Profiling of Secondary Metabolites of Endemic Aromatic Rice in Enrekang Variety of Pulut Mandoti Emas (PME)

Hafsan Hafsan[1], Masriany Masriany[1(✉)], Afridha Sari[1], Selis Meriem[1], Eka Sukmawaty[1], and Bram Kusbiantoro[2]

[1] Department of Biology, Faculty Science and Technology, Universitas Islam Negeri Alauddin Makassar (UINAM), Kabupaten Gowa, Indonesia
masriany.musa@uin-alauddin.ac.id
[2] Balai Besar PADI, Kabupaten Subang, Indonesia

Abstract. Rice (Oryza sativa L.) Pulu Mandoti Emas (PME) variety is an endemic rice typical of Enrekang Regency, South Sulawesi which has a distinctive taste and fragrant aroma. The fragrant aroma of rice is generally caused by the content of secondary metabolites. The purpose of this study was to determine the content of secondary metabolites by histochemical analysis and GC-MS on the organs of roots, stems, leaves and seeds and to determine the effect of environmental abiotic factors on the production of secondary metabolites in PME rice varieties. The results showed that from histochemical tests on roots, stems and leaves found the presence of alkaloids, phenolic compounds, terpenoids and lipids in the epidermis, cortex, parenchymal tissue and xylem phloem transport vessels. In the results of the GC-MS analysis using the Solid Phase Micro Extraction (SPME) method, it was found that there were compounds suspected to be important volatile compounds that play a role in producing the aroma and taste of PME, namely benzaldehyde, 2-Methoxy-4-vinylphenol, Furan 2-pentyl-, hexanal, nonenal, 2-nonenal, octanal, 1-Octen-3-ol, anethole. This secondary metabolite profile becomes the basis information for further testing of the metabolite biomarkers responsible for producing the distinctive aroma of PME rice.

Keywords: Aromatic Rice · Secondary metabolite · 2 Acetyl Pirolin · Pulut Mandoti Emas

1 Introduction

Pulu Mandoti Emas (PME) Rice is an aromatic rice found in Enrekang Regency, South Sulawesi. This rice has potential in the development of food crops with a local rice variety that is endemic. Endemic means that if this rice is grown in another region, it will not produce the same quality, taste and aroma as that grown in its original area. This rice has its own uniqueness because it is fluffier glutinous rice with a sharp and distinctive aroma that makes this rice a rice that has a market price above the average price of glutinous rice. PME rice variety grows at an altitude of 700 m above sea level which

T. D. Larasati et al. (Eds.): ICTROPS 2022, ABSR 31, pp. 312–326, 2023.
https://doi.org/10.2991/978-94-6463-180-7_34

is cultivated in 5 Hamlet 2 Villages, Baraka District, Enrekang Regency. In addition, this rice cultivation technique still uses traditional methods and does not use inorganic fertilizers so that the texture and aroma of this rice can be maintained. The pungent smell makes this rice cookable together with ordinary rice with a concentration of 1 L of pulu mandoti rice mixed with 40 L of ordinary rice [1].

One of the villages that produces PME rice is Salukanan Village. Salukanan Village has about 250 hectares of 312 hectares of rice fields planted with Pulu Mandoti rice. This rice only has a harvest period once a year with a vulnerable time of planting to harvesting of 6 months. Based on the data, it shows an increase in the price of rice. It is known that in 2015 the price of Pulu Mandoti rice was Rp. 35,000, 2016 for Rp. 60,000 (Latif *et al.,* 2020) [1], and in 2020 it increased to a price of Rp. 70,000 (Karim, 2020)[2]. Sukmawaty *et al.,* (2022) reported that the rizosphere bacteria in vegetative phase of PME was dominantly by *Candidatus koribacter* while at the reproductive phase was dominant with *Aquisphaera* [3].

PME rice is one of the aromatic rices that is in great demand by the public with its distinctive and strong taste and aroma. However, this variety of rice has not been explored much, including the various types of compounds contained in it. Therefore, it is necessary to identify the analysis of the content of secondary metabolites both histochemical and metabolomically.

1.1 Research Time and Location

This research was conducted in May - September 2021. Observations of abiotic factors were carried out when sampling at one of the rice fields in Gandeng Hamlet, Salukanan Village, Baraka District, Enrekang Regency. Samples of plant organs were prepared and histochemical tests were carried out at the Botanical Laboratory of UIN Alauddin Makassar. Secondary metabolite analysis was carried out at the Flavour Laboratory of the Indonesian Center for Rice Research (BBPadi) Subang, West Java.

2 Methods

This research was conducted in two main stages, namely: Histochemical test and Volatile metabolite analysis. Histochemical test was carried out according to the method used by Sari (2021), Trimanto and Hapsari (2021) which consisted of alkaloid, terpenoid, lipid and phenol test [4].

