

# Metabolomic and sensomic approach to characterize key aroma of aromatic mangoes from Indonesia

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<sup>1</sup>Department of Agricultural Product Technology, Faculty of Agricultural Technology, Universitas Jember. Jl. Kalimantan Tegalboto 37, Jember 68121, East Java, Indonesia. Tel./fax.: +62-331-337422, \*email: maria\_belgis@yahoo.com

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**Abstract.** Belgis M, Witono Y, Giyarto, Sadek NF, Masruroh EL, Herlina. 2023. *Metabolomic and sensomic approach to characterize key aroma of aromatic mangoes from Indonesia. Biodiversitas 24: 2242-2250.* Garifta Merah, Garifta Orange, and Agri Gardina 45 are new mango cultivars with a strong aroma and attractive color from Indonesia. This study investigated aromatic constituents and sensory profiles using a combination of metabolomic and sensomic approaches of Garifta Merah, garifta Orange, and Gardina 45. Volatile compounds were extracted using Head Space-Solid Phase Microextraction (HS-SPME), then identified using Gas Chromatography-Mass Spectrometry (GC-MS). Sensory descriptive Rate All That Apply (RATA) and hedonic tests were used for sensory attributes profiling and aroma-liking evaluation. Monoterpene hydrocarbons were the predominant volatile compound in all cultivars; however, it was also accompanied by esters in Garifta Orange and Agri Gardina 45. The Principal Component Analysis (PCA) classified those cultivars into different groups. Garifta Merah was characterized by higher  $\alpha$ -pinene, while Garifta Orange was characterized by high ethyl butanoate. Agri Gardina was in the separate group characterized by  $\beta$ -trans ocimene, ethyl butanoate, and isopentyl butanoate. The identified compounds in all cultivars had Odor Active Value (OAV)>1 and correlated to their sensory profile described by assessors. Garifta Orange was perceived to have the highest overall aroma liking among all cultivars. It was presumably due to the high ethyl butanoate content related to the pineapple, mango, fruity, and fermented attributes.

**Keywords:** Aroma, mango, metabolomic, sensomic

## INTRODUCTION

Mango (*Mangifera indica* L.) is reported as a native crop from South Asia that has a unique taste and aroma (Souza et al. 2018). The mango and the banana have become the world's most popular tropical fruit (Maldonado-Celis et al. 2019). Among various types of mangoes in Indonesia, Garifta Merah, Garifta Orange, and Agri Gardina 45 are superior varieties that the Indonesian Agricultural Ministry released. They have attractive yellow to reddish-colored skin with a stronger and more pleasant aroma than others.

Aroma is one of the essential qualities affecting consumers' decisions in mango selection, especially for the new variant in the market. Thus, the evaluation of mango aroma attributes is critical. Mango aroma was developed from volatile compounds, which differ significantly among cultivars. Volatile compounds are mainly secondary metabolite products of metabolism, which can be used as markers of species, cultivars, or varieties. Several factors affect the levels and composition of volatile compounds in mango fruit, such as genotype, pre-post harvesting treatments (handling, storage condition, and chilling injury), and harvest maturity. Several glycosidically-bounded volatile compounds are liberated during ripening, such as terpene alcohols, nor-isoprenoid derivatives, and aromatic alcohols (Thiruchelvam et al. 2020).

A mixture of volatile compounds forms a mango aroma, and each compound serves a distinct odor. Unique aroma characteristics among cultivars depend on their combinations, concentrations, and ratios. They collaborated to develop unique flavors through cumulative, synergistic, and masking effects. The responsible volatiles for mango aroma were terpenoids, alcohols, aldehydes, esters, carbonyl, and nitrogen-containing compounds (Ma et al. 2018), in which terpenes were the primary constituents. Li et al. (2017) investigated the volatile diversity of 25 cultivars from China, America, Thailand, India, Cuba, Indonesia, and the Philippines. It was found that the compositions and proportions of terpenes are significant differences in those cultivars. Another study by Liu et al. (2020) reported that  $\gamma$ -terpinene, 1-hexanol, hexanal, terpinolene trans-2-heptenal, and *p*-cymene were responsible compounds for the aroma of five mango varieties in China. This study also revealed that the unique flavor of those mangoes was greatly affected by aldehydes and terpenes compounds.

The compound impact and sensory profile of Garifta Merah, Garifta Orange, and Agri Gardina 45 could be identified and determined using metabolomic and sensomic approaches. Metabolomics is a systematic study of specific chemical fingerprints regarding various cellular metabolic processes (Onuh and Qiu 2021), while sensomic concerns describing foods' sensory attributes at a molecular level (Vrzal and Olšovská 2019). Combining these methods

could obtain the responsible compounds for specific sensory characteristics and classify the commodity based on their character. A previous study by Belgis et al. (2017) successfully applied these approaches to reveal that durian (*Durio zibethinus*), and several lai cultivars (*Durio kutejensis*) were characterized into different groups. Lai has a less intense sulfury, fruity, and sweet aroma than durian, presumably due to the lower sulfurs and esters in lai than durian.

