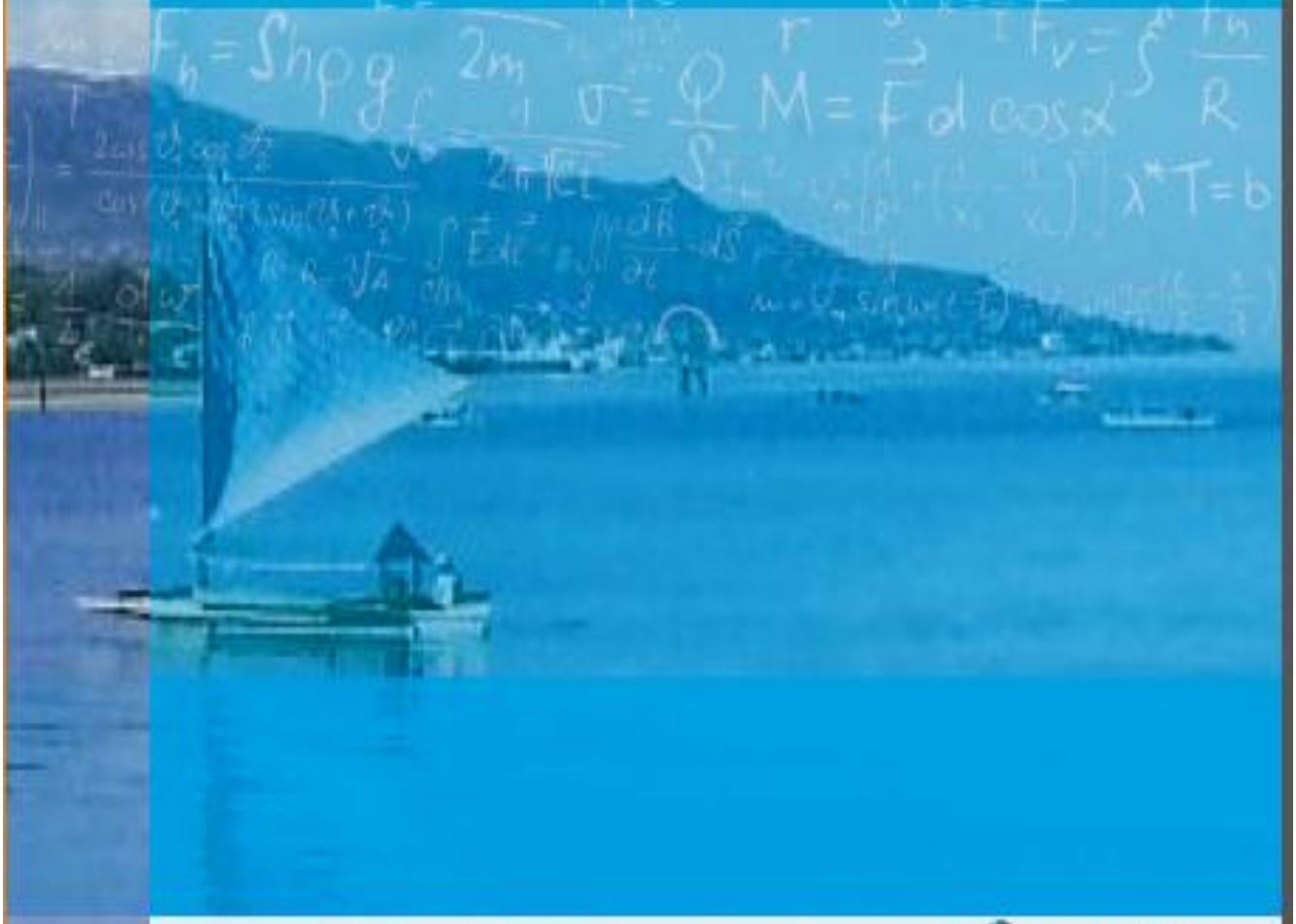
Basic Science
for Sustainable Marine Development

PROCEEDING
INTERNATIONAL SEMINAR 2015

Ambon, 3-4 June 2015



Organized by
Faculty of Mathematics and Natural Sciences
Pattimura University

PROCEEDINGS

1st International Seminar of Basic Science, FMIPA Unpatti - Ambon
June, 3rd – 4th 2015

ISBN : 978-602-97522-2-9

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October 2015

Welcoming Address by The Organizing Committee

The honorable, the rector of Pattimura University

The honorable, the vice rector of academic affair, Pattimura University

The honorable, the vice rector of administration and financial affair, Pattimura University

The honorable, the vice rector of planning, cooperation and information affair, Pattimura University

The honorable, all the deans in Pattimura University

The honorable, the key note speakers and other guests.

