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RESEARCH ARTICLE

Characterization of volatile compounds from processed pineapple waste by Static Headspace Gas Chromatography-Mass Spectrometry (SHS-GC-MS/MS)

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ABSTRACT

The volatile compounds from processed pineapple waste (PPW) from two common cultivars of pineapple in India (Queen and King) were studied using Static headspace Gas Chromatography-Mass Spectrometry (SHS-GC-MS/MS). From the processed waste of Queen cultivar, a total of about 55 to 60 volatile compounds were identified; majorly esters (Methyl 2-methyl butyrate (50%), Methyl butyrate (9%), Ethyl 2-methyl butyrate (6%) and methyl hexanoate (2%), followed by terpenes and a few alcohols, alkanes, aldehydes, acids, lactones and other classes of compounds in minor quantities. From processed waste of King cultivar, a total of 60 to 65 volatile compounds were identified; again majorly esters (Methyl hexanoate (27%) followed by methyl butyrate (18 %) and methyl 2-methyl butyrate (15%), methyl octanoate (4%) in addition to terpenes, alcohols, alkanes, aldehydes and other classes of compounds present in minor concentrations. The study indicated that volatile compounds of pineapple waste was quite comparable to that of the pineapple juice and has the potential for utilization.

Keywords: Processed pineapple waste, static headspace, gas chromatography, volatile compounds, queen cultivar

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INTRODUCTION

Pineapple, *Ananas comosus* [L.] Merr., is a species from Bromeliaceae family grown in the tropical and subtropical regions of the world and it is known for its fruit (Lobo and Yahia 2017). It is the third major fruit crop in the world after banana and citrus in terms of production. The major varieties include 'Smooth Cayenne', which is the largest cultivated variety worldwide, followed by 'Singapore Spanish' ranking second for its commercial importance. A large quantity of pineapple throughout the world is used for salads, concentrates, juices, and jams. In India, a wide range of pineapple varieties are available; however, the 'King' and the 'Queen' are the most popular commercial cultivars. The 'King' cultivar is a late-maturing pineapple variety and is the leading commercial cultivar for juice production and is also suitable for canning. The fruit with light yellow colored flesh weighs 2-3 kg and is big in size, oblong in shape and slightly tapering towards the crown. The 'Queen' cultivar of pineapple is an early maturing cultivar, commonly used for the table as well as for processing purposes. It is deep golden yellow, less juicy than the 'King' variety, crisply textured with pleasant aroma and flavor (Sahu et al. 2017).

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Among the various pineapple products, the juice is largely consumed around the world either in the form of single strength, reconstituted or concentrated and in different blended compositions to obtain new flavors in beverages (Maria et al. 2008). The by-products generated during processing of pineapple include peel, core, residual pulp, stem, crown, and leaves, which constitute about 50% of total weight (Ketnawa et al. 2012). All this waste is rich in components like starch, reducing sugar, non-reducing sugar, total sugar, protein, ascorbic acid, etc (Hemalatha and Anbuselvi 2013). The pineapple leftover after juice extraction and filtration is a valuable source of particulate matter, fibre, lignin, cellulose, hemicelluloses, soluble solids, etc (Botelho et al. 2002).

The juice industry normally disposes of this leftover as ' waste' or as a source of fibre or as animal feed (Sreenath et al. 1994). However, this waste may contain volatile aroma compounds like that of pineapple juice, which indicates its potential for due exploration. Pineapple, as an aromatic fruit is known for unique appealing flavor due to its various volatile constituents like esters, aldehydes, alcohols, ketones, lactones, terpenes, terpenoids, hydrocarbons, etc, in general. (Marta et al. 2010). The extraction and characterization of volatile compounds from pineapple waste have been investigated by Barretto et al. (2013). These investigators used different extraction techniques like simple hydro-distillation, hydro-distillation using nitrogen gas and solid phase microextraction for extraction of flavor volatiles for their identification and characterization. It has been reported that pineapple residue contains a reasonable amount of flavor volatiles similar to that of pineapple juice. The volatile compounds of core and pulp of smooth Cayenne pineapple using headspace-solid phase microextraction (HS-SPME) and gas chromatography-mass spectrometry (GC/MS) were investigated by Wei et al. (2011). They identified the different odor impact compounds like ethyl 2-methyl butanoate, 2,5-dimethyl-4-hydroxy-3(2H)-furanone (DMHF), decanal, ethyl butanoate, nonanal, etc. possessing high odor activity values that are dominating in pineapple.

