

Investigation of Aroma Compounds of Queen Anne's Pocket Melon (*Cucumis melo* L. ssp. *dudaim*) Juice

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Abstract

Queen Anne's pocket melon (*Cucumis melo* L. ssp. *dudaim*) also known as Fragrant melon fruit stands out among other melon cultivars due to its unique, particularly exotic, and dainty odour. The volatile components of Turkish pocket melon juice were isolated by purge and trap extraction methods and detected by gas chromatography-mass spectrometry (GC-MS). According to GC-MS results, a total of 36 volatile compounds were identified and quantified including 12 acids, 11 alcohols, 6 aldehydes, 5 esters, and 2 ketones. The most prominent volatile compounds in pocket melon juice were found to be 3-penten-2-ol, 3-hydroxy-2-butanone, ethyl butanoate, and ethyl 2-methylbutanoate. The results showed that a wide variety of aroma groups contributed significantly to the distinctive odour of Turkish pocket melon juice.

Keywords: *Cucumis melo* L. ssp. *dudaim*, pocket melon, aroma, volatile compounds, GC-MS

1. INTRODUCTION

Melon (*Cucumis melo* L.), a member of the Cucurbitaceae family, is widely cultivated and consumed in temperate, subtropical and tropical regions all over the world (Kesh and Kaushik, 2021; Lecholocho et al., 2022). The popularity of melon is associated with its fragrance and aromatic properties, as well as its nutritional content including antioxidants, vitamins, polyphenols, and minerals (Soltani, 2021). Melon fruit is used in the food, cosmetic and pharmaceutical industries (Gomez-Garcia et al. 2020).

Melons are divided into two subspecies based on the hypanthium's hairiness: subspecies *melo* with hair on the hypanthium; and subsp. *agrestis* without any hair on the hypanthium. Botanical groups belonging to the subsp. *agrestis* are found in eastern Asia from India to Japan and the subsp. *melo* from India to Europe and the New World

(Jeffrey, 1980). This species is subdivided into seven botanical variants: cantaloupensis, reticulatus, inodorus, flexuosus, conomon, chito, *dudaim* and the *dudaim* variant includes the Queen Anne's pocket melon. These groups, which do not have much economic importance in the United States are grown in some local markets (Boyhan et al., 2017). *Dudaim* melons, known as Queen Anne's Pocket Melons, are a special type of melon (Hatami et al., 2019) and although they are not sweet-tasted, they are highly preferred for their intense and pleasant aroma, decorative beauty, and medicinal properties (Soltani et al., 2010; Paris et al., 2012). They are generally grown for ornamental or aromatic uses from Türkiye and the Caucasus to Afghanistan (Aubert et al., 2006).

Aroma is one of the most important factors in fruit quality and consumer preference (Tang et al.,

2010). Pocket melons have a very unique fragrant, exotic, and musky scent, often described as a blend of melon, pineapple, and jasmine. In Victorian times, these melons were carried in the pocket as a body fragrance. It is said that Queen Anne herself carried one, which would explain why common names such as Her Majesty's melon have been applied (Aubert and Pitrat, 2006). Despite the attractiveness of their velvety rind, these melons have a whitish, flabby, tasteless pulp and are barely edible (Aubert et al., 2006). This type of melon is characterized by small, round, reddish-yellow fruits with a diameter of about 6 cm, and smooth and velvety skin, with dark yellow streaks (Hatami et al. 2019; Paris, 2012).

Maturity at harvest is one of the most important factors affecting fruit quality, especially in melons, due to the presence of climacteric and non-climacteric melon groups. Maturity at harvest shows a great influence on the sugar content, volatile content, and texture of the melon fruit (Beaulieu and Lancaster 2007; Perkins-Veazie et al. 2012; Hatami et al. 2016).

The studies about the determination of pocket melon volatiles are still limited in the literature. The oldest known research conducted by Shu et al. (1995) reported that compounds with low odour threshold values like (Z)-3-hexenol, 3,6-nonadienol and some esters contributed to the specific odour of pocket melons. In another study, Aubert and Pitrat (2006) studied the rind aroma of the melon in detail and discovered that volatile levels in the rind were higher than in the flesh. Alcohols, thioether esters, and acids were reported to be likely the main contributors to the Queen Anne's Pocket Melon's distinctive aroma, especially in the rind, as their abundance and low odour threshold suggests.

