



PROCEEDING

The 2nd International Seminar of Basic Science
Natural Science For Exploration The Sea-Island Resources
Ambon, May 31st 2016



Organized by
Faculty of Mathematics and Natural Science
Pattimura University



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The 2nd International Seminar of Basic Science

“Natural Science for Exploration The Sea-Island Resources”

Poka-Ambon, 31st May 2016

**Mathematic and Natural Science Faculty
Universitas Pattimura
Ambon
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2nd edition

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Welcoming Address By The Organizing Committee

Today, We have to thank the The Almighty Allah SWT for the implementation of this international seminar. This is the second seminar about Basic Science in The Faculty of MIPA Pattimura University. The seminar under the title “Natural Sciences for Exploration the Sea-Island Resources” will be carried out on May 31st 2016 at Rectorate Building, Pattimura University. There are 200 participants from lecturers, research institute, students, and also there are 34 papers will be presented.

My special thanks refer to the rector of Pattimura University and the Dean of MIPA Faculty, Prof. Dr. Pieter Kakissina, S.Pd., M.Si. I also would like to express my deepest gratitude to Prof. Amanda Reichelt-Brushett, M.Sc., Ph.D. ; Kazuhiko Ishikawa, Ph.D. ; Nicolas Hubert, Ph.D. ; Prof. Dr. Kirbani Sri Brotopuspito ; Prof. Dr. Marjono, M.Phil. ; Gino V. Limon, M.Sc., Ph.D. as the keynote speakers.

The last, We hope this international seminar usefull for all of us, especially Mollucas People and very sorry if any mistake. Thank you very much.

Dr. La Eddy, M.Si.

Chairman of Organizing Committee

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Opening Remarks By Dean of Mathematic and Natural Sciences Faculty

I express my deepest gratitude to The Almighty God for every single blessing He provides us especially in the process of holding the seminar until publishing the proceeding of International Seminar in celebrating the 18th anniversary of MIPA Faculty, Pattimura University. The theme of the anniversary is under the title “Natural Sciences for Exploration the Sea-Island Resources”. The reason of choosing this theme is that Maluku is one of five areas in Techno Park Marine in Indonesia. Furthermore, it is expected that this development can be means where the process of innovation, it is the conversion of science and technology into economic value can be worthwhile for public welfare especially coastal communities.

Having the second big variety of biological resources in the world, Indonesia is rich of its marine flora and fauna. These potential resources can be treated as high value products that demand by international market. Basic science of MIPA plays important role in developing the management of sustainable marine biological resources.

The scientific articles in this proceeding are the results of research and they are analyzed scientifically. It is expected that this proceeding can be valuable information in terms of developing science and technology for public welfare, especially people in Maluku.

My special thanks refer to all researchers and reviewers for your brilliant ideas in completing and publishing this proceeding. I also would like to express my gratefulness to the dies committee-anniversary of MIPA Faculty for your creativity and hard working in finishing this proceeding, God Bless you all.

Prof. Dr. Pieter Kakisina, S.Pd., M.Si.

Dean of Mathematic and Natural Sciences Faculty

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“The 2nd International Seminar of Basic Science 2016”

Hotel Mutiara Ambon

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ANALYSIS AROMATIC COMPOUNDS OF CITRONELLA OIL BY USING NEAR INFRARED SPECTROSCOPY (NIRS) AND GAS CHROMATOGRAPHY-MASS SPECTROSCOPY (GC-MS)

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ABSTRACT

Generally, near infrared spectrum can give information concerning macromolecular high order-vibration energy like carbohydrate, fat and protein. Aromatic citronella oil is composed of compounds with carbon chains of which also has a vibration energy characteristic that is a stretch of near infrared energy range. This paper gives information about characteristics of citronellal, geranial, and citronellol as well as aromatic compounds using of near infrared transfectans spectroscopy that operated on 4,000-10,000 cm^{-1} . Aromatic compounds has been carried out measurements on citronella oil using NIR spectroscopy and gas chromatography mass spectroscopy (GC-MS). Method of measurement i.e. through measuring of spectrum of aromatic compounds on pure aromatic citronella oil, and the aromatic citronella oil also analyzed using of GC-MS. The results showed absorption area aromatics citronella oil ranges between 4000-6200 cm^{-1} . Results of the analysis by GC-MS showed fifteen aromatic compounds contained in the citronella oil with highest percentage citronellal compound (32.9%), geraniol (30.4%) and citronellol (14.3%) compounds respectively, as well as other twelve compounds of as the derivatives of citronellal, geranial and citronellol compounds as much as 22.4%.

