some anti-microbe compound, so the endophytic fungi could be used for natural antibiotic production purpose. Many endophytic fungi have been reported to produce novel antibacterial, antifungal, antiviral, anti inflammatory, anti tumor, and other compounds belonging to the alkaloids, steroids, flavonoid and terpenoid[7][8]. The medicinal plants where the endophytic fungi live could produce the same antimicrobe compounds. Thus, if the antimicrobe compounds can be acquired from endophytic fungi, it could eliminate the need to harvest and extract the slow growing and relatively rare trees. The antimicrobe compounds can be obtained from the endophytic fungi liquid cultures.

Secondary metabolite produced by this plant contains: saponin, terpenoid, polyphenol, flavonoid, and tannin[9]. It is important to examine the compounds in secondary metabolite produced by each endophytic fungi species from *Talinum paniculatum* (Jag.) Gaertn. Tissue, to know, whether, the endophytic fungi could also be used to examine the antibacterial activity of each endophytic fungi species metabolite. This research is done to: 1) examine the antibacterial activity of each endophytic fungi species metabolite towards *Escherichia coli* and *Bacillus subtilis*; 2) determine the endophytic fungi species that have the highest antibacterial activity towards *Escherichia coli* and *Bacillus subtilis*; 3) to analyze the secondary metabolite content produced by each endophytic fungi species.
2. METHODS

2.1 Materials

Endophytic fungi species isolates: *Fusarium semitectum*, *Aspergillus candidus*, *Colletotrichum accutatum*, *Colletotrichum coccodes*, *Fusarium lateritium*, *Xylohypha sp.*, and *Colletotrichum gloeosporioides* isolated from *Talinum paniculatum* (Jag.) Gaertn., bacteria isolates: *Escherichia coli* and *Bacillus subtilis*, Potato Dextrose Agar (PDA) medium, Potato Dextrose Broth (PDB), Ofloxacin, Steriled destilled water, Alcohol 95%, Paper blank disc, Centrifuge, Laminar Air Flow (LAF), Autoclave, and Inoculation needle.

2.2 Procedure

Endophytic Fungi Culture Preparation

The secondary metabolite could be obtained from endophytic fungi species liquid culture\(^3\). Each fungi isolated inoculated on PDA plate medium and incubated in 26°C – 27°C during 7x24 hours. Then each endophtic fungi colony were cutted in 5 pieces in the size of 1 x 1 cm\(^2\) each, the, inoculated in Potato Dextrose Broth (PDB) medium then shaked in 120 rpm rate and incubated in 26°C – 27°C during 7x24 hours. Afterwards, the liquid culture were centrifuged in 300 rpm during 10 minutes. The supernanatant were used to detect the content of each fungi secondary metabolite i.e: alkaloid, flavonoid, terpenoid, and tannin produced by each endophytic fungi species . The supernatant is also used to examine the antimicrobial effect of each endophytic fungi species toward *Escherichia coli* and *Bacillus subtilis*. The supernatant of each endophytic fungi species in volume 20 μl uses to examine the antimicrobacterial effect in vitro\(^{10}\).

The Antibacterial Effect Examination

The antibacterial effect of each endophytic fungi liquid culture towards *Escherichia coli* and *Bacillus subtilis* were examined by disk diffusion method. The sterile paper disk were dripped with 20 μl supernantant of aech endophytic fungi liquid culture. Then each paper disc were put on *Escherichia coli* and *Bacillus subtilis* culture put on Nutrient Agar (NA) plate medium surface. The *Escherichia coli* and *Bacillus subtilis* culture were inoculated first in Nutrient Broth (NB) and incubated in 37°C for 18 hours, standardized with MacFarland 0,5. Positive control use 5 μg/ml Ofloxacin and Potato Dextrose Broth (PDB) medium as negative control. The plates were incubated at 37°C for 1x24 hours and the inhibition zone was measured and compared with the control.

The Quantitative Screening of Some Secondary Metabolite from Endophytic Fungi Species Isolated from Java Ginseng

The quantitative screening of some secondary metabolites, i.e: alkaloid, flavonoid, terpenoid, and tannin produced by seven endophytic fungi species isolated from Java Ginseng was done by spectrophotometry method.

3. RESULTS AND DISCUSSION

The research result shows that the secondary metabolite of each endophytic fungi, i.e: *Fusarium semitectum*, *Aspergillus candidus*, *Colletotrichum accutatum*, *Colletotrichum coccodes*, *Fusarium lateritium*, *Xylohypha sp.*, and *Colletotrichum gloeosporioides* have an antibacterial effect towards *Escherichia coli* and *Bacillus subtilis* (Fig. 1).

![Figure 1. The Growth Inhibition Zone of Secondary Metabolite of Each Endophytic Fungi Species towards (a) Escherichia coli and (b) Bacillus subtilis. The treatment results of each endophytic fungi: (1) Colletotrichum acutatum, (2) Colletotrichum coccodes, (3) Colletotrichum gloeosporioides, (4) Xylohypha sp., (5) Fusarium semitectum, (6) Fusarium lateritium, (7) Aspergillus candidus, (8) Positive control with Ofloxacin, (9) Negative control with Potato Dextrose Broth.](image-url)
Bacillus subtilis. Aspergillus candidus secondary metabolite produce the highest growth inhibition zone diameter compared with the other fungi species towards Escherichia coli, i.e: 14.24 mm and towards Bacillus subtilis, i.e: 14.38 mm.

The difference effect of each endophytic fungi species secondary metabolite towards Escherichia coli and Bacillus subtilis were analyzed. Table 1 shows the LSD 1% analysis result.

Table 1. LSD 1% of The Difference of Each Endophytic Fungi Species Secondary Metabolites Effect towards Escherichia coli and Bacillus subtilis

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Average</th>
<th>Notation</th>
</tr>
</thead>
<tbody>
<tr>
<td>C. gloeosporoides E. coli</td>
<td>0.707</td>
<td>ab</td>
</tr>
<tr>
<td>Negative Control (PDB) E. coli</td>
<td>0.707</td>
<td>ab</td>
</tr>
<tr>
<td>Negative Control (PDB) B. subtilis</td>
<td>0.707</td>
<td>ab</td>
</tr>
<tr>
<td>Xylohyphae sp. E. coli</td>
<td>1.090</td>
<td>bc</td>
</tr>
<tr>
<td>C. coccodes B. subtilis</td>
<td>1.176</td>
<td>bc</td>
</tr>
<tr>
<td>C. acutatun B. subtilis</td>
<td>1.208</td>
<td>bc</td>
</tr>
<tr>
<td>C. acutatun E. coli</td>
<td>1.225</td>
<td>bc</td>
</tr>
<tr>
<td>C. gloeosporoides B. subtilis</td>
<td>1.261</td>
<td>bc</td>
</tr>
<tr>
<td>Xylohyphae sp. B. subtilis</td>
<td>1.294</td>
<td>c</td>
</tr>
<tr>
<td>C. coccodes E. coli</td>
<td>1.332</td>
<td>c</td>
</tr>
<tr>
<td>F. lateritium E. coli</td>
<td>2.673</td>
<td>d</td>
</tr>
<tr>
<td>F. lateritium B. subtilis</td>
<td>2.676</td>
<td>d</td>
</tr>
<tr>
<td>F. semitectum E. coli</td>
<td>2.790</td>
<td>de</td>
</tr>
<tr>
<td>F. semitectum B. subtilis</td>
<td>2.971</td>
<td>e</td>
</tr>
<tr>
<td>A. candidus E. coli</td>
<td>3.838</td>
<td>f</td>
</tr>
<tr>
<td>A. candidus B. subtilis</td>
<td>3.857</td>
<td>f</td>
</tr>
<tr>
<td>Positive control (Ofloxacin) E. coli</td>
<td>5.587</td>
<td>g</td>
</tr>
<tr>
<td>Positive control (Ofloxacin) B. subtilis</td>
<td>5.907</td>
<td>h</td>
</tr>
</tbody>
</table>

Table 1 shows that each endophytic fungi secondary metabolite have significant different effect towards growth inhibition zone diameter of Escherichia coli and Bacillus subtilis. The Aspergillus candidus secondary metabolite have the highest antibacterial effect towards Escherichia coli as well as Bacillus subtilis as shown with f notation. The clear zone around the paper disc proved that Escherichia coli and Bacillus subtilis cells was dead, because the fungi metabolites have been diffused to the medium around the paper disc.

Based on the analyze result of secondary metabolites content produced of each endophytic fungi species shows that secondary metabolites contains: Alkaloid, Flavonoid, Terpenoid, and Tannin with different content between the fungi species. Alkaloid, Flavonoid, Terpenoid, and Tannin are antibacterial effect compound that caused the death of Escherichia coli and Bacillus subtilis. So, it could inhibit the growth of the bacteria colonies. Aspergillus candidus have the highest contents of each antibacterial compound in the secondary metabolites, so it cause this endophytic fungi have the highest antibacterial activity towards Escherichia coli and Bacillus subtilis (Table 2).

Table 2. The Analyze Result of Secondary Metabolites Content Produced by Each Endophytic Fungi Species

<table>
<thead>
<tr>
<th>Isolate code</th>
<th>Species</th>
<th>Alkaloid</th>
<th>Secondary Metabolite (g/L)</th>
<th>Flavonoid</th>
<th>Terpenoid</th>
<th>Tannin</th>
</tr>
</thead>
<tbody>
<tr>
<td>A1</td>
<td>F. semitectum</td>
<td>0,45</td>
<td>0,94</td>
<td>0,04</td>
<td>0,37</td>
<td></td>
</tr>
<tr>
<td>A2</td>
<td>A. candidus</td>
<td>0,69</td>
<td>2,22</td>
<td>0,06</td>
<td>0,80</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
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<td></td>
<td></td>
<td></td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>R1</td>
<td>C. acutatum</td>
<td>0.59</td>
<td>1.93</td>
<td>0.05</td>
<td>0.70</td>
<td></td>
</tr>
<tr>
<td>R2</td>
<td>C. cocodes</td>
<td>0.57</td>
<td>1.87</td>
<td>0.05</td>
<td>0.68</td>
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</tr>
<tr>
<td>R3</td>
<td>F. lateritium</td>
<td>0.52</td>
<td>1.16</td>
<td>0.05</td>
<td>0.44</td>
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</tr>
<tr>
<td>D1</td>
<td>Xylophypha sp.</td>
<td>0.48</td>
<td>1.01</td>
<td>0.04</td>
<td>0.39</td>
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</tr>
<tr>
<td>D2</td>
<td>C. gloeosporioides</td>
<td>0.56</td>
<td>1.82</td>
<td>0.05</td>
<td>0.66</td>
<td></td>
</tr>
</tbody>
</table>

This research result proved that each endophytic fungi species isolated from *Talinum paniculatum* tissue could produce secondary metabolite contains: alkaloid, flavonoid, terpenoid, and tannin as same as produced by the host plant. So, if people need to take these bacterial compound, it could eliminate the need to harvest and extract the plant. There are differences of the antibacterial compound content between each fungi species.

This research result also show that each endophytic fungi species secondary metabolite have an antibacterial activity towards *Escherichia coli* and *Bacillus subtilis*. Each endophytic fungi species could inhibit *Escherichia coli* as well as *Bacillus subtilis* colony growth with differences in antibacterial activity. *Aspergillus candidus* secondary metabolite have the highest antibacterial activity based on the growth inhibition zone diameter of *Escherichia coli* as well as *Bacillus subtilis*. This fact was appropriate with the quantitative screening result of secondary metabolites produced by seven endophytic fungi species.

Alkaloid could bound with DNA, inhibit activity of some enzyme. i.e: estrease, DNA-polymerase, and RNA polymerase, and also inhibit cellular respiration\([13]\). Flavonoid colud cause cell membrane damaged and inactivity enzymes that play role in cellular metabolism\([12]\). Tannin could dissolved the lipid part of cell wall and caused structural protein denaturation of bacterial cell membrane\([13]\) \([14]\). Terpenoid caused protein structure damage on cell wall as well as on cell membrane. Cell membrane damaged caused decrease of cell membrane semipermeability. This fact caused metabolis inhibition and afterwards caused cell growth inhibition.

4. CONCLUSION

The conclusions are: 1) The secondary metabolites of seven endophytic fungi species have an antibacterial activity towards *Escherichia coli* and *Bacillus subtilis*; 2) *Aspergillus candidus* have the highest antibacterial activity towards *Escherichia coli* and *Bacillus subtilis*; 3) The secondary metabolite contents produced by each endophytic fungi species are: alkaloid, flavonoid, tannin, and terpenoid. *Aspergillus candidus* produce the highest content of the four compound compare with another species.

5. REFERENCES


Optimization of DNA Isolation and PCR Protocol for ISSR Analysis of Cultivar of *Durio zibethinus* Murray. to Reveal The Genetic Diversity

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Abstract – The problem of DNA isolation of durian is on the existence of contaminants such as proteins, polysaccharides, and polyphenol from the isolation process. The contaminants will interact with proteins and nucleic acids then form a gelatinous matrix which disrupts the entire of PCR reaction. Inter-simple sequence repeats (ISSR) is a molecular marker that requires a high degree of DNA purity. In addition, this marker requires appropriate DNA concentration in the amplification process. These problems problem can be solved through optimization of DNA isolation and PCR protocol. The research was conducted at the Laboratory of Molecular Biology, State University of Semarang. Samples of 15 accessions of local durian were taken from ex-situ collection of Hortimart Agro Centre, Central Java. DNA isolation procedure and PCR amplification protocol using a method both are modified. DNA isolation results are checked for their quality in qualitative by electrophoresis of agarose gel, and quantitative by spectrophotometric method. Primers used are four primers of Pusat Kajian Buah Tropika (PKBT). The results of DNA isolation are visible smear because of the contaminants. The DNA genome has size ranges between 9000 base pair (bp) to 10000 bp. DNA concentrations ranged from 11.7 to 247.5 ng/µl. A good ratio of the DNA purity to protein is 1.8-2.0. Purity obtained ranged from 1.223 to 2.008. The best of DNA genomic purity is accession of number eight. The optimum conditions PCR process, namely formula 1, consisted of 1 µl of DNA template, PCR master mix go green taq 6.25 µl, nuclease free water 4.25 µl, primary PKBT 1 µl of 10 mM, the total amount of 12.5 µl. The stages PCR consisted of predenaturation in 94 °C 4 minutes, denaturation in 94 °C 30 sec, annealing in 36 °C-53/54 °C 30 sec, extension in 72 oC 1 minute, final extension at 72 °C 5 minutes, and the number of cycles is 35x. The annealing temperature was obtained by this formula. The primer of PKBT 2 and 3 have a temperature of 53 °C and primer of PKBT 8 and 9 have a temperature of 54 °C. Those primers successfully amplified the DNA and showed polymorphism.

1. INTRODUCTION

Indonesia is megadiversity country that has high value of biodiversity in the world. The number of 30 accessions of local durian have been identified in the world, 14 accessions are endemic in Borneo. One of accessions that can be consumed is *Durio zibethinus* Murray. The number of 28 accessions of *D. zibethinus* have been known as superior durian (Reza, 2002). *D. zibethinus* is widespread in Indonesia and develop into many varieties with different characteristics and genotypes. A large number of local durian varieties in Indonesia causes the difficulty for them to be analyzed. This difficulty is caused by a lack of information about the characteristics of each variety (Uji, 2005).

Inter-simple sequence repeat (ISSR) is a widely used as molecular markers to reveal the diversity of plants. ISSR markers produced by amplification using polymerase chain reaction (PCR) with microsatellites primers. The advantages of ISSR are technically faster, less expensive, and able to detect the genetic diversity without knowing the information of genome analyzed (Kumar et al., 2009). ISSR molecular markers have been used to create the fingerprint of *Garcinia mangostana* (Widiastuti et al., 2013), the identification of the density of kinship in tangerines (Yuliant, 2010), and the identification of the genetic diversity of local durian in Thailand (Vanijaijiva, 2011). The Hortimart Agro Centre is a garden collections of local durian located in District of Bawen, Semarang Regency, Central Java Province. This research is the preliminary test for optimizing the process of DNA isolation and PCR protocol, so it can produce good method for DNA isolation and PCR protocol.
2. METHODS

2.1 Location and Sample

This research was conducted at the Laboratory of Molecular Biology and Research Laboratory, Department of Biology, Faculty Mathematic and Natural Science, Universitas Negeri Semarang. Samples are local durian from ex-situ collections in Hortimart Agro Centre, Central Java. A total of the sample is 15 accessions (Table 1).

<table>
<thead>
<tr>
<th>No</th>
<th>Accession</th>
<th>No</th>
<th>Accession</th>
<th>No</th>
<th>Accession</th>
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<td>1</td>
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<td>Yomodipati</td>
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<td>7</td>
<td>Pendowo</td>
<td>12</td>
<td>Mustiko</td>
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<td>Pancasona</td>
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<td>Ontoseno</td>
<td>13</td>
<td>Banowati</td>
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<tr>
<td>4</td>
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<td>9</td>
<td>Surtikanti</td>
<td>14</td>
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<td>5</td>
<td>Suryo</td>
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<td>15</td>
<td>Sugriwo</td>
</tr>
</tbody>
</table>

2.2 DNA Isolation

DNA isolation procedure using the CTAB method modified (Vanijajiva, 2011). It consists of four stages, namely collecting leaves and extraction, purification, precipitation, and the DNA quality test.

Method 1. DNA isolation method with CTAB modified by the addition of PVP without liquid nitrogen. The durian leaves weighed 1 gram then crushed and added extraction buffer consisting of 2% CTAB; 0.02 M EDTA pH 8.0; 0.1 M Tris-HCl pH 8.0; 1.4 M NaCl; β-mercaptoethanol 0.3%. Then added with 0.1 gram of PVP. The scour incubated in water bath at 30°C for 30 minutes. Then, it was cooled at room temperature for 5 minutes. The CIIA then added as many as 24:1 of the solution. The solution was centrifuged at 3000 rpm for 15 minutes. The supernatant was transferred to a new microtube and then added with a volume of 0.5 M NaCl. The solution added a 2x volume of cold ethanol 95% then inverted and incubated in the freezer for 10 minutes. The next steps are centrifuged at 3000 rpm for 6 minutes. The supernatant formed was then removed until remaining pellets at the end of the microtube. DNA pellet was washed with 70% of cold ethanol and then centrifuged at 4000 rpm for 5 minutes. DNA pellets were dried at room temperature for one night. DNA pellets dried and reconstituted with TE buffer. The first stage of purification, the isolated DNA solution added 3 µl of RNase (10 mg/ml) and then incubated in a water bath at 37 °C for an hour. The solution was then added 3 µl of proteinase K (1 mg/ml) then incubated at 37 °C for 30 minutes. The solution was then added phenol and chloroform then centrifuged at 14,000 rpm for 15 minutes. The supernatant was transferred into a new microtube, then added 25 µl of 5 M NaCl and 250 µl of 70 % ethanol and incubated at -20 °C for an hour. The solution was then centrifuged 8000 rpm 10 minutes. DNA pellets dried and then dissolved in TE buffer.

Method 2. The second method of DNA has some modified steps such as removing the disposal of bottom leaf surface trichomes and using liquid nitrogen. Durian leaves weighed 1 gram then crushed and added extraction buffer consisting of 2% CTAB; 0.02 M EDTA pH 8.0; 0.1 M Tris-HCl pH 8.0; 1.4 M NaCl; β-mercaptoethanol 0.3%. Then, the solution incubated in water bath at 60 °C for 30 minutes. The solution was then added phenol: chloroform: isooamyl-alcohol (PCI) as many as 25:24:1. The solution was inverted until the colour on the bottom layer became green. The solution is then centrifuged at 10,000 rpm for 15 minutes. The solution in the uppermost layer was transferred to a new microtube. The transferred supernatant was added 100% of cold isopropanol as many as 0.6x volume of the solution. Then, it was inverted and centrifuged at 4000 rpm for 5 minutes. The pellets were formed was washed with 70% of cold ethanol and then centrifuged at 4000 rpm for 5 minutes. The washing with cold ethanol do as much as 3 times, and then the supernatant was discarded and the DNA pellets were dried at room temperature for one night. They were dissolved in TE buffer and stored at -20 °C. The purification stages have similar procedures such as DNA isolation, but there are an addition of 2.5 µl of RNase then incubated for 30 minutes in a water bath. The addition of RNase is to degrade the RNA.

2.3 PCR Protocol

The DNA amplification used peqSTAR 2X thermocycler. Primers used are ISSR primers of Tropical Fruit Research Central Laboratory (PKBT) collection as many as four primers. The amplification procedure used a method of Widiausti et al. (2013) which has been modified. The composition of PCR which has been used in PCR-ISSR process includes DNA template, ISSR primer, PCR master mix go qtaq green promega and nuclease-free water. PCR protocol 1. The dilution was done in order to obtain DNA with a concentration of 5 ng/µl. The PCR process consisted of 2 µl (5 ng) of DNA template, 6 µl of PCR master mix go qtaq green, 4 µl of nuclease-free water, 0.5 µl (5 Mm) of ISSR primer, a total of 12.5 µl. PCR protocol 2. The dilution was done in order to obtain DNA with a concentration of 10 ng/µl. The composition of PCR included 1 µl (10 ng/µl) of DNA samples, 6.25 µl of PCR master mix go qtaq green, 4.25 µl of nuclease-free water, and 1 µl (5 Mm) of ISSR primer, a total of 12.5 µl.

The stages of PCR process included pre-denaturation at 94 °C for 4 minutes, denaturation at 94 °C for 30 seconds, annealing at 36-53/54 °C for 30 seconds, the extension at 72 °C for 1 minute, and final extension at 72 °C for 5
minutes, and a total of 35 cycles. The amplification products were separated using a 1.2% of agarose gel. The size of each DNA bands compared with 50 bp DNA ladder.

3. RESULTS AND DISCUSSION

3.1 Comparison of DNA Isolation Using Method 1 and Method 2

Both methods are modified to obtain genomic DNA with the best quality. Modifications carried out with the addition of PVP, CIAA, NaCl, PCI, and disposal of trichomes on the lower leaf surface. The disposal of trichomes on the lower leaf surface is to reduce brown color on the pellet while grinding through liquid nitrogen is to accelerate and expand the destroyed cell walls, in order to obtain the higher number of DNA isolated. The results of DNA isolation using method 1 still looks a smear and shows the presence of contaminants. The DNA genome size of local durian range from 900 base pair (bp) to 10000 bp. Marker DNA used came from Thermo Scientific consists of 5000 bp, 2000 bp, 850 bp, 400 bp and 100 bp. Here is an electropherogram of DNA isolation using both method 1 (Figure 1) and method 2 (Figure 2).

Electropherogram of DNA isolation using method 1 shows the six accessions generate DNA bands with pretty good intensity, namely the accession number 4, 6, 7, 8, 10, and 11 (Figure 1). Almost all of the lanes still produce RNA which is characterized by a thickened band on the tail. In addition, the accession number 1, 2, 3, 5, 12, and 15 did not produce DNA at all. The DNA degraded due to the milling process is too strong and too fast in centrifugation step. The DNA bands in lane 9 and 13 showed a smear. It is suspected there is a broken of DNA caused by their grinding process that is too strong. A luminescence on each well indicate that there are contaminants such as polysaccharides and proteins. The electropherogram of DNA isolation using method 2 shows there are 15 lanes on the visualization (Figure 2). DNA bands visualized quite well on each lane. However, the intensity of which is produced in each lane is difference. Some accessions still have the smear bands, namely the accession of number 7, 8, 13. Almost all of the bands from each accession still exhibit residual RNA, except for the accession of number 3, 6, 10, 11, and 12. Overall, the second method produces the better of DNA isolated than the first method. Electropherogram of DNA isolation using both method 1 and method 2 showed different results. The DNA with thick bands showed high concentrations, whereas the DNA with smear bands showed the presence of the bonds between the DNA molecule is cut off so the genomic DNA cut to be a smaller size. The bond between the molecules is lost can be caused by physical movements excess, which can occur in the process of DNA isolation, centrifuges, or even because the temperature is too high, freezing, thawing, and as a reaction to certain chemicals, so that the size of the DNA fragments are cut into short size. The purity value of DNA isolated using this method from Vanijajiva (2011) modified has a pretty good range, which is between 1.223 to 2.008 and DNA concentration between 11.7 to 247.5 ng/µl (Table 1).

![Figure 1 Metode 1 Isolasi DNA](image1.jpg)

![Figure 2 Metode 2 isolasi DNA](image2.jpg)

Table 1 Value purity and concentration of DNA

<table>
<thead>
<tr>
<th>No</th>
<th>Accession</th>
<th>A260/280</th>
<th>Conc (ng/µl)</th>
<th>No</th>
<th>Accession</th>
<th>A260/280</th>
<th>Conc (ng/µl)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Jagal Bilowo (1)</td>
<td>1,626</td>
<td>45,7</td>
<td>9</td>
<td>Surtikanti (29)</td>
<td>1,628</td>
<td>51,35</td>
</tr>
<tr>
<td>2</td>
<td>Ajimah (4)</td>
<td>1,732</td>
<td>47,45</td>
<td>10</td>
<td>Bima (32)</td>
<td>1,403</td>
<td>44,1</td>
</tr>
<tr>
<td>3</td>
<td>Pancasona (7)</td>
<td>1,472</td>
<td>70,2</td>
<td>11</td>
<td>Yomodipati (33)</td>
<td>1,579</td>
<td>11,7</td>
</tr>
<tr>
<td>4</td>
<td>Ponconoko (9)</td>
<td>1,555</td>
<td>16,1</td>
<td>12</td>
<td>Mustiko (37)</td>
<td>1,311</td>
<td>46,55</td>
</tr>
<tr>
<td>5</td>
<td>Suryo (13)</td>
<td>1,711</td>
<td>42,7</td>
<td>13</td>
<td>Banowati (38)</td>
<td>1,420</td>
<td>28,1</td>
</tr>
<tr>
<td>6</td>
<td>Pasopati (15)</td>
<td>1,393</td>
<td>247,5</td>
<td>14</td>
<td>Cokro (39)</td>
<td>1,223</td>
<td>66,05</td>
</tr>
<tr>
<td>7</td>
<td>Pendowo (17)</td>
<td>1,844</td>
<td>58,15</td>
<td>15</td>
<td>Sugriwo (42)</td>
<td>1,745</td>
<td>32,9</td>
</tr>
<tr>
<td>8</td>
<td>Ontoseno (20)</td>
<td>2,008</td>
<td>35,95</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

The DNA concentration is known from the absorbance at a wavelength of 260 nm. The absorbance value of 1.0 at a wavelength of 260 nm is equivalent to 50 ng mL⁻¹ as double-stranded DNA. The DNA purity value to protein contamination including RNA known from the ratio of A260 to A280. The good ratio for the purity DNA to the protein is 1.8-2.0 (Kundu et al., 2011). The DNA isolation results indicate that concentration of DNA isolated ranged from 11.7 ng/µl to 247.5 ng/µl. The accession which has the highest concentration of DNA is Pasopati. The purity of the protein from the DNA isolation results ranged from 1.223 to 2.008. The genomic DNA purity of Ontoseno is quite well among the other accessions.
3.2 Comparison of PCR Protocol 1 and PCR Protocol 2

The difference between the protocol both of PCR 1 and PCR 2 are the concentration of DNA, the number of PCR master mix go to ta green, nuclease-free water, and primer concentration. Here is a comparison of the electropherogram of DNA amplification using protocol PCR 1 and PCR 2 (Figure 3).

![Figure 3 Comparison of PCR Protocol 1 and PCR Protocol 2](image)

DNA amplification depending on the concentration and purity of DNA template, taq polymerase, primer size, and degree of homology between primer and DNA template. The purity of the DNA template is very important because it can affect the amplification reaction and can inhibit the action of the DNA taq polymerase. Each primer generates the different bands with their thickness and clarity. The concentration of DNA and primer effect on amplification process. If the DNA concentration is too low (5 pg/50 µl) will not produce bands, whereas the concentration of 10 pg-1 ng/50 µl produces a different pattern. In addition, the storage of DNA in a TE buffer that is too long caused the rest of the EDTA contained in the amplification reaction. EDTA can bind Mg\(^{2+}\), consequently, the enzyme is inhibited and the DNA band is not clear when it was run in agarose gel (Weising et al., 1995). According Padmalatha and Prasad (2006) and Harini et al. (2008) primer concentration are too low or too high can cause no amplification. In addition, a low ratio between the primer and DNA causing the products are not consistent even not formed (Ali et al., 2006).

This research used four primers of PKBT, namely PKBT 2, PKBT 3, PKBT 8 and PKBT 9 with the annealing temperature by optimization results for PKBT 2 and 3 is 53 °C, while PKBT 8 and 9 is 54 °C (Figure 4).

![Figure 4 Annealing temperature of primer PKBT 2, 3, 8, and 9](image)

Annealing is one of the factors that influence the success of amplification. If the temperature is too high will cause failure of amplification because it does not happen the annealing between primer and DNA template, otherwise, if the temperature is too low causes the primer will attach to the other side of the DNA template, consequently the DNA bands formed have a low specificity (Ricardo, 2011). The annealing temperature is an important parameter and needs to be optimized.

4. CONCLUSIONS

The results of DNA durian isolated using the second method of Vanijajiva (2011) shows quite good in quality, which has a purity of between 1.223 to 2.008, and the concentration of DNA between 11.7 to 247.5 ng/µl. Optimization of PCR protocol for durian has been successfully determined. The second protocol of PCR is best protocol because it can produce the specific of DNA bands. Primer of PKBT 2, 3, 8, and 9 have amplified DNA of local durian with the annealing temperature of PKBT 2 and 3 is 53 °C, PKBT 8 and 9 is 54 °C. The bands formed show a different pattern so that the primers could potentially generate polymorphic bands.

5. REFERENCES

The Effect of Plant Growth Regulators and Natural Supplements on In Vitro Shoot Regeneration of *Physalis angulata* L.

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**Abstract** – *Physalis angulata* L. (Ciplukan) is one medicinal plant which is currently being developed as herbal medicine in industrial scale. Tissue culture technique is promising alternative to produce medicinal plant raw material continuously. Natural supplements have been frequently added in tissue culture media in order to save the production cost. The purpose of this study is to observe the effect of two kinds of natural supplements, namely tomato juices and bean sprout extract on shoot regeneration of *P. angulata*. The first nodes excised from two weeks old in vitro shoot were cultured on MS basal medium supplemented with tomato juices (5, 7.5 and 10%) or bean sprout extract (1.25, 2.50 and 3.75%). The potential of natural supplement in shoot regeneration was compared with combination of BAP + IAA for six weeks of culture. The number, length and fresh weight of regenerated shoots were recorded every week and the mean of five replicates explants were calculated. BAP + IAA induced better respond in the number and fresh weight of shoot than both tomato juices and bean sprout extract. Otherwise the highest length of shoot was observed in tomato juices at 5 and 7.5% from three to six weeks of culture which reached three times the shoot length in BAP + IAA. This result shows that application of tomato juices alone in micropropagation media of *P. angulata* could be considered due to their good responsive in shoot regeneration.

1. INTRODUCTION

The genus *Physalis* is a member of Solanaceae, which is classified as an annual shrub. These plants usually found as a wild plant in areas with moist soil on the banks of the river, gardens or fields are being planted peanuts, soy or corn. Since they easily grow, usually, the farmers consider it as a weed, even though it has a great economic importance for its valuable chemical compounds. *Physalis angulata* L. (ciplukan or ceplukan) has been known as medicinal plant.

Nowadays, ciplukan is being developed as herbal medicine in industrial scale. It causes the demand for ciplukan plant, as raw material for herbal medicine has increased. However, since ciplukans are not intensively cultivated, they are usually collected from several regions with diverse growing conditions. Different environmental conditions are possible to produce different plant growth. It certainly cannot guarantee the quality and quantity of chemical compound contents.

Conventional farming has limitations to produce tillers of uniform and free of disease [1]. To ensure the continuous availability and standardized raw material of herbal medicine, the tissue culture technique is a promising alternative. Micropropagation is one method of tissue culture that capable to regenerate large number of new plants which have similar characters with those of the mother plants. The success of in vitro plant regeneration is determined by appropriate culture medium. Plant tissue culture medium generally contains macronutrients, micronutrients, vitamins, amino acids or nitrogen supplements, source(s) of carbon, undefined organic supplements, growth regulators and solidifying agents. Growth and regeneration of plant from in vitro tissue culture can be improved by undefined supplement of organic nutrient from natural origin [2], such as coconut water, banana homogenate, many kinds of fruit juice (papaya, tomato, orange), and bean sprout extract. The aim of this study was to identify the effect of natural supplements and plant growth regulator (PGR) on in vitro shoot induction and multiplication of nodal explants of *P. angulata*.

2. METHODS

2.1 Plant material and explant sources

*P. angulata* seeds derived from ripened fruit were surface sterilized with 70% ethanol for 30 seconds, 30% commercial bleach (5.25% NaClO) for 10 minutes and, subsequently, washed with sterilized water three times.
for 5 minutes each. The seeds were then placed in agar medium containing 1.2 % commercial agar. For germination the culture bottle were incubated in continuous light condition (600 lux) at 23 ± 2 °C. After two weeks the top of seedling which had two cotyledons and first leaves were excised, separated from hypocotyl and cultured on MS0 medium for two weeks. This part increased in height and had 2-3 nodes. The first node of the shoot tip was used as explant for shoot regeneration.

2.2 Shoot regeneration medium and culture condition

Shoot regeneration was conducted on MS basal medium supplemented with 30 g/L sucrose. The culture medium were classified into three groups (Table 1), 1) Murashige and Skoog (MS) medium [3] containing combination of BAP and IAA, 2) agar medium containing tomato juices (5.0, 7.5 and 12.5%), and 3) agar medium containing bean sprout extract (1.25, 2.5 and 3.75%). Tomato juices were obtained from meso and pericarp of ripe tomato which grounded using blender and then added into the MS mineral salt solution. The medium pH was adjusted to 5.8 prior to solidified with 1.2% commercial agar. Subsequently, approximately 10 ml medium was distributed into culture bottle and autoclaved at 121 °C, 1.5 atm for 15 min.

Three nodal explants were inoculated into culture medium and incubated under continuous light condition (600 lux) at 23 ± 2 °C for six weeks. Five replicates were used for each type of shoot regeneration medium. The number and length of shoot were observed every week while shoot fresh-weight was measured at sixth week.

3. RESULTS AND DISCUSSION

In all in vitro media tested direct shoot regeneration from P. angulata nodal explants was observed. The successful shoot, or plantlet regeneration using cutting nodal explants, has been frequently reported such as in sweet potato [4,5]. The nodal segment, which has a pre-existing meristem, is suitable for clonal propagation because it is easily manipulated, has a high proliferation rate and maintains clonal fidelity [6]. The buds/meristems such as shoot tips, nodal segments and nodes from pre-existent buds would readily give multiple shoots with cytokinin such as BAP, zeatin etc. When the nodal explants of in vitro shoot were placed on fresh culture medium the axillary bud will grow directly into a new plantlet. So far nodal explants are the most preferred type of explant for tissue culture. Nodal explants as the best source of multiple shoot induction have also been suggested in case of many medicinal plants.

Based on the group of culture medium the shoot number also could be classified into three groups. Agar medium supplemented with bean sprout extract alone as natural supplement produced the lowest shoot number for six weeks observation (Figure 1A). While agar medium supplemented with tomato juices alone induced higher shoot number than bean sprout extract. MS medium + BAP 2 mg/L + IAA 0.05 mg/L (med I) produced the highest number of shoot per explant from 2-6 weeks of culture. Decreasing BAP into 0.5 mg/L combined with increasing IAA into 0.1 mg/L (med II) produced shoot in lower number. After six weeks of culture bean sprout extract, tomato juice, medium I and medium II produced 1.0-1.4, 2.9-3.1, 4.3 and 7.6 shoot, respectively. BAP was also essential for shoot induction and multiplication of Chlorophytum borivilianum Sant. & Fernandez [7].

![Figure 1](https://example.com/figure1.png)

**Figure 1** The growth of in vitro shoot during six weeks of culture. A. Shoot number. B. Shoot length

Agar medium supplemented with tomato or bean sprout alone induced higher length of shoot than MS medium supplemented with combination of BAP and IAA (Figure 1B). From the second to the sixth week of culture on tomato medium the descending order of shoot length was produced increasing concentration from 5, 7.5 and 10 %. While bean sprout 2.50 % provided higher length of shoot rather than two other concentration (1.25 and 3.75 %). While rich mineral nutrition medium supplemented with plant growth regulator (med I and II) apparently inhibited shoot elongation. On the contrary, during six week of culture shoot length in tomato...
medium sharply increased. Similar to shoot regeneration from nodal explants of sweet potato [5] in this work the lower BAP concentration increased height while an increased concentration decreased shoot height of *P. angulata*. Higher concentration of BAP inhibits shoot elongation of *Chlorophytum borivilianum* Sant. & Fernandez [8]

All regenerated shoot (100 %) in tomato medium and in MS medium + BAP + IAA were still survive after six week of culture (Table 1). While in bean sprout extract medium the survival rate of regenerated shoot only less than 80%.

**Table 1** Effect of plant growth regulators and natural supplement on survival rate of regenerated shoot of *P. angulata* derived from nodal explant after six weeks of culture

<table>
<thead>
<tr>
<th>Medium</th>
<th>% of explants showing shoot regeneration response</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tomato juice (T) 5.0 %</td>
<td>100</td>
</tr>
<tr>
<td>Tomato juice (T) 7.5 %</td>
<td>100</td>
</tr>
<tr>
<td>Tomato juice (T) 10.0 %</td>
<td>100</td>
</tr>
<tr>
<td>Bean sprout extract (BS) 1.25 %</td>
<td>77.8</td>
</tr>
<tr>
<td>Bean sprout extract (BS) 2.50 %</td>
<td>66.7</td>
</tr>
<tr>
<td>Bean sprout extract (BS) 3.75 %</td>
<td>77.8</td>
</tr>
<tr>
<td>MS + BAP 0.5 mg/L + IAA 0.1 mg/L (Med I)</td>
<td>100</td>
</tr>
<tr>
<td>MS + BAP 2 mg/L + IAA 0.05 mg/L (Med II)</td>
<td>100</td>
</tr>
</tbody>
</table>

After six weeks of culture the fresh weight (g/clump) of shoot in medium + PGR was higher than medium supplemented with tomato juice/bean sprout extract (Figure 2). This condition is due to med I and II also produced the higher number of shoot (Figure 1A). In medium + natural supplement shoot fresh weight decreased with increasing concentration of either tomato juice or bean sprout extract. The number and length of shoot in tomato medium (Figure 3A-C) and in bean sprout extract medium (Figure 3D-F) were contrast to the length and shoot number in MS medium + combination of BAP + IAA (Figure 3G-H).

![Figure 2 Shoot fresh weight after six weeks of culture](image)

**Figure 2** Shoot fresh weight after six weeks of culture

![Figure 3 Regenerated shoot of *P. angulata* after six weeks of culture. A-C. Tomato medium, D-F. Bean sprout extract medium, G. MS + BAP 0.5 + IAA 0.1, H. MS + BAP 2 + IAA 0.05](image)
4. CONCLUSIONS

The highest shoot number of \textit{P. angulata} nodal explants was provided by MS medium supplemented with combination BAP + IAA. Agar medium supplemented with tomato juice alone was able to produce long shoot of \textit{P. angulata} with high survival rate although until six weeks of culture the nodal explants only produced 2.6 – 3.1 shoot/explant. On the other hand, bean sprout extract had poor effect on \textit{P. angulata} shoot multiplication and elongation. This work represents an economic protocol for direct shoot regeneration of \textit{P. angulata} without using rich nutrient medium without the addition of synthetic plant growth regulators.

5. REFERENCES

The Effect of Banana Homogenate Supplement on Shoot Regeneration of *Physalis angulata* L. Nodal Explants

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**Abstract** – *Physalis angulata* L. is a herbaceous plant that grows wild, and widely used as an ornamental plant, food, fodder and medicinal plant. This study is aimed to investigate the effect of two types of bananas, namely ambon and raja, added in culture medium on shoot regeneration from nodal explants of *P. angulata*. The study used a randomized block design factorial. The first factor is with two levels bananas types ambon and raja, and the second factor is with four concentrations levels of banana homogenate (0 (control), 2.5 %, 5 % and 7.5 %). The study was conducted in several stages, making use of in vitro medium consisted of medium with and without banana homogenate. Shoot induction and multiplication were used in vitro nodal explant. The parameters in this study are the number of shoots, shoot length, and number of leaves. The results showed that the shoot regeneration responses were affected by the types and concentration of banana. During 8 weeks of culture, the highest number of shoot was obtained from medium supplemented with 5 % of ambon banana homogenate. Raja banana homogenate affected on shoot length of in vitro *P. angulata*. The highest number of leaves per 6 weeks of culture was obtained from medium supplemented with 5 % of ambon banana homogenate. However, the lower concentration of ambon banana homogenate (2.5 %) produced leaves higher number at 7-8 weeks of culture. It can be concluded that 5 % of ambon banana homogenate has better effect on in vitro shoot regeneration and multiplication of *P. angulata*.

1. **INTRODUCTION**

Ciplukan (*Physalis angulata* L.) is an important medicinal plant of family Solanaceae. Otherwise, *P. angulata* can be used as ornamental plant, food and fodder. This species is used medicinally as a treatment for hypertension, tumor and diabetes [1]. Currently, a herbal medicine made from *P. angulata* has been developed on an industrial scale. Hence the need for the plant material to meet the traditional medicine industry increases. Ciplukan has not been widely cultivated. On the other hand, *P. angulata* which grows wild in various locations varied greatly because of the influence of the environmental factors. It also will affect the secondary metabolites produced. Techniques of in vitro culture provide alternative solutions to produce the plant in large number and in relatively short times. It is explained [2]that the success of tissue culture techniques is influenced by several factors, including the composition of the medium of the plant growth. Basal medium commonly used for tissue culture techniques is the Murashige and Skoog (MS) (1962) basal medium. MS basal medium containing elements of macro nutrients (N, P, K, Ca, Mg, and Na) and micro (B, Co, Mn, I, Fe, Zn, and Cu) are complete and widely used for herbaceous plants. In addition to macro and micro nutrients, MS basal medium also contains other energy sources such as sugars, vitamins, amino acids and myo inositol.

In vitro culture medium can be supplemented by organic materials such as a source of sugar, vitamins, amino acids, and plant growth regulator (PGR). Natural organic compounds that have been widely used, among others, coconut milk, yeast extract, malt extract, potato extract, extract of papaya, banana homogenate, tomato juice and juice wotel. The use of natural organic compounds such as an additive in a medium can deliver better growth and morphogenesis [3].

Banana is generally added to the media in vitro in the form of homogenates. Giving banana homogenate can improve regeneration response, the number of shoots per explant and complete plantlets of *Cymbidium pendulum* compare to medium without the addition of banana homogenate [4]. Banana homogenate with 150 g / L and NAA 20 ppm can stimulate and produce an average of the highest number of buds on the black orchids (*Coelogyne pandurata*) [5]. Banana contains carbohydrates, protein, fat, Ca, P, Fe, vitamin A, vitamin B-1, and vitamins that can help the process of regeneration in plants [6]. Some types of bananas namely kepok, mas, raja and ambon can produce growth response in vitro on different Phalaenopsis orchid plantlets [7]. Therefore, this study aimed to know the effect of two types of bananas, namely ambon and raja added in culture medium on shoot regeneration from nodal explants of *P. angulata*. 
2. METHODS

2.1 Preparation of Culture Medium

Seed germination medium is agar medium (12 g / L). In vitro culture medium used MS basal medium started with added stock solution A, B, C, D, E, F, G, Vitamin, Myo-inositol and plant growth regulators (BAP 2 mg / L + IAA 0.05 mg / L). After that Ambon and Raja banana mature skin was peeled and then mashed, pureed banana subsequently weighed and added as much as 2.5, 5 and 7.5 gr in 100 ml of medium (for control medium without the addition of banana homogenate). Sugar 30 g / L was added and diluted with aquades. The solution was stirred, after that the pH was adjusted to 5.8 by addition of NaOH or HCl. Then added agar as much as 12 g / L, then stirred, and add aquades to 1 L. The solution was heated to boiling, then medium 10 ml was added to the culture bottle, closed plastic and rubber tied. Medium was sterilized in an autoclave for 15 minutes at 121 °C and a pressure of 1.5 atm.

2.2 Preparation Explants Source

Fruit of *P. angulata* were collected from Field in Gondang Legi, malang, East Java. The mature fruit of *P.angulata* dried for 2-3 days. Then the dried seeds were germinated on an agar medium in light conditions, with intensity of 500 lux, at 23 ± 2 °C. After two weeks old in vitro seedling the cotyledon and apical buds was excised and then subcultured to MS medium.

2.3 Shoot Induction

Sources of nodal explants taken from shoots at two weeks of age cultured in MS0 medium. Then nodes, were cultured in control medium (MS + BAP 2 mg / L + IAA 0.05 mg / L) and treatment medium (MS + BAP 2 mg / L + IAA 0.05 mg / L + banana homogenate). Each culture bottle filled with 3 nodes, then incubated in a culture room in light conditions (light intensity of 500 lux) at 23±2 °C. The parameters observed every 2 weeks during 8 weeks were number and length of shoot and number of leave. The length of shoot was measured from the base of the buds on the surface of the medium until the tip of shoots.

3. RESULTS AND DISCUSSION

The results of this study show that all parameters, namely the number and length of shoots, and number of leaves, increased in all treatments from 1 until 8 weeks of culture. In control and banana homogenate supplemented medium, the number of shoots increased significantly from the first until 4 weeks. But from 5 weeks, after culture, until 8 weeks of culture, the number of shoot increased slightly. The number of shoots produced in medium supplemented with ABH (Ambon Banana Homogenate) was higher compared to the control and raja banana homogenate, while, the addition of banana homogenate did not affect shoot length.

The number of leave showed continuous increased during 8 weeks. This phenomena looks good on control and with the addition of banana homogenate. The number of leaves from the first until 7 weeks weeks generating the highest number of leaves on the addition of ambon banana homogenate, but at weeks 8 the number of leaves were generated on the control.

Banana homogenate good influence on the number of shoots and number of leaves, but the addition of RBH (Raja banana homogenate) had little influence compared with controls (fig. 1).
Figure 1. The effect of banana homogenate on shoot regeneration *P. angulata*, a) the number of shoots, b) shoot length, c) the number of leaves

* ABH (Ambon banana homogenate) * RBH (Raja banana homogenate)

### 3.1 Shoot Number

The results on the number of shoots showed that the highest number of shoots was observed in ABH (Ambon Banana Homogenate) of 5% during 8 weeks. While at concentrations of 2.5 and 7.5% it had little influence on the number of shoots compared with controls. In addition of RBH (Raja Banana Homogenate) of 5% also resulted the highest number of shoots during 8 weeks, but produced shoots which are lower in number than the addition of ABH treatment with a concentration of 5%. ABH and RBH at 2.5 and 7.5 % did not produce different number of shoot compared with control medium. In week 7 the control decreased the number of shoots for differentiation into a callus.

![Figure 1](image1.png)

**Figure 1.** Effect of banana homogenate treatments in number of shoot of *P. angulata*, a) ABH (Ambon banana homogenate) and b) RBH (Raja Banana Homogenate) with three different concentrations (2.5, 5 and 7.5 %)

Fig 1. showed that the concentration of banana homogenate 5% supported multiplication in vitro *P. angulata* shoot. Organic growth supplements banana homogenate proved beneficial for the multiplication of the cultures besides enhancing plantlets growth of *Cymbidium pendulum* with 0.5 % banana homogenate. Added 0.5 of banana homogenate proved beneficial for the development of healthy shoots growth of *C. pendulum* [4].

### 3.2 Shoot Length

The results of shoot length showed that all treatments increased during 8 weeks. ABH 5% high produced higher number of shoots from week 1 to week 2, but at week 4 to weeks 8 it produced the lowest length of shoots compared with controls and ABH 2.5 and 7.5%. While medium supplemented with RBH 5% produced the highest shoot length during 8 weeks, whereas RBH 2.5 and 7.5 % produced lower length of shoot compared to controls.

![Figure 2](image2.png)

**Figure 1. Figure 2.** Effect of banana homogenate treatments in shoot length of *P. angulata*, a) ABH (Ambon banana homogenate) and b) RBH (Raja Banana Homogenate) with three different concentrations (2.5, 5 and 7.5 %)

### 3.3 The Number of Leaf

The results showed that media with the addition of ABH (Ambon Banana Homogenate) affected to the number of leave. It can be seen that medium supplemented with ABH ambon banana homogenate of 5% produced higher number of leave during 6 weeks of culture. While at 7-8 weeks the highest number of leaves was produced in ABH of 2.5%. Control was higher compared to the RBH (Raja Banana Homogenate), but from week 1 until 2
weeks of culture RBH of 5% produced the highest number of leaf. Banana homogenate significantly increased the number of leaves in *Dendrobium nobile* cultures [8].

![Figure 3. Effect of banana homogenate treatments in number of leave *P. angulata*, a) ABH (Ambon banana homogenate) and b) RBH (Raja Banana Homogenate) with three different concentrations (2.5, 5 and 7.5 %)](image)

The addition of banana homogenate 5% produced shoots with better morphology than other concentration (Fig. 4). ABH 5% had higher number of shoots and leaves, but has a smaller shoots compared to the control and RBH medium. While in RBH medium after 8 weeks culture shoots had thick stems, but many leaves become yellow and abscised.

![Figure 4. Shoot regeneration of *P. angulata* on ABH and RBH treatment with three different concentrations (2.5, 5 and 7.5 %) after 8 weeks of culture](image)

4. CONCLUSIONS

It can be concluded that addition of banana homogenate only affected number of shoots. Addition ABH (Ambon Banana Homogenate) of 5% significantly improve the number of in vitro *P. angulata* shoot. While addition of RBH (Raja Banana Homogrmate) at 5% only affected shoot elongation. The concentration of banana homogenate that supports for regeneration in vitro *P. angulata* is 5%.

5. REFERENCES

Ecophysiological Responses of Sweet Potato Plant (*Ipomoea batatas* (L.) Lam) to Drought Stress

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Abstract – Drought due natural conditions, and recently being intensified by phenomena of global warming, environmental change and uncertainty in global climate conditions, has long been a major problem in worldwide agricultural productions. Therefore, understanding ecophysiological responses of crops to drought stress as well as their tolerant mechanism is important in order to obtain plants which are potential to be cultivated in dry area. We investigated ecophysiological responses of sweet potato plant, varieties Cilembu, to drought stress in four levels of field capacity (fc), i.e. control (80% fc), light drought stress (60% fc), moderate drought stress (40% fc) and severe drought stress (20% fc), using randomized block design and six replicates, for an exposure period of 10 weeks. The result showed that severe drought stress significantly inhibited plant growth, although the plant still survived. Increasing level of drought stress significantly reduced plant dry weight, number of leaves, and chlorophyll content. Relative water content (RWC) of plant leaves, at exposure of severe and moderate stress, reduced by ±10%, compared to control. After ten weeks exposure to severe stress, plants produced proline and glycinebetain concentration significantly higher than in control. It can be concluded that severe drought stress for a period of 10 weeks exposure inhibited plant growth, although the plants still able to survive. Higher concentration of proline and glycinebetaine in drought stress plants, probably, indicate their role as adaptive osmotic solutes which enabling plants to survive. Although the experiment was terminated before sweet potato plant produced tubers, however, it showed good tolerance to drought stress, and therefore sweet potato plant is recommended to be cultivated in relatively dry area, as an alternative to staple food.

1. INTRODUCTION

Drought stress is one of the environmental factors that can interfere with the growth and development of agricultural crops thus inhibiting plant productivity. Drought causes stomatal closure, loss of turgor pressure and interfere with plant growth [1]. Plants have defense mechanism in the face of drought stress such as accumulate compatible solutes which are low molecular weight, highly solube organic compounds and non-toxic like proline and glycinebetaine which serves as osmoprotectant to maintain cell function in conditions of drought [2]. However, plant reaction to stress vary. Plant responses to stress are influenced by the intensity and duration of stress, plant species and stage of development [3].

Sweet potato (*Ipomoea batatas* (L.) Lam) is one among seven important agricultural commodities being prioritized to be developed in Indonesia, and since 1968 Indonesia has been included as big four sweet potato producer country. Understanding ecophysiological repsones of sweet potato to drought conditions is important in order to maintain the production of sweet potato. We investigated ecophysiological responses of sweet potato plant, Cilembu varieties, to drought stress in four levels of field capacity (fc).

2. METHODS

2.1 Experimental Design

Cilembu sweet potato plants were taken from Cilembu village, Sumedang, West Java province. The sweet potato planted with shoot cuttings method. Shoots 15-25 cm in length were taken from three months old plants with short internodes. Shoot cuttings were planted in polybags containing a mixture of 10 kg soil and compost (3:1). The experiment was conducted using randomized block design with six replicates, for an exposure period of 10 weeks. Four levels of field capacity (fc) i.e. control (80% fc), light drought stress (60% fc), moderate drought stress (40% fc) and severe drought stress (20% fc) were examined to the plants. Treatments were given after plants reached 4 weeks old. The experiment was conducted within a period of July to December 2016 in a green house of Institut Teknologi Bandung. Mean temperature, solar intensity and humidity were 30/23 °C, 8907 lux and 74,8% respectively.
2.2 Procedures

Number of leaves were counted after ten weeks of exposure. Plant dry weight were measured after plant materials were dried in the oven at 85°C for 48-72 hours. Relative water content (RWC) of the plant was collected after ten weeks exposure. Ten pieces of leaf, with diameter of 1 cm were weighted as fresh weight (FW), then the leaf pieces were placed in petridish containing distilled water for 24 hours to reach turgid weight (TW), or weight of saturated. Dry weight was determined by weighing dried leaf pieces placed in oven 80- 85°C for 24 hours. RWC was calculated using fraction from Turner:\n
\[
\text{RWC (\%) } = \left( \frac{\text{FW} - \text{DW}}{\text{TW} - \text{DW}} \right) \times 100
\]

Photosynthetic pigments, such as chlorophyll a, chlorophyll b, and total chlorophyll, are determined using Arnon [3]. A total of 0.1 grams of fresh leaves crushed with 10 mL of 80% acetone. The absorbance was measured using visible spectrophotometer at wavelengths of 645 and 664 nm. The results are used to calculate the absorbance of chlorophyll content using the formula in Rocha et al. [6]:

\[
\begin{align*}
\text{Chlorophyll a (mg/gFW)} & = \frac{12.7 A_{663} - 2.69 A_{645}}{1000 \text{FW}} \times V \\
\text{Chlorophyll b (mg/gFW)} & = \frac{22.9 A_{645} - 4.68 A_{663}}{1000 \text{FW}} \times V \\
\text{Total chlorophyll (mg/gFW)} & = \frac{8.02 A_{663} - 2.02 A_{645}}{1000 \text{FW}} \times V
\end{align*}
\]

Where:

V : volume of sample (mL)
A : absorbance
FW : fresh weight of sample (g)

Proline content in the leaf tissue was assayed according to method of Bates et al. [7]. A total of 0.5 grams of fresh leaves was soaked in liquid nitrogen and crushed in a mortar, then homogenized by 10 mL sulfosalicylic acid (3%), and filtered with Whatman #1 filter paper. The 2 mL of the filtrate was treated with 2 mL of ninhydrin acid and 2 mL of glacial acetic acid in a test tube and heated in boiling water (100 °C) for 1 hour. The reaction was terminated by incubating test tube into ice. The reaction was extracted by using 4 mL of toluene, homogenized by vortex for 15-20 seconds. Absorbance was measured at wavelength 520 nm using visible spectrophotometer. Proline levels are determined based on reading of standard solution of pure proline.

Glycinebetaine in leaf tissue was assayed according to method Grieve and Grattan [8]. A total of 0.5 grams of fresh leaves was soaked in liquid nitrogen and crushed in a mortar, then homogenized by 20 mL aquades and shock 24 hours and filtered with filter paper. Filtrate was diluted with 1 M sulfuric acid (H₂SO₄) (1:1). Solution about 0.5 mL was transferred into microtube and cooled in ice for 1 hour. The solution was treated with 0.2 mL KI-I2 cold solution then homogenized by vortex. The mixture was kept at a 4°C for 16 hours. The mixture was centrifuged 10000 rpm for 15 minutes to obtain iodide crystals. Crystals were dissolved in 9 mL 1,2 dichloroethane, allowed to stand for 2 hours and its absorbance was measured at a wavelength of 365 nm.

All the data were recorded as mean ± standard error of six replicate. Mean values of each treatment were analyzed using one-way analysis of variance (ANOVA) (SPSS 16.0) with p<0.05.

3. RESULTS AND DISCUSSION

Plant dry weight and number of leaves decreased with increasing drought stress (Table 1). Plant dry weight decreased by 75% and 87.5% at moderate drought stress, and severe drought stress from control. Compared to control, number of leaves decreased by 63% and 43%, respectively for moderate and severe drought stresses. Growth is an increase in the size of the plant relating to the number and size of the cell. The number and size of cells will be reduced due to reduced water potential inside the cell, causing a decrease in plant growth [9].

Reduction of plant growth under drought stress was estimated as due to decrease of turgor pressure, then inhibit cell division and cell elongation which induce reduction plant growth [1]. Drought stress leads to reduce water uptake by plant roots, which result in reduced water content in tissues. The decline in water levels in the tissues cause a decrease in turgor pressure that will inhibit cell division and elongation, resulting in decreased growth, number of leaves, and dry weight of plants [1][10]. It is estimated that as a mechanism of plants against water deficit in the soil [11].
Level of RWC shows variation of water status in plant leaf\cite{12}. Plants with high RWC suspected as plants resistant to drought\cite{13}. Drought stress had little effect on RWC of cilembu sweet potato. After 10 weeks exposure to drought stress, RWC on severe drought stress decreased 10% from control (Fig. 1A). This interpreted cilembu sweet potato could survive in drought conditions and dry area. Leaf chlorophyll content in sweet potato plant decreased with increasing drought stress. Chlorophyll content in moderate drought stress and severe drought stress significantly decreased by 20% and 40%, respectively (Fig. 1B). Yooyongwech et al. \cite{14} reported decreased levels of chlorophyll occur on three genotypes of sweet potato under drought conditions. The decrease in total chlorophyll content in the plant may be attributed to more denaturation of chlorophyll under drought stress, and lower photosynthesis as reported in Barley \cite{15}. Khayatnezhad and Gholamin \cite{16} reported that decreased levels of chlorophyll in maize under drought stress. Total chlorophyll in chickpea cv.bivaniej also decreases when experiencing drought in vegetative and anthesis phase, but different with total chlorophyll in chickpea cv.ILC482 was not significantly different from control after experiencing drought stress in vegetative and anthesis phase \cite{17}. However, the level of chlorophyll is influenced by the intensity of drought stress and plant genetic (cultivar types)\cite{14}. In moderate drought stress, chlorophyll content slightly declined, which indicates that cilembu sweet potato capable to maintain photosynthesis ability. Under drought stress photosynthetic electron transfer and chlorophyll content also declined in pea plants \cite{19}. The results indicated that chlorophyll content was a more sensitive parameter to drought stress compared to RWC.

\begin{table}
\centering
\caption{Dry weight and number of leaves in cilembu sweet potato plant under drought stress}
\begin{tabular}{|l|l|l|}
\hline
Treatment (field capacity) & Dry weight (g) & Number of leaves \\
\hline
Control (80\%) & 8.79 ± 1.15c & 30 ± 3.0c \\
Light stress (60\%) & 5.71 ± 0.65b & 24 ± 2.0b \\
Moderate stress (40\%) & 2.91 ± 0.52a & 19 ± 2.0ab \\
Severe stress (20\%) & 1.56 ± 0.11a & 13 ± 1.0a \\
\hline
\end{tabular}
\end{table}

Values are means ± standard errors of six replicates. Different lowercase letters in each column indicate significant difference among treatments at P ≤ 0.05

Proline content increased at moderate and severe drought stress (Fig. 2A) 2-fold compared to controls, after ten weeks exposure drought stress. Yooyongwech et al. \cite{14} reported accumulation of proline in three sweet potato genotypes increased up to 44 times higher than control under drought stress. Proline accumulation in chickpea cultivars also increased significantly different from control under drought stress at vegetative only and anthesis stage only, but proline content did not differ from control when chickpea cultivars were experienced drought along vegetative until anthesis stage \cite{17}.

\begin{figure}[h]
\centering
\includegraphics[width=\textwidth]{RWC.png}
\caption{RWC (A) and chlorophyll content (B) Cilembu sweet potato plant (I. batatas (L.) Lam.) 10 weeks exposure drought stress. Different letters represent significant compared with control (p = 0.05).}
\end{figure}

\begin{figure}[h]
\centering
\includegraphics[width=\textwidth]{Proline.png}
\caption{Proline (A) and glycinebetaine (B) content Cilembu sweet potato plant (I. batatas (L.) Lam.) 10 weeks exposure drought stress. Different letters represent significant compared with control (p = 0.05).}
\end{figure}

However, higher concentration of proline in cilembu sweet potato plant, under water stress, indicates the role as adaptive compatible solute. Similar to proline, glycinebetaine accumulation in leaves of cilembu sweet potato increased 80% in severe drought stress from control after ten weeks exposure (Fig. 2B). Glycinebetaine acts as
compatible solute in dealing with drought stress by maintaining turgor pressure. Glycinebetaine also plays a role in protecting the plant physiological processes such as photosynthesis and protein synthesis of the effects of drought \cite{20}. Glycinebetaine accumulation also reported in maize under drought stress\cite{21}. Glycinebetaine content also increased under drought condition in peanuts, which are influenced by the level of stress and the duration with increased 6–8 times compared to control\cite{22}. Under drought stress, plant accumulate organic osmolyte like proline and glycinebetaine to maintain osmotic balance. These compounds replace water in biochemical reactions inside the cell to maintain normal metabolism without any phenotypic abnormality to plant \cite{23}.

4. CONCLUSIONS

Severe drought stress for a period of 10 weeks exposure inhibited plant growth, although the plants were still able to survive. Significantly higher concentration of proline and glycinebetaine in plant grown under drought stress indicates their role as adaptive osmotic solutes which enabling plants to survive. Although the experiment was terminated before cilembu sweet potato plant produced tubers, it showed that the plant has good tolerance to drought stress. Cilembu sweet potato plant is therefore can be recommended to be cultivated in relatively dry area.

5. ACKNOWLEDGEMENT

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Mycorrhizal and Soil Nutrient Characteristics of ITS Green Spaces


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Abstract – Environmental assets are major aspect that should be considered as principal components to support EcoCampus program. Institut Teknologi Sepuluh Nopember (ITS) has launched ITS-EcoCampus program. One of its focus activities is the development and maintenance of campus green spaces. Soil characteristic and fertility become indispensable parameters assuring the campus green spaces sustainability. This study aims to characterize the biological and chemical aspects, which include mycorrhizal identification and macronutrient content analysis in some areas of ITS green spaces, respectively. We detected 3 genus of mycorrhiza in 23 areas of ITS green spaces, including Glomus, Acaulospora and Gigaspora. The observations demonstrated that ITS green spaces have low AMF abundance. However, among those three genus, Glomus species were predominantly found in the areas of research. Supporting this results, the majority of ITS green space areas showed neutral to slightly alkaline pH, which favours Glomus spore germination. Moreover, chemical analysis also demonstrated that the ITS green space areas contain high amount of available phosphorous content and low level of nitrogen. This condition also inhibit arbuscular mycorrhizal development.

1. INTRODUCTION

Human overpopulation and industrialization, which move concomitantly, have made environmental issues become more complex. This phenomenon has raised awareness of both individuals and institutions associated with the sustainability of human life. University, as an educational institution, could play a central role in assuring environmental sustainability. University could provide environmental education or be directly involved as a role model for safeguarding the environment. Institut Teknologi Sepuluh Nopember (ITS) as part of the Indonesian educational institution has decided to take part of the movement by implementing ITS-EcoCampus program. This program includes campus “greening” initiatives, which aims to protect and improve the quality and quantity of the existing vegetation around the ITS green spaces (IGS). The soil characteristics and fertility are indespensable parameters that influence successfulness of development and maintenance of campus green spaces.

Soil fertility is the basis for plant productivity. It depends on the biological, chemical and physical status of soils. In addition, the availability of soil nutrients from organic matter is tightly influenced by the presence of soil microorganisms. One of the microorganisms that forms association with plant root is mycorrhiza. Mycorrhiza is a form of beneficial symbiosis between plant root and fungi which can promote nutrient enhancement [1]. There are two major types of mycorrhiza, which are ectomycorrhiza and arbscular mycorrhiza. In the case of ectomycorrhiza, the association is formed with intercellular fungal penetration into the cortical region of plant root. Meanwhile, arbuscular mychorriza could penetrate both inter- and intracellularly into the root cortex. The latter is commonly found in a wide variety of plants, and is classified into three major classes (Archaeosporomycetes, Glomeromycetes and Paraglomeromycetes), and into five orders including Gigasporales, Archaeosporales, Diversisporales, Glomerales and Paraglomerales [2]. These arbuscular mycorrhizas serve as an important element of soil microbial community in order to improve plant nutrition [3]. Gemma et al (2002) also demonstrated that many plants perform an obligate relationship with arbuscular mycorrhiza in poorly nutrient soils. In addition, the mutual association between these fungi and plant root could increase plant drought tolerance and plant immunity against disease [5]. This study aims to characterize the biological and chemical aspects, which include mycorrhizal identification and macronutrient content analysis in some areas of ITS green spaces, respectively.

2. METHODS

2.1. Mycorrhizal identification

Samples for mycorrhizal characteristics were collected from 23 areas of ITS green spaces (Figure 1). Soil
samples were (100 gr) taken from the rhizosphere area with 30 cm depth. Each sample was then homogenized in 500 ml of water. Subsequently, the samples were then filtered through multiple wet sieving method (0.180; 0.075; 0.063 and 0.038 millimeter). Finally, the suspension was briefly centrifuged and then was added with 7 ml 60% sucrose. Final centrifugation was done at 2000 rpm, for 7 minutes. The supernatants were then filtered through 0.038 mm sieve, and placed into vial bottle before being analyzed under optical microscope. The arbuscular mycorrhizal spores were morphologically identified using INVAM (International Culture Collection of Vesicular-Arbuscular Mycorrhizal Fungi, http://www.invam.caf.wvu.edu).

![Figure 1. Sampling areas.](image)

### 2.2. Macronutrient analysis

Thirteen soil samples were used to investigate pH and macronutrient content, including Nitrogen (N), Phosphorous (P), and Potassium (K), using Kjedahl, Bray I and NH$_4$OAc pH 7 extraction method, respectively.

### 3. RESULTS AND DISCUSSION

We detected 3 genera of mycorrhizal spores in 23 areas of ITS green spaces. These include *Glomus*, *Acaulospora* and *Gigaspora*. These mycorrhiza belong to the Arbuscular Mycorrhizal Fungi (AMF). The observations demonstrated that ITS green spaces have low AMF abundance. However, among those three genus, *Glomus* species were predominantly found in the areas of research (Figure 2).

![Figure 2. Arbuscular Mycorrhizal Fungi (AMF) found in 23 areas of ITS green spaces. AS, area around ITS student dormitory; HK, area around forest campus; K, area around eight pond; ND, area around Nasdec Building; R, area around robotika building; TA, area around alumni garden; UF, Urban Farming](image)
Among those 23 areas of sampling, the R1 (IGS around Robotika building) has highest Glomus spores abundance. Meanwhile, AS2 (area around student dormitory) possessed lowest Glomus spore. Both Gigaspora and Acaulospora spores have been found only in the area of Robotika building (R1 and R2, respectively). This results also suggest that IGS around Robotika building might possess good diversity of mycorrhiza compared to other areas.