This study aimed to separate volatile compounds using head space-solid phase microextraction (HS-SPME). Further analysis was performed qualitatively and quantitatively using GC-MS (Gas Chromatography-Mass Spectrometry). Odor Active Value (OAV) was calculated to determine the aroma contribution of each volatile compound to fruit aroma. The acceptance and attributes of mango aroma were evaluated using the hedonic test and RATA (Rate All That Apply) method. The results of this study may provide a valuable dataset to facilitate the plant breeders to develop mangoes with better and desirable aroma qualities. In addition, the optimization of mango processing could also be achieved by an improved understanding of the critical aroma compounds.

## MATERIALS AND METHODS

### Materials

Garifta Merah, Garifta Orange, and Agri Gardina 45 mangoes used in this study were obtained from Cukurgondang Experimental Field, Indonesian Tropical Fruits Research Institute, Ministry of Agriculture, Pasuruan, East Java, Indonesia (7°39'2"S and 112°54'42" E). They were harvested at the same physiological maturity (90-115 days after flowering, reddish peel color, and smelly). Criteria of fruit selection are no visual defects and no fungal infections. Mangoes were stored at room temperature (27-30°C) at 56-62% RH for 5 days after harvesting for ripening optimization. Sample preparation was initialized by collecting the mango flesh and crushed into pulp. Mango pulp was packed in aluminum foil and stored at -18°C before the analysis, then fully thawed at room temperature before use.

### Chemicals

The chemicals used in this study are sodium chloride (Merck, Darmstadt, Germany) as well as the series of alkenes (C6-C27) and 1,4-dichlorobenzene as internal standards (Aldrich, Steinheim, Germany).

### Volatile compounds absorption

The volatile compounds were absorbed using HS-SPME (DVB/CAR/PDMS) fiber purchased from Supelco, Inc. Bellefonte, PA, USA. Three grams of mango pulp were homogenized with 2.5 mL of 25% NaCl, inserted into the 22 mL vial, and added with 0.01% 1,4-dichlorobenzene as an internal standard. The sample was then homogenized at 40°C for 3 minutes. Next, the SPME syringe was inserted into the vial headspace for 30 minutes before being injected into GC-MS.

### Volatile compound analysis

Mangos' volatile compounds were analyzed using GC-MS (Agilent 7890A-5975C) using an HP-5MS column (30 m x 250 µm x 0.25 µm Agilent, Santa Clara). Helium as carrier gas (constant flow rate of 0.8 mL/min), electron ionization energy was 70 eV and spitless mode. The initial oven temperature was 40°C, and it held for 6 minutes. It was set to rise gradually (6°C/minute) to reach 220°C, then held for 4 minutes. Alkane standard (C6-C27) was injected under the same conditions to calculate the Linear Retention Index (LRI). The volatile compounds were identified by comparing the mass spectra with those of NIST 2.0 databases. Confirmation was carried out by comparing LRI components with LRI found in references.

### Sensory analysis

The assessors in this study were from the Faculty of Agricultural Technology, the University of Jember. One hundred and seven untrained assessors (70 female and 37 male, 18-39 years old) were selected based on their self-declaration of health. The sensory room had a uniform temperature (26-28°C), a source of lighting, and an absence of noise.

The RATA method assessed the sensory aroma descriptive of Garifta Merah, Garifta Orange, and Agri Gardina. The assessment was carried out in eight aroma attributes (honey, sweet, fruity, mango, lemon, floral, fermented, and pineapple), developed in FGD (Focus Group Discussion) session conducted before testing. Each attribute was rated using a 5-point intensity scale (1: very low, while 5: very high). A score of 0 was given when no aroma was perceived (Danner et al. 2018). The hedonic test was also performed to evaluate aroma acceptance using a 7-points scale (1: very dislike, 7: very like).

### Odor Active Value (OAV)

The contribution of each volatile compound to overall fruit aroma was evaluated by odor active value (OAV), which was measured as the ratio between each compound concentration to its detection threshold in water. The OAVs value was calculated using  $OAV = c \cdot t^{-1}$  equation, where "c" was each compound concentration, and "t" was the odor threshold value. The t value was obtained from the available literature.

### Statistical analysis

Principal Component Analysis (PCA) was conducted to map the mango's volatiles and sensory data. It was analyzed using Unscrambler Windows version 9.7 software packages, CAMO A/S, Trondheim, Norway.

## RESULTS AND DISCUSSION

Three mango cultivars in this study had different volatile compounds composition. Garifta Merah, Garifta Orange, and Agri Gardina 45 had 35, 41, and 31 volatile compounds, respectively (Table 1). Volatile compounds in fruit were influenced by genetics, maturity level, environmental conditions, postharvest handling, and storage

(El Hadi et al. 2013). The genetic factor affects the formation of precursors, enzymes, and fragrance formation (Zang et al. 2022). Those mangoes' volatile composition differences were mainly due to genetic differences (varieties) since they had uniform growing locations, maturity, and storage condition.