Keywords: aromatic citronella oil, GC-MS, NIRS

INTRODUCTION

Indonesia is one of the largest essential oil producing countries in the world. In addition there are more than 40 types of essential oils that can be synthesized from a variety of typical plants that grow in Indonesia. Essential oil is one of the results of the rest of metabolic processes in plants which can be obtained from roots, bark, leaves, fruit, seeds (kernel) or flowers by vapor distillation, extraction or compressing so that the liquid state, and volatile (volatile oil). The composition of essential oils consists of the elements carbon (C), hydrogen (H), oxygen (O) and sometimes contain nitrogen (N) and sulfur (S). Never the less the essential oil still has resin and waxin small quantities as components that are not easily to evaporate.

Based on its cluster of compound components, the volatile oil is classified into two group namely hydrocarbon and hydrocarbon oxygenase. Hydrocarbon group comprises of

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the elements hydrogen (H) and carbon (C). Meanwhile, hydrocarbon oxygenase comprises of the elements carbon (C), hydrogen (H) and oxygen (O).

Aromatic citronella oil is included in the group of oxygenase hydrocarbon obtained from the distillation of lemon grasses or aromatic citronella plants (*Cymbopogon nardus* L), major components of the oil are citronellal (C₁₀H₁₆O), Geraniol (C₁₀H₁₈O) and citronellol (C₁₀H₂₀O). Volatile oil plays role in our daily life. Traditionally, the oil and main component as well as derivatives of aromatic citronella oil are used as mosquito repellent, fumigant (inhaled poison), it is also applied as fragrance material in foods, soaps and cosmetics (Nakahara *et al.*, 2003). According Sastrohamidjojo (2004), aromatic citronella oil can be applied in industry, mainly as fragrance of soaps, sprays, disinfectants, polish materials, a variety of technical preparation, and cosmetics. In addition, the aromatic citronella oil also used as an insecticide, nematicide, antifungal, antibacterial, exterminator of warehouse pest, and other fungi contaminant (Oroojalin, *et al.*, 2010; Regalado, *et al.*, 2011; Tombe, *et al.*, 2012; and Rohimatun & Laba 2013).

In Indonesia, production of volatile oil, in particular aromatic citronella, generally done in a simple manner and rarely followed up by a substance testing as part of production quality control. The quality control usually performed by GC-MS analysis and as a result, it is very costly, especially involve large number of samples. On the other hand, it is a time consuming and leaving chemical residues during the analysis process. Therefore, this method felt not support the need of fast and online measurements in the volatile oil industry. One potential method for these needs is Near Infrared Spectroscopy (NIRs). This allow the spectroscopic measurement of spectra on a stretch from 4000 to 10,000 cm⁻¹ which is the area where overtone and combination of vibration corresponding to the fundamentals vibration of O and H clusters that occurred in infrared area. Although the NIR spectrum from samples is rather complex in nature considering the broad area of absorbance as a result of vibration combination through the activity of stretching, deformation or bending that appears adjacently and repetitive, but give a unique characteristic for each compound or sample (Karlinasari, *et al.*, 2012).

NIR spectroscopy is advantageous because it offers non-destructive testing (NDT) to evaluate physical properties, mechanical and chemical (Burns and Ciurzak, 2008). Karlinasari, *et al.*, (2012), state that, NIR spectroscopy is also reliable, inexpensive, fast, and can describe the finger print of products. For this reason, the content of aromatic citronella oil expected to be analyzed using of NIR spectroscopy to make easier the testing method and reducing cost analysis.

This paper reports about the characteristics of NIR spectrum from aromatic citronella oil. In addition, the compounds content in the aromatic citronella oil as measured using of gas chromatography-mass spectroscopy (GC-MS) will be described.

MATERIALS AND METHODS

Material

The samples were aromatic citronella oil from PT. Djasula Wangi. No further treatment of the sample. For the measurements is using NIR and GC-MS respectively contains 2 ml sample.