The distinctive aroma compounds in PME variety were analyzed by Solid Phase Microextraction (SPME) to determine the profile of the volatile compounds contained in the rice. The SPME method has been widely used to determine volatile compounds in a sample, Masriany *et al.*(2020) used this SPME method to determine the profile of secondary metabolites in banana flower volatiles [5].

Analysis of volatile compounds components of rice samples used Fiber Solid Phase Microextraction (SPME) combined with Gas Chromatography Mass Spectroscopy (GC-MS) [6]. In the method using SPME, rice samples are processed in the form of rice. Rice samples are cooked in a ratio of 1:3 rice and water to form rice [7]. 3.5 g of rice sample was put into a 22 ml SPME vial + 5.25 ml millique water. Subsequently, the extraction was carried out by heating at a temperature of 90˚C for 60 min. The fiber used is SPME fiber DVB/CAR/PDMS 2cm. The metabolites were separated by Column DB-Wax (30 m × 250 m × 0.25 m) with Helium carrier gas 0.8 ml/min. The oven temperature was set at 40 ˚C for 0 min then 4˚C/minute and 220˚ for 10 min with a mass range of 29–550 am**.**

2.1 Results

(See Figs. 1, 2, 3 and 4).

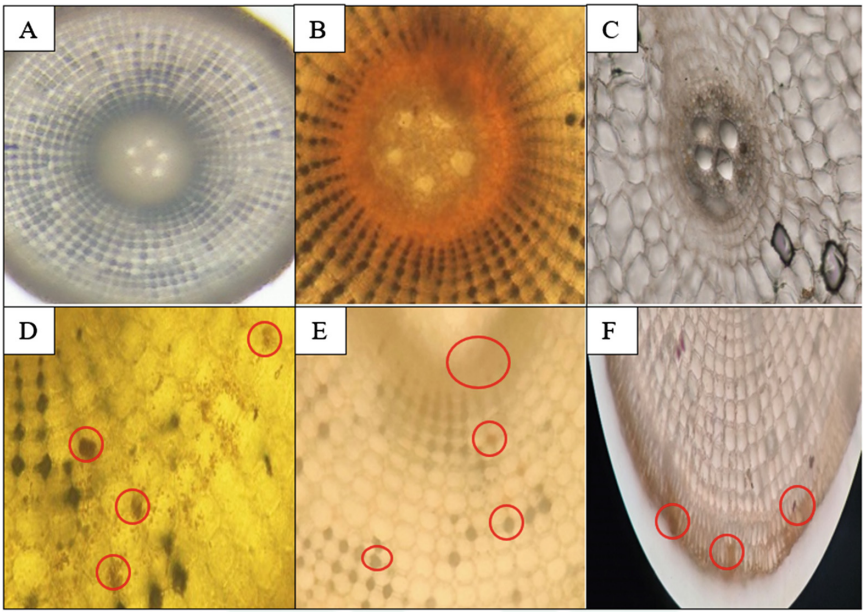

Fig. 1. Histochemical Test of PME Roots: A. Control (10 × 0.25μ magnification), B. Alkaloid test (10 × 0.25μ magnification), C. Alkaloid test (–) (20 × 0.40μ magnification), D. Phenolic test (40 × 0 magnification), 65μ), E. Terpenoid Test (20 × 0, 40μ), F. Lipid Test (10 × 0,25μ magnification)