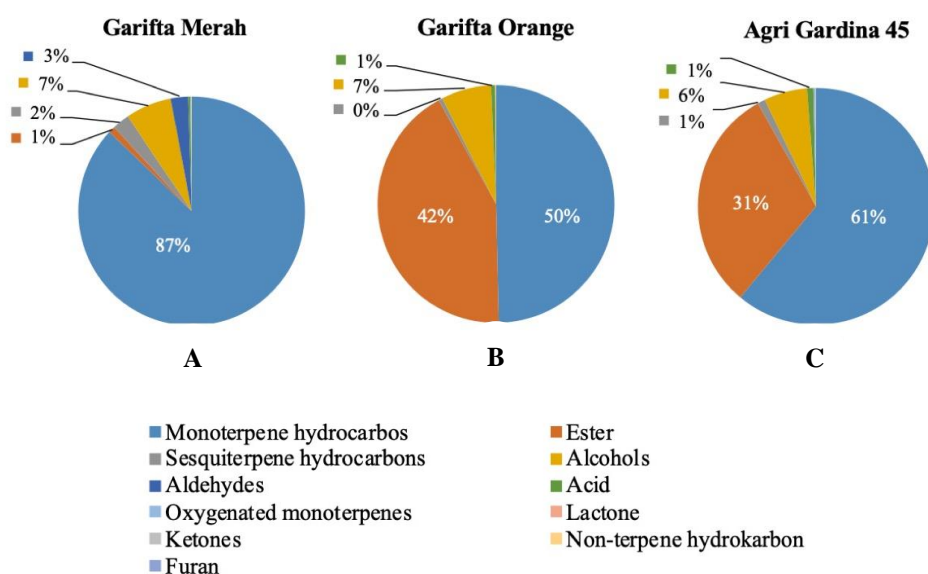
The total relative concentration of volatile compounds in mango varied between 0.72 to 16.77  $\mu\text{g g}^{-1}$  in those three cultivars. Agri Gardina 45 had the highest concentration (16.77  $\mu\text{g g}^{-1}$ ), followed by Garifta Merah (5.39  $\mu\text{g g}^{-1}$ ) and Garifta Orange (0.72  $\mu\text{g g}^{-1}$ ). Pandit et al. (2009) reported that 27 mango cultivars from India had different volatile compositions with total concentrations between 17.62–122.46  $\mu\text{g g}^{-1}$ .

Volatile compound groups of Agri Gardina 45, Garifta Merah, and Garifta Orange were presented in Figure 1. All mangoes contained a relatively high percentage of monoterpene hydrocarbons, sesquiterpene hydrocarbons, esters, aldehydes, alcohols, and acids. Oxygenated monoterpenes, lactones, furan, ketones, and non-terpene hydrocarbons were in small fractions. This result was similar to the finding of Pino et al. (2005) on volatile compound groups of Cuban mangoes. It consisted of monoterpenes hydrocarbon, sesquiterpenes hydrocarbon, esters, lactones, carbonyls, alcohols, and acids. A study by Ma et al. (2018) on core Chinese mango germplasm showed the presence of 114 volatile compounds, including 23 monoterpenes, 16 sesquiterpenes, 29 non-terpene hydrocarbons, 25 esters, 11 aldehydes, five alcohols, and five ketones.

Monoterpenes hydrocarbon and esters were the highest compound groups on these three mango cultivars. Monoterpene hydrocarbons were the highest percentage identified in Garifta Merah (87%), followed by Agri Gardina 45 (61%) and Garifta Orange (50%). The identified monoterpene hydrocarbons were  $\alpha$ -pinene,  $\delta$ -3-

Carene,  $\beta$ -pinene,  $\alpha$ -terpinene, D-limonene,  $\beta$ -thujene,  $\beta$ -phellandrene,  $\gamma$ -terpinene,  $\beta$ -cis-ocimene, m-cymene, terpinolene, and allo-ocimene. Garifta Merah and Garifta Orange had  $\alpha$ -pinene and  $\delta$ -3-Carene as the first and second-largest compounds, but they were not found in Agri Gardina 45 (Table 1). The  $\alpha$ -pinene was correlated to pine and turpentine aromas (Allenspach and Steuer 2021). The  $\alpha$ -pinene was reported as the aroma contributor in Kenyan mango varieties (Wetungu et al. 2018), Tainong mango (Xin et al. 2021), and 20 Cuban mango cultivars (Pino et al. 2005).  $\alpha$ -pinene and terpinolene were the principal components constituting the Chinese mango aroma (Ma et al. 2018). The  $\delta$ -3-Carene contributed to mango-like notes. It was the principal volatile compound in non-Indian cultivars and Cuban mangoes (Pandit et al. 2009; Pino et al. 2005). The  $\beta$ -trans-ocimene was identified only in Agri Gardina 45 as the major volatile compound (Table 1). Zakaria et al. (2018) reported that  $\beta$ -trans-ocimene was one of the chemical markers of Malaysian Harumanis mango. The presence of this compound contributed to the floral aroma.