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NIR Spectroscopy

NIRFLEX solid N-500 near infrared spectroscopy made by BUCHI is used for the measurement of content degree of aromatic citronella oil. The measurement technique used is transreflectance where 2 ml sample is placed in a petri dish and covered with reflector. The sample below reflector has 0.3 mm thickness. The sample is scanned 21 times on 4,000-6,200 cm^{-1} energy range, and then averaged to obtain proper ratio of signal / noise (s/n ratio). Resolution spectrum on this measurement is 4 cm^{-1} . The baseline corrected by performing first derivative, while the improvement of separation enhanced by second derivative. Each level of derivatives started with a smoothing spectrum using Savitsky-Golay polynomial technique of 3 order with 11 framing.

Gas chromatography mass spectrometry (GC-MS)

The constituent components of aromatic citronella oil from *C. nardus* plant obtained from PT. Djasula Wangi, analyzed using of GC-MS SHIMADZU QP-5000. Type of column used was Rastek RXi-5MS with length 30 m and ID 0.25 mm. Tool operation conditions using helium carrier gas with a flow rate of 0.3 ml/min and column pressure at 13.7 kPa. Heating temperature is at 70.0 °C on split injection model. Total flow: 80 ml/min, the column flow 0.50 ml/min and a linear speed 25.9 cm/sec.

RESULTS AND DISCUSSION

Analysis of Aromatic Citronella Oil Components

NIR spectrum acquired using transreflectance techniques with sample geometrical trajectory length is twice of the sample thickness. Therefore, the sample thickness is 0.3 mm, its geometrical length is 0.6 mm shows the NIR spectrum of aromatic citronella oil shown in Figure 1a. The absorption spectrum is shown by a valley. The lower the value the stronger the absorption. From these measurements it appears that the lowest transreflectance is 0.03397 which means that this spectrum has not yet reached its saturation condition, and thus the sample thickness is considered as sufficient. Such as NIR spectrum in general, overtone-vibration absorbance and combination vibrations that appear in NIR area always has a broad absorption feature due to those vibrations that are extremely adjacent.

Wide absorption that occurs on area at approximately 4100-4400 cm^{-1} and 5600-5900 cm^{-1} is a hydrocarbon chain in general identifier. Apparently, there are several noticeable special absorbencies, such as on 4060 cm^{-1} , 4540 cm^{-1} , 4820 cm^{-1} , 5696 cm^{-1} , and 6120 cm^{-1} . Since the other structures of peak spectrum are more difficult to identify, it needs second derivative spectrum. The second derivative spectrum is shown in Figure 1b. It needs to be recognized that the second derivative of spectrum will indicate locations of absorbance that cannot be determined accurately (because it is in form of hill and not of valley shape) on transreflectance spectrum firmly. On this second derivative of spectrum, location of valley shape on transreflectance spectrum will be indicated by the position of peak on the second derivative of spectrum. Meanwhile, position of the valley shape on second derivative of spectrum does not have physical meaning whatsoever.