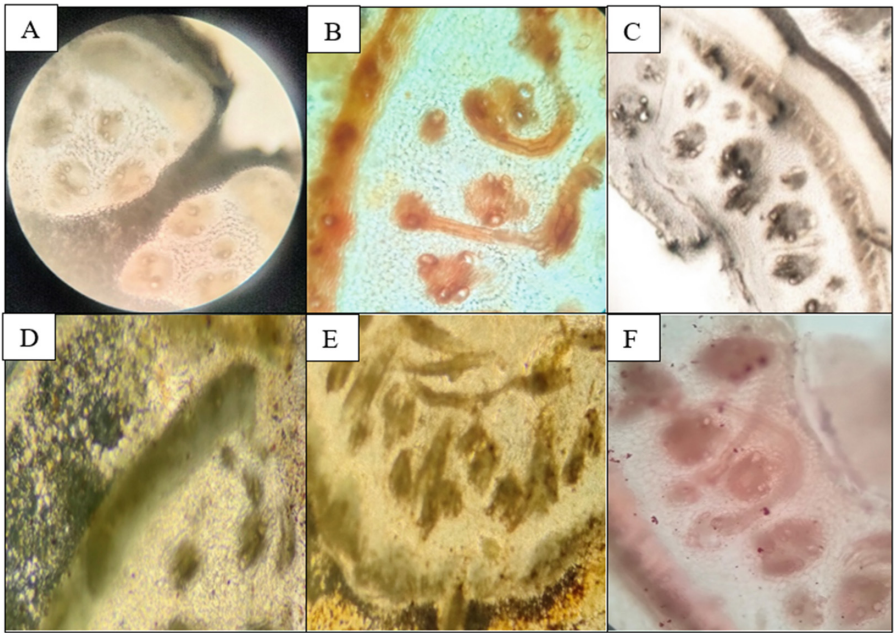

Fig. 2. Histochemical test at the base of the Pulu Mandoti Rice Stem: A. Control (10 × 0.25μ magnification), B. Alkaloid test (10 × 0.25μ magnification), C. Alkaloid test (–) (4 × 0.10μ magnification), D. Phenolic test (10 × 0 magnification). 25μ), E. Terpenoid Test (10 × 0.25μ), F. Lipid Test (10 × 0.25μ magnification)

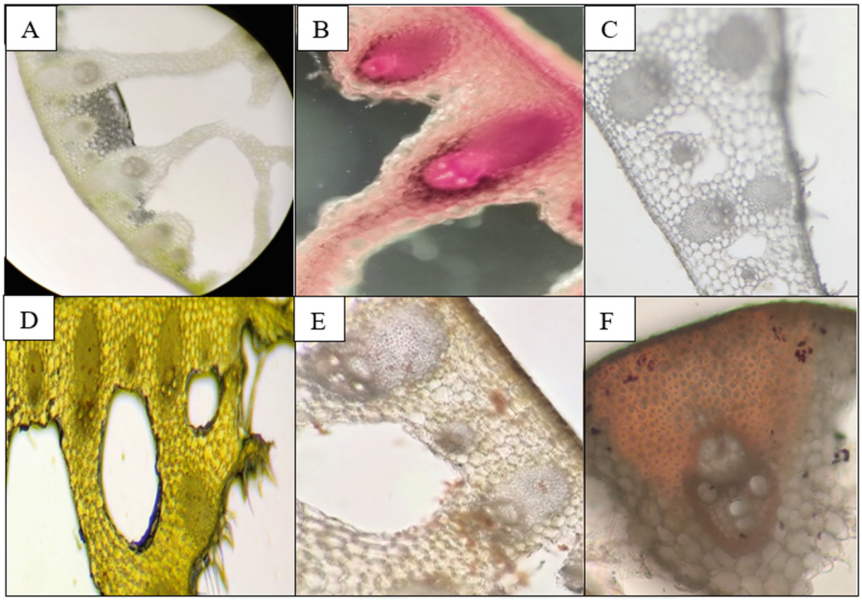

Fig. 3. Histochemical Test of Pulu Mandoti Rice Stem Nails: A. Control (10 × 0.25), B. Alkaloid test (10 × 0.25μ), C. Alkaloid test (–) (10 × 0.25μ magnification), D. Phenolic test (magnification) 10 × 0.25μ), E. Terpenoid Test (10 × 0.25μ), F. Lipid Test (10 × 0.25μ magnification)

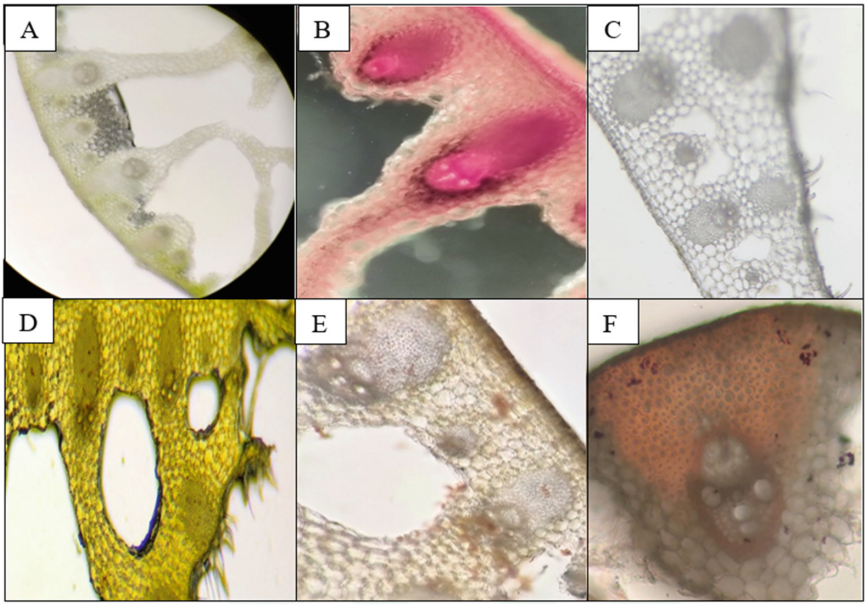

Fig. 4. Histochemical Test of Pulu Mandoti Paddy Leaves: A. Control (10 × 0.25), B. Alkaloid test (10 × 0.25μ magnification), C. Alkaloid test (–) (20 × 0.40μ magnification), D. Phenolic test (20 × 0 magnification),40μ), E. Terpenoid Test (20 × 0,405μ), F. Lipid Test (10 × 0,25μ)