Several techniques have been reported for the extraction and detection of the volatile compounds in past. The method involved in the isolation of volatiles and detection affects the volatile profile of the food at large (Ettre, 2002). Use of static headspace sampling is one such technique that can avoid extensive sample preparation which in the majority of the cases is time-consuming and expensive and at the same time may not give accurate results due to errors in sample preparation. Headspace technique for the isolation of volatiles is fast, highly sensitive; minimizing the unwanted contamination and interferences from the other components present in the sample matrix and highly reproducible. It is also environmental friendly for eliminating the use of a harmful organic solvent in the analysis (principles of green chemistry). The present study involves use of static headspace (SH) technique for the isolation of volatiles from the processed pineapple waste (PPW) from two commonly available cultivars in India i.e. 'Queen and 'King' cultivars for their detection and characterization using Gas chromatograph coupled to a triple quadrapole mass spectrometer (GC-MS/MS).

MATERIALS AND METHODS

Sample preparation

Ripened pineapple fruit (*Ananas comosus*) of the two commonly available cultivars *i.e.* 'Queen and 'King' was purchased from the local market (Azadpur market, Delhi) and stored in a refrigerator at 5 ± 1 ^{o}C. The stem and the crown portions were removed and the fruit without skin was washed in tap water before slicing. The fruit slices (3 x 3 x 1.5 cm) were mashed in a in screw type juice extractor for 3 cycles and the pulp was pressed and filtered using stainless steel juice strainer (mesh) to obtain separation of clear juice and pomace. This leftover was blended in a blender to obtain a homogenous sample and stored in low-density polyethylene plastic bags at -18 ºC. Approximately 2 g of sample was placed in a 20 mL headspace (HS) vial sealed immediately with a Silica/PETF lined septum and aluminum crimp cap and placed in the autosampler tray for analysis.

Static headspace analysis

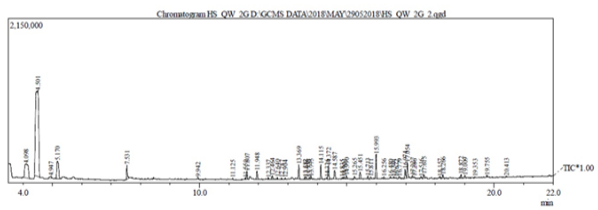
The study was carried out using a Shimadzu GC-MS-TQ 8040 Gas chromatography system coupled with Triple Quadrupole mass spectrometer 2010 ion source and attached to a Shimadzu HS-20 Headspace sampler (loop model). Static headspace was equilibrated at 85 ºC for 30 min with shaking speed of 80 times/min at a gas pressure of 15 psi for 0.5 min during sample heating. A known volume (1ml) of the sample vapors from the headspace gas was injected using a pressure loop system of the headspace sampler which flushes the sample into the transfer line leading to the analytical column. The sample was injected in splitless mode. The splitless inlet temperature was 105 ºC. Separation was performed on a Restak stabilwax column (30 m x 0.25 mm, 0.25 µm film thickness). Helium was used as a carrier gas at a constant flow rate of 1mL/min. The column oven temperature was programmed with an initial temperature set at 40 ºC for 2 minutes and then increased to 220 ºC at 7 ºC min^{-1} and held for 5 minutes.

The mass spectrometer was operated in electron ionization mode with the transfer line temperature at 260 ºC and ion source temperature of 230 ºC. A delay time of 2 minutes was given before recording the chromatogram to avoid the inclusion of solvent peak into the chromatogram. Electron ionization mass spectra were recorded over a mass range of 35 m/z to 500 m/z at 70 eV energy. A solvent delay of 0.2 min was used. The samples were analyzed in duplicate and blank runs were done before each analysis. Compounds were identified against the inbuilt reference mass spectra from the FFNSC and NIST 14 library. The data acquisition was performed in Q3 SCAN mode. The data were acquired to identify the compounds and determine appropriate ions for the later acquisition in SIM mode for future study.