This current study aims to characterize volatile compounds and some general properties of Queen Anne's Pocket Melon cultivated in Türkiye. The volatiles were extracted by the application of purge and trap technique and determined by gas chromatography and mass spectrometry.

2. MATERIALS AND METHODS

2.1. Samples and Chemicals

Fully ripe Queen Anne's pocket melons (*Cucumis melo* L. ssp. *dudaim*) were collected in Mardin province of Türkiye in 2022. Immediately after harvesting, the skin of the fruits was delicately removed using a paring knife and the pulp was mashed with a blender. After blending, the pulp and juice were macerated at 40°C for 4 hours. The juice was centrifuged (Kubota, Tokyo, Japan) at 20°C, 5500 rpm for 10 minutes before the analyses were performed. The standard volatile compounds, dichloromethane, sodium sulfate, and 4-nonanol were purchased from Sigma-Aldrich (Steinheim, Germany).

2.2. Method

2.2.1. General Chemical Analysis

2.2.1.1. Colour Measurement

In the melon juice obtained as a result of centrifugation, colour measurement was performed using Hunterlab (HunterLab Colour Quest, Reston, Virginia, USA) device. The measurement was performed in five repetitions and the results were given with L*, a*, and b* values according to the CIE Lab colour scale. Chroma and hue values were calculated according to the formulas below (Wibowo et al., 2015).

$$\text{Chroma} = \sqrt{(a^*)^2 + (b^*)^2}$$

$$\text{Hue} = \arctan\left(\frac{b^*}{a^*}\right)$$

2.2.1.2. pH and Total Acidity

The pH measurement of the samples was carried out by dipping the pH probe (Mettler Toledo S10, Mettler-Toledo, Switzerland) directly into a sample of pocket melon juice taken in a beaker (Amin et al., 2014). The procedure was repeated three times. The total acidity in samples is determined by Metrohm 800 Dosino 862 Compact Titrosampler (Metrohm AG, Herisau, Switzerland). 10 ml of the centrifuged melon juice samples were taken and 10 ml of pure water was added to it. The mixture was

titrated with 0.1 NaOH until the pH was 8.1. The total acidity is given as mg citric acid/L.

2.2.1.3. Soluble Solid Content (SSC)

The determination of SSC of pocket melon juice was measured with Atago Pocket Digital Refractometer (Pocket refractometer, PAL- α , Atago, Japan) at a temperature of 2°C. The results are given in °Brix.

2.2.2. Aroma Analysis

2.2.2.1. Purge and Trap Extraction

The purge and trap technique was used in the extraction of volatiles from the pocket melon samples. In the purge and trap method, the samples (6 g) were placed in 20 ml glass vials and two holes were drilled in the crimp caps of the vials so that the tip that closes one and brings the carrier gas to the other could pass through. 41.5 μ g of 4-nonanol was added to the samples as the internal standard. The samples that were ready for extraction before the gas flow started were heated in a water bath at 60°C for 10 minutes. Nitrogen gas was passed through the vials at a rate of 50 ml per minute. It was ensured that the substances were attached to the cartridge containing C18 with the help of nitrogen gas. The trap absorbing the volatiles was removed from the samples and kept for 90 minutes in a water bath at 60°C under nitrogen gas flow and the cartridges were washed with dichloromethane solvent. Then, the extracts taken into the evaporation balloons were concentrated with the help of the Vigreux column in order to remove the solution. The liquid obtained in concentrated form was injected directly into the GC-FID and GC-MS-O systems and aroma compounds were determined. The extraction was performed in three repetitions. The amounts of volatiles were calculated by the internal standard method according to Sevindik et al. (2020).

2.2.2.2. GC-MS Analysis

The gas chromatography (GC) system used in a study consisted of Shimadzu QP2020 chromatograph interfaced with a flame ionization detector (FID) and a mass selective detector (MSD)

(Tokyo, Japan) with a DB-Wax column (30m length \times 0.25mm i.d. \times 0.5m thickness, J&W Scientific, Folsom, CA, USA). An aliquot of 2 μ L of the extract was injected in pulsed splitless mode (40 psi; 0.5min) while the injector and FID detectors were set at 270°C and 280°C, respectively. A flow rate of 1.5mL min⁻¹ was used for the carrier gas (helium). The oven temperature of the DB-Wax column was raised from 50 °C to 250 °C at 4°C min⁻¹ with a 10 min holding time. The same oven program was used for the MSD. The MS parameters were as follows: ionization energy of 70 eV, mass range m/z of 30–300 amu., scan rate of 2.0 scans⁻¹, interface temperature of 250°C, and source temperature of 180°C. The aroma compounds were determined by comparison of their retention index and their mass spectra with those of a commercial spectra database (Wiley 11, NBS 75 k) and the instrument's internal library. After identification, 4-nonanol was used as an internal standard. A combination of experimental calibration by internal standard and FID response factors prediction was carried out according to de Saint Laumer et al. (2010). The mean values of the triplicate analysis results were calculated. The mean values of the triplicate analysis results were calculated. Retention indices of the compounds were computed using n-alkane (C8–C32) series (Guclu et al., 2022).