Table 1. Group of Secondary Metabolites of Rice (Oryza sativa L.) PME

Golongan senyawa	RT	Nama Senyawa	Cons (ppb)
Alkana	6,5	2-Methylundecane	0,03
	8,3	Tridecane	0,03
	9,1	Decane, 2,3,6-trimethyl	0,11
	9,3	Dodecane, 4,6-dimethyl	0,17
	9,5	Tetradecane	0,04
	9,7	Undecane, 2-methyl	0,07
	10,2	Undecane, 4,6-dimethyl	0,01
	10,7	3,5-Dimethyldodecane	0,03
	10,8	Dodecane, 2,6,11-trimethyl	0,10
	11,1	Decane, 3,7-dimethyl	0,03
	11,4	Undecane, 5-methyl	0,04
	12,6	Tridecane, 2-methyl	0,04
	12,9	Hexadecane	0,02
	13,5	Decane, 3,8-dimethyl	0,02
	13,9	Pentadecane	0,05
	15,2	Pentadecane, 4-methyl	0,13

(*continued*)

Table 1. (*continued*)

Golongan senyawa	RT	Nama Senyawa	Cons (ppb)
	15,2	Pentadecane, 4-methyl	0,13
	15,4	Nonane, 1-iodo-	0,26
	22,2	Hexadecane, 2-methyl-	0,02
	22,6	5-Ethyl-5-methyldecane	0,06
Alkene	15,9	2-Undecene, 2,5-dimethyl-	0,13
	16,1	5-Undecene	0,02
	18,4	3-Octadecene, (E)-	0,06
	19,0	4-Dodecene, (E)-	0,25
	21,4	1-Heptadecene	0,02
	24,2	8-Heptadecene	0,03
	25,0	1-Octadecene	0,02
	29,5	Z-5-Nonadecene	0,01
	33,7	2-Methyl-7-nonadecene	0,01
Keton	11,9	2-Acetylcyclopentanone	0,04
	12,0	3-Octen-2-one, (E)-	0,02
	13,6	2-Nonanone	0,03
	16,8	2-Decanone	0,05
	17,4	3-Nonen-2-one	0,06
	17,9	6-Undecanone	0,04
	25,9	Cyclodecanone	0,02
	26,9	α-Isomethyl ionone	0,16
	27,4	β-iso-Methyl ionone	0,03
	28,6	α-N-Methyl ionone	0,02
	30,8	β-Methylionone	0,03
	31,3	γ-Nonalactone	0,02
	32,5	2-Pentylcyclopentanone	0,03
	33,8	Perhydrofarnesyl acetone	0,07
Terpen	34,0	α-Cedrene	0,01
	36,3	1-Acetyl-4,6,8-trimethylazulene	0,01
	31,8	β-Chamigrene	0,007

(*continued*)

Table 1. (*continued*)

Golongan senyawa	RT	Nama Senyawa	Cons (ppb)
Aromatic Hydrocarbon	14,3	3-Ethyl-2-methyl-1,3-hexadiene	0,10
	19,3	2-Ethyl-5-isobutylthiophene	0,11
	23,6	α-Bisabolene (Z)	0,04
	23,7	Naphthalene	0,09
	28,6	Butylated hydroxytoluene	0,04
	29,2	Benzene, (3,3-dimethyldecyl)-	0,02
	32,8	Benzene, 1,2,4-trimethoxy-	0,01
	34,1	2,6-Diisopropylnaphthalene	0,04
	34,5	1,7-Diisopropylnaphthalene	0,06
	34,9	Naphthalene, 2-methoxy-	0,05
	35,9	1,3-Diisopropylnaphthalene	0,06
	36,0	1,4-Diisopropylnaphthalene	0,05
	36,2	Naphthalene, 1,2,3-trimethyl-4-propenyl-, (E)-	0,11
	36,5	Naphthalene, 6-methoxy-2-(1-buten-3-yl)-	0,03
	31,8	Isopropyl myristate	0,05
Other Volatile Hydrocarbon	8,5	Decane, 2,4,6-trimethyl	0,03
	14,6	Dodecane, 2-methyl	0,08
	19,7	Tetradecane, 2,6,10 trimethyl	0,05
	24,1	2,6-Dimethyldecane	0,02
Alkohol	10,5	2-Methylcyclopentanol	0,05
	14,2	3,5-Octadien-2-ol	0,04
	15,7	1-Octen-3-ol	0,41
	18,9	Cyclohexanol, 2-tert-butyl-	0,02
	22,3	2-Butyl-2,7-octadien-1-ol	0,03
	23,2	5-(4-Methoxyanilino)-1,3,4-thiadiazole-2-thiol	0,05
	23,4	8-Quinolinol, 2-methyl-	0,05
	24,0	2-Octadecoxyethanol	0,02
	24,6	2-Octyldecan-1-ol	0,03
	25,1	2-Octyldecanol	0,04
	27,5	2-Hexyldecanol	0,03