We have to thank The Almighty God for the blessings that allow this International seminar can be held today. This is the first seminar about MIPA Science in which the Faculty of MIPA Pattimura University becomes the host. The seminar under the title Basic Science for Sustainable Marine Development will be carried out on 3 June 2015 at Rectorate Building, the second floor. There are 250 participants from lecturers, research institute, students, and also there are 34 papers will be presented.

This International seminar is supported by the amazing people who always give financial as well as moral supports. My special thanks refer to the rector of Pattimura University, Prof. Dr. Thomas Pentury, M.Si, and the Dean of MIPA Faculty, Prof. Dr. Pieter Kakissina, M. Si. I also would like to express my deepest gratitude to Dr. Kotaro Ichikawa, the director of CSEAS Kyoto University, Prof. Bohari M. Yamin, University of Kebangsaan Malaysia, Prof. Dr. Budi Nurani Ruchjana (Prisident of Indonesian Mathematical Society/Indo-MS), Dr. Ir. A. Syailatua, M.Sc (Director of LIPI Ambon), and Hendry Ishak Elim, PhD as the key note speakers. We expect that this international seminar can give valuable information and contribution especially in developing basic science for sustainable marine development in the future.

Last but not least, we realize that as human we have weaknesses in holding this seminar, but personally I believe that there are pearls behind this seminar. Thank you very much.

Chairman

Dr. Netty Siahaya, M.Si.

Opening Remarks
By Dean of Mathematic and Natural Science 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 17th anniversary of MIPA Faculty, Pattimura University. The theme of the anniversary is under the title Basic Science for Sustainable Marine Development. 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.

Dean of Mathematic and Natural Science Faculty

Prof. Dr. Pieter Kakisina, M.Si.

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MORPHOLOGICAL DIVERSITY OF NUMEG MOTHER TREES AND SEEDLINGS IN LILIBOOI VILLAGE, AMBON ISLAND

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ABSTRACT

Banda nutmeg (*Myristica fragrans* Houtt) is native to the Moluccas (the Spice Islands) and has been known throughout the world because of its high quality essential oils from the seeds or the maces. In Lilibooi village, Ambon Island, there are very old nutmeg mother trees that show diversity based on their leaves and fruits. The objective of this study was to assess the diversity in nutmeg trees based on leaf, fruit, seed shell and mace morphological markers, and to assess the diversity of seedlings originated from the mother trees. This research used a survey method with selection of mother trees to be observed by purposive sampling. Five mature fruits from each sample mother tree, and 8 months old seedlings in the nursery belonging to the Titasomi farmer group were used as data sources in the observation. Morphological characterization was based on the Tropical Fruits Descriptors as modified, and other information was gathered through a Focus Group Discussion involving nutmeg farmers. The results showed that there were nine mother trees in Lilibooi that showed diversity in terms of the morphologies of leaves, fruits, seeds, and maces. Highly diverse characteristics were shown in leaf shape, leaf tip shape, leaf size, fruit shape, fruit length, shape of fruit stalk, fruit flesh thickness, seed length, mace thickness and mace compactness on the seeds. A high diversity of leaf and stem morphologies was also observed in the seedlings originated from the mother trees.

***Keywords*:** nutmeg, diversity, mother trees, seedlings, Ambon

INTRODUCTION

Indonesia is the center of origin of some species of the genus *Myristica* (Purseglove *et al*., 1981; Weiss, 2002). Banda nutmeg (*Myristica fragrans* Houtt) is native of the Moluccas and was introduced to India and the British since the 18th century. This type of nutmeg is commonly cultivated species in Indonesia, India, Grenada, and Malaysia because its seeds and maces have the best quality (Heyne, 1987). This spice crop has become the attention of the world for along time for its essential oil. The essential oil has an especially rich aroma. The essential oil that is mainly produced from seeds and mace is used for medicinal ingredients and spices.

Banda nutmeg trees are usually dioecious, meaning the female flowers are produced by a female plants that is different from a male plant that produce only male flowers. However, there are some bisexual nutmeg plants with monoecious flowers, which produce fruits without male flowering plants. Krishnamoorthy *et al* (1996) found that from sex segregation of 90 offsprings, 40 unisexual male plants, 45 unisexual female plants and 5 bisexual plants were

obtained. As a consequence of the flowering manner, nutmeg fruits are produced from cross-pollinations. Cross-pollination is one of the causes of diversity. Diversity can be observed visually by morphological and agronomic markers. Soeroso (2012) found 3 accessions of *M. fragrans* Houtt that were different in the forms of fruits, seeds and canopies. This variation indicates the genetic diversity and its potential needs to be explored. Maluku as the center of origin of Banda nutmeg Maluku is expected to have a high genetic diversity.