The Glomus spore (Figure 3) dominance in the area of research may be correlated with the pH of soils. Some studies have also reported that pH of the soil influences diversity of mycorrhiza, and, in some extent, it might tightly affect mycorrhizal infection and germination [6-7]. Our chemical analysis showed that the majority of ITS green space areas showed neutral to slightly alkaline pH (Table 1), which favours Glomus spore germination [8]. In addition, the predominance of Glomus in this study is also in agreement with [9], which stated that Glomus species often dominate agricultural soils and performs a wider adaptation in varied soil condition [10]. Moreover, Glomus has also been reported to fastly produce more spores compared to other genera [11].

![Figure 3. Glomus spores in four different locations. A, lokasi R1 (IGS around Robotika); B, UF (IGS around Urban Farming); C, ND1 (IGS around Nasdec) dan D, HK1 (IGS around Alumni garden)](image)

In term of the micronutrient content of the soils, our result showed that nitrogen content is quite varying, from low category to intermediate (TA2). However, mostly are considered as low level of N. Meanwhile, most locations exhibit high amount of potassium (K), except of UF1 and TA1 which possess low and intermediate level of K, respectively. The same trend has been also observed in the case of phosphorus (P). High to very high amount of P is detected in almost all locations (Table 1).

<table>
<thead>
<tr>
<th>No</th>
<th>Kode</th>
<th>pH</th>
<th>N (%)</th>
<th>P$_2$O$_5$ (ppm)</th>
<th>K (Cmol/ kg)</th>
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<tbody>
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</table>
The overall low abundance of AMF spores in this study may also be correlated with the macronutrient contents of the soils. The majority of ITS green space areas contain high amount of available phosphorous content and low level of nitrogen (Table 1). This condition probably affect the diversity and abundance of AMF [12]. This observation was also in line with [12], which demonstrate that elevated phosphorous has been shown to inhibit arbuscular mycorrhizal development. Liu et al (2016) also stated that plants grown under low P are efficiently colonized by AM fungi. Therefore, a high level of P fertilization seems to inhibit AM symbiosis predominantly by acting on the plant itself rather than on the content of its root exudates or on the fungal partner.

4. CONCLUSIONS

The observations demonstrated that ITS green spaces have low AMF abundance. However, among those three genus, the *Glomus* species were predominantly found in the areas of research. Supporting this results, the majority of ITS green space areas showed neutral to slightly alkaline pH, which favours *Glomus* spore germination. Moreover, chemical analysis also demonstrated that the ITS green space areas contain high amount of available phosphorous content and low level of nitrogen, which also probably inhibit arbuscular mycorrhizal development. Therefore, fertilization management seems to be determined further for avoiding the over-use of P, which probably inhibit mycorrhizal development.

5. REFERENCES

Morphology Response of Alfalfa (*Medicago sativa* L.) based on Level Gamma Ray Irradiation with Tissue Culture Methods

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Abstract – Forage needed in rations of ruminant feed reached 40-80% of the total ration DM, or about 1.5 - 3.0% of the live weight of cattle (Abdullah et al, 2005) [1]. Medjemen et al. (2015) and Wattimen et al (2011) indicated that tissue culture technique is plant propagation technique that is able to produce uniform crop seeds, in large quantities, and in a relatively shorter time [2]. Alfalfa (*Medicago sativa* L.) or Lucerne is a perennial herbaceous legume with superior forage quality. Gamma ray irradiation technique necessary for Alfalfa produces varieties of legume plants that are resistant to drought. Irradiation using gamma rays in the aspect of plant breeding is very beneficial in developing new varieties or clones mutants. This study aims to measure response of the alfalfa plant morphology based on level of gamma ray irradiation. The experimental data were analyzed using analysis of variance (ANOVA), with the differences among treatments were further tested using Duncan Test. Parameters observed were leaf number, plant canopy height, and leaf color. The results of this research show that the potential changes in plant morphology of Alfalfa (*Medicago sativa* L.), as the effects of gamma ray irradiation, indicate that the effectiveness of gamma ray irradiation is in the level 400 Gy based on the morphology factor such as the changes in the plant height and leaf color.

Key words: alfalfa, tissue culture, gamma irradiation, morphology

1. INTRODUCTION

Forage needed in rations of ruminant feed reached 40-80% of the total ration DM, or about 1.5 - 3.0% of the live weight of cattle (Abdullah et al, 2005). Legume plants are one of the forages that is used in animal foods as a protein source that can replace the addition of concentrate food. It is inefficient due to the large portion of the costs of concentrate is close to 100% of the total feed cost. Alfalfa (*Medicago sativa* L.) is a leguminous plant that contains enough nutrients, can potentially be used as food material which rich of protein and chlorophyll. This plant has the potential to be used as for goats, for its production and nutritional values is quite high (Sirait et al, 2011) [3]. Burns et al. (2005), which utilized alfalfa (in the form of hay) for livestock for feeding goats, obtained good results, namely: dry matter intake 207-370 g / 10 kg; BK digestibility of 68.4 to 73% and the digestibility of crude protein of 80.4 to 81.7% [4]. According to studied of Radovic et al. (2009), Alfalfa requires good drainage, with a pH of 6.5 or more, and good soil fertility [5]. This is an obstacle for farmers to grow alfalfa in some areas, especially on the soil conditions that have poor drainage. The development of plant biotechnology, such as tissue culture and induced mutants is done with gamma irradiation on the cultivation of crops in order to maximize seed Alfalfa forage superior, uniform, in large numbers and a relatively short time. The use of energy, such as gamma rays on the plant, will provide a good influence in the field of agriculture, with a dose of gamma radiation treatment on acquired plants that have desirable characteristics such as: production from the high resistance to disease and etc (Batan 2006) [6]. This study aims to measure the response of the alfalfa plant morphology based on the level of gamma ray irradiation.

2. METHODS

2.1 Materials

This study was conducted from December 2016 to January 2017, in the Plant Tissue Culture Laboratory of Animal Feed Science and Technology section Plant Feed and Pasture, Faculty of Animal Husbandry, Bogor Agricultural University. Materials and equipment used were plant seeds Alfalfa (*Medicago sativa* L.), and materials for
sterilization such as 70% alcohol, detergents, NaClO 5.25%, distilled water, MS (Murashige and Skoog), PGR (plant growth regulator) 2,4D (dichlorophenoxyacetic acid) and cytokinin, culture equipments, calipers, Laminar Air Flow Cabinet, as well as equipment used for observation (calipers, LCC (Leaf Color Chart)).

2.2 Procedures

Preparation and Seeding

Preparation of explants: the explants to be used are from seed Alfalfa have been irradiated with gamma rays at National Atomic Energy Agency (BATAN) Patir, South Jakarta, with a radiation level of 0 Gy, 100 Gy, 200 Gy, 300 Gy, 400Gy, and 500 Gy. Alfalfa seeds were sown after 30 days post-irradiation, then sterilized using detergent and rinsed with sterile water until the foam that forms is missing, then sterilized by HgCl2 for 5 minutes, and then rinsed three times until the seeds are ready for sowing (Bonyanpour and Khosh-Khui 2013). Sterile seeds were sown on Murashige and Skoog (MS) basal for 6 weeks [7].

Subcultures of Explants and Growth

Explants have been sowing done subculture on MS medium plus growth regulator (PGR) cytokinin (BAP) 1 mg L⁻¹ in order to grow the plant canopy. The explants with individual differences in the level of gamma rays irradiation (100, 200, 300, 400, 500 Gy) were observed for six weeks in a sterile laboratory with lighting for 16 hours with a light intensity of 700 Lux white light. Parameters observed in this study are leaf number, the height of the plant, leaf color, and the weight of the plant.

3. RESULTS AND DISCUSSION

<table>
<thead>
<tr>
<th>Table 1 Effect of Gamma Rays Irradiation Level Combination (Gy) of the amount Alfalfa Leaf</th>
</tr>
</thead>
<tbody>
<tr>
<td>Time of Growth</td>
</tr>
<tr>
<td>----------------</td>
</tr>
<tr>
<td>0MST</td>
</tr>
<tr>
<td>1MST</td>
</tr>
<tr>
<td>2MST</td>
</tr>
<tr>
<td>3MST</td>
</tr>
<tr>
<td>4MST</td>
</tr>
<tr>
<td>5MST</td>
</tr>
</tbody>
</table>

Explanation: MST = Weeks after planted.

Gamma irradiation up to the level of 500 Gy has not shown significant changes (p < 0.05) in the number of LEAves, which suggest that high levels of irradiation do not indicate that genetic changes linked to productivity on the plant LEAVES alfalfa (Medicago sativa L.). These conditions made possible the genetic instability that allows for the recovery of post alfalfa plants subculture process. In contrast to the research of Sinambela et al (2015) [8] that administration level gamma ray irradiation significantly reduced the number of leaves of onion plants planted in open fields, as well as results of research Cahyo (2015), which showed that irradiation doses given very significantly reduced the number of leaves formed on orchid plantlets in vitro [9]. The higher level WHICH was GIVEN to the orchid plantlets then the ability to review the forming of leaves INCREASES more slowly. This suggests that genetic engineering is relatively secure in term of the stability performance of the plant alfalfa (Medicago sativa L.).
Table 2 Effect of Gamma Rays Irradiation Level Combination (Gy) of the amount Alfalfa Plant Height

<table>
<thead>
<tr>
<th>Time of Growth</th>
<th>Control</th>
<th>100</th>
<th>200</th>
<th>300</th>
<th>400</th>
<th>500</th>
</tr>
</thead>
<tbody>
<tr>
<td>0MST</td>
<td>7.43±3.68</td>
<td>7.32±1.93</td>
<td>9.59±2.14</td>
<td>12.54±3.69</td>
<td>10.18±1.67</td>
<td>15.08±6.43</td>
</tr>
<tr>
<td>1MST</td>
<td>7.94±2.32c</td>
<td>8.41±1.85bc</td>
<td>11.51±2.61abc</td>
<td>14.25±3.27a</td>
<td>13.19±0.61ab</td>
<td>16.23±5.88a</td>
</tr>
<tr>
<td>2MST</td>
<td>13.51±4.84</td>
<td>14.33±4.53</td>
<td>21.91±4.43</td>
<td>18.07±5.55</td>
<td>19.04±1.32</td>
<td>19.99±5.02</td>
</tr>
<tr>
<td>3MST</td>
<td>13.38±2.53c</td>
<td>16.45±2.88bc</td>
<td>27.18±5.69a</td>
<td>18.85±4.85bc</td>
<td>22.42±7.97ab</td>
<td>21.28±5.15abc</td>
</tr>
<tr>
<td>4MST</td>
<td>12.27±1.83b</td>
<td>14.77±4.42b</td>
<td>27.59±6.22a</td>
<td>16.26±5.15b</td>
<td>25.61±5.03a</td>
<td>20.72±9.50ab</td>
</tr>
<tr>
<td>5MST</td>
<td>10.89±1.95b</td>
<td>13.81±6.43b</td>
<td>28.02±6.41a</td>
<td>15.16±5.69b</td>
<td>26.38±5.28a</td>
<td>19.98±9.31ab</td>
</tr>
</tbody>
</table>

Explanation: MST = weeks after planted, values followed by the same letter are not significantly different indicates based on DMRT test at the level of 5%.

Morphological response to high-level Alfalfa plants irradiation of 400Gy significantly effects the total plant height, which means that at this level of gamma irradiation, it indicates improving crop candidate genetic engineering results. This is in contrast to the results of research done by Harsanti (2015) saying that the higher the level of irradiation, the plant growth will decrease [10], and by Sinambela et al (2015) saying that the administration of gamma-ray irradiation at a local variety of onion Samosir significantly reduce the length of the plant. This shows that the high doses causing stunted growth, difficult rooted and no growth. The influence of the growth was caused by physiological damage due to gamma-ray irradiation (Qosim 2007) [11]. This study showed that the Alfalfa plant still has a good vigor after gamma irradiation up to the level of 400Gy.

Table 3 Effect of Gamma Rays Irradiation Level Combination (Gy) of the Alfalfa Leaf Color

<table>
<thead>
<tr>
<th>Time of Growth</th>
<th>Control</th>
<th>100</th>
<th>200</th>
<th>300</th>
<th>400</th>
<th>500</th>
</tr>
</thead>
<tbody>
<tr>
<td>0MST</td>
<td>2.38 ± 0.41</td>
<td>2.50 ± 0.58</td>
<td>2.25 ± 0.50</td>
<td>2.50 ± 0.58</td>
<td>2.00 ± 0.00</td>
<td>2.00 ± 0.00</td>
</tr>
<tr>
<td>1MST</td>
<td>2.50 ± 0.50</td>
<td>2.00 ± 0.00</td>
<td>2.25 ± 0.50</td>
<td>2.00 ± 0.00</td>
<td>2.50 ± 0.58</td>
<td>2.75 ± 0.50</td>
</tr>
<tr>
<td>2MST</td>
<td>2.50 ± 0.50b</td>
<td>2.00 ± 0.00c</td>
<td>2.00 ± 0.00c</td>
<td>2.00 ± 0.00c</td>
<td>3.00 ± 0.00a</td>
<td>2.00 ± 0.00c</td>
</tr>
<tr>
<td>3MST</td>
<td>2.25 ± 0.43</td>
<td>2.00 ± 0.00</td>
<td>2.25 ± 0.50</td>
<td>2.25 ± 0.50</td>
<td>2.75 ± 0.50</td>
<td>2.25 ± 0.50</td>
</tr>
<tr>
<td>4MST</td>
<td>2.33 ± 0.71</td>
<td>1.67 ± 0.47</td>
<td>2.25 ± 0.50</td>
<td>1.90 ± 0.81</td>
<td>2.25 ± 0.50</td>
<td>2.50 ± 0.58</td>
</tr>
<tr>
<td>5MST</td>
<td>2.04 ± 0.58b</td>
<td>1.17 ± 0.69c</td>
<td>3.00 ± 0.00a</td>
<td>1.98 ± 0.84b</td>
<td>2.04 ± 0.39b</td>
<td>3.00 ± 0.00a</td>
</tr>
</tbody>
</table>

Explanation: MST = weeks after planted, values followed by the same letter are not significantly different indicates based on DMRT test at the level of 5%.

Leaf color chart indicates that engineering of gamma rays is not negatively affecting the morphology of color on Alfalfa plants in the first week, and the third week to the fourth week, but significantly different in the third week with leaf color change was seen in the treatment with gamma-ray irradiation on the level 400Gy. This is in contrast to the results of research done by Wardhani (2007) saying that the larger the dose of radiation given then the leaves change color from dark green to yellow-green [12]. The results of this study showed that there are tolerance levels where Alfalfa plant leaf color change to the level of gamma rays irradiation.

Table 4 Effect of Gamma Rays Irradiation Level Combination (Gy) of the Alfalfa Plant Weight

<table>
<thead>
<tr>
<th>Variable</th>
<th>Control</th>
<th>100</th>
<th>200</th>
<th>300</th>
<th>400</th>
<th>500</th>
</tr>
</thead>
<tbody>
<tr>
<td>Plant Weight</td>
<td>0.33 ± 0.04</td>
<td>0.33 ± 0.05</td>
<td>0.50 ± 0.32</td>
<td>0.42 ± 0.07</td>
<td>0.47 ± 0.30</td>
<td>0.32 ± 0.03</td>
</tr>
</tbody>
</table>

The results showed that the level of gamma irradiation does not leave a negative effect on the weight of alfalfa plants, and it means that the alfalfa plant is still able to absorb nutrients properly from the growing media. Treatment of gamma-ray irradiation at a particular dose may improve the biochemical activity of explants such as enzymes and phytohormones so the explants can improve its ability to absorb the mineral from the culture medium (Kadir, 2011) [13]. According to Hoesen (2011), the ratio between the nitrogen compound of ammonium nitrate contained in ms media can influence the process of differentiation, growth, and development of explants [14].
4. CONCLUSIONS

Potential changes in plant morphology of Alfalfa (*Medicago sativa* L.) as the effects of gamma ray irradiation indicates that the effectiveness of gamma ray irradiation is in the level 400 Gy based on the morphology factor such as the changes in the plant height and leaf color. However, the difference doses of gamma irradiation do not impact negatively on morphological responses such as leaf number and weight of the plant.

5. REFERENCES


Distribution Role of Nitrogen and Glutamate on Citrus Canopy (Citrus Hystrix L.) Toward Citronellal Content in Leaves

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Abstract – The extend remobilization of nutrient on crops so far is known well. However, as we assumed that the re-translocation of the particular nutrient as nitrogen has a strong affect to the terpenoid metabolic like citronellal, In many cases, the environment stress, such as drought, induced volatile compound as citronellal leaves in citrus. The aim of this experiment is to observe the distribution of citronellal as consequence the distribution of nitrogen on different layer of tree canopy. The research was conducted by obtaining the samples of leaves, which were collected from different layer of citrus canopy, from 10 samples of trees on each part of field: bottom layer, middle layer and upper layer of canopy. The randomized leaves were collected and analyzed by the HPLC and Spectro-photometer to measure the content of nitrogen, and glutamate distribution, correlated to the citronellal content. The results indicate close correlation of distribution of nitrogen between bottom layer, middle layer, and upper layer, of lemon leaves, to produce glutamate and citronellal. The remobilization of nitrogen from bottom leaves contains of 1.35 ppm, followed by middle leaves, which contains of 1.4 ppm, and the highest content is from the upper leaves, which is 1.65 ppm. As consequence, the glutamate and citronellal increase with the same trend as the those nutrient distributions. The glutamate seems to be the precursor of the secondary metabolic, which was converted into the harvested leaves of citrus. Increasing of 5% glutamate from the upper leaves to the middle leaves, and the bottom leaves, seem to be much accumulated of citronellal content in the bottom leaves.

Keywords: nutrient distribution, upper leaves, middle leaves, bottom leaves, citronellal, glutamate.

1. INTRODUCTION

Citrus hystric L is one family of citrus grown in Indonesia. The citrus type was harvested commonly as fresh leaves which are used as pharmaceutical industries material. However, the quality of secondary oil (from leaves) as a secondary metabolic production, are not reported as well as their growth on different soil condition. The objective this experiment is to study how the transportation of nutrient, particularly on nitrogen as substrate to produce the secondary metabolic (citronellal in citrus), occur. There are differences between content of nitrogen to canopy leaves, in term of citronellal as well as the organic matter production. It would be the fertilization particularly nitrogen is very important, following the age of plant and reproduction of new leaves after pruning.

2. METHODS AND APPROACH

The research was conducted at field of Citrus hystric L., in Kesamben, Blitar, East Java, on rainy season on November-December 2016. The targeted area is located at 8°08'53.2"S 112°22'03.4"E, with elevation of 190 m above sea level. Leaves were collected from 40 plants, which were 4 years old after planted. The sample leaves were pruned from three canopy layers to separates the leaf materials. (upper leaves, middle leaves, and bottom leaves). Each sample was taken about at 200g on each layer per plant. The collection of leaves was repeated from 10 plants, as samples from each quadrant (North, South, East, West). The measurement included:

- Citronellal (total secondary metabolism) in leaves from three layer of canopy, the total nitrogen and glutamate on leaves, total nitrogen and organic acid material in soil, HPLC
3. RESULTS AND DISCUSSION

Based on the graph above, that the totals of nitrogen in leaves are different among the leaves, which placed on upper, middle, and bottom canopy (figure 1.). It is assumed that the distribution of nitrogen depends on where the leaves were placed, which might tranlocate the nitrogen from the bottom leaves to upper in all young leaves. Since the macronutrient (nitrogen) is a faster re-translocated than the micronutrient among the leaves, as a mobile transport in plant (Taiz and Zeiger, 2010), consequently, the carbon cycle as the product photosynthesis process form the simple organic matter higher in bottom leaves and nitrogen accumulation higher concentration in upper leaves. It also present in glutamate production which reduce in moderate and bottom leaves compared to upper leaves. (figure 2).

On the other hand, the organic matter shows the higher on bottom leaves, as results of increasing metabolism during the nitrogen re-translocated, and reducted to carbohydrate and organic molecules (figure 3).
The interestingly that the secondary metabolite as citronellal on the higher concentration in bottom leaves, following by those concentration in other leaves placed assumed that the organic compounds ready to be reducted into the last product (figure 4). Those metabolism address to be re-translocate and storage into vacuoles in old leaves (bottom). The nutrients partitioned on the sink organs which required more vegetative growth. Indeed, the metabolism processes of carbohydrate reduction will be much take place on old leaves or bottom canopy (Barunawati, 2013).

However, the supporting data of nitrogen distributed on region of citrus field, is that the whole of field had the similarly concentration. It means that the distributions of nitrogen and others nutrient do not strongly affect to nutrient absorption and distribution into plant. Thus, the production of citronellal in the citrus plants, as well as the organic compound, were strongly influenced by the distribution of nitrogen and glutamate among the canopy (figure 5).

4. CONCLUSIONS
The re-translocation of nitrogen and assimilation of glutamate effected to content of secondary metabolic and organic matters.

The bottom leaves had the lower of nitrogen and glutamate, and higher of secondary metabolic (assumed citronellal substrate).

The upper leaves had the higher of nitrogen and glutamate and lower of secondary metabolic.

5. REFERENCES
Diversity and Composition of Tree Species of the Secondary Tropical Lowland Forest as a Response to Structure Change of the Meranti-Dangku Landscape, South Sumatra, Indonesia

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Abstract – The diversity and composition of tree species of the tropical lowland forest will change, when there are fragmentation and changes of landscape structure of natural forest, and the dynamics of changes in the composition and diversity of trees species occur in the landscape [1]. However, there are variations among the study areas, which can be determined by tracing the linkage and interaction among indicators of changes in the composition and diversity of tree species [2]. The research objectives are to examine the the trees species diversity forest, and interactions between species on the landscape structure change in the Meranti-Dangku forest, to better understand the effects of landscape modification against individuals and groups of species. We expect that the research findings can be used to enhance the efforts on the forest restoration and biodiversity conservation. We applied a tree vegetation analysis of fragmented natural secondary forest, and conducted observations in 32 units of sample plots, whose size is 20m x 50m. Ecological analysis of all tree species with dbh ≥ 10 cm was conducted by identifying the local name, latin name of the tree species, and its typical characteristics, and then making the sample of herbarium. Hill’s Diversity Number produced a family of number related to diversity, effective species richness, a rare species and the abundance of common species in the population [3]. We also calculated the importance value index of tree species and the Jaccard similarity index, to differentiate the level of the forest succession and the gradation among sub-landscape. The results showed significance of some parameters, which has played important roles in the detecting changes in biodiversity, namely index of species richness and species evenness, and basal area of a tree stand. Three other parameters are not significant. The changes of the tree species composition of the natural forest succession, as respond to the changes of landscape structure, and the gradation among sub-landscape, can be used as a reference on species selection in rehabilitation and restoration of the fragmented natural forest, as a part of the planning on human-modified landscape.

Keyword: Diversity indices, Importance Value Index, Similarity Index, Inter-correlation.

1. INTRODUCTION

The diversity and composition of tree species of the tropical lowland forest will change when there are fragmentation and changes of landscape structure of natural forest, and the dynamics of changes in the composition and diversity of trees species occur in the landscape [1]. However, there are variations among the study areas, which can be determined by tracing the linkage and interaction among indicators of changes in the composition and diversity of tree species [2].

The research objectives are to examine the trees species diversity forest and interactions between species on the landscape structure change in the Meranti-Dangku forest, to better understand the effects of landscape modification against individuals and groups of species. We expect that the research findings can be used to enhance the efforts on the forest restoration and biodiversity conservation.

2. METHODS

2.1 Materials
Work map of field survey in the form of an Indonesia topographical map of 1:50,000 scale, a vegetation cover type map as resulted from interpretation of Landsat TM Imagery acquisition in 2013, GPS handheld, magnetic compass, clinometer Suunto, meter types, phi band, Haga, camera, and materials needed to make herbarium.

2.2 Procedures

We applied a tree vegetation analysis of fragmented natural secondary forest and conducted observations in 32 units of sample plots, whose size 20m x 50m. We measure diameters of all tree species with dbh ≥ 10 cm, identified the local name, latin name of the tree species and its typical characteristics, and next made the sample of herbarium for identification in Herbarium Bogoriensis, LIPI.

To analysis of trees species diversity and composition, we use Hill’s Diversity Number, which produced a family of number related to diversity, effective species richness, effective number of species, evenness of species, and the abundance of common species in the population [4; 3]. We also calculating the tree density and basal areal of tree stand [5], Importance Value Index of tree species [6], Shannon-Wiener Diversity Index, Simpson Dominance Index, and Simpson Evenness Index [7]. We also calculate the Jaccard Similarity Index [8; 7] to differentiate the structur of forest, level of the forest succession and the gradation among sub-landscape.

3. RESULTS AND DISCUSSION

Species Accumulation Curve

Within 32 sample plots, which covers 3.2 ha, we found a total of 774 species and 1,598 trees, and we processed data of species into a species - accumulation curve. We found 341 species and two unidentified species. The species-accumulation curve showed that it has reached the asymptotic number, where the addition of sample plot did not significantly increase the number of species.

Composition and Diversity of Trees Species

We use Hill's Number of Diversity [9] to determine the richness of species, effective number of species (true diversities), abundance distribution of species, and the evenness of species, as well as the structure of the forest trees, i.e. the density of trees and basal area of trees stand (Table 1). We calculate the important value index of tree species, Jaccard's index for measuring the similarity between communities, and Shannon and Wiener Index to differentiat the level of forest succession, entropy, and gradien between the sub-landscape.

The results of the study showed that the increase in the effective number of species (true diversities) correlated with a decrease in evenness of species. This is the identifier of the process of secondary succession of forest ecosystems, which indicates the occurrence of a grouping of the new tree species population distribution (clumpiness).

Importance Value Index of Tree Species

Based on the highest order of the important value index, the main species composition in the sub-landscape of Kapas, i.e. Palaquium gutta-percha (Hook) Ball, Litsea sp, Xanthophyllum rufum, Benn, Shorea parvifolia Dyer, and Castanopsis acuminitissima DC. In the sub-landscapes of Meranti Ulu (specially of you trees class), Meranti Ilir and Dangku are dominated by secondary species or pioneers species, such as Bellucia pentamera Naudin, Bellucia axinanthera Trian, Macaranga gigantea (Rchb. f. & Zoll.) Mull. Arg., Endospermum diadenum (Miq.) Airy Shaw, and Ficus variegata Blume. The degraded natural forest has a small average of an important value index, none of dominant species, and the species composition is similar to the natural forest (old growth dipterocarp forest).

Ecological analysis showed significance of some parameters, which have played significant roles in detecting changes in biodiversity, namely index of species richness, species evenness, and basal area of a tree stand. Three other parameters are not significant. The changes of the tree species composition of the natural forest succession, as respond to the changes of landscape structure, and the gradation among sub-landscape, can be used as a indicator in species selection in rehabilitation and restoration of the fragmented natural forest, as a part of the human-modified landscape.
Barbour states [8] that the value of Shannon's and Wiener Diversity Index can range between 0-7, with criteria: 0-2 (low), 2 (medium), and > 3 (high). Thus, the level of species diversity index of young and mature trees at four locations are classified as high.

Simpson's Dominance Index of Dangku forest was the highest at both young and mature trees. However, for the forest at Dangku had Shannon's and Wiener (Table 2). Diversity Index is lower, at both young and mature trees, compared to a third of other locations. Specifically, the young trees, with a diameter of ≥10–30 cm, at Dangku had the highest Simpson's Evenness Index, compared to an other three locations.

3.1 Discussion

**Correlation between Tree Species Diversity Index**

The calculation of the value of the correlation between the vegetation of the forest biodiversity indices, was conducted based on Hill's Diversity Number, and structure of the forest stand, performed in Kapas, Meranti Ulu and Meranti Ilir, indicating there is a very significant positive correlation \((p=0.01)\) between the number of species in the plot or the richness of species, and the number of effective species (true diversities), and the abundance of species distribution, and as well as between the number of effective species with an abundance of species distribution.

In addition, in Kapas, there was a significant positive correlation \((p = 0.01)\) between a richness of tree species and tree density of forest stands, as well as a highly significant positive correlation \((p = 0.01)\) between the abundance of tree species distribution with a basal area of forest tree stands.

In Meranti Ulu, shows that the richness of species and the effective number of species (true diversities) in Meranti Ulu, both negatively correlated in a very significant \((p=0.01)\) against species evenness. This means that if the total number of species in the plots and the effective number of species increased, so this is precisely indicated that species evenness will decrease. Species diversity of ensembles in undisturbed primary forest was distinctly higher than in disturbed or secondary forest. Disturbed forest with tall trees remaining appeared to represent impoverished subsets of the undisturbed primary forest community [10].

**Importance Value Index of Tree Species**

The analysis to the top ten IVI showed that each group of forest on each location, and a different diameter class was dominated by different species, thus the ability of species to live in a place is very depending on its ability to adapt to environmental conditions of the habitat. Therefore, the environmental conditions are very instrumental in the selection of species to be able to survive in a habitat.

There is no dominant young tree species, where the average IVI is low. This means that in the sub-landscape Kapar has a high species richness, especially for young trees. Old dipterocarp forests in Kapas and Meranti Ulu sub-landscape are managed using silviculture system of the restoration of natural forest ecosystem, the species of young trees and mature trees, have low IVI compared to IVI in secondary forest in sub-landscape of Meranti Ilir and Dangku.

**The Implication against Island Biogeography Theory**

Every ecologist know [11], that, Island Biogeography Theory (IBT) states that the number of species in a habitat Island is governed by a balance of dependence distance colonization and dependence of extinction area, and it predicts that the habitat of the smaller and more isolated will be supported by fewer species.

A fragmented landscape, whether in the dissection phase, dissipation or break apart, then in accordance with the IBT will happen to lose and decline in species richness, species-area curve as that hypothesized by IBT [12].

Deviations against the IBT as stated by [13] are, that, the dynamics of populations and communities are often heavily reinforced by the relatively fragmentary habitat against the natural condition. Likewise, other discoveries are that habitat fragmentation affects different species in different ways. Some species are declining sharply, or disappeared in fragments, remain stable in other area, and increases dramatically, elsewhere [14].

The results of this research also show, that, the young growth tree species in the sub-landscape of Meranti Ulu and Meranti Ilir, and the young growth and mature growth of tree species in sub-landscape of Dangku, were dominated by a secondary tree species, which controlled the growth phases of a tree in the process of forest growth.

<table>
<thead>
<tr>
<th>Tree-diversity Index</th>
<th>Kapas</th>
<th>Meranti Ulu</th>
<th>Meranti Ilir</th>
<th>Dangku</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Shannon's and Wiener Index</td>
<td>4.51</td>
<td>3.04</td>
<td>6.60</td>
<td>5.22</td>
</tr>
<tr>
<td>2. Simpson Dominance Index</td>
<td>0.01</td>
<td>0.06</td>
<td>0.02</td>
<td>0.03</td>
</tr>
<tr>
<td>3. Simpson Evenness Index</td>
<td>0.01</td>
<td>0.04</td>
<td>0.01</td>
<td>0.01</td>
</tr>
</tbody>
</table>

Table 2. Index of tree diversity of young tree (dbh ≥ 10-30 cm) and mature tree (dbh >30 cm) at four locations of the Meranti-Dangku landscape.
succession. The symptoms are so common on the type of vegetation that lead to the climatic conditions and to stability. The species composition of the natural forest, that has developed in the long run, will show physiognomy, fenology, and low power regeneration, and tends to be steady. Thus, the new dynamics of the forest community are manifestly and less conspicuously, where the turning generations, or species regeneration, does not seem as if, as a result of certain rare species are dominant, since all species have to adapt in a prolonged period of time [15].

At the stage of forest climax, then, the IBT is likely to be re-occur [15]. It was observed during this research results that the reduction of the number of populations and species of trees occurred, when the phase of forest fragmentation is Attrition, and an increase in the number of species of trees and evenness index of tree species on the secondary growth forest, during the phase of Dissipation and Dissection [16].

4. CONCLUSIONS

The effect of the change of landscape metrics, especially on the regrowth and regeneration of the forest succession, is significantly to increase the richness of species, the evenness index of tree species, and basal area of tree stand. The increase in the effective number of species (true diversities) was followed by a decline in the evenness index of tree species, and the grouping of the new tree species population (clumpiness). This is an indicator of the occurrence of a succession process of the secondary forest ecosystems.

Dangku sub-landscape has an average value of Shannon’s and Wiener entropy index, an index of tree species richness, a tree density, and basal area, which are higher than in Meranti Ilir sub-landscape. This indicates that the entropy condition for the succession process of secondary forest ecosystems in Dangku sub-landscape relatively more steady than on Meranti Ilir sub-landscape.

Secondary forest of the adjacent sub-landscape of Meranti Ilir and Dangku are experiencing the high pressure from the deforestation and forest degradation., An average value of density of trees stand is almost the same, and has a very significant positive correlation between density of trees stand and index of tree species evenness. However, the Jaccard’s similarity index both sub-landscapes is very low.

The change of landscape metrics led to the decolonization of the composition and diversity of tree species in the succession of secondary natural forest, and significantly cause a rise in the index of tree species evenness, (clumpiness). This indicates that the entropy condition for the succession process of secondary forest ecosystems in Dangku sub-landscape relatively more steady than on Meranti Ilir sub-landscape.

5. REFERENCES

The Enhancement of Vetiver (Vetiveria zizanioides (L.) Nash) Essential Oil Production in The Symbiotic System with Glomus aggregatum N. C. Schenck & G. S. Sm. using Hydroponic Medium with Various Phosphate Content

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Abstract – Glomus aggregatum is an arbuscular mycorrhizae, and its symbiosis with vetiver (Vetiveria zizanioides L. Nash) can enhance plant growth, essential oil production, and variety of essential oil components. The objectives of this study is to evaluate the effect of mycorrhizae interactions with vetiver in hydroponic system on plant growth, phosphorus uptake, and essential oil quality and quantity. Vetiver plants were inoculated with Glomus aggregatum for 4 weeks, acclimatized in Hoagland solution, then planted in three various Hoagland solution (0.1; 0.2; 0.5). The results of this study indicate that the infected plants grown in 0.2-strength Hoagland solution resulted in the highest specific growth rate (0.0381 g/day), phosphorus uptake (99.8%), and total oil content (3.49%). According to these results, it is evident that the symbiosis of Glomus aggregatum with vetiver enhanced plant growth, efficiency of phosphorus uptake, and essential oil production, especially in 0.2-strength Hoagland solution i.e. phosphorus of 6.2 mg P/L or 27.2 mg KH2PO4/L.

1. INTRODUCTION

Vetiver essential oil (Vetiver Root Oil of Vetiveria zizanioides), is one of the unique prime commodities from West Java, particularly in Garut. Vetiver oil production is highly prospected to be continuously developed since its opportunity on the market share is high for both the domestic and overseas markets. As the prime secondary metabolite of vetiver, vetiver oil is basically one of the main ingredients for the manufacture of perfumes and other cosmetics. The secondary metabolites production of a plant can be improved, either abiotically or biotically, one of the methods is by using arbuscular mycorrhizal fungi or AMF. AMF forms symbiosis with the roots of a plant to get nutrients, and the plant in return will also get extra nutrients due to its interaction with the AMF [1]. Many studies have proven that the AMF can enhance growth of several species of plants, including vetiver, such as research conducted by [2]. The growth increase was mainly caused by the enhancement of phosphate intake by the host plant with the help of AMF. Phosphate is an essential element for the plant growth and development, seed production, as well as the growth of the AMF itself. A research had shown that the AMF increased the production of secondary metabolites in medicinal plants [3]. Other research [4] also showed that different species of AMF induced the production of different component in vetiver oil, resulting in a different aroma. Therefore, the research to increase metabolite production and variation of oil component in an interaction of vetiver with AMF is needed in order to evaluate the growth and the nutrient uptake, especially phosphate, as well as the effect of AMF on the quantity and quality of vetiver oil production. Moreover, it is also expected that the optimal phosphate concentrations can be obtained to increase the production and/or quality of vetiver oil. This research was carried out by planting crops hydroponically. Hydroponic system is selected to minimize other variables that can possibly interfere the response of plant and reduce the level of result validation, as the one that had been conducted by [5] and [6].

2. METHODS

2.1 AMF Culture Inoculation on Plants

The experiment was conducted at Jatinangor in December 2015-June 2016. Vetiver (Vetiveria zizanioides (L.) Nash) clumps were obtained from vetiver plantations in Samarang, Garut, West Java. Pure G. aggregatum culture as the AMF was obtained from SEAMEO BIOTROP. The culture was propagated in zeolite and it was made homogeneous with a density of 10-12 spores/gram.

Inoculation of vetiver by AMF was conducted in sterile soil-sand mixtures as the mediums (mixed homogeneously with the ratio of 1:1). Each medium was added by AMF culture as much as 7 grams, and they were then mixed homogeneously. Control plants were the plants which were not inoculated with AMF. The
cultivation was carried out for 4 weeks. Confirmation of plant roots degree of infection by AMF was done by staining procedure. The staining method followed [7] with some modifications.

### 2.2 Plant Cultivation in Hoagland Medium

Acclimatization of plants were carried out in half-strength Hoagland solution for 1 week. The pH range of the solution were maintained from 5.6 to 5.9 [6]. After the acclimatization period, phosphate treatment is applied with various Hoagland strengths: 0.5 (as control) containing 68 mg KH$_2$PO$_4$/L or 15.5 mg P/L; 0.2 containing 27.2 mg KH$_2$PO$_4$/L or 6.2 mg P/L; and 0.1 containing 13.6 mg KH$_2$PO$_4$/L or 3.1 mg P/L [6]. Phosphate compounds were provided in the form of KH$_2$PO$_4$. Three plants of each treatment were harvested in every 6 days during the 18 days experiment.

### 2.3 Plant Growth, Phosphate Concentration, and Oil Content Analysis

Sample plants of each treatment were harvested and the samples was weighed. After that, the plants were divided into leaves and roots. The oil of the roots was extracted with n-hexane by a soxhlet, then it was analyzed by GC-MS. The phosphate concentrations of Hoagland solution were measured at the beginning and end of the experiment. The measurement of phosphorus in Hoagland solution was done using a colorimetric method [8].

### 2.4 Oil Extraction and Terpenoid Analysis

The extraction of oil in the 95% n-hexane was done by using soxhlet at the temperature of 50-70°C for 4 hours. N-hexane were then evaporated by a rotary vacuum evaporator with the operating conditions as follows: the waterbath temperature of 50°C, the vacuum pump pressure of 350 mbar, and evaporator flask rotation speed of 60 rpm. Evaporation was done until the solvent evaporated completely. The oil yield was determined as the oil mass divided by the mass of dry roots (% m/m).

The remaining of vetiver oil inside the evaporator flask was redisssolved with 5 cm$^3$ of 100% n-hexane. The constituents of vetiver oil were identified using GC-MS, with flame ionization detector and a column of 5% phenyl methyl silox (30 x 0.25 mm x 0.25 mm). The components of vetiver oil were analyzed with comparing the samples’ mass spectrum and the mass spectrum of the internal standard in the database.

### 3. RESULTS AND DISCUSSION

#### 3.1 AMF Infection on Plants

The result of the root staining which was taken after inoculating plants with the AMF for a month in a soil medium is shown in Figure 1 (a). This picture shows that the roots of the plant have been successfully infected by *G. aggregatum*. Some parts of the root tissues turned blue because aniline blue colored the infecting hyphae on the between-cells space.

![Figure 1 Results of the staining of vetiver roots which are (a) infected with AMF for 1 month in soil medium, (b) infected with AMF and grown in hydroponic system, (c) not infected (control plant)](image)
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The staining was also carried out on the AMF-infected vetiver roots which was cultivated in the hydroponic system. The result was shown in Figure 1 (b). In Figure 1 (b), the infected tissues were indicated by the red circle. In this figure, it showed that the roots were infected by *G. aggregatum*. However, no vesicle observed. Similar results were obtained in the study conducted by [9]. Compared to Figure 1 (b), Figure 1 (c) showed no blue color in control plant.

The colonization of AMF in vetiver grown in the soil habitat was better than in hydroponic system. The result was supported by a study [5], which showed a minimal interaction between AMF and vetiver that were grown in a hydroponic system. According to [10], the AMF did not grow optimally on plants in hydroponics system due to the insufficient nutrient content in the medium solution, which was mainly consumed by plants to support their growth.

### 3.2 The Effects of Various Hoagland Solution on The Growth of AMF-Infected Vetiver

Figure 2 (a) and (b) are the NI and I plants fresh weight comparison (*‘NI’ code is intended for "without AMF" plants, the ‘I’ is for "with AMF" plants, and the number that follows the code indicates the strength of the Hoagland solution. In Figure 2, bar with full color is for NI, while the bar with hatches is for I). Based on the charts, it is shown that the mean of plant growth at day-18 is increased. I plants generally grew faster than the NI plants.

![Figure 2](image)

**Figure 2** Fresh weight comparison of (a) NI plants and (b) I plants

Table I (a) and (b) showed specific growth rate and doubling time of the non inoculated (NI) and inoculated (I) plants. In Table I (a) and (b), it showed that the specific growth rate of I plants was always higher than the NI plants’, thus doubling time of I plants was lower than the NI plants’. These results demonstrated role of AMF on plant growth in general. NI plants depended only on the root uptake of the nutrients provided by Hoagland solution, whereas I plants were supported by the AMF to absorb nutrients more quickly. This result was similar with the literature [11].

![Table 1](image)

**Table 1** The specific growth rate and doubling time of NI plants (a) and I plants (b)

<table>
<thead>
<tr>
<th>Hoagland Strength</th>
<th>P (mg/L)</th>
<th>Parameters (Average)</th>
<th>Hoagland Strength</th>
<th>P (mg/L)</th>
<th>Parameters (Average)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Specific Growth Rate</td>
<td></td>
<td></td>
<td>Specific Growth Rate</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(g/days)</td>
<td></td>
<td></td>
<td>(g/days)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Doubling Time</td>
<td></td>
<td></td>
<td>Doubling Time</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(days)</td>
<td></td>
<td></td>
<td>(days)</td>
</tr>
<tr>
<td>0.5</td>
<td>15.5</td>
<td>0.0226</td>
<td>30.64</td>
<td>0.5</td>
<td>15.5</td>
</tr>
<tr>
<td>0.2</td>
<td>6.2</td>
<td>0.0220</td>
<td>31.50</td>
<td>0.2</td>
<td>6.2</td>
</tr>
<tr>
<td>0.1</td>
<td>3.1</td>
<td>0.0219</td>
<td>31.65</td>
<td>0.1</td>
<td>3.1</td>
</tr>
</tbody>
</table>

The results of the study conducted by [11] showed that the smaller amount of phosphorus added to the AMF-infected plants, the higher the growth rate. The growth rate of I plants added by 0.2 and 0.1-strength Hoagland were higher than the one that was added by 0.5-strength Hoagland, because in the high level of nutrients, especially phosphorus, AMF colonization did not affect the growth of plants, which might be due to the reduction of the AMF effectiveness to colonize and absorb nutrient in a medium with a high level of nutrient, particularly phosphate [2].

Although the AMF colonization in this experiment showed positive results, the difference was insignificant. The result might be related to the AMF that did not perform in plants with hydroponics system. The weak association that occurred between the AMF with the host plant did not induce the plant growth and quality as expected. These results were consistent with experiment results from study conducted by [10].
3.3 Effects on Plant’s Phosphorus Intake Balance

Table 2 shows data of simple phosphorus mass balances of vetivers which were grown in hydroponic system. In Table 2, the remaining phosphorus in Hoagland solutions was only less than 1%, except for NI 0.5 and I 0.5. The remaining phosphorus quantity in Hoagland solutions, used by AMF-infected plants, tend to be lower than those used by plants that were not infected by AMF. The amount of remaining phosphorus was much larger in 0.5-strength Hoagland solution because the available phosphorus exceeds the plants’ needs, thus the phosphorus was not completely absorbed by plants. Moreover, as the AMF-infected plants’ specific growth rate was higher than the non-infected ones, the remaining phosphorus amount in the Hoagland solution used previously by the AMF-infected plants should be lower than the non-infected plants’. Allegedly, the phosphate was absorbed by plants directly as in NI plants.

<table>
<thead>
<tr>
<th>Hoagland Strength</th>
<th>Parameters</th>
<th>Hoagland Strength</th>
<th>Parameters</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>P provided (mg/300 cm³)</td>
<td>P remained (mg/300 cm³)</td>
<td>P absorbed (mg/300 cm³)</td>
</tr>
<tr>
<td>0.5</td>
<td>4.65</td>
<td>0.22654</td>
<td>4.42346</td>
</tr>
<tr>
<td>0.2</td>
<td>1.86</td>
<td>0.00393</td>
<td>1.85607</td>
</tr>
<tr>
<td>0.1</td>
<td>0.93</td>
<td>0.00332</td>
<td>0.92668</td>
</tr>
</tbody>
</table>

3.4 AMF Effects on Total Essential Oil Content

Terpenoids analysis using neutral red reagent gave positive result, which was shown in Figure 3. It indicated that there were terpenoid compounds in vetiver root tissues, marked by the change of red color to darker [12] around the cortical cells adjacent to the endodermis, on the outer side. This result was supported by study result conducted by [13], which proved that vetiver root tissue produced the essential oil in the innermost layer of the cortical parenchyma tissue. This tissue is located adjacent to the endodermis. Based on the results, it can be concluded that vetiver roots used in this study contains terpenoids compounds. In addition, there was a rhexigen glands formed in the cortical root tissue. It is commonly formed in vetiver roots which accumulate a typical compound of vetiver [14].

![Figure 3 Terpenoids presence and distribution in vetiver root](image)

The essential oil yield from plant roots in day-18 for all treatments was shown in Figure 4. In Figure 4, the highest essential oil yield was produced in I 0.2 treatment, while the lowest one was produced in I 0.1 treatment. The role of AMF was shown by treatment I 0.5 and I 0.2, since it produced higher yield of essential oil than that in NI 0.5 and NI 0.2. Result from treatment I 0.5 was 35.21%, higher than NI 0.5. I 0.2 also produced 35.21%, higher than NI 0.2. However, result obtained from treatment I 0.1 was low. The result could be an indication that I 0.1 allocated its nutrients to grow and survive, so that the nutrients used for the oil synthesis was very low. These results were in contrast to study conducted by [4], G. aggregatum did not increase the essential oil yield, the experiment results also showed that vetivers which formed a symbiosis with G. aggregatum produced essential oil yield 17.96%, lower than the control plants. Allegedly this might be due to different interaction conditions that occur between this studies and the research in literature.

The yield of essential oil obtained from I 0.2 treatment was high, so was the specific growth rate. Therefore, I 0.2 plants used the nutrition to grow and produce oil efficiently. On the other hand, NI 0.5 were the least efficient plants in using supplied nutrients, because with larger amount of nutrition, the vetiver growth and its essential oil...
yield was not higher than the plants from the other treatments, it also left higher amount of phosphates in the hydroponic medium.

![Figure 4 The essential oil yield (% m/m) from plant roots in day-18 for all treatments](image)

### 3.5 AMF Effects on Composition and Variety of The Essential Oil Compounds

The list of terpene compounds and the proportion of compounds contained in essential oils of each treatment is shown in Table 3. The comparison for essential oil obtained from the experiment was the commercial vetiver root oil from Samarang, Garut. The commercial oil contained terpenes (mostly composed of sesquiterpenes) in a larger amount compared with the oil obtained from vetivers which were grown in hydroponic system. However, the oil from this studies contained more compounds, i.e. 39-50 substances, than the commercial one’s, which contains only 29-34 substances. The concentration of particular vetiver sesquiterpenoid compounds in the commercial oil were higher (1-4%) than the concentration of each sesquiterpenoid of the oil obtained from experiments, that was just around 0.2-1.5%. The total content of terpenes in the commercial oil was also much higher (27.9%) than the total content of terpenes in the oil from experiment (less than 5%). These results might be due to the commercial essential oil was harvested from 11-13 months old vetivers [15], while the oil from experiments was obtained from 6 months old plants. Because of the plants used in the experiment was too young, the compounds found in the essential oil might be still in the form of precursor in vetiver root oil biosynthesis.

**Table 3** The list of terpene compounds and the proportion of substances contained in commercial essential oil and oil from plants of each treatment

<table>
<thead>
<tr>
<th>Essential Oils</th>
<th>Terpene Compounds</th>
<th>Compounds in Oil (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td><strong>Terpene</strong></td>
</tr>
<tr>
<td>Commercial</td>
<td>α-copaene</td>
<td>27,9</td>
</tr>
<tr>
<td></td>
<td>caryophylenol-ii</td>
<td></td>
</tr>
<tr>
<td></td>
<td>9,10-dehydro-2-norzizaene</td>
<td></td>
</tr>
<tr>
<td></td>
<td>valencen-12-ol</td>
<td></td>
</tr>
<tr>
<td></td>
<td>dehydroaromadendrene</td>
<td></td>
</tr>
<tr>
<td></td>
<td>ylangene</td>
<td></td>
</tr>
<tr>
<td></td>
<td>β-eudesmene</td>
<td></td>
</tr>
<tr>
<td></td>
<td>isocurcumenol</td>
<td></td>
</tr>
<tr>
<td></td>
<td>alloaromadendrene</td>
<td></td>
</tr>
<tr>
<td></td>
<td>cyclostativen</td>
<td></td>
</tr>
<tr>
<td></td>
<td>khusiloic acid</td>
<td></td>
</tr>
<tr>
<td></td>
<td>α-cedrenal</td>
<td></td>
</tr>
<tr>
<td>NI 0,1</td>
<td>α-vetivone</td>
<td>4,9</td>
</tr>
<tr>
<td></td>
<td>copaene</td>
<td></td>
</tr>
<tr>
<td></td>
<td>β-cadinene</td>
<td></td>
</tr>
<tr>
<td></td>
<td>4,β.h, 5,α.h-cis-eudesma-6,11-diene</td>
<td></td>
</tr>
<tr>
<td></td>
<td>epiprezizaene</td>
<td></td>
</tr>
<tr>
<td>NI 0,2</td>
<td>dehydroaromadendrene</td>
<td></td>
</tr>
<tr>
<td></td>
<td>khusimone</td>
<td>0,9</td>
</tr>
<tr>
<td>NI 0,5</td>
<td>(-)-selina-5,11-diene</td>
<td></td>
</tr>
<tr>
<td></td>
<td>khusimone</td>
<td>2,3</td>
</tr>
</tbody>
</table>
**4. CONCLUSION**

*Glomus aggregatum* can form a symbiosis with vetiver roots, but it is better in a soil medium than in a hydroponic medium. In hydroponics system, vetiver, which interacted with *Glomus aggregatum*, produced a higher growth rate compared with the vetivers which did not form interaction with *Glomus aggregatum*. In general, vetivers interacted with *Glomus aggregatum* in hydroponic systems absorbed and utilized the phosphorus as their nutrient more efficiently than the non-inoculated vetivers. Vetivers interacted with *Glomus aggregatum* also produced higher content of essential oil than the non-inoculated vetivers, although with a smaller content of terpenoids.

**5. REFERENCES**

Potentiality of Plant Combination for Removing Indoor Air Pollutants: Fundamental Physiology of *Euphorbia milii*, *Sansevieria trifasciata*, and *Dieffenbachia seguine* – A Review

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Abstract – The indoor air pollution could be caused by physical, chemical and biological pollutants. Many studies showed the ability of an individual plant to absorb volatile chemical compounds. In addition, several plants also had the ability to emit phytochemical compounds that functioned as anti-microbes. However, the report of plant combination for air pollutant removal was little. This review explains the plant combination potential to be applied as indoor air pollutant removal which is included *Euphorbia milii*, *Sansevieria trifasciata*, and *Dieffenbachia seguine*. Three different plants are chosen mainly based on their ability to absorb polar and non-polar gaseous pollutants. The ability of the plant to produce phytochemical and the type of photosynthesis of plant also be used as consideration.

1. INTRODUCTION

The indoor air pollution could be influenced by outdoor air pollution, and also directly caused by anthropogenic activities, such as cigarette smoking, cooking, and painting, as well as furniture volatile emission [1-4]. In addition, the activities of unhealthy animals and humans might emit airborne microorganisms in indoor air environment [5], [6]. The phytoremediation is a low-cost technology to mitigate the indoor air pollution. Several studies described that the individual plant effectively removed airborne pollution in the closed system as a model of the indoor air environment [7], [8]. However, it was a lack of report for applying plant combination to remove air pollutants. Therefore, a physiological-based review of the plant combination potential for removing indoor air pollution is important to give a new insight.

2. METHODS

This review is arranged from several references.

3. DISCUSSION

The individual plant had been proven to absorb volatile organic compounds (VOCs). The previously studies mentioned that *Euphorbia milii* absorbed formaldehyde, trimethylamine, and benzene; *Sansevieria trifasciata* absorbed toluene, ethylbenzene, benzene, and trimethylamine; *Dieffenbachia seguine* absorbed formaldehyde and xylene. In addition, *D. seguine* was able to produce phytochemical for anti-bacteria and fungi [7-9]. *E. milii* is a crassulacean acid metabolism (CAM)-cycling species which close their stomata at night under the absence of pollutant (normal condition). The implication of night gas exchange was concluded based on its stem capability of CO$_2$ absorption. Under the presence of trimethylamine as a pollutant, *E. milii* mainly absorbed trimethylamine by its stem although the stomata also had a role [10]. *S. trifasciata* is a CAM plant that opens its stomata at night for CO$_2$ absorption under normal condition. However, it is different from *E. milii* in which, in the case of the presence of trimethylamine, the *S. trifasciata* mainly absorb trimethylamine by its stomata [11]. Meanwhile, *D. seguine* is a C3 plant that absorbs CO$_2$ at day under normal condition. Although the mechanism of aerial part absorption of the plant for removing VOCs had not clearly mentioned, this plant had an important role in controlling biological contaminants [9]. The combination of the three formerly mentioned plants may compete with an electronic device such as air conditioning-equipped with ultraviolet germicidal irradiation as air purifier from chemical and biological air pollutants.

4. CONCLUSIONS

The combination of *E. milii*, *S. trifasciata*, and *D. seguine* is potential to remove polar and non-polar air pollutants and also potential to control biological contaminants. Consider the type of photosynthesis, the combination of plants may balance the CO$_2$ emission from each plant respiration during absorbing the air pollutants.
5. REFERENCES

Plants in *Ai Tahan*, Traditional Medicine of the Tetun Ethnic Community in West Timor Indonesia

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Abstract – For thousands of years, plants become important part of man's efforts to prevent, cure, and treat various diseases. Many people in developing countries still rely on plant-based traditional medicines. Therefore, it is important to conduct researches in botanical, chemical and pharmacological fields, to support the prospect of utilizing the medicinal plants in modern society. The purposes of this study were to collect, identify, and documenting the plants used in “ai tahan” traditional medicine of the Tetun ethnic community in Belu, West Timor Indonesia. The study was conducted by using field method, and then the data were collected by an open and semi-structured interviews, observations, and documentations. Eleven traditional healers namely “makdok”, or “dok”; or “dauk”; were involved in this study. 6 are men and 5 are women, aged between 40-60 years. The results were analyzed and presented in descriptive qualitative and quantitative. The study revealed 142 species of plants belong to 62 families used in the various formulas of “ai tahan”. The plants consist of 71 were trees, 18 shrubs, 15 lianas, 33 herbs, 2 ferns, and 1 parasite, orchid and also lichen. Fabaceae/Leguminosae is the dominant family (12.7%), followed by Moraceae (6.3%), Euphorbiaceae (5.8%), Poaceae (3.5%), Sterculiaceae, Solanaceae, Rubiaceae, Apocynaceae, Arecaceae, Asteraceae, Compositae and Convolvulaceae (2.8%, respectively). Almost all of plants used as medicine are non-cultivated plants. The leaves are the most commonly used part (28.2%), followed by stem bark (24.7%), roots (18.3%), a mixture of roots, stem barks, leaves, etc. (16.9%), fruits or seeds (6.3%), heartwood (2.1%), bulbs (1.4%), and clove, rhizome and sap (0.7%, respectively). In preparing of the medicine, plant part(s) processed in a simple way as brewed or boiled to drink; boiled for bathing, mashed to the sick place of the body, refined and mixed with coconut oil to rub and massage, or just chewed to be rubbed on the affected body part. There are about 369 formulas to treat various ailments and complaints. *Garuga floribunda*, *Melia dubia*, *Hyptis pectinata*, *Jatropha curcas*, *Helicteres isora*, *Ficus hirta*, and *Melanolepis multiglandulosa* are the plants often found in various formulas of “ai tahan”. The Tetun ethnic community of Belu, West Timor has a culture of traditional medicine that is mainly based on their local plants, and continued through generations by oral instruction. The recording and the preservation of the knowledge in the written form are necessary.

Key words: Medicinal plant, traditional medicine, Tetun ethnic group, Timor, field method.

1. INTRODUCTION

Every ethnic group community has local knowledge, such as the traditional medicine [1-6]. World Health Organization defined the term of "traditional medicine" is the summary of the knowledge, skills, and practices based on the theories, beliefs and experiences indigenous to different cultures that are used to maintain and improve health, as well as to prevent, diagnose, and treat physical and mental illnesses [7-10]. Besides, by Indonesian Government Regulation No. 102/2014 on Traditional Health Services, the traditional drug is an ingredient in the form of plant, animal, and mineral materials, galenic formulas, or mixtures of these materials that historically used for treating many diseases and injuries.

For thousands of years, plants have become an important part of a human effort to treat various diseases [11-24]. In the Timor island, native people have the knowledge of traditional medicines and use it in the prevention and treatment of diseases. An ethnic group of West Timor Indonesia which called “Tetun” dominantly lives in Kabupaten Belu and Malaka. They still use traditional medicine and often engage it in various rituals of traditional medicine. Traditional medicine practice in the Tetun ethnic group usually performed by a shaman or traditional health practitioner (traditional healer) called “makdok” or “dok” or “dauk”, or the older people who also have the knowledge and skill about traditional medication. In the treatment practice, the traditional health practitioners often use various ingredients of herbal medicine [25, 26], called “ai tahan” (*ai* = tree/wood, *tahan* =
leaf; Tetun language in Belu) or “ai fuan” (fuan = fruit), or “kwa” (Tetun in Malaka) or “ai moruk” (moruk = bitter, bitter wood; Tetun in East Timor).

Some ethnomedicine knowledge in many indigenous communities is going to be disappeared. The ethnomedicine information is continued orally by generations and it would be faded by modern health care information [27]. Therefore, it is necessary to preserve the public’s knowledge about traditional medicine, especially in the Tetun ethnic group. The effort starts from invention, utilization, cultivation, and re-excavation of the local knowledge of traditional medicine. Study of the use of plants by traditional communities is followed by the identification of its phytochemicals study, preclinical study, and clinical trial. It is an important approach to discover and develop new drug candidates from the source of traditional medicine.

2. METHOD

2.1 Study area

A research for recording the medicinal plants and traditional medicine prescriptions was performed by Simon Mali (the co-author) during 16 weeks, from April to July 1991, in 5 villages (desa) namely Naitimu, Lookeu, Naekasa, Tukuneno and Jenilu, in Kecamatan Tasifeto Barat, Kabupaten Belu (124°40' – 125°15' E and 8°7’ – 9°23’ S). The study area was located at the central of Timor Island, East Nusa Tenggara Province, Indonesia. The villages were randomly determined according to the place where the Tetun ethnic group live.

2.2 Ethnomedicinal data collection

Ethnomedicinal data were collected by means of open and semi-structured interviews, observations and documentations. The interview was designed to focus on the local names of plants, their various medicinal applications, the parts of the plant used, and the methods of preparation and administration to the patients. Eleven shamans or traditional health practitioners or traditional healers (6 men and 5 women) were involved in this study; 4 from Naitimu, 1 from Naekasa, and 2 from Lookeu, Tukuneno, and Jenilu, respectively. Selection of these traditional health practitioners was done by purposive sampling, according to the local community leaders’ (village head and cultural leaders) recommendation.

3. RESULTS AND DISCUSSION

Traditional medicinal plants

The family name of the plants, vernacular names, botanical names, plants forms, plant parts used, and use of the plants in the ethnomedicine are shown in Table 1. We use the original terminology as it was presented by the informants to avoid any misinterpretation. The study revealed 142 plant species belongs to 62 families which were used for medicinal purposes by the traditional healers of the Tetun ethnic community in Kabupaten Belu. The Fabaceae/Leguminosae family is the dominant family (18 species, 12.7%), followed by Moraceae (9 species, 6.3%), Euphorbiaceae (8 species, 5.6%), Poaceae (5 species, 3.5%), Sterculiaceae, Solanaceae, Rubiaceae, Apocynaceae, Arecaceae, Asteraceae, Compositae and Convolvulaceae (4 species, 2.8%, respectively). Of the total 142 species, the trees were the plant form used most frequently (71 species, 50.0%), followed by herbs (33 species, 23.2%), shrubs (18 species, 12.7%), lianas (15 species, 10.6%), ferns (2 species, 1.4%), and parasite, orchid and lichen (1 species, 0.7%, respectively).

Plant part used

According to the frequency of the plant part used, the leaves are the plant parts most commonly used (40 species, 28.2%), and then followed by stem bark (35 species, 24.7%), roots (26 species, 18.3%), a mixture of roots, stem barks, leaves, etc. (24 species, 16.9%), fruits or seeds (9 species, 6.3%), heartwood (3 species, 2.1%), bulbs (2 species, 1.4%), and clove, rhizome and sap (1 species, 0.7%, respectively). The different parts of a plant are usually used for the treatment of different ailments or disorders.