(*continued*)

Table 1. (*continued*)

Golongan senyawa	RT	Nama Senyawa	Cons (ppb)
	27,9	Ethanol, 2-(dodecyloxy)-	0,01
	28,1	2-Hexyldodecanol	0,01
	28,7	5-Thiophen-2-yl-2H-pyrazol-3-ol	0,03
	29,6	Dicyclopentenyl alcohol	0,01
	30,1	6-Dodecanol	0,02
	30,3	Tricyclo[5.2.1.0(2,6)]dec-3-en-10-ol	0,03
	31,5	Z,Z-3,13-Octadecadien-1-ol	0,02
	33,4	Metacetamol	0,02
	33,6	Tetramethyl decynediol	0,01
Aliphatic Aldehyde	5,1	Hexanal	1,01
	10,6	Octanal	0,11
	11,6	2-Heptenal, (Z)-	0,10
	13,8	Nonanal	0,34
	14,8	2-Octenal, (E)-	0,19
	16,3	2-Heptenal, 2-propyl-	0,03
	18,0	2-Nonenal, (Z)-	0,06
	22,0	2-Octenal, 2-butyl-	0,52
	25,8	2,4-Decadienal, (E,E)-	0,03
	29,0	3,4-Hexadienal, 2-butyl-2-ethyl-5-methyl-	0,01
Aromatic Aldehyde	5,8	Glutaraldehyde	0,05
	17,5	Benzaldehyde	0,07
	23,1	Di-tert-dodecyl disulfide	0,03
	38,0	Galaxolide	0,09
	38,2	Versalide	0,09
	38,9	2-Hexylcinnamaldehyde	0,02
	44,9	Ambrettolide	0,01
Ester	24,8	Benzyldimethylcarbinyl acetate	0,06
	27,7	1,3-Pentanediol, 2,2,4-trimethyl-, diisobutyrate	0,06
	29,7	Verdyl acetate	0,02
	29,8	Geranyl isovalerate	0,01
	31,5	Isoamyl salicylate	0,01
	31,8	Isopropyl myristate	0,05

(*continued*)

Table 1. (*continued*)

Golongan senyawa	RT	Nama Senyawa	Cons (ppb)
	33,1	Amyl salicylate	0,01
	35,6	n-Hexyl salicylate	0,08
	39,2	Diethyl phthalate	0,01
	40,9	Methyl-trans-oleate	0,01
	41,9	Methyl linolelaidate	0,01
	47,7	Benzyl salicylate	0,01
	44,4	Benzyl benzoate	0,004
	35,2	1-(4-Benzylphenyl)ethanone	0,03
Fenol	32,1	Phenol, 3,5-di-tert-butyl-	0,03
	33,3	4-Methoxy-2-tert-butylphenol	0,007
	35,3	2-Methoxy-4-vinylphenol	0,001
	38,1	Phenol, 2,4-di-tert-butyl-	0,07
	26,	Anethole	0,06
Carbocylic acid	32,3	Caprylic acid	0,05
	34,9	Nonanoic acid	0,02
	37,3	Caprinic acid	0,003
	42,8	Lauric acid	0,006
	46,0	Myristic acid	0,11
	48,6	Pentadecylic acid	0,003
	51,8	Palmitic acid	0,61
Furan	6,1	Furan, 2-butyl-	0,03
	8,7	Furan, 2-pentyl-	0,13
	8,9	Furan, 2-hexyl-	0,14

3 Discussion

Histochemical tests were carried out on vegetative samples of rice (Oryza sativa L.) PME variety using 4 types of samples, namely roots, stem bases, stem nodes and leaves. Histochemical test is a test used to detect the location of secondary metabolites in plant tissues. Histochemical results were determined based on the reaction of plant tissue and reagents in producing a color change [4].

3.1 Alkaloid Test

In the histochemical test of alkaloids all samples (roots, stems and leaves) showed positive results. Wagner's reagent reacts with plant tissue to produce a reddish-brown

or yellow color change in samples that are positive for alkaloid confirmation. This brown color is formed from the reaction of iodine and I- ions to produce I3- ions. The presence of K+ ions in Wagner's reagent forms covalent bonds with nitrogen to form potassium-alkaloids [8]. At the root of this compound is found in the endodermis and stele layers, at the base of the stem it is found in the epidermis layer, parenchyma tissue and transport bundles, in the nail part it is found in the epidermis layer, parenchyma tissue and transport bundles and in the leaves this compound is found in the lower part of the stem. Parenchyma and cortex. Alkaloids are active compounds that contain nitrogen. Alkaloid compounds are found in mesophyll cells and idioblasts. These compounds are often found in roots, but can also be identified in the stems and rhizomes [9].