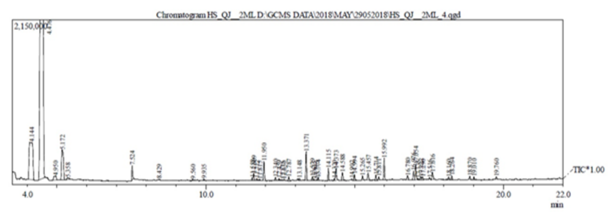
RESULTS AND DISCUSSION

The total ion chromatograms for the volatiles in the sample of both processed waste and the juice from the 'King' and the 'Queen' cultivar are given in Fig 1. The volatile compounds identified in the PPW and their odor description are listed as per Table 1 (queen cultivar) and Table 2 (king cultivar) indicates the various volatile compounds and their odor description. Based on the percent peak area of the various peaks obtained in the total ion chromatogram (TIC), esters represented the main class of aroma volatile compounds in PPW of both cultivars, followed by terpenes and alkanes. The volatile esters compounds identified in this study were in agreement with the previous studies conducted by Elss et al. (2005), Takeoka et al. (1989), Zheng et al. (2012), Wei et al. (2011) and Steingass et al. (2016) whereby, it has been reported that esters and their derivatives are primarily responsible for pineapple aroma. As indicated in Table 1, the analysis of the volatiles in the headspace of both i.e. the waste as well as the juice of 'Queen' cultivar indicated the presence of 55-60 volatile compounds, in which several ethyl and methyl esters such as methyl butyrate, ethyl butyrate, methyl 2-methyl butyrate, ethyl 2 methyl butyrate, methyl hexanoate, Ethyl hexanoate, methyl octanoate, methyl decanoate followed by hydroxy and other esters like dimethyl 2-hydroxy-2-methyl succinate, Methyl 4-decanoate, Methyl 2-methyl-3-oxo butanoate were predominating (consisting of about 85% of the total volatiles). From the results, it can clearly be said that the volatile compounds present in the PPW are almost similar to that present in the pineapple juice, though with variation in the concentration levels. However, the concentration of hydroxy and acetoxy esters may increase during pineapple ripening (Engel et al., 1989). Based on the percent peak area, the compounds with the highest concentration in the processed waste were methyl 2-methyl butyrate (50 %) followed by methyl butyrate (9 %), Ethyl 2-methyl butyrate (6 %) and methyl hexanoate (2%). Next to the esters, Terpenes (constituting about 8 % of total volatiles) like α - cubebene, β - cubebene, (+)-Sativene, β - Cedrene, 1,3,5,8-undeca tetraene, α - murolene, δ - cadinene, β - elemene, Bicyclogermacrene, etc. act as a second group of dominant odor constituents in pineapple fruit, similar to the findings of Wei et al. (2011). Besides the above-mentioned groups, other volatiles detected were alcohols (1 %), aldehydes (0.5 %), acids (0.3%), lactones (0.3 %) and certain alkanes (5 %). Among lactones, γ-hexalactone was identified in pineapple were present in leftover and juice of queen cultivar is contributing for a sweet aroma. Similarly, hexanoic acid responsible for typical pineapple aroma is also in higher concentration in PPW than juice.

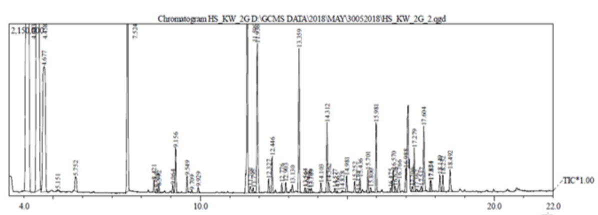
Fig 1. The total ion chromatograms (TIC) for the volatiles in the sample of both processed waste and the juice from the 'King' and the 'Queen' cultivar.



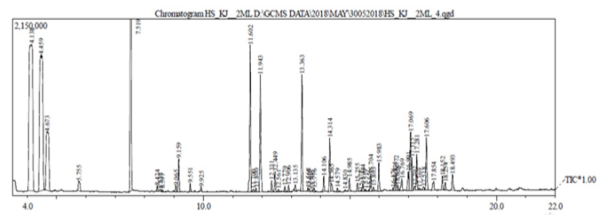
a. TIC for the Volatiles in Processed waste (Queen Cultivar)



b. TIC for the Volatiles in Juice (Queen Cultivar)



c. TIC for the Volatiles in Processed waste (King Cultivar)



d. TIC for the Volatiles in Juice (King Cultivar)