Esters had been known to give a sweet and fruity aroma to fruits. This group was identified as the second major compound in Garifta Orange (42%) and Agri Gardina 45 (31%) but low in Garifta Merah (1%) (Figure 1). It resembled the ripe Brazilian Palmer mangoes with esters as their main aroma marker. However, no esters were found in 27 Indian mango cultivars Pandit et al. (2009). Several identified esters in these three cultivars were ethyl acetate, methyl butanoate, ethyl butanoate, butyl acetate, ethyl cyclopropane carboxylate, propyl butanoate, ethyl crotonate, butyl butanoate, ethyl hexanoate, ethyl 2-hexanoate, isopentyl butanoate, pentyl butanoate, ethyl heptanoate, butyl 2-butanoate, butyl hexanoate, hexyl butanoate, ethyl octanoate, isopentyl hexanoate, ethyl decanoate, ethyl dodecanoate, and ethyl tetradecanoate.



**Figure 1.** Percentage of volatile compounds of: A. Garifta Merah, B. Garifta Orange, C. Agri Gardina 45 mangoes

**Table 1.** Relative volatile compounds (ng g<sup>-1</sup>) identification of Garifta Merah, Garifta Orange dan Agri Gardina 45 mangoes

Group	Peak No.	Compounds	RI	Garifta Merah	Garifta Orange	Agri Gardina 45
Aldehydes	7	Hexanal	1079	2.73	nd	nd
	40	2-Nonenal, (E)-	1537	77.36	nd	nd
	43	2,6-Nonadienal, (E,E)-	1587	54.12	nd	nd
Alcohols	2	Methyl alcohol	901	nd	nd	27.23
	3	Ethanol	940	200.86	47.65	945.47
	11	1-Butanol	1150	nd	nd	16.00
	38	3-Hepten-1-ol	1489	36.30	0.56	nd
	51	3-Nonen-1-ol, (Z)-	1690	53.52	0.75	nd
	53	3,6-Nonadien-1-ol, (E,Z)-	1757	58.70	nd	nd
	56	Phenylethyl alcohol	1918	3.43	0.27	nd
Monoterpenes hydrocarbons	10	$\alpha$ -Pinene	1146	2818.65	214.79	nd
	12	$\delta$ -3-Carene	1156	584.54	57.47	nd
	13	$\beta$ -Pinene	1159	228.95	11.81	nd
	15	$\alpha$ -Terpinene	1182	178.74	7.54	nd
	16	D-Limonene	1188	310.05	18.74	60.47
	17	$\beta$ -Thujene	1196	170.90	11.29	14.37
	18	$\beta$ -Phellandrene	1206	32.87	2.42	nd
	21	$\beta$ -Trans-ocimene	1235	nd	7.71	9549.36
	23	$\gamma$ -Terpinene	1242	25.89	1.97	37.92
	24	$\beta$ -Cis-ocimene	1253	11.76	0.80	584.31
	27	m-Cymene	1270	15.94	4.68	nd
	28	Terpinolene	1277	318.68	18.51	nd
	32	Allo-ocimene	1372	0.73	0.04	nd
Sesquiterpenes hydrocarbons	39	$\alpha$ -Gurjunene	1530	6.84	nd	nd
	44	Caryophyllene	1596	77.05	2.41	25.24
	47	$\alpha$ -Caryophyllene	1670	45.77	1.44	12.10
	49	$\beta$ -Selinene	1721	nd	nd	119.60
	50	$\alpha$ -Selinene	1726	1.26	nd	24.80
Oxygenated monoterpenes	41	Eucarvone	1558	3.07	nd	nd
	42	Linalol	1550	nd	nd	50.40
	54	p-Cymen-8-ol	1848	2.56	nd	nd
Lactones	48	$\gamma$ -Caprolactone	1704	nd	nd	8.01
	58	$\gamma$ -Nonalactone	2035	0.25	0.06	nd
Furan	20	2-Pentyl- furan	1227	nd	0.15	nd
Ketones	59	Furaneol	2043	nd	0.10	nd
Non-terpene hydrocarbon	52	Naphthalene	1740	4.04	0.68	nd
Esters	1	Ethyl Acetate	882	35.75	13.46	30.58
	4	Methyl butanoate	979	nd	nd	22.55
	5	Ethyl butanoate	1035	nd	228.42	1793.91
	6	Butyl acetate	1072	1.68	nd	nd
	8	Ethyl cyclopropane carboxylate	1102	nd	2.06	nd
	9	Propyl butanoate	1119	nd	1.54	12.62
	14	Ethyl crotonate	1161	nd	20.56	401.54
	19	Butyl butanoate	1216	11.43	1.28	864.68
	22	Ethyl hexanoate	1238	nd	8.07	nd
	25	Ethyl 2-hexenoate	1260	nd	0.86	nd
	26	Isopentyl butanoate	1265	nd	2.85	1377.71
	29	Pentyl butanoate	1316	nd	nd	7.40
	30	Ethyl heptanoate	1335	nd	0.30	nd
	31	Butyl 2-butenate	1351	nd	0.15	16.49
	33	Butyl hexanoate	1413	nd	nd	66.07
	34	Hexyl butanoate	1416	nd	0.48	79.15
	35	Ethyl octanoate	1435	nd	nd	141.56
	37	Isopentyl hexanoate	1459	nd	nd	79.97
	46	Ethyl decanoate	1640	1.46	11.66	139.31
	55	Ethyl dodecanoate	1824	nd	10.72	82.42
	60	Ethyl tetradecanoate	2052	0.37	2.40	40.77
Acids	36	Acetic acid	1452	14.64	0.50	nd
	45	Butanoic acid	1630	nd	3.41	146.98
	57	Ethyl hexanoic acid	1962	2.02	0.11	nd