3.2 Phenolic Test

The reagent trichloride ($FeCl_3$) and sodium bicarbonate in the phenol test showed a dark green to black color change reaction in the positive tissue containing phenol. $FeCl_3$ can produce color changes due to the formation of complex compounds between Fe^{3+} ions and phenolic compounds such as tannins. The Fe^{3+} ion acts as the central atom which, when combined with tannins which have O atoms, can coordinate the central atom and form ligands [10]. Positive test results were found in the root, base and nodes of stems and leaves. In roots and stems, phenolic compounds were found in the cortex layer, while in the root nodes were found in parenchyma and phloem tissue and in leaves found in the cortex layer. The results of the research by Dewi *et al.* (2020) showed that the betel stalk *(Piper betle* L.) found phenolic compounds in parenchyma tissue and cortex, while in the leaves it was detected in parenchyma tissue [11].

3.3 Terpenoid Test

Cupric acetate reagent which reacts with plant tissue produces a yellow-brown color change in terpenoid positive samples. Staining with 5% copper acetate was able to produce a color change from the terpenoid histochemical test results to a yellow-brown color. Terpenoid compounds are synthesized through Acetyl CoA in mevalonic acid [10]. Histochemical test results showed that all positive vegetative samples contained terpenoid compounds. In the root samples, terpenoid compounds were found in the cortex and stele. Terpenoid compounds at the base of the stem were found in the epidermis and parenchyma, in the nail part it was found in the parenchyma tissue and in the leaves, terpene compounds were found in the cortex. Research by Dewi *et al.* (2020) reported that the terpenoid compounds in betel leaf were found in the parenchyma tissue and lamina, while in the positive betel stem it was found in the parenchyma tissue, cortex and pith. On the stems of Tagetes sp [11]. Terpenes were found in the cortical parenchyma tissue near the stem epidermis.

3.4 Lipid Test

Sudan III reagent reacts with plant tissue to produce a reddish color change in positive samples. Sudan III reacts with lipids so that it will produce complex compounds with

a red color change if there are lipid compounds contained in the test sample. Based on the lipid test, all samples (roots, stems and leaves) were positive for lipid compounds. Lipid compounds in roots can be detected in the epidermis, at the base of the stem it can be detected in the epidermis and transport tissue, in the root nodes it can be detected in the phloem and in leaves, lipid compounds can be detected in the epidermis and cortex. Maghfiroh *et al.* (2018) explained that in the results of histochemical tests on olive leaves (*Olea europaea*) where lipophilic compounds with fatty properties were localized in the epidermis [12]. The results of Trimanto *et al.* (2018) showed lipid in Curcuma rhizome were detected in parenchyma tissue and transport bundles which were marked by the presence of a red color detected in the rhizome tissue[4].

3.5 Volatile Metabolite Analysis

Based on the results of GC-MS that have been carried out on (Oryza sativa L.) PME, 13 types of volatile compound groups were obtained consisting of 59 alkane compounds, 11 alkene compounds, 14 ketone compounds, 3 groups of compounds. Terpenes, 14 aromatic hydrocarbon groups, 4 other volatile hydrocarbon groups, 20 alcohol groups, 10 aliphatic aldehydes, 7 aromatic aldehydes, 12 esters, 5 phenolic compounds, 7 carboxylic acid compounds and 3 furan group.

1. Quantification of Alkanes and alkenes

   Based on the observations that have been made, there are 59 types of compounds identified into the alkane group, namely decane, undecane, tetradecane, pentadecane, tridecane, dodecane, heptadecane, octadecane, nonane, hexadecane and tridecane. Many alkane compounds have been detected in rice. According to Hinge *et al* (2016) alkane compounds can be identified in the leaves and seeds of rice plants with the percentage of alkanes in the range of 11–13% more in the seeds and 7–9% in the leaves [13]. A total of 15 alkane compounds identified in rice grains showed that this group did not contribute to the aromatic effect [6]. Furthermore, his research also explained that alkane and alkene group compounds are associated with lipids. However, when these lipid compounds are synthesized at high concentrations, this accumulation will affect the quality, this is because these compounds do not have an aromatic effect on rice but instead produce an unpleasant aroma in rice.