Identification of nutmeg is an important step for future improvement of this plant as well as for its conservation. Characterization of plants can help to identify the diversity as well as to determine the morphological markers. Morphological markers used may be a simple marker, such as shape, color, size, and weight. Fruit characteristics, such as fruit flesh thickness is related to its utilization for making candy and pickles, whereas seed and mace sizes are related to the content level of essential oil that is produced. Information about each mother tree and uniformity of seedlings that are derived is important to determine the mother trees that need to be maintained as seed sources. The objectives of this study are: (1) to obtain data on morphological variability of leaves, fruits, seeds and mace from several mother trees, and (2) to assess the variability on seedlings originated from the mother trees.

METHODS

This research was conducted in Lilibooi village, West Leihitu Subdistrict, Central Maluku. This research employed a survey method of determining the mother trees by purposive sampling in the nutmeg forest. While morphological data retrieval 8 month old seedlings in the garden in the village farmer groups Titasomi Lilibooi. Networking information includes nutmeg morphological characteristics based on Tropical Fruits Descriptors (IPGRI, 1980) as modified by Soeroso (2012), as well as other information based Focus Group Discussion.

Nine mother trees (9 accessions) as sources of seeds, as as assigned by the BPSB Maluku (Seed Certification Board), used in this characterization study. From each mother trees, 5 mature fruits and 5 full-size leaves were taken for characterization. Sixty seedlings were sampled from approximately 10,000 seedlings in the nursery. Characterization of leaves, fruits and seeds included shape, color, texture, and size. Characterization of seedlings was conducted on plant height and leaf morphology. Qualitative data collected were then tabulated and presented in the form of tables and figures. Descriptive analysis was subsequently conducted on these data.

RESULTS AND DISCUSSIONS

Morphological Diversity of Leaves

Qualitative characteristics such as leaf shape, leaf surface texture, shape of leaf tip, and leaf size (Table 1) showed the diversity among the nine accessions of the mother trees in Lilibooi village. High diversity among accessions in leaf shape, leaf surface texture, leaf tip shape and leaf size were indicated by 3-4 phenotypic classes. The characteristics of the other leaf morphological traits had only two phenotypic classes. LBooi-6, LBooy-7 and LBooi-9 accessions have oblong elliptical leaf shape and pointed leaf tip with a small leaf size, but the three accessions differs in the appearances of leaf, shape of petiole base and leaf surface texture. Meanwhile, the other six accessions have an elliptical and oval leaf shape with relatively larger leaf sizes. These different leaf morphological characteristics that among the accession indicate the presence of genetic diversity, and can be used as a morphological marker for each mother plant.

Table 1. Morphological Characteristics of Leaves of Nutmeg Mother Trees

No. of Mother Tree	Leaf Shape	Leaf Surface Texture	Leaf Color	Leaf Tip Shape (Sharpness)	Shape of Petiole Base	Leaf Size (cm^2)
LBooi-1	elliptical	smooth, dull & not bumpy	dark green	pointed (1.0-1.5 cm)	looped	large (111-120)
LBooi-2	elliptical	smooth, shiny & bumpy	dark green	pointed (> 1.5 cm)	looped	medium , (100-110)
LBooi-3	0val	smooth, dull & bumpy	green	pointed	looped	very large (>120cm)
LBooi-4	Oval	smooth, shiny & not bumpy	dark green	short (< 1.0 cm)	looped	very large
LBooi-5	elliptical	smooth, dull & moderate	green	pointed	looped	large
LBooi-6	oblong elliptical	smooth, dull & not bumpy	green	slightly pointed	looped	small (<100)
LBooi-7	oblong elliptical	smooth, shiny & moderate	dark green	slightly pointed	looped	small
LBooi-8	oval	smooth, shiny & bumpy	dark green	short	looped	large
LBooi-9	oblong elliptical	smooth, shiny & not bumpy	dark green	slightly pointed	flat	small

The largest leaf size was shown by LBooi-3, LBooi-4 mother tree accessions followed by LBooi-8, which indicated productivity potentials. Large leaf size contributes to generating more photosynthic product which in turn will enable increased fruit as well as oil productivity. According Wardiana *et al* (2008) the size of the leaf is also one of the important characters for selection, in addition to the canopy size, branching and tree height.