Formulation, preparation, and administration of the traditional medicine

There are about 357 types of formulas of various medicinal plant materials are used to treat more than 100 ailments and complaints. The parts of the plant are processed and used as a single material, or more common, as a mixture with the other plant material. Most of the formulas (recipes) of the traditional medicine consist of two or more plants and even found a formula that contains seven types of plants. Plants could be a major component in a formula for the treatment of an ailment or complaint and become an additional component in the formula for others. Based on the content of the various formulas of traditional medicine, it was found that Garuga floribunda is the most frequently found in various formulas (14 formulas), followed by Melia dubia (12 formulas), Hyptis pectinata (9 formulas), Jatropha curcas (8 formulas), and Helicteres isora, Ficus hirta and Melanolepis multiglandulosa (7 formulas of each plant).
### Table 1. Plants in ai tahan formulas

<table>
<thead>
<tr>
<th>Family name and vernacular name</th>
<th>Scientific name</th>
<th>Plant form</th>
<th>Plant part used</th>
<th>Use in ethnomedicine</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>ACANTHACEAE</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ai matamuk tasi</td>
<td>Barleria cristata</td>
<td>Shrub</td>
<td>Roots</td>
<td>Horak modo lakan (male genital disease, swelling at the tip of the penis)</td>
</tr>
<tr>
<td>Aimatamuk</td>
<td>Barleria prionitis</td>
<td>Shrub</td>
<td>Leaves</td>
<td>Infected wounds</td>
</tr>
<tr>
<td>Tali mano nunun</td>
<td>Thunbergia alata</td>
<td>Liana</td>
<td>Roots</td>
<td>Cough with phlegm, cough with blood, bloody urine</td>
</tr>
<tr>
<td><strong>AMARYLIDACEAE</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Kilat maromak</td>
<td>Crinum asiaticum</td>
<td>Herb</td>
<td>Bulb</td>
<td>Beriberi</td>
</tr>
<tr>
<td>Koke hudi</td>
<td>Uvaria rafia</td>
<td>Shrub</td>
<td>Stem bark</td>
<td>Heart problem, dysentery</td>
</tr>
<tr>
<td><strong>APOCYNACEAE</strong></td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>Kroki mutin</td>
<td>Alstonia scholaris</td>
<td>Tree</td>
<td>Stem bark, sap</td>
<td>Cough, cough with blood, boils/ulcers</td>
</tr>
<tr>
<td>Kroki metan</td>
<td>Alstonia villosa</td>
<td>Tree</td>
<td>Stem bark</td>
<td>Wormy, lumbago/back pain</td>
</tr>
<tr>
<td>Klitit mean</td>
<td>Wrightia pubescens</td>
<td>Tree</td>
<td>Fruits</td>
<td>Difficult to urinate</td>
</tr>
<tr>
<td>Klitit mutin</td>
<td>Wrightia calycina</td>
<td>Tree</td>
<td>Stem bark, leaves</td>
<td>Headaches, hemarthoids</td>
</tr>
<tr>
<td><strong>ARECACEAE/PALMAE</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Akadirun</td>
<td>Borassus flabellifer</td>
<td>Tree</td>
<td>Leaves</td>
<td>Nosebleed</td>
</tr>
<tr>
<td>Bet</td>
<td>Cyrtostachys lakka</td>
<td>Tree</td>
<td>Fruits</td>
<td>Old- or infected wounds</td>
</tr>
<tr>
<td>Tali, akar</td>
<td>Corrypha gebanga</td>
<td>Tree</td>
<td>Leaves</td>
<td>Asthma, cough with phlegm</td>
</tr>
<tr>
<td>Nawa, tzuawawa</td>
<td>Arrangia pinnata</td>
<td>Tree</td>
<td>Roots</td>
<td>Difficult to urinate</td>
</tr>
<tr>
<td><strong>ASCLEPIADACEAE</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fuka</td>
<td>Calotropis gigantea</td>
<td>Shrub</td>
<td>Roots, sap</td>
<td>Asthma, ulcers, heart problem, gonorrhea</td>
</tr>
<tr>
<td><strong>ASTERACEAE</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Asukar mutin</td>
<td>Siegesbeckia orientalis</td>
<td>Herb</td>
<td>Roots</td>
<td>Cough with phlegm, irregular menstruation, gonorrhea</td>
</tr>
<tr>
<td>Aidoss metan</td>
<td>Hypitis pectinata</td>
<td>Herb</td>
<td>Roots, stem bark, leaves</td>
<td>Cough, cough with blood, fever, new injuries, epilepsy</td>
</tr>
<tr>
<td>Sirikaut lotu</td>
<td>Bidens bidentata</td>
<td>Herb</td>
<td>Leaves</td>
<td>Beriberi</td>
</tr>
<tr>
<td>Ai hak</td>
<td>Erigeron sumatensis</td>
<td>Herb</td>
<td>Leaves</td>
<td>Wounds, hemorrhoids</td>
</tr>
<tr>
<td><strong>BOMBACACEAE</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Kuan, nitas</td>
<td>Bombax malabaricum</td>
<td>Tree</td>
<td>Fruits</td>
<td>Heart problem, diarrhea, rheumatism</td>
</tr>
<tr>
<td>Nunak</td>
<td>Cordia subpubescens</td>
<td>Tree</td>
<td>Stem bark, leaves</td>
<td>Thin body after childbirth, cough with blood, chicken pox (herpes), itchy</td>
</tr>
<tr>
<td><strong>BRYOPCEAE</strong></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>Lumut</td>
<td>Aerobryopsis longissima</td>
<td>Lychen</td>
<td>Whole plant</td>
<td>Gonorrhea, diarrhea</td>
</tr>
<tr>
<td><strong>BURSERACEAE</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Feu</td>
<td>Garuga floribunda</td>
<td>Tree</td>
<td>Stem bark</td>
<td>Postpartum care, fever, itchy, irregular and excessive menstruation, new injuries, old wounds, diarrhea, fractures, bleeding childbirth, rheumatism</td>
</tr>
<tr>
<td><strong>CACTACEAE</strong></td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>Klatun metan</td>
<td>Opuneta sp.</td>
<td>Shrub</td>
<td>Bark</td>
<td>Cough with blood, gonorrhea</td>
</tr>
<tr>
<td><strong>CAESALPINIACEAE</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Frasukten inan</td>
<td>Cassia alata</td>
<td>Shrub</td>
<td>Leaves</td>
<td>Postpartum care, ringworm, new injury</td>
</tr>
<tr>
<td><strong>CASUARINACEAE</strong></td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Kakeke</td>
<td>Casuarina junghuhniana</td>
<td>Tree</td>
<td>Stem bark, roots, fruits</td>
<td>Cough, wormy, male genital diseases</td>
</tr>
<tr>
<td><strong>COMBRETACEAE</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lese, klee</td>
<td>Terminalia edulis</td>
<td>Tree</td>
<td>Stem bark</td>
<td>New wound, infected/old wounds</td>
</tr>
<tr>
<td><strong>COMPOSITAE</strong></td>
<td></td>
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</tr>
<tr>
<td>Fafok</td>
<td>Blumea balsamifera</td>
<td>Shrub</td>
<td>Leaves</td>
<td>Fever</td>
</tr>
<tr>
<td>Ai dois mutin</td>
<td>Ageratum conyoides</td>
<td>Herb</td>
<td>Roots</td>
<td>Fever, new injury/wound</td>
</tr>
<tr>
<td>Kaulele</td>
<td>Emilia sonchifolia</td>
<td>Herb</td>
<td>Leaves</td>
<td>Wounds, cholera</td>
</tr>
<tr>
<td>Kraukidan</td>
<td>Elephanta balsamifera</td>
<td>Herb</td>
<td>Roots</td>
<td>Lumbago/back pain</td>
</tr>
<tr>
<td><strong>CONVOLVULACEAE</strong></td>
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</tr>
<tr>
<td>Talitehuk kasoruk</td>
<td>Ipomoea hederifolia</td>
<td>Liana</td>
<td>Roots, leaves</td>
<td>Asthma, itchy, heart problem, gonorrhea</td>
</tr>
<tr>
<td>Kaboen mean</td>
<td>Operculina turpethum</td>
<td>Liana</td>
<td>Roots, leaves</td>
<td>Clean breast milk, bloody urine, bleeding childbirth, excessive menstruation</td>
</tr>
<tr>
<td>Talitehuk kabuar</td>
<td>Ipomoea obscura</td>
<td>Liana</td>
<td>Roots, leaves</td>
<td>Itchy, gonorrhea</td>
</tr>
<tr>
<td>Fehuk tasi</td>
<td>Ipomoea pescapae</td>
<td>Liana</td>
<td>Roots</td>
<td>Infected wound</td>
</tr>
<tr>
<td><strong>CUCURBITACEAE</strong></td>
<td></td>
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</tr>
<tr>
<td>Kolokoen moru</td>
<td>Gymnopetalum leucosticum</td>
<td>Liana</td>
<td>Leaves</td>
<td>Ringworm, scabies</td>
</tr>
<tr>
<td><strong>CYPERACEAE</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fahiflower inan</td>
<td>Cyperus dilatus</td>
<td>Herb</td>
<td>Roots</td>
<td>Heart problem</td>
</tr>
<tr>
<td><strong>DILLENIACEAE</strong></td>
<td></td>
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<td></td>
</tr>
<tr>
<td>Buku</td>
<td>Dillenia pentagyna</td>
<td>Tree</td>
<td>Stem bark</td>
<td>Headache, migrain</td>
</tr>
<tr>
<td><strong>DIOSCOREACEAE</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Kuan rahuk</td>
<td>Dioscorea pentaphylla</td>
<td>Liana</td>
<td>Bulb</td>
<td>Baby dies in the womb, the placenta did not come out, fracture</td>
</tr>
</tbody>
</table>

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### EUPHORBIACEAE
- **Manumeak lotu, renes** *Phylanthus niruri* Herb Roots, leaves Clean breast milk, bloody urine, burns
- **Nirak, tubi tahak** *Macaranga tanarius* Tree Sap, stem bark Ulcers, irregular menstruation, female genital diseases, constipation
- **Kur mutin** *Gelonium glomerulatum* Tree Stem bark Fever in children, scabies, ringworm, jaundice
- **Badut malaka** *Jatropha curcas* Shrub Stem bark Cough with blood, gonorrhea, diarrhea
- **Donu, kakak lamak** *Melanotepis multiglandulosa* Shrub Stem bark Itching, gonorrhea, fracture
- **Klan, klan** *Breynia cernua* Shrub Leaves Itchy, sore eyes
- **Badut malaka mean** *Jatropha gossypifolia* Shrub Stem bark Heart problem, dirty tongue, bleeding childbirth, excessive menstruation, blood added
- **Ai tahan lalek** *Euphorbia tirucalli* Tree Sap Scabies

### FABACEAE/LEGUMINOSEAE
- **Kru** *Cassia siamea* Tree Stem bark Cough
- **Kenfaek** *Bauhinia malabarica* Tree Stem bark Cough, cough with phlegm, coughing with blood, ear pus/inner ear infection
- **Besak** *Acacia leuca* Tree Stem bark Postpartum care, inflammation of the gums, irregular menstruation, dirty tongue, bleeding childbirth, excessive menstruation, blood added
- **Ai na** *Pterocarpus indicus* Tree Stem bark, sap
- **Sukaer** *Tamarindus indica* Tree Leaves Chicken pox (herpes), gonorrhea, sore eyes, sarampa
- **Taun fuik** *Indigofera sp.* Shrub Leaves Epilepsy
- **Ai fahu kahen** *Albizia lebeckioides* Tree Stem bark Cramps and stiffness, injury at the breast, dysentery, fractures
- **Kidalu** *Albizia procera* Tree Stem bark Stinky nose
- **Lalima wai** *Cassia fistula* Tree Stem bark, leaves Stinky nose, headache, ringworm, constipation
- **Sakiki asu ikun** *Uvaria lagopodioides* Herb Roots The fetus bleed out, bleeding childbirth, excessive menstruation, miscarriage
- **Santuku, sama tuku** *Albizia chinesis* Tree Stem bark Itchy, diarrhea
- **Kakabar lotu** *Desmodium gangeticum* Herb Roots The fetus bleed out, diarrhea
- **Haan fuik** *Phaseolus lunatus* Liana Leaves Infected wounds
- **Bakuro** *Accacia farnesiana* Shrub Fruits Infected wounds
- **Dik fuik** *Erythrina variagata* Tree Stem bark Fever
- **Faei matametan** *Adenanthera microperma* Tree Seeds Green snake venom antidote
- **Sakiki mean** *Flemingia strobilifera* Herb Roots Bloody urine, lumbugo, earache
- **Kakaut** *Mimosa invisa* Shrub Leaves Cold sweat and panting breath

### HYDROCARCYTACEAE
- **Ai bilan bala** *Ottelia alismoides* Herb Roots Inflammation of the gums
- **LABIATAE**
  - **Silasi fuik** *Basilicum polystachyon* Herb Leaves Clean breast milk
  - **Aibada** *Ocimum sanctum* Herb Leaves Bloated, excessive fart

### LAURACEAE
- **Tahih mulalek** *Cassiya filiformis* Liana Whole plant Jaundice, hemorrhoids

### LENTIBULACEAE
- **Samodo daren** *Utricularia aurea* Liana Roots *Beriberi*, heart problem, lumbugo
- **LILIACEAE**
  - **Lisamaromak** *Eleutherina americana* Herb Clove Cough with blood, bloody urine, dysentery

### LOGANIAECEAE
- **Bakumoru** *Strochnis ligustrina* Tree Stem bark, heart wood Cleaning up dirty blood after childbirth

### LORANTHACEAE
- **Kenluhu, ai hun lalek** *Scirrula atropurpurea* Parasite Leaves Headache, scabies, hemorrhoids

### LYTHACEAE
- **Ai samodo fuik** *Amannia microcarpa* Herb Roots Cough
- **MALVACEAE**
  - **Fau** *Hibiscus tilaceus* Tree Leaves Clean breast milk, fever
  - **Kakuit lotu** *Urena lobata* Herb Roots Heart problem
- **MELIACEAE**
  - **Samer** *Melia dubia* Tree Stem bark, roots, leaves Asthma, cough with phlegm, wormy, jaundice, lumbugo

### MYRTACEAE
- **Bubur** *Eucaliptus alba* Tree Stem bark Cough, cough with phlegm, stinky nose
- **Koya, koiwa** *Psidium guajava* Tree Leaves Influenza, gonorrhea, dysentery, diarrhea
- **Ai siba, ai we** *Eugenia sp.* Tree Leaves Body weakness, easy to sick

### MORACEAE
- **Saltu** *Ficus callosa* Tree Stem bark Asthma, infected wounds at the heads of the children
- **Hali muti** *Ficus retusa* Tree Roots Heart problem, hemorrhoids
- **Hali mear** *Ficus nitentifolia* Tree Roots Chronic fever
- **Mammanus** *Ficus septica* Tree Leaves The fetus bleed out, diarrhea
- **Mahar, knmahar** *Ficus variegate* Tree Stem bark Gonorrhea
- **Kanuku, kenuku** *Ficus pubinervis* Tree Stem bark New wound/injury
<table>
<thead>
<tr>
<th>Family</th>
<th>Common Name</th>
<th>Type</th>
<th>Part Used</th>
<th>Medical Uses</th>
</tr>
</thead>
<tbody>
<tr>
<td>MUSACEAE</td>
<td>Hudi faufuk</td>
<td>Herb</td>
<td>Stem</td>
<td>Beriberi, fever, new injury</td>
</tr>
<tr>
<td>ORCHIDACEAE</td>
<td>Orchid</td>
<td>Orchid</td>
<td>Leaves</td>
<td>Fever in children</td>
</tr>
<tr>
<td>OXALIDACEAE</td>
<td>Moat nu</td>
<td>Herb</td>
<td>Leaves</td>
<td>Fever in children</td>
</tr>
<tr>
<td>PANDANACEAE</td>
<td>Pandanus tectorius</td>
<td>Tree</td>
<td>Roots</td>
<td>Horak hudi dubu (female genital disease, swelling and injury at the clitoris)</td>
</tr>
<tr>
<td>PAPILIONACEAE</td>
<td>Tuhak rekas</td>
<td>Liana</td>
<td>Stem bark</td>
<td>Itchy, scabies</td>
</tr>
<tr>
<td>Kade</td>
<td>Mucuna pruriens</td>
<td>Liana</td>
<td>Stem, sap</td>
<td>New wound</td>
</tr>
<tr>
<td>Aibua</td>
<td>Desmodium tricusentrum</td>
<td>Herb</td>
<td>Leaves</td>
<td>Back pain/lumbago</td>
</tr>
<tr>
<td>PEPERACEAE</td>
<td>Fuik meateian</td>
<td>Piper sp.</td>
<td>Leaves</td>
<td>Cramps and stiffness</td>
</tr>
<tr>
<td>Kunus alete</td>
<td>Piper cubeba</td>
<td>Liana</td>
<td>Leaves</td>
<td>Itchy, sore eyes, diarrhea, rheumatism</td>
</tr>
<tr>
<td>PITTOSPORACEAE</td>
<td>Pittosporum timorense</td>
<td>Tree</td>
<td>Stem bark</td>
<td>Beriberi, diarrhea</td>
</tr>
<tr>
<td>PHIACEAE</td>
<td>Au fui</td>
<td>Shrub</td>
<td>Roots</td>
<td>Wormy</td>
</tr>
<tr>
<td>Fafenok</td>
<td>Saccharum spontaneum</td>
<td>Herb</td>
<td>Leaves, roots</td>
<td>Itchy, diarrhea</td>
</tr>
<tr>
<td>Lanok</td>
<td>Thymelia gigantea</td>
<td>Herb</td>
<td>Roots</td>
<td>Insane</td>
</tr>
<tr>
<td>Ikiri</td>
<td>Andropogon aciculatus</td>
<td>Herb</td>
<td>Roots</td>
<td>Centipedes- and millipedes- antidote</td>
</tr>
<tr>
<td>Hae manlain</td>
<td>Imperata cylindrica</td>
<td>Herb</td>
<td>Roots</td>
<td>Back pain/lumbago</td>
</tr>
<tr>
<td>POLYPODIACEAE</td>
<td>Manlaras</td>
<td>Drymaria quercifolia</td>
<td>Leaves, stem</td>
<td>Asthma, fever</td>
</tr>
<tr>
<td>Bibirusan dikur</td>
<td>Platycladium bifurcatum</td>
<td>Fern</td>
<td>Leaves</td>
<td>Fever with headache</td>
</tr>
<tr>
<td>PONTEDIERACEAE</td>
<td>Talas ran</td>
<td>Herb</td>
<td>Stem</td>
<td>Inflammation of the gums</td>
</tr>
<tr>
<td>RHAMNACEAE</td>
<td>Fahun</td>
<td>Zygophyus timorensis</td>
<td>Tree</td>
<td>Cough, cough with phlegm</td>
</tr>
<tr>
<td>Ai tabako</td>
<td>Pithecellobium junghuhnianum</td>
<td>Tree</td>
<td>Stem bark</td>
<td>Stinky nose</td>
</tr>
<tr>
<td>Kakehi, kabuka</td>
<td>Zygophyus mauritianus</td>
<td>Tree</td>
<td>Leaves</td>
<td>Heart problem</td>
</tr>
<tr>
<td>RIZOPHORACEAE</td>
<td>Leon, fuk bada</td>
<td>Carallia brachiata</td>
<td>Tree</td>
<td>Fever accompanied by cough</td>
</tr>
<tr>
<td>Rosaceae</td>
<td>Bakat</td>
<td>Rhizophora stylosa</td>
<td>Tree</td>
<td>Horak hudi dubu</td>
</tr>
<tr>
<td>Rubaceae</td>
<td>Ai besi kulit maar</td>
<td>Parinarium corymbosum</td>
<td>Tree</td>
<td>New wound</td>
</tr>
<tr>
<td>Rubiaceae</td>
<td>Kabanase</td>
<td>Wendlandia barkilli</td>
<td>Tree</td>
<td>Thin body after childbirth, cough with blood, fever, irregular menstruation, heart problem</td>
</tr>
<tr>
<td>Kafiri modok</td>
<td>Nauclea orientalis</td>
<td>Tree</td>
<td>Stem bark</td>
<td>Cough with phlegem</td>
</tr>
<tr>
<td>Katimun, ketimun</td>
<td>Guettarda speciosa</td>
<td>Tree</td>
<td>Leaves</td>
<td>Nosebleed, green snake venom antidote</td>
</tr>
<tr>
<td>Kafiru mutin, kafiru manen</td>
<td>Nauclea orientalis</td>
<td>Tree</td>
<td>Stem bark</td>
<td>The fetus bleed out, bleeding childbirth, excessive menstruation</td>
</tr>
<tr>
<td>Rutaceae</td>
<td>Asderok, aslerok</td>
<td>Aegle marmelos</td>
<td>Shrub</td>
<td>Old- or infected wounds</td>
</tr>
<tr>
<td>Dilafatu</td>
<td>Atalanta trimera</td>
<td>Shrub</td>
<td>Leaves</td>
<td>Horak abano (female genital disease, women genital disease, swelling and injury at the labia mayora), green snake venom antidote</td>
</tr>
<tr>
<td>Samydaceae</td>
<td>Ai batar, abatar</td>
<td>Caseria moluccana</td>
<td>Tree</td>
<td>Heart problem</td>
</tr>
<tr>
<td>SANTALACEAE</td>
<td>Kamenlin</td>
<td>Santalum album</td>
<td>Tree</td>
<td>Postpartum care, itchy, new wounds, earache</td>
</tr>
<tr>
<td>Sapindaceae</td>
<td>Sukabi</td>
<td>Schleichera oleosa</td>
<td>Tree</td>
<td>Itchy, constipation, bleeding childbirth, excessive menstruation</td>
</tr>
<tr>
<td>SOLANACEAE</td>
<td>Kaut matakmodok</td>
<td>Solanum melongena</td>
<td>Herb</td>
<td>Toothache, ringworm, jaundice</td>
</tr>
<tr>
<td>Babotek</td>
<td>Datura metelo</td>
<td>Shrub</td>
<td>Fruits</td>
<td>Horak abano, scabies</td>
</tr>
<tr>
<td>Kaut alas</td>
<td>Solanum torvum</td>
<td>Herb</td>
<td>Fruits</td>
<td>Scabies</td>
</tr>
<tr>
<td>Kaut hibisusun</td>
<td>Solanum mammosum</td>
<td>Herb</td>
<td>Roots</td>
<td>Rheumatism</td>
</tr>
<tr>
<td>STERCULIACEAE</td>
<td>Te dulis</td>
<td>Helicteres isora</td>
<td>Shrub</td>
<td>Cough, cough with blood, goiter, diarrhea</td>
</tr>
</tbody>
</table>
TILLIACEAE

<table>
<thead>
<tr>
<th>Genus</th>
<th>Species</th>
<th>Type</th>
<th>Common Name (Indonesian)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ai niitaen</td>
<td>Sterculia arceolata</td>
<td>Tree</td>
<td>Body weakness, easy to sick</td>
</tr>
<tr>
<td>Aibano, abano, nitas</td>
<td>Sterculia foetida</td>
<td>Tree</td>
<td>Horak abano</td>
</tr>
<tr>
<td>Paulon</td>
<td>Kleinovia hospital</td>
<td>Tree</td>
<td>Infected wounds at the head of children</td>
</tr>
</tbody>
</table>

ULMACEAE

<table>
<thead>
<tr>
<th>Genus</th>
<th>Species</th>
<th>Type</th>
<th>Common Name (Indonesian)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Krau matan</td>
<td>Elaeocarpus petiolaris</td>
<td>Tree</td>
<td>Leaves</td>
</tr>
<tr>
<td>Ai besi kulit nias</td>
<td>Celtis wightii</td>
<td>Tree</td>
<td>Old- or infected wounds</td>
</tr>
<tr>
<td>Ai te</td>
<td>Celtis sinamomea</td>
<td>Tree</td>
<td>Heart wood</td>
</tr>
</tbody>
</table>

UMBELIFERAE

<table>
<thead>
<tr>
<th>Genus</th>
<th>Species</th>
<th>Type</th>
<th>Common Name (Indonesian)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aimata ust</td>
<td>Centella asiatica</td>
<td>Liana</td>
<td>Wound at the breast</td>
</tr>
</tbody>
</table>

VERBENACEAE

<table>
<thead>
<tr>
<th>Genus</th>
<th>Species</th>
<th>Type</th>
<th>Common Name (Indonesian)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fulis manen</td>
<td>Vitex pubescens</td>
<td>Tree</td>
<td>Itchy</td>
</tr>
<tr>
<td>Ai sui sa, tateka</td>
<td>Lantana camara</td>
<td>Shrub</td>
<td>New wound/injury</td>
</tr>
</tbody>
</table>

VITACEAE

<table>
<thead>
<tr>
<th>Genus</th>
<th>Species</th>
<th>Type</th>
<th>Common Name (Indonesian)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ai darek losuk</td>
<td>Leea acquta</td>
<td>Herb</td>
<td>Swelling and injuries to the thigh slit</td>
</tr>
</tbody>
</table>

ZINGIBERACEAE

<table>
<thead>
<tr>
<th>Genus</th>
<th>Species</th>
<th>Type</th>
<th>Common Name (Indonesian)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Kinur</td>
<td>Curcuma domestica</td>
<td>Herb</td>
<td>Sprains, whiplash, burns, scalded, fractures</td>
</tr>
</tbody>
</table>

People of the Tetun ethnic group in West Timor have a local knowledge of traditional medicine to prevent, cure, and maintain many diseases. This local knowledge is developed by their experience in dealing with various diseases and other health problems in their life. The main component of the traditional medicine of the Tetun ethnic is medicinal plants. This is in line with the term "ai tahan" (leaf) or "ai fuan" (fruit/seed) which they use to call "medicine". The choice of using specific plant is based on the past generation’s knowledge.

Medicinal plants are prepared in various formulas (recipes) which are obtained from their native area. Almost all the plants used as the medicine are wild type taken from the nearest forest, except Areca catechu, Piper sp. and Musa sp. The Tetun ethnic people believe that the wild plants are more effective than the cultivated plants [1].

Some medicinal plants used in the traditional medicine of the Tetun ethnic group are also known and used elsewhere in Indonesia. The plants are used to treat some diseases or ailments as other ethnicities do, but there are some differences. The differences are correlated to the people’s experience in dealing with various diseases and other health problems before, and also by the content of the pharmacologically active metabolites of plants which are different resulted from the growth place [28,29].

4. CONCLUSIONS

Tetun ethnic group in West Timor has a local knowledge of traditional medicine that was mainly based on local (native) plants. The traditional medicine culture is performed by traditional healers, but it is on disappearing process as an impact of modernization. It is only orally transmitted rather than written. It is important to record the Tetun traditional medicine to continue its existence.

5. REFERENCES


Environmental Science and Technology
The Use of Natural-Ingredient Medium for Culturing Locally-
Isolated Bacillus sphaericus and Its Toxicity Against Anopheles 
Larvae in Lombok Island, Indonesia

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Santi Pristianingrum3

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Abstract – The aim of the study was to explore toxicity of locally-isolated B. sphaericus cultured in natural-ingredient medium (ie. soybean milk, coconut milk and fish flour) widely found in Lombok Island, Indonesia. The bacteria was then assayed against Anopheles larvae naturally captured from the island. All ingredient were prepared in 30% concentration (v/v or w/v) and used to cultivated B. sphaericus. The cultures were incubated aerobically at 30oC with 180 rpm shaking for 72 hours. Fish flour medium resulted the highest bacterial concentration, followed by coconut milk medium and soybean milk medium was the lowest. However, the endospore concentration percentage was reached by B. sphaericus cultured on coconut milk medium, followed by soybean medium and fish flour medium. The highest mortality rate was showed by B. sphaericus cultured in fish flour medium, followed coconut milk medium and the lowest showed by soybean medium. The highest price was showed by fish flour medium, followed by soybean milk medium and the lowest price was coconut medium. In conclusion, natural-ingredient medium could be used to grow locally-isolated entomopathogenic B. sphaericus to be used for suppressing Anopheles larvae.

Keyword: Bacillus sphaericus, natural-ingredient medium, Anopheles, larvae and Lombok Island.

1. INTRODUCTION

Malaria is still becoming a health problem in Lombok Island, Indonesia. There are roughly 13,000 people suffering from Malaria in Lombok Island, Indonesia [1]. Malaria was caused by Plasmodium parasite and spread by Anopheles mosquito. The highest prevalence of malaria in Lombok Island are among people inhabit theareanear the beach in the island [2].

The use of biological agent is recommended for larval control in Integrated Mosquito Management. A biological agent such as microbe is considered effective for suppressing mosquito larvae and safe for the environment. One bacterium species popular for combating Anopheles larvae is Bacillus sphaericus. B. sphaericus (now its name becomes Lysinnibacillus sphaericus) is aerobic rod bacteria that capable of producing specific toxins targeting Culex and Anopheles larvae. The toxins are produced when the bacterium enters the sporulating stage. This species is widely used in some countries, along with chemical-based preparations to control mosquito larvae [3].

In this study, we cultured Lombok Island local isolate of B. sphaericus (Isolate Bs-MNT) [4] in 3 natural-ingredient media (soybean, coconut milk, and fish flour) widely found in Lombok Island, Indonesia. The cultured bacterium was tested against naturally-captured Anopheles larvae to observe its toxicity.

2. METHODS

Local isolate of B. sphaericus (Bs-MNT) was refreshed to standard solid medium NYSM consisted of Nutrient Agar, yeast extract, MgCl2, MnCl2 dan CaCl2 [5]. The culture was incubated at 30 oC for 24-48 hours.

The natural ingredient medium preparation was done by mixing soybean milk, coconut milk and fish flour with distilled water (all prepared in 30% v/v or w/v). Two full loops of B. sphaericus isolate was subcultured in the natural-ingredient liquid medium. The cultures were incubated at 30 oC shaking at 180 rpm for 72 hours. After reaching 72 hours, cultures were counted for cell concentration and endospore percentage.

Bioassay/toxicity analysis was carried out based on standard procedure [6]. Lethal concentration (LC) values in 24, 48 and 72 hours were calculated from larval death data using Probit Analysis [7] applying MINITAB V16.
Statistical Analysis Software for Windows [8]. Bioassay was also carried out using B. sphaericus cultured on NYSM medium for comparison. LC value was stated in cell/mL.

3. RESULTS AND DISCUSSION

In this study, we used and compared the potential of soybean milk, coconut milk and fish flour as natural ingredient medium to culture local isolate of B. sphaericus (isolate Bs-MNT) as a replacement for synthetic culture media. Those materials were highly abundant in Lombok Island, Indonesia and very cheap in price.

After 72 incubation in testing medium, B. sphaericus cell concentration and endospore percentage were counted. They are presented in Table 1 as follows.

<table>
<thead>
<tr>
<th>Material</th>
<th>Cell Concentration (cell/mL)</th>
<th>Endospore (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>NYSM (standard)</td>
<td>3.53 x 10⁸</td>
<td>65.4</td>
</tr>
<tr>
<td>Soybean milk</td>
<td>8.90 x 10⁷</td>
<td>48.0</td>
</tr>
<tr>
<td>Coconut milk</td>
<td>6.00 x 10⁸</td>
<td>57.6</td>
</tr>
<tr>
<td>Fish flour</td>
<td>8.90 x 10⁷</td>
<td>33.5</td>
</tr>
</tbody>
</table>

The highest cell count was found in B. sphaericus cultured in fish flour medium, followed by coconut milk and soybean milk respectively. However, the highest endospore concentration was showed by B. sphaericus cultured in coconut milk medium, followed by Soybean milk and Fish flour medium, respectively.

The mortality rate of B. sphaericus cultured in the natural-ingredient medium is presented in Figure 1 as follows.

![Figure 1. Mortality rate of B. sphaericus cultured in natural-ingredient medium](image)

(A: Soybean milk; B: Coconut Milk; C: Fish flour; D: NYSM standard medium)

B. sphaericus grew in standard NYSM medium (Fig. 1D) showed maximum larval mortality rate of 80% on first 24 hours, then gradually declined until 10⁻⁴ dilution. On 48 and 72 hours, it reached maximum rate (100%). These rates retained through 10⁻⁴ dilution, then declined gradually. B. sphaericus cultured in natural ingredient material that showed 24-hour larval killing were fish flour and coconut milk medium (85% and 75%). Soybean milk medium (Fig 1A) did not show larval killing in the first 24 hours. These values were gradually declined until 10⁻⁴ dilution. On 48 and 72-hour observation fish flour medium (Fig 1C), coconut milk medium (Fig 2B) and soybean milk medium reached their maximum values of 97-100%. These values were declined gradually until 10⁻⁴ dilution.

Bioassay of B. sphaericus against Anopheles larvae in 3 natural-ingredient was presented in Table 2 as follows.
Table 2. Lethal concentration values of *B. sphaericus* cultured in natural-ingredient medium

<table>
<thead>
<tr>
<th>Medium</th>
<th>LC50-24h</th>
<th>LC90-24h</th>
<th>LC50-48h</th>
<th>LC90-48h</th>
<th>LC50-72h</th>
<th>LC90-72h</th>
</tr>
</thead>
<tbody>
<tr>
<td>NYSM (Standard)</td>
<td>1.20 x 10^3</td>
<td>4.49 x 10^7</td>
<td>9.97 x 10^3</td>
<td>3.78 x 10^3</td>
<td>5.65 x 10^2</td>
<td>2.51 x 10^3</td>
</tr>
<tr>
<td>Soybean milk</td>
<td>-</td>
<td>-</td>
<td>1.02 x 10^6</td>
<td>5.24 x 10^6</td>
<td>3.50 x 10^3</td>
<td>6.71 x 10^4</td>
</tr>
<tr>
<td>Coconut milk</td>
<td>1.25 x 10^7</td>
<td>4.22 x 10^7</td>
<td>8.21 x 10^7</td>
<td>2.91 x 10^4</td>
<td>4.43 x 10^3</td>
<td>2.26 x 10^4</td>
</tr>
<tr>
<td>Fish flour</td>
<td>9.08 x 10^8</td>
<td>3.64 x 10^7</td>
<td>6.28 x 10^8</td>
<td>2.61 x 10^7</td>
<td>9.56 x 10^2</td>
<td>2.73 x 10^3</td>
</tr>
</tbody>
</table>

In the first 24 hour observation, *B. sphaericus* that grown in fish flour medium showed the highest toxicity, followed by *B. sphaericus* grown in coconut milk medium. The toxicity of *B. sphaericus* grown in fish flour medium was higher than that of *B. sphaericus* grown in standard NYSM medium. However, *B. sphaericus* grown in soybean milk failed to kill *Anopheles* larvae tested.

In 48 and 72 hour observation, *B. sphaericus* grown in fish flour medium showed about 10^3 increase in its LC values and *B. sphaericus* grew in coconut milk medium showed about 10^2-10^4 increase in its LC values as well. However, these LC values increase were lower than that of *B. sphaericus* grown in standard NYSM medium that reached about 10^4-10^5 increase in LC values.

The high toxicity in the first 24 hours showed by *B. sphaericus* grown in Fish flour medium showed the advantage of this medium. Its components seemed readily utilized by *B. sphaericus* to synthesize all toxins needed to kill the larvae. *B. sphaericus* grown in standard NYSM medium reached its toxicity peak at 48-hour observation.

Fish flour medium that contained 65-75% of protein [9] could stimulate the highest concentration in *B. sphaericus* culture (8.90x10^8 cell/mL) compared to another medium (soybean and coconut milk medium). However, this medium only stimulated the lowest endospore percentage (33.5%). This medium also showed the higher toxicity against *Anopheles* larvae compared to another medium. Interestingly, soybean medium that contained 35-56% of protein [10] showed the lowest cell concentration (3.53x10^8 cell/mL) and the lowest toxicity (0% at the first 24-hour testing) compared to another medium. However, coconut milk medium that contained 3.3% of protein [11] showed lower cell concentration after fish flour medium (57.6%) could stimulate higher toxicity against *Anopheles* larvae compared to soybean medium. This indicated that protein was not the only component needed to stimulate high toxicity in *B. sphaericus* if cultured in the natural-ingredient medium. Other components such as minerals and vitamin also play roles in growth and toxin synthesis.

The price comparison is presented in Table 3 as follows.

Table 3. Price comparison among all testing medium and standard NYSM medium

<table>
<thead>
<tr>
<th>Medium</th>
<th>Type</th>
<th>Price/L (in IDR) (Approx)</th>
</tr>
</thead>
<tbody>
<tr>
<td>NYSM (Standard)</td>
<td>Synthetic</td>
<td>150,000 or more</td>
</tr>
<tr>
<td>Soybean milk</td>
<td>Natural</td>
<td>8,000</td>
</tr>
<tr>
<td>Coconut milk</td>
<td>Natural</td>
<td>7,000</td>
</tr>
<tr>
<td>Fish flour</td>
<td>Natural</td>
<td>15,000</td>
</tr>
</tbody>
</table>

The materials chosen in this study were based on their availability and price in Lombok Island, Indonesia. Soybean, coconut, and fish are widely abundant in this island and inexpensive to purchase. Soybean and coconut are planted on almost all area of Lombok Island. Fish flour is made from fish captured from the nearby sea. Coconut milk medium showed the lowest price of all natural-ingredient medium tested. Its price (/L) was so low that reach only about 1/21 of the standard NYSM medium price. It was followed by soybean medium and fish flour medium that was 1/19 and 1/10 of the standard NYSM medium price. Besides the prices, all natural ingredient medium had a similar advantage in producing high toxicity of the testing *B. sphaericus* compared to standard NYSM medium. The low price of the natural-ingredient medium in return can suppress the cost for large scale bio-larvicide production. Hopefully, in the near future, the *B. sphaericus*-based larvicide is affordable for most users in the community and mosquito larval control is no longer problem in developing countries, especially in Indonesia.
4. CONCLUSIONS

In conclusion, the tested natural-ingredient medium (soybean milk, coconut milk, and fish flour) can be applied as candidates for alternative/natural culture medium to grow entomopathogenic B. sphaericus and showed high toxicity culture against Anopheles larvae.

5. REFERENCES

Plant Microbial Fuel Cells (PMFCs): Green Technology for Achieving Sustainable Water and Energy

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* Corresponding authors: bimastyajisurya@gmail.com

Abstract – Sustainability of water and energy is a major problem that is getting more attention because it is very closely related to the human well-being and quality of life. The next few years, Indonesia will get energy and water resources problems. Plant-microbial fuel cells (PMFCs) is one of the alternative green technology that can be used to support the sustainability of other technologies in terms of both energy and clean water production. Bibliometric methods approach used to obtain relevancy and get the latest information of PMFCs. This paper describes the status of energy and water resources, technological developments PMFCs and how PMFCs challenges and development opportunities in Indonesia. This paper reviews the basic understanding of PMFCs development and the way to implement PMFCs in Indonesia.

1. INTRODUCTION

Increasing energy needs and access to clean water is a global issue that is thriving today. Both of these are indicators of human well-being and quality of life. Global energy needs to be increased by 7% annually in line with world population growth. In Indonesia, crude oil and coal diminishing due to over-exploitation in the past. This can lead to a social and economic slump in the various sectors of society. Therefore, the biggest challenge for Indonesia is how to manage the resources of existing energy alternatives and to create new energy sources to replace petroleum hydrocarbon resources. The use of renewable energy in Indonesia such as biomass (22%), water (2%), natural gas (18%) and geothermal (1%) is still quite low when compared with petroleum (39%) and coal (18%) [1].

Indonesia is a tropical archipelago with abundant water resources. About 6% of the total water resources in the world are in Indonesia. Even so, access to clean water in several places in Indonesia is still low. This occurs because (1) water is abundant but can not be consumed, (2) drought due to extreme weather and climate, and (3) water sources that are difficult to access. Among the five major islands (Sumatra, Java, Sulawesi, Bali and NTT) in Indonesia, only Sumatra island that has surplus water. This raises the competition and rivalry among communities. Water quality degradation can occur as a result of their domestic activities, agriculture and mining, deforestation and industrial activity [2]. One solution to this problem is how to increase the efficiency and effectiveness of water treatment technologies.

Plant Microbial Fuel Cells (PMFCs) or also known as plant-assisted sMFCs; Plant power®; and MFCs Constructed Wetland (CW-MFCs) have attracted worldwide attention because it can generate electricity directly from the aquatic plant [3,4]. Those acronyms can be stated as part of PMFCs or PMFCs in advanced. Energy conversion using microorganisms has several advantages such as environmentally friendly, sustainable and able to work in a large range of concentration and a wide variety of substrate. PMFCs are known to generate electricity directly and through constant electron capture resulting from organic compounds conversion. The small efficiency of electricity production Sediment MFCs (SMFCs) led the investigators to seek other alternatives to improve it. PMFCs is known to have electricity production 18 times greater than conventional SMFCs. PMFCs able to improve the water quality due to microorganism metabolism and plant absorption. PMFCs is known to be able to treat some wastewater such as textile waste, domestic, farm, ranch, and tapioca. Some options such as membrane bioreactor technology reclamation, wetland, membrane filtration, and lagoon pools require investment costs and vast land. PMFCs may be one option to compete with those technologies [3,5]. PMFCs's potential to produce clean energy and sustainable water becomes the main subject in this paper. A critical review of some research that has been done as well as deep analysis against PMFCs technology is expected to be the first step in the development of PMFCs in Indonesia. PMFC has a lot of potentials to be developed, so it is worthy to be the object for further studies.

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2. PLANT MICROBIAL FUEL CELLS (PMFCs)

2.1 Basic Concept of PMFCs

Plant-microbial fuel cells are newly emerging technology which generates solar energy as electricity from living plants. In this system, electricity generation achieved by the combination of electricity generation by bacteria through oxidation of organic compounds and deposition of organic compounds by plants into the rhizosphere. The configuration of a PMFCs is shown in Figure 1. To date, there is four basic design of PMFCs, namely the sediment type microbial fuel cell [6], the plant microbial fuel cell [7], the flat-plate plant microbial fuel cell [8], and tubular plant microbial fuel cell [9].

Plant Microbial Fuel Cells (PMFCs) is an alternative technology that converts complex organic compounds into simpler ones using living plants and microorganisms consortium that can be found in roots of plants and soil. In principle, living plant performs photosynthesis process which produces organic compounds such as glucose and sucrose to grow and multiply. Most of the organic compounds are then discharged into the soil through the roots. Microorganisms around the roots will naturally convert organic compounds to produce energy. The excesses of the process are the electrons released into the environment. Some microorganisms such as Geobacter sp., Rhodobacter sp. and Shewanella sp. is able to transfer electrons out of his body (exoelectrogens). Electron transfer serves to maintain the stability of biochemical processes and gain energy from the difference in the number of electrons on the inside and outside of the cell (diffusion). The free electrons are captured by the electrode then causing direct electric current [3,4].

![Figure 1 Rice Paddy Field MFC (Left) and Plant sMFC (Right)](image)

Description of the reaction that occurs in PMFC process:

a. Photosynthesis
   \[
   \text{CO}_2 + 6\text{H}_2\text{O} \rightarrow \text{C}_6\text{H}_{12}\text{O}_6 + 6\text{O}_2 \]

b. Transport of organic compounds in the anode chamber

c. Oxidation of organic compounds by microorganisms in the anode chamber
   \[
   \text{C}_6\text{H}_{12}\text{O}_6 + 6\text{H}_2\text{O} \rightarrow 6\text{HCO}_3^- + 30\text{H}^+ + 24e^- \]

d. Reduction in cathode chamber
   \[
   6\text{O}_2 + 24\text{H}^+ + 24e^- \rightarrow 12\text{H}_2\text{O} \]

2.2 PMFCs as Energy Sources

The first publication which is shown that PMFCs potentially to use as an energy source was demonstrated by Strik et al. (2008). In this paper reported that under Western European climate conditions, the theoretical maximum of power generation of PMFCs was 21 GJ/ha/year (5,800 kWh/ha/year). Further, Deng et al. (2012) stated that the prospective annual gross PMFC outputs were approximately around 1,266–1,638 kWh/ha depends on the types of plants. However, to use a PMFC as a renewable energy source, the power density is a key performance parameter. The newly report from Strik et al. (2011) estimated that the ultimate power density from a PMFC was 1.6-3.2 W/m² plant growth area (PGA) [7,10,11]. This value is comparable to conventional renewable bio-energy source with crop harvesting, which achieves net power density up to 220 mW/m² PGA [12].

<table>
<thead>
<tr>
<th>References</th>
<th>Plants</th>
<th>Electron donor</th>
<th>Power Density (mW/m²)*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Strik et al., 2008</td>
<td>Glyceria maxima</td>
<td>Rhizodeposits</td>
<td>67</td>
</tr>
<tr>
<td>De Schamphelaire et al., 2008</td>
<td>Oryza sativa ssp. Indica</td>
<td>Rhizodeposits</td>
<td>33</td>
</tr>
<tr>
<td>Timmers et al., 2010</td>
<td>Spartina anglica</td>
<td>Rhizodeposits</td>
<td>79</td>
</tr>
<tr>
<td>Timmers et al., 2010</td>
<td>Spartina anglica</td>
<td>Rhizodeposits</td>
<td>100</td>
</tr>
<tr>
<td>Helder et al., 2012</td>
<td>Spartina anglica</td>
<td>Rhizodeposits</td>
<td>220</td>
</tr>
<tr>
<td>Helder et al., 2015</td>
<td>Spartina anglica</td>
<td>Rhizodeposits</td>
<td>155</td>
</tr>
<tr>
<td>Wetser et al., 2015</td>
<td>Spartina anglica and Phragmites australis</td>
<td>Rhizodeposits</td>
<td>1.3-18</td>
</tr>
</tbody>
</table>

*plant growth area (PGA)
In practice, Table 1 provided increasing trend of PMFCs power density reported in the literature from 67 mW/m² in 2008 to up to 240 mW/m² in 2015, from which one can expect a promising prospect for the use of PMFC as a practical energy in the near future. The highest power density achieved was 240 mW/m², obtained with PMFC using *Spartina anglica* salt marshes. Even though the power density is still lower than what was earlier estimated, it is potential to improve the PMFCs performance still wide open. For example, recently in the field application, PMFC were commercially applied at 100 m² PGA scale to power LEDs [17].

### 2.3 PMFCs as Wastewater Treatment Units

The study on PMFCs as wastewater treatment units is still limited due to this technology is still emerging. Table 2 shows a performance of PMFCs on different types of wastewater in current studies. The first study of PMFCs as wastewater treatment unit was conducted by Yadav et al. (2012). In this paper, the constructed wetland combined with MFC was able to degrade the methylene blue dye up to 75% based on COD removal [18]. The same trend on COD removal efficiency was achieved when Zhao et al. (2013) and Doherty et al. (2015) treating swine wastewater in the PMFC reactor [19,20]. Great results on removing Azo dye from wastewater were also shown by Fang et al. (2013) and Fang et al. (2015) [21,22]. Further, the outstanding results were achieved when a PMFCs applied to treat the domestic wastewater (90-95% of COD removal) as shown by Villasenor et al. (2013) and Liu et al. (2014) [23,24,25]. The very recent research also shown that PMFCs was able to remove the heavy metal (Cr (VI)) from the wastewater up to 99% [25]. PMFCs is able to reduce COD with an efficiency of more than 100% (using 1-day aeration addition) with an initial concentration of 314,8 mg / l. PMFCs also can lower the concentration of ammonium and substrate by 97% in anaerobic conditions and 40% under aerobic conditions [26]. Another study conducted by Srivastava et al., mentioned that PMFCs has the efficiency to over 90% for glucose substrate and substrate initial input of 0.5 g/l. PMFCs efficiency ranges vary depending on the system, the substrate and the material used. With COD input range 180 - 1500 mg / l, may be set aside PMFC 64-100% COD [27]. Since the PMFCs have a possibility to be applied in the broad range of wastewater, it seems that this technology is very promising to be used as wastewater treatment units in the near future.

<table>
<thead>
<tr>
<th>References</th>
<th>Type of wastewater/ Pollutants</th>
<th>Initial COD (mg/l)</th>
<th>COD removal efficiency (%)</th>
<th>Maximum power density (mW/m²)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Yadav et al., 2012</td>
<td>Methylene blue dye</td>
<td>500 – 1500</td>
<td>75</td>
<td>7.44</td>
</tr>
<tr>
<td>Fang et al., 2013</td>
<td>Azo dye</td>
<td>150 and 180</td>
<td>85.7</td>
<td>5.62</td>
</tr>
<tr>
<td>Villasenor et al., 2013</td>
<td>Synthetic domestic wastewater</td>
<td>13,9; 31,1; 61,1 / g/m²</td>
<td>90.0–95.0</td>
<td>20.76</td>
</tr>
<tr>
<td>Zhao et al., 2013</td>
<td>Swine wastewater</td>
<td>1058.45 ± 420.89</td>
<td>76.5</td>
<td>12.37</td>
</tr>
<tr>
<td>Liu et al., 2014</td>
<td>Synthetic domestic wastewater</td>
<td>50 – 1000</td>
<td>95</td>
<td>44.63</td>
</tr>
<tr>
<td>Doherty et al., 2015</td>
<td>Swine slurry</td>
<td>411 – 854</td>
<td>64</td>
<td>10.51</td>
</tr>
<tr>
<td>Fang et al., 2015</td>
<td>Azo dye</td>
<td>135 ± 10</td>
<td>85.7</td>
<td>1.84</td>
</tr>
<tr>
<td>Oon et al, 2015</td>
<td>Synthetic wastewater</td>
<td>314,8</td>
<td>40 – 90</td>
<td>6.12</td>
</tr>
<tr>
<td>Srivastava et al, 2015</td>
<td>Synthetic wastewater</td>
<td>180 – 1500</td>
<td>&gt; 90</td>
<td>320,8</td>
</tr>
<tr>
<td>Habibul et al., 2016</td>
<td>Chromium (VI)</td>
<td>9,5 and 19</td>
<td>99*</td>
<td>-</td>
</tr>
</tbody>
</table>

*a based on Cr (VI) removal efficiency

### 3. CHALLENGES AND FUTURE PERSPECTIVE

Currently, PMFCs still in the stage of product development and in-depth research related to the upscaling process. In Netherlands, PMFCs used in conjunction with a green roof because it can restrain the rate of flow of rain water, add more aesthetic value, improve biodiversity and air quality as well as keep the indoor temperature remains stable. It is estimated, electricity production was 3.2 W / m² so as to reduce dependence on electricity consumption of fossil energy. PMFCs can also be integrated with a particular wetland e.g., constructed wetland, fields, places of recreation, and pool. Additionally, PMFCs can reduce greenhouse gas emissions such as CO₂ and CH₄ thus potentially to be developed in countries such as Indonesia [26,27]. In connection with the issue of clean water, PMFCs can be a solution for monitoring water quality in remote areas. PMFCs can be used as an energy source of in situ measurement tools which it capable of providing a stable electricity all the time. The challenges of implementation of PMFCs as a biosensor is a convenience, price, and capability of use in the field. Studies related to the efficiency of the system is still being developed to get the lowest price and eligible to be traded [28].

PMFCs still has a variety of limitations, mainly from the electrical output generated. Factors that affect the electrical output and efficiency of wastewater treatment is organic loading, redox conditions, roots/wetland plants, microorganisms around rhizosphere, configurations, and electrode material. The kind of organic compound affects the efficiency of the substrate which is converted by microorganisms into electricity. The
larger the COD, the greater the microorganisms produce electrons. Even so, there is an optimal COD must be determined in order to obtain optimum processing results anyway. Redox conditions are affecting the voltage gradient that exists between the two compartments (anode-cathode). This occurs due to the difference in oxygen concentration between the two compartments. By keeping the DO, the optimal redox conditions can be maintained. Another factor to note is the importance of the wetland plants. Wetland plants should have a good root system, because of the more the roots, the better the efficiency at the cathode. Root is the place to supply oxygen and organic compounds produced by plants. Microorganisms greatly affect the overall process. Symbiotic mutualism of consortium microorganisms in PMFCs system needs to be maintained and controlled. Reactor configuration influence on the process, the greater the distance between the electrodes, the greater the obstacles that occur in the electrical circuit. In addition, the rate of water flow and the electrode material is also capable of affecting the efficiency of electricity generation [3,4].

Research on PMFC in Indonesia is still quite rare. So, there is a chance for finding the optimal configuration and adapting this technology to Indonesia’s environment. PMFCs can not work alone as a treatment of wastewater and require integration / hybrid from a wide range of other technologies to enhance efficiency. MFCs constituent materials are still quite expensive and hard to find in Indonesia such as proton exchange membrane and cation exchange membrane. Stability and long-term performance of the MFC system are also an important issue that needs to be studied further. PMFCs fouling problems in the media also need to be reviewed in order to obtain optimal conditions for the process PMFCs. Interdisciplinary cooperation is needed to get answers to the problems of energy and water supply in the future.

4. CONCLUSIONS

This paper describes the status of water and energy in Indonesia and serves PMFCs alternative technology as a solution to the energy crisis and water. PMFCs is a converter of organic compounds directly into electricity at the same time lowering the concentration of pollutants in wastewater. PMFCs challenge ahead is how to improve process efficiency and reduce investment costs to be incurred and other limitations. The need for collaboration and interdisciplinary research in order PMFCs can be applied in Indonesia as one of the alternative producers of renewable energy technologies.

5. REFERENCES

Travel Time Difference between Estimated and Observed Values of the 2011 Trans-Oceanic Tohoku Tsunami

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²Center for Earth Science Studies, Department of Physics, Universitas Negeri Surabaya, Surabaya 60231, Indonesia
*latifah.cholifah@gmail.com

Abstract – Difference in travel times between estimates from numerically simulated tsunami waves and records from field observations is vital for predicting tsunami arrival times at shorelines hence minimizing disaster risks. In this study, we provide estimated travel times of the 2011 Tohoku tsunami and compare them with observed travel time records provided by NOAA for a number of DART stations. We then find a systematic increase in the calculated travel time difference, termed as travel time delay, with increasing epicentral distance. The linear dependence of travel time delay upon epicentral distance suggests that the trans-oceanic tsunami wave in nature encountered geophysical disturbances, mainly due to the elastic effects of the solid Earth on bottom topography. This source of disturbance contributes to tsunami speed variations, leading to a time delay of up to 17 minutes for the wave to reach the first peak after the origin time of distant observations.

1. INTRODUCTION

The travel time of tsunami wave propagation from a tsunami source to a place where a station is located is one of important tsunami parameters. In the context of hazard mitigation, knowledge of accurate tsunami travel times is required to accurately predict arrival times of a tsunami at shorelines that is useful for minimizing disaster risks. Previous work [2, 3, 4, 6] have confirmed that gigantic, trans-oceanic tsunamis propagating across the Pacific with a travel distance of more than thousands of kilometers away from the epicenter revealed systemic differences in tsunami travel times between simulated and observed waves. The differences were then arguably attributed to tsunami speed variations in nature while advancing over large distances compared with the non-dispersive speed of a tsunami predicted by the long, surface gravity wave approximation, as addressed in [2, 3, 5]. In this study, we extract estimates of travel times from tsunami waveforms discussed in relevant work [6] and compare them with those obtained from field records. In turn, we examine travel time delay in place of the calculated travel time difference in relation to epicentral distance.

Here we provide basic assumptions commonly made for the case of shallow-water, surface gravity waves in numerically modeled tsunamis that include a homogeneous fluid of seawater with no density stratification and flat-bottomed sea floor. Such assumptions lead to the theoretical, non-dispersive speed $c$ of a propagating tsunami as follows

$$c = (gH)^{1/2}$$

(1)

where $g$ denotes the gravitational acceleration and $H$ is the ocean depth. For a typical ocean depth of $H \sim 5$ km, the tsunami speed is about 720 km/h. However, as discussed in [2] the propagating speed is limited by a factor originated from loading effects of seafloor deformation on the bottom topography as given by

$$c = (gH)^{1/2} (1 - \beta)^{1/2}$$

(2)

where $\beta$ is the factor representing such effects that result in the speed reduction relative to (1) by as much as 1%. On the basis of (2), we argue that tsunami speed variations occur while propagating over large distances across the ocean, as also suggested by [3, 5]. These variations in speed lead to apparent tsunami travel time differences between values estimated from numerical simulations and those obtained from direct observations [4, 6].

2. METHODS

For the case of the 2011 Tohoku tsunami examined in this study, we have utilized a number of numerical data providing travel time datasets visually derived from the first peak of simulated tsunami waveforms recorded at various geographical locations of the Deep-ocean Assessment Reports of Tsunamis (DART) stations discussed by [6]. These datasets represent estimates of tsunami travel times, and in comparison with the observed values of
travel times taken from either the National Oceanic and Atmospheric Administration (NOAA) or similar stations in [6], the data sets allowed us to calculate travel time differences simply. The differences in travel times were tabulated and classified into three regions of observations, depending upon a particular DART location measured from the source, namely near-field, intermediate-range, and distant measurements. We argue here that the data from near-field measurements were values provided by DART codes positioned less than 3,000 km away from the epicenter. Intermediate observations were then obtained from DART stations located between 3,000 km and 12,000 km, comprising the majority of the field data used in the current study. Monitoring of far-field regions for this case was provided by DART stations located more than 12,000 km away. All the differences were termed as tsunami travel time delay plotted against epicentral distance to see systematic delays, as the wave propagates away from the source to large travel distances across the Pacific. The results are discussed within the context of tsunami speed variations as predicted in (2) by [2] and also addressed by [3, 5], as well as reported by [4, 6] as the apparent travel time delay for distant tsunamis.

3. RESULTS AND DISCUSSIONS

As previously stated, Table 1 describes a total of 13 DART stations located at different geographical positions used in this study, covering three regions of field observations with increasing epicentral distances and providing a list of corresponding estimated and observed travel times. The difference in travel times was then defined as travel time delay, measured in minutes.

Table 1. The data for the 2011 Tohoku event with travel time delay is defined as the travel time difference.

<table>
<thead>
<tr>
<th>DART Code</th>
<th>Observational Location</th>
<th>Latitude</th>
<th>Longitude</th>
<th>Epicentral Distance (km)</th>
<th>Estimated Travel Time (h)</th>
<th>Observed Travel Time (h)</th>
<th>Travel Time Delay (min)</th>
</tr>
</thead>
<tbody>
<tr>
<td>21413</td>
<td>near-field</td>
<td>30.51° N</td>
<td>152.12° E</td>
<td>1,246</td>
<td>1.29</td>
<td>1.30</td>
<td>0.6</td>
</tr>
<tr>
<td>21415</td>
<td>near-field</td>
<td>50.18° N</td>
<td>171.85° E</td>
<td>2,670</td>
<td>3.14</td>
<td>3.18</td>
<td>2.4</td>
</tr>
<tr>
<td>52402</td>
<td>intermediate</td>
<td>11.88° N</td>
<td>154.12° E</td>
<td>3,165</td>
<td>3.71</td>
<td>3.76</td>
<td>3.0</td>
</tr>
<tr>
<td>52403</td>
<td>intermediate</td>
<td>4.05° N</td>
<td>145.59° E</td>
<td>3,828</td>
<td>4.91</td>
<td>4.97</td>
<td>3.6</td>
</tr>
<tr>
<td>46409</td>
<td>intermediate</td>
<td>55.30° N</td>
<td>211.48° E</td>
<td>5,344</td>
<td>6.71</td>
<td>6.78</td>
<td>4.2</td>
</tr>
<tr>
<td>52406</td>
<td>intermediate</td>
<td>5.29° S</td>
<td>165.00° E</td>
<td>5,388</td>
<td>6.71</td>
<td>6.80</td>
<td>5.4</td>
</tr>
<tr>
<td>51407</td>
<td>intermediate</td>
<td>19.59° N</td>
<td>203.41° E</td>
<td>6,183</td>
<td>7.63</td>
<td>7.74</td>
<td>6.6</td>
</tr>
<tr>
<td>51425</td>
<td>intermediate</td>
<td>9.51° S</td>
<td>183.76° E</td>
<td>6,839</td>
<td>8.11</td>
<td>8.23</td>
<td>7.2</td>
</tr>
<tr>
<td>46411</td>
<td>intermediate</td>
<td>39.35° N</td>
<td>232.98° E</td>
<td>7,486</td>
<td>9.23</td>
<td>9.35</td>
<td>7.2</td>
</tr>
<tr>
<td>43412</td>
<td>intermediate</td>
<td>16.07° N</td>
<td>253.00° E</td>
<td>10,619</td>
<td>13.40</td>
<td>13.59</td>
<td>11.4</td>
</tr>
<tr>
<td>51406</td>
<td>intermediate</td>
<td>8.48° S</td>
<td>234.97° E</td>
<td>10,828</td>
<td>13.37</td>
<td>13.59</td>
<td>13.2</td>
</tr>
<tr>
<td>32411</td>
<td>far-field</td>
<td>4.99° N</td>
<td>269.16° E</td>
<td>12,741</td>
<td>16.57</td>
<td>16.82</td>
<td>15.0</td>
</tr>
<tr>
<td>32412</td>
<td>far-field</td>
<td>17.97° S</td>
<td>273.61° E</td>
<td>14,816</td>
<td>18.90</td>
<td>19.18</td>
<td>16.8</td>
</tr>
</tbody>
</table>

The data listed in Table 1 in particular columns providing distances to which the wave travels from the epicenter and time delays with which the simulated wave is predicted to arrive earlier at larger distances indicate a linear dependence of the delays upon epicentral distances. This is not surprising in that in regions far from the tsunami source the observed speed predicted in (2) is slowed down by ~ 1% from its value derived from the long, surface gravity wave approximation due to topographic variation effects along tsunami propagation. We provide a plot of the linear dependence of the time delay on the travel distance in Figure 1 below.

Figure 1. Travel time delay as a function of epicentral distance for the 2011 Tohoku event.
Some interesting features are drawn from careful examinations on the plot. At a near-field region of observation, particularly for travel distances of less than 1,000 km, the time delay is insignificant, meaning that there is no apparent travel time difference between the simulated and observed waves. This is supported by [1] stating that tsunamigenic earthquakes recorded at stations in the radius of less than 1,000 km from the earthquake epicenter are classified into local events. We argue here that this covered travel distance by such events is, to some extent, not much influenced by the geometry of the elastic, solid Earth and hence giving no delayed propagation times. The near-field observations cover a travel distance of up to 3,000 km away from the source and correspondingly take a travel time of up to 4 hours for the trans-oceanic wave with a typical speed of 720 km/h to travel across the ocean with only limited amount of energy loss and with no significant perturbations with respect to changes in its waveforms [4, 5, 6]. For this region, the speed variation is responsible for a slight increase in the time delay of up to 1.0%.

For regions in the range 3,000 – 12,000 km away from the source (intermediate regime – open blue circles in Figure 1), the travel time delay appears in a manner reflecting that the estimated travel times are systematically shortened relative to the observed values. The delays are found to be in the range 3.0 – 15.0 minutes, in good agreement with some of the previous work [4, 6]. As travel distance increases, more geophysical disturbances upon the tsunami wave propagation come into play reducing the speed, relative to (1), to a varying value of 1.3 – 1.6%. This speed reduction is effectively consistent with a maximum value of 1% obtained from numerically predicted speed given in (2) as discussed by [2, 3] and also addressed by [7].

At remote locations where the giant wave travels to a distance of more than 12,000 km away from the source or equivalent to a traveling time of more than 16 hours, a further reduction in speed gives a longer time delay of more than 15.0 minutes, visually seen as two green open circles in Figure 1. However, the longer delays found in these field observations give no significant change in the proportion of speed reduction relatively compared with the wave speeds relevant to the observed travel times. In other words, for the distant measurements as listed in the last two rows of Table 1 a maximum value of the time delay of 16.8 minutes only corresponds to a reduction in the speed of approximately 1.5%, lying in the range of values given in the intermediate regime.

The results for all regions of space for tsunami monitoring considered in this study is a bit surprising in the sense that while the time delay taken up by the computed and observed waves increases the proportion of the reduced speed is relatively fixed with the small reduction only at a value of 1.0 – 1.6%, in good agreement with [2, 3, 7]. It should be noted here that there are at least two logical consequences that can be drawn from these findings. Firstly, it is true to say that tsunami wave speed varies primarily with the external, local parameters, such as bathymetry effects on the topography induced by seafloor deformation [4, 6, 7]. Secondly, the reduction in tsunami speed in correlation with the propagation of a tsunami wave indicates that some amount of energy is lost. The loss of energy needs clarifying regarding calculations of energy dissipation incompressible fluids that go beyond of the present study and therefore this issue is promising for future work.

4. CONCLUSIONS

We have calculated the travel time delay defined here as the difference in travel times between simulations and observations for the 2011 Tohoku, trans-oceanic tsunami. We have divided the Pacific into three zonal divisions, namely near-, intermediate-, and far-field observations at which the time delay is determined. The delay is found to increase linearly with increasing epicentral distance and is then measured to be of up to 17 minutes estimated from the origin time for distant DARTs managed by NOAA. The tsunami speed variation in nature is believed to cause such delays, where the observed speed is reduced by 2% from its value calculated using the shallow-water wave theory. The speed reduction is primarily caused by the Earth elasticity in terms of topographic variations of the ocean.

5. REFERENCES

A Systematic Decrease in Tsunami Amplitude with Epicentral Distance

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Abstract – The dynamics of tsunami wave propagation in the open ocean includes discussion on the behavior of tsunami amplitude while propagating over time and distance. In this study, we use data records of sea surface displacement considered as the wave amplitudes of the 2011 Tohoku tsunami monitored by 14 DART buoys at various positions across the Pacific to examine tsunami energy decay in terms of the decrease in the amplitude with increasing epicentral distance. The data reveal the fact that the amplitude decreases sharply in the near-field observation and decreases gradually while covering a travel distance of up to 15,000 km away from the source. This suggests that there are at least two different mechanisms of energy release in near-field and distant tsunamis but the details of the mechanisms go beyond of the present study. The amplitude attenuation is then discussed in the context of dispersive tsunamis with the importance of frequency-dependent speed in the far-field only.

1. INTRODUCTION

The need for a better understanding of the behavior of tsunami wave energy and amplitude with respect to travel time and distance from a particular tsunami source while a tsunami wave propagates in the open ocean is crucial for the development of the reliable tsunami early warning system and hazard assessment [1, 6]. Prediction of water run-up as the giant wave approaches a coastline requires knowledge of accurate tsunami amplitude measured in the ocean. Although previous work [2] has suggested that a large tsunami could travel great distances across the ocean with only limited amount of energy loss, careful examination and analyses of tsunami waveforms from observations and simulations have clearly indicated a systematic decrease in the maximum wave amplitudes of a propagating tsunami with increasing epicentral distance [3, 5, 7].

In the limit of non-dispersive long wave propagation theory, the phase speed of a tsunami is only determined by the external parameter, namely the ocean depth \( H \). For this approximation; the tsunami speed \( c \) is given by

\[
    c = (gH)^{1/2}
\]

where \( g \) is acceleration due to gravity. Regardless of the Earth’s imperfectly spherical surface, g is considered constant across the globe and hence for a relatively constant depth of the open oceans the tsunami generated by a large earthquake moves at a constant speed with no loss of kinetic energy during its propagation, independent of zonal divisions of observation.

However, from records of travel times, the speed given in (1) remains true for only near-field measurements, and it starts to slightly deviate from the predicted value in (1) for adequately distant tsunami observations [3, 5, 7, 8]. It follows that for global trans-oceanic tsunami propagation there may be energy decay due to dispersion effects. Previous studies [6, 8] have addressed these effects and suggested the significance of frequency-dependent speed for dispersive tsunamis, particularly for far-field observations. As wave energy is proportional to the square of wave amplitude, the energy decay of a tsunami wave can be examined in terms of tsunami amplitude decrease while advancing over large distances in the ocean. In this preliminary study, we show the spatial distribution of the decreased amplitude from the 2011 Tohoku event and discuss a functional relationship that is likely possible for the decrease in the observed amplitudes with respect to travel distances measured from the epicenter.

2. METHOD

We have used a major tsunami event, namely the 2011 Tohoku tsunami as a case study reported in this study. The mega-tsunami event was generated by a large earthquake with a magnitude of \( M_w \) 8.9 and its hypocenter off the northeastern-coast of Japan that occurred on March 11, 2011, at 5:46 UTC, causing a devastating giant wave initially formed only 5 minutes after the main-shocks. The epicenter of the tsunami was at 38.3° N and 142.4° E.
We have then utilized field data records of vertical sea surface displacement of disturbed seawater monitored by 14 Deep-ocean Assessment Reports of Tsunamis (DART) buoys distributed over various geographical positions around the Pacific rim, from nearby the source at 1,246 km away (DART-21413) to a distance of 14,816 km away from the epicenter (DART-32412). The data records in the form of tsunami waveforms from which tsunami amplitudes were estimated are made freely available at [http://www.ngdc.noaa](http://www.ngdc.noaa) managed by the National Geophysical Data Center (NGDC) as part of the National Oceanic and Atmospheric Administration (NOAA). The observed amplitudes given by all the DART buoys are then plotted against travel distances measured from the source to examine the observed variations in tsunami amplitude with increasing epicentral distance.

3. RESULTS AND DISCUSSIONS

Table 1 below describes a total of 14 DART buoys located at different geographical positions across the Pacific used in this study, covering broad regions of tsunami observations from the nearest position, DART-21413, to the far station, DART-32412. A list of the epicentral distance measured in kilometer away from the tsunami source and its corresponding observed amplitude measured in meter from the sea surface is provided.

<table>
<thead>
<tr>
<th>DART Code</th>
<th>Latitude</th>
<th>Longitude</th>
<th>Epicentral Distance (km)</th>
<th>Observed Amplitude (m)</th>
</tr>
</thead>
<tbody>
<tr>
<td>21413</td>
<td>30.51° N</td>
<td>152.12° E</td>
<td>1,246</td>
<td>0.76</td>
</tr>
<tr>
<td>21415</td>
<td>50.18° N</td>
<td>171.85° E</td>
<td>2,670</td>
<td>0.25</td>
</tr>
<tr>
<td>52405</td>
<td>12.88° N</td>
<td>132.33° E</td>
<td>3,001</td>
<td>0.36</td>
</tr>
<tr>
<td>52402</td>
<td>11.88° N</td>
<td>154.12° E</td>
<td>3,165</td>
<td>0.30</td>
</tr>
<tr>
<td>52403</td>
<td>4.05° N</td>
<td>145.59° E</td>
<td>3,828</td>
<td>0.28</td>
</tr>
<tr>
<td>46409</td>
<td>55.30° N</td>
<td>148.51° W</td>
<td>5,344</td>
<td>0.20</td>
</tr>
<tr>
<td>52406</td>
<td>5.29° S</td>
<td>165.00° E</td>
<td>5,388</td>
<td>0.17</td>
</tr>
<tr>
<td>51407</td>
<td>19.59° N</td>
<td>156.58° W</td>
<td>6,183</td>
<td>0.16</td>
</tr>
<tr>
<td>51425</td>
<td>9.51° S</td>
<td>176.24° W</td>
<td>6,839</td>
<td>0.10</td>
</tr>
<tr>
<td>46411</td>
<td>39.35°N</td>
<td>127.02° W</td>
<td>7,486</td>
<td>0.18</td>
</tr>
<tr>
<td>43412</td>
<td>16.07° N</td>
<td>107.00° W</td>
<td>10,619</td>
<td>0.13</td>
</tr>
<tr>
<td>51406</td>
<td>8.48° S</td>
<td>125.03° W</td>
<td>10,828</td>
<td>0.07</td>
</tr>
<tr>
<td>32411</td>
<td>4.99° N</td>
<td>90.84° W</td>
<td>12,741</td>
<td>0.11</td>
</tr>
<tr>
<td>32412</td>
<td>17.98° S</td>
<td>86.39° W</td>
<td>14,816</td>
<td>0.04</td>
</tr>
</tbody>
</table>

We here provide a plot of the tsunami amplitude against the epicentral distance measured from the source in Figure 1 below and discuss consequences from the apparent trend in the plot. We focus on providing arguments based on physical grounds that are sensible for the explanation of the decreased trend in the observed amplitudes with increasing epicentral distances.

![Tsunami Amplitude vs Epicentral Distance](image)

**Figure 1. Tsunami amplitude as a function of epicentral distance for the 2011 Tohoku event.**

The data points in Figure 1 indicate that there are two possible mechanisms of energy release in the context of mechanical energy conservation during the rapid generation and propagation of a tsunami wave in the ocean. The first mechanism is likely to the extent within a few hours (of about 4 hours) calculated from tsunami origin time. This duration is estimated at the time when sea surface elevation was recorded at 0.76 m by DART-21413 at about 1,250 km away from the source to the time when the mean sea level change was measured to be 0.30 m given by
a group of three stations: DART-21415, 52405, 52402, where the average position is about 3,000 km away from the epicenter. This period of time where the energy decay was rapid is considered as the initial stage.

The initial stage of the energy release is arguably linked to mechanical energy available for the tsunami wave generation and is dominated by the gravitational collapse of initial potential energy that is directly converted into kinetic energy during a phase of the tsunami generation. This phase could extend to several hours of propagation and in Figure 1 it takes approximately 4 hours, which is relevant to a travel distance in the near-field observation, from zero points to a distance of approximately 3,000 km away from the source if the tsunami speed is taken to be in the order of 750 km/h.

Following the initial development of the gigantic wave, the second stage occurred during the tsunami passage in the ocean from the time when the propagating wall of seawater reached a quasi-steady state at the upper end of the near-field station (of about 3,000 km away) to more than 14,000 km away from the epicenter, proportional to a total time of approximately 15 hours traveling across the Pacific, spanning over basin-wide. This wider range of observations is named intermediate to distant observations. In this stage, the actual rate of the energy decay and hence the amplitude attenuation is much slower than the previous stage, indicating that there may be the interplay between potential energy change and its corresponding change in kinetic energy. This energy budget, however, is out of the scope of the present study and at present, it is enough to say we leave this challenging issue for future work.