Note: nd: not detected

Ethyl butanoate was the highest ester group in Agri Gardina 45 (1793.91 ng g<sup>-1</sup>) and Garifta Orange (228.42 ng g<sup>-1</sup>) (Table 1). This compound was associated with a sweet and fruity aroma. The isopentyl butanoate and ethyl crotonate were also identified and known to contribute to the pleasant aroma. The isopentyl butanoate or isoamyl butyrate was associated with a fruity, pineapple, and banana-like aroma (Zhu and Yu 2020), while ethyl crotonate was associated with a fruity, sour, caramel one (Mamede et al. 2017).

Alcohols were 6-7% of the total volatile group in all samples, including alcohol compounds were ethanol, methyl alcohol, 1-butanol, 3-hepten-1-ol, 3-nonen-1-ol, and phenylethyl alcohol. Ethanol was reported to give the apple juice and banana aroma (Burdock 2010), and it was found in relatively high amounts compared to other alcohol compounds in 3 cultivars. Several terpene and aromatic alcohols were also detected. Some of them were butanol, (Z)-3-hexenol, 1-hexanol, and 1-hexadecanol were present in notable amounts in many studied cultivars (Pino et al. 2005).

The sesquiterpene hydrocarbons group was also present in all samples in low proportion (Table 1). The relative concentrations of sesquiterpene hydrocarbons of Garifta Merah, Agri Gardina 45, and Garifta Orange were 2%, 1%, and less than 1%, respectively (Figure 1). In this study, the sesquiterpene hydrocarbon compounds included  $\alpha$ -gurjunene, caryophyllene,  $\alpha$ -caryophyllene,  $\beta$ -selinene, and  $\alpha$ -selinene. Caryophyllene and  $\alpha$ -caryophyllene were found in three mango cultivars, and they contribute to a clove-like aroma (Sulistyoningrum et al. (2017). Among the sesquiterpene hydrocarbons,  $\beta$ -caryophyllene,  $\alpha$ -humulene, and eremophilene were predominant in most Cuban mango cultivars (Pino et al. 2005).

Garifta Merah was the only cultivar with aldehydes groups; their concentration was 3% of the total group (Figure 1). Hexanal, 2-nonenal, and 2,6-nonadienal were aldehyde compounds identified in this study (Table 1). These compounds have been known to correlate with a green and grassy aroma. Pino et al. (2005) reported that hexanal compounds provide a grassy, green, and fresh-green odor, while 2-nonenal provides a cucumber aroma in Macho and Obispo mangoes. Chen et al. (2015) also reported that 2,6-nonadienal was responsible for green and

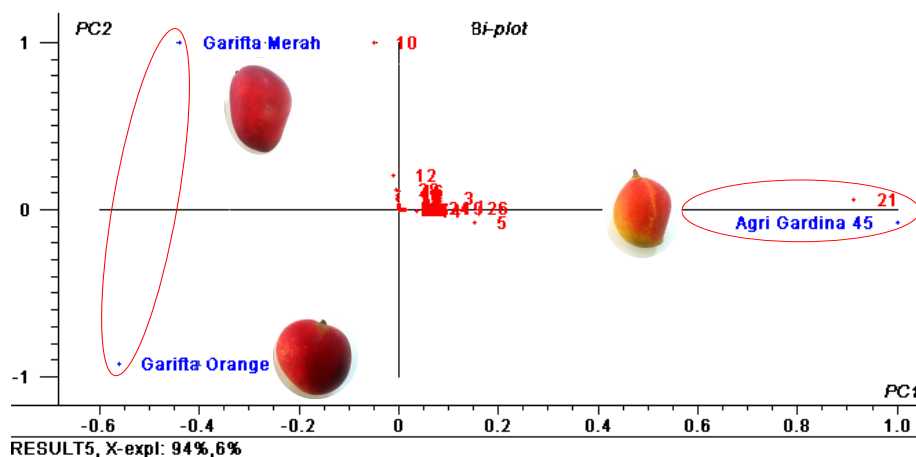
vegetable aromas. Some Cuban mango cultivars contained quantifiable amounts of aldehyde compounds, including hexanal, (E)-2-hexenal, (E, Z)-2,6-nonadienal, (E)-2-nonenal, (Z)-11-pentadecenal, pentadecanal, hexadecanal, 2,5-dimethyl-4-methoxy-3(2H)-furanone, and (E)- $\alpha$ -ionone (Pino et al. 2005).  $\gamma$ -terpinene, 1-hexanol, hexanal, terpinolene trans-2-heptenal, and p-cymene were also responsible compounds for the aroma of Chinese mangoes (Liu et al. 2020).