   In alkene compounds, 11 types of compounds were identified. The alkene compounds consist of undecene, octadecene, dodecene, heptadecene and nonadecene. Many alkene compounds are produced in the leaves. According to Hinge *et al* (2016) about 6–7% of alkene compounds are mostly produced in the seeds and about 6–10% of alkene compounds are mostly produced in the leaves [13].
2. Ketone Quantification

   Based on Table 1, the GC-MS table obtained 14 compounds of the ketone group including 2-acetylcyclopentanone, 3-octen-2-one, (E)-, 2-oonanone, 2-decanone, 3-nonen-2-one, 6 -undecanone, cyclodecanone, -isomethyl ionone, -iso-methyl ionone, -N-methyl ionone, -methylionone, -nonalactone, 2-pentylcyclopentanone and per-hydrofarnesyl acetone. Compounds that are classified as ketones are compounds that give rice a fruity aroma. Many ketone compounds are found in the generative phase during seed maturation [13]. Compounds such as alcohol, aldehydes,

ketones are components that are responsible for the aroma profile of aromatic rice [14]. The compound 3-Octen-2-one, (E) is a compound from the ketone group which is commonly found in the leaves and seeds of aromatic rice cultivars [13]. Some of the ketone compounds identified in black rice include 3-Octen-2-one which produces a rose aroma and 2-nonanone which produces a fruity aroma [13]. Choi *et al* 2018 was found 7 volatile ketone compounds in cooked black rice samples including 2-nonanone with a fruity, cheesy aroma, 3-octen-2-one with a creamy, oily, earthy aroma, 2-pentadecanone compound. With the aroma of jasmine, celery, compound 6,10,14-trimethyl-2-pentadecanone with the aroma of fresh jasmine, celery, compound 6-methyl-1-3-5-heptadien-2-one with the aroma of coconut, cinnamon, compound 5- pentyl-2(5H)-funanone with minty aroma and compound (5E)-6,10-dimethyl-5,9-undecadien-2-one with fruity, green, waxy aroma.

3. aldehyde quantification

   The aldehyde group of compounds is a compound that contributes to the aroma of aromatic rice. Based on the analysis results found in Table 4.1, there are 2 groups of aldehyde class compounds consisting of aliphatic aldehydes and aromatic aldehydes. A total of 10 types of compounds classified as aliphatic aldehydes consist of hexanal, octanal, 2-heptenal, nonanal, 2-octenal, 2-heptenal 2-propyl, 2-nonenal, 2-octenal 2-butyl, 2,4-decadienal, 3, 4-hexadienal 2 butyl-2-ethyl-5-methyl. While in the aromatic aldehyde group, the types of compounds obtained include glutaraldehyde, benzaldehyde, di-tert-dodecyl disulfide, galaxolide, versalide, 2-hexylcinnamaldehyde and ambrettolide. According to Hinge *et al* (2016) heptanal compounds contribute to the aroma of rice [13].

   Heptanal compounds were significantly found in aromatic rice cultivars when compared to non-aromatic rice. Heptanal compounds are volatile compounds derived from fatty acids that accumulate with the aroma of green grass. Heptanal compounds are mostly produced in the vegetative stage of rice. This is due to the presence of fatty acids which increase during vegetative growth and decrease during the maturation phase. According to Nadaf *et al* (2016) heptanal compounds are the key that give aromatic rice a floral aroma [15].

4. Alcohol Group

   Alcohol group compounds are a type of compound group that also contributes to the aromatic properties of aromatic rice. Based on the results identified, there were found as many as 20 compounds belonging to the alcohol group including 2-Methylcyclopentanol, 3,5-Octadien-2-ol, 1-Octen-3-ol, Cyclohexanol, 2-tert-butyl-, 2-Butyl- 2,7-octadien-1-ol, 5-(4-Methoxyanilino)-1,3,4-thiadiazole-2-thiol, 8-Quinolinol, 2-methyl-, 2-Octadecoxyethanol, 2-Octyldecan-1-ol , 2-Octyldecanol, 2-Hexyldecanol, Ethanol, 2-(dodecyloxy)-, 2-Hexyldecanol, 5-Thiophen-2-yl-2H-pyrazol-3-. Alcohol comes from the formation of secondary hydroperoxide decomposition of fatty acids which increase a sweet taste, floral and fruity aroma found in rice [13]. Among the alcohol compounds identified, there was a compound 1-Octen-3-ol with the highest cons (ppb) among other alcohol group compounds, namely 0.41. Identified alcohol compounds found in black rice, including 1-Octen-3-ol which produces mushroom aroma, 1-pentanol which produces plastic aroma and 1-heptanol 1-heptanol with green aroma.

The compound 1-Octen-3-ol is an alcohol group that has been identified as being found in more aromatic rice cultivars than non-aromatic rice. 1-Octen-3-ol has a strong herbal aroma and acts as a compound that affects the smell and aroma of aromatic rice. The compound 1-Octen-3-ol produces an aroma in the form of a mushroom smell in several studies on the aroma of rice. The compound 1-Octen-3-ol has a role in the defense against the growth and development of rice plants. In a study conducted by Kanchiswamy *et al* (2015) it was found that the compound 1-Octen-3-ol has the potential as an attractant for insects such as mushroom-eating beetles. In addition, this compound also plays a role in plant resistance to pathogenic fungi [16].