Another interesting argument for the decreased trend in the observed amplitude with travel distance comes from a theoretical point of view. Instead of using the non-dispersive, long surface gravity wave approximation where the speed is predicted in (1), some modelers have used dispersive tsunamis to modeling tsunami propagation. The inclusion of dispersion effects results in different features as these effects play a crucial role in modifying the energy and hence the corresponding amplitude for the distant propagation of global trans-oceanic tsunami [6, 8]. For this approach, the speed of a tsunami wave is written in terms of the dispersion relation as follows,

\[ c^2 = \frac{g}{k} \tanh(kH) \]

where \( k = 2\pi/\lambda \) and \( \lambda \) represents the wavelength of a tsunami wave. Thus, the speed of a propagating tsunami can be different for various wavelengths of dispersive tsunamis. Equation (2) is, therefore, the general form of (1) in the sense that if \( kH \) is extremely less than unity than the shallow-water approximation applies, modifying (2) to be (1). Using the concept of frequency dispersion as a measure of the spread of tsunami energy over time and space along the direction of advancing front owing to different wavelengths, [6] convincingly demonstrated that waveforms developed from simulations had slightly lower amplitudes relatively compared with those obtained from observations in all cases considered. It was found that dispersion effects accumulate in time and space, and thus such effects are less important for near-field observations and become important for distant tsunamis.

While [4] have argued that the tsunami energy density decreased with the vertical length scale, \( i.e., \) the depth of the ocean, the results of the present study clearly show that the open-ocean amplitude is likely to be inversely proportional to the epicentral distance, with a proportionality constant that is not yet clearly explained. Further reduction in the tsunami energy includes other external factors, such as the irregularity of bottom topography and shallow coastal shelves, as discussed by [1].

All the above arguments explaining the decay of the energy and hence the observed amplitude in this study have been an increasing issue in recent years for the development of reliable tsunami prediction off and on shores in the context of reducing hazard risks. Tsunami warnings are important not only for people living in local regions within the perimeter of the near-field measurements but also lives in remote areas within the distant observations. This is in line with the findings of some previous work [1, 6, 8], addressing the significance of dispersion effects on limiting the energy propagation particularly for the far-field only. Therefore, enhanced tsunami prediction of arrival times and tsunami heights using accurate simulations by adding factors, such as the frequency content of tsunami sources and the geometry of subsurface structures in the governing equations describing the generation and propagation of a trans-oceanic tsunami is of fundamental importance for the far-field tsunami measurements.

4. CONCLUSIONS

We have examined tsunami wave energy decay in terms of tsunami wave amplitude attenuation with distance measured from the epicenter. The datasets of the observed amplitudes and the corresponding epicentral distance were provided by 14 DARTs operated by NOAA around the Pacific rim that monitor the 2011 Tohoku tsunami from different zonal divisions of observations. The results indicate that the observed amplitude decreases rapidly during the initial stage of the tsunami generation relevant to the near-field and decreases steadily for the rest of the stage reaching the far-field points of measurements. The role of wave dispersion effects owing to the frequency content of the wave is found to be vital particularly for distant tsunami monitoring.

5. REFERENCES


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The Impact of Cocoa Plantation Management Towards The Diversity of Ground Cover Vegetation and Soil Chemical Properties

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Abstract – Farming management will certainly affect the environmental condition. It can influence the diversity of species in the area and the quality of the soil. Monoculture system in the farming method can affect the loss of species richness and diversity of plant. Soil degradation will increase in the area which is planted with homogenous stands while the diversity of ground cover vegetation is decreasing. However, the information on the diversity of ground cover vegetation and soil chemical properties in the cocoa plantation is still limited. This study aims to determine the diversity of ground cover vegetation and soil chemical properties. The study was conducted at Katibung district, South Lampung Regency. The sampling design to assess the diversity of ground cover vegetation was random using quadrat method by counting the number of species present and the number of individual of each species within the plot. Soil chemical properties were analyzed at the Laboratory of Soil Science, Faculty of Agriculture, University of Lampung. The parameters analyzed include soil pH, organic C, total-N, P2O5 available, CEC, base saturation, Al saturation. Information on the cocoa plantation management obtained through interviews and questionnaires using purposive sampling method. The result shows that most farmers use monoculture practices. Farming maintenance and fertilization are not routinely carried out. Most of the farmers use both chemical fertilizer and organic manure to fertilize the soil. Some of them use only the organic manure or the chemical fertilizer, whereas a few of them do not perform fertilizing to the soil. Soil analysis shows that CEC and organic carbon are at low level. The CEC is low so that the nutrient provided is poor. CEC can be worked up by adding organic matter such as compost or manure. Shannon-Wiener index of ground cover vegetation in the area is 2,21 which means the ground cover vegetation was at a moderate level. The condition was quite stable. For conserving the environment, the diversity of ground cover vegetation should be preserved. Managing the growth of ground cover vegetation is suggested in order to maintain the sustainability of the environment.

Keywords: cocoa, diversity, soil, ground cover vegetation

1. INTRODUCTION

Cocoa is one commodity that is widely grown in Indonesia. Cocoa planting area has reached 1,622,600 ha in the year of 2015 [1]. In South Lampung Regency, cocoa is developed with support from local government facilities. Cocoa plantation area managed by smallholder in South Lampung Regency became 16.084 ha in 2014 [2].

Farming management will certainly affect the environmental condition. It can influence the diversity of species in the area [3] and the quality of soil [4]. Monoculture system in farming practice can affect the loss of species richness and diversity of plant [4]. Soil degradation will increase in the area which is planted with homogenous stands while the diversity of ground cover vegetation is decreasing [5].

The ecological balance is important in order to maintain the sustainability of cocoa production [6]. However, information on the diversity of ground cover vegetation and soil chemical properties in the cocoa plantation is still limited. This information is very useful to determine the appropriate method to manage the plantation because each area requires its own method to achieve sustainable agricultural practices [7]. This study aims to determine the diversity of ground cover vegetation and soil chemical properties. The results can be used as a basis for the management of the sustainable cocoa plantation.
2. METHODS

2.1 Study Site
The study was conducted at Katibung district in South Lampung Regency. This area produces the highest cocoa bean for the regency. Katibung district was located between 105°14’ and 105°45’ E and 5°15’ and 6° S. This area had 3.834 ha cocoa plantation grown by smallholder farmers.

2.2 Diversity Observation
The sampling design to measure the diversity of ground cover vegetation was random using quadrat method. There were 27 plots of 1 x 1 m quadrat samples. Plots were used to count the grass, shrub, and seedlings with a height of less than 1,5 m. The method used was count/list quadrat count by counting the number of individual present and the number of individuals of each species in the plot [8]. The diversity of ground cover vegetation was analyzed through Shannon-Wiener diversity index and can be categorized as Odum [9].

\[ H' = - \sum \frac{n_i}{N} \log \frac{n_i}{N} \]

Figure 1. Shannon-Wiener diversity index formula

The stability of a community depends on the diversity of its growth, by means more higher diversity, more stable community. Vegetation diversity reaches a high level when the value of H’ is more than 3. A moderate level is when the value of H’ is in the range of 2 to 3. The biodiversity is said to be low if the value of H’ is less than 2.

2.3 Chemical Soil Properties
The soil samples were taken by disturb sample method at a depth of 0-20 cm. Soil chemical properties were analyzed at the Laboratory of Soil Science, Faculty of Agriculture, University of Lampung. The parameters analyzed include soil pH, organic C, total-N, P2O5 available, CEC, base saturation, salinity, Al saturation. The result can be used to determine the suitability of the land to cultivate cocoa.

2.4 Cocoa Plantation Management
Information on the cocoa plantation management was obtained through interviews and questionnaires using purposive sampling method. The respondents were smallholder cocoa farmers. They own about 0,5 to 3 ha cocoa plantation. The information collected is about the management of their land and plantation.

3. RESULTS AND DISCUSSION

3.1 Diversity of Ground Cover Vegetation
There were 19 species of ground cover vegetation found in the area (Table 1). *Spigelia anemia* was the most dominant vegetation found as ground cover vegetation. There were also *Clitoria alternate* and *Setaria app.* whose abundantly covering the ground.

Analysis of Shannon-Wiener index showed that the ground cover vegetation diversity is 2,21. This may imply that ground cover vegetation under the cocoa plantation was in the category of quite in abundance. This means that the condition was quite stable with middle environment pressures.

### Table 1. Diversity of ground cover vegetation under cocoa plantation

<table>
<thead>
<tr>
<th>Species</th>
<th>RF</th>
<th>RD</th>
<th>IVI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acalypha indica</td>
<td>2,02</td>
<td>4,04</td>
<td>1,01</td>
</tr>
<tr>
<td>Algarum compositus</td>
<td>15,15</td>
<td>1,01</td>
<td>7,07</td>
</tr>
<tr>
<td>Clome sp.</td>
<td>2,02</td>
<td>2,02</td>
<td>1,01</td>
</tr>
<tr>
<td>Clitoria ternatea</td>
<td>2,02</td>
<td>2,02</td>
<td>1,01</td>
</tr>
<tr>
<td>Cocos nucifera</td>
<td>2,02</td>
<td>2,02</td>
<td>1,01</td>
</tr>
<tr>
<td>Cyperis rotundus</td>
<td>2,02</td>
<td>2,02</td>
<td>1,01</td>
</tr>
<tr>
<td>Digitaria sp.</td>
<td>2,02</td>
<td>2,02</td>
<td>1,01</td>
</tr>
<tr>
<td>Dryopteris sp.</td>
<td>2,02</td>
<td>2,02</td>
<td>1,01</td>
</tr>
<tr>
<td>Euphorbia xerophytica</td>
<td>13,13</td>
<td>24,24</td>
<td>8,08</td>
</tr>
<tr>
<td>Ficus septica</td>
<td>2,02</td>
<td>2,02</td>
<td>1,01</td>
</tr>
<tr>
<td>Melastoma decipiens</td>
<td>14,78</td>
<td>14,78</td>
<td>5,51</td>
</tr>
<tr>
<td>Musa paradisiaca</td>
<td>14,78</td>
<td>14,78</td>
<td>5,51</td>
</tr>
<tr>
<td>Peperomia pellucida</td>
<td>2,02</td>
<td>2,02</td>
<td>1,01</td>
</tr>
<tr>
<td>Phyllanthus nir urva</td>
<td>14,78</td>
<td>14,78</td>
<td>5,51</td>
</tr>
<tr>
<td>Paspalum conjugatum</td>
<td>2,02</td>
<td>2,02</td>
<td>1,01</td>
</tr>
<tr>
<td>Saururus cohnghyus</td>
<td>2,02</td>
<td>2,02</td>
<td>1,01</td>
</tr>
<tr>
<td>Setaria app.</td>
<td>2,02</td>
<td>2,02</td>
<td>1,01</td>
</tr>
<tr>
<td>Spigelia anemia</td>
<td>2,02</td>
<td>2,02</td>
<td>1,01</td>
</tr>
<tr>
<td>Theobroma cacao</td>
<td>2,02</td>
<td>2,02</td>
<td>1,01</td>
</tr>
</tbody>
</table>

RF = Relative Frequency; RD = Relative Density; IVI = Importance Value Indeks
3.2 Soil Chemical Properties

Soil analysis showed that some soil properties were in the category of suitable for cultivating cocoa, but some indicated that the land was less suitable as cocoa plantation (Table 2).

<table>
<thead>
<tr>
<th>Soil Chemical Properties</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH H₂O</td>
<td>5.73</td>
</tr>
<tr>
<td>N (%)</td>
<td>0.10</td>
</tr>
<tr>
<td>P₂O₅ (ppm)</td>
<td>28.12</td>
</tr>
<tr>
<td>CEC (me/100g)</td>
<td>6.26</td>
</tr>
<tr>
<td>Base saturation (%)</td>
<td>31.95</td>
</tr>
<tr>
<td>Organic carbon (%)</td>
<td>0.82</td>
</tr>
<tr>
<td>Al saturation (%)</td>
<td>0.81</td>
</tr>
</tbody>
</table>

When the soil properties are compared with the class of land suitability for cocoa, the value of P and Al saturation are in the category of suitable 1 (S1). The pH value, N, and base saturation are in the category of suitable 2 (S2), but the value of CEC (cation exchange capacity) and organic C are in the category of suitable 3 (S3).

3.3 Smallholder Management on Cocoa Plantation

The land area managed by each family was on the average of 1 ha. It was about 43.8% families managed 1 ha of cocoa plantation. 32.5% families managed less than 1 ha, and the rest (23.8%) managed more than 1 ha of cocoa plantation. Some farmers cultivated only the cocoa plant, but around 90% of the farmers grew other commodities in very small quantities in their cocoa fields. Some of the farmers also raised coconut, chili, Jeong Koi (Archidendron pauciloquy), Pete (Parkia species), clove (Syzygium aromaticum), or durian (Durio sp.) in the field.

Few farmers did not give fertilizer to their field. Around 18.8% of the farmers gave only chemical fertilizer, while the other 25% using both organic manure to fertilize their land. Most of them (53.8%) used both organic and chemical fertilizer. The amount of fertilizer used by each farmer was not the same.

Some farmers were already aware of the importance of organic fertilizer to the soil. It can be seen from the majority of the farmers (70%) which had made compost either in groups or individually. The farmers did the composting by utilizing residue materials from cropping and pruning or by adding manure to the pile of farming waste.

Residual materials from cropping and pruning were not only used to make compost. Some of it were used as livestock feed. Around 7.5% farmers used the residues both for livestock and to make compost. Unfortunately, 32.5% farmers hadn’t utilized the residues at all.

Treatment of pest and plant diseases was still dominated by administering chemicals (insecticides, herbicides, fungicides). Few farmers (2.5%) began to use natural material to overcome plant diseases, such as the use of turmeric, while many of them (37.5%) did not do any treatment against pests and plant diseases.

Most farmers did not regularly perform maintenance for the plant. The spacing varied in every field. Most of them used monoculture system. However, there are other crops that were planted irregularly and in amounts far less than the main crop.

3.4 Discussion

Cocoa plantation management as described gave the result of ground cover vegetation biodiversity in the region get into the category of moderate level. The ecological balance is important in order to maintain the sustainability of cocoa production [6]. However, monoculture system in farming method could affect the loss of species richness and diversity of plant [4]. The mix crops in agricultural have been suggested [7] in order to create a sustainable agriculture. Vegetation cover is also known to give provision of ecosystem services [10]. Mix-crop in cocoa plantation usually uses coconut, banana, or perennial trees (i.e. Gliricidia septum, Durio sp., Parkia speciosa) [11], [12]. For conserving the environment, the diversity of ground cover vegetation should be preserved or even enhanced. Cultivating a variety of ground cover plants can protect the land from degradation, maintain soil moisture, increase the content of organic matter, reduce run-off and increase carbon sequestration [13]. This area needs a suitable pattern of a mixed cropping system for the sustainability of the environment. Environmental stability must be considered to maintain the sustainability of cocoa plantation.
As a result of cocoa management practices of smallholder farmers for years, the CEC and organic carbon element fall into the low level (Table 2). CEC is an element that is closely related to soil fertility [14]. Soil with a high CEC is able to absorb and provide better nutrients than soil with a low CEC [15]. Soil fertility will be increased if the majority of the elements is dominated by alkaline cations, Ca, Mg, K, and Na [15, 16]. In addition, element C is one of the essential nutrients required by plants because it improves the soil physical properties, CEC, the water-holding capacity, and helps tie up particles in the aggregate [16]. The lack of C leads to less optimal plant growth. Thus, the elements of CEC and C that are low in this location indicates the less fertile soil to cultivate cocoa. CEC can be worked up by adding organic matter such as compost or manure. Farmers have started using crop residues or trimming to provide organic matter to the soil. The ground cover vegetation can also be harvested as organic matter to make compost. Thus, the carbon content of dead plant matter can be returned again to the ground.

Cocoa was not the only commodity cultivated by the most cocoa farmers in Katibung district. Most of the farmers (90%) planted another commodity to provide additional income. They usually grew petal, durian, clove, and mainly banana. Ground-cover vegetation was still considered to be useless as a weed. Whereas, the ground cover vegetation is potential to support other sectors. Cleome sp. and Spigelia anemia are potential plants for medicine [17]. S. Anthelmia extract can also be used to control worms in cattle [18]. Acalypha indica indicates extract is believed to be used against malarial vector, Anopheles Stephens [19, 20]. Ageratum conicoid's extract can be used as a natural insecticide for controlling Spodoptera literal larvae (usually attack agricultural crops) [21]. Crassocephalum trepiddies can be domesticated and consumed as a vegetable [22]. In traditional folk medicines, the rhizome of Cyperus rotundus is used to treat stomach diseases, bowel disorder, and inflammatory diseases [23]. Digitaria sp., Setaria spp., Paspalum conjugated are usually used as livestock feed [24, 25]. The extract of E. hiroe is a promising antibacterial against Aeromonas hydrophilia in vitro [26], Staphylococcus Aureus and Escherichia coli [27]. Ficus septic is usually used by Tolitoli tribe for medicine [28]. There are many benefits from the leaves and roots of Ficus septic such as curing skin diseases, appendicitis, ulcers, snake bites, poison fish, and prevention of asthma [29]. Peperomia pellucida extract has been tested to decrease uric acid levels in animal and has already been used by the public [30]. Phyllanthus Nir Urf has been used empirically by the public to cure many diseases such as gonorrhea, ureter infection, stomach ache, toothache, fever, dysentery, diuretics, and diabetes [31]. Thus, farmers can start to manage the growth of ground cover vegetation. When the fertility of the soil needs to be improved, then the ground cover vegetation would be better to be returned to the soil in the form of compost. Meanwhile, when the plants are considered quite excessive, then the plants can be harvested for use as needed.

4. CONCLUSIONS

As a result of cocoa management practices for years, ground cover vegetation diversity in the region falls into the category of moderate level. For conserving the environment, the diversity of ground cover vegetation should be preserved or even enhanced. In addition, soil condition for a particular element (CEC and organic C) is at a low level. It can be said that the land is less conducive to cultivate cocoa. It will be better to manage ground cover vegetation in the area. Ground-cover vegetation can be harvested as a compost material to improve soil condition or as a beneficial crop (feed for livestock, herbal medicine, insecticide, etc.). Furthermore, ground cover vegetation will maintain the environmental stability.

5. REFERENCES

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Long Fermentation Effect on Liquid Fertilizer for Spinach, Green Mustard, Watermelon and Banana Peel Waste Toward Nutrient Ingredients of Phosphor and Potassium with Effective Microorganism4 (Em4) Bioactivator Addition

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Abstract-Spinach, green mustard, watermelon and banana peels are vegetables and fruits that have high nutrients content. Currently, these vegetables and fruits are not fully used as organic fertilizer, even though the content of phosphorus and potassium is high. This research aims to determine the content of macronutrients of Phosphorus (P) and Potassium (K) of liquid organic fertilizer after fermentation with Effective Microorganism-4. This research is completely randomized design (CRD) with the treatment of three level fermentation time (4, 8 and 12 days) and commercial fertilizer as a control. Follow the fermentation is the analysis of levels of total P and total K, using the spectrophotometric method. The results showed that the fermentation time (4, 8, 12 days) gave different level of total P and total K, namely 0.0282%, 0.0271%, 431%, respectively of total P; and 0.3033%, 0.4290%, 0.4236% of total K. In comparison with control (0.06% of total P and 0.49% of total K), these results are lower. The optimal of long fermentation to get the highest total content of P is 12 day. Meanwhile, the optimal of long fermentation to get the highest total content of K is 8 day.

Keywords: Fermentation, macronutrients, phosphor, potassium.

1. INTRODUCTION

Fertilizer has very important role in agriculture. It helps the plants to grow and the soil fertility to be increased, but it is different with inorganic fertilizer. The overuse of inorganic fertilizer could leave the chemical residues in the soil which dangerous for its condition. Encounter this problem; there is a solution which is the utilization of liquid organic fertilizer. It is proved that liquid organic fertilizer is friendly environment also increase the fertility of the soil. The best ingredients of liquid fertilizer which uses organic waste are from the waste of fruits or vegetables because of their high content of water.

Sriningsih research (2014), got the result of banana peel utilization with the EM4 addition to be liquid fertilizer was having Phosphor content 106.53 ppm and Potassium content 1686.60 ppm [1]. Furthermore, this banana peel utilization as liquid fertilizer was innovated with the addition of green mustard, spinach, and watermelon peel. Utilization of those additional ingredients is in order to increase the content of P and K in the fertilizer. Meanwhile, the prime ingredients of the fertilizer are included as cost less ingredient and easy to get. Also, it is rich in minerals.

In the process of making liquid fertilizer, there is a need of bacterial to bound the unsure of nitrogen, phosphor, potassium, and some other unsure. Effective Microorganism-4 (EM4) is a microorganism that can be used and was used for this research. Its role is in the fermentation process. There is another important factor which is the fermentation time. The fermentation time function is giving the time to the microorganism to outline organic elements in liquid fertilizer so that can be absorbed by neighboring plants. Mujiatul research (2013), got the result as fermentation time affect the amount of N, P, and K content in liquid fertilizer from tofu’s waste with Mexican marigold addition. Mujiatul got the highest amount of N, P, and K content from 4 days fermentation process with N content of 732 ppm, P content of 840.6 ppm, and K content of 7189.8 ppm [2].

There are various ingredients that can be turned into liquid fertilizer. The waste of our surroundings can be utilized. So that a number of soil nutrients or organic elements and the microorganisms also has many varieties. For it, there should be further research to be conducted to analyze the content of any other liquid fertilizer from different ingredients so specific benefit can be known and delivered to the people who needs for the good use.

The purpose of the research is to know the total of Phosphor and Potassium content in liquid fertilizer as the result of the fermentation of spinach, green mustard, watermelon peel, and banana peel with EM4 addition and to know how long does the fermentation takes time to get the highest amount of total phosphor and potassium content in the liquid fertilizer.
2. METHODS

The type of this research is a quantitative descriptive research with experimental design. The experimental design is Completely Randomized Design (CRD) with a factorial. The free variable of this research is the fermentation time (4 days, 8 days, and 12 days). The bound variable of this research is Phosphor content (P) and Potassium content (K) in the liquid fertilizer. Meanwhile, the controlled variable of this research is the mass of the primer ingredients, EM4, sugary water or molasses, and each total of P and K commercial liquid fertilizer.

The first step of this research is to make the sample of the liquid organic fertilizer that will be fermented with the continuation of testing the result of fermented liquid fertilizer. The procedures began cutting the primary ingredient, the second mix all the ingredient, and then prepare the fermentation solvent with EM4 and molasses. The next steps are anaerobic fermentation process, and the end analyzed the research of the fermentation.

Primary parameter that will be observed is the phosphor and potassium content in the liquid fertilizer using spectrophotometry method. Phosphor using spectrophotometer UV-Vis and Potassium using spectrophotometer AAS. Data that is collected will be statistically analyzed through Analysis of Variance (ANOVA) One Way with SPSS application. The value of F critical for $\alpha = 0.05$. If the value of $F_{\text{obs}} > F_{\text{crit}}$, it will be significant [3].

3. RESULTS AND DISCUSSION

These are the average of total Phosphor (P) and Potassium (K) content in the liquid fertilizer as the result of spinach, green mustard, banana peel, and watermelon peel fermentation (in percentage %) from different fermentation time.

<table>
<thead>
<tr>
<th>No</th>
<th>Treatment</th>
<th>Measurement Result (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>U1</td>
</tr>
<tr>
<td>1</td>
<td>4 days fermentation</td>
<td>0.0277</td>
</tr>
<tr>
<td>2</td>
<td>8 days fermentation</td>
<td>0.0275</td>
</tr>
<tr>
<td>3</td>
<td>12 days fermentation</td>
<td>0.0418</td>
</tr>
</tbody>
</table>

Note: U1 : 1st Repetition  
U2 : 2nd Repetition  
U3 : 3rd Repetition

From the result of laboratory analyses about the phosphor content, the researcher got a different average percentage of total phosphor content according to the treatment.

Average total phosphor content from 4 days, 8 days, and 12 days fermentation time (in %) are 0.0282; 0.0271; and 0.0437. The phosphor content from commercial liquid fertilizer is 0.06%. If the result to be compared with commercial liquid fertilizer (positive control), the total phosphor content from fermentation process is much smaller.

According to 3.1 table, the highest amount of total P content in liquid fertilizer from spinach, green mustard, banana peel, and watermelon peel fermentation is from 12 days fermentation treatment with $437 \text{ ppm (0,0437%) }$ and the lowest amount of total P content is from 4 days fermentation treatment with $282 \text{ ppm (0,0282%) }$.

Also from the data analysis, the researcher got the result that the total phosphor content in liquid fertilizer is significantly affected by the fermentation time. The factor that possibly significantly affect the data is the activity of micro bacteria Lactobacillus sp., Streptomyces sp., cellulose-degrading fungus, and yeast that can reform phosphor which leaves the total P content raise [4]. Other than that, it can be affected by the substrates in organic used.

As for the potassium content laboratory analyses, the data also had differences according to the fermentation time treatment. Average total Potassium content in liquid fertilizer from spinach, green mustard, banana peel, and watermelon peel fermentation can be seen in 3.2 table.
Table 3.2. Average Total Potassium Content in Liquid Fertilizer as the result of spinach, green mustard, banana peel, and watermelon peel fermentation

<table>
<thead>
<tr>
<th>No</th>
<th>Treatment</th>
<th>Measurement Result (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>U1</td>
</tr>
<tr>
<td>1</td>
<td>4 days fermentation</td>
<td>0.3198</td>
</tr>
<tr>
<td>2</td>
<td>8 days fermentation</td>
<td>0.4284</td>
</tr>
<tr>
<td>3</td>
<td>12 days fermentation</td>
<td>0.2050</td>
</tr>
</tbody>
</table>

Note:
- U1: 1st Repetition
- U2: 2nd Repetition
- U3: 3rd Repetition

Average total potassium content from 4 days, 8 days, and 12 days fermentation time (in %) are 0.3033; 0.4290; and 0.4236. The potassium content from commercial liquid fertilizer is 0.49%. If the result to be compared with commercial liquid fertilizer (positive control), the total potassium content from fermentation process is much smaller.

From the data analysis, the researcher got the result that the total potassium content in liquid fertilizer is not significantly affected by the fermentation time. Another way, the researcher can say that through all the treatment, there were no significant differences.

4. CONCLUSIONS

The conclusions from the research are:

1. Total phosphor content in liquid fertilizer from spinach, green mustard, banana peel, and watermelon peel with EM4 addition for 4 days, 8 days, and 12 days fermentation are 0.0282%, 0.0271%, and 0.0431%. Total potassium content in liquid fertilizer from spinach, green mustard, banana peel, and watermelon peel with EM4 addition for 4 days, 8 days, and 12 days fermentation are 0.3033%, 0.4290%, and 0.4236%.

2. Optimum fermentation time to get the highest amount of total phosphor content is 12 days, and total potassium content is 8 days.

5. REFERENCES

Effect of Time’s Giving of Arbuscular Mycorrhiza Fungi (Amf) for Koro Hijau (Macrotyloma Uniform) Growth as Pioneer Plants for Soil Fertility Ex-Lime Mine Reclamation

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Abstract – One of the reclamation efforts of an ex-lime landmine is by planting a certain pioneer’s plant in order to restore soil fertility. This solution would be more effective with the presence of symbiosis between pioneer plants with Arbuscular Mycorrhiza Fungi (AMF). This research was conducted to identify the influence of time’s giving of AMF for the growth of Koro Hijau and examine it’s symbiosis mutualism, as well as examine the effect of giving AMF for Koro hijau’s growth compared to control plants (without AMF). This research is an experimental research. The research was conducted to 24 plant samples which consisted of 3 treatments and 1 control which was designed using one factor ANOVA as its statistical analysis become one-factor research that was examining the effectiveness of giving AMF fertilizer in a different time. AMF was given in three different time sequences with a one-week interval as three different application treatments by spreading around in near its roots plant. The data was collected in every 5 days for 50 days by measuring the length of the stem, a number of leaves and stem diameter. Root length and its AMF infection rate were measured at the end of the experiment. The results showed that the time’s giving of AMF is significantly affected the growth of Koro Hijau and the first week (M1) have proven most effective in mutualistic symbiosis with the host plant, and the effect of growth is greater compared to control plants (K).

Keywords: Koro hijau, AMF, lime soil, fertility.

1. INTRODUCTION

Karst area is an area that covers 10% of the earth's surface [1]. In Indonesia, one of karst area which is currently known is Gunung Sewu Karst region, Gunung Kidul, Yogyakarta. Gunung Sewu karst can be categorized as open type (bare karst) which is characterized by karst formations which are a phenomenon of karst topography. Its typical form of conical hills is not found in other karst areas around the world [2]. The existence of karst areas in Gunung Sewu is currently experiencing a serious threat of damage due to limestone mining without adequate reclamation efforts.

The problem of ex-lime mine can be resolved with some efforts, one of them is by planting pioneer plant species. One of the pioneer plants which has a great potential improving the condition of mine lands is Koro Hijau (Macrotyloma uniform). Koro hijau is a plant from the legume family. It grows in many countries, especially in tropics and sub-tropics region [3]. It also grows well in dryland region with temperatures between 25-35 °C. It is adaptable to different types of soil including sand soil, gravel, and poor mineral soil. Its growth is enormously fast. According to an observations which have been made in India showed that within 6-7 weeks after planting, this plant is capable of producing fresh forage as much as 5-12 tons per hectare [4].

Koro hijau is a legume which is able to associate in mutualistic symbiosis with AMF. In its symbiosis, host plants provide a fixed supply of sugar to the fungus. Meanwhile, fungi increase the surface area for the up taking of water and also supply the plant with phosphates and other minerals absorbed from the soil. AMF also secrete growth factors that stimulate the roots to grow and branched, such as an antibiotic that helps protect plants from pathogens in the soil [5]. AMF has an important role for the region nutrient-poor soils, assisting the decomposition of organic waste, translocating of some nutrients such as nitrogen and phosphorus from the soil to the roots and water networks [6].
2. MATERIAL AND METHOD

1.1 Time and Place

This study was held from June to October 2016 in Department of Biology Education Laboratory of Sanata Dharma University, Yogyakarta.

1.2 Material and Tools

Materials used in this research are the seeds of Koro hijau, grumusol soil, limestone soil, soil cytosol, AMF fertilizer, natural pesticides, and water, KOH 10%, HCl 1%, staining solutions, trypan blue 0.05%, and Immerse oil.

The tools used in this research are polybag, hoes, spades, bamboo, water sprayer, hygrometer, acidity meter, ruler, digital caliper, raffia, gloves, scales, buckets, microscope, petri dish, pipette, cutter, glass objects, autoclave, stationery, and cameras.

1.3 Research Method

This research is an experimental research. The research was conducted to 24 plant samples which consisted of 3 treatments and 1 control which was designed using one factor ANOVA as its statistical analysis become one-factor research that was examining the effectiveness of giving AMF fertilizer in a different time.

3. RESULTS AND DISCUSSION

Based on data from the mean’s growth of Koro hijau shows that treatment of M1 has the best growth rate compared to the growth rate of M2 and M3. Treatment M1, M2, and M3 have rates below and the same (especially in mean diameter stem) compared to K as a negative control (Table 1).

Based on ANOVA test, stem length has a value of sig probability 0.605 > 0.05. It shows that the treatment of the AMF at different times does not significantly affect the growth of stem length compared to the negative control due to the difference in length stem is small. Meanwhile, for the probability of leaves number and stem diameter value is sig 0,036 and sig 0.033 <0.05. It shows that the treatment of the AMF at different times and significantly different effect on the growth of Koro hijau compared to negative control treatment because there is a difference between the numbers of leaves and stem diameter. Based on the observations of AMF on root infection, M1 and M2 treatment have 52.4% and 59.8% which are categorized high rate of infection, and M3 treatment has 30.6% infection which is categorized has moderate infection rates.

<table>
<thead>
<tr>
<th>No</th>
<th>Treatment and Control</th>
<th>Stem length (Cm)</th>
<th>Leaf number</th>
<th>Stem diameter (Cm)</th>
<th>AMF Infection %</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>M1</td>
<td>61.70</td>
<td>22</td>
<td>0.29</td>
<td>52,4</td>
</tr>
<tr>
<td>2</td>
<td>M2</td>
<td>44.30</td>
<td>19.75</td>
<td>0.21</td>
<td>59,8</td>
</tr>
<tr>
<td>3</td>
<td>M3</td>
<td>47.20</td>
<td>16</td>
<td>0.23</td>
<td>30,6</td>
</tr>
<tr>
<td>4</td>
<td>K</td>
<td>74.30</td>
<td>32</td>
<td>0.29</td>
<td>-</td>
</tr>
</tbody>
</table>

Note: M1: First week, M2: Second week, M3: Third week, K: Negative Control (without AMF).

The given of AMF has a significant effect on the growth of Koro hijau. Toward stem length indicator, AMF treatment has a probability value greater than 0.05. Both indicators of leaf and stem diameter have a significant influence on the growth of Koro hijau and significantly different compared to the negative control. Based on the results of mean growth, M1 treatment has higher mean compared to M2 and M3 treatment’s mean. It is related to AMF which has important roles in accelerating the definitive physiological phase and increasing the durability of
the plant at the beginning of planting. Hanafiah also reported that the growth of Koro hijau as a legume is dependent on the association of AMF [7].

Influence of time’s giving of AMF for Koro hijau could also be seen by observing the root morphology and its length compared to the level of AMF infection. The thickness of the roots shows the level of mutuality between the roots of host plants with AMF. AMF will form a blanket outside and inside the roots, in the interstellar between cells of the epidermis and cortex [8]. Furthermore, the AMF also intensively will produce external hyphae on the roots of host plants (figure 2). It optimizes the process of photosynthesis, absorbing water and nutrients from the soil. From the results, M1 treatment has the thickness roots level compared with M2 and M3 treatment as well as negative control (figure 1). The level of thickness and length of the roots cannot be separated from the rate of AMF infection. Based on the development of roots, treatment of M3 has the greatest root length. However, it does not show the characteristic form of mutualism between the AMF and host plant. Good root growth enables plant roots to absorb nutrients around the roots. AMF works by covering the exterior root near the top root, and the hyphae work like a coat. It is even able to spread up to 8 m [9]. Another hypha will penetrate the host plant root cell membrane to form a network of nutrient absorption. Based on the characteristics of the development of AMF root infections, the early development of AMF symbiosis with the host plant showed a negative relation to geotropism [10].

AMF utilization for the growth of Koro hijau as a pioneer plant for reclamation of ex-lime mine is needed. The region of ex-lime mine does not have characteristics or capacity which support the growth of the plant because it has poor soil quality. However, by inventing AMF association and the addition of organic fertilizer at planting media allows plants to obtain sufficient nutrients [11]. AMF offers major advantages in plants that live in the poor soil. It is also said that without the help of AMF to absorb nutrients, many plant communities are not able to survive. Khasa et al. (2009) reported that the CMA increase the mobilization of nutrients for plants. In addition, the CMA also increased mobilization of nutrients in the soil that has a low concentration of nutrients number.

Soil fertility levels in some region commonly have some reservations. Generally, the requirements for soil fertility is determined by the availability of water, microorganisms, soil biota, the number of species of plants and soil types. In addition, the soil fertility is also affected by the availability of humus, clay and neutral acidity [12]. Lacking some of those reservations can affect the level of soil fertility. According to Khasa (2009), the symbiosis between the AMF and the plant like Koro Hijau can precisely increase soil fertility. AMF is generally able to correlate to various types of soil and microorganisms. In association with plants, AMF helps plants photosynthesizing, binding C and N element and distributing them into the surrounding. Those elements eventually are eventually needed to improve soil fertility.

4. CONCLUSIONS

Time’s giving of AMF affect the growth of Koro hijau and M1 application treatment is the most influence in symbiosis mutualism between AMF and Koro hijau compare to M2 dan M3 treatments. In addition, the effect of giving of AMF for Koro hijau’s growth is greater than plants control (without AMF).

5. REFERENCES


Spatial Variability of Diurnal Precipitation over Southern of Sumatra during 2009–2010

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2 Department of Physics, Faculty of Mathematics and Natural Sciences, University of Sriwijaya, Palembang, Indonesia
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Abstract – This study was designed to examine spatial variations of diurnal precipitation over Southern of Sumatra for a period of 1 January 2009–31 December 2010. The Indonesian maritime continent is surrounding by many islands and sea. Therefore, the spatial variation of its precipitation will be strongly influenced by the interactions between land and adjacent sea. The data of this study were provided by the Tropical Rainfall Measuring Mission (TRMM) Precipitation Radar (PR) and the National Centers for Environmental Prediction (NCEP)/the National Centers for Atmospheric Research (NCAR) reanalysis. In other, to evaluate the spatial variations of the diurnal cycle of precipitation and other climate variables, the diurnal climatology analysis is focused on the area of 0.5°S – 6.5°S and 95°E – 112°E. The study result shows that the intensity of precipitation over the ocean is much higher than that over the land. High precipitation over the ocean region occurred in the early morning during 0400–1000 Local Time (LT), while lower precipitation was observed in the evening (1900–2200 LT). On the other hand, the rainfall peak over the land was observed in the evening (1900–2200 LT) and the low rainfall was occurring in the daytime (1300–1600 LT). In addition, high precipitation both over the land and over the ocean is associated with the upward motion. Based on the hovmöller diagram, the precipitation intensity was concentrated near the coast, but it shows offshore migration during the daytime.

1. INTRODUCTION

Indonesia is a maritime continent surrounded by two oceans and two continents. This position makes Indonesia climate is influenced by Monsoon wind that flows over the Indonesian continent. The thermal difference between land and sea regions will cause the pressure difference between land and sea regions. It corresponds to the annual movement of the sun. In addition, local and regional characteristics of the lands in Indonesia were also influenced by convection variations in Indonesian continent. In concludes, the interaction between the Monsoon wind and complex topography were caused high variation of the spatial and temporal precipitation intensity over Indonesian continent [1]. In addition, other global climate factors also influence rainfall patterns in Indonesia, such as the El Niño–Southern Oscillation (ENSO) and the Indian Ocean Dipole (IOD) events. Those phenomena also contribute to the interannual climate variations. Both the climate events are important because of their large environmental and societal impacts, globally and regionally [2].

Precipitation displays high space-time variability that requires frequent observations for adequate representation [3]. Rain gauge observations yield relatively accurate point measurements of precipitation but suffer from sampling errors in representing real means and are not available over most oceanic and unpopulated land areas [4]. However, nowadays, rainfall data required for a wide range of scientific applications can be achieved through meteorological satellites [5]. Meteorological satellites expand the coverage and time span of conventional ground-based rainfall data for a number of applications by which all hydrology and weather forecasting are made [6]. The use of remote sensing, which has better spatial and temporal resolution data, in a study about rainfall and its spatial relationship to ENSO and IOD in Indonesia, thus offers an exciting opportunity.

Tropical Rainfall Measuring Mission (TRMM) Precipitation Radar (PR) satellite launched in 1997 because it has the advantage of detecting raindrops directly from space, which allows us to reveal the rainfall diurnal variation accurately over the globe [7]. A considerable amount of validation has been performed to certify that the estimated near-surface rainfall rate is reliable [8]. TRMM satellite data have good performance to representing rain gauge data that is shown by correlation coefficient both of the data is significant [9].

Seasonal and interannual variability over the Indonesian Maritime Continent including Sumatra Island have been examined using long term statistical data [10]. Relation between rainfall amount at Kotatabang and background
wind circulation for a limited period and also study about diurnal land-sea rainfall peak migration over Sumatra Island has been also documented [11]. These studies have described unique characteristics of both regional and temporal rainfall over this area, but the spatial variation of diurnal precipitation over Southern of Sumatra has not been addressed. This study presents the diurnal variations of rainfall cycle and its regional climate impacts over the Indonesian Maritime Continent, especially around Southern of Sumatra.

2. METHODS

2.1 TRMM PR satellite data

The precipitation data were provided by the TRMM-PR satellite with spatial and temporal resolutions of $0.1^\circ \times 0.1^\circ$ which contains an hourly gridded Network Common Data Form (NetCDF) format dataset. In other, to evaluate the spatial variation of the diurnal cycle of precipitation, the diurnal climatology was calculated for a period of 1 January 2009 through 31 December 2010. The analysis is focused on the area of $0.5^\circ$S – $6.5^\circ$S and $95^\circ$E – $112^\circ$E.

2.2 Other climate data

Objective reanalysis data, that are the vertical and horizontal wind, the specific humidity, and the vertical velocity data, provided by the National Centers for Environmental Prediction (NCEP) the National Center of Atmospheric Research (NCAR) reanalysis. These data were used to analyze diurnal variability. The spatial and temporal of those data are $2.5^\circ \times 2.5^\circ$ and 6-hour, respectively. Interpolation data is needed in these data. It is to set same data period between precipitation and other climate data which were 6-hours interpolated to 8-hour data. In addition, to evaluate the spatial variation of the diurnal variability of these data, the calculations have been done for the same time period and area coverage of all the data.

3. RESULTS AND DISCUSSION

This study has been examined diurnal variation of precipitation over the Southern Sumatra region. The spatial variation of diurnal precipitation is shown in Fig 1. The intensity of precipitation over the ocean is much higher than that over the land. High precipitation over the ocean region occurred in the early morning during 0400 until 1000 Local Time (LT). The intensity of rainfall area is concentrated along the coastal area of Sumatra Island at 0400 LT, then expands its area to the ocean region at 1000 LT. Whereas, lower precipitation was observed in the evening (1900–2200 LT). Rainfall intensity over the Northwestern of Java Sea and Karimata Strait is almost cleared up in the evening rain.

Figure 1. The high and low intensity of diurnal precipitation over the ocean region

On the other hand, the intensity of precipitation over the land region shows more rainfall in the evening than that in the daytime. Figure 2 shows that the rainfall peak over the land was observed in the evening which starts at 1900 LT until reach its peak at 2200 LT. The rainfall area covers most of the Southern Sumatra land at 2200 LT. But, most of strong precipitation intensity was shown over the coastal sea region at this time. Rainfall over the land region expands its area into the ocean region and the low precipitation over the land was occurring in the daytime (1300–1600 LT). The clear up of the rainfall intensity over the land was observed at 1300 LT.
We also examine moisture flux which is generated by zonal and meridional wind and specific humidity data to relate background conditions to spatial variation of diurnal rainfall intensity. Moisture flux convergence area is concentrated in the offshore region (0100 LT–0400 LT), then migrate to the land region along the northwestern Sumatra Island at 1600 LT and increase with time until meet its peak at 1900 LT (not shown). Based on the hovmouller diagram of precipitation and moisture flux data that is present in figure 3, the precipitation intensity is concentrated near the coastal region, but it shows offshore migration during the daytime. The area of increased (decreased) precipitations associated to the enhanced (reduced) moisture flux. Most of strong intensity of moisture flux appears in the ocean region during 13 00 LT until 19 00 LT. Spatial distribution of rainfall intensity is also influenced by vertical and horizontal wind that is blow towards and backwards the ocean and land regions. The westerly wind is stronger than easterly wind. It causes moisture flux from the ocean migrate towards inland regions during 13 00–19 00 LT, then migrates toward the ocean until the late evening (not shown). We also have examined wind convergence in this study that is present in figure 4. Clear convergence over Sumatra Island appears at 00 00 LT. This condition maintains the wind divergence until reach its peaks at 04 00 LT. The convergence is observed in the daytime that is start at 13 00 LT over the northwest Sumatra. In addition, strong convergence appears mostly in large islands, for example, Sumatra and Kalimantan. This suggests that most of the convective clouds over the land region in the daytime. On the other hand, the convective cloud maintains its area over the ocean region in the evening. This result is consistent with the study of diurnal land-sea rainfall peak migration that is done by Mori et al. This study result shows that the convergence field appears 4–6 hours earlier than that of the strong
precipitation intensity over both the land and ocean regions. In addition, Convergence wind is associated with the upward motion calculation (not shown). Consistent and strong upward motion of moisture air show at 13 00 LT. Based on the figure 4a, consistent upward motion maintains its coverage area over 98°E to 105°E. These results indicate that upward motion is developed over northwestern Sumatra and also along the northwest coastal region of Sumatra Island. In addition, high precipitation both over the land and over the ocean is associated with the upward motion.

4. CONCLUSIONS

Spatial variations of diurnal precipitation cycle over Southern Sumatra have been examined using TRMM and objective reanalysis data, and the results are summarized as follows:

1. The intensity of precipitation over the ocean is much higher than that over the land. High and low intensity of the precipitation over the ocean is about 1 mm/hour and above and 0.2 mm/hour, respectively. On the other hand, High and low intensity of the precipitation over the land is about 0.5 mm/hour and 0.1 mm/hour, respectively.

2. Based on hovmoller diagram, spatial variability of precipitation is caused by the migration of the rainfall intensity from the coastline toward the land region in the daytime and toward the offshore in the nighttime.

3. The convergence field appears 4-6 hour earlier than that of rainfall over both the land and sea regions.

5. REFERENCES

Biopigment Tracing and its of Mangrove *Rhizophora mucronate* Leaf and Bark Waste and Its Application in Batik Fabrics With Various Fixation Methods

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**Abstract** – The purpose of this research is to determine the pigments in the bark and leaves of mangrove (*Rhizophora mucronata*), to analyze rate of color fastness in batik, level of consumer interest, and most effective pricing for the resulting batik products. This research focused on the leaves and barks of mangrove plants. The leaves and barks were extracted to be used as dyeing agents, after which the batik fabrics were fixated using alum, limestone, and lotus. This descriptive research employed UV-Vis spectrophotometry to identify pigment and Adobe Photoshop computer program to perform color fastness analysis. The result showed that chlorophyll is a pigment which contributes to the color green in leaves. Photochemical test results also show that the pigment associated to the blackish-green hue was tannin, and that which was associated with red was flavonoid and quinone. The various colors were applied in batik dyeing process, which resulted in a range of color from yellow to dark brown. Colorfastness test of batik dyed with mangrove-derived pigments on wash air dry, wash press dry, and wash sundry using calico as fabric showed that alum and limestone fixated fabric had the least color fastness, while conversely, lotus-fixated fabric displayed good color retention.

**Keywords**: Bio pigment, mangrove waste, fixation, batik

1. INTRODUCTION

An alternative to synthetic dye is a natural dye, most of which are derived from plants. Within the tissue of plants is pigment which gives distinctive colors based on its chemical structure. Such chemical structures include chlorophyll, carotenoid, tannin or anthocyanin [1]. Fashion and batik fabric design using natural dye have added retail value because they are artistic, have unique colors, and are environmentally friendly, adding the impressions of ethnicity and exclusiveness. One of the most viable natural dye materials, which is also proven to be environmentally friendly, is mangrove waste. For example, different colors can be obtained by processing leaf, twig, and root waste of mangrove trees. Colors resulted from such processing are in the hue range of yellow, red and brown [2].

The importance of information of pigments on mangrove (*Rhizophora mucronata*) natural dye which is used as a treatment for batik and consumer interest as well as ideal pricing for naturally-dyed batik from mangrove waste. Providing the importance of such information, this study aims to: identify the pigment contained in the leaf and bark of mangrove waste; determine the color fastness of batik fabrics treated with mangrove-based dye.

2. METHODS

2.1 Sampling, Extraction Process, Pigment Identification and Phytochemical Screening

Samples of mangrove leaves and barks were collected from Mangunharjo village, Semarang city. Dried mangrove leave and bark samples were then extracted. The study focused on 18 colors extracted from mangrove leaves and barks as natural dye for batik fabrics, fixated by alum, limestone and lotus with three repetitions.

The extraction of mangrove (*Rhizophora mucronata*) leaves and barks was carried out in two parts in this research. The first part was synthesizing natural dye from mangrove waste [3] and extraction by ethanol 95% solution [4][5]. The second part included several processes, namely: batik painting, batik dyeing, fixation, batik wax removal by repeatedly boiling and fabric drying. Pigment identification using UV-Vis spectrophotometer was carried out by observation of absorbance spectrum pattern, maximum wavelength (\(\lambda_{max}\)), and percentage of the difference in height between the 3rd and the 2nd peaks (%III/II) [6]. Ethanol extracts of mangrove leaves and barks were put into phytochemical screening to determine the secondary metabolites compounds [7].
2.2 Color Fastness Test for Batik Fabrics

This test was performed to determine the color quality of batik fabrics with three treatments: wash-air and dry, wash-iron and dry, wash and sun dry. Obtained image data were coded in accordance to the treatment used for each fabric sample, after which the said fabric was scanned under Canon scanner. Data from fabric scanning were then processed using photoshop to obtain RGB (Red/Green/Blue) [8].

The color fastness test was performed to gauge the color quality obtained from each fixation process using fractional factorial design (3x2) experimental method in SPSS 10 where the first factor was fixation compound (alum, lotus, limestone) and the part of mangrove (leaves and barks). Research data were analyzed with F test on 5% significance.

3. RESULTS AND DISCUSSION

The screening in this research showed that the mangrove leaves and barks contained several distinct secondary metabolite compounds as listed in Table 2.

### Table 2. Result of Leaf and Skin Phytochemical Test

<table>
<thead>
<tr>
<th>Types of Mangrove Waste</th>
<th>Class of Secondary Metabolite Compound</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Tannin</td>
</tr>
<tr>
<td>Leaf</td>
<td>+ +</td>
</tr>
<tr>
<td>Bark</td>
<td>+ +</td>
</tr>
</tbody>
</table>

Note: (-) = not detected  (+) = positive  (+++) = strong positive

3.1 Batik Natural Dye from Mangrove (*Rhizopora mucranata*) Waste

The study of mangrove plant (*Rizophora*) waste in the form of leaves and barks as source of natural dye, fixated with alum, limestone and lotus, resulted in various colors, which are listed in Table 3.

### Table 3. Colors from Mangrove Natural Dye with Different Fixations

<table>
<thead>
<tr>
<th>Sample</th>
<th>Fixation</th>
<th>Alum</th>
<th>Limestone</th>
<th>Lotus</th>
</tr>
</thead>
<tbody>
<tr>
<td>Leaves</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Leaves</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bark</td>
<td></td>
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<td></td>
</tr>
</tbody>
</table>

Fixation by alum resulted in light brown color, which appeared faded and pale. Fixation by limestone resulted in light brown with more brightness, whereas fixation by lotus resulted in dark brown color.

3.2 Pigment Identification by UV-Vis Spectrophotometer

The results of UV-Vis Spectrophotometer, shown in Figure 1, showed the existence of a and b chlorophyll. Chlorophyll a was detected in mangrove leaves, with 666 nm wavelength in peak I and an absorbance rate of 2.29. Chlorophyll b was detected in mangrove barks, having 396 nm wavelength in peak I and 665 nm wavelength in peak II and an absorbance rates of.

![Figure 1. Chlorophyll a and b spectra on leaves and barks of mangrove.](image-url)
3.3 Discussion

Phytochemical screening of mangrove leaves and barks with FeCl₃ showed strongly positive results on tannin by opaque, blackish green coloration. Phytochemical screenings, such as the one employed in this study, is carried out by adding reagents to sample extracts. The screening is a classical method to determine simple phenol compounds, done by adding 1% FeCl₃ dissolved in water, upon which positive results will be shown by distinct green, red, purple, blue or black coloration [4]. The blackish green coloration from the screening results was caused by the complex reaction between tannin and Fe³⁺ ions. Tannin is defined scientifically as a polyphenol compound with high molecule mass and possesses hydroxyl and other clusters (such as carboxyl), making complex formation with proteins and other macromolecules under specific environmental conditions. Mangrove contains tannin, phthalate, and furfural which act as media for dye [10]. There is a recurring interest in developing natural dye in many industrial nations recently owing to the fact that it is environmentally friendly and safer to use, in keeping with many clean production and products policies. As secondary metabolites, tannin have a number of uses such as for insecticides and fungicides for plants, for antiseptic due to its protein-controlling properties, for alkaloid poisoning antidote, for a reagent in detecting gelatin, alkaloid, and protein as well as for preservative agents in leather products.

Based on the results in Table 3, various fixations resulted in different colors. Based on the results from this study, the application of Rhizophora mucronata mangrove leaves and bark in the production of natural dye resulted in a range of color from pale to dark brown. The application of leaves and barks of Sorensasia alba, Rzophora sp, Aveccenia sp, Ceripos decandra, and Lumicera sp showed relatively similar range of dominant colors, namely pale to dark brown [2]. Producing natural dye from several mangrove species resulted in various colors [11]. Sonneratia Caseolaris yielded greenish brown, the fruit of Cerbera Manghas resulted in light brown, the leaves of Cerbera Manghas resulted greenish yellow. Natural colors are derived from natural pigments produced by plants and animals [12]. Natural colors produced by mangrove plants have been identified as greenish brown, light brown and yellowish green. The degree of the hues and the resulting dye colors are determined by the base ingredient, fixation agents, and the number and duration of dipping during the fabric dyeing process. In this study, barks were obtained from a relatively juvenile mangrove trees (aged between 3 and 4 years old), which affected the resulting colors. Fixation agents used in the fabric dyeing process also contributed to the colors; alum would bring up natural colors, limestone resulted in darker colors and lotus gave out darker colors or stronger hues. These different results were attributed to the reaction between tannin and Fe³⁺-metal, which results in complex salt (ferric tannate) appearing brown in color. Ferric sulphate reacts with oxygen to form another ferric formation, resulting in darker colors. Cotton fabric fixated using lotus resulted in the darkest coloration, brownish green, compared to those fixated with alum and limestone [13]. Complex potassium and aluminum sulfate from the reaction between tannin and lotus fixer tend to result in darker colors albeit not with the fibers, blocking out the dye and reducing interaction with the fabric.

The results of the analysis observed from the pattern line, as seen in Figure 3, showed the identification of chlorophyll a and b. Chlorophyll a was identified in the mangrove leaves with 666 nm wavelength at peak I and 2.29 absorbance rate. Mangrove barks were shown to contain chlorophyll b with 396 nm wavelength at peak I, 665 nm wavelength at peak II, with 2.804 absorbance rate. The color fastness test in this study was performed to determine the color quality as a result of the use of various fixers in the fixation processes. The color fastness rate was concluded by RGB value, obtained from Photoshop image data processed through experimental factorial pattern statistics (3x2) using SPSS 1.8. The mean of RGB value found for mangrove leaf and bark-based batik was measured by using Levene’s test, resulting in Sig. 0.46 (>0.05) which indicated that all of the fixation treatments had insignificant impact in color fastness, with wash-sun dry treatment. The mean of RGB data also showed different color fastness rate resulting from the use of each fixer. Mangrove leaf-based dyed batik, fixated with alum under the treatment of wash-air dry, showed a very significant color bleaching. It was suspected that during the fixation process, alum did not allow maximum absorbance of tannin in mangrove by the fabric.

Analysis of the RGB data values of mangrove leaf-based natural dye colors resulted, with wash-iron dry treatment, showed a Levene’s sig. 0.30 (>0.005). This indicated that there was no significant color bleaching in the use of this dye with all fixation methods under the aforementioned fabric treatment. The use of different fixers also resulted in different color fastness rate, with limestone fixation showing the most sensitive towards color bleaching in mangrove leaf-based dyed fabric under wash-iron dry treatment, and alum fixation showing the least color fastness in mangrove bark-based dyed fabric under similar treatment. These results were possibly caused by the intense heat applied during the iron dry treatment, which prevented color bond between the tannin and the fabric from properly forming.

The color fastness analysis of mangrove leaf and bark-based natural dye, each fixated with alum and limestone showed significantly low color fastness under three treatments (wash-air dry, wash-iron dry, and wash-sun dry). Low color fastness rate is attributed to the severance of bond between the acrosome and the fabric, significantly lowering the dye absorbance rate of the fabric, resulting in surface coloring [8]. On the other hand, the usage of
lotus as a fixer resulted better color fastness for the end product, since lotus increased the color fastness rate of the dyed fabric by locking the dye molecules with the fabric. This proved that the contents of mangrove leaves and barks were viable as fabric natural dyeing materials. Tannin, which is capable of forming strong bonds with fabrics, contributed to the best results in this study. Tannin as a compound falls into mordant coloring agent, which combines easily with metal oxides to form lasting dye. These mordant compounds possess good capacity in forming bonds between the fabrics and the dye used, increasing the affinity of the dye to the fabric. The fixation process performed after the dyeing process is finished, is meant to reinforce the dye to the fabric, which will make better lasting colors.

The treatment which caused the most significant color bleaching found in this study for all mangrove-based dye with all fixation methods was the wash-sun dry treatment. It was suspected that sunlight posed significantly adverse impact towards color stabilization during the drying process. The impact of sunlight toward the pigment of red yeast rice, in which lowered colorization intensity was observed due to the damage in active pigments [14]. This decline of color intensity was represented by lower spectrum absorbance rate from the kinetic energy caused by the damage. Ultraviolet light and the energy from sun exposure attacked chains of color molecules and could cause breakdown in the bonds. The damage in color molecule chain contributed to lower color fastness rate because the color carrier cluster in the fabric became inactive. The most significant factor contributing to the color fastness in the molecule stability of the dye material under sunlight exposure.

4. CONCLUSIONS

Based on the research, it can be concluded as follows: The content of pigment in mangrove leaves and barks by spectrophotometer UV-Vis indicated the existence of chlorophyll a and b. In mangrove leaves, it was indicated the content of chlorophyll a, while in mangrove bark it indicated the content of chlorophyll b. Fabrics fixated with alum and limestone showed resulted in the least color fastness, after three post fixation treatments of wash the content of chlorophyll a, while in mangrove bark it indicated the content of chlorophyll b. Fabrics fixated spectrophotometer UV for determination color fastness in the molecule stability of the dye material.

5. REFERENCES

Hydrochemical Study of Groundwater Quality in Jepara Coastal Plain and Lowland

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Abstract – Jepara coastal plain and lowland are located along Java Sea in the Java Island which is selected a appropriate field to study about hydrochemical of groundwater quality in the unconfined aquifer. The primary purpose of the study is to assess the groundwater quality of unconfined aquifer. Physical characteristic of groundwater such as pH, Electrical Conductivity/EC, Salinity, and Dissolved Oxygen/DO value will explain to the actual condition of groundwater quality. It will then combine with the assessment of major cations and anions content. Moreover Piper diagram provides more detail explanation about groundwater facies in the study area. There were around thirty-five (35) groundwater depth measurement and analyze physical properties while around twenty-eight (28) of them were analyzed the major cations and anions content to define groundwater facies. Water table flows from the mountain in the southeast to coastal plan and lowland in the northwest. Some physical characteristics of groundwater in the unconfined aquifer show pH around 5.0-7.3, the maximum EC up to 1,614 μS/cm in Bandengan (close to the coast), salinity in the range 0-5.2 mg/L as well as DO from 0.1 to 2.1 mg/L. The water type is alkaline water with predominantly hydrogen bicarbonate and chloride. Meanwhile Sodium is the most cation content while Chloride indicates the highest anion contents.

1. INTRODUCTION

Java Island as the highest percentage of urban growth in Indonesia occurs in the coastal cities [1]. The increasing population in the coastal cities affects an enormous stress on groundwater as the most precious natural resources. Some environmental problems related to the groundwater exploitation in the recent years are seawater intrusion, degradation of groundwater quality, lowering groundwater level, flooding, and subsidence. Jepara coastal plain and lowland are located along Java Sea in the Java Island which is selected a appropriate field to study about hydrochemical study of the groundwater quality in unconfined aquifer. Population are increased from 1.1 million in 2010 to 1.2 million in 2015 or around 1.5% of annual population growth rate [2]. To fulfill daily need of fresh water, groundwater is withdrawn via dug wells in the unconfined aquifer. The primary purpose of the study is to assess the groundwater quality of unconfined aquifer. Physical characteristic of groundwater such as pH, Electrical Conductivity/EC, Salinity, and Dissolved Oxygen/DO value will explain to the actual condition of groundwater quality. It will then combine with the assessment of major cations and anions content. Moreover the Piper diagram provides more detail explanation about groundwater facies in the study area.

Regionally, stratigraphy of Jepara coastal plain and lowland from young to old consist of Alluvium (Qa) and Muria tuff (Qvtm). Alluvium is spread along coastal plain that consists of unconsolidated material from gravel to clay. Meanwhile, Muria tuff is found from the slope of Muria Mountain in the southeast to the lowland area in northwest of Jepara [3].

2. METHODS

Hydrogeological mapping was the basis to conduct some hydrogeological characteristics by measuring groundwater depth and collecting groundwater samples via dug wells as shown in Figure 1. There were around thirty-five (35) groundwater depth measurement and analyses physical properties of groundwater quality such pH, EC, Salinity, and DO by using the water level meters and the WTW Handheld of pH (3210) meters, Conductivity and salinity (3110) meters, and Oxygen (3310) meters. Indeed, around twenty-eight (28) groundwater samples were analyzed the major cations (Ca²⁺, Mg²⁺, Na⁺, and K⁺) and anions (Cl⁻, HCO₃⁻, and SO₄²⁻) content to define groundwater facies.
3. RESULTS AND DISCUSSION

People in Jepara coastal plain and lowland utilize dug wells to fulfill daily need of fresh water. Groundwater is abstracted via dug wells in the unconfined aquifer. The groundwater depth has a range from 0 to 14 m depth based on the measurement in the hydrogeological mapping. The maximum depth is found in Bondo village (Bangsri sub-district), SG-13 (Figure 1), while the groundwater depth in Telukawur village (Tahunan sub-district), SG-09, is approaching the ground surface. Groundwater flows from the mountain in the southeast to coastal plan and lowland in the northwest. Groundwater levels start from close to the ground surface up to 36 m above sea level (Figure 2).

Physical analyses of groundwater samples conduct pH around 5.0-7.3 (Figure 2b). The minimum value of pH is located in Kalaman village (Kembang sub-district), SG-34, while sample SG-29 in Mororejo village (Mlonggo sub-district) provide the maximum of pH value. The average of pH is 6.2 indicate the groundwater samples in the range of standard for water drinking based on the regulation of Ministry of Health the Republic of Indonesia [4]. Meanwhile, conductivity value shows that the highest value is found in Bandengan village (SG-31) as shown in Figure 2c. It is up to 1.614 μS/cm. This sample is located close to the coast. The highest value may be caused by the interaction between fresh and seawater. Indeed, the salinity value is in the range 0-5.2 mg/L. The highest salinity is found in sample SG-01 that is located in Bulu village (Jepara Kota sub-district) in the city center. Salinity in the city center to the southwest shows higher value than other region that is indicated by tight contour (Figure 2d). Moreover, DO values are from 0.1 to 2.1 mg/L (Figure 2e). DO are effected by the source, temperature of water, treatment and chemical or biological processes in the distribution system [5].

Based on the major cations and anions content analyses, ion sodium (Na$^+$) has the highest cations content (Table 1). It is up to 2,454 mg/L while Mg$^{2+}$ shows the minimum cations content. Sodium is highly soluble chemical element and naturally found in groundwater. The location of this sample is close to the coastline. Meanwhile, Chloride ion (Cl$^-$) is the highest anion contents in groundwater samples up to 3,176 mg/L. Sodium and chloride are the most abundant ions commence in the atmosphere above the oceans, and atmospheric deposition of chloride and sodium from the oceans is the highest along coastal areas [6] as occurs in Jepara coastal plain. It obviously indicates that salt-water intrusion into wells in coastal plain area affect elevated sodium and chloride levels in groundwater. To identify the groundwater facies, the Piper diagram [7] describes the electrical equivalent percentage (meq%) composition of different ions (Figure 2f). The major cations concentration in groundwater are in the decreasing order as Na$^+$ > Mg$^{2+}$ > Ca$^{2+}$ > K$^+$ while anions concentration are in the
decreasing order as $\text{HCO}_3^->\text{Cl}^->\text{SO}_4^{2-}$. The water type is alkaline water with predominantly hydrogen bicarbonate and chloride.

![Figure 2](image)

Figure 2 Result of hydrogeological mapping including physical and chemical analyses of groundwater samples. Groundwater flows from southeast to northwest (a), pH around 5.0-7.3 (b), conductivity value is up to 1,614 $\mu$S/cm (c), Salinity in the range 0-5.2 mg/L (d), as well as DO around 0.1-2.1 mg/L (e), water type is alkaline water with predominantly hydrogen bicarbonate and chloride (f).

Table 1 Summary statistics of major ions content

<table>
<thead>
<tr>
<th>Ion</th>
<th>$\text{Na}^+$ (mg/L)</th>
<th>$\text{K}^+$ (mg/L)</th>
<th>$\text{Ca}^{2+}$ (mg/L)</th>
<th>$\text{Mg}^{2+}$ (mg/L)</th>
<th>$\text{Cl}^-$ (mg/L)</th>
<th>$\text{HCO}_3^-$ (mg/L)</th>
<th>$\text{SO}_4^{2-}$ (mg/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Min</td>
<td>2.25</td>
<td>2.00</td>
<td>5.63</td>
<td>1.95</td>
<td>4.50</td>
<td>54.1</td>
<td>0.10</td>
</tr>
<tr>
<td>Ave</td>
<td>320</td>
<td>16.7</td>
<td>40.1</td>
<td>42.4</td>
<td>308</td>
<td>432</td>
<td>8.3</td>
</tr>
<tr>
<td>Max</td>
<td>2,454</td>
<td>94</td>
<td>354</td>
<td>313</td>
<td>3,176</td>
<td>985</td>
<td>32.3</td>
</tr>
<tr>
<td>SD</td>
<td>615</td>
<td>19.3</td>
<td>64.1</td>
<td>64.7</td>
<td>753</td>
<td>254</td>
<td>8.58</td>
</tr>
</tbody>
</table>

Note: Min: Minimum; Ave: Average; Max: Maximum, SD: Standard Deviation

4. CONCLUSIONS

Groundwater plays role in fundamental aspect for water drinking. Dug wells spread in the most Jepara coastal plain and lowland area which abstract groundwater in the unconfined aquifer. Hydrogeological mapping result that groundwater flows from the mountain in the southeast to the coastal plan and lowland in the northwest. Physical parameters of groundwater conduct that pH has a range of around 5.0-7.3 and the maximum EC is up to 1,614 $\mu$S/cm. Salinity is in between from 0 to 5.2 mg/L, while DO levels are 0.1-2.1 mg/L. Chemical analyses of cations and anion content result that the water type is alkaline water with predominantly hydrogen bicarbonate and chloride. Salt-water intrusion into wells in coastal plain area affect elevated sodium and chloride levels in groundwater samples.

5. REFERENCES


Application of 2-D Finite Element Model to Determine Channel Embankment Design

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Abstract – Tanggok River floodplain area is situated in Gresik Regency, within the development of JIIPE or Java Integrated Industrial and Port Estate. It is considered as high-value place as the Grand Estate Marina City/ GEM City is developed in this area. The average floodplain elevation is +2.5m whereas the water surface elevation/ WSE at observed maximum discharge during the highest tidal flood is 2.84. This made the area is prone to inundation at least for 3 to 6 hours due to the mixed, predominantly semi-diurnal tide type. A steady state condition was chosen to observe the current patterns and WSE in Tanggok River. The simulation resulted Tanggok River segment which across the GEM City development area has an average WSE between +2.79m to +2.88m. This value means that the area is prone to the inundation with the depth of 0.4m. Current magnitude of the river is the safety standard between 0.3 to 0.4 m/s. The important elevation in this design are: maximum embankment elevation +3.50m; design WSE +2.84m; and minimum elevation for the drainage system must be located at +2.90m to assure the run-off could be drained during the maximum condition.

1. INTRODUCTION

JIIPE is the only integrated industrial estate which will have its own railway, toll road connection, and deep sea port located in Gresik Regency, East Java Province. GEM city is designed as the first development of JIIPE complex, serve as central business district/ CBD area [1]. It is located in Tanggok River and Mireng River floodplain. The average floodplain elevation is +2.5m whereas WSE is 2.84m at observed maximum discharge during the highest tide flood. Tide type in the study location is categorized as mixed, predominantly semi-diurnal tide. It means the floodplain are prone to inundation at least for 3 to 6-hours retention time during the peak flood condition. Hydrodynamic analysis is urgently needed to understand the circulation patterns and WSE. Ascertaining the magnitude of vector current and WSE is paramount in designing river structures, in this case, is channel dimension. The objective of this analysis is to observe how changes in flood discharge and tidal WSE in Tanggok and Mireng River system affect current patterns and WSE in water bodies which related to the safety of the developed area.