3. RESULTS AND DISCUSSION

3.1. General chemical properties of pocket melon

The mean values of the general chemical analysis are shown in Table 1 for Turkish pocket melon juice. pH, titratable acidity soluble solid content (SSC) of juice samples were determined as 5.61, 3.86 mg citric acid/L, and 6.2% respectively. Similarly, Lignou et al. (2013) reported pH values changing between 5.20 and 6.55 for four different melon (Galia and acidic) varieties. The authors indicated that muskmelons coded as acidic had lower pH values and this affected the sensory evaluation of

samples resulting in more sour and sharp sensations. Another study comparing different melons in terms of physicochemical properties investigated eleven commercial varieties from *C. melo* var. cantaloupensis, *C. melo* var. inodorus, and *C. melo* var. dudaim classes (Guler et al., 2013). The authors reported that dudaim pocket melons

had lower pH (5.17) value than the others (6.07-6.86). The pocket melon had also the highest acidity among the group with 5.42 g citric acid / 100g while inodorus cv. had 1.16 and cantaloupensis cv. had 0.92 g citric acid / 100g acidity.

Table 1. General composition of pocket melon juice

Analysis	Pocket melon juice
pH	5,61±0,01
Titrateable acidity (mg citric acid /L)	3,86±0,02
SSC (%Brix)	6,2±0,10
Colour	
L*	50,48±0,01
a*	8,65±0,02
b*	37,59±0,05
Chroma	38,57±0,01
Hue	13,16±0,01

Consumers' perceptual acceptance of the fruit is significantly influenced by colour. acceptance of the fruit is significantly influenced by colour and this parameter is of importance for many reasons. L*, a*, and b* colour values of the pocket melon were determined as 50.48, 8.65, and 37.59 respectively. Chroma and hue values were found to be 38.57 and 13.16. In a previous study by Fundo et al. (2018), different parts of cantaloupe melons including juice, pulp, peel, and seed were investigated in terms of physicochemical characteristics, bioactive compounds, and antioxidant activity. The L*, a*, and b* values of different parts of melons were between 37.80-57.12, -8.91-13.61, and 24.38-30.27 respectively. A similar set of results to our findings were reported for the pulp colour of cantaloupe melons with levels of L*:51.16, a*:9.10, and b*:30.27. Additionally, the results being all positive were associated with the presence of β -carotene.

3.2. Volatile compounds of pocket melon samples

36 compounds were identified and quantified by GC-MS following purge and trap extraction including 12 acids, 11 alcohols, 6 aldehydes, 5 esters, and 2 ketones. The relative abundances of the main volatiles in the pocket melon juice are shown in Table 1. The general total of aroma compounds was quantified as 10245,7 $\mu\text{g/L}$. As shown in Table 1, alcohols and acids were qualitatively and quantitatively the main components, accounting for 42 and 21% of the total volatiles quantified in pocket melon juice. Among all the volatiles, 3-penten-2-ol (1918 $\mu\text{g/L}$), 3-hydroxy-2-butanone (1408 $\mu\text{g/L}$), ethyl butanoate (638 $\mu\text{g/L}$), and ethyl 2-methylbutanoate (507 $\mu\text{g/L}$) were determined with the highest concentrations.