5. Phenol quantification

Based on the results of GC-MS obtained 5 types of compounds belonging to the class of phenol compounds including Phenol, 3,5-di-tert-butyl-, 4-Methoxy-2-tert-butylphenol, 2-Methoxy-4-vinylphenol, Phenol, 2,4-di-tert-butyl- and Anethole. Phenolic compounds are one type of compound that is responsible for the quality of aromatic rice. According to Bryant and McClung (2011), compounds such as alcohol and phenol contribute to producing a sweet, floral or fruity aroma in rice. There are 3 main phenolic compounds identified in cooked black rice including 4-vinylphenol, guanicol and 2-methoxy-4-vinylphenol [6].

Aprotosaie *et al* (2016) that anethole is a compound that contributes to the aroma that is widely used in the food industry and acts as a bioactive compound in the form of anti-inflammatory, anti-carcinogenic, immunomodulatory, and so on. 2-Methoxy-4-vinylphenol is a phenol group compound that contributes to the aroma of aromatic rice [17]. That the compound 2-Methoxy-4-vinylphenol was found in aromatic rice cultivars that contributed to the aroma but in their study this compound was not found in the seeds. The compound 2-Methoxy-4-vinylphenol was also detected as a key aroma compound recently identified from Japanese sweet rice wine. This compound gives it a spicy aroma [13].

6. Furan quantification

Furan group compounds are also volatile compounds that play a role in the quality of aromatic rice aroma. Based on the table above, there are 3 types of compounds classified as furan compounds including Furan, 2-butyl-, Furan, 2-pentyl- and Furan, 2-hexyl-. Choi *et al* (2018) was found that there were 2 main volatile compounds of the furan group, namely Furan, 2-butyl- and Furan, 2-pentyl-[18]. Setyaningsih *et al* (2019), other compounds that are the key volatile compounds that determine the quality of rice are Hexanal and Furan compounds, 2-pentyl. 2-pentyl- is a furan group compound that can produce fruity, floral and nutty flavors detected in the leaves and seeds of rice cultivars [19]. This compound is also able to produce aromatic properties such as caramel in rice [13].

7. Terpenoid, Ester, Carboxilic acids

Based on the results contained in Table 1, there were 3 compounds classified as terpenes, 12 ester and 7 compounds belonging to carboxylic acid. Terpenes are one of the volatile compounds that contribute to the aromatic contribution. Terpenes that have been identified include -Cedrene, 1-Acetyl-4,6,8-trimethylazulene and -Chamigrene. Terpenes in rice can produce a specific aroma in the vegetative stage, but these compounds decrease at the stage of seed maturation. Terpene compounds in rice consist of monoterpenes, sesquiterpenes, diterpenes and triterpenes where

monoterpenes and sesquiterpenes are aromatic compounds that are mostly found in rice leaves, while triterpenes are usually found in rice husks. Monoterpenes are volatile compounds that play a major role in the aromatic quality of rice. Terpenes play a role in the characteristic taste and aroma of fruits and spices. These compounds also act as endogenous defenses in plants [13].

Based on the reference results regarding the analysis of aromatic compounds in Pulu Mandoti rice, it is suspected that several volatile compounds that affect the taste, aroma and quality of Pulu Mandoti rice include benzaldehyde, 2-methoxy-4-vinylphenol, furan, 2-pentyl-, hexanal, nonenal, 2-nonenal, octanal, 1-octen-3-ol, anethole. Volatile compounds are chemical compounds that are volatile. These compounds are then what can distinguish between aromatic and non-aromatic rice. The taste and aroma of each aromatic rice is different. This is due to the presence of key compounds that become markers that distinguish one rice from another. Although there is one volatile compound as a marker for the specific aroma of rice, there are also compounds that associate one compound with other compounds in producing a distinctive aroma in rice. Volatile compounds, of course, have many roles in plants. This compound will produce an aroma that can affect animal behavior [20]. Volatile compounds play a major role in producing the distinctive taste and aroma of aromatic rice. In addition to taste and aroma, volatile compounds also contribute to endogenous and exogenous defense systems in plants [13].

4 Conclusion

1. Based on histochemical tests on the roots, stems and leaves of the rice, "Pulut Emas Mandoti" contains alkaloids, phenolic, terpenes and lipids which are mostly found in the epidermis, cortex, stele, parenchyma tissue and vascular tissue.
2. Based on the volatile metabolite of profile data, it is suspected that the metabolites that have the potential to be candidates for biomarkers of distinctive aroma in "PME Emas" rice include: benzaldehyde, 2-Methoxy-4-vinylphenol, Furan, 2-pentyl-, Nonenal, 2-Nonenal, octanal, 1-Octen-3-ol.

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