Table 1. Volatile Constituents identified in the processed pineapple waste (Queen Cultivar) using SHS-GC-MS/MS

Class	Retention Time (min)	Components	% Peak area[c]		% matching from the Library		Earlier identified in pineapple juice/pulp	Odor description (As reported in the literature)
			Juice	Left-over	Juice	Left-over		
Esters	4.09	Methyl butyrate	11.70	9.27	97[a]	97[a]	√	Apple, sweet, toast[d]
	4.50	Methyl 2-methyl butyrate	63.16	50.34	97[a]	97[a]	√	Pineapple[d], fruit, apple like[e]
	4.94	Ethyl butyrate	0.86	0.87	95[b]	94[a]	√	pineapple [d], fruity[e]
	5.17	Ethyl 2-methyl butyrate	4.84	6.04	97[a]	97[a]	√	pineapple heart [d] ,fruity [e]
	5.35	Acetyl propionyl	0.28	-	93[a]	-	-	n.d.
	7.53	Methyl hexanoate	1.02	2.06	93[a]	91[a]	√	Fruity, ester-like[f]

	8.42	Ethyl hexanoate	0.09	-	92[b]	-	√	Pineapple[h]
	9.56	Methyl heptanoate	0.08	-	93[b]	-	√	n.d.
	9.94	Methyl 2-hydroxy 2-methylbutanoate	0.12	0.31	93[a]	91[a]	√	woody[g]
	11.60	Methyl octanoate	0.50	1.20	97[a]	97[a]	√	fruity, winy, orange [d]
	12.57	Methyl 2,4-hexadienoate	0.10	-	78[a]	-	-	n.d.
	13.63	(S)-Methyl 3-hydroxybutanoate	0.10	-	79[a]	-	√	Mild, fruity, apple, winey
	14.32	Methyl 3-(methylthio) Propionate	0.17	-	86[a]	-	√	oniony, fruity at low level[b]
	14.37	methyl 2-methyl-3-oxobutanoate	0.96	2.33	87[a]	86[a]	√	n.d.
	17.13	Methyl linolenate	0.10	-	77[a]	-	-	n.d.
	15.99	Methyl decanoate	-	1.33	-	85[a]	√	oily winey fruity floral[j]
	12.46	Ethyl octanoate	-	0.63	-	87[a]	√	Fruity, winy[h]
	13.77	Methyl (E)-2-octenoate	0.19	0.42	93[a]	92[a]	√	fruity pear skin green cooked [j]
	14.58	Ethyl 3-acetoxy butyrate	0.67	1.59	82[a]	82[a]	√	fruity green galbanum pin eapple [j]
	15.99	Methyl 4-decanoate	1.56	3.79	93[a]	93[a]	-	n.d.
	16.67	Ethyl trans 4-decenoate	-	0.12	-	86[a]	√	Fruity, pear like[j]
Terpenes	11.81	Undecatriene-1,3,5-triene	0.10	-	89[b]	-	√	Fresh, pineapple like[e]
	11.94	(-)-Aristolene	1.57	1.53	88[a]	87[a]	-	n.d.
	12.33	α – bulnescene	0.25	0.29	88[a]	87[a]	-	n.d.
	13.36	α – copaene	2.16	2.34	96[a]	96[a]	-	n.d.
	14.99	Guaia-6,9-diene	0.44	0.47	83[a]	87[a]	-	n.d.
	15.26	β - cubebene	0.29	0.40	92[a]	85[a]	√	Fruity[m]
	13.14	Cyclosativene	0.20	-	91[a]	-	√	n.d.
	15.71	(+)-sativene	0.39	0.44	88[a]	88[a]	-	n.d.
	15.81	Modhephene	0.24	0.33	93[a]	90[a]	-	n.d
	12.78	1,3,5,8-undecatetraene	0.28	0.15	93[a]	87[a]	√	fruit ,fresh, pineapple[e]
	17.28	β – maaliene	0.25	0.16	90[a]	88[b]	-	n.d.
	17.61	α - muurolene	0.66	0.78	94[a]	94[a]	√	Woody[j]
	18.16	δ - cadinene	0.21	0.24	90[b]	83[a]	√	Wood, dry, mild[m]
	18.26	β - elemene	0.34	0.57	99[a]	89[a]	√	Herb, wax, fresh[n]