2. METHODS

2.1 Modelling Approach

The numerical simulations of the Tanggok and Mireng River were performed using the Resources Management Associated-RMA 2, a 1D/2D hydrodynamic model using the finite element method. RMA2 was maintained by the US Army Corps of Engineers (USACE) Engineering Resources Development Center (ERDC), the writer of the program is Ian King from Bringham Young University [2]. RMA2 is a 2D-depth averaged finite element hydrodynamic (FEM) numerical model. It computes horizontal velocity and WSE components for subcritical at free-surface 2D flow fields. It computes a FEM solution of the Reynolds form of the Navier-Stokes equations for turbulent flows. Friction is calculated with the Manning’s or Chezy equation, and eddy viscosity coefficients are used to define turbulence characteristics. RMA2 operates under the hydrostatic assumption; meaning accelerations in the vertical direction are negligible. RMA2 computes the WSE and horizontal velocity on the node point within a FEM which represent studied water body such as: river, estuary, reservoir, or port basin. Governing equations in RMA2 were depth-integrated equations of mass conservation and momentum equation [3].

Mass Equation

\[ \frac{\partial H}{\partial t} + H \left( \frac{\partial U}{\partial x} + \frac{\partial V}{\partial y} \right) + U \frac{\partial H}{\partial x} + V \frac{\partial H}{\partial y} = 0 \]

Momentum Conservation Equation on x-axis
\[
H \frac{\partial U}{\partial x} + HU \frac{\partial U}{\partial x} + HV \frac{\partial U}{\partial y} = 2H \omega V \sin \phi - \frac{\partial H}{\partial x} + \frac{\partial^2 H}{\partial x^2} + \frac{\partial^2 H}{\partial y^2} + KW^2 \cos \phi - \frac{gH \left( \frac{\partial^2 U}{\partial x^2} + \frac{\partial^2 U}{\partial y^2} \right)}{R^2} (U^2 + V^2)^{1/2}
\]

Momentum Conservation Equation on y-axis

\[
H \frac{\partial V}{\partial x} + HU \frac{\partial V}{\partial x} + HV \frac{\partial V}{\partial y} = 2H \omega U \sin \phi - \frac{\partial H}{\partial y} + \frac{\partial^2 H}{\partial x^2} + \frac{\partial^2 H}{\partial y^2} + KW^2 \sin \phi - \frac{gH \left( \frac{\partial^2 V}{\partial x^2} + \frac{\partial^2 V}{\partial y^2} \right)}{R^2} (U^2 + V^2)^{1/2}
\]

Where \( h \) is water depth, \((u,v)\) the local velocity in Cartesian coordinate, \((x,y,t)\) Cartesian coordinate and time, \( \rho \) is density of fluid, \( E \) is eddy coefficient, for \( xx \) is normal direction on x-axis surface \( yy \) is normal direction on y-axis surface and for \( xy \) and \( yx \) is shear direction on each surface, \( g \) acceleration due to gravity, \( an \) elevation of bottom, \( n \) Manning’s roughness \( n \)-value, \( 1.486 \) is the conversion from SI (metric) to non-SI units, \( \zeta \) empirical wind shear coefficient, \( V \) wind speed, \( \psi \) wind direction, \( \omega \) rate of earth’s angular rotation, and \( \Phi \) local latitude.

2.2 Hydrodynamic Modelling

Bathymetry of study site was used as water depth input, it is considered as the primary simulation data. It is a local sounding which cover the north-south area of mixed lands use plan (Fig 1.) Model area was formed in semicircular domain boundary at the downstream with a total number of elements in triangular form of 9551 with 21124 nodes; it is quadratic element type. Primary tidal data used in this model was a tide measured in Manyar, Gresik in 07° 05' 53.8" latitude and 112° 37' 43.9" longitude for 15 days from November 11th 2012. Least square method was used to analyze the tidal record. It was shown that the tide is mixed, predominantly semi-diurnal with Formzhal value of 2.38. The specified flowrate of Tanggok and Mireng River was used in two flow types, maximum discharge and minimum discharge (Table 1.) Simulation was conducted at steady state condition with river flow as the upstream boundary condition and the tidal WSE as the downstream condition [4].

There are two conditions to approach the objective of this study: Scenario A with maximum discharge and highest flood tide; and Scenario B with minimum discharge and highest flood tide. These scenarios were chosen to evaluate the embankment design in extreme condition for GEM City drainage planning. Further drainage design must consider the tidal and backwater effect on Tanggok and Mireng River. Numerical domain is started from Tanggok upstream at UTM/ Universal Transverse Mercator coordinate X= 673684.73 and Y = 92174717.45.

<table>
<thead>
<tr>
<th>River</th>
<th>Minimum Discharge (m³/second)</th>
<th>Maximum Discharge (m³/second)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tanggok</td>
<td>13.75</td>
<td>19.2</td>
</tr>
<tr>
<td>Mireng</td>
<td>50</td>
<td>70</td>
</tr>
</tbody>
</table>
Figure 2. Distance orientation on model compared to study location (x is length of the river measured from datum 0, river upstream)

Figure 3. WSE and velocity vector in every scenario (A and B)
3. RESULTS AND DISCUSSION

The monitoring of simulation was conducted mainly in Tanggok River. As can be seen in Fig 2, the main focus of the study is in yellow circle area. Tanggok River and intersection between Tanggok River downstream and Mireng River also interesting location to be look for the phenomenon. Embankment is one of the river structure which is main and the most important to protect lives and people’s welfare to the inundation caused by flooding. The structure chosen for the study site is earth fill embankment. Due to the behavior of that type of embankment, so that it must fulfill the criteria such as the embankment must be able to retain water pressure, it has to be stable, and the crest must above the extreme water level [5].

As can be seen in Fig 4. and Fig 5. the scenario A WSE graph shown the maximum WSE is in 2.88m and the lowest in +2.81m whereas scenario B graph shown the maximum WSE is in 2.83m and the lowest in +2.79m. It can be concluded that the area is prone to inundation as height as 0.4m. In order to protect the surroundings, the maximum elevation of +2.9m was chosen as the basis of design. Moreover, with the observation length to 3,856m the WSE difference from x=om to x=3,856m computed as 0.2m the elevation basis of +2.9m is reasonable. Current magnitude of the river is the safety standard between 0.3 to 0.4 m/s. The embankment typical dimension is tailored to the design discharge and computed water level. Based on Indonesian standard or SNI, the embankment must have a freeboard of 0.6m and berm width of 2.0m, the berm slope is chosen as 1:2. Because of the WSE design is in +2.90m, drainage bed elevation must be placed in minimum elevation of +2.90m to guarantee the developed area is clear of the inundation.

4. CONCLUSIONS

Simulation was conducted at steady state condition with river flow as the upstream boundary condition and the WSE as the downstream condition. The simulation resulted Tanggok River segment which across the GEM City development area has an average WSE between +2.79m to +2.88m. This value means that the area is prone to inundation with the depth of 0.4m. Current magnitude of the river is the safety standard between 0.3 to 0.4 m/s. The embankment must reach the following requirement, such as: 0.6m freeboard; embankment width of 2.0m; berm slope of 1:2. The drainage bed elevation must be placed in minimum elevation of +2.90m to guarantee the developed area is clear of the inundation.

5. REFERENCES

Examination of Coffee Pulp Waste for Medium in Cellulase Production by Aspergillus Species

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Abstract – An isolate, identified as Aspergillus species can grow well by introducing coffee pulp medium as carbon and nitrogen source in solid state fermentation without any nutrient added. This genus optimum produced cellulase in five days of fermentation at 37°C. Purification, gave the yield 0.13% and 477 fold. The enzyme stable at pH 3.5 to 6.5 and below 55°C, while the optimum activity at pH 5 and 50°C respectively. It is suggested that coffee pulp waste can be used as a cheap medium for cellulase production.

1. INTRODUCTION

Indonesia as the world's fourth largest exporter of coffee, so necessary to adopt a careful control and integrated waste either in the process of coffee production. In the coffee processing, 45% of the polysaccharides-rich coffee pulp were produced. In Indonesia, a huge amount of coffee pulp (CP) and estimated at least 81 million tons were released annually. Research proved that CP waste can be utilized as a material for bioethanol [1] [2]. It was reported that CP can be utilized as substrate in production of protease, xylanase, and endoglucanase production [3] [4] [5]. Other investigations, CP can be modified as absorbent, activated carbon [6] [7], and another industrial purpose as well [8] [9]. However, microbial research utilization of these potential wastes is still limited and less attention. A promising strategy through microbial utilization of CP to produce enzyme cellulase is one of environmentally friendly and possibly to increase the economic value of this biomass.

2. METHODS

1.1 Optimizing and Harvesting of Crude Cellulase Production of Aspergillus sp. through Solid State Fermentation of Coffee Pulp

Ten gram of water saturated CP in 500 mL flask was sterilized, inoculated with Aspergillus sp., and then incubated at 30°C. To optimize cellulase production, the activity crude cellulase was daily examined. To harvest crude cellulase was done by 1% NaCl extraction on 200 ml water, containing 0.01% natrium azide (v/v), followed by shaking 120rpm at room temperature for 12 hours. To recover crude cellulase from suspension, filtration using 40µm glass filter was done. Then to remove remaining cells or debris from crude cellulase filtrate, the centrifugation at 8000rpm for 10 minutes. The supernatant was dialyzed using PS MidiKros Filter Modules, 10 kD against 20 mM acetate buffer pH 5. Solution as a crude cellulase was then stored at 4°C till used for examination of cellulase activity. To produce large scale crude cellulase, 100gr of CP was prepared in 5L flask. And for recovery of crude cellulase, the same previous procedure steps were used.

1.2 Examination of Crude Cellulase Activity

The activity crude cellulase was examined by measuring of reducing sugar against 1% carboxy methyl cellulose (CMC) during hydrolysis. The CMC substrate solution was prepared in 20 mM acetate buffer pH 5. The reaction mixtures of 50µL crude cellulase and 100µL substrate were homogenized and incubated at 37°C for an hour. Quantification of reducing sugar released, the method of Somogy-Nelson was employed [10] [11].

1.3 Cellulase Purification

Purification was done in 25°C room temperature 20mM acetate buffer pH 5. All purification steps were described in results and discussion.

1.4 Effect of Temperature and pH on Cellulase Optimum Activity

Optimum Temperature (°C) and pH on purified cellulase activity were examined in a range temperature 30 to 70 °C and pH 3 to 7.5. The pH 3 to 5 and pH 5 to 7.5 acetate and phosphate buffers 20 mM were used. Optimum activity of purified enzyme was measured using Somogy-Nelson method.
1.5 Temperature and pH Effect on Cellulase Stability

Stability of purified cellulase activity on temperature and pH were examined in a range temperature 30 to 70°C and pH 3 to 7.5. The pH 3 to 5 and pH 5 to 7.5 acetate and phosphate buffers 20 mM were used. Stability of cellulase was measured using the same method above.

3. RESULTS AND DISCUSSION

Optimum production of cellulase activity was obtained after 5 days solid state fermentation at room temperature. At that time, observation showed that Aspergillus sp. grow rapidly even though no any nutrient added during fermentation, and some liquefy form appeared in CP medium. As the cultivation time increased, the liquefied forms also increased. Mean, Aspergillus sp. released some extracellular enzymes which possible hydrolysed CP actively. Some research reports that Aspergillus sp. secreted extracellular enzymes in board spectrum [12] [13] which capable utilized biomass as carbon and nitrogen source [8] [14] [15].

Cellulase activity by quantifying on reducing sugar production revealed that gave optimum in 5 days fermentation and no significant increasing on enzyme activity in 6-7 days fermentation as shown in Figur 1. The optimum reducing sugar production was achieved 1.4μg/ml. Base on this results, large scale fermentation was done for crude enzyme source in cellulose purification.

![Figure 1. Optimization of Cellulase Production of Aspergillus sp. in Solid State Fermentation of Coffee Pulp](image1)

Purification of cellulase was started by precipitated of dialyzed crude cellulase in 65% saturated ammonium sulfate and centrifuged at 12000 rpm for 20 minutes. The precipitates were dissolved in buffer. Remaining ammonium sulfate was removed by overnight dialysis against 20mM acetate buffer at 4°C. This solution was loaded onto DEAE Toyopearl 650M anion exchanger open column pre-equilibrated with buffer and eluted with 0–0.5M NaCl linear gradient. Active fractions (Figure 2) were pooled and dialyzed against buffer to remove NaCl. As shown in Figure 2, the active peak which had cellulase activity was subsequently loaded on Pharmacia FPLC (Fast Protein Liquid Chromatography) using DEAE Cellulofine A100 column (anion exchanger). The linear gradient of NaCl from 0.1 M to 0.3 M used. Further step, the active fraction was purified on super dex 75 pg (gel chromatography).

![Figure 2. Purification on DEAE Toyopearl 650M column pre-equilibrated with acetate 20mM pH 5 buffer and eluted with linear gradient 0-0.5 M NaCl](image2)

The purification procedure of cellulase summarized in Table 1 resulted in 0.13% yield, 477 fold purification. The enzyme production through solid state fermentation usually gave a good yield. However, in this study resulted in
lower yields which probably caused by optimal conditions for solid state fermentation has not fulfilled [16]. Some reports described that the production of enzymes under solid state fermentation depends on several factors. Significant increase in cellulase production influenced by increasing in inoculum concentration. Physical factors such as moisture, temperature, and pH are critical factors in solid state fermentation which influence growth, metabolism, biosynthesis and enzyme secretion as well [17] [18] [19].

### Table 1. Cellulase Purification Step

<table>
<thead>
<tr>
<th>Purification Step</th>
<th>Total ABS-280 (unit)</th>
<th>Total Activity (unit)</th>
<th>Spec. act. (unit)/ABS280</th>
<th>Yield (%)</th>
<th>Fold</th>
</tr>
</thead>
<tbody>
<tr>
<td>Crude enzyme</td>
<td>995000</td>
<td>16267</td>
<td>0.016</td>
<td>100</td>
<td>1</td>
</tr>
<tr>
<td>Ammonium sulfate</td>
<td>218000</td>
<td>14587</td>
<td>0.067</td>
<td>21.91</td>
<td>4</td>
</tr>
<tr>
<td>Precipitation</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>DEAE Toyopearl 650M</td>
<td>94035</td>
<td>12997</td>
<td>0.138</td>
<td>9.45</td>
<td>8</td>
</tr>
<tr>
<td>DEAE Cellulofine A100</td>
<td>8976</td>
<td>11980</td>
<td>1.335</td>
<td>0.90</td>
<td>82</td>
</tr>
<tr>
<td>Superdex 75G</td>
<td>1256</td>
<td>9789</td>
<td>7.794</td>
<td>0.13</td>
<td>477</td>
</tr>
</tbody>
</table>

The effect of pH on the cellulase activity was measured after 1-hour incubation at 37°C of each enzyme in 1 ml 1% CMC substrate at various pH values. As shown in Figure 3 (A), purified cellulase exhibited maximum activity at pH 5 and retained nearly 100% activity in a pH range of 3-6.5 after 60 minutes exposure to corresponding pH values. The enzyme had optimum activity at 50°C and is nearly 100% stable below 55°C after 30 minutes exposure to respective temperatures shown in Figure 3 (B).

Figure 3. Effect of pH (A) and Temperature (B) on Activity and Stability of Cellulase

4. CONCLUSIONS

Coffee pulp waste as a medium for the production of cellulase has been proven. This research despite getting a low yield, but the advantage was noted that during cellulase production via solid state fermentation no requirement the addition of any nutrient. Therefore it is necessary to do further research on the optimization of the cellulose based CP production so that the production efficiency will be increased and feasible in industrial scale.

5. ACKNOWLEDGEMENTS

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6. REFERENCES

Decolorization of Black Liquor Using Coagulation-Flocculation and *Trametes versicolor* F200

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**Abstract** – Black liquor is harmful to the aquatic ecosystem if discharged directly into waters because it contains high COD (194.458 mg/L) and high pH (12.89). The purpose of this research was to compare two methods, coagulation-flocculation and combination of coagulation-flocculation and biological method using *Trametes versicolor* F200, to decolorize black liquor. Coagulation-Flocculation process was conducted by using Iron (II) sulfate and polyacrylamide anionic with different ratio (1:1; 1:2; 1:3; 2:1; 2:2; 2:3; 3:1; 3:2 and 3:3 g). The optimum condition for decolorization of black liquor using coagulants and flocculants was obtained in ratio 3:3. It has able to decolorize black liquor 68 %, reduced COD 20.450 mg/L, and decreased pH 5.09. Further, the decolorization of black liquor using combination method of coagulation-flocculation and *Trametes versicolor* F200 on 10th days was 77 % and resulted COD 19.033 mg/L, and pH 3.43. This fungus has able to decolorize black liquor because it contained ligninolytic enzymes such as LiP, MnP, and Lac at 1959, 15, 137 U/L, respectively. Further, mycelial mass of fungus and concentration of glucose during decolorization were 0.1203 g and 36245.45 ppm, respectively.

1. INTRODUCTION

The utilization of oil palm empty fruit bunches (OPEFB) to produce bioethanol consist of three processes: pretreatment, hydrolysis, and fermentation. The pretreatment process is needed to break lignin and hemicellulose bonding. The one method for pretreatment process is usage of Sodium Hydroxide (NaOH). This process generates high amounts of black liquor wastewater. Bioethanol from 600 kg of OPEFB produced 3000 liters of black liquor [1]. The black color is mainly due to degradation process of lignocellulose, and it is an indirect measurement of the amount of lignin compound in the effluent. Lignin is a complex heterogeneous phenyl propanoid biopolymer containing a diverse array of stable carbon-carbon bonds with aryl/alkyl ether linkages and may be cross-linked to hemicellulose [2]. Black liquor was harmful to the aquatic ecosystem such as fish, plankton, and daphnia if it discharges directly into waters [3,4] because it contains high COD (194.458 mg/L) and high pH (12.89). Black liquor contains organic compounds 78 % and inorganic compounds 22 % [5]. Lignin is the biggest content in black liquor. Black liquor wastewater has been minimized in many technologies such as acidification [6], coagulation-flocculation using photocatalysis [7], poly aluminium chloride [8], membrane filtration [9], and biological method by *Schizophyllum commune* [10], *Trametes elegans* [11], and *Trametes versicolor* U97 [8,12]. White-rot fungi (WRF) were used to decolorize black liquor because the fungus secrete one or more enzymes such as *Lignin peroxidase* (LiP), *Manganese peroxidase* (MnP) and *Laccase* (Lac) [13]. The purpose of this research was to compare two methods, coagulation-flocculation using Iron (II) sulfate and polyacrylamide anionic and combination coagulation-flocculation and biological method using *Trametes versicolor* F200, to decolorize black liquor.

2. METHODS

2.1 Chemicals

The chemical used in this research were black liquor, *Trametes versicolor* F200 from InaCC Indonesian Institute of Sciences, Potato Dextrose Agar (PDA), Iron (II) sulfate, polyacrylamide anionic, glucose, KH2PO4, K2HPO4, CuSO4, MnCl2, Na2MoO4, FeCl3, ZnCl2, thiamine hydrochloride, 2,6-dimethoxyphenol (2,6-DMP), malonate buffer, MnSO4, H2O2, LiP buffer, syringaldazine, sodium acid buffer, phenol, sulphuric acid, digestion liquid, and potassium bromide (KBr).

2.2 Procedures

2.2.1 Decolorization of Black Liquor using Coagulation-Flocculation

Coagulation-Flocculation process was conducted by using Iron (II) sulfate and polyacrylamide anionic with different ratio (1:1; 1:2; 1:3; 2:1; 2:2; 2:3; 3:1; 3:2 and 3:3 g). The coagulant was mixed with diluted black liquor
(1:1 %v/v) for decolorization, three minutes by using Jar Test Mixer at 200 rpm and then it was mixed with the flocculant for eight minutes at 50 rpm. Total suspended solid (TSS), chemical oxygen demand (COD), and pH before and after decolorization were also measured [14].

### 2.2.2 Decolorization of Coagulated Black Liquor and *Trametes versicolor* F200

*Trametes versicolor* F200 was maintained on potato dextrose agar (PDA) at 25°C for 7 days. It was inoculated in 100 ml liquid medium in erlenmeyer flask and pre-incubated for 7 days under a static condition at 25°C [8]. After the culture period, the extracellular fluid was blended and added to coagulated black liquor and then incubated for 3, 7, 10, 14 and 20 days. After incubation, the sample was filtrated to get supernatant that measured for mycelial mass of fungus. The filtrate was measured for enzyme activity (LiP, MnP, Lac) [15,16,17], decolorization of black liquor, chemical oxygen demand (COD) [14], and concentration of glucose [13].

### 3. RESULTS AND DISCUSSION

#### 3.1 Decolorization of Black Liquor using Coagulation-Flocculation

The optimum condition for decolorization of black liquor using coagulants and flocculants optimum was obtained in ratio 3:3. It has able to decolorize black liquor 68 %, reduced COD 20.450 mg/L and decrease pH 5.09 (Fig 1 and Fig 2).

![Figure 1](image1.png)  
**Figure 1.** Effect of coagulant-flocculant mass for decolorization and TSS efficiency black liquor

![Figure 2](image2.png)  
**Figure 2.** Effect of coagulant-flocculant mass for COD and pH black liquor

Most of the suspended particles in wastewater carry a negative charge in an aqueous medium, so that added inorganic metal salts as coagulant and flocculant will hydrolyze rapidly in wastewater to form cationic species, which are adsorbed by negative charged colloidal particles, resulting in simultaneous surface charge reduction and formation of micro-flocks. [18]. The addition of high-dose coagulant would make decreasing of pH effluent, because the iron (II) sulfate in the water will be hydrolyzed and forming an acid, according to the reaction:

\[
\text{FeSO}_4 + 3 \text{H}_2\text{O} \rightarrow \text{Fe(OH)}_3 + 3 \text{H}^+ + \text{SO}_4^{2-}
\]

This research showed that the increasing coagulant dose makes the floc easy to be precipitated, so the decolorization and TSS will be increased. Increasing the coagulant and flocculant ratio caused decreasing COD because needs increasing oxygen as oxidizing the wastewater. Characterization of black liquor by FTIR described below (Fig 3).

![Figure 3](image3.png)  
**Figure 3.** Characterization of Black Liquor by FTIR
Characterization using FTIR showed the aromatic group (C-H) at wave number 877 cm\(^{-1}\), The alkene group (C-H) at wave number 1427 cm\(^{-1}\), the aromatic group (C=C) at wave number 1585 cm\(^{-1}\). The hydroxyl group phenol (O-H) at wave number 2283 cm\(^{-1}\) and the hydroxyl group carboxylic acid (O-H) at wave number 3464 cm\(^{-1}\).

3.2 Decolorization of Coagulated Black Liquor and *Trametes versicolor* F200

Based on data from procedure 3.1, the optimum coagulant: flocculant dose is 3:3 ratio, after that used *Trametes versicolor* F200. The result showed that decolorization of black liquor using combination method of coagulation-flocculation and *Trametes versicolor* F200 on 10\(^{th}\) days was 77 % and resulted COD 19.033 mg/L, and pH 3.43 (Fig 4 and 5). Lignin is the biggest compound in black liquor [5], although lignin units are relatively simple molecules provided with a phenyl-propane backbone, the tri-dimensional structure of the resulting polymer is very complex and non-repeating, a feature with important consequences for the biological method. Only extracellular enzyme can access the highly variable polymer surface. In addition, the diversity of the lignin interunit bonds prevents their specific cleavage by the enzyme. Only highly sophisticated organisms are able to achieve this object. Of these, filamentous fungi and particularly white rot fungi are the most efficient for lignin degradation [19]. *Trametes versicolor* F200 is one of the white rot fungi which is used in this research. The larger LiP enzyme activity than MnP and Laccase showed that LiP play role as the main enzyme for decolorization of black liquor. The oxidation of lignin involves efficient biocatalysts that use strong oxidant as electron acceptors, for example, H\(_2\)O\(_2\) and dioxygen. Besides, small molecules known as redox mediators have to be oxidized first to obtain a high oxidant-to-reductant ratio and it turns oxidize aromatic structures in lignin. Another advantage of these small molecules is their diffusibility in the lignocellulosic structure. The LiP is stronger oxidant than MnP [20]. The LiP enzyme activity until 10 days has been increasing, while after 10 days, have been decreasing.

The LiP is the initiator and enzymes take responsibility for the breakdown of lignin inside the black liquor. The LiP has the ability to oxidize aromatic compounds in lignin. It is believed that this action is done by catalyzing the transfer of an oxygen atom from a hydrogen peroxide molecule to an aromatic ring and resulting a radical cation which spontaneous causing reaction with nucleofil and oxygen, and enzyme process breaking C-C and C-O bond in lignin. [21]. The mechanism of lignin degradation by LiP enzyme is described in Fig 6. The fungus has a different growth period, where the metabolic activity has several phases of its growth. In the beginning phase, the growth phase and the metabolic activity will be declined after fungi growth passed the peak phase, it was called the lower phase. That phase has a significant impact to produce an enzyme by fungi used to decolorize black liquor. Fig 7 showed that concentration of glucose have been increasing until 7\(^{th}\) days incubation (36245.45 ppm) due to the fungus using the wastewater as the nutrient source, while after that the concentration of glucose was decreased because the fungus needs more nutrient source from glucose to decolorize black liquor.
4. CONCLUSIONS

The optimum condition for decolorization of black liquor using coagulants and flocculants was obtained in ratio 3:3. It has able to decolorize black liquor 68%. Increasing of coagulant dose makes the flocs easy to be precipitated so that the decolorization will be increased. Decolorization of black liquor using combination method of coagulation-flocculation and *Trametes versicolor* F200 on 10th days was 77%. The best method for decolorizing black liquor in this research was the combination coagulation-flocculation and *Trametes versicolor* F200.

5. ACKNOWLEDGEMENT

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6. REFERENCES

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Detection of Organic and Inorganic Waste Anomaly in Homogeneous Soil Using Electrical Resistivity Tomography Method Wenner β-Schlumberger Configuration

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Abstract – In the subsurface exploration investigations there are many methods used, one of them is Electrical Resistivity Tomography (ERT). ERT method can measure the electrical properties of the material below the earth surface based on resistivity value of the material by injecting electric current and measure the potential at the surface. Based on the data obtained then will be inputted into RES2DINV software for final processing of 2D image. This research is a laboratory scale, to test the use of Wenner β-Schlumberger configuration in particular to detect the presence of anomalies in a homogeneous soil. In the test, the system will be made a wooden box (size 120 cm x 30 cm x 20 cm) containing homogeneous soil. In addition, the system will be given two anomalies; those are organic waste anomaly which has been made by clumping the rotten of leaves and inorganic waste anomaly which has been made by clumping the drying thin of plastics. All anomalies will be placed in different position in the system. The result shows that organic waste anomaly has the resistivity value is smaller than resistivity value of inorganic waste anomaly. The differentiation of both resistivity value is caused by the presence of water content which contained in both anomalies. In the begining of making process of all anomalies, inorganic waste anomaly has been made by clumping the rotten of leaves. It means, the leaves is in moist condition and has water content more larger than inorganic waste anomaly that has been made by the drying thin of plastic. Based on the references, material that has excessive of water content has small of resistivity value. In addition, the result also shows that all anomalies can be detected in the system.

Keywords: ERT, Wenner β-Schlumberger configuration, Anomaly, RES2DINV

1. INTRODUCTION

In the field of imaging method, Electrical Resistivity Tomography (ERT) is one of a method that is widely used in exploration field especially subsurface exploration [1,2]. The measurement of ERT method is based on the value of resistivity of the material by injecting electric current and measure the potential at the surface [3,4,5].

Based on the research before by Yunus (2016) has been developed the configuration of ERT. The development is Wenner β-Schlumberger configuration for detecting wood, stone, and wet soil anomaly [6,7]. In this research is a laboratory scale that makes a development further from the research before. The development was varying the kind of anomalies that will be detected; those anomalies are organic waste and inorganic waste.

The result shows that Wenner β-Schlumberger configuration can detect The differentiation of resistivity value and position of all anomalies (organic waste and inorganic waste). Based on this research, if we look at the simple physics concept that is used, hoping in the future can be referenced for experimental activity in school or College. In addition, in the future can be references for researchers that want to exploring and detecting the waste on underground.

2. METHODS

2.1 ERT for Under Ground Detection

ERT method is an imaging method that can measure the electrical properties of the material below the surface that are based on the value of the resistivity of the material by detecting from the surface [8]. ERT is performed by injecting an electric current to a pairs electrodes and measuring the potential data to the other pairs electrodes [9,10]. From the results of current and potential measurements then can be obtained resistance value each layer below the measuring point [11].
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Figure 1. Potential Around current injecting point on Earth’s surface [12]

The distribution of current is spread out over hemispherical surface with an area $2\pi r^2$, so that:

$$J = \alpha E.$$  \hfill (1)

$$J = \frac{I}{A}; \quad \sigma = \frac{1}{\rho}; \quad E = \frac{V}{r}$$  \hfill (2)

$$V_r = \frac{\rho I}{2\pi r}.$$  \hfill (3)

$\rho$ resistivity of the surface (Ohm-m), $V$ is electric potential in distance $r$ (Volt), $I$ is electric current supply (A), $J$ electric current density (A/m$^2$), $\sigma$ is conductivity of the system (Ohm$^{-1}$m$^{-1}$), and $E$ is an electric field (V/m).

2.2 Wenner $\beta$-Schlumberger

Wenner $\beta$-Schlumberger configuration is the configuration which has spacing pattern rules. The electrodes $C_2$-$C_1$ (current electrodes) and electrode $P_1$-$P_2$ (potential electrodes) placed respectively at the same distance of $n\alpha$. While the electrodes $C_1$-$P_1$ placed at a distance $\alpha$ (as shown in Figure 3).

Figure 3. Wenner $\beta$-Schlumberger configuration [6]

$$V = \frac{\rho I}{2\pi} \left( \frac{1}{R_1} - \frac{1}{R_2} \right).$$  \hfill (4)

By substituting the value of electrodes distance,

$$\Delta V = V_1 - V_2.$$  \hfill (5)

$$\Delta V = \frac{\rho I}{\pi} \frac{n^2}{a(n+1)(2n+1)}.$$  \hfill (6)

$$\Delta V = \frac{\rho I}{K}.$$  \hfill (7)

$\rho$ is resistivity (Ohm-m), $\Delta V$ is electric potential between electrodes $P_1$-$P_2$ (Volt), $I$ electric current between electrodes $C_1$-$C_2$, dan $K$ geometric factor (m).

2.3 Procedures

This research is a laboratory scale that has been created in Laboratory of Physics, University of Palangka Raya. In the test, will be made a wooden box system (size 120 cm x 30 cm x 20 cm) that containing homogeneous soil. Then, will be used Wenner $\beta$-Schlumberger configurations by measuring range is 100 cm and the smallest spacing value is 4 cm ($\alpha = 4$ cm). This measurement has been created by moving the potential electrode at the
constant current electrodes, then the electrode displacement currents on the space followed by the displacement of the potential electrode along the system up to that last point of measurement in the system [6,7].

In the test, have been made two different systems. First system is homogeneous soil or system without anomaly and second system is non-homogeneous soil or system with anomalies. The anomalies that used in this research are organic anomaly and inorganic anomaly. Organic anomaly which has been made by clumping the leaves that was drying and decomposed to spherical form with diameter 11 cm. And inorganic waste anomaly which has been made by clumping thin plastics to tube form with length 11 cm and diameter 11 cm. In this research, all anomalies are placed with depth 5 cm from the surface. In addition, all anomalies are placed in different position. Organic waste anomaly was placed in position 32 cm, and inorganic waste anomaly was placed in position 75 cm.

3. RESULTS AND DISCUSSION

Figure 6 describes the 2D image of system without anomalies with resistivity value measured in the range of 73.3 Ωm-204 Ωm. Based on the figure can be seen that the bottom of the system detected a small resistivity, it indicates the distribution of water seeping into the bottom of the system.

Figure 7 describes the 2D image of system with anomalies. The results obtained that the range of resistivity values measured between 58.3 Ωm-252 Ωm. This Change happened because the anomalies exist in the system. Clearly, the inorganic waste anomaly detected in position range 22 cm-38 cm with a range of resistivity value between 224 Ωm-252 Ωm, while the organic waste anomaly in the position range 60 cm-72 cm with a range of resistivity values between 18.7 Ωm-26.7 Ωm. Based on these results can be seen that the differences of all anomalies resistivity value can be detected as organic waste anomaly that has the smallest resistivity values, and...
inorganic waste anomaly has the largest resistivity value. The differentiation of both resistivity value caused by water content that more larger distributed in organic waste anomaly, so that make resistivity value on it become smaller. Surely in this study also contained an error value in the detection of 9.6 % for the system without anomalies and 7.9 % for systems with anomalies. This error value can be referenced for researchers in the stage of analyzing the data and accuracy of detection of real position and real resistivity.

Figure 7. Result of the 2D image with anomalies by Wenner-Schlumberger configuration

4. CONCLUSIONS

Based on the results that by variated two different kind anomalies (organic waste and inorganic waste), shows that Wenner β-Schlumberger configuration can detect all of the anomalies are exist in the system. In addition, the differentiation of resistivity value for both anomalies is also can be detected which as organic waste anomaly that has the smallest resistivity values, and inorganic waste anomaly has the largest resistivity value. The differentiation of both resistivity value caused by water content that more larger distributed in organic waste anomaly, so that make resistivity value on it become smaller. Surely in this study also contained an error value in the detection of 9.6 % for the system without anomalies and 7.9 % for systems with anomalies. This error value will be referenced for researchers in the stage of analyzing the data and accuracy of detection of real position and real resistivity.

5. REFERENCES

[6]. P. Yunus, 2016, Indonesia: Bogor Agricultural University.
TRMM Data Correction using Semiparametric Model

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Abstract – The observation rainfall data needed for forecasting are not always completely available. This problem could be corrected based on the rainfall satellite data from Tropical Rainfall Measuring Mission (TRMM) which is available with spatial and temporal high resolution. However, this satellite data cannot be used directly to forecast rainfall. This paper concerns with the correction of TRMM data using PCA and semiparametric modeling so the corrected data can be used to substitute the observation data. The result is compared to the result of transfer function model (Silmi, 2015) based on the values of AIC, correlation (r), and RMSEP. The results of quadratic semiparametric model with six knots (AIC=1394; r=0.87; RMSEP=77) and transfer function model (AIC=1155; r=0.86; RMSEP=76) were relatively similar. However, the first model is preferred. The process of semiparametric modeling was faster than that of the transfer function modeling.

Keywords: Semiparametric, Penalized Spline, Linear Mixed Model

1. INTRODUCTION

One of climate phenomenon is rainfall. This data can be viewed through the stations. Sometimes, the data generated from the stations are needed very quickly, but the data are not always completely available. Those problems can be overcome by using Tropical Rainfall Measuring Mission (TRMM) satellite data. Data of the TRMM satellite is a grid data derived from the imaginary rectangular forms. So, TRMM satellite data expected to have high correlation and autocorrelation and we need an appropriate statistical analysis method to solve the problem. Validation and correction of TRMM data modeling are necessary to see the results of the estimation accuracy of the rainfall observations at the stations. To validate and make correction to TRMM data, [4] used geometric and linear models and [3] used transfer function with principal component analysis. Another method which can be potentially used for the same purposes is semiparametric modeling. This model have been used in statistical down-scaling to predict rainfall and gave good results [2]. This research concerns with the use of semiparametric model to TRMM satellite data correction.

2. METHODS

2.1 Data

This research used the rainfall data (mm) in Sukadana Station (rainfall station in Indramayu District) as dependent variables and TRMM Satellite data (mm) as independent variables. Each of these data is monthly data from January 1998 to December 2008. The size of TRMM domain used in this research was 4 × 4 square grid (0.25° × 0.25° for each grid) from 108.125° to 108.375° East longitude and from 6.25° to 6.375° South latitude above the area of Sukadana station. The grids of TRMM data as independent variables are grid7, grid8, grid12, and grid13 (Fig. 1) with the altitude of satellite orbit of 403×10^3 m.

![Figure 1 Position of TRMM grids](image-url)
2.2 Methods

The steps of the analysis in this research were:
1. Identifying multicollinearity in TRMM data based on the value of variance inflation factor (VIF) which is more than 10 and the problem is overcome by principal component analysis (PCA).
2. Determining the pattern of functional relationship between rainfall and TRMM data by plotting rainfall with the component score resulted from PCA in the first step. The plots are built based on various degrees of freedom to fit the data pattern. The optimum degrees of freedom is determined using the GCV (generalized cross validation) minimum criterion.
3. Generating spline basis, which includes:
   a. Determine the number of knots
      For example, there is a p-spline models:
      \[ f(x) = \beta_0 + \beta_1 x + \cdots + \beta_p x^p + \sum_{k=1}^{q} \beta_{j+k}(x - K_k)^{p} \]
      where \( \beta = (\beta_0, \ldots, \beta_p, u_{p1}, \ldots, u_{pq}) \) is a spline regression coefficient vector, \( p \geq 1 \) is a positive integer, \( (w)^{p} = v^{p}I(w \geq 0) \) is truncate basis function, and \( K_1 < \cdots < K_q \) is fixed knot \([1]\). The model in equation (1) has \( m \) degrees of freedom and \( q \) knots. Thus, the number of knots in the model can be formulated as:
      \[ q = m - v = m - (p + 1) \]
      where \( q \) is the number of knots, \( m \) is the number degrees of freedom the smoothing spline, \( v \) is the number of model parameters, and \( p \) is the degree of truncate basis function. The distance between knots can determined using formula:
      \[ s = n/(q + 1) \]
      where \( s \) is the distance between the nodes, \( n \) is the number of observations and \( q \) is the number of knots.
   b. Generate truncate power basis function for penalized spline. Formula for this function is
      \( (x - K_k)^{p} \) if \( x \geq K_k \)
      \( 0 \) if \( x < K_k \)
      where \( x \) is independent variable, \( K_k \) is the k-th knots on the independent variable, and \( p \) is the highest rank penalized spline model.
4. Estimating the parameters based on linear mixed model
   Parameter estimation is based on linear mixed model which needs the following two design matrices \( X \) and \( Z \) according to equation (1).
   \[
   X = \begin{bmatrix}
   1 & x_1 & \cdots & x_1^p \\
   \vdots & \vdots & \ddots & \vdots \\
   1 & x_n & \cdots & x_n^p 
   \end{bmatrix}
   \]
   \[
   Z = \begin{bmatrix}
   (x_1 - K_1)^p & \cdots & (x_1 - K_q)^p \\
   \vdots & \ddots & \vdots \\
   (x_n - K_1)^p & \cdots & (x_n - K_q)^p 
   \end{bmatrix}
   \]
5. Comparing the result of semiparametric model to that of the transfer function model (Silmi, 2015) based on the values of AIC, correlation (r), and RMSEP (Root Mean Square Error of Prediction).

3. RESULTS AND DISCUSSION

Based on VIF values, all grids of TRMM data are with VIF more than 10. It indicates that there are multicollinearity problem in this data. Therefore, it required the dimension reduction of this data. The result of PCA suggests that one principal component (PC1) captures 97% proportion of total variance of 4 predictor variables. The pattern identification of relation between rainfall as response variable and PC1 as predictor variable was determined by selection the optimum degrees of freedom with GCV minimum. The result shows that the optimum degrees of freedom was eight and the pattern of relation between rainfall and PC1 is showed in Fig. 2.
Based on AIC, correlation (r), and RMSEP values we concluded that the quadratic semiparametric model with six knots was the most preferred model. This model was used to predict rainfall one year ahead and Fig. 3 compared the predicted and actual rainfall.

The results of semiparametric models and transfer function models were relatively similar. However, the process of semiparametric modeling was faster than that of transfer function modeling. The comparison between semiparametric model and transfer function model is presented in Table 1.

<table>
<thead>
<tr>
<th></th>
<th>semiparametric model</th>
<th>transfer function model</th>
</tr>
</thead>
<tbody>
<tr>
<td>AIC</td>
<td>1393.70</td>
<td>1154.20</td>
</tr>
<tr>
<td>R</td>
<td>0.87</td>
<td>0.86</td>
</tr>
<tr>
<td>RMSEP</td>
<td>76.60</td>
<td>75.47</td>
</tr>
</tbody>
</table>

4. CONCLUSIONS

The quadratic semiparametric model with six knots was the most preferred model. Semiparametric models and transfer function models gave relatively similar results. However, the semiparametric modeling process was faster than the transfer function modeling process.

5. REFERENCES

Simulation on the Effect of the Cover Soil And the Starting Time of the Landfill Operation To Predict the Leachate Quality in the Landfill Site

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Abstract – The degradation model was used as an appropriate method to predict the leachate quality generated from the landfill site. This model was applied to the Supit Urang Landfill site in Malang Municipality, Indonesia. This model was used with assumptions and approaches that considered the landfill conditions and climate of a tropical area.

The biodegradation model that predicts the leachate quality uses the assumptions of the dissolution and degradation model by developing relationships based on the first-order rate equation. In the model calculation, three cover soil conditions were considered to evaluate the influence of a cover soil on the leachate quality and quantity in the landfill. Four cases simulations for the time the waste dumping started were considered to show the effect of variations in the climatic conditions on the prediction of leachate quantity.

The prediction of the leachate quality showed that the effect of the application of cover soil was not that significant for the leachate concentration at the bottom of the landfill. The time that the waste dumping started had no significant influence on the leachate concentration at the bottom of the landfill, only the timing of the leachate production was different. The results of the leachate quality prediction can be summarized and used as the basis for starting a waste dumping operation.

1. INTRODUCTION

Leachate in landfills is considered to be a factor of environmental risk and represents a key consideration during the design and operation of a landfill. The factors affecting the chemical composition and the production rate of leachate include the characteristics of the waste (initial composition, particle size, density and so on). The other factors are an interaction between the percolating landfill moisture, the hydrology and climate of the site, the landfill design and the operational variables, and microbial processes. The entire factors are taken place during the stabilization of the waste, and the stage of the landfill stabilization.[1]

It is widely accepted that moisture content is important in controlling the decomposition of waste within landfill sites and that the rate of decomposition generally reduces with a reduction in moisture content. Moisture content affects landfill decomposition through a variety of different ways. Water is the medium through which biochemical reactions take place, including the microbiological decomposition of refuse.

In the case of landfill process modeling, the significance of local factors such as the waste composition, disposal method and protection systems against potential impacts, the heterogeneity of the medium, as well as the several physical and biochemical phenomena that need to be considered (meteorology, liquid and gas movement, biological and chemical degradation of the waste, aging of materials), makes the development of models applicable to different landfill facilities difficult.

Several computer programmers for estimating leachate generation have been developed, for example: Hydrology Evaluation Leachate Performance (HELP), FULLFILL, and SOILINER. All of them are based on the so-called Water Balance Method (WBM) developed by the U.S. Environment Protection Agency. [2][3]

The aim of this paper is to make the model for prediction of leachate quality from the landfill and to evaluate the effect of the cover soil on leachate generation pattern and the effect of beginning of landfill operation time on leachate generation pattern. The combination degradation model in this paper is carried out in the tropical condition and the open dumping landfill site from the beginning of the waste placed. The influence of the water content, the depth of evaporation, field capacity, design and operation of landfill that were appropriate with the tropical climate and landfill condition was used as the consideration of the model.
2. METHODS

2.1 Assumption of the BOD degradation in the waste

The combination of water balance and degradation model is applied as the method that appropriates with the leachate quantity and quality generated from the landfill site. This model is applied at the Supit Urang Landfill site in Malang Municipality, Indonesia. This model is applied with some assumption and approaching that considering the condition landfill and climate of a tropic area.

The BOD biodegradation model that derives the leachate quality uses assumptions of the dissolution and degradation Model with the developing the relationships based on the first-order rate equation as follow:

\[ \Delta C_j = C_{in, j} \times P_{in, j} - C_{out, j} \times P_{out, j} \]

Where:
- \( C_j \) = the concentration in the cell at time \( t_j \) (mg/l)
- \( C_{in, j} \) = the concentration that enter the cell from the cell above at \( t_j \) (mg/l)
- \( C_{out, j} \) = the concentration that leave the cell to the cell below at \( t_j \) (mg/l)
- \( P_{in, j} \) = the discharge that infiltrate to the cell from the cell above at \( t_j \) (l)
- \( P_{out, j} \) = the discharge that leave the cell to the cell below at \( t_j \) (l)

The \( \Delta L_j \) and \( \Delta C_j \) was the dissolution and degradation of BOD in material amount that occurred in the cell at \( t_j \).

The Dissolution of BOD in the Cell was calculated as follow:

\[ L = L_0 \times e^{-k_L \times t} \]

Where:
- \( L \) = initial weight of contain BOD in the waste (kg – BOD / kg – waste)
- \( L_0 \) = weight of contain BOD in the waste after process dissolution occur (kg – BOD / kg – waste)

And the degradation process was formed as follow:

\[ C = C_0 \times e^{-k_C \times t} \]

Where:
- \( C \) = initial concentration in the cell (mg/l)
- \( C_0 \) = concentration in the waste after process degradation occur (mg/l)

2.2 Procedures

In the calculation of model, three conditions of cover soil were considered because the model wants to evaluate the influence of cover soil to the leachate quality and quantity in a landfill. The case 1 is an application of landfill without cover soil; it is called fully open dumping, which is a usual manner in Indonesia and other developing countries. Case 2 is application of cover soil in the top of waste layer that implemented at 10 days after the waste first time dumped. Case 3 is intermediate cover soil; cover soil was applied in the middle of waste dumped, implemented at 5 days after the first layer of waste dumped. In case 3, the cover soil is also implemented in the top of a waste layer.

Three case simulations for time of starting dumping the waste were considered to show the variation of the climate condition to the prediction of leachate quantity. Case 1 the starting time is in the beginning of wet season. The case 2 is started in the middle of wet season. While the case 3 is started in the beginning of dry season and case 4 was implemented in the middle of dry season.
3. RESULTS AND DISCUSSION

The biodegradation process in the waste depends on the condition of water content. According to the Chapter 3, the water content of waste below 50%, biodegradation slightly slowly occurred. Since the water content in the waste of the landfill was influenced by the infiltration water from the rainfall, the biodegradation process in the waste was directly related to the process of infiltration that occurred in the landfill. In the upper layer of the landfill, the evaporation was influence the condition of water content. In the dry season, the water content decrease and become zero in the end of the season. The degradation in that period was not occurred.

The result of leachate quality prediction showed the effect of the application cover soil in the landfill site not so significant for leachate concentration in the bottom of the landfill. The top cover soil and intermediate cover soil application will prevent the evaporation from the waste. The water content was stable in that condition. The stable condition prepared the suitable condition for continue of degradation. The BOD concentration in the bottom of the landfill site was not so different, only the time start to degrade was delayed in case of the start in the middle dry season of waste dumped. In the condition of without cover soil, in the upper layer the water content was low and the degradation was terminated. However, the influence of evaporation only occurred in the top layer, and the degradation occurred in the layer beneath of 0.5 meter.

The time to start to dump the waste had no significant influence to the leachate concentration in the bottom of the landfill, only the time occurrence of the leachate concentration in first time was different. The landfill without cover the in the middle wet season and in the beginning of dry season had similar trend and time of concentration increase. The condition occurs because the water still available to provide the condition of degradation process. In the beginning of wet season the increasing of leachate concentration was delayed until 50 days because in that time, amount rainfall still low and not sufficient for increase the water content of 50%. In the middle of dry season the delay increased around 300 days. The delay of increasing the leachate was caused by insufficient available infiltration water from the rainfall to support the process of degradation.

Some consideration can be summarized from the result of the leachate quality prediction that can be used as the base of the beginning of waste dumped operation. Since the application of the top cover soil and intermediate cover had similar pattern of degradation compare with the open dumping landfill, the application of top cover soil and intermediate cover can be implemented in the beginning of dry season and in the middle of wet season. The application of intermediate cover was not recommended to apply in the beginning of wet season, because the effect of degradation was low when it was applied. In the middle of dry season, to provide the condition that suitable for degradation process, the implementation of top cover soil can be delayed until the rainfall came. The condition of BOD concentration was same with the conditions without cover soil. And the intermediate cover soil was not recommended to apply, because it will delay the degradation process for the long time.

![Figure 2](image2.png)

**Figure 2** Leachate Quality at the bottom of landfill in the Beginning of Wet Season

![Figure 3](image3.png)

**Figure 3** Leachate Quality at the bottom of landfill in the Middle of Wet Season
4. CONCLUSIONS

The combination of degradation model in this paper was carried out in landfill site from the beginning of the waste placed. Some conclusion were derived from the model to comply leachate generation due to the condition of climate in the tropical area, especially Indonesian condition.

In case of the effect of cover application, it will cause in the delay of time of rising BOD concentration for case of beginning of dry season. Time delay application of intermediate cover is 100 days compared with the application without soil cover. For the other case, the effect of cover application did not cause in the delay. There was no difference in maximum BOD concentration between no-cover and cover application in any dumping time.

Since the leachate treatment plant capacity was depend on the leachate quantity and quality, it was necessary to consider the type of landfill (without cover soil, top cover soil, or intermediate cover soil landfill). The time of starting to start dump the waste was one of consideration factor. The combination of the application the cover soil and timing to start dump the waste will produce the different pattern of leachate quantity and quality. The plan of the leachate treatment plant must be considering the application of the cover soil and timing to start dump in order determine the appropriate dimension and capacity plan.

5. REFERENCES


A Model of Multi-Stage Water Allocation for Estimating the Irrigated Crop Production

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Abstract – The problem with the evaluation and optimization of irrigation crop production scheme is how to estimate the harvest yield at the end of the cropping season based on the input values of the irrigation water supply distributed along the season time window. In this article, a model of multi-stage water allocation for estimating the irrigated crop production is presented is discussed. This mathematical function will take water allocation time series as the input and produce the rate of harvest as the output. The applications of the model have been conducted on various optimization schemes of reservoir operation, using Dynamic Programming and Stochastic Simulation. The overall performance of the model can be said as being satisfactory.

1. INTRODUCTION

Principally, optimization can be applied when the number of available resources intended to be allocated are less than the maximum possible number of allocations of resources. Accordingly, the problem of optimization can be summarized as how to allocate these available resources so that the overall benefit is maximum (or the overall cost is minimum). In the optimization of multi-stage irrigation water allocation during an entire cropping season for maximizing the irrigated crop production at the end of the season, a function is needed for expressing the amount of irrigated crop production. This function of crop production will take in the values of multi-stage irrigation water allocation during the cropping season as the input of decision variables. One such example is the Jensen’s Equation [1] which relates the water stress to yield. In this article, a practical model for estimating the irrigated crop production with the time series of water allocation as the input is discussed.

This model of multi-stage water allocation for estimating the irrigated crop production has been introduced as an article in a journal in 2007 [2]. The model since then has been applied for optimization in the various reservoir operation and water allocation models using dynamic programming and stochastic simulation. The model has been precisely calibrated for a various number of stages. The model also has been subjected to verification study using field and experimental data. This article will discuss the progress of the model so far and examine its development in the future.

2. METHODS

2.1 Construction of the model

The basic idea for the model is the general form of relationship between applied water and crop yield [3] as can be seen in the Fig.1.

![Figure 1. The Original Curve.](image)

In the original curve, the relationship between the Relative Water Application $AWr$ (range 0 → 1) and the Relative Crop Production $Yr$ (range 0 → 1) is the manifestation of the reality that the Water Application of zero will result in the Production of zero, and the Water Application of 100% will result in the Production of 100%. But this model means that the entire cropping season is represented by just one stage. To expand this relationship into a multi-stage cropping season, the Product Function comes into the scene as in Eq.(1).
\[ Y_{ri} = Y_{r1} \times Y_{r2} \times Y_{r3} \times Y_{r4} \times \cdots \times Y_{rn} \]  

(1)

\[ Y_{ri} = AWR_i \rightarrow [AWR_i]^e \rightarrow [\sin (AWR_i)^e \times \Pi/2)]^e \rightarrow \sin [AWR_i \times \sin (AWR_i, 2\Pi) \times a - \sin (AWR_i, 2\Pi) \times b] \times e^{(c \times d)} \]  

(2-5)

\[ Y_{ri} = \sin (AWR_i \times \sin (AWR_i, 2\Pi) - b) \times (1 - b \sin (AWR_i, \Pi))^e \times e^{(c \times d)} \]  

(6)

Figure 2. Steps for fitting the Original Curve.

There are five steps (equations) for fitting the shape of the Original Curve:

1. It begins with the simplest equation Eq.(2) of \( Y_{ri} = AWR_i \) resulting in CURVE-1 with reverse curvature.
2. To flip up the reversed curvature, a power coefficient of \( e = 0.03 \) is added, resulting in CURVE-2.
3. To move the hump upward, a sine function and a coefficient of \( d \) are introduced, resulting in CURVE-3.
4. To straighten the lower curve, the second sine function and a coefficient of \( a \) are added, resulting in CURVE-4.
5. The final step is by adding the third sine function and two more coefficients of \( b \) and \( c \). The resulting CURVE-5 is quite similar with the Original Curve. At this point, the values of \( a = 0.06, b = 0.25, c = 1.3, d = 0.15 \), and \( e = 0.99 \).

Hence the proper model of multi-stage water allocation for estimating the irrigated crop production are the Eq.(6) and Eq.(1). The input for the model are the rate of irrigation water allocation at every stages (AWr). The output is the rate of irrigated crop production (Yr). To get at the real crop production (in ton or monetary unit), the resulting Yr is to be multiplied with the standard (maximum) crop production.

### 2.2 Calibration of the model

The coefficient of \( a, b, c, d, \) and \( e \) of the model have been calibrated for various number of stages in a cropping season. The procedure is to minimize the sum of square error of Yr using ten points in the AWR axis. The results are presented in the Table 1.

<table>
<thead>
<tr>
<th>Coefficients of the Model</th>
<th>4</th>
<th>5</th>
<th>6</th>
<th>7</th>
<th>8</th>
<th>12</th>
<th>24</th>
<th>36</th>
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<tbody>
<tr>
<td>a</td>
<td>0.119420</td>
<td>0.119419</td>
<td>0.119422</td>
<td>0.119419</td>
<td>0.119424</td>
<td>0.119419</td>
<td>0.119415</td>
<td></td>
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<tr>
<td>b</td>
<td>0.241071</td>
<td>0.241065</td>
<td>0.241088</td>
<td>0.241064</td>
<td>0.241066</td>
<td>0.241119</td>
<td>0.241108</td>
<td>0.240991</td>
</tr>
<tr>
<td>c</td>
<td>2.233352</td>
<td>2.233401</td>
<td>2.233180</td>
<td>2.233408</td>
<td>2.233391</td>
<td>2.232846</td>
<td>2.232924</td>
<td>2.234148</td>
</tr>
<tr>
<td>d</td>
<td>0.090427</td>
<td>0.090425</td>
<td>0.090436</td>
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<tr>
<td>e</td>
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<td>2.552648</td>
<td>2.233610</td>
<td>1.488346</td>
<td>0.744481</td>
<td>0.496490</td>
</tr>
</tbody>
</table>

The results shows that for various number of stages, all coefficients are practically constant except of \( e \) which become smaller as the number of stages become larger.
2.3 Verification

The verification study has been done in 2010 using aggregate data of agriculture and hydrology from the USA [4]. The agricultural data consist of the crop production in the irrigated and non-irrigated areas, and these data have been obtained from the website of USDA. The hydrologic data consist of the amount of precipitation in the crop areas, and these data have been obtained from the website of NOAA. These data have been used to perform a statistical test to see if there is any significant difference between $Y_r$ values from the model and from reality.

<table>
<thead>
<tr>
<th>STATE</th>
<th>CROPS FOR VERIFICATION</th>
<th>CORN</th>
<th>COTTON</th>
<th>SORGHUM</th>
<th>SOYBEAN</th>
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</thead>
<tbody>
<tr>
<td>Sample size</td>
<td>Colorado</td>
<td>266</td>
<td>Arkansas</td>
<td>Colorado</td>
<td>Arkansas</td>
</tr>
<tr>
<td>$Y_{r\text{average}}$ (model/real)</td>
<td>0.2634 0.2500</td>
<td>0.2500 0.7064</td>
<td>0.7984 0.4436</td>
<td>0.4389 0.7103</td>
<td>0.7012 0.9840</td>
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<tr>
<td>Z-score</td>
<td>1.605 8.392</td>
<td>0.278</td>
<td>0.840</td>
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<tr>
<td>Sample size</td>
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<td>221</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>$Y_{r\text{average}}$ (model/real)</td>
<td>0.5157 0.5090</td>
<td>0.7242 0.7911</td>
<td>0.6405 0.6362</td>
<td>0.6866 0.6635</td>
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</tr>
<tr>
<td>Z-score</td>
<td>0.658 5.366</td>
<td>0.491</td>
<td>1.671</td>
<td></td>
<td></td>
</tr>
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<td>Sample size</td>
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<tr>
<td>$Y_{r\text{average}}$ (model/real)</td>
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<td>0.7204 0.7837</td>
<td>0.6463 0.6582</td>
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<tr>
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<tr>
<td>$Y_{r\text{average}}$ (model/real)</td>
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<td>1.521 1.797</td>
<td>0.659</td>
<td>0.154</td>
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<td></td>
</tr>
</tbody>
</table>

The test shows that in three crops (corn, sorghum, and soybean) there is no significant difference between the model and the reality. Even for cotton, the values of average $Y_r$ have no big difference. Therefore, the model is valid for various kinds of crops with a various start of planting.

The verification study also has been done in 2011 using the data from experiment with corn and rice [5]. The corn and rice were grown up under the canopy of transparent plastic sheets so the water application can be controlled and recorded. At the end of the cropping season, the crop yields were measured accurately. The results show that specifically the model can be stated to be valid for various water applications (irrigation + precipitation) during the cropping season for various crops.

2.4 Application

The model of multi-stage water allocation for estimating the irrigated crop production can easily fit to various schemes of irrigated crops. For example, the rice’s 4 month long cropping season can be divided into 8 stages (½ monthly) or 12 stages (½ monthly). Therefore it can be directly implemented to one cropping season dynamic programming model. In this case the usual “+” algebra operator in the recursive equation is replaced by the “x” operator. For optimization of a single reservoir, dynamic programming is good because of the sequential decision making and the ability for dealing with non-linearity [6]. A model of stochastic dynamic programming has been developed for stream water quality management incorporating a fuzzy decision model [7].

For the simulation model, the required input of AWR, series can be simulated, in a deterministic or stochastic way. The model then will calculate the resulting rate of crop production $Y_r$. To obtain the real crop production (in ton or monetary unit), the $Y_r$ must be multiplied with the standard (maximum) crop production. The resulting simulated values of $Y_r$ can be used for the purpose of optimization or evaluation.

The standard (maximum) crop production is the real production when the crop receives the standard (full) water supply, while others factors are normal. It may be not readily available, but it can be obtained statistically by collecting the available record of crop production. If the record if long enough, then the maximum value of production can represent the standard crop production.

The simulation model can incorporate the well-known methods such as GA (Genetic Algorithm) or SA (Simulated Annealing). The GA and SA are both belong to the stochastic simulation for optimization methods. For example, the Genetic Algorithm model has been applied as the optimization technique for modeling an optimal operation strategy of an irrigation reservoir dealing with various crops [8].

Table 2. Statistical Test for Verification of the Model.
3. RESULTS AND DISCUSSION

There are some points about the model which can be discussed here.

1. The model of Multi-Stage Water Allocation here is consist of two functions. The first function is the AWri function which has three sine functions in it. These sine functions are for the adjustment of the resulting Yr-AWri curve to be similar with the shape of the original curve. The second function is the product function of Yr, which is simply the multiplication (product) of all Yr from each stage.

2. This model of Multi-Stage Water Allocation has been proposed in 2007 (in an Indonesian Language journal article). At the time the proposed name is Model Sinus-Perkalian (in Indonesian) or Sine-Product Model (in English). However, it seems that the term of Sine-Product has also been used for some mathematical terms.

3. The model is well fit into the Dynamic Programming optimization model since both have the same structure of multi-stage. The model can also be used properly with the simulation model, where the series of simulated water application can be inputted into the model to produce the simulated rate of crop production Yr.

4. For a certain amount of irrigation water, the Yr is at the maximum when the AWri is distributed homogeneously during the entire cropping season. This is in accordance with the real practice to rotate the insufficient irrigation water homogeneously among the irrigated crops.

5. When just one of the AWri is 0, then the final Yr is also 0. It is like when the irrigation water supply is failed in just one stage (½ month for example) in the cropping season, then the resulting harvest of the season is also failed. It may be true, but it needs some retrospections before can be implemented.

6. For a certain number of stage, the current model has a uniform (identical) AWri function. But in reality, the value of the function’s parameters can be varied between stages. It can be subjected for research studies in the future.

4. CONCLUSIONS

The conclusions which can be drawn so far are:

1. The estimation of irrigated crop production by using the model of Multi-Stage Water Allocation will give realistic values of Yr, although probably not quite the accurate ones.

2. The model of Multi-Stage Water Allocation can be applied well in the optimization model of Dynamic Programming and Simulation.

3. The model of Multi-Stage Water Allocation is still open to further investigation especially in the area of (1) Inter-Stages Variation in the Awri function, (2) Variation in the time width of stages, and (3) Minimum Limit of Irrigation Water Application.

5. REFERENCES


Analysis Of Strategy Transportation Demand Management
To Solve Traffic Congestion In Cilegon City

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Abstract – Traffic congestion in some cities in Indonesia such as Jakarta, Surabaya, Medan, Bandung and Yogyakarta is getting worse. Many studies conducted in order to resolve the congestion problem by improving the supply side in response to the development of demand side. Unfortunately, this approach is only effective for a short time interval only and does not resolve comprehensively. The approach is starting to be done by many cities in Indonesia to manage traffic congestion is Transportation Demand Management (TDM). TDM, through a variety of strategies, provide a solution to the problem of urban transport by departing from the demand side.

Cilegon City has implemented several strategies TDM to slightly unravel the existing congestion problems. Some programs that have been implemented is Car Free Day, Ridesharing, pedestrian improvement, bus routes and parking pricing.

1. INTRODUCTION

Problems of transportation in Indonesia usually arise because of the imbalance between the capacity of transport facility (supply side) with the movement of people (demand side) is. There is a gap between the addition of the movement of people with the condition of the infrastructure. Perceived result is traffic congestion that often occurs is clearly visible in the form of long queues, delays and noise and air pollution well.

The conventional approach has always been used by urban transport planners and decision-makers is to accommodate any growth in transportation needs in the form of increased capacity and efficiency of the system infrastructure network. Policy development of urban transport infrastructure system in Indonesia using this conventional approach (Predict and provide) provided must be abandoned and replaced with a new approach that Predict and Prevent or fortuneteller and prevent. One way to do is to do business or management on the needs of transportation known to the Transportation Demand Management (TDM).

2. TDM CONCEPT

Transportation Demand Management (TDM) or often called Mobility management is defined by victoria transport policy institute (VTPI, 2009), Columbia are strategies that change the travel behavior (how, when and where people travel) in order to increase the transport system efficiency and Achieve specific planning objectives.

Completion of transportation problems by using conventional approaches propose various policies to improve the transportation infrastructure systems that can accommodate the size of the transportation needs without the slightest consideration the social, environmental and operational needs. But with TDM approach proposed several attempts to minimize or mitigate the transportation needs so that the movement generated still within the boundary condition of social conditions, environmental and operational.

There are three basic concepts of the application of several different TDM strategies for creating more efficient transportation, namely:

1. The concept of how
   This concept encourages people to discourage the use of private vehicles into public transport or changes of single occupancy vehicles (SOV) to high occupancy vehicles (HOV)

2. The concept of when
   This concept is geared to change the travel time of usage peak-hours toward the off peak hour

3. The concept of where
   Where the concept of focusing on where the destination or route selection on routes that are solid to be applied as road pricing policy that some users are forced to avoid the route.

In other words, the policy pursued in the implementation of the concept of TDM should be able to lead to a shift in some of the impact of a movement in space and time as follows (Tamin, 1999):

- The impact of the time shift
The process of movement occurs in the same location, but at different times

- The impact of the route shift
  The movement occurred at the same time, but on a different route
- The impact of the mode shift
  The process of movement occurs in the same location and time, but on different modes of transportation
- The impact of the destination location shift
  The movement took place on location, time and mode of transportation are the same, but with a different destination location

3. RESULTS AND DISCUSSION

Some common application of TDM strategy and has already been done by other countries with a fairly good success rate (Lanarc, 2009) are as follows:

1. Ride sharing
Ride sharing can reduce the impact of the use of vehicles on the road and on the environment very easily. The trick is to group people who want to travel to the same destination into groups by using one vehicle for each group. This condition will reduce the number of private vehicles on the streets. Ride sharing itself subdivided into two forms of carpooling and vanpooling.

2. Bus Rapid Transit (BRT)
BRT is defined as the transportation of high-quality customer-oriented, which provides urban mobility is fast, convenient and inexpensive (GTZ, 2002). BRT has another name that is well known in some places such as the Bus System Capacity (High-Capacity Bus Systems), System Bus High Quality (High-Quality Bus Systems), Metro Bus, Surface Subway, Bus System Express (Express Bus Systems), and Systems busway.

The characteristics of the BRT (GTZ, 2002):
- Raising and lowering the passengers quickly
- Withdrawal charges efficiently
- Stops and stations were comfortable
- Clean bus technology
- Integration mode
- Identity of modern marketing
- Excellent customer service

Some cities in the world also have a lot to develop strategies BRT to resolve transportation problems such as:
- In Asia: Istambul, Nagoya, Taipei, Jakarta
- In Europe: Bradford, Eindhoven, Ipswich, Leeds
- In latin America: Kuritiba, Bogota, Sao Paulo
- In north America: Ottawa, Los Angeles, Miami, Vancouver
- In Australia: Brisbane, Adelaide

Kuritiba experience in Brazil as one of the best examples that illustrate the application of BRT integration between transport and urban planning. With a total population of 1.5 million, Kuritiba has developed 65 km of busway along five routes and forwarded by the feeder 340 connected to terminals at strategic locations with a total length of 185 km service.

3. Walking and Cycling
Walking and cycling are important in supporting sustainable transportation program. Almost all good travel a short distance and long distance running must have involved activities. Pedestrian design for good walkers certainly makes a more humane city. In many ways, walking and cycling is a good way and ideal for surrounding a town.

The last few decades, European countries have implemented rules that support the NMT (Non-motorized Transportation) program. Copenhagen, Denmark has found the social benefits of the pedestrian. Initially happened a heated discussion before Copenhagen, improve pedestrian and cycling facility (pedestrianization), although in the end the project was declared a success. conducted continuously and increased cycling network, over time the room that had belonged to motorists of traffic is now the space owned by Copenhagen.

Curitiba, Brazil displays one of the many development best pedestrian zones. Pedestrian built in Curitiba adjacent to mass transit locations and homes, also within walking distance of restaurants and other businesses. The city has been combining the zones by means of an efficient bus that can carry more passengers. Integration between mass public transport with pedestrian zones bring many benefits to society, as well as lower transportation costs.
4. **Alternative Work Schedules**

Alternative work schedules are divided into (VTPI, 2009):

- **Flextime**
  Flextime is intended as a convenience to employees so that employees get the flexibility in the arrangement of work time daily. For example, employees generally work from 08:00 until 16:00 pm, with the ease of these employees can work from 7:30 pm until 15:30 pm or 8:30 pm to 16:30 pm.