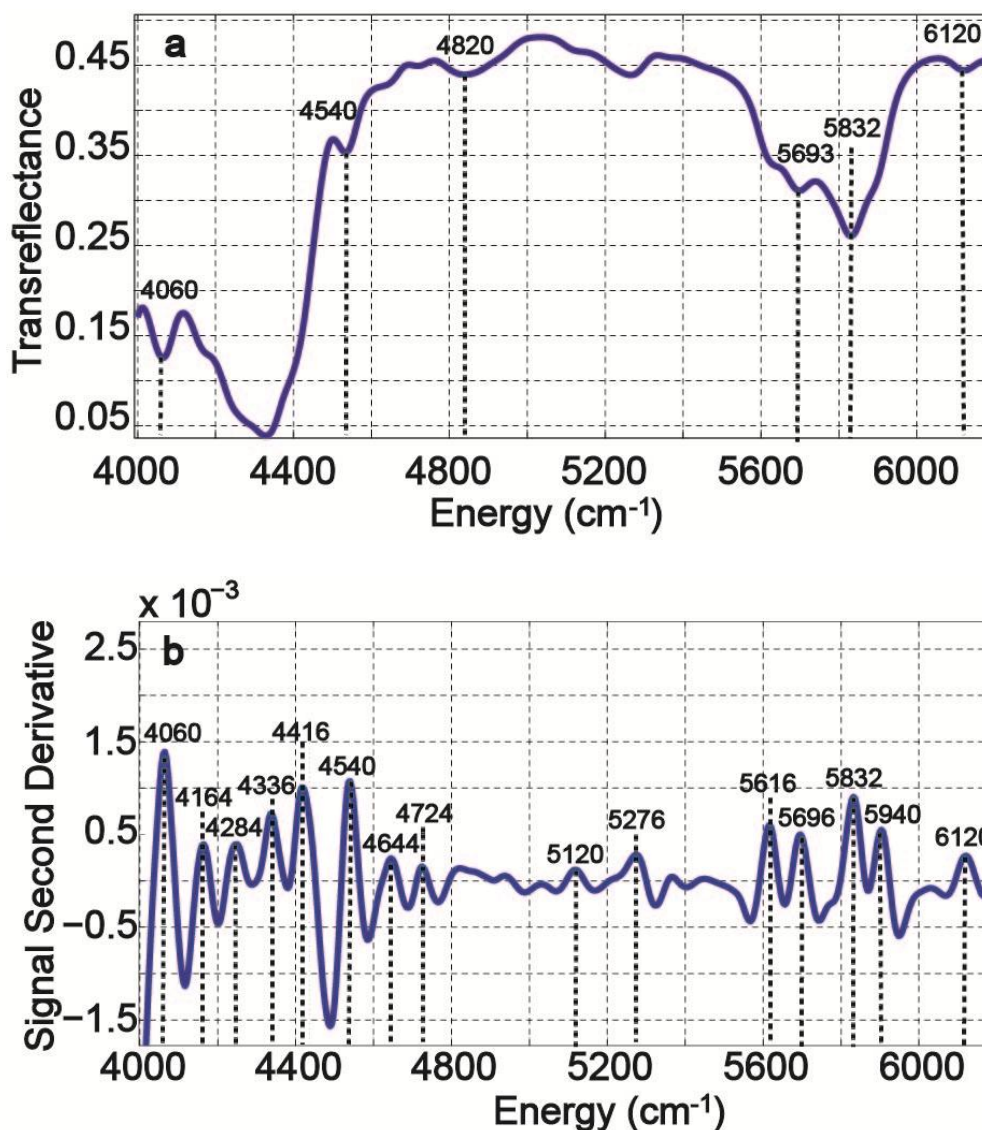


Figure 1. a. Transreflectance Spectrum, b. Second Derivative spectrum of aromatic citronella oil

On Figure 1b, it can be seen that there are several peaks. Those peaks show that aromatic citronella oil has an absorbance at 4060, 4248, and 4644 cm⁻¹ (C-H aromatic), 4336, 5904, 6120 cm⁻¹ (C-H aliphatic hydrocarbon compound), 4416, 4540, 5616, 5696, 5832 cm⁻¹ (C=O aromatic), 5276 cm⁻¹ (O-H polyvinyl alcohol). This data is supported by the results of Pazitny, *et. al.*, (2000); Workman and Weyer, (2007); Gaydou, *et. al.*, (2011). This results show that there are O-H and C=O clusters as characteristics of aromatic citronella oil compounds (consisting of C, H and O clusters). According to Guenther (2006), composition of aromatic citronella oil consists of types of terpen (fraction with low boiling level), citronellal (C₁₀H₁₆O), mixture of citronellal (C₁₀H₂₀O) and geranial (C₁₀H₁₈O) (*rhodinol*), types of ester, alcohol, *sesquiterpen*, and alcohol *sesquiterpen*.

Data tabulation of identification result of absorbance peaks on aromatic citronella oil is presented at table 1.

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Table 1. Chemical constituents of citronella oil pure by Workman dan Wayer (2008)

No	Wavenumber (cm ⁻¹)	Functional Groups	Type of Material
1	4060	C-H Aromatic C-H (aryl)	Aromatic compounds
2	4164, 4248	C-H Aromatic C-H (aryl)	Aromatic compounds
3	4336	C-H methylene C-H	Aliphatic compounds C-H, hydrocarbons
4	4416	O-H/C-H cellulose (OH and C-O)	Aromatic compounds
5	4540	C-H / O-H combination	Aromatic compounds
6	4644	C-H Aromatic C-H (aryl)	Aromatic compounds
7	4724	O-H/C=O the combination of the methanol	Methanol O-H
8	5120	C=O esters and acids (C=OOR)	Esther and acids compounds
9	5276	O-H hydrogen bonding between water and exposed polyvinyl alcohol OH	Aliphatic compounds C-H, hydrocarbons
10	5616	C-H methylene (CH ₂)	Aromatic compounds
11	5696, 5832	O-H/C-H=O combination	Aromatic compounds
12	5904	C-H methyl C-H, (CH ₃)	Aliphatic compounds C-H, hydrocarbons
13	6120	C-H vinyl C-H, associated with (CH ₂ =CH-)	Aliphatic compounds C-H, hydrocarbons