	16.78	δ- selinene	0.39	0.30	77[b]	75[a]	-	n.d.
	17.15	trans-Cadina-1(6),4-diene	-	0.24	-	83a	-	n.d.
	16.77	cis-Thujopsene	-	0.30	-	75[a]	-	n.d.
Alcohol	11.12	1-hexanol	-	0.19	-	94[b]	√	Chemical, winey[m]
	12.90	1-octen-3-ol	-	0.21	-	85[a]	-	earthy, green, oily, fungal[j]
	14.83	1-octanol	-	0.24	-	89[a]	√	Soapy[m]
	16.59	Nonanol	-	0.20	-	93[a]	√	Oily, floral, powerful[m]
Acid	19.75	Hexanoic acid	0.19	0.36	90[a]	90[a]	√	Pineapple[d]
Alkanes	11.55	n-Tetradecane	0.19	0.20	79[b]	83[b]	-	alkane[n]
	13.57	Pentadecane	0.26	-	94[a]	-	-	n.d.
	15.45	1-Iodotridecane	0.58	0.35	84[a]	80[a]	-	n.d.
	17.22	Heptadecane	0.27	0.32	93[a]	95[a]	-	n.d.
	12.64	2-methyloctacosane	-	0.24	-	85[a]	-	alkane[n]
	18.87	Heneicosane	0.27	0.75	96[a]	88[a]	-	alkane[n]
	16.26	n-eicosane	-	0.19	-	89[a]	-	alkane[n]
	19.35	n-hexacosane	-	0.16	-	84[b]	-	alkane[n]
Aldehyde	13.72	n-Decanal	0.13	0.26	84[a]	88[a]	√	Beef, musty, marine, cucumber[m]
	16.48	Phenylacetaldehyde	-	0.23	-	79[a]	√	Green, clover, honey, cocoa[j]
	16.59	Nonanal	0.24	-	94[a]	-	√	Piney, floral, citrusy [m]
Others	12.45	2-ethyl-4-methoxy cyclohexanone	0.19	-	81[a]	-	-	Fruity, rummy, tobacco, woody
	14.11	Cycloheptosiloxane, tetradecamethyl-	0.76	1.82	85[a]	85[a]	-	n.d.
	16.97	Cyclooctasiloxane, hexadecamethyl	0.80	1.41	91[a]	91[a]	-	n.d.
	17.05	Cyclohexene, 3(3-methyl-1-butenyl)-,(E)	1.27	2.61	90[a]	90[a]	-	n.d.
Lactone	17.51	γ- hexalactone	0.10	0.34	82[a]	82[b]	√	sweet, caramel, toast [d]

[a] NIST 18 Library, [b] FFNSC 1.3 Library [c] peak area percentage, [d] Sinuco et al. (2004), [e] Tokimoto et al. (2005), [f] Zheng et al. (2012), [g] Umano et al. (1992), [h] Pino et al. (2013), i Burdock (2005), [j]The good scents company, [k] Berger et al. (1985), [m] Aroma descriptors, [n] Terry et al. (1984), n.d., not detected.

Table 2. Volatile Constituents identified in the processed pineapple waste (King Cultivar) using SHS-GC-MS/MS