The acid groups were identified at 1% concentration in Garifta Orange and Agri Gardina 45, while in Garifta Merah was less than 1% (Figure 1). Acetic, butanoic, and ethyl hexanoic acids were in different proportions in all samples. Acetic acid was associated with vinegar and grape aroma, while butanoic acid was with green-fruity and apple-like notes (Dias et al. 2016). Pino et al. (2005) reported that hexadecanoic acid was the principal acid found in 20 Cuban mango cultivars.

Lactones, oxygenated monoterpenes, non-terpene hydrocarbons, furans, and ketones were less than 1% (Figure 1). The  $\gamma$ -caprolactone was found in Agri Gardina 45, while  $\gamma$ -nonalactone was found in Garifta Merah and Garifta Orange. Those compounds were associated with coconut odor (Wu et al. 2022). The identified oxygenated monoterpenes in Garifta Merah were carvone and p-cymene-8-ol, while linalool was in Agri Gardina 45. Linalool was known to be associated with floral aroma (Bechen et al. 2022). Non-terpene hydrocarbon identified in Garifta Merah and Garifta Orange mangoes was naphthalene, and Furaneol and 2-pentyl-furan were found only in Garifta Orange.

### Principal Component Analysis (PCA)

Although the total contents for each volatile group in the three mango cultivars had previously shown, the specific volatile compounds of each cultivar were identified by Principal Component Analysis (PCA). PCA is a method to reduce the dimensions of multivariate data sets (Zakaria et al. 2018). This study used PCA to determine each cultivar's dominant volatile compounds as secondary metabolites. The result of PCA analysis showed that both PC (Principal Compound) accounted for 100% (PC1: 94%; PC2: 6%) of total diversity (Figure 2).



**Figure 2.** PCA biplot of Garifta Merah, Garifta Orange, and Agri Gardina 45 volatile compounds. In total, 100% of the data variation is represented by PC1 (94%) and PC2 (6%)



Agri Gardina 45 mango was positively positioned on PC1 that characterized by some pleasant volatile aromas, such as  $\beta$ -trans ocimene (floral and herbaceous), ethyl butanoate (sweet and fruity), and isopentyl butanoate (fruity, pineapple, and banana-like) (Zakaria et al. 2018; Zhu and Yu 2020). The other two cultivars were on the left side of PC1, i.e., Garifta Merah was in a positive PC2 characterized by higher  $\alpha$ -pinene (no 10). The Garifta Orange was in negative PC1. The PC2 position of this cultivar was characterized by high ethyl butanoate.

### Odor Active Value (OAV)

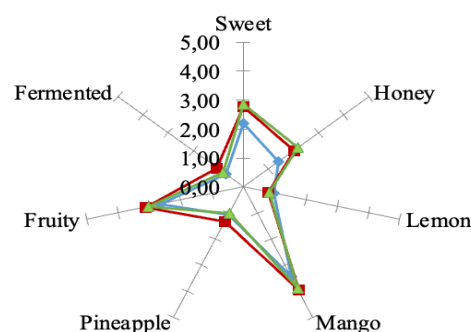
The contributions of volatile compounds depend not only on the amounts of compounds but also on the odor threshold value. Some studies used the Odor Active Value (OAV) calculation to assess the contribution of each compound to fruit aroma by comparing the compound concentration with its odor threshold. OAV values higher than one were considered an active fruit aroma contributor (Wu et al. 2018). The OAV values of volatile compounds identified in Garifta Orange and Garifta Merah are presented in Table 2. The result showed that those cultivars had different aroma compound contributors.

Two-nonenal-E; 2-nonadienal- (E, E); 3,6-Nonadien-1-ol, (E, Z)-;  $\alpha$ -pinene;  $\beta$ -pinene; and terpinolene were compounds that had OAV>1 in Garifta Merah (Table 2). These compounds have previously been reported to correlate with 'green and pine' aromas. Compounds with OAV>1 in Garifta Orange were  $\alpha$ -pinene, propyl butanoate, ethyl butanoate, ethyl hexanoate, and isopentyl butanoate (Table 2). Pleasant and fruity-related compounds were found as OAV>1 in Agri Gardina 45: propyl butanoate,  $\beta$ -trans-Ocimene,  $\beta$ -cis-Ocimene, linalool, ethyl butanoate,

and isopentyl butanoate (Table 2). Aroma contributors found in all samples were different from that of 5 Chinese mango cultivars reported by Liu et al. (2020), showing that  $\gamma$ -terpinene had the highest OAV with the range of 3.04-10.04, followed by  $\beta$ -phellandrene (2.41-3.41), hexanal (1.10-16.97), and 1-nonanal (5.37-56.2).

### Sensory aroma profiles

The RATA sensory test characterized the sensory profile of three mango cultivars. It was considered a sensory method that could improve sample description and discrimination (Giacalone and Hadelund 2016). Opperman et al. (2017) reported that RATA had a similar configuration to Descriptive Analysis (DA). RATA provide an excellent alternative to time and resource. The spider web descriptive sensory test by the RATA method is presented in Figure 3.