Morphological Diversity Fruits, Seeds and Maces

Nutmeg fruits from the nine mother trees showed diverse morphologies. The fruit shapes were generally spherical, and only 3 accessions had oval and ovate fruit shapes. Fruit length and diameter were also diverse. Long fruits were found in some mother tree accessions such as LBooi-6 and LBooi-9 (Table 2). The shapes of fruit bases were found to be a special characteristic of each accession. Some had straight, bending forward, or with a bump on the back of the fruit bases, as those seen Lbooi-5 and LBooi-7 accessions.

The colors of fruit skin color varies from light yellow, yellow to yellowish-green, with smooth and dull skin surface texture. Grooves on the fruit medians also varies from flat, deep, or very deep, and most of the mother tree fruits tended to have flat slope (Table 2). The Research results of Soeroso (2012) showed that the nutmeg accessions from Patani and Tidore also had a high diversity in term of fruit shape, ripe fruit color and seed shape.

The parts of a nutmeg fruit consists of fruit flesh, mace, seed shell and seed. The description of Banda nutmeg fruit is as follows: round to slightly oval shaped with fruit length between 1-10 cm, thin to moderately thick fruit flesh with creamy white flesh color. The seeds have a hard seed coat (shell) and covered by seed arilus, which is better known as mace, with an aromatic characteristic by its myristicin content (Arrijani, 2005; de Guzman and

Siemonsma, 1999). The utilized parts of the nutmeg fruit consist of fruit flesh, seed shell, mace and seed content (Purseglove *et al*, 1981). Nutmeg accessions from Lilibooi which have thick fruit fleshes. Fruit fleshes from the four accessions of nutmeg with the thick flesh characteristic can be used as raw material for processed snack (nutmeg pickle). In some places in Maluku, the fruit flesh has been processed into fruit candies, pickles, nutmeg wine, and jam, in a home industry it has not been widely recognized by the public.

Table 2. Morphological Characteristics of Fruits of Nutmeg Mother Trees

Number of Mother Tree	Fruit Shape	Fruit Diameter /Length	Fruit Skin Color	Fruit Flesh Thickness (cm)	Fruit Base Shape	Groove Fruit Median
LBooi-1	round/spherical	4.7cm / 5.8cm	Yellow	1.0	straight	rather deep
LBooi-2	round	4.9cm /5.1cm	light yellow	0.9	straight	flat
LBooi-3	oval	4.3cm /6.0cm	Yellow	1.0	straight	rather deep
LBooi-4	round	5.0cm /6.0cm	Yellow	1.1	bended forward	flat
LBooi-5	short round	4.8cm /5.2cm	yellowish green	1.2	bended forward with a bump	flat
LBooi-6	round	4.7cm /6.5cm	light yellow	1.1	bended forward	flat
LBooi-7	short round	4.6cm /5.2cm	Yellow	1.0	bended forward with a bump	flat
LBooi-8	round	5.1cm /5.5cm	light yellow	1.2	straight	flat
LBooi-9	ovate	5.2cm /6.2cm	Yellow	1.3	bended forward with a bump	deep

As a source of seeds, Banda nutmeg in Lilibooi village had seed characteristic with diameter of 2.10-2.70 cm and with seed lengths varies among accessions. Based on the seed length and diameter, the accessions with large seed size included LBooi-1, LBooi-2, LBooi-3 and LBooi-4. Thick maces were found in LBooi-4 and LBooi-8, which were supported by fullness of maces covering the seed. It is expected that the accessions with large seed sizes and with thick maces will have a great potential in relation to oil production. Although, research of Soeroso (2012) proved that thick red mace of *M. fragrans* Houtt from North Maluku produced nutmeg oil yield lower than the thin red mace. LBooi-3 and other four nutmeg mother trees have thin maces. In Lilibooi village, nutmeg plants with thin maces were used as the mother trees because they produced more fruit yields compared to the plants with thick maces. Nutmeg oil is an essential oil that can be obtained from nutmeg seeds and maces by distillation method. This essential oil has the distinctive quality due to the presence of the main component in the aromatic essential oil nutmeg called myristicin. Soeroso (2012) found the highest myristicin content from maces of *M. Fragrans, M. succedanea* Reinw and

Myristica sp. Oil content of seeds of M. fragrans Houtt ranged from 10.92 to 11.57 % , while the oil content of maces ranged from 19.60 to 21.30 %.