- **Compressed Workweek (CWW)**
  This condition is defined as an employee can work less hour in one day but longer next week. Suppose that in the first week of an employee working for 40 hours (5 days x 8 hours / day), the employee can add hours of work in one day so having extra time off or reduce working hours in a day so there is an extra day.

- **Staggered Shifts**
  The timing of this shift is intended to reduce the number of employees who come and go at any one time. For example, by giving different time shift, the shift time division may be 08:00 to 16:00 pm, 8:30 to 16:30 pm and 9:00 until 17:00 pm.

Examples of successful implementation of Alternative Work Schedules namely by Transamerica Financial Corporation in Los Angeles. The company has been implementing flextime program since 1974. There are 90% of the approximately 3,700 employees of the company which has been harnessing flextime program. More than two-thirds of employees who use flextime to arrive before 07.30 to avoid congestion downtown and they free themselves from the congestion in the afternoon anyway.

Another example is what was done by the Southern California Association of Government (SCAG). SCAG offer a compressed workweek and the program turned out 95% of employees took advantage of this program. Employees work nine hours a day and get a holiday on his Friday time. The advantage obtained in the form of satisfaction of employees who have an impact on work productivity thus directly also have an impact on company performance.

5. **Telecommuting**

This strategy allows the worker to finish the job at home or elsewhere closer, it means that the process of fulfilling the requirements are not always met with the movement process. The need is information and services can be met by using the mode of telecommunication. The use of email facilities, fax and internet will help reduce the amount of movement due to the process of fulfilling the needs of informational can be done without a person having to move.

Application of TDM strategies in an area closely related to the characteristics of the region and the linkages between each strategy. So election strategy set is not necessarily based on a ranking of judgment or ability to cope with congestion is quite large but need further assessment in the light of the characteristics and relationship strategies.

**USE TDM IN CILEGON**

Some TDM strategy implemented in Cilegon are:

1. **Ride Sharing Application** as one of the strategies TDM via transport employees. Cilegon City is the base of the steel industry, energy, and chemicals in Banten Province. Multi-national corporations with a large number of employees such as PT. Krakatau Steel Group, PT. Indonesia Power and chemical companies largely utilizes the application of ride sharing through employee shuttles to transport its employees.

2. **Implementation of the Car Free Day in Cilegon** is held every Sunday from 06.00 until 10:00 pm housed in the Yasin Beji Street in Cilegon.

3. **Utilization pedestrian path on the South Ring Road Cilegon**. Since the beginning of 2014, on the South Ring Road Cilegon already constructed a pedestrian path with a width of up to 5 meters to accommodate pedestrians. Until this year the construction of the pedestrian path along the South Ring Road continues. Construction of pedestrian path is one part of a TDM strategy called pedestrian improvement.

4. **Bus route** is one application of TDM strategies that held by Cilegon City. Implementation of this program is in the form of bans on public bus to pass the main road of Cilegon City at 06.00 until 18.00 pm.

4. **CONCLUSIONS**

Completion of transportation problems by using conventional approaches (Predict and Prevent) can no longer be used. Cities in Indonesia it time to implement a more comprehensive method and sustainable. Approach through Transportation Demand Management strategy which emphasizes the principle predict and Prevent is the best solution.
the policy pursued in the implementation of the concept of TDM lead to the occurrence of some of the impact that the movement of the time shift, route shift, mode shift and shift the location of the destination.

Some of the major cities in the world have menerap TDM strategies with a pretty good success rate. Therefore it is appropriate to the current city of Cilegon has been able to implement several strategies TDM. Only in its application needs to synergize with other policies to be more effective

5. REFERENCES

Phytoremediation of Chromium Polluted Water Using Water Hyacinth (*Eichhornia crassipes* (Mart.) Solms), Water Lettuce (*Pistia stratiotes* L.), and Water Hyssop (*Bacopa monnieri* L.) in a Simulated Constructed Wetland

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**Abstract** – Tannery factories often discharged chromium-containing wastewater to a nearby river which caused chromium pollution in the river. At high concentration, chromium is known to cause toxic effects to living organisms. There are some alternatives to remediate chromium-containing wastewater. The use of constructed wetland to remediate chromium-containing wastewater has been applied in many countries as a cost-effective alternative for wastewater treatment. In this experiment, we evaluated the efficiency of three aquatic plants i.e., water hyacinth (*Eichhornia crassipes* (Mart.) Solms), water lettuce (*Pistia stratiotes* L.), and water hyssop (*Bacopa monnieri* L.) in removing chromium from the medium. Concentrations of chromium (K₂Cr₂O₇) used in this experiment were 0 (control), 10, 30, and 50 mg/L. The experiment was conducted in a simulated constructed wetland using aquatic plant system, for a period of nine days exposure. Water hyacinth showed the mean tolerant plant to chromium solution as the plant showed healthy after nine days exposure, whilst water lettuce and water hyssop showed chlorosis in their leaves. In terms of relative growth (RG), however, water hyssop showed the highest RG, whilst in terms of bioconcentration factor (BCF), it was water lettuce that showed the highest value. The highest chromium concentration in all plants was obtained on the 9th day of exposure at 50 mg/L Cr. The highest chromium removal efficiency from water (90.23%) was obtained by *E. crassipes* at 10 mg/L, whilst the removal efficiency for *P. stratiotes* and *B. monnieri* were 86.62% and 45.7% respectively. The three species therefore can be categorized as hyperaccumulator for chromium.

Keywords: chromium, phytoremediation, constructed wetland, aquatic plant system, water hyacinth, water lettuce, water hyssop

**1. INTRODUCTION**

Tannery factories often discharged chromium-containing wastewater to a nearby river which caused chromium pollution in the river, such as the case in Sukaregang, Garut, West Java. At high concentration, chromium is known to cause toxic effects to a living organism, and Cr (VI) is known to be carcinogenic [1]. Chromium is a nonessential element for plants. Hence there is no particular mechanism for Cr uptake. Chromium is an inorganic contaminant which must physically be removed from the system or converted into inert forms [2].

There are some alternatives to remediate chromium-containing wastewater. The use of constructed wetland to remediate chromium-containing wastewater has been applied in many countries as a cost-effective alternative for wastewater treatment [3]. We conducted phytoremediation experiment to evaluate the effectiveness of three aquatic plants, namely water hyacinth (*Eichhornia crassipes* (Mart.) Solms), water lettuce (*Pistia stratiotes* L.), and water hyssop (*Bacopa monnieri* L.) in removing chromium from wastewater using constructed wetland (aquatic plant system).

**2. MATERIALS AND METHODS**

Water hyacinth (*E. crassipes*), water lettuce (*P. stratiotes*), and water hyssop (*B. monnieri*) were used in this experiment. Chromium-containing wastewater was made using K₂Cr₂O₇ solution. A simulated constructed wetland was made from a plastic bucket (5 L) filled with soil in the bottom and water as media of an aquatic plant system. Plants were acclimatized and grown in the soil–water system with an addition of 10% (v/v) Hoagland nutrient solution. Plants were treated with 0, 10, 30, and 50 mg/L of K₂Cr₂O₇ solution. Distilled water was added every three days to compensate the water loss through transpiration or evaporation. Plants were
harvested at 3rd, 6th, and 9th day of exposure. Plant samples were washed and rinsed then air dried at room temperature to determine fresh weight. Plant samples were separated into each organ parts (root, shoot, leaf), dried at 105°C to determine dry weight. Parameters measured were relative growth (RG) based on plants fresh weight, chromium concentrations in plants and media. Bioconcentration and translocation factor were also calculated. Plants and water samples were digested using wet ashing digestion method with HNO₃ and 30% H₂O₂[4]. Chromium concentrations were analyzed using Atomic Absorption Spectrophotometer. Relative growth value was calculated using equation 1 [5]. Chromium removal efficiency was calculated using equation 2 [6].

\[ RG = \frac{\text{Final fresh weight (g)}}{\text{Initial fresh weight (g)}} \]  

\[ \% \text{ efficiency} = \frac{[\text{metal}]_{\text{initial}} - [\text{metal}]_{\text{final}}}{[\text{metal}]_{\text{initial}}} \]  

3. RESULTS AND DISCUSSION

3.1 Relative Growth (RG)

All species showed a decrease in RG values compared to control (Figure 1). The decrease in RG value indicates that the toxicity of chromium on the plants increases with increasing chromium concentration. B. monnieri has the highest value of RG (1.016 ± 0.104), while P. stratiotes has the lowest value (0.513 ± 0.076) after nine days exposure to 50 mg/L chromium. Zewge [9] observed that heavy metal can disturb plants organ function which inhibit plants growth. Chlorotic leaves and lack of nutrients uptake by roots were shown as a symptom of growth inhibition. Plants exposed to chromium at high concentrations will decrease its rate of metabolism. Hence, its cell division will be inhibited, therefore, the RG value will decrease [10]. Peralta et al. in Oliveira [11] showed that hexavalent chromium in low concentration would increase the root growth compared to control in Medicago sativa.

![Figure 1. Relative growth values after 9 days of chromium exposure](image)

3.2 Chromium Concentrations

Chromium concentration in plants increased following the increase in chromium concentration in the medium (Figure 2). Other experiments also reported the same tendency [6, 12, 13]. P. stratiotes showed the highest chromium concentration (10.463 ± 3.678 mg/g) after 9 day of exposure to 50 mg/L chromium in the medium. As
a comparison, the highest chromium concentration for *E. crassipes* was $3.389 \pm 1.423$ mg/g and in *B. monnieri* $1.32 \pm 0.215$ mg/g. Satyakala and Jamil [12] reported that *P. stratiotes* that was exposed to chromium for three days at 50 mg/L, its chromium concentrations in root and shoot were 28 mg/g and >10 mg/g respectively. *E. crassipes* reported can accumulate 4.18 mg/g chromium after 42 days exposure [13].

![Figure 2. Chromium concentration in plants in 9 days of chromium exposure](image)

Figure 2. Chromium concentration in plants in 9 days of chromium exposure

![Figure 3. Morphology of the plants after exposure to 50 mg/L Cr within 9 days period (a) *E. crassipes* (b) *P. stratiotes* (c) *B. monnieri*](image)

Figure 3. Morphology of the plants after exposure to 50 mg/L Cr within 9 days period (a) *E. crassipes* (b) *P. stratiotes* (c) *B. monnieri*

A plant can be regarded as hyperaccumulator if it can accumulate more than 1 mg/g of Cu, Co, Cr, Ni, and Pb, or more than 10 mg/g of Fe, Mn, and Zn in its dry matter [14]. The results showed that all three aquatic plants used in this experiment can be categorized as hyperaccumulator for chromium. Although the chromium concentration in *P. stratiotes* was higher than others, however, the morphology of *P. stratiotes* showed signs of wilting and chlorosis due to the toxicity of chromium. Whilst *E. crassipes* showed no such symptoms until the end of this study (Figure 3). Satyakala and Jamil [12] reported that *P. stratiotes* that exposed to chromium at 25 and 50 mg/L showed chlorosis and detachment of leaves. The experiment conducted by Sinha [15] and Shukla [16] using *B. monnieri* showed the same tendency as this experiment along with the same symptoms of chromium toxicity such as, chlorophyll and protein content reduction. Histological analysis conducted by Saikia [17] using 50 mg/L showed chromium accumulation in the cortical tissue with a particular pattern. It concluded as strong reduction in meristematic activity and hypertrophy of cortical parenchyma.
Chromium removal efficiencies by three species tend to increase as exposure time became longer and chromium concentration was increased. The highest chromium removal efficiency was obtained at 9th-day exposure to 10 mg/L by *E. crassipes*. From the previous study [18], it has been observed that chromium removal occurred significantly at the beginning of exposure. It has been proposed that there are two different mechanisms to remove chromium from water. The mechanism that occurs rapidly is a direct chromium adsorption process by roots, whilst the slow mechanism is a reduction of Cr(VI) to Cr(III) which will be precipitated by roots exudates and absorbed by plants.

![Figure 4. Chromium removal efficiency](image)

Chromium concentration in soil tends to increase with increasing concentration of chromium in the medium and longer exposure time (Figure 5). Chromium sedimented on the surface of the soil. It was found that chromium concentrations in soil using *E. crassipes* and *P. stratiotes* was not significantly different. The presence of iron and manganese hydroxides in soil can cause immobilization of chromium in soil [19].
Figure 5. Chromium concentration in soil in 9 days of chromium exposure

Bioconcentration factor (BCF) and translocation factor (TF) were parameters used to assess the ability of plants to tolerate and accumulate heavy metals. A small value of TF indicated that the ability of plants to translocate metals is minimum [20], so most of the metals absorbed by plants accumulated in roots. Water lettuce showed the highest BCF value than others (Table 1). The longer exposure time, the higher BCF values are obtained. Zhou et al. [6] reported that P. stratiotes has a BCF value of 632 with chromium exposure at 10 mg/L on 9th day and E. crassipes was reported to have BCF value of 225 at 10 mg/L after 42nd day of exposure [13]. The results also showed the same tendency as other experiments [6,13] where the BCF value decreased due to increased chromium concentration in the medium. Since the TF values that obtained were > 1, these plants were suitable as a phytostabilization agent. It seems that phytoremediation mechanism that happened in the plants was rhizofiltration.

Table 1. BCF and TF values

<table>
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<tr>
<th>Cr conc. in medium (mg/L)</th>
<th>Exposure time (days)</th>
<th>E. crassipes BCF</th>
<th>TF</th>
<th>P. stratiotes BCF</th>
<th>TF</th>
<th>B. monnieri BCF</th>
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</table>

4. CONCLUSION

The three species can be regarded as hyperaccumulator for chromium. Morphology of E. crassipes showed better tolerance to chromium than two other species. Water hyacinth also showed slightly higher chromium removal efficiency, 90.23%, than the other two species, i.e. 86.62% for P. stratiotes, and 45.7% for B. monnieri.

5. ACKNOWLEDGEMENT

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6. REFERENCES


Decolorization of Black Liquor through Environmentally Friendly Method by *Trametes versicolor* F200

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Abstract – Black liquor is produced from bioethanol pretreatment process. This waste has a high hardness value and difficult to be decomposed. It is harmful to an ecosystem and has high Chemical Oxygen Demand (COD). One way of handling this waste is biodegradation by using fungi. The fungus used in this study was *Trametes Versicolor* F200. This paper aims to compare the ability of *Trametes Versicolor* F200 to decolorize original black liquor (without treatment) and coagulated black liquor. The result showed that decolorization of original black liquor by *Trametes versicolor* F200 was 85.96% and decolorization coagulated black liquor by *Trametes versicolor* F200 was 94.30%.

1. INTRODUCTION

Bioethanol pretreatment process produces black liquor that has high Total Suspended Solid (TSS) and Chemical Oxygen Demand (COD). Black liquor contains 78% organic components and 22% inorganic components, which the organic component contains lignin amount to 37.5% [1]. The black color produced from black liquor is caused by the presence of lignin compounds. Lignin compounds are hydrophobic, have rigid structures and difficult to be decomposed so a few microorganisms that can degrade. Black liquor has potential to pollute the environment, especially harmful for aquatic ecosystems, such as fish, daphnia and plankton [2,3,4].

One of the efforts for handling black liquor is using microorganisms. Fungi are microorganisms which are biodegradable and can secrete oxidative enzymes which can reduce the toxicity of pollutant. White rot fungi (WRF) are effective for degrading lignin compounds contained in black liquor [5]. One fungus belonging to the White-rot fungi is *Trametes Versicolor*. White-rot fungi have three ligninolytic enzymes: lignin peroxidase (LiP), manganese peroxidase (MnP), and laccase [6]. These enzymes are used to degrade the complex structure of lignin. The aims of this study were to determine the ability of *Trametes versicolor* F200 to decolorize black liquor and coagulated black liquor in liquid medium. Further, the enzyme activity of fungus and COD concentration in black liquor were also measured.

2. METHODS

2.1 Chemicals

Materials used in this research was black liquor produced from bioethanol pretreatment process in RC Chemistry LIPI, *Trametes Versicolor* F200 from InaCC Research Center for Biology, Malt Extract Agar (MEA) and other chemicals were obtained from Wako Japan, include: Poly Aluminium Chloride (PAC), polyacrylamide, glucose, KH$_2$PO$_4$, K$_2$HPO$_4$, H$_2$SO$_4$, phenol, 2,6-dimethylphenol (2,6-DMP), a buffer solution LiP, H$_2$O$_2$ buffer, MnSO$_4$, malonate buffer solution, syringal arudazin, sodium acid, and K$_2$Cr$_2$O$_7$.

2.2 Procedures

Firstly, *Trametes Versicolor* F200 was cultured in Petri dishes containing malt extract agar (MEA) and incubated at 25°C for 7 days. Then the fungus was inoculated into 100 ml of a liquid medium containing: glucose 10 g L$^{-1}$ KH$_2$PO$_4$ 0.5 g L$^{-1}$, K$_2$HPO$_4$ 0.6 g L$^{-1}$, and concentrate at pH 4.5. After the stage of pre-incubation at 25 °C for 7 days, diluted black liquor has been added and then the sample was incubated for 15 days. Second, treated black liquor from coagulation-flocculation process was added to the cultivated *Trametes Versicolor* F200 and then incubated for 15 days. The coagulant-floculant used were Polyaluminium Chloride (PAC) and Polyacrylamide. Decolorization, concentration of glucose, COD, mycelial mass, and enzymatic activity of fungus were analyzed at the day 0, 3, 5, 7, 10, 14, and 21.

2.2.1. Mycelial mass

After incubation, *Trametes versicolor* F200 was separated from the medium using a filter paper. After that, dried at 55°C for 28 hours and weighed [7].
2.2.2. Glucose concentration

200 mL of black liquor was added with 200 mL of 5% phenol solution and 2 mL of H₂SO₄. After that, it was incubated at 30°C for 30 minutes. Concentration glucose was measured by using UV-Vis spectrophotometer at wavelength 492 nm [8].

2.2.3. Enzyme activity

Enzymatic activity of Trametes versicolor F200 was measured by monitoring MnP, LiP and Laccase activity. Activity of Manganese peroxidase (MnP) was measured at wavelength 470 nm by monitoring oxidation of 20 mM 2,6-dimethylphenol (2,6-DMP) in 50 mM malomat buffer solution, 20 mM MnSO₄ and 2 mM H₂O₂ [9]. Activity of Lignin peroxidase (LiP) was measured at wavelength 310 nm by monitoring the formation of 2 mM H₂O₂ and LiP buffer solution [10]. Activity of Laccase was measured at wavelength 525 nm by monitoring the oxidation of syringal arudazin and sodium acid buffer [11]. All the reaction occurs after 1 minute measurement and at temperature 20°C. Activity of enzyme are express in units U/I, to describe the amount of enzyme required to oxidize 1 mol of substrate for 1 minute.

2.2.4. Measurement of Chemical Oxygen Demand (COD)

Black liquor was added with 1,5mL of digestive solution and 2,5 ml of H₂SO₄. After that, the sample was boiled at 150°C for 2 hours. COD was measured by using UV-Vis spectrophotometer at wavelength 600 nm [12].

2.2.5. Calculation decolorization

The concentration of black liquor before and after decolorization was determined by absorbance at wavelength 393 nm using a UV-Vis spectrophotometer (UV 2120 Optizen). Decolorization percentage was calculated by the following equation:

\[
\text{Decolorization (\%) } = \frac{C_c - C_s}{C_c} \times 100\%
\]

Where Cc is initial concentration of black liquor (ppm) and Cs is the final concentration of black liquor (ppm).

3. RESULTS AND DISCUSSION

In this study, black liquor has a maximum wavelength at 393 nm. Before black liquor decolorized using Trametes versicolor F200, it was divided into non-treatment and treatment (pretreated by coagulation-flocculation method). PAC was used as coagulant because PAC has efficient in lower dosage and effective coagulant in wastewater treatment with suspended particle [13]. In this study, black liquor has base condition with pH approximately 12. Coagulant PAC effectively worked in acid condition at pH 5.5. Suspended particles in wastewater will carry negative charge, inorganic coagulant form cationic spesies which will be absorbed by negative particles in wastewater, to form microflocs [14]. Polyacrylamide anionic was used as floculant, PAC and polyacrylamide were added in varying 1, 2 and 3% dosage to black liquor. Sludge weight of particle suspended from black liquor reach highest value with addition PAC and polyacrylamide were 3%. Polyacrylamide anionic has able to bring suspended particles in wastewater and agglomerate the slow settling microflocs formed by coagulant to form larger flocs [15]. Decolorization of black liquor by coagulation-flocculation method after addition PAC and Polyacrylamide 3:3 was 97.88%. Decolorization of black liquor was increased after addition PAC and Polyacrylamide in large amount.

![Fig 1. Decolorization of Black Liquor by Coagulation-Flocculation Method](Image)

Trametes Versicolor F200 has been able to decolorize black liquor by using secretion of ligninolytic enzymes. In this study, the highest enzyme activity produced by Trametes versicolor F200 was LiP. LiP has ability for breakdown lignin on black liquor. LiP will be oxidize lignin though transfer oxygen atom from hydrogen peroxide to an aromatic ring (lignin). LiP activity and decolorization of black liquor and treated black liquor
were optimum at 10 days incubation. LiP activity during decolorization of black liquor and treated black liquor were 301.081 U/l and 1039.677 U/l, respectively. Decolorization of black liquor and treated black liquor were 85.96% and 94.30%, respectively.

Fig 2. Decolorization of Black Liquor and Treated Black Liquor and LiP Activity of *Trametes versicolor* F200

Mycelial mass of fungus increased was coincided with decreasing of glucose concentration during the period of incubation. *T. versicolor* F200 used glucose as a carbon source for growth. Black liquor was not affected for growth of *T. versicolor* F200. Black liquor has rich of carbon but black liquor was not used as an energy source and growth substrate. The presence of an alternate energy source such as glucose was used white rot-fungus (WRF) for secrete enzyme. This enzyme was used for decolorized black liquor, called secondary metabolism [16,17,18]. The presence of metabolisable substrates such as glucose, will be affected for biomass to produce enzymes in liquid cultures [19].

Fig 3. Mycelial Mass of *Trametes versicolor* F200 and Concentration Glucose in Liquid Medium During of Black Liquor and Treated Black Liquor Decolorization

Fig 4. COD of Original and Treated Black Liquor
In this study, COD was decreased with the duration of incubation, that means hardness of black liquor was getting lower. COD reduction of black liquor and treated black liquor after 10 day incubation were 250.75 mg/L and 170.35 mg/L, respectively.

4. CONCLUSIONS

In this study, the combination of PAC and polyacrylamide in ratio 3:3 is effective for treatment coagulation-flocculation. *Trametes Versicolor* F200 secrete ligninolitic enzyme for decolorizing black liquor. In this study, LiP has the highest enzyme activity during the period of incubation. LiP activity and decolorization of black liquor and treated black liquor were optimum at 10 days incubation. LiP activity during decolorization of black liquor and treated black liquor were 301.081 U/l and 1039.677 U/l, respectively. Decolorization of black liquor and treated black liquor were 85.96% and 94.30%, respectively. Mycelial mass of fungus increased which coincided with decreasing of glucose concentration during the period of incubation. *T. versicolor* F200 using glucose as a carbon source for growth, which on growth phase fungus secrete enzyme used for decolorize black liquor. COD was decreased with the duration of incubation, that means hardness of black liquor was getting lower.

5. REFERENCES

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The Geometric Planning of Double Track Railway At Rangkasbitung - Serang

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Abstract – The capacity of single track railway Rangkasbitung - Serang line current could not serve the high demand and the passing train intensity is relatively low. So the additional railway track comes up as a propose solution in Rangkasbitung - Serang with double track railway as the plan to provide more train trips and to raise the number of passengers served. This research aimed to plan the geometry of double track railway which consist of horizontal and vertical alignment on Rangkasbitung-Serang’s path (km 79 + 694 up to km 113 + 446) as far as 33,752 km according to the existence path which refers to Rules and regulation railway construction project (Government Regulation No 10, 1986). The results showed that the geometric planning for double track railway on cross Rangkasbitung - Serang belongs to the class of track 3, with the annual passing tonnage value = 5,380 million tons/year and maximum speed (Vr) = 65 km/hours and the operating speed (Vopr) = 59 km/hours. 25 arches are set in horizontal alignment with one bend kind Spiral-Spiral (SS), 19 bends kind Spiral-Circle-Spiral (SCS) and 5 bend types Full Circle (FC). Also, 27 arches are set in vertical alignment with 14 curved vertically through the convex (rising) and 13 curved concave verticals (down).

1. INTRODUCTION

The geometry of the railway is the shape and size of the rail road, either in the longitudinal direction or in the widening direction which includes the width of the rail, the gradient, the horizontal alignment and the vertical alignment, the elevation of the rail and the widening of the rail [1].

At the present time, Rangkasbitung - Serang line is along the major stream to access the port of Merak which is connecting Java and Sumatra island. The capacity of current single track railway could not serve the high demand and the passing train intensity is relatively low. So the additional railway track comes up as a propose solution in Rangkasbitung - Serang with double track railway as the plan to provide more train trips and to raise the number of passengers served. Moreover, the construction of double track on this track can also increase the intensity of the train that crossed into the area of Banten and can shorten the travel time, at once to reduce the burden on the highway, because the usual cargo transported by truck can be reduced by the addition of transport of goods by rail, it is because the frequency of freight trains will be increased when the double track is already in operation.

The geometry of the railway is planned based on the speed of the plan, as well as the size of the train that passes it with regard to safety, comfort, economy and equity with the surrounding environment [2]. This research aimed to plan the geometry of double track railway which consist of horizontal and vertical alignment on Rangkasbitung-Serang’s path (km 79 + 694 up to km 113 + 446) as far as 33,752 km according to the existence path which refers to Rules and regulation railway construction project (Government Regulation No 10, 1986)

2. METHODS

2.1 Reference planning

This research according to the existence path which refers to Rules and regulation railway construction project (Government Regulation No 10, 1986)

2.2 Procedures

Firstly, is the collection of primary data and secondary data obtained from the relevant agencies, it is useful to determine the precise planning of the geometry of the new railway. Second, Calculation of geometry planning double track railway, at this stage is the planning phase of railway double railway geometry across Rangkasbitung - Serang, with reference to the existing path trace, which consists of horizontal and vertical alignment. And lastly, drawing the curve scheme on the horizontal alignment and the vertical alignment [3,4,5].
3. RESULTS AND DISCUSSION

Cross carrying capacity (T) is the number of conveyance assumptions that pass through a cross within a period of one year. The cross carrying capacity reflects the type and total load and the speed of the train passing in the cross. The carrying capacity (T) is called the unit of ton/year [6].

Table 1. Daily Traffic Railways Rangkasbitung - Serang

<table>
<thead>
<tr>
<th>Source</th>
<th>PT. Kereta Api Indonesia Daop 1 Jakarta [7].</th>
</tr>
</thead>
</table>

Tp = 2294 Ton, Tb = 6406.400 Ton, T1 = 1212 Ton, K1 = 1.4, Kb = 1.5, S = 1.1 then:

\[ TE = Tp + (Kb \times Tb) + (K1 \times T1) = 13600.400 \text{ (Ton/days)} \]

\[ T = 360 \times S \times TE = 5.386 \text{ (Million Tons/Year)} \]

Based on Daily Traffic Railways Rangkasbitung - Serang, Obtained the value of cross carrying capacity (T) = 5.386 (million tons / year), then:

Table 2. Class of Railway

Source: PD. 10 Perencanaan Kontruksi Jalan Rel Bab I Pasal 4a Hal.2 [8].

3.1 Horizontal alignment planning

Operating speed = 59 Km/hour

Maximum speed = 65 Km/hour

Plans speed for planning Fingers & Curved transition:

\( V_{R1} \) Plans speed = Maximum speed = 65 Km/hour

Plans speed for planning elevation railway:

\( V_{R2} = 1.25 \times \text{operating speed} = 1.25 \times 59 \text{ Km/hour} = 73.75 \text{ Km/hour} \)

1.1.1 Planned components arch to bend P1 Spiral - spiral (SS) =

With value \( \Delta = 56°21'00'' \sim 6.350^\circ \) and \( R = 500 \text{ m} \):

\( Lc = 0, \Theta s = 3.175^\circ, \text{Ls} = 55.437 \text{ m}, \text{L} = 110.873 \text{ m}, \text{Xc} = 55.420 \text{ m}, \text{Yc} = 1.024 \text{ m}, p = 0.257 \text{ m}, k = 27.727 \text{ m}, T = 55.476 \text{ m}, E = 1.026 \text{ m} \)

1.1.2 Lanned components arch to bend P2 Spiral – Circle – Spiral (SCS) =

With value \( \Delta = 73°55'48'' \sim 73.930^\circ \), \( \text{Ls} = 70.200 \text{ m} \) and \( R = 300 \text{ m} \):

\( \text{Ls} = \text{Lh} = 70.200 \text{ m}, \text{Lc} = 317.052 \text{ m}, \Theta s = 6.701^\circ, \Theta c = 60.528^\circ, \text{L} = 457.452 \text{ m}, \text{Xc} = 70.104 \text{ m}, \text{Yc} = 2.738 \text{ m}, p = 0.688 \text{ m}, k = 35.098 \text{ m}, T = 261.395 \text{ m}, E = 76.330 \text{ m} \)

1.1.3 Planned components arch to bend P4 Full Circle (FC) =

With value \( \Delta = 52°59'56'' \sim 52.999^\circ \), \( \text{Ls} = 26.650 \text{ m} \) and \( R = 800 \text{ m} \):

\( \Theta c = 26.500, \text{Lc} = 740.303 \text{ m}, \text{L} = 740.303 \text{ m}, T = 398.856 \text{ m}, E = 93.916 \text{ m} \)

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3.2 Vertical alignment planning

Vertical alignment is planned based on the following data:

Plans speed (Vr) = 65 km / hour
Minimum radius = 6000 m (for Vr ≤ 100 km / hour)

1. Vertical alignment planning P1

with concave slope of 4 %, then:

Lv = 24 m, Ev = 0.012 m, Xm = 12 m

1.1.4 Vertical alignment planning P2

with convex slope of 10 %, then:

Lv = 60 m, Ev = 0.075 m, Xm = 30 m
4. CONCLUSIONS
The results showed that the geometric planning for double track railway on cross Rangkasbitung - Serang (km 79 + 694 up to km 113 + 446) belongs to the class of track 3, with the annual passing tonnage value = 5,380 million tons/year and maximum speed (Vr) = 65 km/hours and the operating speed (Vopr) = 59 km/hours. 25 arches are set in horizontal alignment with one bend kind Spiral-Spiral (SS), 19 bends kind Spiral-Circle-Spiral (SCS) and 5 bend types Full Circle (FC). Also, 27 arches are set in vertical alignment with 14 curved vertically through the convex (rising) and 13 curved concave (down).

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Performance Analysis of Dijkstra and A* Algorithm to Determine Shortest Path of Hexapod Fire Fighting Robot

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Abstract – In this paper we use Dijkstra and A* algorithm to analyze of Hexapod firefighting robot performance. We use two evaluation criteria of an algorithm: time and space complexity. Time complexity is the time taken by an algorithm to process the data. While, space complexity is the time needed by an algorithm to process the command. By using KRPAI (Indonesian Firefighting Robot Contest) we get Dijkstra algorithm need 6.757,16 µs in average of execution time. Whereas, A* algorithm need an average of 4.270,72 µs to find a solution. Furthermore, Dijkstra algorithm needs 35.374,4 bytes memory to determine shortest path. Whereas, A* algorithm needs 14.192 bytes memory. We hope this study can improve the robot performance in the future. Therefore, someday firefighting robot can be implemented in real life and can reduce the fire disaster victims.

1. INTRODUCTION

The hexapod firefighting robot is a mobile robot which is moved by six feet locomotion. Compared with the wheeled type of firefighting robot, hexapod firefighting robot has limited speed movement of the robot. However, hexapod can be easily manipulated [1]. Accordingly, to improve the performance of the robot, it needs an algorithm to support the searching method to the nearest room from the start point. Therefore, the robot will increase the opportunities in finding fire on available rooms.

Firefighting hexapod used for extinguish a fire on the middle of KRPAI track. The robot must find the fire less than five minutes. One kind of method that can be applied in this case is A* and Dijkstra method. The microcontroller used by the robot has 20kb SRAM memory capacity. In addition, microcontroller clock speed is 72 MHz. Therefore, the existence of an algorithm in support of the robot’s ability to find a solution is needed. Dijkstra algorithm and A* algorithm is the algorithms which use the distance between points which pre-designed as a variable. Based on the characteristic, Dijkstra algorithm and A* algorithm were considered capable to improve the performance of firefighting hexapod to finding the nearest point.

In previous studies, it was found that each algorithm is considered potential to improve the performance of hexapod firefighting robot. Compared with other types of algorithms, both Dijkstra and A* algorithm have a relatively better performance. For example Dijkstra algorithm has a better memory usage and less time complexity compared to ant colony algorithm [1], in other case Dijkstra Algorithm has more efficient execution time compared to ant colony in case Surabaya City Map [6]. Whereas in another study show that A* algorithm produces less execution time and memory efficiency than the breadth first search algorithm [4].

2. METHODS

In this study, we use Dijkstra and A* algorithm method. Dijkstra algorithm uses the actual distance of each point on the map. However, the A* algorithm uses the actual distance and heuristic distance from current point to the destination point.

Dijkstra algorithm using greedy principle, which means that the algorithm uses the shortest side to be included in the solution set for each step. Dijkstra algorithm will begin the search at the point which has the shortest value from the starting point [2]. Here is the pseudocode of dijkstra algorithm.

```plaintext
for each vertex v in Graph: // Initialization
    dist[v] := infinity // initial distance from source to vertex v is set to infinite
    previous[v] := undefined // Previous node in optimal path from source

dist[source] := 0 // Distance from source to source
Q := the set of all nodes in Graph // all nodes in the graph are unoptimized - thus are in Q

while Q is not empty: // main loop
    for each vertex v in Graph: // Initialization
        dist[v] := infinity // initial distance from source to vertex v is set to infinite
        previous[v] := undefined // Previous node in optimal path from source

    min := minimum(dist, Q) // Get the vertex with the lowest distance
    remove min from Q // Remove from Q
    for each neighbor w of min
        relaxation: if dist[w] > dist[min] + weight(min, w) then
            dist[w] := dist[min] + weight(min, w)
            previous[w] := min
```

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u := node in Q with smallest dist[ ]
remove u from Q
for each neighbor v of u:  // where v has not yet been removed from Q.
    alt := dist[u] + dist_between(u, v)
    if alt < dist[v]    // Relax (u, v)
        dist[v] := alt
        previous[v] := u
return previous[ ]

A* algorithm is one type of Best First Search algorithm, that combines Uniform Cost Search and Greedy Best-First Search algorithm [3]. A* algorithm using actual costs and estimated costs. The following is the pseudocode of A* algorithm.

// A*
initialize the open list
initialize the closed list
put the starting node on the open list (you can leave its f at zero)
while the open list is not empty
find the node with the least f on the open list, call it "q"
pop q off the open list
generate q's 8 successors and set their parents to q
for each successor
    if successor is the goal, stop the search
    successor.g := q.g + distance between successor and q
    successor.h := distance from goal to successor
    successor.f := successor.g + successor.h
    if a node with the same position as successor is in the OPEN list \ which has a lower f than successor, skip this successor
    if a node with the same position as successor is in the CLOSED list \ which has a lower f than successor, skip this successor
    otherwise, add the node to the open list
end
push q on the closed list
end

3. RESULTS AND DISCUSSION

The Dijkstra and A* algorithm will be implemented on map made before, show on Figure 1.
Based on the map will be calculated the distance between each zone using Euclidean equation.

\[ f(x) = \sqrt{(x_2 - x_1)^2 + (y_2 - y_1)^2} \]

Therefore, we will get the real distance between each zone that has been connected and heuristic distance from the current zone to the goal point. The calculation of the distance between each zone will be calculated at the midpoint of each zone. Based on these calculations, Dijkstra and A* algorithm may calculate the estimated distance to the nearest room from the start point. Therefore, distance of each zone shown on Figure 2.

![Figure 2 (Map Design)](image)

Therefore, the results of the execution of the algorithm are as follows:

1) **Time Complexity:** This criterion is the execution time required by each algorithm to find a solution for each problem that has been given [5]. Execution time data on each algorithm is shown in Table III.

<table>
<thead>
<tr>
<th>Start Zone</th>
<th>Solution Zone</th>
<th>Processing Time (microseconds)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Start 0</td>
<td>Room 4</td>
<td>Dijkstra: 7868</td>
</tr>
<tr>
<td></td>
<td></td>
<td>A*: 4056</td>
</tr>
<tr>
<td>Start 1</td>
<td>Room 2</td>
<td>Dijkstra: 6436</td>
</tr>
<tr>
<td></td>
<td></td>
<td>A*: 4837</td>
</tr>
<tr>
<td>Start 2</td>
<td>Room 1</td>
<td>Dijkstra: 6373</td>
</tr>
<tr>
<td></td>
<td></td>
<td>A*: 3790</td>
</tr>
<tr>
<td>Start 3</td>
<td>Room 4</td>
<td>Dijkstra: 6666</td>
</tr>
<tr>
<td></td>
<td></td>
<td>A*: 3970</td>
</tr>
<tr>
<td>Start 4</td>
<td>Room 1</td>
<td>Dijkstra: 6457</td>
</tr>
<tr>
<td></td>
<td></td>
<td>A*: 4696</td>
</tr>
</tbody>
</table>

The table records execution time for each algorithm. The execution time is obtained from the accumulated time for each algorithm in the process of searching. The computation time carried out on the internal timer in the robot microcontroller.

2) **Space Complexity:** This criterion shows memory usage for each algorithm in the process of searching [5]. The calculation process performed by accumulating every function that is being executed by the microcontroller. Total consumption of memory needed by each function obtained from ELF file produced by the compilation process of the robot program. Record data of memory consumption for each algorithm is shown in Table IV.
### Table 2 (Memory Consumption Data)

<table>
<thead>
<tr>
<th>Start Zone</th>
<th>Solution Zone</th>
<th>Memory Consumption (kByte)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Dijkstra</td>
</tr>
<tr>
<td>Start 0</td>
<td>Room 4</td>
<td>42784</td>
</tr>
<tr>
<td>Start 1</td>
<td>Room 2</td>
<td>33120</td>
</tr>
<tr>
<td>Start 2</td>
<td>Room 1</td>
<td>32960</td>
</tr>
<tr>
<td>Start 3</td>
<td>Room 4</td>
<td>34784</td>
</tr>
<tr>
<td>Start 4</td>
<td>Room 1</td>
<td>34224</td>
</tr>
</tbody>
</table>

### 4. CONCLUSIONS

Based on the data that has been discussed before, each algorithm could find a solution to any problems that have been given. Therefore both algorithms can be said to be complete. However, on the results produced within the searching process. Dijkstra algorithm has more margin calculation results compared to manual calculation. Meanwhile, the A* algorithm has a lower margin than the manual calculation. Dijkstra algorithm have 5.5cm difference average and A* algorithm have 0 difference on each searching process solution. Therefore, A* algorithm can be said more optimal than Dijkstra algorithm.

In addition, Dijkstra algorithm have more execution time than A* algorithm in find a solution. On the other side, memory consumption of Dijkstra algorithm is more than the required memory of A* algorithm. Then, A* algorithm have better space and memory complexity than Dijkstra algorithm.

Over all, A* algorithm has better performance than Dijkstra algorithm. A* algorithm excellent on three criteria of algorithm performance those are optimality, time complexity, and memory complexity. And both algorithm have meet completeness criteria.

### 5. REFERENCES


Digital Elevation Model for Mapping Physical Vulnerability to Tsunami Hazard Using Geospatial Approach

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Abstract – DEM data followed by other physical parameters in the same pixel size described five levels of tsunami vulnerability. Hydrology analysis using DEM data produced stream distribution that play as a flooding canal for tsunami wave to hinterland area. Moreover, the surface analysis produced elevation and slope data that applied as one of the parameters in weighted overlay through Geographical Information System. The study applied the analysis of Digital Elevation Model (DEM) data which is obtained for Aster GDEM V2 in disaster mapping. The analysis described potential use of global DEM data in the resolution of 1 arc second in performing the physical vulnerability map to tsunami hazard in the coastal area. All parameters of the physical vulnerability described different value in pixel weight. Elevation performed as the highest weight compared to other parameters. The combination of this physical tsunami vulnerability map and social tsunami vulnerability analysis will perform all tsunami vulnerability assessment. This concept can also be applied in the vulnerability assessment of coastal hazard, and will be our further study.

1. INTRODUCTION

A tsunami can be defined as a series of waves generated in a water body by a disturbance that vertically displaces the water. These disturbances push water upwards, sideways, or downwards to generate huge waves in the ocean. The waves have extremely long wavelengths and period. Tsunami waves can propagate across the ocean, and affect the coastal area, also inundate those areas. Mostly tsunami caused by earthquakes and landslide, or volcanic eruptions [1]. Physical vulnerability mapping is one of the important approaches to reducing the impact of natural disasters. Together with hazard assessment and the element at risk or exposure, vulnerability assessment is one of the parameters in risk analysis [2];[3]. The combination of hazard risk, societal vulnerability, physical vulnerability and the limited capacities of communities to reduce the impacts of the hazard resulting in disaster. Some research related to the tsunami risk mapping was applied Digital Elevation Model (DEM) in order to create the parameter of elevation, slope, and also the existence of stream for the tsunami vulnerability assessment. Geographical Information System (GIS) approaches have proven effective for creating seamless DEMs of bathymetry and topography at a finely scaled resolution [1]. This study tried to assess tsunami vulnerability area using physical vulnerability parameters that created from DEM data.

2. METHODS

2.1 Study Area and Datasets

The coastal area of Malang and Lumajang at the South Coastal of East Java Province was applied as study areas (Figure 1). The study applied a resize pixel of DEM collected from the Advanced Spaceborne Thermal Emission and Reflection Radiometer (ASTER) Global Digital Elevation Model Version 2 (GDEM V2) (Aster GDEM V2) for creating physical parameters of tsunami vulnerability. The pixel size or spatial resolution of this DEM was 1 arc second.

Aster GDEM is a product of The Ministry of Economy, Trade, and Industry (METI) of Japan and the United States National Aeronautics and Space Administration (NASA). The improved GDEM V2 adds 260,000 additional stereo-pairs, improving coverage and reducing the occurrence of artifacts. The refined production algorithm provides improved spatial resolution, increased horizontal and vertical accuracy, and superior water body coverage and detection [4]. Surface and hydrology analysis in geospatial processing also applied for generating tsunami vulnerability map, includes elevation, slope, water accumulation, and river or stream proximity. The data analysis was done using geospatial analysis as well as GIS approach.
2.2 Methods

The current study applied 1 arc-second resolution of DEM data. DEM was in raster format and applied down-scaled process to increase the spatial resolution of raster DEM. This raster DEM could also create contour map. Some of the previous studies applied the combination of spatial multi-criteria analysis and Analytic Hierarchy Process (AHP) together with DEM data processing for assessing the affected area by tsunami waves [5];[6];[7]. Criteria of elevation, slope, water accumulation, and water direction were created from DEM data and were examined to create a map of physical vulnerability to tsunami hazard (Figure 2). Tsunami vulnerability map was generated using weighted overlay in geospatial analysis. The weight of the criteria was calculated through pair-wise comparison of AHP.

![Figure 1 Map of study area](image1)

![Figure 2 Flow diagrams of methods](image2)

3. RESULTS AND DISCUSSION

Raster re-classification of each parameter was done using geospatial analysis concept. Each pixel of DEM data had been calculated based on the physical vulnerability range to tsunami hazards. Tsunami vulnerability level was illustrated in five level of vulnerability (very low to very high). The range of elevation was applied; (1) less than 5 meters for very high level; (2) 5-10 m for high; (3) 10-15 m for moderate; (4) 15-20 m for low; and (5) more than 20 m for very low [8]. Moreover, the slope was defined as the rate of maximum change in the z-value in each cell of the raster DEM. The range of values in the output depends on the type of measurement units used. The range of slope in percent was applied; (1) 0-2 for very high level; (2) 2-6 m for high; (3) 6-13 m for moderate; (4) 13-20 m.
for low; and (5) more than 20 m for very low [9]. The result from DEM processing for elevation and slope analysis is presented in Figure 2 and Figure 3.

Figure 3 DEM processing for elevation based on tsunami vulnerability level

Figure 4 DEM processing for slope based on tsunami vulnerability level

The flow accumulation function calculates the accumulated flow as the accumulated weight of all pixels flowing into each downslope cell in the output raster. Tsunami vulnerability mapping using this parameter was applied in the area of Tohoku Japan after the 2011 Japan tsunami [7]. Pixels with a high flow accumulation are considered to be areas of concentrated flow and may be used to identify stream or water canal (Figure 5).

Both elevation and slope in the eastern part of the study area more described as a flat area with a low range of elevation. In addition, based on the water accumulation map, the coastal area illustrated few feature of stream compared to the west part (coastal area of Malang). Together with DEM data, land cover map supported in providing data for the density of vegetation along the coastal area.

Figure 5 DEM processing hydrology analysis for water accumulation

The calculation of all criteria trough weighted overlay in GIS analysis is described on tsunami vulnerability map (Figure 6). The map described that the area with a very high level of tsunami vulnerability was spread in the flat area, mostly in the coastal area of Lumajang, where identified as the area with a low density of vegetation. Different type of coastline (coastal morphometry) also plays as an important parameter in the analysis of vulnerability area. This study illustrates that although there was an island that acts as barriers to the tsunami, but its existence did not significantly reduce the impact of waves. In Malang coastal area, high level of tsunami
vulnerability identified around the area that covers by a barrier island (Sempu Island). This area identified as a residential area and well known as the center of marine fisheries activities.

![Figure 6 Physical tsunami vulnerability map in the coastal area of Malang and Lumajang](image)

4. CONCLUSIONS

Tsunami vulnerability mapping was an effort to reduce the impact of tsunami hazards. By mapping the vulnerable area and potentially inundated area caused by the tsunami wave, we can prepare for risk map of the tsunami. The use of DEM in preparing basic data for analyzing and assessing vulnerability and risk area to tsunami hazards performed good result. This information is important for disaster mitigation related to the preparation of both evacuation route and evacuation building. By adding more parameters that could be affected by the final result of tsunami hazard mapping, better early warning of tsunami hazard could be generated. Results that performed in this study could be very important for tsunami mitigation and early warning system.

5. ACKNOWLEDGEMENT

Aster GDEM is a product of The Ministry of Economy, Trade, and Industry (METI) of Japan and the United States National Aeronautics and Space Administration (NASA). The authors thank Ministry of Research, Technology and the Higher Education Republic of Indonesia for the financial support.

6. REFERENCES

Implications of Kendeng Fault to Seismic Hazard Potential in Malang

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Abstract – Recent study has revealed the existence of earthquake source in East Java region that comes from Kendeng fault. The fault proved still active with movement of about 5 mm/year. The implications of Kendeng fault to seismic hazard potential in Malang region can be estimated by adopting the deterministic seismic hazard analysis method (DSHA). The DSHA stages consist of identifying and characterizing earthquake sources, determining earthquake sources to site distance parameter, determining the controlling earthquake and defining earthquake hazard produced at site by the controlling earthquake. Implications of Kendeng fault to seismic hazard potential in Malang region by applying the DSHA gave peak ground acceleration (PGA) values ranging from 0.1 to 0.22 g, also the spectral acceleration in T=0.2 and T=1 sec periods ranging from 0.25 to 0.55 g and 0.12 to 0.24 g respectively. To the south, the seismic hazard potential that caused by Kendeng fault gradually decreases, and reached the minimum near the coastal area. Nevertheless, southern Malang region has earthquake hazard potential originating from south of Java subduction zone and background earthquake source.

1. INTRODUCTION

Malang region had a population of nearly 4 million people [1], is the second largest city in East Java. Malang is also famous as city of education. Based on seismotectonic, Malang and surrounding areas affected by south of Java subduction zone and also local and regional faults in surrounding areas. The historical seismicity recorded some destructive earthquakes in Malang and surrounding areas, including the October 20, 1958 earthquake with magnitude of 6.7 which felt VII - VIII Modified Mercalli Intensity (MMI) in Malang, the February 19, 1967 earthquake with magnitude of 6.2, the September 28, 1998 earthquake with magnitude of 6.3 [2] and the November 16, 2016 earthquake with magnitude of 6.2 [3]. Seismic risk in Malang region got worse by increasing of its vulnerability caused by its population growth and infrastructure expansions.

Generally, East Java is divided into six physiographic zones (see Figure 1), one of which is Kendeng anticlinorium [4]. Kendeng zone includes the mountain range with east - west lengthwise direction which lies directly to north of Ngawi sub zone. This mountain is composed by intensively deformed marine sedimentary rocks which formed anticlinorium. These mountains have a maximum width of 40 km [5], extending from Ungaran volcano in the west to the east through Ngawi until Mojokerto area. Below the surface, the continuation of this zone can still be followed up under southern Madura.

Based on differences in stratigraphy and tectonic intensity, Kendeng zone is divided into three sections covering West Kendeng, Central Kendeng and East Kendeng [4, 5]. West Kendeng covers area between Mount Ungaran until around Purwodadi with the oldest outcrops aged Oligo-Miocene and represented by Pelang formation. These rocks containing volcanic materials. These areas have complicated geological structure i.e. many thrust faults. Central Kendeng covers area from Purwodadi up to Mount Pandan. The oldest outcrops aged middle Miocene. These area consists of turbidities sediments (sea), represented by Kerek and Kalibeng formations. East Kendeng consists of latest Cenozoic sediment exposed between Mount Pandan and Mojokerto, aged Pliocene and Pleistocene. Its geological structure is folds with the folding axis shifted to the north and subducting eastward.

Recently, the understanding of seismic hazard in East Java has increased. Based on geoscience studies, the number of known crustal fault sources increased. Recent study has revealed the existence of earthquake source in East Java. The earthquake source is Kendeng fault. The Kendeng fault passes through southern outskirts of Surabaya, and traverses a 300 km length westward of East Java. Thus fault was still active with a movement about 5 mm per year [6] and will be a new threat for earthquake hazard potential in East Java.

Generally, there are two possible approaches for estimating the seismic hazard, i.e deterministic seismic hazard analysis (DSHA) and probabilistic seismic hazard analysis (PSHA). Both methods can complement each other thereby providing additional insight into seismic hazard. This aim of the study is to analyze the implications of Kendeng fault to seismic hazard potential in Malang region through a reliable DSHA methodology. The results
of the study are expected to contribute to earthquake disaster mitigation especially for the Malang region.

![Figure 1. Physiographic map division of Central and East Java [4].](image)

2. METHODS

The implications of Kendeng fault to earthquake hazard potential in Malang region is determined by adopting deterministic seismic hazard analysis (DSHA) method. DSHA is an approach to evaluate site specific seismic hazard that is influenced by maximum hazard from controlling sources affecting the site specific study [7]. DSHA involves the development of a particular seismic scenario upon which a ground motion hazard evaluation is based. DSHA method can be described into four stages. First, identification and characterization of earthquake sources. Source characterization includes defining source’s geometry and earthquake potential. Second, determination of a source to site distance parameter for source zone. In DSHA, generally the shortest distance between source zone and the site of interest is selected. The next stage is determination of controlling earthquake. Generally, it have been selected earthquake that is expected to produce the strongest level of shaking. The last stage is defining seismic hazard at the site produced by the controlling earthquake, usually in terms of the ground motions. To know the implications of Kendeng fault in Malang region, we assumed that the controlling earthquake came from Kendeng fault zone.

For seismic hazard analysis, the potential maximum magnitude that can be generated from Kendeng fault is important and is an input key. The maximum magnitude indicates the highest potential of accumulated strain energy to be released in the region or seismic source. Alternatively, the maximum magnitude is an upper limit or the largest possible earthquake that may produce the highest seismic hazard scenarios for the region [8]. One method that can be used to estimate the potential maximum magnitude is calculated by the equation proposed by Wells and Coppersmith [9] as shown in Equation 1. The maximum Magnitude ($M_w$) is a function of fault surface rupture length ($S_{RL}$).

$$M_w = 5.08 + 1.16 \log (S_{RL})$$  \hspace{1cm} (1)

Characteristics magnitude ($M_{char}$) can also be determined by its relationship with the recurrence interval magnitude characteristics ($T_{char}$) and slip rate ($S_r$) as shown in Equation 2 [9, 10].

$$T_{char} = 10^{0.59M_{char} - 0.99} / S_r$$  \hspace{1cm} (2)

Estimation of seismic hazards due to Kendeng Fault was determined by using a ground motion prediction equation (GMPE). The GMPE express the seismic hazards in forms of peak ground acceleration (PGA) and spectral acceleration. In this study, seismic hazard evaluated by using GMPE from Abrahamson and Silva [11]. This GMPE was derived by using a database of 655 recording from 58 earthquakes. Empirical response spectral attenuation relations are derived from average horizontal and vertical components for shallow earthquakes in active tectonic regions. PGA and spectral accelerations are functions of magnitude, distance, and other variables related with ground motion on the hanging wall and footwall of dipping fault. The function adopted from Abrahamson and Silva [11] for spectral ordinates at rock sites is the following:

$$\ln{(y)} = f_1(M_w, r_{rup}) + F_f(M_w) + HW f_2(M_w, r_{rup})$$  \hspace{1cm} (3)

where, $y$ is the peak ground acceleration (PGA) or spectral acceleration in g, $M_w$ is moment magnitude, $r_{rup}$ is the closest distance to the rupture plane in km, $F$ is the fault type (1 for reverse, 0.5 for reverse/oblique, and 0 otherwise), and $HW$ is a dummy variable for sites located on the hanging wall (1 for sites over the hanging wall, 0 otherwise).
The distance from earthquake source to site can be estimated as the epicentral distance \(r_{eb}\), hypocentral distance \(r_{hypo}\) and closest distance to the rupture plane \(r_{rup}\) as shown in Figure 2. The source to site distance in GMPE proposed by Abrahamson and Silva [11] is defined as the closest distance to the rupture plane \(r_{rup}\).

![Figure 2. Source to site distance parameter.](image)

3. RESULTS AND DISCUSSION

The implication of Kendeng fault to seismic hazard potential in Malang region is evaluated by applying deterministic seismic hazard analysis (DSHA). The results are in form of peak ground acceleration (PGA) and spectral accelerations at T=0.2 and T=1 sec periods maps as shown in Figure 3 and 4 respectively. The deterministic peak ground acceleration map showed values about 0.1 to 0.22 g. The Kendeng fault implies relatively high peak ground acceleration in Malang region. The highest peak ground acceleration value is in northern Malang and gradually decrease to the south then reach the minimum one in the coastal area. This revealed the impact of Kendeng fault to seismic hazard potential in Malang.

Deterministic peak ground acceleration in Malang region were relatively high when compared with previous study by Irsyam et al. [12] which the deterministic peak ground acceleration value in Malang region less than 0.1 g. In their study, Irsyam et al. [12] did not include Kendeng fault in the calculation of deterministic peak ground acceleration. The spectral acceleration at period T= 0.2sec map showed the highest values (ranging from 0.25 to 0.55 g). The spectral acceleration at period T=1 sec map showed a slightly higher values than the deterministic peak ground acceleration map. The third maps showed a similar pattern, which is the lowest value in the south (coastal area), gradually increase, and reached the maximum in the northern Malang.

Compared with calculation results of probabilistic seismic hazard analysis as adopted in SNI 1726:2012 [13], deterministic peak ground acceleration values in Malang region due to Kendeng Fault are still relatively lower. It is because peak ground acceleration (PGA) in SNI 1726:2012 also involved other sources, i.e. subduction and background earthquake sources. The southern parts of Malang region have a greater potential hazard from southern Java megathrust earthquake zone and also from background earthquake sources.

![Figure 3. Deterministic peak ground acceleration (PGA) in Malang region as implication of Kendeng fault.](image)
Figure 4. Spectral acceleration at T=0.2 and T=1 sec periods in Malang region as implication of Kendeng fault.

4. CONCLUSIONS

Implications of Kendeng fault to seismic hazard potential in Malang region by applying deterministic seismic hazard analysis (DSHA) gave peak ground acceleration at bedrock values around 0.1 – 0.22 g and spectral acceleration at T=0.2 and T=1 sec periods around 0.25 – 0.55 g and 0.12 – 0.24 g respectively. The Kendeng fault implies a relatively high peak ground acceleration and spectral acceleration in northern Malang and gradually decrease to the southern and reach the minimum one in the coastal area. Still, the southern parts of Malang have a greater seismic hazard potential from southern Java megathrust earthquake zone and also from background earthquake source.

5. REFERENCES

The Possibility of VLF Method for Measuring and Mapping Peatland

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Abstract –This paper introduces a geophysical method as an alternative for peat thickness measurement. It uses electromagnetic waves of very low frequency (VLF) which is often used for subsurface mapping but has not done much to peat mapping. This method is expected to be used for measuring the thickness of the peat due to the ability of propagation of VLF waves that can penetrate the ground. Besides, its equipment is light weight, portable and without contact with the ground. Therefore, the aim is to provide an overall review on the possibility of using VLF electromagnetic wave method for thickness measurement and mapping of peat. This paper is based on desk study, exploring related literatures that shows the capability of VLF method and the related characteristic of peat and analyze the possibility to relate them.

From the study, it is known that VLF electromagnetic is a high-powered radio wave of 15-30 kHz broadcasted by radio stations over the word for submarines navigation. Because of the high power and low frequency, the signal can propagate around the world and atmosphere and also penetrate the ground till a hundred meter. The signal exhibit as a primary electromagnetic field and it generates the secondary electromagnetic field from the material of the ground. The difference material of the ground –especially in terms of electrical conductivity or resistivity- will exhibit different secondary electromagnetic field. Some experiments have proved that peat and clay mineral has different electrical conductivity and resistivity. The resistivity of peat increase as organic content increase. It is above 600 Ω-meter while the dry soil is lower than 200 Ω-meter and water is between 200-600 Ω-meter. It is concluded that the VLF method possibly applied to measure the thickness and mapping peat.

1. INTRODUCTION

Peat land is important resource not only due to its role as a counterweight to the global climate, but also its functions as a unique ecosystem that support for wild habitat and biodiversity. Also, in terms of its water holding capacity, peat land exhibits as a sponge that is necessary for hydrological buffer.

Problems due to improper use of peat has been widely reported by experts. Tidal peat land in Pulau Rimau, South Sumatra, (Armanto, et al., 2008), the Mega Rice Estate Project in Kalimantan (Limin, 2006) are samples of the fail peatland project. The most excited problems is peat fire. Unlike common fire, peat fire cause much more carbon releasing into the atmosphere and spacially damaging (Agus & Subiksa, 2008). In short, the sustainable peat is very important. Therefore, the government policy in peat restoration and management need to be supported and get reinforcement. One form of such support is the provision of accurate data and information on the extent and thickness of the peat.

So far, the report on the extent and thickness of the peat in Indonesia still vary. That is caused by probably different standards, tools and methods in peat measurement and also due to the dynamical characteristic of the peat itself.

Indonesia’s peatlands are widespread in numbers of major islands, particularly in Sumatra, Kalimantan and Irian Jaya. The extent reaches approximately 10.8% of the total land area (Wahyunto, et al., 2003). Various sources and research institutions reported a total area of Indonesia's peat in different amounts. Wetland International reported 20.6 million hectares (Wahyunto, et al., 2003), the International Peatland Society (2016) reported only 11.5 million hectares and The Center for Agricultural Resources Research and Development agree that it is 14.9 million hectares (Ritung et al., 2011). Furthermore, it is said peat does have the dynamics or change, especially when used as cropland.

This paper discusses a new possibility as an alternative for peat thickness measurement. It is based on geophysical method, namely the use of an Electromagnetic Wave of Very Low Frequency (VLF-EM). The
method is called VLF method. The VLF-EM is originally generated for submarine navigation and communication but then used for other purposes due to its capability in penetrating earth surface and propagating in very long distance.

The use of VLF method for peat measurement is reasonably proposed, considering the fact that the commonly known method of peat thickness measurement is digging or boring. Therefore, when there is a tool and method that is able to withdraw information from under ground without digging, then it will be very helpful and effective.

2. METHODS

This paper was written based on desk study method. The literatures related to the VLF-EM and its capability in subsurface detection were explored, as well as ones related to the characteristic of peat that allow it to be detected using VLF method. The relation between them was analyzed to look for the possible match.

3. RESULTS AND DISCUSSION

3.1 The Basic of VLF Electromagnetic

VLF-EM stands for Very Low Frequency of Electromagnetics. It is part of radio wave in the rage of 3-30 kHz and corresponding wavelengths from 100 to 10 Km, respectively. The higher the frequency of electromagnetics the more penetrating capacity in terms of line-of-sight propagation while the lower frequency the more penetrating regarding influence the media to disturbance or oscillation. The VLF tends to penetrate in terms of influence media to disturbance or oscillation (Ramo, et al. 1994).

The VLF-EM is generated by various natural and human-made sources. The natural sources are volcanic eruptions, dust storms, tornadoes, aurora, meteor and lightning discharges while the human-made sources are VLF radio transmitter, wireless’ antennas, and nuclear explosion. Unlike the natural, the VLF-EM generated by transmitter is regular, definite and originally used as a carrier wave to transfer codes for radio navigation, time radio station and secure military communication (Wikipedia).

3.2 VLF Method in Geophysics

The use of electromagnetic method in geophysics has begun since 1920s in US and Canada where the conductive metal deposits was detected in contrast to the surrounding resistive rock. The first geophysical VLF instrument was EM16, manufactured by Geonics on 1973 (W.M. Telford, L.P. Geldart, 2004).

The VLF method utilize 15 – 30 kHz of VLF electromagnetic generated by radio transmitters. The transmitted wave propagates through the earth's surface as ground wave and sky wave, having bounced and refracted so that it is capable of propagating away from the transmitter. The emitted magnetic and electric field role as primary field. This primary field, then generates secondary field due to the induced current (Eddy current) flowing in the conductor in the soil. The arising secondary field is dependent to the electrical properties of objects in the ground and the surrounding areas. The resultant of the primary field and the secondary field is measured by the equipment. The primary field is considered homogeneous, so the changes in resultant field depends only on the change in the secondary field, so that the nature of electrical conductive objects below the surface can be estimated (Totok Wijayanto, et al. 2015)

There are two kinds of VLF method, the active and the passive one. The active method uses the VLF transmitted by its instruments, so never be a problem with the loss of signal from the global transmitter. Also, there is a possibility to adjust the direction of transmitted signal in accordance to the direction of the observed object. Conversely, the passive method uses VLF from global radio stations. There are around 40 radio stations transmit the VLF-EM spread throughout the world. Therefore, the passive VLF method often faces the problem off-signal and improper direction of coming signal.

3.3 The use VLF method in subsurface mapping

The use of VLF method in subsurface exploration and mapping has been reported by some researchers. There are as follows:

b. Fault mapping, done by Beamish (1994) and Ganerød et al., (2006)
c. Groundwater Exploration (Teeuw, 1995), (Bernard & Valla, 1991) and (Sundararajan, et al., 2007)
d. Mineral exploration; chrome ore (Bayrak, 2002), Ag-Pb-Zn deposits, sulfides (Paterson & Ronka, 1971), etc.
3.4 Peat Conductivity and Estimated Response to VLF

Peat is a layer of organic material in soil, result of partially decomposed vegetation that has accumulated in a water-saturated environment and in the absence of oxygen. As an organic, peat contains a lot carbon which bonded in great chemical compounds, The carbon is around 56,30-58,31% of peat mass (Prayitno & Setyawan, 2011), it also contain a lot of water in natural condition. Peat can hold water 13 times as much of its dry weigh (Ricardo et al., 2011) that allows it to function as a flooding buffer during rainy season and to act as a water deposit storage during dry season.

Here are some characteristics of peat that can enable the use of VLF method in peat measurement;

- Peat soil is acidic, contains many dissolved hydrogen ions, pH 3-5, result of anaerobic decomposition of organic material that produces phenolic and carboxylic acids. The presence of water within the peat and the ions dissolved in peat water will influence conductivity and resistivity of peat. Therefore, wet and dry peat will exhibit different responses to the VLF-EM. In general, the more water the more conductive the more electrical current flowing in it and the more arising secondary field detected by VLF receiver.

- Peat soil is generally located on the top of mineral soil. Mineral soil contains clay and sand minerals, while peat soil consist of organic materials in different decomposition level. Olhoeft (1985) examined the electrical response of different minerals and organic soil on several low frequencies. He drew the conclusion that different mineral and organic material shows different response of conductivity and resistivity. More or less similar achievement was reported by Asadi (2009), Ponziani, et al. (2011), Comas et al., (2015).

- One of the minerals commonly detected by VLF instrument is sulfide. Among sulfide is pyrite. It is frequently found on the boundary between peat and mineral soil beneath the peat.

3.5 The Method of Peat Thickness Measurement.

The method should be used for peat measurement is not quite different from common geophysical VLF exploration. The survey lines are designed perpendicular to transmission direction in order to reach the maximum effect to the underground conductive object. The measurement is done at a certain interval along the survey line. Then the data have to be recorded are: tilt angle (α) and ellipticity (ε). The tilt, also called in-phase or real, is the amount of polarized angel of secondary field to the vertical primary field. Whilst the ellipticity which is called quadrature or imaginary is the ratio of elliptical axes on polarization plane. The collected data then have to be analyzed using Fraser and Karous-Hjelt filter in order to get a picture of location distribution and peat depth or thickness along survey line.

There will be two possible approaches in peat thickness measurement.

1. In case there are sulfides under peat layer, then the depth of detected sulfides is considered as peat thickness.
2. In case no sulfides under peat layer, the peat thickness would be recognized from vertical profile, result of filtering analysis.

Next, the map of peat thickness could be made by interpolating the thickness among survey lines, and GIS would be properly applied for this step.

4. CONCLUSION

The different conductivity between peat and mineral soil and the fact that sulfide minerals are found under the peat layer bring the possibility to use the VLF method in peat thickness measurement and mapping.

5. REFERENCES


Utilization of Crude Palm Oil To Produce Biolubricant Through Process of Epoxydation, Hydroxylation And Acetylation

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Abstract – The aims of this research is to determine the optimum operating conditions and to study the reaction kinetics of bio-lubricant production. Biolubricant has been produced using palm oil as a raw material through epoxydation, hydroxilation and acetylation processs. The highest conversion was a reaction at a temperature of 80 °C and at 25 minutes with the conversion value of 64.14%. In this research, also the reaction energy activation of reaction is 4,834.4 J/mole. The reaction enthalpy at a temperature of 70 °C and 80 °C are of 1,981.5 J/mole and 1,898.3 J/mole respectively.

1. INTRODUCTION

Production of lubricants made from vegetable oil (bio-lubricant) is as part of efforts to reduce the use of lubricant made from petroleum. This bio-lubricant can be used motor vehicles and heavy equipment. The raw materials of bio-lubricants can be derived from vegetable oils such as palm oil. Vegetable oil is available in large quantities and can be renewed, and sustainable. Bio-lubricants has good characteristics: stable to heat and evaporation, low emissions. The term bio-lubricants applies to all lubricants, which are both rapidly biodegradable and non-toxic to humans and other living organisms, especially in aquatic environments [1]. Waste cooking oil (WCO), which otherwise finds no immediate potential utilization can be successfully used to synthesize bio-lubricant [2]. Plant seed oils are renewable alternatives to petrochemicals, but they cannot be used in their raw form except they are suitably modified [3]. The production of biolubricants from palm oil and Jatropha oil through two stages of transesterification has been studied [4]. Utilization of vegetable oil to be converted to polyurethanes has been carried out [5]. The exploiting fatty acids in the preparation of biobased polyols and polyurethanes has been carried out [6]. The aims of this research are to determine the optimum operating conditions and to study the reaction kinetics of bio-lubricant production.