**Figure 3.** Spider web sensory attributes intensity of Garifta Merah (—), Garifta Orange (—), Agri Gardina 45 (—) mangoes

**Table 2.** Odor Active Value (OAV) value of volatile compounds

Compounds	OT (ppb)	OAV			Description
		Garifta Merah	Garifta Orange	Agri Gardina 45	
Ethyl acetate	5 <sup>a</sup>	7	3	6	Fruity, brandy-like, pineapple
Ethanol	8 <sup>a</sup>	25	6	118	Alcohol-like
Ethyl butanoate	0,1 <sup>a</sup>	-	2284	17939	Sweet, fruity, pineapple
Hexanal	4,1 <sup>a</sup>	1	-	-	Green, fatty, grassy
Propyl butanoate	0,8 <sup>a</sup>	-	2	16	Pineapple and apricot
$\alpha$ -Pinene	2,5 <sup>a</sup>	1127	86	-	Pine, turpentine, resin-like
$\delta$ -3-Carene	770 <sup>b</sup>	1	<1	-	Sweet
$\beta$ -Pinene	140 <sup>a</sup>	2	<1	-	Turpentine (a dry woody or resinous)
D-Limonene	4 <sup>a</sup>	78	5	15	Pleasant, lemon-like
Butyl butanoate	87 <sup>a</sup>	<1	<1	10	Fruity (pear-pineapple-like)
$\beta$ -Trans-Ocimene	34 <sup>c</sup>	-	<1	281	Floral
Ethyl hexanoate	0,3 <sup>a</sup>	-	27	-	Fruity ( pineapple-banana)
$\gamma$ -Terpinene	3 <sup>b</sup>	9	1	13	-
$\beta$ -cis-Ocimene	34 <sup>c</sup>	<1	<1	17	Herbaceous, sweet
Isopentyl butanoate	0,13 <sup>b</sup>	-	22	10598	Fruity, pineapple, banana
Terpinolen	200 <sup>b</sup>	2	<1	-	Sweet-piney
Ethyl octanoate	5 <sup>a</sup>	-	-	28	Pleasant, fruity
2-Nonenal, (E)-	0,1 <sup>a</sup>	774	-	-	Fatty
Linalool	4 <sup>a</sup>	-	-	13	Floral
2,6-Nonadienal, (E,E)-	0,09 <sup>a</sup>	601	-	-	Tallowy, green vegetable
Caryophyllene	64 <sup>a</sup>	1	<1	<1	Woody-spicy, dry, clove-like
Butanoic acid	240 <sup>a</sup>	0	<1	1	Butter-like
$\gamma$ -Caprolactone	7 <sup>a</sup>	-	-	1	Coconut and fatty
3,6-Nonadien-1-ol, (E,Z)-	10 <sup>a</sup>	6	-	-	Pleasant, sweet, cucumber, honey with oily nuance
Phenylethyl alcohol	0,015 <sup>a</sup>	228	18	-	Rose-like, honey

Note: OT: odor threshold (ppb) from <sup>a</sup>(Burdock, 2010), <sup>b</sup>(Pino and Mesa 2006), <sup>c</sup>(Tamura et al. 2001)

Eight aromas that assessors detected were honey, sweet, fruity, mango, lemon, floral, fermented, and pineapple. Those attributes were associated with several volatile compounds identified using GC-MS (Table 1) and the OAV (Table 2). The sweet, fruity, and pineapple aroma was correlated with ethyl butanoate, isopentyl butanoate, and ethyl crotonate (Zhu and Yu 2020). The mango and lemon aroma were associated with  $\delta$ -3-Carene (Pandit et al. 2009; Baccati et al. 2021). The presence of  $\beta$ -trans-Ocimene was responsible for the floral aroma detected by assessors (Zakaria et al. 2018).

Garifta Merah, Garifta Orange, and Agri Gardina 45 had different intensities of all aroma attributes. The intensity of honey, sweet, mango, and fruity characteristics of Garifta Orange and Agri Gardina 45 mango was higher than that of Garifta Merah. Those attributes might be the contributions of ethyl butanoate and isopentyl butanoate compounds with  $OA V > 1$ ; both have  $OA V > 10.000$  (Table 2). The relative concentration of ethyl butanoate was  $228.42 \text{ ng.g}^{-1}$  and  $1793.91 \text{ ng.g}^{-1}$  in Garifta Orange and Agri Gardina 45, respectively. Isopentyl butanoate contributed to the aroma and had a relative concentration of  $2.85 \text{ ng.g}^{-1}$  (Garifta Orange) and  $1377.71 \text{ ng.g}^{-1}$  (Agri Gardina 45).