Table 2. Aroma compounds of pocket melon juice

No	LRI	Compounds	Concentration (µg/L)	Identification
Alcohols				
1	1142	1-Butanol	187±0,8	LRI, MS, Std
2	1166	3-Penten-2-ol	1918±43,5	LRI, MS, Std
3	1202	2-Methyl-1-butanol	569±12,8	LRI, MS, Std
4	1291	2-Hexanol	762±8,8	LRI, MS, Std
5	1308	2-Methyl-2-buten-1-ol	225±6,3	LRI, MS, Std
6	1343	1-Hexanol	102±5,0	LRI, MS, Std
7	1369	(E)-3-Hexen-1-ol	406±28,8	LRI, MS, Std
8	1840	Benzyl alcohol	56,3±4,2	LRI, MS, Std
9	1873	2-Phenylethanol	26,5±1,8	LRI, MS, Std
10	1949	1-Dodecanol	21,6±0,6	LRI, MS, Tent
11	2099	2-Phenoxy ethanol	22,1±1,8	LRI, MS, Std
Acids				
12	1421	Acetic acid	490±1,3	LRI, MS, Std
13	1514	Propanoic acid	75,8±6,7	LRI, MS, Std
14	1547	2-Methyl-propanoic acid	218±4,3	LRI, MS, Std
15	1600	Butanoic acid	110±5,3	LRI, MS, Std
16	1644	2-Methylbutanoic acid	68,2±3,2	LRI, MS, Std
17	1706	Pentanoic acid	30,1±2,6	LRI, MS, Std
18	1815	Hexanoic acid	370±3,4	LRI, MS, Std
19	1922	Heptanoic acid	66,1±3,5	LRI, MS, Std
20	1927	(E)-3-Hexenoic acid	10,3±0,3	LRI, MS, Tent
21	2030	Octanoic acid	127±9,9	LRI, MS, Std
22	2135	Nonanoic acid	427±11,6	LRI, MS, Std
23	2240	Decanoic acid	154±11,2	LRI, MS, Std
Aldehydes				
24	1079	Hexanal	455±26,9	LRI, MS, Std
25	1376	Nonanal	79,9±4,7	LRI, MS, Std
26	1481	Benzaldehyde	44,1±1,6	LRI, MS, Std
27	1508	(E)-2-Nonenal	60,0±0,8	LRI, MS, Std
28	1557	(E,Z)-2,6-Nonadienal	242±1,0	LRI, MS, Std
29	1662	(E)-2-Octenal	15,9±0,9	LRI, MS, Std
Esters				
30	1041	Ethyl butanoate	638±23,9	LRI, MS, Std
31	1055	Ethyl 2-methylbutanoate	507±9,7	LRI, MS, Std
32	1417	Ethyl-(methylthio)acetate	71,4±0,5	LRI, MS, Tent
33	1493	Ethyl 3-hydroxybutyrate	102,5±3,0	LRI, MS, Std
34	1500	Ethyl 2-hydroxyisobutyrate	110±4,3	LRI, MS, Tent
Ketones				
35	1225	3-Hydroxy-3-methyl-2-butanone	71,0±1,9	LRI, MS, Std
36	1263	3-Hydroxy-2-butanone (acetoin)	1408±11,8	LRI, MS, Std

LRI: retention indices on DB-WAX column. Concentration mean values based on two repetitions as µg/L. Identification methods of identification; LRI: (linear retention index), Std: (chemical standard); MS: (mass spectra), Tent: Tentatively identified by MS.

Alcohols, as previously stated, formed a major part of the volatile profile with a total concentration of 4294 µg/L. Within this group, 3-penten-2-ol, 2-hexanol, 2-methyl-1-butanol, and (E)-3-hexen-1-ol had the highest amount.

Alcohols contribute positively to food flavour at low concentrations. These compounds are formed by the metabolic activities of microorganisms or the conversion of carbonyl compounds to alcohols (Reineccius, 2016).

Similarly, most of the alcohols found in this study were also reported for several melon cultivars including cantaloupes, inodorus, Dulce, Vedrantaïs, Noy Yizre'el, Tam Dew, and Rochet (Gonda et al., 2010, 2013, 2016; Guler et al., 2013). Apart from the alcohols mentioned before, benzyl alcohol, 2-phenylethanol and 1-hexanol were reported to contribute the overall aroma of melons (Gonda et al., 2010; Shi et al., 2020; Majithia et al., 2021).

Volatile acids had the quantitatively most numbers of compounds including acetic acid, propanoic acid, 2-methyl-propanoic acid, butanoic acid, 2-methylbutanoic acid, pentanoic acid, hexanoic acid, heptanoic acid, (E)-3-hexenoic acid, octanoic acid, nonanoic acid, and decanoic acid. Acetic acid and acids derived from the lipid oxidation reactions were found in higher amounts. Acetic acid was reported to be formed from the carbohydrate oxidation or fermentation pathways (Gonzalez et al., 2007; Qian et al., 2016).