2. METHOD

2.1 Chemicals

The chemicals used in this work were refined palm oil, 30% H₂O₂, methanol, anhydride acetic acid, glacial acetic acid, sulfuric acid 2%, bentonite, sodium hydrocarbonate, violet crystal indicator, HBr 0.1 N, indicators phenolphthaleine, ethanol, 0.1 N NaOH, and distilled water.

2.2 Procedures

A volume of 200 ml of palm oil and 40 ml of acetic acid (purity 99%) were put in a reactor. This mixture then was heated at 70 °C. The hydrogen peroxide (H₂O₂) and sulfuric acid of were added drop by drop to palm oil. The reaction was carried out for 120 minutes. After completed reaction, the mixture was then cooled. Epoxy compound then was purified by addition of water of 200 ml and then saturated by addition of NaHCO₃ solution in water of 100-200 ml. A 150 ml of epoxy and 100 ml of methanol was reacted to produce polyol at 40 °C for 120 minutes. Bentonite was used as a catalyst. Finally, 60 ml of polyol compound was converted to bio-lubricant by the addition of 6 ml of anhydride acetic acid of 97% at a temperature of 70 °C and 80 °C. Bentonite was used as a catalyst. The polyoester sampling was analyzed for each 5, 10, 15, 20, 25 minutes. Figure 1 represents the scheme of experimental procedure of biolubricant production.
3. RESULT AND DISCUSSION

Table 1 represents the physical and chemical characteristics of the standard of refined palm oil, and palm oil as raw material respectively. Based on Table 1, it can be seen that the quality of palm oil used in this study meet the standard value so that palm oil can be used as a raw material for making polyolester through epoxidation and hydroxylation process.

Table 1 The result of palm oil quality used in this research

<table>
<thead>
<tr>
<th>No</th>
<th>Parameter</th>
<th>Unit</th>
<th>Standard of Refined palm oil</th>
<th>Result of this research</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Density</td>
<td>kg/m$^3$</td>
<td>923</td>
<td>922</td>
</tr>
<tr>
<td>2</td>
<td>Viscosity</td>
<td>centipoise</td>
<td>49.4</td>
<td>49.5</td>
</tr>
<tr>
<td>3</td>
<td>Oxiran Number</td>
<td>% O</td>
<td>0.04</td>
<td>0.041</td>
</tr>
<tr>
<td>4</td>
<td>Acid Number</td>
<td>mg NaOH/gr of oil</td>
<td>0.704-0.728</td>
<td>0.720</td>
</tr>
</tbody>
</table>

Table 2 represents the physical and chemical characteristics of epoxy and polyl compounds. The physical and chemical properties of intermediate products (epoxy and polyol) were changed. After hydroxilation reaction, there were the changes of physical and chemical characteristics between epoxy and polyol. Based on Table 2, the density decreased from 939.1 kg/m$^3$ (epoxy) to 924.0 kg/m$^3$ (polyol). But both densities of epoxy and polyol are greater than the density of the raw material (922 kg/m$^3$). For the value of kinematic viscosity, at temperatures of 40 °C, the kinematic viscosity of epoxy compound (56.1 cSt) is higher than the kinematic viscosity of polyol (40.5 cSt). This different kinematic viscosity of both compounds is caused by differences in the structure of molecular between the epoxy compound and polyol compound. At a higher temperature (100 °C), there was a decreasing of kinematic viscosity for each type of epoxy compound and polyol compound. This phenomena is due to the increasing of temperature that the distance between the molecules was longer. In addition, the movement of each molecule will more rapid due to an increasing in the kinetic energy of each molecule. The viscosity index value between the epoxy compound and polyol produced quite different. The viscosity index is a number that indicates changes in viscosity due to temperature change. As we know, this viscosity index can describe the stability of a chemical substance or physics to changes in temperature. If the viscosity index greater the viscosity, the stability of the compound is better. Stable properties of these compounds indicate that the bonds between the compounds present in these substances are also more stable. Stable properties of a compound are very important in a lubrication system of an engine. Therefore, with stable properties, the lubrication properties of the machine can work well, either at low temperatures or at high temperatures. The viscosity index of the epoxy compound (162.1) obtained was greater than the viscosity index of polyol (143.5). This different viscosity index can be said that in terms of stability against temperature, the epoxy compound is better than polyol compound. However, for the lubricating oil to be produced later is a polyoester compound. The changes...
in the chemical properties of the epoxy compound into a polyol compound comes from the change of oxyrane numbers. The difference is very clear in physical properties between epoxy and polyol in the term of oxyrane number. The oxyrane numbers of epoxy compound of 4.3 %O are greater than the oxyrane number of polyol of 36% O. This condition due to the epoxy compound had oxyrane-oxygen groups, while for polyol, the oxyrane group (COC) change into COCH₂COH group.

Table 2 Physical and Chemical Characteristics of Epoxy and Polyol Compounds

<table>
<thead>
<tr>
<th>No</th>
<th>Physical and Chemical Characteristics</th>
<th>Unit</th>
<th>Value</th>
<th>Epoxy</th>
<th>Polyol</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Density (at 40 °C)</td>
<td>kg/m³</td>
<td>939.1</td>
<td>924.0</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>Kinematic Viscosity (at 40 °C)</td>
<td>cSt</td>
<td>56.1</td>
<td>40.5</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>Kinematic Viscosity (at 100 °C)</td>
<td>cSt</td>
<td>14.0</td>
<td>7.7</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>Viscosity Index</td>
<td>-</td>
<td>162.1</td>
<td>143.5</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>Oxyran Number</td>
<td>% O</td>
<td>4.3</td>
<td>0.36</td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>Acid Number</td>
<td>mg NaOH/mg</td>
<td>0.91</td>
<td>1.22</td>
<td></td>
</tr>
<tr>
<td>7</td>
<td>Hydroxil Number</td>
<td>mg NaOH/mg</td>
<td>169.1</td>
<td>351.0</td>
<td></td>
</tr>
</tbody>
</table>

Table 3 shows density, kinematic viscosity, viscosity index and an acid number of produced polyester. According to the Table 3, the minimum density of polyolester compound was of 931.5 kg/m³ at temperature 70 °C and at a reaction time of 10 minutes and a maximum value of 937.0 kg/m³ at a temperature of 80 °C and at reaction time for 20 minutes. The minimum kinematic viscosity of polyolester at 40 °C measurement was of 53.97 cSt at temperature reaction of 70 °C, and for a reaction time of 10 minutes. The maximum kinematic viscosity of polyolester at 40 °C measurement was of 58.50 cSt at temperature reaction of 80 °C and at a reaction time of 25 minutes. At a temperature of 100 °C, a minimum kinematic viscosity of polyolester was the value of 12.69 cSt at a temperature of 70 °C and for a reaction time of 20 minutes and a maximum kinematic viscosity of 15.88 cSt at a temperature of a fraction of 80 °C, and at a reaction time of 25 minutes. The kinematic viscosities are higher than the standard value of SAE 5W30. The minimum viscosity index of polyolester was of 160.0 at a temperature of 70 °C and for a reaction time of 20 minutes and a maximum viscosity index of polyolester of 164.7 was found at a temperature of 80 °C and for a reaction time of 5 minutes.

Table 3 Density, Kinematic Viscosity, Viscosity Index and Acid Number of polyester

<table>
<thead>
<tr>
<th>No</th>
<th>Acetylation Variable</th>
<th>Temperature °C</th>
<th>Time (minute)</th>
<th>Density (kg/m³)</th>
<th>± 62.3</th>
<th>± 10.7</th>
<th>± 163</th>
<th>Acid Number (mg NaOH/mg sample)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td></td>
<td>5</td>
<td>931.9</td>
<td>54.29</td>
<td>13.62</td>
<td>162.7</td>
<td>0.54</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td></td>
<td>10</td>
<td>931.5</td>
<td>53.97</td>
<td>14.04</td>
<td>164.6</td>
<td>0.36</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td></td>
<td>70</td>
<td>933.4</td>
<td>54.54</td>
<td>14.12</td>
<td>163.6</td>
<td>0.41</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td></td>
<td>20</td>
<td>933.6</td>
<td>54.79</td>
<td>12.69</td>
<td>160.0</td>
<td>0.64</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td></td>
<td>25</td>
<td>933.5</td>
<td>54.34</td>
<td>14.50</td>
<td>164.6</td>
<td>0.67</td>
<td></td>
</tr>
<tr>
<td>6</td>
<td></td>
<td>5</td>
<td>931.7</td>
<td>54.36</td>
<td>14.80</td>
<td>164.7</td>
<td>0.58</td>
<td></td>
</tr>
<tr>
<td>7</td>
<td></td>
<td>10</td>
<td>932.4</td>
<td>54.31</td>
<td>15.07</td>
<td>164.6</td>
<td>0.52</td>
<td></td>
</tr>
<tr>
<td>8</td>
<td></td>
<td>80</td>
<td>933.6</td>
<td>54.15</td>
<td>14.60</td>
<td>164.5</td>
<td>0.77</td>
<td></td>
</tr>
<tr>
<td>9</td>
<td></td>
<td>20</td>
<td>937.0</td>
<td>55.40</td>
<td>15.49</td>
<td>164.6</td>
<td>0.68</td>
<td></td>
</tr>
<tr>
<td>10</td>
<td></td>
<td>25</td>
<td>936.9</td>
<td>58.50</td>
<td>15.88</td>
<td>163.0</td>
<td>0.33</td>
<td></td>
</tr>
</tbody>
</table>

The value lubricating viscosity SAE 5W-30 is of ± 163. If the viscosity index value of a lubricant does not meet the standards, then the polyolester compound can be added an additives to increase viscosity stability. The minimal of acid number (0.33 mg NaOH/mg) was found at 80 °C and reaction time of 25 minutes. The maximum acid number (0.77 mg NaOH/mg) was obtained at 80 °C and reaction time of 15 minutes. If acid number of biolubricant is small, so the quality of biolubricant is better. High acid number of lubricant used can cause corrosion in the machine. To minimize the acid number close to zero, then the polyolester compound can be added an additives such as pH-neutralizing compound.
Figure 2 represents the reaction conversion of polyol to polyester at temperature 70 °C and 80 °C for 5, 10, 15, 20 and 25 minutes of reaction time.

According to the Figure 2, increasing of reaction temperature, the conversion of polyol to polyoester (bio-lubricant) was increased. Also, increasing of reaction time, the conversion of polyol to polyoester was increased. The highest conversion was a reaction at a temperature of 80 °C and at 25 minutes with the conversion value of 64.14%. In this research, also the reaction energy activation of reaction is 4,834.4 J/mole. The reaction enthalpy at a temperature of 70 °C and 80 °C are of 1,981.5 J/mol and 1,898.3 J/mol respectively.

4. CONCLUSIONS

The conclusion of this work are:

a. The conversion value of polyol to polyester through acetylation process was increased if reaction temperature and reaction time increased.

b. The highest conversion of polyol to polyoester (64.14%) was obtained at 80 °C and reaction time of 25 minutes.

c. The reaction activation energy E was found of 4,834.4 J/mol, enthalpy of acetylation reaction at 70 °C and at 80 °C was obtained of 1,981.5 J/mol and of 1,898.3 J/mol respectively

5. REFERENCES

Sub Surface analysis using Geo-Electrical Resistivity Methods (Schlumberger and Wenner Configuration) at the Umaboco Area, Natabora Village, District of Barique, Manatuto Regency, East Timor (Timor-Leste)

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* Corresponding authors: [adisusilo@ub.ac.id]

Abstract – Sounding acquisition Geo-electrical Resistivity, using Schlumberger and Wenner configurations were applied over the “Sekolah Pendidikan Pertanian” (SPP, Agricultural) Natarbora, District of Barique, Manatuto Regency, East Timor (Timor-Leste) in December 2008. The purpose of this research was to describe the condition of sub-surface rock formations including the depth and thickness and also to determine the potential aquifer in the research area. Another purpose was to compare the two configurations at the same sounding points. Resistivity-meter OYYO type MCOHM-EL Model 2119D was used for the acquisition. It displays the voltage, current, and pseudo-resistance directly. Data processing using Ipi2win were performed to obtain the resistivity, thickness, and depth values of sub-surface rock formations. The interpretation was supported by geological map of Study Area and rock resistivity chart. The results were, the resistivity value ranges from 0.1 to 14400 Ωm using Schlumberger Configuration, and 0.4 to 21700 Ωm using Wenner Configuration. The interpretation of these values are clay, sand, gravel, sandstone, and limestone. Based on Schlumberger Configuration, the most potential aquifer layers are sands and gravels, located at the S-1 point in the depth of 19 – 93 m with 74 m thickness. Wenner Configuration shows the most potential aquifer layers are sands and gravels, located at the W-1 point in the depth of 13.8 – 102 m with 88.6 m thickness. The interpretation of Schlumberger and Wenner configuration have the similar aquifer layers, which are sands and gravels. However, these two configurations have different result on the depth and thickness of a aquifer layer.

1. INTRODUCTION

Sekolah Pendidikan Pertanian (SPP, Agricultural) Natarbora, District of Barique, Manatuto Regency, East Timor (Timor-Leste) has an area of 180,000 m2. Barique District is a vast lowland area and located at an altitude of 32 meters above sea level. Its area is 134 km2 and 45% is utilised as agricultural land. Administratively Barique Sub-district is adjacent to Laclubar Subdistrict and Manatuto Subdistrict in the north, south coast of Timor Sea in the south, Regency of Viqueque in the east, Regency of Manufahi in the west. Geologically, the rocks at the study site are a part of the Suai (Qs) formation, i.e., rock formations composed of loose sediments of sand and gravel sizes up to the skeletal, containing fossil foraminiferal and molluscan shells, old Pleistocene to Resen, deposited in littoral environments to shallow seas. The thickness of the unit is more than 600 meters [1]. The minimal availability of water at the research site which is the centre of agricultural education in Timor-Leste is interesting to examine its subsurface conditions. One geophysical method that can be used to determine aquifer depth is the geodey resistivity [2]. The geoelectric resistivity method has several configurations that are utilised for either sounding (vertical) or mapping (lateral) acquisitions. For the determination of aquifer depth, the configuration of Schlumberger and Wenner is configured for sounding (vertical) acquisition. By using both of the configurations are expected to provide the same information for subsurface conditions and the position of the aquifer at the study site.

2. METHODS

2.1 Aquifer And Resistivity Methods

The purpose of this study is to determine the depth of the aquifer to support the need for water availability at the school. The aquifer is derived from the word aqua which means water and fere meaning to contain, thus the aquifer is defined as a water carrier layer or a permeable layer [3]. The rocks that serve as the best water carrier layers are sand, crust, and gravel. Whereas, 90% of the aquifers consist of unconsolidated rocks, mainly gravel and sand [4]. The method used is the configuration of geoelectrical resistivity method of Schlumberger and Wenner. In principle, the geoelectric method uses four electrodes, i.e., two current electrodes and two potential electrodes. The current
is injected into the earth through two current electrodes, then the response of a potential difference between the current is measured by two potential electrodes [5]. Subsequent calculations of the resistivity of all sub-surface rocks according to geometry factor (k) and the electrode configuration use the equation below.

\[
\rho_a = \frac{\Delta V}{I} 2\pi \left[ \left( \frac{1}{r_1} - \frac{1}{r_2} \right) - \left( \frac{1}{r_3} - \frac{1}{r_4} \right) \right]
\]

\[
\rho_a = \frac{\Delta V}{I} k \quad [1]
\]

2.2 Procedures

The study was conducted in the area of Sekolah Pendidikan Pertanian (Figure 1) at the coordinates of 08°33'59" LS and 125°59'59" BT on December 15-20, 2008. Geoelectric measurements were performed with six sounding points for the Schlumberger and Wenner configurations respectively. For Schlumberger configuration the distance between the current electrode (AB / 2) equal to 0,25-200 m with the distance between electrode potential (MN / 2) equal to 1-30 m. For the configuration of Wenner, the distance between the current electrode (AB = 3MN) is 3-630 m with a potential electrode spacing (MN = a) of 1-210 m. After the data is obtained, data processing using IP2win software is performed. Subsequent interpretation is conducted based on a geological map of the research area and reference resistivity table.

![Figure 1. (a) Research Location Map, (b) Wenner (W1-W6) and Schlumberger (S1-S6) configuration measurement points in the area of Sekolah Pendidikan Pertanian](image)

3. RESULTS AND DISCUSSION

There are six sounding points for each Schlumberger configuration and Wenner configuration. Based on the results of the resistivity data interpretation for the six sounding points, the approximate substructure rocks of the study sites consist of: Sand and Gravel (<45Ωm), Sandstone (46-100 Ωm), Clay (101-300 Ωm), and limestone (> 300 Ωm). All the rocks are evenly distributed across the six sounding points with varying thickness and depth. The possible rocks as aquifers are sand and gravel. To ease the comparison of the results between the Schlumberger and Wenner configurations, the results of the resistivity interpretation for the same sounding point is used in sounding 1 (S1 and W1) points.
Figure 2. Results of data processing IP2win (a) Wenner sounding point 1, (b) Schlumberger sounding point 1

Figure 2 shows the results of data processing for sounding point 1 for both Wenner configuration and Schlumberger configuration. Both configurations show five layers of rock. From the interpretation of the five layers of the rock, there is a difference of the estimation between point S1 and W1 in the first layer. The first layer at point S1 is assumed as Clay having resistivity value 260 Ωm at depth 0-1.46 m with thickness 1.46 m, whereas at point W1 is allegedly Sandstone having resistance value 91.3 Ωm at depth 0-0.5 m and 0.5 m thickness. For the second layer until the five dots S1 and point W1 show the same layer type but with different values of depth and varying thickness.

Figure 3. 2-dimensional cross-sectional overlay of the Wenner (W1) and Schlumberger (S1) sounding layers of the sounding point 1

Figure 3 shows a 2-dimensional cross-section for W1 and S1 as well as the overlay results of both configurations. The difference in the first layer for W1 shows sandstone while for S1 shows clay. Potentially aquifer layer is sand and gravel. The sand and gravel layer for S1 is at depth 19-92.9 m with layer thickness 73.9 m, while for W1 there is at depth 13.8-102 m with layer thickness 88.6 m. Based on these results, it is known that the Schlumberger and Wenner configurations show good and similar results for sounding acquisition. However, both configurations still have differences, namely the depth variation and the thickness of each layer interpreted.

4. CONCLUSION

Based on the results of the interpretation of the Schlumberger and Wenner configurations, the study area is estimated to have a rock array consisting of Sand and Gravel (<45 Ωm), Sandstone (46-100 Ωm), Clay (101-300 Ωm), and limestone (> 300 Ωm). The possibility of rocks that can be served as aquifers are sand and gravel. Sand and gravel are found at all sounding points in either Wenner or Schlumberger configuration with varying depth and thickness. The interpretation of the results also shows that both configurations can indicate good results for sounding acquisition techniques and provide similar information even with different depth and thickness variations.
5. REFERENCES


Study On Mineralization Zone of Southern Blitar By Means of Magnetic Anomaly

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Abstract – Study on mineralization zone of Blitar district by means of magnetic data has been done. Magnetic and position data acquisition were acquired 195 data respectively by using Proton Precession Magnetometer and GPS distributed in all of Blitar district included 11 sub-district. Data distributed in geographic coordinates (112.027311°BT; 8.33114°LS) and (112.43811°BT; 8.12586°LS). Magnetic data processing has been done by some corrections i.e daily, drift, and IGRF(International Geomagnetic Reference Field) to obtain the total magnetic anomaly. From this data processing has been obtained the simple Bouguer anomaly value in the range from -650nT until 800nT that distributed in all of the sub-district. By means of magnetic data processing and interpretation has been obtained that the zero crossing of magnetic anomaly distributed at Bakung - Tambahrejo Beach - Panggungrejo - Binangun - Panggungrejo - Sutojayan as shown contact plane of tuff and limestone-andesite at South and alluvium at North, Kesamben - Southern Malang as shown some local fault, Wonotirto (close zero crossing) as shown intrusion zone of dacite-quartz diorite-tonalite) to andesite-breaks tuff, and Kademangan (close zero crossing) as shown contact plane of tuff and limestone-andesite. The intrusion is a major cause of various mineralization that has magnetic contrast anomaly.

Key words: Magnetic anomaly, Southern Blitar, Mineralization Zone.

1. INTRODUCTION

Administratively, the district Blitar lies between Kediri district in the northern, Malang regency in the eastern, Indian Ocean in the southern, and Tulungagung district in the western part.

While geographically, Blitar district has located between geographical coordinates 111°40' - 112°10' in East longitude and 7° 9'31" in south latitude or have an area about 1588.79 km². Blitar located at an altitude of 167+ meter above sea level. Brantas River divides the Blitar district into two parts, namely north Blitar region has an area of 898.94 km² and South Blitar with an area of 689.85 km². Southern Blitar region has infertile soil because the soil tend to contain limestone, while northern Blitar region has fertile soil because in this area there is Kelud volcano is still active with the eruption and volcanic ash as an adequate number of river streams. Blitar has 22 districts with 7 districts located in southern Blitar and 15 districts located in North Blitar.

In Physiographic, southern Blitar region is part of the southern mountains [1]. Southern mountains extend from Wonosari area, also known as mountain Sewu in Central Java to Blambangan Peninsula in East Java is flanked by Yogyakarta depression zones on the west side and Pasirian depression zone or Lumajang in the east. In the northern of this mountains is lined Quaternary volcanoes on Solo zone, while in the southern of this mountains is the Indian Ocean. southern Blitar region itself constrained by Tulungagung district on the west side, the Kediri district on the north side, Malang district on the east side, and the Indian Ocean on the south side. In general, these mountains have Tertiary age (Oligocene, Miocene, Pliocene up) and Quaternary, several formations intruded by dacite, quartz diorite and tonalite [2]. As a result of the intrusion, the rock surrounding suffered hydrothermal alteration and make propitiation and mineralization. Due later in the area, there are many mineralized zones that produce propilite, pyrite, and other ore minerals. Some mineral or mineral resources resulting from the above process are often found in southern Blitar.

Metallc minerals, in general, are safe products as a result of a process of differentiation and crystallization of magma that occurred during the formation of igneous activity will end. This process begins with the initial break through a hydrothermal solution of high concentration in rocks older age. When the solution in through cracks and/or fractures, there was a process of alteration in these rocks. The process of alteration occurs at high temperature and pressure, followed by changes in the mineral composition of the rock, due to the inclusion of hydrothermal solution. Alteration processes can be classified into 3 types [3], i.e:
- **Argilitisation**: Alteration that changes the mineral feldspar into clay minerals in rocks intruded.
- **Silisication**: Alteration that resulting silica mineral, can be as fine-grained mineral quartz, chalcedon or other types of silicate compounds.
- **Propilitisation**: Alteration that resulting in the mineral group.

At the cooling took place, along with the chemical and physical reactions, precipitates newly formed minerals. This mineral kind depending on the type hydrothermal solution, kind of rock that intruded, and the rate of the cooling process that occurs in the rocks themselves. Diverse rock mineral content, will produce some kind of newly formed minerals. The end result of this process, it will accumulate in the pores of rocks, fractures/faults and/or perlapisan rock. Meanwhile, the area's main characteristic alteration is the formation of CO$_2$ which is the product of the alteration process. The existence and amount of CO$_2$ depend on the type of metal, magnesium and/or potassium rocks intruded. In this case, the alteration process followed by the formation of pyrite (FeS$_2$). This event usually found around bodies of sulfide minerals. Another possible sulfide minerals such as chalcopyrite formed (CuS$_2$) and galena (PbS).[4]

Physically, in the alteration zone has lower value in density, susceptibility, and resistivity than the similar parameters in fresh rock (not experienced alteration) in the surrounding. This happens because of the alteration zone which is the accumulation of metallic minerals would have a value of low resistivity or high conductivity values compared with non-metallic minerals. The existence of contrasting physical values which led to the application of integrated geophysical methods can be used to determine the distribution of the subsurface mineral [5]. Therefore, efforts to optimize the real potential of this district should continue to be done, both the identification, inventory and design management.

Based on the above, the research will be conducted to determine the cause of the spread of various types of rock mineral or mineralized zones contained in Blitar district, especially the southern part Blitar using gravity. The use of magnetic method in this study on the grounds that the mineralized zone generally will have a significant magnetic anomaly contrast with the surroundings.

### 2. METHODS

The study was conducted through a series of activities which include data acquisition, processing, and interpretation. Magnetic data acquisition is distributed in the southern Blitar and lies on the geographical coordinates geographic coordinates (112.02731°BT; 8.33114°LS) and (112.43811°BT; 8.12586°LS) with the points number of measuring data 195. The data is distributed along the roads that can be passed either by vehicle or by foot. Data is also focused on the discovery outcrop locations and locations of mining that has been operating at the study site. The main equipment that used in this study is the PPM (Proton Precession Magnetometer) in nT accuracy rate. The distance between the measuring point to measuring point the other is approximately between 1km to 2km.
Data acquisition is done by looping daily to the local base station. The distributions of the magnetic data are shown in figure 1.

Magnetic data processing has been done by some corrections i.e daily, drift, and IGRF (International Geomagnetic Reference Field) to obtain the total magnetic anomaly. [6].

3. RESULTS AND DISCUSSION

Based on the contour map of the total magnetic field anomaly after diurnal correction (H_D) and International Geomagnetic Reference Field (IGRF) correction (H_0), it can be seen that the values of the total magnetic field intensity result from data acquisition that was originally distributed on 44605.8 nT value up to 46847.2 nT changed distributed into value of -650 nT up to 800 nT as shown in Figure 2.

The changes in the distribution of these values indicate that the intensity delta value is the result of differencing from the intensity of daily correction (diurnal) (H_D) to IGRF (H_0) against the results of field intensity measurement data acquisition field (H) as shown in equation (1).

\[
\Delta H = H - \left( \frac{(t_n - t_{aw})}{(t_{ak} - t_{aw})} (H_{ak} - H_{aw}) \right) - H_0
\]

In this cases, \(\Delta H\) is a value of the magnetic field intensity has been corrected from the influence of diurnal (H_D) and global magnetic intensity (H_0) of IGRF. While \(t_n\) is the nth measurement time, \(t_{aw}\) is the time measurement at the first data, \(t_{ak}\) is the time measurement at the last data, \(H_{aw}\) is the value of the total magnetic field intensity measurement of the first data, and \(H_{ak}\) is the value of the total magnetic field intensity measurement of the last data in one looping. Based on the value of \(\Delta H\), the dipole properties of the distribution of the magnetic field intensity value becomes clear increasingly in the view.

Figure 2. The contour of total magnetic anomaly and zero crossing.

Total magnetic anomaly contour shows the positive or negative of the total magnetic field distribution. The distribution of positive and negative values indicate the presence of dipoles anomalies (figure 2). Geologically, southern Blitar region indicates the presence of intrusion zones of dacitic - quartz diorite - tonalite against the andesite and tuff breccia rocks at the early mesosine [2]. In addition, there are areas of contact between limestone - andesite with tuff in the south and alluvium in the northern and eastern part of the limestone bedrock - some local fault also.

By means of zero crossing analysis from magnetic data contour, shown that the zero crossing of magnetic anomaly distributed at Bakung - Tambakrejo Beach - Panggungrejo - Binangun - Panggungrejo - Sutojayan as
shown contact plane of tuff and limestone-andesite at South and alluvium at North, Kesamben - Southern Malang as shown some local fault, Wonotirto (close zero crossing) as shown intrusion zone of dacite-quartz diorite-tonalite) to andesite-breksi tuff, and Kademangan (close zero crossing) as shown contact plane of tuff and limestone-andesite. The presence of intrusive rocks and some local faults cause the formation of various types of minerals with magnetic contrast value and its spread throughout the south and surrounding of Blitar area.

As we know, that Blitar district has about 16 mines and mineral potential, either as a primary or secondary potential, i.e: kaolin, bentonite, limestone, manganese, feldspar, sand iron, copper, ball clay, onyx stone / calcite, zeolite , mountain stone, sand stone, tras, gold, piropolit, and chalcedon [2].

4. CONCLUSIONS

By means of magnetic data processing and interpretation has been obtained that the zero crossing of magnetic anomaly distributed at Bakung - Tambakrejo Beach - Panggungrejo - Binangun - Panggungrejo - Sutojayan as shown contact plane of tuff and limestone-andesite at South and alluvium at North, Kesamben - Southern Malang as shown some local fault, Wonotirto (close zero crossing) as shown intrusion zone of dacite-quartz diorite-tonalite) to andesite-breksi tuff, and Kademangan (close zero crossing) as shown contact plane of tuff and limestone-andesite. The intrusion is a major cause of various mineralization that has magnetic contrast anomaly.

5. REFERENCES

Characterization of Brick Artifact from Candi Agung Site, Barito River-Kalimantan

Susilo, T. B1,2, Rohman, T1, Maslamah, F1, Soesanto, O2, and Sunarningsih1

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Abstract – Austronesian was the greatest a nation of seafaring in the world. Indonesian was 80% of this nation. There were 12 sites of human settlement along of the Barito river in Kalimantan, including the Candi Agung temple site. This study improved an analysis of brick artifact archeological based on FTIR and XRF which was processed by Analysis Nuclear Network (ANN). Based on grouping by ANN for XRF and FTIR data shown that three samples have closed composition with ceramics from Hubei site (Taxi period) and Shanghai site (Liangzhu period), and the calculations of the brick artifact was 200°C, respectively. Analysis of brick artifact was an extinction by FTIR and XRF that support by ANN. This idea was useful to reconstruction and traced of history/prehistory of a human.

Keyword: FTIR, XRF, and ANN

1. INTRODUCTION

The archaeological artifact remain was a history evidence, and the history was a recognized tool to nation’s culture and the origin of the nation. The great of some historian, as like Seeley say “am w involved word van te born” and Toynbee state that history is a vision of God's creation on the move. Gandhi says “a nation's culture resides in the hearts and in the soul of its people” and Soekarno advice that “don’t forget a history”.

The origin of the nation can be traced to nation’s culture based on archeological remain, including material culture and non-material culture. Human prehistoric can not be writing, while that was a recognize based on archeological remain. The research of archeological chemistry was an extinction, but in Indonesian, the archeological research started since Holland colonial, example P.V. van Stein Callenfels, R. von Heine Geldern, A. N. J. Th. Van Hoop, and H. R. van Heekeren [1].

The human settlement of Banjar site was the part of Austronesia’s site. At along of the Barito river, there were 12 sites of human settlement of Banjar, including the site where the brick artifacts were discovery at Candi Agung site [2]. The research to brick artifacts by spectroscopy was rarely conducted, except for the analysis of wooden artifact and ceramics [3]. Therefore, the research of artifact archeological was a puzzle that crucial to traced and recognized of the culture of human history/prehistory. In this paper, we present a detail FTIR and XRF spectroscopic analysis of the chemical changes occurring in the brick artifact.

2. METHOD

The Fourier Transform Infrared (FTIR) spectra were recorded on Bruker Optic IFS66/s/S interferometer equipped with an attenuated total reflectance (ATR) unit. The range number waves were 650-4000 cm⁻¹ and the typical experimental condition utilized a resolution of 4 cm⁻¹, a velocity of 6-10 kHz, a gain of 16x, an apodization Black Harris 3-term, a Mertz phase correction and zero filling 2, on a double sided, forward-backward acquisition mode. A KBr beam splitter was used for the M-IR source. Subsequently, aliquots of approximately 2 mg artifact were ground and pressed into a KBr pellet, and the infrared spectra were measured on a Perkin-Elmer Spectrum One instrument [4]. While XRF (X-ray Fluorescence) was a non-destructive method. The application advantage was compatibility for analysis of distinction artifact as like brick artifact [5]. ANN was used to processing XRF and FTIR data, as reported studies the dispersal and clustering the characterization of artifact Chinese potteries by Ma [3].
3. RESULT AND DISCUSSION

3.1 Characterization by XRF

Our data of characterization, the XRF data brick fresh (T1, T2, T3 in Fig. 1a) and artifact (B1, B2, B3 in Fig. 1b) shown in Figure 1. The analysis of chemical composition between China’s ceramic, Italian’s ceramic and brick artifact (table 2). The clustering shown that related to between China’s site and Candi Agung site were closed. FTIR data showed the presence of OH stretching vibration, H-O-H bending vibrations, kaolinite, Si-O quartz, Fe-O of hematite, microcline, and wollastonite. Based on table 2, that was processed by ANN, shown the prediction related between the composition artifact and Chinese’s artifact. Based on processing for XRF by ANN, result shown that there are three samples Candi Agung have closed composition with ceramics from Hubei site (Taxi period) and Shanghai site (Liangzhu period) after the data XRF elaborated with Ma’s database (2000) [3].

![Figure 1. (a). Brick fresh and (b) Brick artifact](image)

Table 1 XRF data from brick artifact (B1, B2 B3) and raw material (T1, T2 T3).

<table>
<thead>
<tr>
<th>No</th>
<th>code sample</th>
<th>Locus Site</th>
<th>SiO2 (%)</th>
<th>Al2O3 (%)</th>
<th>Fe2O3 (%)</th>
<th>CaO (%)</th>
<th>MgO (%)</th>
<th>TiO2 (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>B1</td>
<td>Candi Agung</td>
<td>56,12</td>
<td>25,15</td>
<td>13,57</td>
<td>1,93</td>
<td>1,60</td>
<td>1,02</td>
</tr>
<tr>
<td>2</td>
<td>B2</td>
<td>Candi Agung</td>
<td>60,05</td>
<td>19,68</td>
<td>11,02</td>
<td>1,53</td>
<td>1,64</td>
<td>0,94</td>
</tr>
<tr>
<td>3</td>
<td>B3</td>
<td>Candi Agung</td>
<td>55,38</td>
<td>16,66</td>
<td>10,12</td>
<td>1,69</td>
<td>2,99</td>
<td>0,69</td>
</tr>
<tr>
<td>4</td>
<td>T1</td>
<td>Candi Laras</td>
<td>55,38</td>
<td>17,70</td>
<td>4,31</td>
<td>1,46</td>
<td>1,66</td>
<td>0,88</td>
</tr>
<tr>
<td>5</td>
<td>T2</td>
<td>Candi Agung</td>
<td>56,84</td>
<td>19,15</td>
<td>8,66</td>
<td>1,27</td>
<td>1,46</td>
<td>0,91</td>
</tr>
<tr>
<td>6</td>
<td>T3</td>
<td>Pulang Pisau</td>
<td>59,99</td>
<td>23,44</td>
<td>4,08</td>
<td>1,23</td>
<td>1,60</td>
<td>0,93</td>
</tr>
<tr>
<td>7</td>
<td>T4</td>
<td>Panggandingan</td>
<td>58,35</td>
<td>13,90</td>
<td>5,39</td>
<td>1,91</td>
<td>1,56</td>
<td>0,72</td>
</tr>
</tbody>
</table>

Table 2. The resulting analysis of clustering based on XRF data by NMM

<table>
<thead>
<tr>
<th>No</th>
<th>Cluster Cina</th>
<th>Cluster Italia</th>
<th>Sample Artifact</th>
<th>Locus</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>0,12146</td>
<td>0,86589</td>
<td>B1</td>
<td>Candi Agung</td>
</tr>
<tr>
<td>2</td>
<td>0,70476</td>
<td>0,29275</td>
<td>B2</td>
<td>Candi Agung</td>
</tr>
<tr>
<td>3</td>
<td>0,86421</td>
<td>0,13515</td>
<td>B3</td>
<td>Candi Agung</td>
</tr>
<tr>
<td>4</td>
<td>1,1374</td>
<td>-0,13437</td>
<td>T1</td>
<td>Candi Laras</td>
</tr>
<tr>
<td>5</td>
<td>0,74966</td>
<td>0,2479</td>
<td>T2</td>
<td>Candi Agung</td>
</tr>
<tr>
<td>6</td>
<td>0,73649</td>
<td>0,26031</td>
<td>T3</td>
<td>Pulang Pisau</td>
</tr>
<tr>
<td>7</td>
<td>1,1358</td>
<td>-0,13242</td>
<td>T4</td>
<td>Panggandingan</td>
</tr>
</tbody>
</table>

3.2 Characterization by FTIR

Figure 2, 3 and 4 were the spectrum of B1, B2, and B3, respectively. The analysis FTIR vibrational assignment to received shown in table 3. The absorption band at 1628 VS and 3449 VS cm⁻¹ were due to the H-O-H binding of molecule water. The indicate there was a content of kaolinite on all artifact. It was reported that the band around 1034 cm⁻¹ due to red clay origin with the strong intensity [Palanivel 200] and this peak were present in the Ba, B2, and B3.
Figure 2. Spectra FTIR B1

Figure 3. Spectra FTIR B2

Figure 4. Spectra FTIR B3

The appearance absorption peak in the 787 and 764 cm\(^{-1}\) indicate that the quartz was present in all samples as reported by Palanivel. Palanivel (2009) state that the spectra peak 548S and 532 W were Fe-O hematite, except in the B2. The peaks based on database Palanivel means that samples B1 and B3 was fired on in an oxidizing condition. This indicates that artifact Candi Agung (Kalimantan) and artisans of Sembiankandiyur (India) have been similarity of technique and well aware of the firing of potteries in an oxidizing atmosphere. The spectra
peak at around 471 S was microcline in the B2 as reported by Palanivel (2009) [7] and studies carry out to mineral clay by Farmer [6].

Tabel 3. Characterization by FTIR

<table>
<thead>
<tr>
<th>Sample code [Palanivel, 2009]</th>
<th>ARS</th>
<th>Firing temperature (°C)</th>
<th>Artefact</th>
<th>Assignment</th>
</tr>
</thead>
<tbody>
<tr>
<td>No</td>
<td></td>
<td>200</td>
<td>400</td>
<td>600</td>
</tr>
<tr>
<td>Wavenumber (cm⁻¹)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>3422 S</td>
<td>3445 W</td>
<td>3410 W</td>
<td>3419 W</td>
</tr>
<tr>
<td>ARS</td>
<td>1647 W</td>
<td>1647 W</td>
<td>1645 VW</td>
<td>1633 VW</td>
</tr>
<tr>
<td>3</td>
<td>1040 VS</td>
<td>1038 VS</td>
<td>1045 VS</td>
<td>1054 VS</td>
</tr>
<tr>
<td>4</td>
<td>795 W</td>
<td>778 W</td>
<td>796 W</td>
<td>795 W</td>
</tr>
<tr>
<td>5</td>
<td>778 W</td>
<td>778 W</td>
<td>778 W</td>
<td>778 W</td>
</tr>
<tr>
<td>6</td>
<td>694 VW</td>
<td>694 VW</td>
<td>695 VW</td>
<td>694 VW</td>
</tr>
<tr>
<td>7</td>
<td>668 VW</td>
<td>668 VW</td>
<td>668 VW</td>
<td>-</td>
</tr>
<tr>
<td>8</td>
<td>639 VW</td>
<td>640 VW</td>
<td>642 VW</td>
<td>642 VW</td>
</tr>
<tr>
<td>9</td>
<td>581 VW</td>
<td>-</td>
<td>582 VW</td>
<td>584 VW</td>
</tr>
<tr>
<td>10</td>
<td>535 VW</td>
<td>534 VW</td>
<td>535 VW</td>
<td>534 VW</td>
</tr>
<tr>
<td>11</td>
<td>517 VW</td>
<td>517 VW</td>
<td>510 VW</td>
<td>-</td>
</tr>
<tr>
<td>12</td>
<td>465 S</td>
<td>467 S</td>
<td>463 S</td>
<td>464 S</td>
</tr>
<tr>
<td>13</td>
<td>3421 W</td>
<td>3421 W</td>
<td>3421 W</td>
<td>3421 VW</td>
</tr>
<tr>
<td>14</td>
<td>1639 VW</td>
<td>1645 VW</td>
<td>1640 VW</td>
<td>1640 VW</td>
</tr>
<tr>
<td>15</td>
<td>1085 VS</td>
<td>1086 VS</td>
<td>1087 VS</td>
<td>1084 VS</td>
</tr>
<tr>
<td>16</td>
<td>796 W</td>
<td>796 W</td>
<td>796 M</td>
<td>796 M</td>
</tr>
<tr>
<td>17</td>
<td>778 W</td>
<td>778 W</td>
<td>778 M</td>
<td>778 M</td>
</tr>
<tr>
<td>18</td>
<td>696 VW</td>
<td>696 VW</td>
<td>696 VW</td>
<td>696 VW</td>
</tr>
<tr>
<td>19</td>
<td>583 VW</td>
<td>585 VW</td>
<td>585 VW</td>
<td>584 VW</td>
</tr>
<tr>
<td>20</td>
<td>535 VW</td>
<td>534 VW</td>
<td>534 VW</td>
<td>535 VW</td>
</tr>
<tr>
<td>21</td>
<td>464 M</td>
<td>465 M</td>
<td>463 M</td>
<td>463 M</td>
</tr>
</tbody>
</table>

ARS = non calcsinations; VS = very strong; S = strong; M = Medium; W = weaks; VW = very weak; B1 = Brick no 1 of Candi Agung site; B2 = Brick no.2 of Candi Agung site; B3 = Brick no.3 of Candi Agung site.
3.3 Firing Temperature of Brick Artifact

Table 4. The distance of firing temperature of B1, B2, and B3 artifact from Candi Agung Site with Palanivel (2009) by ANN processing.

<table>
<thead>
<tr>
<th>No</th>
<th>1 (200°C)</th>
<th>2 (400°C)</th>
<th>3 (600°C)</th>
<th>4 (800°C)</th>
<th>Result Clustering</th>
<th>Assignment</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>0.92716</td>
<td>0.13178</td>
<td>0.10382</td>
<td>-0.16457</td>
<td>1</td>
<td>B1</td>
</tr>
<tr>
<td>2</td>
<td>0.94041</td>
<td>0.08935</td>
<td>0.10283</td>
<td>-0.13804</td>
<td>1</td>
<td>B1</td>
</tr>
<tr>
<td>3</td>
<td>0.68215</td>
<td>0.25435</td>
<td>0.15267</td>
<td>-0.09211</td>
<td>1</td>
<td>B1</td>
</tr>
<tr>
<td>4</td>
<td>1.82410</td>
<td>-0.18487</td>
<td>-0.07132</td>
<td>-0.56952</td>
<td>1</td>
<td>B2</td>
</tr>
<tr>
<td>5</td>
<td>2.64170</td>
<td>-0.50743</td>
<td>-0.23753</td>
<td>-0.90418</td>
<td>1</td>
<td>B2</td>
</tr>
<tr>
<td>6</td>
<td>2.64170</td>
<td>-0.50743</td>
<td>-0.23753</td>
<td>-0.90418</td>
<td>1</td>
<td>B2</td>
</tr>
<tr>
<td>7</td>
<td>1.43020</td>
<td>-0.14605</td>
<td>0.00991</td>
<td>-0.29524</td>
<td>1</td>
<td>B3</td>
</tr>
<tr>
<td>8</td>
<td>1.59150</td>
<td>-0.60896</td>
<td>-0.00145</td>
<td>-0.00026</td>
<td>1</td>
<td>B3</td>
</tr>
<tr>
<td>9</td>
<td>0.76937</td>
<td>-0.14726</td>
<td>0.15422</td>
<td>0.20354</td>
<td>1</td>
<td>B3</td>
</tr>
</tbody>
</table>

Table 4. was a result of analysis FTIR data in the B1, B2, and B3 by ANN. This method was useful for clustering of FTIR data. The similarity of XRF data what was conducted for studies dispersal and clustering of artifact Chinese by Ma (2000) [3]. In ANN modeling, the eight samples and each sample was twelve of peak spectra FTIR. By ANN, the firing temperature was found that all samples related to close in at 200°C. This statement was rare because before there was no reported by the researcher, how the prediction of firing temperature of an artifact based on FTIR data [8].

4. CONCLUSION

Based on the grouping of Analysis Networks Method on the XRF data states that the samples of B1, B2, and B3 have closed composition with Chinese ceramics. The samples of B1 closest to the site of Hubei, Taxi period while B2 and B3 samples close to the site of Shanghai, Liangzhu period. FTIR states the prediction of that firing temperature of artifacts bricks were at 200°C.

5. ACKNOWLEDGEMENT

Highest thank to head of The office of Banjarmasin Archeology and Grant PUPT 2016.

6. REFERENCES:


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Evaluation and Development of Network Distribution of Clean Water PDAM Unit Lawang

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Abstract – PDAM Lawang is a region company that has a function to supply clean water for Lawang sub-district. The percentage of service, loss of water and the non-optimal utilization of discharge become the problems of service. At this time PDAM unit Lawang take water from Polaman Source of 70 l/s and distributed to three tanks. In the existing condition on 2015 Polaman Source’s service can serve 4988 SR. Every village has different percentage of service between 25 – 65%. This study is planned to will be performed continuously until 2030. Simulation of the development distribution network using WaterCAD v8i program was conducted under non-permanent condition and simulation time for 24 hours with intervals 1 hour. The goal of developing this plan to improve service up to 65% for Lawang Village and Bedali Village, up to 85% for Kalirejo Village and Sidodadi Village respectively. Moreover, it is important to minimize the water losses only for 20% of total production, increase Polaman Source’s discharge from 70 liters/s up to 80 liters/s in 2030, the installation of new pipelines with parallel method to the existing pipeline which have large velocity. Based on the simulation result of WaterCAD v8i, it has been shown that the network distribution of clean water in PDAM unit Lawang until 2030 has fulfilled the requirement for the technical planning of distribution systems. The cost of the development step I is 2.173.000.000 IDR and for the development step III is 2.011.500.000 IDR.

Keywords: clean water, pipelines, WaterCAD V8i

1. INTRODUCTION

PDAM unit Lawang uses 5 clean water sources to meet service, one of them is Polaman Source. Discharge is drawn from Polaman Source of 70 liters/second. On existing condition 2015 PDAM unit, Lawang zone service of Polaman Source may serve 4 villages with a total 4988 SR. Every village having different percentage services between 25% -65%.

2. METHODS

It takes several systematically step to achieve the expected goals. The systematically step work study as follows: Collecting Data, analysis population data and the number of PDAM services, evaluate the hydraulic conditions using software WaterCAD V8i, development for clean water need and hydraulic conditions using software WaterCAD V8i, calculate the budget plan at the development stage.

In this study, it’s use 3 storage location. Projection calculation needs clean water to the taps unit Lawang:

a. Domestic and Non-Domestic Needs
   Need for clean water is composed of two kinds of needs of domestic and non-domestic needs. Based on the assumption Lawang unit taps, clean water needs of the existing conditions of 80 l / person/day, but at this stage of development will be increased to 100 l / person/day.

b. Losing Water
   Figures loss of water is considered reasonable or within the tolerance limit is 20%.
Table 2.2. Existing Clean Water Needs

<table>
<thead>
<tr>
<th>Stock</th>
<th>Village</th>
<th>Average Clean Water Needs</th>
</tr>
</thead>
<tbody>
<tr>
<td>BNA Stock</td>
<td>Lawang</td>
<td>5.74</td>
</tr>
<tr>
<td></td>
<td>Kalirejo</td>
<td>9.80</td>
</tr>
<tr>
<td></td>
<td>Total</td>
<td>15.54</td>
</tr>
<tr>
<td>Bunder Stock</td>
<td>Bedali</td>
<td>6.16</td>
</tr>
<tr>
<td>Sidodadi Stock</td>
<td>Sidodadi</td>
<td>6.28</td>
</tr>
<tr>
<td></td>
<td>Total Served</td>
<td>27.98</td>
</tr>
</tbody>
</table>

Source: PDAM Unit Lawang

According to the energy conservation theory of Bernoulli’s law that if no energy passes or is received between two points in a closed system, its total energy remains constant. This can be explained in figure 1 and figure 2.

3. RESULTS AND DISCUSSION

For simulation we separate 2 condition for clean water needs, first condition for evaluation criteria second is for development stage. Watercad V8i Analysis Result can be shown in below.

From the results of running WaterCAD for BNA Stock obtained:

1. The maximum pressure occurs when the minimum water requirement is at 00.00 which amounted to 5.52 atm, while the minimum pressure occurs at the time of 3.77 atm 07.00. These results are in accordance with the planning criteria (minimum pressure of 0.50 atm and a maximum pressure of 8 atm).

2. Headloss smallest gradient occurred at 00:00 of 0.25 m / km, while the largest gradient headloss occurred at 07.00 by 5.97 m / km. These results are in accordance with the planning criteria (terms headloss gradient 0-15 m / km).

3. Lowest speed occurred at 00.00 by 0.19 m / sec and a top speed of 07.00 occurred at 1.02 m / sec. These results are in accordance with the planning criteria (speed requirements from 0.1 to 2.5 m / sec).

From the results of running WaterCAD Bunder Stock obtained:

1. The maximum pressure occurs when the minimum water requirement is at 00.00 which is 1.13 atm, while the minimum pressure occurs when 07.00 0.52 atm. These results are in accordance with the planning criteria (minimum pressure of 0.50 atm and a maximum pressure of 8 atm).
2. Headloss smallest gradient occurred at 00:00 of 0.13 m / km, while the largest gradient headloss occurred at 07.00 by 3.04 m / km. These results are in accordance with the planning criteria (terms headloss gradient 0-15 m / km).

3. Lowest speed occurred at 00:00 of 0.13 m / sec and a top speed occurred at 07.00 by 0.74 m / sec. These results are in accordance with the planning criteria (speed requirements from 0.1 to 2.5 m / sec).

**From the results of running WaterCAD Sidodadi Stock obtained:**

1. The maximum pressure occurs when the minimum water requirement is at 00.00 which amounted to 1.98 atm, while the minimum pressure occurs at the time of 07.00 at 0.86 atm. These results are in accordance with the planning criteria (minimum pressure of 0.50 atm and a maximum pressure of 8 atm).

2. Headloss smallest gradient occurred at 00.00 by 0.20 m / km, while the largest gradient headloss occurred at 07.00 by 4.82 m / km. These results are in accordance with the planning criteria (terms headloss gradient 0-15 m / km).

3. Lowest speed occurred at 00.00 by 0.17 m / sec and a top speed of 0.91 occurred at 07.00 m / sec. These results are in accordance with the planning criteria (speed requirements from 0.1 to 2.5 m / sec).

After simulation we can show how clean water need in every development stage. Simulation result can be shown in table 2.3.

<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>BNA (41 ltr/s)</td>
<td>Lawang Barat</td>
<td>1.34</td>
<td>1.34</td>
<td>1.34</td>
</tr>
<tr>
<td></td>
<td>Lawang Timur</td>
<td>8.37</td>
<td>10.23</td>
<td>12.09</td>
</tr>
<tr>
<td></td>
<td>Kalirejo</td>
<td>13.02</td>
<td>15.45</td>
<td>18.00</td>
</tr>
<tr>
<td><strong>∑ Clean Water Needs BNA</strong></td>
<td></td>
<td><strong>22.73</strong></td>
<td><strong>27.03</strong></td>
<td><strong>31.44</strong></td>
</tr>
<tr>
<td>Bunder (22 ltr/s)</td>
<td>Bedali</td>
<td>10.09</td>
<td>12.73</td>
<td>15.51</td>
</tr>
<tr>
<td>Sidodadi (17 ltr/s)</td>
<td>Sidodadi</td>
<td>8.23</td>
<td>9.50</td>
<td>10.77</td>
</tr>
<tr>
<td><strong>∑ Clean Water Needs Stock</strong></td>
<td></td>
<td><strong>41.06</strong></td>
<td><strong>49.26</strong></td>
<td><strong>57.72</strong></td>
</tr>
</tbody>
</table>

Source: Calculation

Based on the simulation result, it has been shown that the network distribution of clean water in PDAM unit Lawang until 2030 has been fulfilled the requirement for the technical planning of distribution systems. The cost of the development step I is 2,173,000,000 IDR and for the development step III is 2,011,500,000 IDR.

**Table 3.1. Draft Budget Plan 2030**

<table>
<thead>
<tr>
<th>No</th>
<th>Description of activities</th>
<th>Total Price (Rp)</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>Procurement Pipe &amp; Pipe accessories</td>
<td>326,155,000.00</td>
</tr>
<tr>
<td>B</td>
<td>Excavation Work, Landfill &amp; Compaction and Piping</td>
<td>455,990,000.00</td>
</tr>
<tr>
<td></td>
<td><strong>TOTAL</strong></td>
<td><strong>782,145,000.00</strong></td>
</tr>
</tbody>
</table>

Conversion rates in 2030 **2,011,500,000.00**

Source: Calculation
4. CONCLUSIONS

Based on the analysis that has been done, it can take some conclusions as follows:

1. Results of the evaluation of pipeline systems on the existing condition with the help of the program obtained Watercad V8i there are several pipes in the distribution network system in areas of study that have not qualified a minimum speed of 0.1 m / s and headloss gradient of more than 15 m / km. Pressure on the existing condition has qualified a pressure of 0.5 to 8 atm.

2. Planning the development of clean water distribution network taps Source Polaman Lawang unit carried out until 2030 with three stages of development, namely:
   - In the first phase of development in 2020 carried out an increasing number of services to 45% of the total population for the village and village of Lawang Bedali, while for the Village and Village Kalirejo Sidodadi to 65% of the total population. At this stage also replacement pipe with smaller diameter on pipes with flow conditions which has a speed of less than 0.1 m / sec and an enlarged diameter of the pipe in the pipe with the flow conditions have Headloss Gradient more than 15 m / km.
   - The results of the evaluation of pipeline systems in a state of expansion until 2030 with the help of the program showed Watercad V8i distribution network system in the study area is feasible and can function well enough to meet the needs of customers.

3. Great budget costs taps Lawang unit during the development stage of:
   - Costs incurred in the development of Phase I in 2020 amounted to Rp 2.173 billion
   - The development of phase II 2025 no changes or the addition of clean water distribution network so as not calculating budget plan.
   - Costs incurred in the development phase III in 2030 amounted to Rp 2,011,500,000.00.

5. REFERENCES

Investigation of Flood Potential Areas In Bengkulu City

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Abstract – During the last five years, several floods have stricken some areas in Bengkulu City. They happened due to the geomorphology of Bengkulu City which is hilly. Hence Bengkulu City becomes a potential city of being stroke by floods during rainy season every year. In addition, some factors, including a high rainfall (over 3000 mm) and tidal flood for both all rivers and sea, contribute to a high frequency of flood occurrence. This study was aimed to map the areas of potential flood in Bengkulu City. Supporting data had been collected directly from field survey through spatial zoning approach based on position, contour, land elevation, altitude, and distance from river as well as from sea. We used GPS type Epoch™ 10 L1 for obtaining data of coordinate and altitude. The data of distance from the rivers and seashore line have been assessed through GoogleEarth. All data have been scaled for describable calculation. From the acquired data, several figures have been drawn to map of flood risks for some areas in Bengkulu City. Additional results of the study were that flood could potentially happen due to rainfall which exceeds average rainfall, especially for some areas such as the districts of Muara Bangkahulu and Singaran Pati.

Keywords: geomorphology, flood, tidal flood, rainfall.

1. INTRODUCTION

Bengkulu City is a city in Indonesia which is located in equator. Climate change in Bengkulu City has been significant during the last ten years which had caused several floods in the city. Climate change had also been affected by climate change is Quebec, Canada, which experienced increased hydrological risk at extreme level that resulted in some flood disaster [1]. National Atmosphere Research Center, Boulder USA published a report that demonstrated the direct effect of global warming on rainfall change and heavy rain occurrences that resulted in flood disasters [2].

In Equatorial zone, flood frequency increased due to climate change which may then related to economic growth. Nigeria is one example of equatorial country which economy has been worsen by floods. There was a linear relationship between flood and economic growth in North Nigeria [3]. In Asian countries, relationship between climate change and floods has been shown on modification of temperature increase which affect on flood disaster and decrease in economic growth index [4]. Flood effect is not only on reduction of economic growth, but also on the destruction of infrastructure. In the case of irrigation construction, flood can cause total loss of plant harvest. In addition, a flood may also create insurgence of diseases [6]. Figure 1 presents one effect of the flood which caused road damage.

Flood disaster can be caused by nature or human activity; hence it is a continuous human problem all over the world, in the past, at present and in the future. Naturally, earthquakes might increase flood risk in an area. Observation of water path deformation in a Christchurch before and after an earthquake by using LIDAR demonstrated a decline in surface area between 0.5 to 1 meter [5].

Bengkulu City is located in the west coast site of Sumatra Island at coordinate of 102°14’-102°22” East dan 3°45”- 3°59” South with hilly ground surface. Such kind of topography is conducive for flood occurrence whenever there are rainy days. Topographical change is one parameter for calculation of flood risk in an area [7]. The magnitude of topographical change makes Bengkulu City as prone for flood incident. This was added with the presence of a big river which estuary is located in Bengkulu seashore. During rainy seasons, the river surfaces increase even flood some land surface. The magnitude of these floods was attenuated by the presence of high tide of Indonesia Sea through inhibition of river flow in the estuary. These conditions often create floods which occur in every rainy season. With the increase in economic activity of Bengkulu City, floods should be stopped soon.
Another consideration of floods occurrence is the increase of Bengkulu City population due to urbanization. This urbanization creates a typical city problem, especially the pronoeness to social disorder. A research reported that Social Susceptibility Index in a city increase with the increase in its population [8]. For these reasons, identification of areas which is prone to floods in Bengkulu City is primary important to be conducted. Figure containing zonation of flood disaster potential in Bengkulu City can be used as a reference by policy makers in Space Arrangement Office, for planning of city development especially for building people settlement, house development, hospital, office etc. In results, people settlement can be plotted and the number of casualties when floods disaster can be minimalized.

2. METHODS

2.1 Flood

Flood is a natural phenomenon which usually occurs in an area where river flow occurred outside the riverbank. Flood can be defined as the presence of water which covers a wide area. In a broader sense, flood can be seen as a hydrological cycle. Theoretically, a flood is water flow or water settlement which causes economical disruption or even loss of human life [9]. Another theory said that flood is a river water flow debit which occurs in a relatively greater than usual as caused by rain that occur in upper area or continuously in a area for a long time so that the water can be deposited in the river body, and hence outflow and submerged surrounding area [10]. The relationships between settled surface water, percolated water and evaporated water can be formulated as follow:

\[ X = Y - (A + B) \]  

In the equation (1): \( X \) = settled water; \( Y \) = rain fall; \( A \) = percolated water; and \( B \) = evaporated water

2.2 Procedures

a. Location

Data collection in this research was located in Bengkulu City. The location was divided into zonatings with distance from each other was about 300 - 500 meter. In every zone, coordinate, altitude and its tophography were drawn, as presented in Figure 2.

b. Data acquisition

Data for mapping of flood disaster potential included coordinate data, altitude, distance from the closest river, distance from coast line, and condition of zone surface. Data was obtained directly in the location by using Geographic Information System (GIS) type Epoch™ 10 L1. The shortest distances between the location and river or coastline were measured by using Google Earth.
Figure 2. Location of flood proneness research in Bengkulu City

Table 1. Data of locations: coordinate, altitude, distance from coast line, distance from river, and topographic condition of areas in Bengkulu City

<table>
<thead>
<tr>
<th>No</th>
<th>Coordinate</th>
<th>Altitude</th>
<th>Distance from coast-line</th>
<th>Distance from river</th>
<th>Topography</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Latitude</td>
<td>Longitude</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
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<td>633</td>
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<td>64</td>
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<tr>
<td>4</td>
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<td>102.256209</td>
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<td>5</td>
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<td>102.255644</td>
<td>14</td>
<td>1119</td>
<td>6557</td>
</tr>
</tbody>
</table>

**c. Data processing**

Acquired data was processed by using the following scoring:
- For altitude data: 0–5 m was scored as 10, 6–10 m as 8, 11–15 m as 6, and those >15 m as 4.
- For distance from the shoreline data: 0–400 m was scored as 10, 401–800 m as 8, 801–1200 m as 6, and >1200 m as 4.
- For distance from the river data: 0–300 m was scored as 10, 301–600 m as 8, 601–900 m as 6, and >900 m as 4.
- For land topography: low land was scored as 8, flat land as 5 and high land as 2.

Scoring was conducted by using Arcgis to give indices and to classify areas in Bengkulu City into three zone classes according to its proneness for flood disaster. In resulted figure, areas with high flood potential were marked with red color, zones with medium flood potential with yellow and zones with low flood potential with green.

**3. RESULT AND DISCUSSION**

Classification of the 182 investigated locations in Bengkulu City according to their proneness to a flood as indicated by different colors were presented in Figure 3. About 30% areas were classified as high risk of flood potential – indicated by red color, 40% areas as medium flood potential – indicated by yellow color, and about 30% areas as no potential of flood – indicated by green.
In this research, within 30% zones with high-risk potential of flood in Bengkulu City, beside those areas were located at low elevation, some areas being located in close proximity with Bengkulu river, as indicated by red color in these areas. Similar research also reported that climate change has caused coastal zones to become areas that prone to flood as affected by high tidal of sea surface which reach into estuary; and hence inhibit river water flow to the sea and then create a flood to the land area [11]. For this reason, a model should be developed to anticipate flooding effect of the river flood to the surrounding lands [12]. In addition, flood disaster was not only affected by high rainfall; river bank sensitivity also contributes to the disaster [13]. These three statements [11, 12, and 13] indicated that the distance of a location the shoreline and its distance to a river significantly affected flood disaster proneness of the location.

Several theories formulated the causes of flood might be fail when observed in reality. A theory which stated that high tides of sea surface easily occur during dry season was not proven in Bengkulu City. We found that the contradictory occurred in Bengkulu City, as high tides often occurred during the rainy season as indicated by high frequency of flood cases. When rain happened in the upland, water that flow to the sea was inhibited by sea water which is in high tide condition. This condition then resulted in the diversion of the river water flow to areas surrounding the river located in low elevation. Another relatively recent theory which create enthusiastic discussion is that an earthquake can cause flood. This theory was proven by the occurrence of earthquake in New Zealand in 2016. Land deformation at water path in Christchurch City before and after earthquake was detected by using LIDAR. LIDAR analysis revealed that there were a subsidence of land that increased flood potential between 0.5 to 1 meter [14]. This detection concluded that earthquake vibration can create flood incident in an area. This phenomenon is a new reality which developed that a theory should be tested continuously.

Previously, earthquake has been proven to cause landslide, tsunami, and liquefaction. Earthquake research in Oregon Coast, United Stated of America, to identify the effect of big earthquake that occurred at previous 300 – 500 years ago on landslide during the last 2 – 9 years demonstrated that an earthquake induce the occurrence of landslide. The greatness of landslides were depended on the strength of occurred earthquake [15]. Furthermore, in the case of earthquake in California, there was an empirical correlation between earthquake strength and landslide and coast erosion [16]; whereas, Chi-chi earthquake caused thousands of landslide spots, in which each area covers wider than 625 m²and a total of more than one kilometer square [17]. Another research stated that earthquakes that occurred in Middle America often induced landslide incidence which magnitude were proportionally correlated with the strength of the earthquake. Volumes of landslide varied among countries in Middle America. Big landslides often occured in Costarica, El Salvador, Guatemala and Panama.

Discussion regarding flood in Bengkulu City become interesting as its occurrence increases yearly in recent years. The volume of water flood increased, both in previously occurred location and in new areas, where there was no flood experience. This research used a primary data that was acquired directly, giving the advantage of this research over the others. Floods which occurred with increasing frequency and volume that occurred in the last five years, indicating increasing threats for the near future. It is predicted that Bengkulu City population will doubled in the next ten years; this increases the social susceptibility index in the city. The increasing population coupled by the increasing flood water volume, it is predicted that future flood will bring more losses of both human and material. For this reason, Bengkulu City government should arranged an accurate mitigation decision to minimize the expected loss when future flood will take place.

![Figure 3. Map of flood potential in Bengkulu City](image-url)
4. CONCLUSIONS

The threat level of flood disaster is predicted to increase significantly as indicated by increasing flood volume every year. With increasing the population of Bengkulu City, this trend warns its community to prepare necessary measures to minimize casualties and loss of material whenever flood accidentally occurs. The threat of more floods in areas that is still close to the beach and close to the river flow.

5. REFERENCES


The Utilization Potential of Radon as Earthquake Precursor in Indonesia

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* Corresponding authors: [angga.sprayogo@gmail.com]

Abstract – A Survey of published literature and simple calculation was taken to identify and catalog earthquake precursors observed in several regions of Indonesia such as West Java, Yogyakarta, and Central Sulawesi. The earthquake precursor selected for analysis anomaly of Radon emissions that trapped in rocks and would be released when faults moves prior to earthquake. The published literature was searched to document the statistic of each reported nearby earthquake by spatial extend, time, duration, amplitude, and signal. This anomaly obtained by running average and standard deviation method, where the earthquake magnitude was plotted by time functions equal to radon concentration data. Along 2016, in Palolo Station, Central Sulawesi, within radius 20 km there are 9 of 10 earthquakes were preceded by increase of radon concentration (anomaly) In Tadulako, 38 of the 84 earthquakes in radius of 50 km which preceded anomalous. In Yogyakarta, 10 of the 18 earthquakes were preceded by anomalous. While in the Pelabuhan Ratu, also found some of the earthquake that preceded by radon anomalous. This results shows the correlation between earthquake and radon with the pattern that has not completely consistent, primarily correlations between radon anomalous with parameters of earthquake.

1. INTRODUCTION

A Survey of published literature was undertaken to identify and catalog earthquake precursors observed in several regions of Indonesia such as West Java, Yogyakarta, and Central Sulawesi. Survey of published literature was accomplished because the Indonesia region was located at the confluence of tectonic plates and faults thus vulnerable to earthquake that cause harm in the society. The earthquake precursor selected for analysis anomaly of Radon Emissions. Radon that trapped in rocks would be released when faults moves prior to earthquake occurs [1,2,3]. Radon is one of natural radioactive element that comes from Uranium series. Radon can be easily diffused from groundwater or rocks due to compression and natural migration [4,5]. While deformation in the pre earthquake zone is occurred, it would be formed microcrack as radon emission which could be detected by sensor. Radon anomaly can be observed in the ground gasses measurement near the fault before until after earthquakes occurred. For a short period anomaly, the observation must be continue to correlate anomaly with the earthquake. Observation and research of radon as earthquake precursor has been done in some country, i.e. Japan, India, Italy etc [2,6].

Figure 1 a) Increase of Radon for Bandirma and Saroz Earthquake [6] and b) Schematic cross section of radon out flux mechanism in the Tashkent groundwater basin [7]

While in Indonesia, research with sample topics was initiated by Research and Development Center of Agency for Meteorology Climatology and Geophysics (BMKG) since 2011. For this purpose, BMKG has also build station and radon detector in West Java, Yogyakarta, and Palu. Development of precursor earthquake monitoring station was built by BMKG be done with consideration of the existence of local faults existing in their respective region. For example, Cimandiri Fault in West Java, Opak Fault in Yogyakarta region, and Palukoro Fault in region of
Central Sulawesi. Long-term expectations with this station is disaster risk reduction has been recognized as of symptoms that occur before an earthquake.

2. METHODS

The compilation from published literature and self analysis about precursors in Pelabuhan Ratu, Yogyakarta, Tadulako and Palolo was searched to document the statistic of each Reported nearby earthquake precursors ie spatial extend, time, duration, amplitude, and signal/noise ratio [4]. Several information of anomalous precursory signals were sought. This anomaly obtained by running average and standard deviation, where the earthquake magnitude was plotted by time functions equal to radon concentration data [9,10].

The anomaly information from the literature analyzed in order determine the characteristics of suspected anomaly precursor, statistically and relate to earthquake parameters. Determination of anomaly is based on percntation of increase to the mean diurnal value (background). A precursor can be assumed as anomaly if its value was higher than mean diurnal value +/- deviation standart. Based on Cicerone experiments [4], 83% of earthquake observation were reported that radon was increase before earthquake occur, relatively to the radon's mean diurnal values. The following figure show location of radon detector installation as monitoring of earthquake precursors in BMKG.