Class	Retention Time (min)	Components	% Peak area[c]		% matching from the Library		Earlier identified in pulp pineapple[c]	Odor description (As reported in the literature)
			Juice	Leftover	Juice	Leftover		
Esters	4.08	Methyl butyrate	18.59	20.13	97[a]	97[a]	√	Apple, sweet, toast[d]
	4.45	Methyl 2-methyl butyrate	15.02	17.25	97[a]	97[a]	√	Pineapple[d], fruit, apple like[e]
	5.15	Ethyl 2-methyl butyrate	-	0.06	-	89[b]	√	pineapple heart [d],fruity [e]
	5.75	Methyl Pentanoate	0.57	0.61	96[a]	96[a]	√	Odour not detected by experts [d]
	7.52	Methyl hexanoate	27.76	22.29	96[a]	96[a]	√	Fruity, ester-like[f]
	8.42	Ethyl hexanoate	0.17	0.27	96[a]	97[b]	√	Fruity, fruity, pineapple, banana[k]
	8.52	Methyl 5-hexanoate	0.09	0.09	93[a]	93[a]	√	n.d.
	9.06	Methyl (E)-4-hexenoate	0.20	0.19	93[a]	94[a]	√	n.d.
	9.15	Methyl (Z)-3-hexanoate	0.97	0.95	96[a]	96[a]	√	n.d.
	9.54	Methyl heptanoate	0.24	0.34	95[a]	96[b]	√	sweet, fruity, green, waxy, apple[j]
	9.70	Methyl (E)-2-hexanoate	-	0.05	-	91[a]	√	green, fruity, pineapple, earthy[j]
	9.92	Methyl 2-hydroxy 2methylbutanoate	0.14	0.15	94[a]	94[a]	√	woody[g]
	11.59	Methyl octanoate	4.52	8.00	98[a]	98[a]	√	fruity, winy, orange[d]
	12.44	Methyl 4-octanoate	0.62	0.82	92[a]	90[a]	√	n.d.
	12.56	Methyl sorbate	0.07	-	85[a]	-	-	sweet, fruity , spicy, green, anise [j]
	12.90	Methyl (Z)-3-octanoate	0.20	0.22	81[a]	93[a]	√	n.d.
	14.10	Dimethyl propanedioate	-	0.22	-	93[a]	-	n.d.
	13.56	Methyl heptacosanate	0.26	0.11	85[a]	82[a]	-	n.d.
	14.31	Methyl 3-methylthio-propionate	1.68	1.56	97[a]	97[a]	√	meaty, oniony, fruity at low level[b]
	14.36	Methyl 2-methyl-3-oxobutyrate	0.32	0.25	81[a]	81[a]	-	n.d.

	14.57	Ethyl 3-acetoxybutyrate\	0.12	0.11	81[a]	83[a]	√	Mint like[g]
	15.51	Methyl 2,4 (E,Z)-decadienoate	0.07	-	78[a]	-	-	n.d.
	15.43	Methyl decanoate	-	0.34	-	94[a]	√	Sweet, oily, nut like[m]
	15.98	Methyl 4-decanoate	0.86	1.42	95[a]	94[a]	√	n.d.
	16.47	Methyl 3-hydroxy hexanoate	0.93	0.11	87[a]	93[b]	√	ripened pineapple-like[g]
	17.06	Methyl 3-hydroxy pentanoate	2.53	-	80a	-	√	Honey like[g]
	17.85	2,4-dimethyl-3-pentanol acetate	-	0.22	-	74[a]	-	n.d.
	16.62	1-octen-4-ol acetate	0.17	0.15	79[a]	77[a]	-	n.d.
	18.49	Methyl 7-oxooctanoate	0.65	0.57	77[a]	77[a]	-	n.d.
	16.57	2-hydroxyisocaproic acid acetate	0.46	0.48	88[a]	89[a]	-	n.d.
Terpenes	8.59	Trans β- ocimene	0.05	0.14	93[a]	95[a]	√	Herbaceous, tropical, sweet[m]
	11.93	(-)-Aristolene	4.11	3.75	88[a]	98[a]	-	n.d.
	11.79	(E,E)-1,3,5-undecatriene	0.11	0.11	94[a]	94[a]	√	Fresh, pineapple like[e]
	12.32	α- Bulnescene	0.34	0.30	89[a]	89[a]	-	wood[n]
	12.77	1,3,5,8 undecatetraene	0.16	0.19	93[a]	94[a]	√	Fresh, pineapple like[e]
	14.98	Guaia -6,9-diene	0.44	0.36	89[a]	90[a]	-	n.d.
	15.25	β- cubebene	0.30	0.26	92[a]	95[b]	√	Fruity[m]
	15.37	Caryophyllene	0.05	0.04	90[a]	92[a]	√	Wood, spicy [m]
	17.60	α-muurolene	1.87	1.67	95[a]	95[b]	√	Woody[j]
	18.14	δ-cadiene	0.44	0.39	91[a]	95[b]	√	Wood, dry, mild[m]
	13.13	Cyclosativene	0.24	0.22	94[a]	94[a]	√	n.d.
	13.35	α- copaene	3.62	3.15	96[a]	96[a]	√	Wood, spice[n]
	13.76	γ- maaliene	0.11	0.13	72[a]	74[a]	-	n.d.
	15.70	α-Cedrene	0.52	0.48	88[a]	88[a]	-	wood[n]
	15.80	Modhephene	0.15	0.12	92[a]	95[a]	-	n.d.
	16.76	γ- gurjunene	0.53	0.42	89[a]	88[b]	√	Wood, balsamic[n]