Acetic acid, a crucial volatile compound responsible for the strong pungent and sour odours, was also reported before in pocket melon which was evaluated as sour by sensory panelists (Hernandez-Gomez et al., 2005; Saftner et al., 2006; Guler et al., 2013). Hence, this compound is thought to be a possible contributor to the particular sour flavour of pocket melons. The other lipid oxidation-derived acids were also reported previously in several melon cultivars with the lead of nonanoic acid (Shu et al., 1995; Majithia et al., 2021).

Ketones were the following substantial group determined in pocket melon juice. Although, only two compounds were detected as ketones (3-hydroxy-2-butanone and 3-hydroxy-3-methyl-2-butanone), particularly one of them, 3-hydroxy-2-butanone, had one of the highest concentrations (1408 µg/L) among the whole volatiles. This compound was also reported to be one of the constituents found in high amounts in melon cultivars like Protea and Vector from Italy, and pocket melon from

France and Türkiye (Senesi et al., 2002; Aubert and Pitrat, 2006; Guler et al., 2013). Additionally, it was reported to be one of the crucial key odorants of melons, bananas with butyric acid odour, of guavas with acidic and pungent odours (Jordán et al., 2001, 2003; Selli et al., 2012). Acetoin is a short-chain ketone derived from pyruvate and citrate metabolism pathways (Mallia et al., 2005) giving a buttery odour note to the products and this compound is commonly used in foods, cosmetics and chemicals for flavour and fragrance (Cheng, 2010; Xiao and Lu, 2014).

Esters are significant fragrance components that give many fruits their distinctive fruity, flowery scent characteristics. According to reports, these substances are created either through the metabolism of amino acids or the oxidation of fatty acids (Belitz et al., 2009). Alcohol dehydrogenase and alcohol acid transferase enzymes have also reportedly been shown to be useful in the production of esters. A total of five ester compounds including ethyl butanoate, ethyl 2-methylbutanoate, ethyl-(methylthio)acetate, ethyl 3-hydroxybutyrate, and ethyl 2-hydroxyisobutyrate were found in the pocket melon juice. Ethyl butanoate had the highest concentration and is followed by ethyl 2-methylbutanoate. These two compounds were reported in several melon cultivars previously and they were attributed to a fruity odour with a pineapple undertone and a powerful green, fruity apple-like odour respectively. They were reported to be the most abundant aliphatic esters in the Cantaloupe variety over Inodorus (Hayata et al., 2003; Saftner et al., 2006; Beaulieu and Lancaster, 2007; Guler et al., 2013). Another important volatile ester was found to be in a thioester form, ethyl (methylthio)acetate. Thioesters are of importance due to their low threshold values and are commonly found in melon cultivars. Wyllie and Leach (1992), who studied the aroma profile of Makdimon melon, indicated that ethyl (methylthio)acetate had an impact on the aroma of ripe fruit. Similarly, Sakamoto et al.

(2002) identified sulfur-containing compounds in the aromatic volatiles of *C. melo* L. cv. Miyabi and reported that ethyl(methylthio)acetate had a grassy cucumber-like odour.

As for the last group detected in pocket melon juice, six aldehydes including hexanal, nonanal, benzaldehyde, (*E*)-2-nonenal, (*E,Z*)-2,6-nonadienal and (*E*)-2-octenal had also affected the volatile profile. It has been reported in studies that these aldehydes are formed as a result of oxidative degradation of unsaturated fatty acids and that they are likely to contribute to the aroma of fruits due to low odour threshold values (Buttery, 2010; Oliviera et al., 2010). Of these aldehydes determined, Schieberle et al. (1990) reported that (*E,Z*)-2,6-nonadienal and (*E*)-2-nonenal appeared both in the aromagram of muskmelons and cucumber and they were attributed as key odorants.

4. CONCLUSION

The present research demonstrated the aroma profile of Turkish Queen Anne's pocket melon (*Cucumis melo* L. ssp. dudaim) juice. The volatile components of Turkish pocket melon were composed of five different aroma groups with alcohols, acids, and ketones forming 77% of the overall profile. According to GC-MS results, the most abundant volatile compounds were found to be 3-penten-2-ol, 3-hydroxy-2-butanone, ethyl butanoate, and ethyl 2-methylbutanoate. The results showed that our findings were correlated with the literature and the researches should be enhanced to evaluate and elucidate the physicochemical structure of Queen Anne's pocket melon grown in Türkiye.

CONFLICT OF INTEREST STATEMENT

No potential conflict of interest was reported by the authors.

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