![Figure 2 Location of Radon Detector as Earthquake Precursor Monitoring Station in a) Yogyakarta [1], b) Central Sulawesi [11] c) West Java](image)

3. RESULTS AND DISCUSSION

Variation of radon gas values in the ground in which related to the seismic activity was commonly including 3 phase, those are increase, up to highest top, and then decrease again, depend on the earthquake epicenter distance from the observation station. Whether it is radon measurement far inside the earth or near to the surface, radon concentration changing that may be occur are indications of changes inside the earth which some of them will be associated with the earthquake [12,13]. Radon anomaly is the increase in value of concentration of radon gas suddenly over the limit of daily average based on the half-life of radon (3.8 days). Along the 2016 within a radius 20 km in Palolo there are 9 of the 10 earthquakes were preceded by anomalous. In Tadulako there are 38 of the 84 earthquakes in radius of 50 km which preceded anomalous. In the region of Yogyakarta that 10 of the 18 earthquakes were preceded by anomalous. Variation of radon gas concentrations indicated a change, in form of concentration increase in the same period with the frequency of earthquakes increase. The observation of radon gas was consistent with previous studies that said the increase in radon concentration prior to large earthquakes [6,1]. The figure below is example of the data analysis, radon from BMKG monitoring conducted since 2011 by the method of standard deviation in the three regions in Indonesia.
Figure 3 Radon Concentration in West Java from a) Lembang [8] and b) Sukabumi [4]

Figure 4 Radon Concentration in Yogyakarta from a) Piyungan and b) Pundong

Figure 5 Radon Concentration in Central Sulawesi from a) Tadulako Station and b) Palolo Station in 2016

Result of radon monitoring and analysis was show in fig 3, fig 4 and fig 5. Based on the analysis results from radon observations in Yogyakarta, was found more than 55% local earthquakes occurred preceded by the
appearance of anomalies/precursores of earthquakes in range of 4 up to 10 days before earthquake. The concern was the increased concentration of radon relatively equal with increase frequency or intensity of earthquakes.

The analysis of radon concentration data in region of Central Sulawesi earthquake carried on up to 50 km radius from Tadulako observation station. The results indicate that as many as 38 of the 84 events, followed by anomalous radon earthquake that occurred 7 times in the same time frame. At the Palolo station, from 18 events of earthquake, more than 66% was preceded by increasing value of radon concentration. And from both of station, for narrow radius was found existing increase in the percentage of the earthquake that preceded by the earthquake precursors.

While in the Pelabuhan Ratu, Sukabumi, in 2012 based on Pakpahan et al research [4], also found 4 earthquake that preceded radon anomalous. The sign is the increase in emissions of radon are detected 1.5 to 60 times the normal value daily average over the span of 3 to 40 days before the earthquake. On Mustika research [2] from total of 41 Earthquakes were observed, amount of radon rising trend of the which is Considered as the earthquake precursors were 21 anomalies. This means that in the current study, the successive radon as an earthquake precursor was 51%.

Based on the analysis of the observation of radon concentration monitoring in West Java, Yogyakarta and Central Sulawesi, known the average more than 60% increase of radon concentration is followed by severe earthquakes. This indicates that the concentration of radon has the potential to be further investigated in efforts observation of precursors of earthquakes in Indonesia. It is required is continuity of data, longer research time and the consideration of other seismic factors to obtain analysis to conclude the pattern of precursors that occurs through observation of radon gas concentrations in Indonesia.

4. CONCLUSIONS

Research results on the radon and earthquakes that has already begun in Indonesia, especially West Java, Yogyakarta, and Central Sulawesi shows the correlation between both of them with the pattern that has not completely consistent. So it necessary to develop long-term research with various parameter, primarily to see correlations between radon anomalous as precursor with the parameters of earthquake.

5. REFERENCES


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Earthquake hypocenter relocation using double difference method in Aceh Region

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* Corresponding authors: [thomas.hardy@bmkg.go.id]

Abstract – Sumatra located over Southeast Asia plate where the meeting place of Indo-Australian plate and the Eurasian plate. This fact contributes to Sumatra being the islands prone to earthquake tectonic. Aceh is one the region in which often sustain earthquake. The latest big earthquake on 7th December 2016, M6.5 shook the districts of Pidie Jaya, involve 104 people's deaths, over 43,000 peoples evacuated, and more than a hundred building damaged. One of the effort to assist earthquake mitigation is to determine high precision earthquake hypocenter. We propose double-difference algorithm to relocate hypocenter to get high precision hypocenter. A double-difference method based on the fact if the distance between the two earthquake hypocenter is smaller than the distance between the event with the station observer, then the ray path between the source and the seismic recording station is almost equal to the length of ray paths. We use 2088 earthquake events from BMKG catalog, within area 1.48 – 6.04 N and 94.44 - 98.84 E, from 2009 to end of January 2017. Results of hypocenter relocation indicate a better hypocenter, with a histogram of the frequency distribution of travel time residual and RMS after relocation close to 0 and earthquakes with a certain depth (fixed depth = 10 km) has been relocated.

Keywords: Aceh Region, double difference, earthquake hypocenter, travel time residual

1. INTRODUCTION

Sumatra located over Southeast Asia plate where the meeting place of Indo-Australian plate and the Eurasian plate [1],[2]. It is characterized by three tectonic system, which is Sunda Megathrust, Mentawai Bachtrust, and Great Sumatran Faults [3], shown in Figure 1. This fact contributes to Sumatra being the islands prone to earthquake tectonic. Aceh is one the region in which often sustain earthquake.

Figure 1. Tectonic Setting of Sumatra (www.earthobservatory.sg).

The latest big earthquake on 7th December 2016, M6.5 shook the districts of Pidie Jaya, involve 104 people's deaths, over 43,000 peoples evacuated, and more than a hundred building damaged. List of destructive earthquake that occurred in Aceh Region can be shown in figure 2.
Relocation of earthquake hypocenter is important for obtaining a very accurate hypocenter location which is needed for mapping of earthquakes vulnerability, velocity structure study, global and local studies of seismicity analysis, detail structural analysis as well as identification of the fault zone, distribution and orientation of microfracture [5],[6]. All the effort is to assist earthquake mitigation. One of the efforts to assist earthquake mitigation is to determine high precision earthquake hypocenter. [7]

We propose double-difference algorithm to relocate hypocenter to get high precision hypocenter. A double-difference method based on the fact if the hypocentral separation between two earthquakes is small compared to the event-station distance and the scale length of velocity heterogeneity, then the ray paths between the source region and a common station are similar along almost the entire ray path. In this case, the difference in travel times for two events observed at one station can be attributed to the spatial offset between the events with high accuracy.

2. METHODS

We use 2088 earthquake events from BMKG catalog, within area 1.48 – 6.04 N and 94.44 - 98.84 E, from 2009 to end of January 2017 that can be shown as seismicity data in Aceh Region in Figure 3. Because of lack of arrival phase data and minimum criteria number of observer station (4 stations) so we got 1641 event.

The double difference technique takes advantage of the fact that if the hypocentral separation between two earthquakes is small compared to the event-station distance and the scale length of velocity heterogeneity, then the ray paths between the source region and a common station are similar along almost the entire ray path. In this case, the difference in travel times for two events observed at one station can be attributed to the spatial offset between the events with high accuracy.
3. RESULTS AND DISCUSSION

After relocation process, we got 1585 event (76% of original data relocated). It means that the process of relocation still has strong relation with original data (shown in Figure 4). Epicenter location before relocation process can be shown in figure 4a, while epicenter after relocation can be shown at Figure 4b.

![Figure 4. Epicenter location before and after relocation process](image)

Epicenter location changes before and after relocation maximum are about 0.3 km. It means there are no significant changes in epicenter location before and after relocation. The dominant directions of epicenter shift are from southwest to northeast after relocation process, approximately 60 events. We can see the distance and direction of epicenter location before and after relocation from compass and rose diagram (shown in Figure 5a & 5b)

![Figure 5. Compas Diagram (a) and Rose Diagram (b)](image)

We try to illustrate changes in depth after hypocenter relocation process by making two vertical cross section slice line A-A’, and B-B’ that illustrate the analysis of changes in depth of hypocenter especially in Siemelue Island and Great Sumatran Faults. (shown in Figure 6)

![Figure 6. Vertical cross section line of hypocenter relocation](image)
Figure 7. Vertical cross section A-A’ and B-B’

From figure 7, we can see changes in depth vertical cross section A-A’ and B-B’ that hypocenter with fix depth (10 km) had relocated more vary in distribution and can describe the tectonic setting in Siemelue Island and Great Sumatran Faults.

Figure 8. Residual travel time histogram

Figure 9. Root Mean Square (RMS) histogram
The frequency distribution of residual travel time and RMS (Figure 8 and Figure 9) before and after the relocation of the hypocenter indicate highly significant differences. The amount of residual travel time and RMS distribution close to 0, indicates that after the relocation resulted in a better position of hypocenter and near the actual hypocenter.

4. CONCLUSIONS

Earthquake hypocenter relocation using the double different method in Aceh Region yield hypocenter location more precise, proved from the amount of frequency residual travel time and RMS close to 0, fixed depth (10 km) can be relocated more vary and relocated hypocenter related with the tectonic setting in Aceh Region.

5. REFERENCES

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Metabolite Analysis of Rice Endophytic Bacteria Response to Osmotic Stress

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Abstract – The study was conducted by observing the five isolates of endophytic rice (IM-1, IM-3 IM-10, IM-11, IM-12) on the condition of osmotic stress. Analysis of metabolite using Gas Chromatograph Mass Spectrometer QP 2010 SE. All isolates were able to grow under osmotic stress conditions with different growth patterns. The isolates were able to synthesize osmoprotectants in osmotic stress especially pantothenic acid with different percentages. But for osmoprotectant trigonelline and dehydrovomifoliol not all isolates were able to synthesize.

Keywords : stress, osmotic, bacteria, endophytic, rice.

1. INTRODUCTION

Microorganisms commonly used as the active ingredient are biological fertilizer nitrogen-fixing microbes, phosphate dissolver and stabilizing aggregate (Rao, 1982). Wood. (2014) proposed that in order to determine the pattern of interaction of endophytic bacteria with plants, there is a need to understand the mechanism of interaction. Interaction between endophytic bacteria with the rice plant is a new form of association that can potentially leads to the use of endophytic bacteria as biological agency. The pattern of these interactions have not been widely studied so that there are still many unknown aspects regarding the potential and mechanisms of the endophytic bacteria in rice plants.

Results of previous studies have obtained 32 isolates of endophytic bacteria of some varieties of rice paddy. The bacteria have the potential as agents of biological fertilizer in red rice, black rice, mentik wangi and glutinous red rice. However, these studies have not evaluated their potential as a biological fertilizer (biofertilizer) on osmotic stress or drought conditions and mechanisms of their interactions with the rice crop as well as the production technology as a biological fertilizer. Therefore, this study aimed to gain superior endophytic bacteria isolates that are tolerant to osmotic stress or drought that have a potential as a biological fertilizer on dry land ready for use by farmers.

2. METHODS

2.1 Materials

The study was conducted by observing the five isolates of endophytic rice (IM-1, IM-3 IM-10, IM-11, IM-12). The bacteria were grown on M63 + 0.5 M. M63 medium consisted of 100 mM KH₂PO₄, KOH 75 mM, (NH₄)₂SO₄ 15 mM, MgSO₄ 0.16 mM, FeSO₄ 3.9 μM and 10 mM D-glucose (Cheung, et al. (2009).)

2.2 Procedures

The growth of the isolates was observed under condition of osmotic stress using media M63 + 0.5 M NaCl. The Growth was observed spectrophotometrically with wavelength of 420 nm and observed every 2 hours until it reached stationary growth. The data obtained is displayed in the form of the growth curve and the tendency of growth is analyzed.

The endophytic bacteria isolates were grown in liquid medium M63 + 0.5 M NaCl as osmotic stress for 5 days. Cells were harvested by centrifugation at 4000 rpm for 5 minutes, and the 2.5 ml of the supernatant obtained was taken and was then extracted with 2.5 ml of absolute ethanol in strong centrifuge and concentrated with 100 ul cold ethanol. It was then centrifuged again at a speed of 4000 rpm for 5 minutes and the pellet was resuspended by using 100 ml of absolute ethanol and ready to be analyzed. Samples were analyzed using GCMS QP 2010 SE, with a column of type ZB - AAA (10 ml x 0.25 mmL.D (Phenomenex Inc.), injection quantity 1ul, evaporation temperature of 280 °C, helium as carrier gas, a mass range of m/z 45 - 400 (3.33 u / sec) and flow speed in the column of 0.6 ml / min (Aggio, et al. 2014).
3. RESULTS AND DISCUSSION

3.1 Analysis of Rice Endophytic Bacteria Growth In Osmotic Stress

Endophytic bacteria growth can be observed by its OD (Optical Density) value or absorbance value generated. The OD value is the value that indicates the level of bacterial populations in a medium. In conditions of osmotic stress of 0.5 M NaCl, isolates IM-10 showed a very rapid growth phase (fast grower) compared to other isolates, as well as rapid exponential phase at 8 hours after inoculation. IM-1 isolates had relatively short adaptation phase and relatively fast growth phase. Isolates IM-3 IM-11 and IM-12 showed the relatively slow adaptation phase because it took time for them to adapt until the age of 6 hours, but did not show different exponential points. The same relative growth occurred in isolates IM 3 and IM-11. Cheung, et al. (2009); stated that the adaptation phase (lag phase) is an early period of life of the population of bacteria when cells begin to adjust to the new environment, before the start of exponential growth.

3.2 Analysis of Osmoprotectant Compounds Using Gas Chromatograph Mass Spectrometer (GC MS)

The results of the analysis of extracellular metabolites by GC-MS showed, all isolates were able to synthesize osmoprotektan pantoteic acid, while for others osmoprotektan, bacteria synthesize different patterns (Table 1).

Table 1. Osmoprotektan compounds synthesized by endophytic bacteria

<table>
<thead>
<tr>
<th>Osmoprotektan Compounds</th>
<th>The Concentration of Osmoprotektan (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>IM-1</td>
</tr>
<tr>
<td>Pantothenic acid</td>
<td>0.169</td>
</tr>
<tr>
<td>Trigonelline</td>
<td>0.047</td>
</tr>
<tr>
<td>Dehydrovomofoliol</td>
<td>0.072</td>
</tr>
</tbody>
</table>

Pantoteic acid. It shows that pantotic osmoprotektan acid is most effective to deal with osmotic stress. Pantoteic acid is an amide compound derived from amino acids that can function as osmoprotectants to cope with osmotic stress. Osmoprotectant compounds are dissolved osmotic compounds that serve to balance the osmotic pressure of cell cytoplasm depending on the environment (Wood, 2014).

Trigonelline (N-methylnicotinic acid) is a metabolite of nicotinamide. Trigonelline is an an alkaloid with the chemical formula C7H7NO2. This is a zwitterion formed by methylation of the nitrogen atom of niacin (vitamin B3) (Rojasa, et al. 2014). The rice endophytic bacteria isolate that produced the highest trigonelline was IM-1 with a concentration of 0.047%.

Dehydrovomofoliol is a product of oxidation, which is an intermediate in the synthesis of the inhibitor-abscisic acid in the germinating plant growth (Aspedon, et al., 2006). The rice endophytic bacteria isolate that produced the highest pantothenic acid was IM-12 at a concentration of 0448%.
4. CONCLUSION
All isolates (IM-1, IM-3 IM-10 IM-11 IM-12) were able to grow under osmotic stress conditions with different growth patterns. The isolates were able to synthesize osmoprotectants in osmotic stress especially pantothenic acid with different percentages, whereas for osmoprotectant trigonelline and dehydrovomofoliol not all isolates were able to synthesize.

5. ACKNOWLEDGEMENT
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6. REFERENCES
Instrumentation and Measurement
Prototype to Improve The Effectiveness of E-Toll System Using Height Detectors On Car Using Ultrasonic Sensor Hc-Sr04 and Arduino Uno

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Abstract – E-Toll system on a freeway in Indonesia is a new thing and still have carried out a review of the effectiveness of the system of E-Toll. Implementation of E-Toll system that exists today, on the one hand, it makes a queue of vehicles was reduced. However, on the other hand, it creates new problems. One of the problems arising from the E-Toll system is the addition of a queue of vehicles at the time of payment online queue of vehicles that do not apply the system of E-Toll. Though a freeway should have no such problems. With the E-Toll improve the effectiveness of systems that exist, the problem queue of vehicles on the lane queue of vehicles that do not apply the system of E-Toll can be resolved.

1. INTRODUCTION
In Indonesia, the use of toll still uses the conventional system is to take a ticket and pay the money in cash. This condition will take about 7-10 seconds. Not seldom toll payment queue is causing a huge traffic jam, especially in big cities. The Indonesian government has implemented more modest payments using an e-toll system that can reduce the time of payment of up to 4-7 seconds.

E-Toll is an electronic card to use to pay an entrance fee of toll roads in some areas of Indonesia. E-toll users need only attach the card to pay tolls within 4 seconds, faster than when paying in cash which takes 7 seconds. The use of e-toll also reduces operational costs because it costs to collect, deposit, and transfer cash to and from banks. In addition to being the first step in the modernization of the collection of money, the use of e-toll is also intended to reduce violations (moral hazard) due to the toll attendant did not receive direct payments.

However, the use of e-toll is only enjoyed by private vehicle alone or vehicle type 1 only and only in big cities. Then the e-toll also requires us to buy the card and pay it in the bank, so it is too complicated and takes on another day.

So the need to optimize the e-toll system that can be enjoyed by all types of vehicles and enables users of toll roads.

Grouping vehicles used in the toll using the number of axles. We tried to classify vehicles based on height. Then premises using HC-SR04 ultrasonic sensor, the sensor detects the class of vehicles from various vehicle heights.

2. METHODS
2.1 Design
The design of Research to be conducted is make 2 bolt with attaching by 2 ultrasonic sensor HC-SR04 as a reference from all category of vehicle. Because every vehicle has a different height and length, then each vehicle category will be separated by the different height of the vehicle. Design of research as follows:
2.2 Procedures

Ultrasonic wave is a wave with frequency above sound frequency. Ultrasonic sensor consists of transmitter ultrasonic circuit and receiver ultrasonic circuit. HC-SR04 is Ultrasonic circuit transmitter and receiver that can transmitting ultrasonic signal. Signal be taken out will radiated from transmitter. When ultrasonic signal is touch the obstacle, then signal will reflected and go through receiver. Received signal continued to microcontroller for processed height and length of vehicle. The result of measuring height and length of vehicle displayed by LCD 16x2 and decision of closed or opened gate. Servo will open the gate if fulfill the category and closed if not.

3. RESULTS AND DISCUSSION

Result of Research say that if obstacle with $14 \pm 2$ cm go under sensor, LCD respond with typing Golongan I and servo open the gate. It continuous until Golongan V and closed if obstacle under 12 cm. Result can be seen in table 1 as follows.

<table>
<thead>
<tr>
<th>Category</th>
<th>Example</th>
<th>Height Vehicle in Scale (cm)</th>
<th>LCD Respond</th>
<th>Servo Respond</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>Sedan, Jeep, Family Car</td>
<td>14 ± 2</td>
<td>Golongan I</td>
<td>OPEN</td>
</tr>
<tr>
<td>II</td>
<td>Truck with 2 Axle</td>
<td>18 ± 2</td>
<td>Golongan II</td>
<td>OPEN</td>
</tr>
<tr>
<td>III</td>
<td>Truck with 3 Axle</td>
<td>22 ± 2</td>
<td>Golongan III</td>
<td>OPEN</td>
</tr>
<tr>
<td>IV</td>
<td>Truck with 4 Axle</td>
<td>26 ± 2</td>
<td>Golongan IV</td>
<td>OPEN</td>
</tr>
<tr>
<td>V</td>
<td>Truck or else with 5 Axle</td>
<td>30 ± 2</td>
<td>Golongan V</td>
<td>OPEN</td>
</tr>
<tr>
<td>VI</td>
<td>Motorcycle</td>
<td>&lt; 12</td>
<td>Dilarang Masuk</td>
<td>CLOSED</td>
</tr>
</tbody>
</table>

4. CONCLUSIONS

The prototype E-Toll system using HC-SR04 has been made and can select all type of vehicle according to the height of the vehicle. This prototype can minimalizing human error to determine category of vehicle. And of course this prototype can reduce cost to make it will be cheaper.

5. REFERENCES


The Optimum Wave Length From The ICP-OES For Analyze Mg and Ca Tracer in Coral Skeleton of Porites lutea as Sea Surface Temperature Proxies in Kondang Merak Beach, Malang, Indonesia

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Abstract – ICP (Inductively Coupled Plasma) is one of an analytical technic for detection trace metals in sea environment. ICP-OES measure the intensity of radiation trace element that was atomized and ionized in high temperature. Optical emission spectrometer can analyze simultaneous radiation from specific element then converted to elemental concentration. One of the strengths of ICP-OES can measure 80 different elements in a single analyze with high precision (ppm). A couple of software to run this machine is a software Thermo Scientific Qtegra Intelligent Scientific Data Solution. Coral structured by calcium carbonate that was deposited with the slow mechanism. Poritids coral is one type of massive slow growing coral, their growth form usually has annual pattern was called annual pattern that illustrated by banding in coral skeleton showed a high-low density growth rate. The coral banding provides a various information such as sea temperature, climate condition and pollution in the past time. One of geochemical tracer in coral skeleton was Mg/Ca that been proxies of sea surface temperature. The aim of this study was to know the optimum of wavelength to analyze Mg and Ca tracer of Porites lutea in ICP-OES. The methods sequences begun by coral sampling, coral bleaching, radiograph used X-ray, sample preparation and calculation of coral growth rate, milled and analyzed. About 300 μg coral skeleton was diluted in 240μl of nitrite acid (HNO3) 25 %, then analyzed and the result was comparing with 5 standard solutions. The results showed the best wavelength for analyzed Mg and Ca were Mg285.213(118) with R² = 0.9547 and Ca237.690(90) with R² = 0.9999. The result of ratio of Mg/Ca is around 3.18 – 4.78 mmol/mol

1. INTRODUCTION

Estimating the sea surface temperature (SST) is an important component to understanding past environmental conditions (paleoceanographic), the researchers discovered two elements, they were Mg and Ca that derived from coral aragonite. These elements are very sensitive to changes in seawater temperature. Several analytical tools began to be created to enhance accurately and precision of the minor element of Ca in coral. Some several instruments that have the capability to measure of the minor element to calcium ratio in coral carbonate were inductively coupled plasma-mass spectrometry (ICP-MS) and inductively coupled plasma-optical emission spectrophotometry (ICP-OES). The basic principle used in ICP-OES instruments are emission spectrophotometer, which analyzes the wavelengths that are characteristic of each element. The material to be analyzed by ICP-OES instrument must be the tangible solution. Measurement elements by ICP-OES based on the nature of the elements, if given the thermal energy will cause the valence electrons excited from the ground state to a higher level of energy, but a few moments later the electron will return to the ground state, releasing large emission intensity is proportional to the concentration elements. ICP-OES can measure 80 different elements in a single analyze with high precision (ppb).

Climate change is a change in the behavior patterns of climate over long periods of naturally occurring, and the speed of climate change is presumably due to human activities (NOAA, 2007). The reports of the Intergovernmental Panel on Climate Change (IPCC) in 2007 affirmed that during the last 8000 years before the era of industrialization around 1750s, atmospheric CO₂ concentration increased by only 20 ppm, the concentration of CO₂ in 1750 was was 278 ppm and in 2005 was 379 ppm. The CO₂ concentration was increased about 100 ppm for 100 yrs. Guldberg et al., (2007) made an estimation the concentration of CO₂ in the atmosphere will reach 500 ppm and global temperatures will rise 2 °C in the year 2050-2100.
Two great effects of global warming were the increasing of sea surface level and acidification of the seawater. The high temperature will increase coral stress and trigger of coral bleaching and disease. Decreasing pH of seawater became a serious threat to calcareous biota including coral the main builder of the reef. It explains that the attachment between the reef with changes in global temperatures. Sea surface temperatures and salinity are key climate parameters. Currently available data from both direct measurements only up decades ago, while making a future climate change predictions for the coming of temperature and salinity data are needed in a long-time scale. The use of the elements Mg and Ca which is in the coral skeleton that serves as a proxy can be used to reconstruct the climate over hundreds of years ago (Cahyarini et al., 2009).

Coral skeleton composed of calcium carbonate precipitated by slow mechanisms, during the deposition process most of the heavy metal ions that enter the marine organisms through the food chain will be deposited in the coral skeleton along with the formation of aragonite structure. Ions of heavy metals will continue to be deposited in a coral skeleton for new growth will cover the old framework (Ali et al., 2010). Porites sp often used in determining the climate change study this is because these corals have large colonies of different bands and the annual and annual band is evident compared with other massive corals, and can grow for hundreds of years. Band on the coral provides a variety of information such as sea temperature, climate and pollution in the past (Cobb et al., 2003).

The content of geochemistry in modern coral, the Mg/Ca is known as a proxy for temperature. The previous researchers have proven that the Mg/ Ca in modern coral contained a track record of temperature in the surrounding environment (Mitsuguchi et al., 1996; 2002; Watanabe et al., 2001). To measure the concentrations of Mg and Ca is in coral skeletons using ICP-OES, the first thing that must be determined standard wavelengths. The most optimum wavelength is the wavelength that has the most excellent regression approaches 1. The aim of this study was to know the optimum of the wavelength to analyze Mg and Ca tracer of Porites lutea in ICP-OES.

2. METHODS

2.1 Standards

Before analyzing the content of Mg and Ca in coral skeleton, five standard solutions with different concentrations were prepared. Standard solution 1 were Ca: 5 ppm, Sr: 0.1 ppm and Mg: 0.01 ppm; standard solution 2 were Ca: 10 ppm, Sr: 0.2 ppm and Mg: 0.03 ppm; standard solution 3 were Ca: 20 ppm, Sr: 0.4 ppm and Mg: 0.05 ppm; standard solution 4 were Ca: 30 ppm, Sr: 0.6 ppm and Mg: 0.08 ppm; standard solution 5 were Ca: 40 ppm, Sr: 0.8 ppm and Mg 0.1 ppm. To destructive carbonate in coral skeleton has been used HNO3 25% and dissolved in DD water.

2.2 Procedures

Coral Porites lutea was taken from the flat reef area of Pantai Kondang Merak with size 30 cm in diameter and 13 cm in high. The massive coral P. lutea then cut into slabs with 1 cm thickness and soaked with 1 liter chlorine for 2 days until whitened. A piece of coral then dried using an oven with a temperature of 60 °C for 24 hours. After a perfectly dry slab of coral then exposed using X-rays to see the pattern of coral polyp and annual growth band of coral to determine the direction of growth as a guide track subsampling. By using a milling machine the coral slab drill 1 mm thick or called as subsampling. Each subsampling representative of 1 month of coral growth. Results subsampling produces 100 subsamples. Subsample powder that was obtained then analyzed the content of the elements Mg and Ca. Before analyses process each subsample (300 mg coral powder) dissolved into HNO3 25%, this preparation refers to a method performed by Watanabe (2001).

Measurement of standard solutions and samples using ICP-OES instruments ICAP 7000 series conducted at the Laboratory of Marine, Industry and Environment, BATAN. Standard solution and a subsample that was created, and then analyzed using ICP-OES instruments integrated with Thermo Scientific Qtegra Intelligent Scientific Data Solution. Standard solution vial is placed on the bottle and sorted on autosampler racks. The intensity of each element will be measured by ICP-OES with a wavelength of Ca (393.366; 396.847; 422.673; 317.933; 318.128; 370.603 and 373 690) and the wavelength has been used for Mg (279.553; 280.270; 285.213 and 279.079). Analysis 5 standard solutions will then generate a regression value of each wavelength. The wavelength used is the wavelength which has the highest value of regression. Regression of the most highly regarded as the most optimum wavelength for metal analysis of Mg and Ca contained in coral skeletons.

Here are the specifications of ICP-OES instruments ICAP 7000 series:

a. Fuel: Argon
b. This type of detector: Segmented-array of charge-coupled-device detector (SCD)
c. Plasma flow: 12 liters/minute
d. Nebulizer flow: 0.5 Liter / minute
e. RF Power: 1150 watts
f. Pump: 1.5 mL / min
3. RESULTS AND DISCUSSION

Based on the analysis of the optimum wavelength of 5 standard solution with ICP-OES instrument ICAP 7000 were Ca393.366 (R² = 0.9991); Ca396.847 (R² = 0.9991); Ca422.673 (R² = 0.9994); Ca317.933 (R² = 0.9998); Ca370.603 (R² = 0.9999); Ca318.128 (R² = 0.9998) and Ca373.690 (R² = 0.9999). Wave length of Mg279.553 has R² = 0.9523; Mg280.270 (R² = 0.9507); Mg285.213 (R² = 0.9547) and Mg279.079 (R² = 0.7904) respectively.

![Figure 1. Kurva Kalibrasi Unsur Mg 285.213](image1)

![Figure 2. Kurva Kalibrasi Unsur Ca 373.690](image2)

A total of 100 subsamples were measured with a wavelength of Mg and Ca were 373.690 and 285.213. The results showed concentrations of Mg and Ca contained in a subsample produce ratios of Mg/Ca was 3.18-4.78 mmol/mol. Mg/Ca’s ratio has been used to be a proxy of paleoclimatic, in the tropical area many previous of this ratio in range 3.77-5138 mmol/mol (Wei, 2000), 3.3-4.8 mmol/mol (Yu, 2005), and Mitsuguci (2003) in the range of 3.2 – 4.4 mmol/mol. Another wavelength was used by Arman (pers. Com) in Japan for Mg was 280.270 and Ca was Ca 370.603 or Ca 318.128 for optimum result.

4. CONCLUSIONS

ICP-OES can analyze the composition of up to 80 elements simultaneously with concentrations of up to parts per million (ppm). The most optimum wavelengths for analysis of the elements Mg and Ca in this coral was Mg285.213 R² = 0.9547 and Ca373.690 R² = 0.9999. The results of the ratio of Mg/Ca using these wavelengths minimum value of 3.18 mmol/mol and the highest value of 4.78 mmol/mol. In further experiments the ratio of Mg/Ca linked to sea surface temperature for studying past climate (paleoclimatology) of the content of coral geochemistry.

5. ACKNOWLEDGMENT

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6. REFERENCES


Heart Signal Measurement Design Base on Arduino with LabView

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Abstract – Some of human health’s indicators are heart signal. The designed tool determined the heart signal. It used ECG Shield Olimex sensors to measure the heart signal using LabView software. The measurement of ECG signal used Six Lead Method. Three electrodes used to measure the signal. Where the three electrodes placed in left ankle, right wrist, and left wrist. Three electrodes were Shield ECG showed the P, Q, R, S and T signal in heart signal. The signal amplitude were P wave was 0,13 mV, Q wave was 0,12 mV, R wave was 0,18 mV, S wave was 0,11 mV, and T wave was 0,12 mV.

Keywords: ECG Shield Olimex, Six lead Method,PQRST wave, Labview.

1. INTRODUCTION
Heart disease is still the number one of the cause of death in the world. The WHO’s Data in 2012 showed at least 17,5 million people or 34% death in the world because of this disease. In the meantime 80% of the death cause heart disease was heart attacks and stroke [1]. The successful of medication for heart disease depended on speed treatment. So the monitoring devices needed to handle the heart disease especially for the man with heart attack history [1].

Common heart monitoring device was electrocardiogram (ECG) used to assess the heart. The main advantages ECG were the capability to capture cardiac abnormalities, easy available in the hospital, and noninvasive test. The ECG signal could identify the information of heart such as heart rate, heart rhythm, heart muscle, heart attack history, coronary artery disease, or conduction abnormalities.

The data that gathered with ECG were significant and important. The data could indicate a heart attack or another possible coronary artery disease. ECG signal could show the earlier information to indicate heart disease. Extra measurement should be used to determine the anomalies, disease and best treatment. One of another tool to measure the heart signal was Echocardiogram.

Analog ECG captured the heart signal to obtain useful and information through the logarithmic chart. The chart related the function of the heart signal represent the voltage change of heart in real time. It showed particular value in the clinical cases such us Myocardial Infraction (heart attack), Pericarditis, Electrolytic transformations, Auricular and ventricular hypertrophy, Arrhythmias, Cardiac medicine effects, especially digital and quinidine, Generalized suffering affecting heart and blood, etc.

The portable digital medication tool needed to record the data and analyze the heart signal for accuracy and best treatment. The tool could show the heart signal in real time. Its data would be stored in digital format so it could be analyzed easily. The digital tool would show the heart signal in real time with period of time. It would be help the physician for medication and treatment in heart disease. Analog ECG took couple of minutes to show heart signal and digital ECG would show the signal immediately. So that, used a tool to determine heart rate in a short time and result value for an accurate reading. Measurement of heart rate using ECG olimex module and the result of the measurements are displayed on the labview program.

2. METHODS
Hardware used in this research were Arduino Uno R3, Electrode, Ultrasound Gel and Personal Computer (PC)
a. Arduino Uno R3

Arduino is a microcontroller module based on AT Mega the function to process data from any sensors or other input. Arduino Uno R3 at this research have any function such as for compute data from ECG module with analog input then convert digital output require function Analog Digital Converter (ADC) and serves to integrate the sensor data and then sent to the PC via serial port communication and a serial monitor.
b. Electrode

In this research electrode used to contact with the object or medium they contain no metal. The electrode made by conductor such as gold, silver, copper and aluminium. Electrode can integrated with Shield ECG Olimex. The electrode sent wave signal to Shield ECG Olimex and then processed.

c. Ultrasound Gel

Ultrasound gel serves as a catalyst to help speed up the process but not to react and to help clarify the object to be inspected waves making it easier to detect waves via electrodes.

d. Personal Computer (PC)

The instrument used in this study using PC hardware. PC in this research used to program the Arduino Uno R3 board and program labview as monitoring data and graphics.

Software used in this research were Arduino Ide 1.6.7, ElecGuru40 and Labview.

a. Arduino Ide 1.6.7

Arduino IDE has been used to make listing program then compile and upload the program on Arduino uno R3. Serial monitor was used to simulate the results of the program have been made.

b. ElecGuru40

In this research elecguru40 used to test current and Olimex Shield ECG electrodes. The software showed the Arduino base hardware in good condition. The aims were to determine what it's working according to need is to display the ECG waveform.

c. Labview

Labview is one of the visual programming language that uses icons instead of text to create applications. In this study labview is used to view the ECG wave, the wave analyzing and processing the data to make it easier in terms of readability.

2.1 Prototype ECG with Labview

This research includes several blocks of the design of hardware and software used to make the process of monitoring the heart rate. In this research use multiple sensors such as electrodes, ECG Shield Olimex and ultrasound gel, as a major component in the process of monitoring the heart rate. All data is the result of the detection sensor connected to the Arduino Uno microcontroller module to be processed and sent to the program interface in the PC using Labview.

2.2 Six Lead Method

1. Lead I = recording the potential difference between the right (RA) and left arm (LA), right-hand negative (-) and the left hand is positively charged (+).
2. Lead II = recording the potential difference between the right (RA) and left leg (LF), right-hand negative (-), and the left leg positively charged (+).
3. Lead III = recording the potential difference between the left arm (LA) and left leg (LF), right-hand negative (-) and the left hand charged (+).
4. Lead aVL = recording electrical potential on the left hand (LA), the left hand is positively charged (+), right hand (RA) and left leg (LF) formed indifferent electrode (zero potential).
5. Lead aVF = record the electrical potential of the left leg (LF), the left leg positively charged (+), the left hand (LA) and right hand (RA) zero.
6. Lead aVR = recording electrical potential at the right hand (RA), the right hand the positive (+), the left hand (LA) and left leg (LF) zero.

3. RESULTS AND DISCUSSION

Olimex Shield ECG testing is used to view and process signals from the sensor readings electrode on heart rate that occurs. Electrode sensor is used to detect cardiac signals on the pulse that existed at the wrists and ankles.

Figure 1 Shield EKG Olimex
Testing shield done for 3 times to find the appropriate cardiac signal characteristics. Early cardiac signals measured using elecguru software as a test tool to function properly and also as referesni early before going on labview software. Pictures of the test results shown in Figure 4 below.

![Figure 2 Signal pulse using elecguru](image)

Based on these tests may show that the results are measured by the sensor signal is not much different from the characteristics of cardiac signals. There is a wave interval P, Q, R, S and T at different distances generate the same wavele

Heart rate measurement using six leads. Six leads have PQRST wave, signal analyzers six leads can be calculated from the distance of each interval and the amplitude of the wave PQRST. In experiments using labview captured ECG data is as follows:

![Figure 3 Six Leads active](image)

Figure 3 shows the sixth lead active and viewed simultaneously. The graph looks perfectly clearly establish the P wave, Q, R, S and T.

The graph shown labview already provided in the form of a data logger to be converted into the form of a notepad or ms. Excel to get the number of readings labview.

![Figure 4 Data Logger in MS Excel](image)

On the data logger name six leads into the name analog inputs because of a program that was created on labview. Making calculations on the data logger does not have the slightest error because percent of data retrieval of measurement results in real time. To check the data logger is accurate or not, it can be made in the form of graphs that can be analyzed from each wave.

<table>
<thead>
<tr>
<th>Wave</th>
<th>Lead I</th>
<th>Journal</th>
</tr>
</thead>
<tbody>
<tr>
<td>P</td>
<td>0.13</td>
<td>0.015 – 0.12</td>
</tr>
<tr>
<td>Q</td>
<td>0.12</td>
<td>0.00 – 0.16</td>
</tr>
<tr>
<td>R</td>
<td>0.18</td>
<td>0.02 – 0.13</td>
</tr>
<tr>
<td>S</td>
<td>0.11</td>
<td>0.00 – 0.36</td>
</tr>
<tr>
<td>T</td>
<td>0.12</td>
<td>0.06 – 0.42</td>
</tr>
</tbody>
</table>
In the all Table obtained the calculation of the amplitude of each wave, the wave compared with the reference journal. Obtained from the calculation have the same results and accurate. On the wave of P, Q, R, S and T and Lead measurement results obtained there who have little difference, and there is that does not have the difference.

4. CONCLUSIONS

The result has been recorded with LabView and saved with Microsoft Excel data format. Heart signal showed voltage amplitude of PQRST wave. Fig 5. showed the PQRST wave from lead aVR, lead aVL and lead aVF. The signal represented from 3 second signal in heart signal from wrists and ankle. Amplitude from PQRST wave captured from the signal. Amplitude PQRST compared with normal amplitude ECG waveform [2].

5. REFERENCES

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A Low Cost Distributed Wireless Monitoring System
Implementing Bluetooth and Android Based Portable Devices

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Abstract – Nowadays, Android-based devices are becoming more and more popular portable electronic tools that are applied for many purposes. This research is aimed to develop a distributed wireless monitoring system of physical parameters by implementing Bluetooth and Android-based portable devices. The activity in this research includes design and development of a monitoring system, acquiring data and analysis. Some sensors were implemented to measure temperatures from several locations and connected to several microcontrollers at different locations. The microcontrollers would periodically read the temperatures from the sensors and send the digital data in a block of a data frame to a mobile phone as the portable device. It could be seen from some series of experiments that a low cost distributed wireless monitoring system could be realized. A small problem was however identified as the developed application on the portable device occasionally missed one or more characters sent by the microcontrollers. This problem was solved by the use of a block check character to recognize the integration of the data.

1. INTRODUCTION

Nowadays, Android-based devices are becoming more and more popular portable electronic tools that are applied for many purposes. Besides their uses as communication, many other applications including remote monitoring and control are commonly developed. Many Android-based devices include Bluetooth radio hardware. By using this hardware, a low-cost monitoring and control applications can be built; that is by running an intended Android application in the devices without additional hardware. As Bluetooth communication is a type of wireless communication, a distributed wireless monitoring system can be carried out with ease. Applications implementing Bluetooth technology are increasing during this time. Bluetooth is even developed and applied for IoT (the Internet of Things) where a large scale of a network containing electronic devices can be built [1]. Some application implementing Bluetooth technology is discussed in [2, 3, 4, 5].

This research is aimed to develop a distributed wireless monitoring system of physical parameters by implementing Bluetooth and Android-based portable devices. Some microcontrollers are implemented to perform acquisition of physical parameters and control the transmission of data to the portable device. The output of this research is a low cost distributed wireless monitoring system. This research is related to the previous research explained in [6].

2. DESIGN AND DEVELOPMENT

The activity in this research includes design and development of a monitoring system, acquiring data and analysis. Fig. 1 shows the design and developed monitoring system. Some DS18S20s were implemented to sense temperatures from several locations and connected to the microcontrollers. Outputs of the sensors were connected to the digital inputs of the microcontrollers. The microcontrollers, which were positioned at different locations, would periodically read the temperatures from the sensors and send the digital data in a block of a data frame to a mobile phone as the portable device. Wireless transmission of data between the microcontrollers and the mobile phone was enabled through the use of a Bluetooth module (HC05) for each microcontroller. An Android application was developed to receive and display the data from the microcontrollers on the mobile phone.

In the development of the system, the following software was utilized:

1. AVR Studio 7. This is an application program that was used to design and build the intended application that will run on the microcontrollers.
2. RAD Studio XE8. This software can be used to build an application that will run either on computers or portable devices such as mobile phones. In this research, the intended monitoring application that will run on the portable device was designed and developed using the RAD Studio XE8.
3. Serial Monitor. This is an application that was used for debugging and testing tool. The digital data sent
by the microcontrollers can be checked on a computer through the use of this software.

**Figure 1. The design and developed monitoring system.**

Fig. 2 depicts the flowchart of the developed program for the microcontrollers. It can be seen from this figure that the microcontrollers will perform endless and periodic temperature reading from the sensors. After that, the temperatures are periodically sent by the microcontrollers through the Interrupt Service Routine (ISR) mechanism. In the program for the mobile phone, a timer is implemented for checking the input stream containing the data from the microcontrollers; see Fig. 3. Whenever data are available, the routine will write and display them on the memo.

**Figure 2. Flowchart of the program in the microcontroller.**

**Figure 3. Flowchart of the program in the portable device.**

### 3. EXPERIMENTATION

The experiments in this research were divided into two steps:

1. Reading the temperature sensors.
2. Communication using the Bluetooth modules.
3. Temperature monitoring using the mobile phone.

The first experiment was intended to make it sure that the microcontrollers would send the block of the data frame correctly to the Bluetooth modules. For this purpose, a computer with the serial monitor application was used to receive the block of the data sent by the microcontroller. Fig. 4 shows the string of data sent by one of the microcontrollers where its measured temperatures using three sensors; see the column with the titles of CH1, CH2, and CH3. These temperatures were measured in degrees Celsius.

![Image of the string of data sent by one of the microcontrollers](image-url)

**Figure 4. The string of the data sent by one of the microcontrollers.**
In the second experiment, the microcontrollers were connected serially to the Bluetooth modules as depicted in the Fig. 5. The TX and RX pins related to the internal USART in the microcontrollers were utilized. The baud rate for serial communication was set to 9600 Baud. Then, the blocks of the data containing temperatures were sent to the Bluetooth module periodically. A mobile phone application called BlueTerm was used and run as terminal emulator to receive the data sent by the microcontroller over Bluetooth. The result of the experiment showed that all characters contained in the block of the data could be received and shown in the BlueTerm window. To be able to receive the data, the Bluetooth serial adapter in the mobile phone needed to be turned on.

The final experiment was intended to perform monitoring implementing all microcontroller modules and developed mobile application. In this experiment, six temperature sensors were used and positioned at different locations and two microcontroller modules as well as two Bluetooth modules were implemented. The monitoring was carried out in three steps:

1. Step 1: With time interval of 1 Second, from 15:40:44 to 15:41:02.

The setting of the time interval was carried out through the developed mobile application. Results of monitoring process which were displayed in the form of strings were saved in the memory of the mobile phone as text files. Figure 6 is an illustration of how the whole system was implemented during the final experiment.

4. DISCUSSIONS

Strings of the data as shown in Fig. 4 indicate that the developed application for microcontroller as well as its hardware could perform measurement of temperatures from several connected sensors. The microcontroller was able to recognize the existence of the connected sensors automatically and in this case, the sensors marked with CH1, CH2 and CH3 were detected. To be able to recognize the existence of the sensors, the microcontroller was programmed to perform a polling request of each sensor’s ID. The string ‘x,xx’ in the Fig. 4 indicates that the certain sensors were not available at that moment.

The block of the data sent by the microcontroller was formatted as depicted in the Fig. 7. The block is initiated by the character ‘.’. Following this character, measurement results for every sensor are added. A checksum (CS) is also implemented to detect the integration of the data. The end of the frame is indicated by character ‘E’.

![Figure 5. The connection between the microcontrollers and the Bluetooth modules.](image)

![Figure 6. The experiment using the whole system.](image)

![Figure 7. The format of the transmitted serial data.](image)

The performance of the whole system can be seen from the result of the final experiment where several monitoring processes were carried out. Several screenshots of the developed mobile application that was run on the mobile phone during the final experiment can be seen from Fig. 8. From left to right, the images show the monitoring data (temperatures in degrees Celsius) using time intervals of 1, 10 and 60 Second(s). In this case, the monitoring processes could be performed as expected. In the memo of the displayed interface, in each row, the 1st and 2nd characters (in this case S1 and S2) represent the IDs of the microcontroller modules. The 4th to 9th
characters (for instance 160539) indicate the measurement times, in the format of HH:MM:SS. The following characters contain information of temperatures for active channels.

Figure 8. Monitoring data displayed on the mobile phone using the developed application.

To be able to collect the data from several microcontroller modules, a polling method was implemented in the developed mobile phone application. In this method, a timer with its OnTimer event would periodically check incoming data through a stream from all connected Bluetooth modules; see Fig. 3.b. During the first development of the application, a small problem was however identified as the application occasionally missed one or more characters sent by the microcontrollers. The missing characters would cause failure on the related measurement results. To solve the problem, a checksum character was added in the format of the data block as previously depicted in the Fig. 7. The value of this character is calculated from the 2nd to the 72nd characters in the data block. By using this additional information, the mobile phone application could recognize the integration of the data block.

As the monitoring in this system was done implementing the Bluetooth technology, the monitoring could be easily performed through mostly portable devices containing Android Operating System. In this case, additional hardware interfaces would not be needed for users as most current mobile phones include Bluetooth interfaces. The use of the Bluetooth also enabled the distributed measurement as the microcontroller modules could be easily positioned separately.

5. CONCLUSIONS

It could be seen from some series of experiments that the system could generally work as expected. In this case, a low cost distributed wireless monitoring system could be realized. A small problem was however identified as the developed application on the portable device occasionally missed one or more characters sent by the microcontrollers. This problem arose as the application used a polling technique with the use of a timer to collect the data from the microcontrollers. The problem was solved by the use of a block check character to recognize the integration of the data.

6. REFERENCES

Induction Motor Protection and Starting System Using Programmable Logic Control Based on Star-Delta Method

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Abstract – The three-phase induction motors for applications in industrial machines have been widely used because it has a simple construction that is easy to maintain. The main problem is a three-phase induction motor starting current is high, for starting the motor absorbs a current of six to seven times the size of the full-load current. The starting phenomenon increases the voltage drop on the bus that caused some problems, such as the trip by a safety relay that is not expected. When the operator of three-phase motors disorder can occur in the form of high starting currents, current overload, short circuit, phase imbalance and phase failure regardless of the form of the supply phase induction motor. Starting currents generated from the wye-delta starting the generator coupling load of 1.76 A with a span of 2.2 seconds. Starting with a wye-delta method can reduce the starting current will be lower by 47.69% compared directly connected delta. The test results disruption, PFR works if one phase disorder or abnormal conditions. MCB automatically disconnect the system from the power source in case of a short circuit that has been tested. TOR trip after 1 minute 23 seconds after braking is made.

1. INTRODUCTION

Three-phase induction motors are used for applications in the machinery industry and have been widely used in the industry because it has a simple construction that is easy to maintain. The weakness of the three-phase induction motor is sufficiently high starting currents and low starting torque, to overcome it is necessary to use a method of starting off the right that can lower starting current [1]. During the process of starting, the motor requires a current of six to seven times the size of full-load current. The starting phenomenon, causing a voltage drop on the bus that caused some problems, such as the trip by a safety relay that is not expected. Therefore, the starting method is needed to reduce the starting current [2].

The three-phase induction motor has large initial flow characteristics but this can be resolved with some setting methods, one of which is using the starting of the star-delta method. The system is simple because it does not require a lot of support equipment and can be applied to any type of three-phase induction motor, but it is not uncommon on the starting method star delta there is a problem among others at the time of transfer of the star to the delta. This affects system performance. Thus, necessary to make a system of star delta starting of three phase induction motor based on plc (programmable logic controller) [3].

Automation systems grow increasingly complex [4]. The development of automation forming automation system toward a particular structure and establish an automated system. In the field of automation, PLC (Programmable Logic Control) is known as one of the tools of control. This tool not only able to replace the use of a relay as a control tool, but it also has many additional control functions. During this time, for the control of these systems are still using a conventional relay as a controller. One problem that often arises is that if one relay is broken, then the system will automatically stop and will only be run if the relay has been repaired. This process usually takes a long time. In addition, if the system is to be renewed, the whole system must be improved. Meanwhile, if using PLC time repair and renewal system is relatively short because simply replacing the system program can run again [5].

In all types of electrical systems always appear problems or disorders that can not be avoided, especially in induction motors. In all types of electrical systems always appear problems or disorders that can not be avoided, especially in induction motors. Disturbances that are often experienced by three phase induction motor, among others, over current that occurs when the engine first turned on, overload which occurs when the motor load, a short circuit between phases or phase to neutral, unbalance between phases, under voltage, and phase failure caused phase separated. The disturbance of three-phase induction motor is as often operated beyond capacity and a lack of regular maintenance by the operator or technician causing damage that results in the failure of the operation of the three-phase induction motors [6]. Based on the types of disturbances, in this study made a protection system on star delta starting of a three-phase induction motor in case of trouble starting of the three-phase induction motor.
2. RESEARCH METHODS

2.1 Design and Development Equipment

A general description of the instrument can be seen in (Figure 1) below.

Rotary encoders are electromechanical devices that can monitor the movement and position. Rotary encoder composed of a thin disk that has holes in the circular disc [7]. The relationship between the frequency and the rotational speed of the rotary encoder can be seen in Equation (1) [8].

\[ \text{Rotational Speed} = \frac{\text{Frequency}}{\pi} \text{ (rps)} \]  

(1)

Frequency to voltage converter circuit (F / V) serves as a counter-frequency output of the encoder and turn it into a voltage. The resulting voltage is directly proportional to the number of times the count. Based on LM2917 datasheet magnitude of the generated voltage can be calculated using Equation (2) [9].

\[ V_{out} = V_{sup} \times R_1 \times C_1 \times f_{in} \]  

(2)

2.2 Design of Power Circuit

The power circuit wiring diagram is shown in (Figure 2).

Starting with this method is the most commonly applied to motors three-phase induction large capacity. At the starting of this method aims to avoid big surprises asut flow [10]. At the time relationship Save the current flowing in the motor can be seen in equation (3).

\[ I_{LF} = \frac{V_{EF}}{Z} \text{; and } V_{EF} = \frac{V_1}{\sqrt{3}} \]  

(3)

While on the delta method of current flowing in the motor can be seen in equation (4).

\[ I_{\Delta} = \frac{V_{EF}}{Z} \text{; and } V_{EF} = \frac{V_1}{\sqrt{3}} \]  

(4)

It should be remembered that if you use this method, the switch is connected in the star should not be left too long, but must be converted into the delta method (switch function only star to reduce the high current at the time of starting of course). If the switch is left in the position of the star, the motor current will be determined by the
weight of the motors, with a current that will make the motors to overheat and eventually burn. Because of the position of the motor nominal current star is smaller than the nominal current on the delta-notch [10].

From (Figure 2) is described, contactor K1 is used as the main contactor. K2 Contactors are used as a switch delta and used as a switch K3 contactor star. When the system starts to work the motor works with relation stars where contactors K1 and K3 are working, reaching a maximum speed of the motor to work with the delta method where contactors K1 and K2 are working.

The PLC program design by GX-Developer software, the first condition when the operation process and the second condition nuisance when doing simulations. In the operation process conditions standby button serves as a prelude to starting the system, if the stand by the button is not pressed, the system can not work. The button serves to turn the motor on with the starting star delta. Impaired consists of three kinds, if an interruption occurs then the motor stops rotating and disturbance indicator lights up. Armpits during the indicator system can be restarted before pressing the restart button, when the restart button, the system can be revived. If no interruption occurs then the motor still spins on and off by pressing the off button.

3. RESULTS AND DISCUSSION

The design prototype delta star starting of three-phase motors that have been made can be seen in (Figure 3).

![Figure 3. Prototype Starting of Star-Delta Method](image)

From the testing that has been done nominal motor rotation speed of 1479 rpm for relations Reviewed by measuring motor speed using a tachometer. Motor speed in rps units are $1479 \text{ rpm} \times 0.01666667 = 24.65 \text{ rps}$

Using equation (5) and (6) are used to determine the value of the frequency of the motor speed in rps units are:

$$24.65 = \frac{Frequency}{16} (\text{rps})$$

$$Frequency = 24.65 \times 36 (\text{Hz}) = 887.4 \text{ Hz}$$

Measurement of frequency values is read at 884.6 Hz. Tests conducted to determine the performance of equipment failure relay phase protection that serves to secure the electric motor of voltage imbalance disorders such as under voltage, over voltage and voltage off. Tests conducted by the percentage of overvoltage and under a voltage of 5% and 10%. The results of data collection in the form of the voltage value shown in (Table 1).

<table>
<thead>
<tr>
<th>Tolerance</th>
<th>Phase Voltage (Volt)</th>
<th>Unbalance Voltage (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>R-S</td>
<td>S-T</td>
<td>T-R</td>
</tr>
<tr>
<td>+5%</td>
<td>404</td>
<td>399</td>
</tr>
<tr>
<td>+10%</td>
<td>406</td>
<td>418</td>
</tr>
<tr>
<td>-5%</td>
<td>361</td>
<td>361</td>
</tr>
<tr>
<td>-10%</td>
<td>347</td>
<td>340</td>
</tr>
</tbody>
</table>

PFR is set according to the value of the voltage that has been tested, with a value of 360 V under voltage and over voltage of 400 V. The results of the testing PFR are shown in (Table 2).

<table>
<thead>
<tr>
<th>No</th>
<th>Phase Voltage (Volt)</th>
<th>Note</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>R-S</td>
<td>S-T</td>
</tr>
<tr>
<td></td>
<td>404</td>
<td>399</td>
</tr>
<tr>
<td>1</td>
<td>406</td>
<td>418</td>
</tr>
<tr>
<td>2</td>
<td>361</td>
<td>361</td>
</tr>
<tr>
<td>3</td>
<td>347</td>
<td>340</td>
</tr>
<tr>
<td>4</td>
<td>386</td>
<td>382</td>
</tr>
</tbody>
</table>
From the test results show that in the (Table 2), found that the security PFR works well in accordance with the setting voltage is used, the PFR will trip after 5 seconds as a time of tolerance.

From a comparison chart that uses the current delta method has a very high starting currents with a rated current of 2.843 A. At the starting of the current delta-star-delta can be reduced to 1,397 A, the starting using the delta method save occur during 1.1 seconds. Displacement stars to delta work with the current settings of the motor speed reaches 1180 rpm or 80% of its nominal speed. Testing the starting star-delta with a load coupling with the generator has decreased the speed while the motor relation stars of 873 rpm, the condition is different from the testing without a load so that the motor can not move from the condition of the star to the condition of delta, in order to move from the condition of star into delta then setting sensor changed to the maximum speed of the motor to the load current stars relations. At the time of starting the motor is turned currents generated at the starting of this amounted to 1.76 A condition star, and 1.666 A on the delta method. When the motor operates in star method, the motor current is 1.3 A. The move to the delta method occurred during 2.2 seconds after the motor is turned on, when the motor runs in the delta motor speed up to 1384 rpm and the average current flowing in the motor of 1.1 A.

4. CONCLUSIONS
The conclusion of this research is at the starting of star delta starting current is reduced by 47.69% from the starting method of delta directly, and starting currents generated from the starting of star delta load generators coupling of 1.76A with a span of 2.2s. The third protection device works well, PFR works if one phase disorder or abnormal conditions. MCB work directly decided the system from the power source in case of a short circuit which has been in testing. TOR trip after 1 minute 23 seconds after the braking is made.

5. REFERENCES
The Effect of pH and Temperature On The Performance of Lead (II) Selective Electrode Based on S-Methyl N-(Methylcarbamoyloxy) Thioacetimidate

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Abstract – The effect of pH and temperature on the performance of lead (II) selective electrode membrane sensor based on a new S- methyl N-(methylcarbamoyloxy) thioacetimidate has been developed. S- methyl N-(methylcarbamoyloxy) thioacetimidate was chosen as a ligand in the membrane electrode, commonly named as methomyl. This compound was selected due to its tendency to form stable chelates with heavy metals ions of environmental concern. The membrane electrode consist of mixture a methomyl as ionophore, polyvinylchloride (PVC) and dioktilftalat (DOP) dissolved in tetrahydrofuran (THF) (1:3 w/v). It showed a good Nernstian slope of 29.2 mV/decade with linear concentration range of lead between 10^-5 M - 10^-1 M, and detection limit of 1.4 x10^-5 M or equal to 2.07 ppm of lead. By using the electrode, the objectives of this research were to learn the effect of pH and temperature on the performance of lead (II) selective electrode based on methomyl. The effects of pH and temperature was determined by measuring potential response of lead solution buffered by acetate solution at pH 2 - 8 and temperature of 20 - 50°C. The result showed that the performance of the sensor could be used optimally at pH 7, and it is not influenced by temperature range of 20 - 50°C.

Keywords : methomyl, ionophore, PVC membrane pH, potentiometry

1. INTRODUCTION

Lead (Pb) is one of the highly toxic heavy metal, which is still widely used in human activity that is in the process of mining, smelting and refining of metals, the result of industrial waste and emissions of motor vehicles [1]. Based on these activities, lead pollution is increased, causing environmental damage and excessive exposure in humans will lead to hearing loss, nervous system disorders, anemia and mortality [2], so that need require lead monitoring to prevent dangerous levels of lead poisoning and environmental damage.

The method of analysis of lead is often used atomic absorption spectrophotometry (AAS) methods [3]. This methods has advantages can analyze samples at low concentrations, high sensitivity and the use of relatively quickly. However, this methods has drawbacks for sample analysis of which can not be used for field analysis and require different cathode lamps for different metals. Additionally AAS methods less practical when used for analysis in the field. Therefore, these problem can be overcome using other simple methods such as potentiometry. Potentiometric methods commonly used is using the Ion Selective Electrode (ISE). Ion Selective Electrode is a working electrode that is able to measure selectively against certain ion. Potential that will measurably changed reversibly reactivity of ions determined [4]. Ion Selective Electrode is a very important part in the electrochemical sensor system, this is because the sensor tip contains an active material (ionophore) which the thermodynamic and kinetic events [5].

Lead (II) selective electrode have been developed by M. Ghaedi et al, 2011 [6] by using organic materials as ionophores. Other membrane support material used is polyvinylchloride (PVC), dibutylphthalate (DBP). Nernst Factor value resulting from these studies are still not close to the theoretical price (for the divalent ion is equal to 29.58 mV / decade of concentration) that need to be modified membrane of lead (II) selective electrode to produce Nernstian electrode.

Based on the results of previous studies, needs to be further developed lead (II) selective electrode with modification of ionophores. In this study, ionophores used are S-Methyl-N (Methylcarbamoyloxy) Thioacetimidate. This compounds was selected as ionophores due to interact very strongly to metal ions and have a tendency to form a stable chelate with heavy metals ions of environmental concern[7]. Lead (II) selective electrode based on methomyl has been developed by Noviana (2014). Based on these studies, the price Nernstian slope of 29.3 mV / decade concentration. This research obtained that lead (II) selective electrode based on S-methyl-N (methylcarbamoyloxy) thioacetimidate have wide linier concentration range with 1x10^-5 - 1x10^-1 M and the detection limit of 1.4x10^-5M or equivalent with 2.07 ppm lead (II). However, this sensor can
not directly applied for field measurement because it is not necessary to learn about the influence of environmental factors on the performance lead (II) sensor.

Several environmental factors that may affect the performance of lead (II) selective electrode such as pH and temperature. In this research, optimization of pH to determine the optimum conditions that can be used to detect lead to the maximum. Range pH used was 2 - 8 with the use of the addition of acetate buffer. This condition used because the ion lead (II) is stable in the pH range 3-7, and for methomyl is stable at pH neutral or slightly acidic atmosphere [13]. The effect of temperature have been studied in ranges between 20-50 °C which is the optimum temperature of the membrane methomyl. The membrane electrode consist of mixture a methomyl as ionophore, polyvinylchloride (PVC) as polymer and dioktilftalat (DOP) as plasticizer dissolved in tetrahydrofuran (THF) solvent (1:3 w/v). Variables of this research consist of range pH and temperature of the solution test.

2. METHODS

2.1 Materials and Equipment

The materials used in this study are S-Methyl-N-(Methylcarbamoyloxy) ThioacetimidatePb(NO$_3$)$_2$ pa (EmErck), dioktilftalat (DOP) (Sigma), polvininiklorida (PVC) (Sigma), tetrahdrofurant (THF) (E.merck), CH$_3$COOH pa (E.Merck), CH$_3$COONa pa (E.Merck), NaNO$_3$ p.a (E.Merck), HNO$_3$ 50% v/v p.a (EmErck), HNO$_3$ 65% (v/v) p.a (E.Merck), alcohol 96% (v/v) hydrochloric acid (HCL), sodium hydroxide (NaOH) and aquadest. Equipment used in this study are : lead (II) selective electrode based on S- methyl N-(methylcarbamoyloxy) thioacetimidatehave in characterization, electrode Ag/AgCl type HI 5313 Hanna, potentiometer (SchootGeräte CG model 820), pH meters (Hanna), alligatorclamps, analytical balance (Adventurer AR model 2130), centrifuges, oven, desiccator, the sample bottles, hot plate, and glass equipment

2.2 Fabrication lead (II) selective electrode based S-methyl-N-(methylcarbamoyloxy) thioacetimidate

Manufacture of membranes is done by mixing the constituents of the membrane which include S-methyl-N (methylcarbamoyloxy) thioacetimidate, PVC and DOP. After it was dissolved with THF with a ratio of 1:3 (% w/v). Membrane coated on Pt wire with a thickness of 0.3-0.5 mm followed by heating at 50 °C for 12 hours. Platinum wire that has been coated membrane is cooled and immersed with a solution of Pb(NO$_3$)$_2$ 1.5 M.

2.3 Potential measurement lead (II) selective electrode at pH 2-8

Solution of Pb(NO$_3$)$_2$ 10$^{-5}$-10$^{-1}$ M are taken as much as 25 mL, pH 2-8 potential was measured after 30 seconds. Lead (II) selective electrode potential effect of pH measurement performed three repetitions. Deviation of Nernst Factor value show that pH give the effect on the performance of lead (II) selective electrode based on S-methyl-N (methylcarbamoyloxy) thioacetimidate.

Temperature measurement lead (II) selective electrode at optimum pH

Measurements were made to solution Pb(NO$_3$)$_2$ 1x10$^{-5}$ - 1x10$^{-1}$ M by setting the temperature variation 20; 25; 30; 35; 40; 45; 50 °C. Decrease the temperature using a cooling solution in a bowl of ice cubes and an increase in temperature using an electric heater and thermostat settings using the temperature rise. The temperature of the solution was observed by using a thermometer. Deviation of Nernst Factor value show that temperature give the influence on the performance of lead (II) selective electrode based on S-methyl-N (methylcarbamoyloxy) thioacetimidate.

3. RESULTS AND DISCUSSION

3.1 Fabrication lead (II) selective electrode based S-methyl-N-(methylcarbamoyloxy) thioacetimidate

As for the basic characterization lead (II) selective electrode based on methomyl has produces Nernst Factor of 29.26 mV / decade of concentration, with a linear concentration range of 10$^{-5}$-10$^{-1}$ M and is able to detect cations lead up to 1.4 x 10$^{-5}$ mol/L, equivalent to 2,07 ppm of lead.

3.2 The effect of pH on performance lead (II) selective electrode based S-Methyl-N (Methylcarbamoyloxy) Thioacetimidate

In this study to determine the effect of pH on the performance of ESI lead (II) type coated wire-based S-methyl-N (methylcarbamoyloxy) thioacetimidate, was measured at a concentration of 10$^{-5}$ - 10$^{-1}$ M at pH 2, 3, 4, 5, 6, 7, and 8. The results showed that the effect of pH on performance of lead (II) selective electrode based on methomyl with measurement of lead nitrate solution concentration of 10$^{-5}$-10$^{-1}$ M at pH 2-8 is based on the
measured potential value can be known Nernst Factor. Then made the relationship curve between pH (x-axis) with the Nernst Factor as shown in Figure 1.

![Figure 1. Effect of pH on performance lead (II) selective electrode based on methomyl](image)

Theoretical Nernst Factor value for divalent ions is 29.6 mV/decade of concentration, with a minimum limit of 24.6 mV/decade of concentration and maximum 34.6 mV/decade of concentration. When the value is below the limit of Nernst Factor minimum or above the maximum value of Nernst Factor so the lead (II) sensor electrode is not Nernstian.

In Figure 1 shows that the pH approaching Nernst Factor is the pH of 7 is approximately 28.73 mV / decade of concentration. At pH 2, 3, 4 and 8 have Nernst Factor value below the minimum limit of the theoretical Nernst Factor is equal to 8.93 mV / concentration decade, 13.47 mV / concentration decade, 14.4 mV / concentration decade and 8.87 mV / concentration decade. At pH 5 and 6 have a Nernst Factor value of 26.6 mV / decade and a concentration of 28.27 mV / decade concentration. So the optimum conditions lead (II) selective electrode based on S-methyl-N (methylcarbamoyloxy) thioacetimidate achieved at pH 7. Because at pH 7 lead (II) selective electrode provides Nernst Factor value are approaching the theoretical Nernst Factor value is 28.73 mV / concentration decade.

3.3 The Effect of temperature on performance lead (II) selective electrode based on methomyl

Selective electrode capability test lead (II) ions in response to the lead (II) in the solution of the analyte carried out in the temperature range 20-50°C with variation margin testing every 5°C rise. The following graph of temperature relationship with Nernst Factors presented in Figure 2.

![Figure 2. Effect of temperature to performance lead (II) selective electrode based on methomyl](image)

Based on data obtained show that the temperature range of lead (II) selective electrode have Nernst Factor value that still about Nernst Factor value theoretical that allowed that is a range of 29.7 ± 5 mV / decade of concentration. Nernst Factor optimum value obtained at temperatures of 30°C. This condition caused by the mobility of ions in the test solutions is more increase so the potential response generated more better. The rising temperatures above 30°C gives the potential changes cause the value of the Nernst Factor is more lower, although still within the range allowed Nernst Factor value. In this study the methomyl membrane used with PVC as polymers that have a resistance to the effects of temperature. Therefore, the resulting membrane remained stable up to temperatures of 50 °C with Nernst Factor values still are within the range allowed.
4. CONCLUSIONS

Based on research that has been done, it can be concluded as follows:

1. Lead (II) selective electrode based on S-methyl-N (methylcarbamoyloxy) thioacetimidate have a Nernstian value optimum at pH 7 with Nernst Factor value of 28.73 mV / decade of concentration.

2. Performance lead (II) selective electrode based on S-methyl-N (methylcarbamoyloxy) thioacetimidate stable at range of temperature 20 – 50 °C.

5. REFERENCES

[1]. Fardiaz, S., 1992, Polusi Air dan Udara, Penerbit Kanisius, Jakarta
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