	16.98	γ-muurolene	0.73	0.55	91[a]	93[a]	√	Herbal, woody, spicy[j]
	17.27	β- maaliene	1.10	0.95	93[a]	94[b]	-	n.d.
	18.15	β- cadinene	0.44	-	91[a]	-		woody[j]
	17.36	Germacrene D	0.06	0.06	91[a]	91[a]	-	Wood, spice[n]
	17.51	β-acoradiene	0.15	0.17	-	80[a]	-	n.d.
	18.25	β-elemene	0.23	0.37	90[a]	92[a]	√	Herb, wax, fresh[n]
	17.28	β-calarene	1.10	-	93a	-	-	n.d.
Alcohols	13.62	2-ethyl 1-hexenol	0.07	0.03	91[a]	84[a]	-	n.d.
	14.82	1-octanol	0.06	0.04	94[a]	95[a]	√	soapy[m]
	14.66	Linalool	-	0.03	-	90[a]	-	Fruity [m]
Aldehyde	11.70	Nonanal	0.14	0.23	94[a]	96[a]	√	Piney, floral, citrusy [m]
	13.70	Decanal	0.07	0.10	88[a]	83[a]	√	Beef, musty, marine, cucumber[m]
Alkane	17.20	Heptadecane	0.18	0.14	94[a]	95[a]	-	Alkane[n]
	13.56	3-methyl Nonane	0.11	-	81[a]	-	-	hot milk, soap, green[n]
	17.82	Nonane-3-one	0.44	0.22	77[a]	74[a]	-	hot milk, soap, green[n]
Other	7.67	Tricholoromethane	5.55	8.45	98[a]	98[a]	-	n.d.
	14.10	Cycloheptosiloxane, tetradecamethyl-	0.45	-	72[a]	-	-	n.d.

[a] NIST 18 Library, [b] FFNSC 1.3 Library [c] peak area percentage, [d] Sinuco et al. (2004), [e] Tokimoto et al. (2005), [f] Zheng et al. (2012), [g] Umano et al. (1992), [h] Pino et al. (2013), i Burdock (2005), [t]he good scents company, [k] Berger et al. (1985), [l] Tokimoto et al. (2014), [m] Aroma descriptors, [n] Terry et al. (1984), n.d., not detected.