Table 3. Characteristics of Seeds and Mace of Nutmeg Mother trees in Lilibooi

Mother tree No.	Seeds with Shell		Mace	
	Diameter (cm)	Length (cm)	Mace Thickness (mm)	Mace Compact-ness on Seeds
LBooi-1	2.40	3.20	1.00	sparse
LBooi-2	2.70	3.35	1.00	very sparse
LBooi-3	2.30	3.10	0.50	very sparse
LBooi-4	2.60	3.20	1.62	dense
LBooi-5	2.50	3.05	1.00	very sparse
LBooi-6	2.20	3.00	1.00	very sparse
LBooi-7	2.10	2.70	1.25	sparse
LBooi-8	2.25	2.85	1.80	very dense
LBooi-9	2.25	2.80	1.25	sparse

Morphological Performances and Development of Nutmeg Seedlings

Seedling performance generally showed the presence of morphological diversity in the nurseries. Seedling leaf shapes were elliptical, oblong elliptical, obovate and oblong, with oblong elliptical leaf shape being the dominant one. Leaf surface textures also varied into 4 phenotypic classes, with smooth dull and not bumpy surface being the dominant. Most of the seedlings had pointed leaf tips, with a green leaf color; although those with dark green and yellowish green leaves were also found. Most of the seedlings had small leaves less than 70 cm. Seedling height ranging from 25 to 35 cm was dominant. Three leaf morphological performances of seedlings that were different from the mother trees included leaf shapes i.e. obovate and oblong obovate, yellowish-green leaf color and leaf size.

The differences in morphological performance in the seedlings were caused by genetic and environmental factors. The causes of diversity due to genetic factors were: (1) the seeds were originated from a mixture of nine mother trees, (2) the relatively close spacing of trees (6 x 8 m) which allowed cross pollination among the mother trees, (3) genetic recombination due to sexual reproduction. The environmental factors that affected morphological performances of seedlings were (1) 8 month old seedlings were still grown in poly bags of 12 x 18 cm with limited soil media, (2) the distance between adjacent seedlings were very close, (3) fertilization and pest control were limited. These factors inhibited the growth and development of the seedlings as shown by their short height and small leaf size and the yellowish green leaf color.

The results of this study showed that the uniformity of seedlings developed by Titasomi farmer groups was not guaranteed. Seedling uniformity is one of indications of seedling superiority to become productive trees. Seedling production may be a problem when the trees from them have low and diverse yields. This is because the high morphological diversity of seedlings can not guarantee their yield superiority. However, diversity of seedlings from the selected mother plants that are isolated from unwanted pollen may be able to guarantee their superiority.

Table 4. Morphological Performances of Nutmeg Seedlings from Mother Trees

Number of Samples	Leaf Shape				Leaf SurfaceTexture			
	Elliptical	Oblong Elliptical	Obovate	Oblong Obovate	Smooth, shiny & bumpy	Smooth, shiny & not bumpy	Smooth, dull & bumpy	Smooth, dull & not bumpy
60 seedlings	15%	63%	7%	15%	7%	8%	25%	60%

Continuation of Table 4.

Number of Samples	Leaf Color			Leaf Size				Plant Height		
	Green	Dark green	Yellowish green	Very large (> 100cm^2)	Large (85-100cm^2)	Medium (70-84cm^2)	Small (< 70cm^2)	Tall (> 35cm)	Medium (25-35cm)	Short (<25 cm)
60 seedlings	65%	17%	18%	5%	15%	25%	55%	17%	55%	28%

Male trees in Lilibooi nutmeg plantation have been cut before reaching flowering stage in the past. Therefore, the trees that produce fruits are possibly female, monoecious (male and female flowers on the same plant), or trimonoecious plants (male, female and hermaphrodite flowers on the same plant). Nutmeg is usually a dioecious plant, however, occasionally monoecious and trimonoecious types are found (Purseglove *et al*., 1981; Soeroso, 2012).

CONCLUSIONS:

1. Nine nutmeg mother trees in Lilibooi village showed morphological diversities in leaves, fruits, seeds and maces. Mother trees of Lbooi-4 and Lbooi-8 had the larger size of leaves and fruits than the others, also their maces were thicknest. Lbooi-2 showed the biggest size of seeds, leaves and its mace thickness was moderate, and its fruit flesh was most thin. Whereas Lbooi-6 and Lbooi-9 were related in its potentials on fruit size and fruit flesh thickness.
2. Diversity among the seedlings from the mother trees was found in all characterized leaf morphologies. The morphological variability of seedlings that were not found in the mother trees were obovate and oblong obovate leaf shapes, and yellowish green leaf color.

ACKNOWLEDGEMENTS

We thank Titasomi Farmer Group in L Lilibooi and Ir. J. W. Hetharion (head of the group) for permitting us to conduct this study and collect data in their nurseries and nutmeg plantations.

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