The data in Table 1 shows that the components of aromatic citronella oil consist of six large groups that is aromatic compounds either composed of C-H cluster chains or C-H-O cluster chains, C-H aliphatic compounds, hydrocarbons, O-H methanol, C=O ester compounds and O-H alcohol polyvinyl. Based on element of composing compounds, the oil composed of carbon (C), hydrogen (H) and oxygen (O).

Analysis of Aromatic Citronella Oil Componends Using of GC-MS

The aromatic citronella oil is analyzed using mass spectroscopy-gas chromatography and interpretation of results of GC-MS analysis of aromatic citronella oil is presented at Table 2.

From the analysis result of GC-MS, fifteen mass spectrums can be obtained which shows the existence of fourteen compounds, because on the 4th and 6th peak are the same compound. From those mass spectrums, it shown that three peaks with a relatively high spillover are citronellal, geraniol and citronellol compounds. Analysis result of the aromatic citronellal oil is supported by statement of Sastrohamidjojo (2004), that the chemical components in the aromatic citronellal oil are complex, but the most important components are citronellal, citronellal and geraniol.

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Table 2. Composition of aromatic citronella oil compounds based on the result of GC-MS.

No	Base Peak (%)	Compound name
1	32.87	citronellal (C ₁₀ H ₁₆ O)
2	30.44	Geraniol (C ₁₀ H ₁₈ O)
3	14.31	Citronellol (C ₁₀ H ₂₀ O)
4	4.18	Linalil acetate (C ₁₂ H ₂₀ O ₂)
5	3.69	Limonene (C ₁₀ H ₁₆)
6	3.32	Citronellil (C ₁₂ H ₂₂ O ₂)
7	3.15	Alpha-Copaene (C ₁₅ H ₂₄)
8	2.03	Citral (C ₁₀ H ₁₆ O)
9	1.54	delta-Cadinene (C ₁₅ H ₂₄)
10	1.22	alpha-Amorphene (C ₁₅ H ₂₄)
11	1.20	beta-Elementene (C ₁₅ H ₂₄)
12	0.78	alpha-Murolene (C ₁₅ H ₂₄)
13	0.72	Cyclohexane (C ₆ H ₁₂)
14	0.55	Germacrene (C ₁₅ H ₂₄)

Result of other research also support this finding as done by Koba et al., (2009), it shows that the highest percentage is Citronellal (35.5%), followed by Geraniol (27.9%) and Citronellol (10.7%). According to Guenther (2006), Javanese aromatic citronellal oil contains the following chemical components: citronellal (32-45%), geraniol (12-18%), citronellol (11-15%), geraniol acetate (3-8%), citronellal acetate (2-4%), limonene (2-4%), cadinen (2-4%) and the rest of them (2-3,6%) are *citric, Cavikol, eugenol, elemol, cadinol, vanillin, Kamfen, α -Pinen, linalool, β -Kariofilen*. Analysis result of aromatic citronellal oil obtained from PT. Djasula

Wangi shows that it contains the following components: citronellal (32.87%), geraniol (30.44%) and citronellol (14.3%), and other components are the derivatives compound of citronellal, geraniol and citronellol. The analysis result using of GC-MS shows the same result with previous research about the analysis of Javanese aromatic citronellal components.

CONCLUSION

Based on the results, it can be concluded that NIRs can be used for characterizing aromatic citronellal oil. The broad absorption that occurs on area at approximately 4100-4400 cm⁻¹ and 5600-5900 cm⁻¹ characterizes the existence of hydrocarbon chains in general. Apparently, there are several observable special absorptions on aromatic citronellal oil spectrum such as at 4060 cm⁻¹, 4540 cm⁻¹, 4820 cm⁻¹, 5690 cm⁻¹ and 6120 cm⁻¹.

GC-MS analysis has been performed to show that the aromatic citronellal oil at least contains at least fourteen compounds with the main compounds are citronellal (32.87%), geraniol (30.44%) and citronellol (14.3%), and the rest compounds (22.4%) are the derivatives compounds of citronellal, geraniol and citronellol.

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