As per Table 2, the 'King' cultivar showed the presence of 60-65 volatile compounds consisting of esters (about 77% of total volatiles) like Methyl hexanoate (27 %) followed by methyl butyrate (18 %) and methyl 2-methyl butyrate (15 %), methyl octanoate (4 %) were predominating; followed by terpenes (about 16.85% of total volatiles) majorly being cyclosativene, α-muurolene, δ- cadinene, α- copene, caryophyllene, 1,3,5,8- undecatetraene, (E,E)-1,3,5-undecatriene, β-ocimene, etc. There was a difference observed in the concentration levels of the volatiles observed in the two different cultivars i.e. the 'King' and the 'Queen' as indicated by the percent peak area. However as far as fruit aroma is concerned the quantity and quality of volatiles and its odour threshold plays a major role for contributing to specific flavor. Several other esters like Methyl sorbate, Dimethyl propanedioate, Methyl heptacosanate Methyl 2-methyl-3-oxo butyrate, Methyl 2,4 (E,Z)-decadienoate, Methyl 7-oxo octanoate, 2-hydroxy isocaproic acid acetate, etc. were earlier not been identified and reported in pineapple and no suitable description is available in the literature about the odour. There were 9 esters and 10 terpenes present in the processed waste of both the cultivars i.e. the 'King' and 'Queen' respectively. These ester compounds include methyl butyrate, methyl 2-methyl butyrate, ethyl 2-methyl butyrate, methyl hexanoate, methyl octanoate, methyl 4- decanoate, methyl 2-hydroxy 2-methyl butanoate, Methyl 2-methyl-3-oxobutyrate, Ethyl 3-acetoxybutyrate, etc and are in accordance with the findings of Takeoka et al. (1989) who identified flavor volatiles from pineapple crown, pulp, and whole fruit by dynamic headspace sampling. However, there would always be a variation in the volatile constituents with the season, cultivar, stage of maturity (Steingass et al. 2015), handling and transportation conditions (Steingass et al. 2014), processing, etc. Wei et al. (2011) identified a total of 44 volatile compounds in pineapple pulp and 31 in the pineapple core out of which 21 compounds were common in the pulp and the core with the different esters and terpenes as the predominating ones. Certain sesquiterpene hydrocarbons like *a*-Copaene, α-patchoulene, γ-gurjunene, germacrene D, α-muurolene, and δ-cadinene are also responsible for their contribution towards fruit spicy odor (Berger et al. 1983) which have also been found in juice and leftover used in this study. Barretto et al. (2013) studied the extraction and characterization of volatile compounds from the Pineapple waste and reported that volatiles from the waste possesses mostly of esters (35%), ketones (26%), alcohol (18%), aldehydes (9%) and acids (3%). They found that using the hydrodistillation technique at inert atmosphere helped in the extraction of higher amounts of odor-active compounds like decanal, ethyl octanoate, acetic acid, 1-hexanol, γ-hexalactone, γ-octalactone, δ-octalactone, γ-decalactone, and γ-dodecalactone. The studies conducted by Tokimoto et al. (2014) identified that pineapple possesses major volatiles like Ethyl acetate, Methyl 2-methyl propanoate, Ethyl 2-methyl propanoate, 2,3, Butanedione, and other products methyl 2 and 3-methyl butanoates, ethyl butanoate, ethyl 2-methyl butanoate, Ethyl hexanoate, octanal, (z)-1,5,-octadien-3-one, 1-(E,Z)3,5,Undecatriene, 1,3,5,8, Undecatetrane, 4-methoxy-2,5-dimethyl-3-(2H) furanone, Butanoic acid, β- Damascenone, γ-Octalactone, δ-Octalactone, γ-Nonalactone, 4 hydroxy-2,5-dimethyl-3(2H)-furanone, γ-decalactone, δ-decalactone, vanillin, γ-dodecalactone responsible for their typical flavor. The leftover of the king cultivar showed the presence of two minor hydrocarbons like 1-(E,Z) 3,5,Undecatriene and 1,3,5,8- Undecatetrane which were not detected in the 'Queen' cultivar but are one of the important contributors of pineapple aroma due to their low odour threshold (Berger et al., 1985).

The aroma compounds are generally determined by odor activity values (OAV). Based on the OAV calculated previously by Zheng et al. (2012), Takeoka et al. (1989), Pino et al. (2003), it can be said that methyl 2-methyl butyrate, methyl butyrate, ethyl 2-methyl butyrate, methyl hexanoate, nonanal, decanal, etc. may be the odor-active constituents contributing to the aroma of pineapple. Based on the results of this qualitative study, it can clearly be proposed that the PPW could be utilized as a source of natural pineapple flavor. Recent studies have shown that flavoring substances like limonene possess toxicological effects in animal studies (Ravichandran et al. 2018). The USFDA in October 2018 has banned 7 different synthetic flavoring substances from the food additives list due to its carcinogenic properties. Therefore, it is always a better alternative to utilize the natural flavor extraction from the fruit by-products. Since the leftover possess a reasonable amount of natural flavor volatiles which are similar to the pineapple juice, further quantification of this volatiles needs to be carried out in future work.

CONCLUSION

The present study deals with the identification of the various volatile compounds in PPW of two commonly available cultivars in India 'Queen' and 'King' using Static headspace Gas chromatography-Mass spectrometer (SHS-GC-MS/MS) which is rapid, highly sensitive and requires minimal sample preparation. The results indicated the presence of different types of esters, terpenes, aldehydes, alcohols, alkanes, acids, lactones and other class of compounds. The presence of many of the volatiles corroborates with the studies carried out earlier and the findings therein. Majority of odor-active volatiles like methyl 2-methyl butyrate, methyl butyrate, ethyl 2-methyl butyrate, methyl hexanoate, nonanal, decanal, etc. that were detected in the pineapple juice was also present in processed waste though at lower concentration level. From the studies carried out, it is evident that the leftover of the processed pineapple has the tremendous potential to be utilized for the extraction of flavor compounds that can be utilized by the food industry.

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