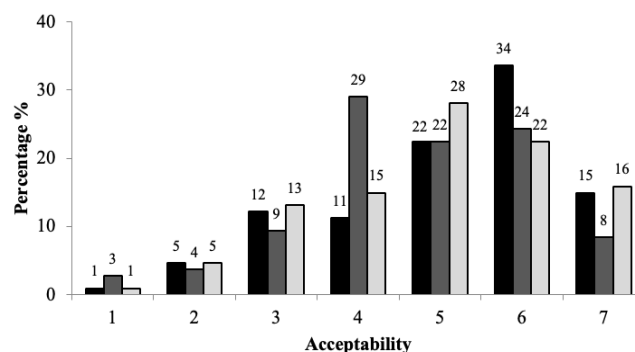
The PCA biplot of the RATA test is presented in Figure 4. The results showed that Garifta Merah, Garifta Orange, and Agri Gardina 45 were separated based on their sensory profile. As mentioned before, it might be attributed to some volatiles that characterized each mangoes cultivars. The position of Garifta Merah was in negative PC1 and PC2, and the lemon aroma characterized this cultivar. While Garifta Orange was in positive PC1 and PC2, in which the attribute of fermented fruit, mango, and floral aroma were the characteristics of this mango. As opposed to the other two mangoes, Agri Ardina 45 was characterized by a strong sweet and honey aroma in positive PC1 and negative PC2.

### Aroma acceptance

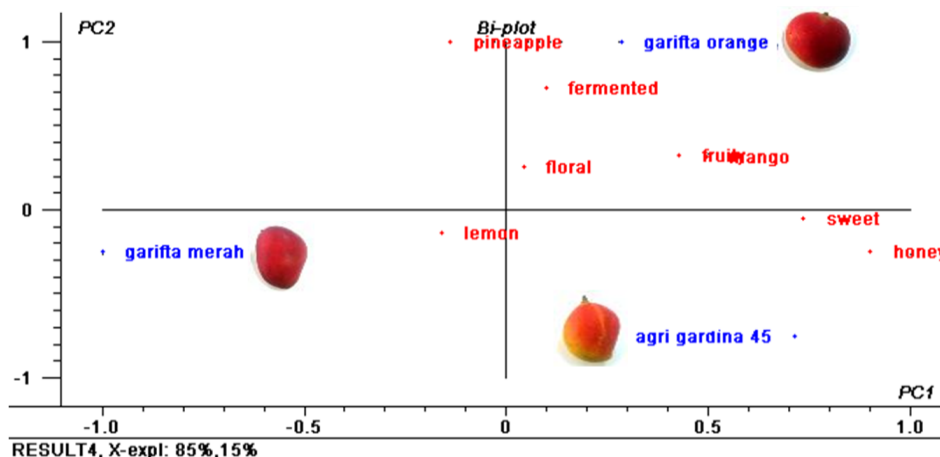
The aroma acceptance of samples was evaluated using the hedonic test with a 7-point scale, ranging from very

dislike to very like. Figure 5 presented the frequency percentage of the hedonic scale score of overall aroma acceptance of Garifta Merah, Garifta Orange, and Agri Gardina 45. The response frequencies of all samples were more concentrated in a score of 5-7 (like to very like). It means that the aroma of those cultivars was well-accepted by assessors.

The total acceptance response (score 5-7) of Garifta Orange (71%) was the highest among cultivars, followed by Agri Gardina 45 (66%) and Garifta Merah (54%). The result showed that the overall aroma of Garifta Orange and Agri Gardina 45 was more acceptable than that of Garifta Merah. It might be due to pleasant aroma compounds, i.e., ethyl butanoate, isopentyl butanoate, and ethyl crotonate, present only in Garifta Orange and Agri Ardina 45 (Table 1). Garifta Orange was characterized by high ethyl butanoate in the PCA biplot of volatile compounds (Figure 2). It might correlate with the RATA PCA, indicating that Garifta Orange was characterized by pineapple, mango, fruity, and fermented aroma attributes (Figure 4). These results were probably the underlying reason for the highest liking of overall aroma acceptance for Garifta Orange.



**Figure 5.** Frequency of overall acceptance scores of Garifta Orange (■), Garifta Merah (■), and Agri Gardina 45 (□) mangoes (1: very dislike; 7: very like)



**Figure 4.** PCA biplot RATA sensory of Garifta Merah, Garifta Orange, and Agri Gardina 45 mangoes. In total, 100% of the data variation is represented by PC1 (85%) and PC2 (15%)

From the results of this study, it can be concluded that metabolomics based on volatile compounds can determine differences between mango cultivars/varieties. Garifta Merah, Garifta Orange, and Agri Gardina 45 have different compositions of volatile compounds and sensory aroma attributes. These characteristics were interrelated in several cultivars. Garifta Merah was characterized by a high content of  $\alpha$ -pinene (pine, turpentine-like, and resin-like); however, it was sensory characterized by the lemon aroma. Ethyl butanoate was a volatile compound of Garifta Orange responsible for its specific fruity aroma. The high content of  $\beta$ -trans ocimene, ethyl butanoate, and isopentyl butanoate characterized Agri Gardina 45. The presence of ethyl butanoate correlated to its' specific sweet aroma. Those compounds were aroma contributors since their OAV values were  $>1$ . The overall aroma of Garifta Orange was the most liked. It indicated that assessors in this study preferred a sweet and fruity mango aroma.

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