Identification of the flavor profiles of Chinese pancakes from various areas using smart instruments combined with E-noses and E-tongues

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Abstract: In order to reveal the flavor characteristics of Chinese pancakes, the aroma and taste compounds of seven traditional Chinese pancakes were identified. The results showed that electronic nose (E-nose) analysis with PCA could successfully distinguish the aroma profiles of seven Chinese pancakes; the principal components PC1 and PC2 represented 75.74% and 23.2% of the total variance (98.94%) respectively. Meanwhile, the discrimination index of taste profiles of seven Chinese pancakes based on electronic tongue (E-tongue) analysis with LDA was 99.32%; the discriminant factors DF1 and DF2 represented 94.99% and 4.33% of the total variance respectively. Furthermore, GC-MS results demonstrated that thirty-three flavor compounds were identified in seven Chinese pancakes, including aldehydes, alcohols, alkanes, acids, and aromatics. Among the flavor components, aldehydes with ROAVs higher than 1 contributed most significantly to the overall aroma, such as (E,E)-2,4-nonadienal present the largest contribution in Qingzhou pancake, Jinan and Yishui pancake; hexanal present the largest contribution in Shenxian and Gaomi pancake; nonanal and benzeneacetaldehyde present the largest contribution in Linqu and Tai'an pancake, respectively. The umami and sweet taste amino acids were the most abundant in all the Chinese pancake samples, and Qingzhou pancake had relatively high amino acid content. The content of glucose was higher than maltose in Gaomi, Shenxian, and Tai'an pancakes, whereas the content of maltose was higher than glucose in Linqu, Qingzhou, Jinan, and Yishui pancakes. These results indicated that the aroma and taste profiles of Chinese pancakes differed significantly in terms of their flavor compound composition. The presented results could be beneficial for providing a comprehensive method for flavor profile identification of traditional whole-grain-based staples such as Chinese pancakes.

Keywords: Chinese pancake, flavor profile, E-nose, E-tongue, smart instrument, identification

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1 Introduction

Presently, more and more traditional Chinese food products become labeled with their geographical origins. Geographical classification is helpful for branding strategy purposes to help consumers select their food items[1]. The flavor characterization of traditional food could be an important attribute for consumers to accept and be used to guarantee the authenticity and classification of foods[2]. Normally, food flavor is a combination of aroma and taste. The aroma is characterized by volatile compounds with different olfactory characteristics in the human olfactory mucosa, while the taste is produced by non-volatile water-soluble substances stimulating taste receptors in the human taste buds[3-5]. Aldehydes, ketones, alcohols, esters, acids, hydrocarbons, sulfur, and other volatile compounds have been reported as flavor substances in foods[6,7]. The raw materials and processing technology will affect the final flavor of the product. Each grain has different contents in proteins, lipids, or small molecule carbohydrates which contain its unique flavor active compounds and flavor precursors. Small molecule carbohydrates and amino acids are the most important flavor precursors to produce volatile flavor compounds in the process of processing[8-10], which will cause changes in flavor and texture.

Since sensory assessment performed by humans inevitably presents several limitations, smart instrumental sensory analysis is an innovative and effective method to comprehensively evaluate the quality of food at present. Artificial electronic nose (E-nose) and electronic tongue (E-tongue) based on the mechanism of mammalian olfaction and taste are smart analytical instruments that can quickly and accurately characterize the overall quality and properties of food samples[11,12]. Various components of the E-nose are designed to function as mechanical analogs of the biological nose. The structural variation of odoriferous molecules is exploited for detection by sensors in the E-nose. E-tongue is capable of determining food quantitative composition and recognizing different food tastes. Moreover, the artificial sensorial assessment of analyzed food products can be easily correlated to human perception. Complex data sets from E-nose and E-tongue signals combined with multivariate statistics represent rapid and efficient tools for the classification, recognition,

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and identification of samples, also for the prediction of concentrations of different compounds[13,14]. Wang et al.[15] investigated the taste properties of rice wines of different geographical origins by electronic tongue. Shi et al.[16] investigated the flavor profiles of braised sauce spareribs with different preparations using electronic nose and gas chromatography-mass spectrometry. Zhang et al.[17] characterized volatile compounds and sensory characteristics of golden pompano fillets by GC-MS combined with an electronic tongue and electronic nose. Furthermore, there are challenges associated with the extraction, separation, and quantitation of aroma compounds due to the complexity of food and their low concentrations in products, as well as the low odor thresholds. Solid-phase micro-extraction (SPME) is suitable for low molecular weight compound isolation. Solid-phase micro-extraction (SPME) can extract low molecular weight volatile compounds, more efficient extraction of volatile flavor compounds. SPME is a new extraction technology widely used to sample volatile components with the advantages of compound extraction and sampling, fast and simple operation, low sample consumption, and good repeatability. It shows the versatility for different matrices sampling, however, the majority of detection utilizes SPME as quantitative or semi-quantative methods[18-21].

Chinese pancake (also known as Jianbing) is a traditional staple food especially popular in northern China. There are many types of Chinese pancakes nowadays. It can be made from any variety of cereals, from wheat to millet, buckwheat, and even rice. Chinese pancakes are regarded as typical whole-grain foods with high nutritional value because the raw materials mostly retain the natural aleurone layer, embryo, and endosperm[22-24]. Chinese pancakes are crispy, thin, and round, usually consumed as a staple food with meat paste, egg, sauce, and vegetables rolled inside. It is well-recognized that flavor plays a significant part in the consumer acceptance of pancakes. There are various factors influencing the flavor of Chinese pancakes, such as raw material, mixing and blending, thermal cooking process, etc., of which the thermal cooking process is the first complex factor to be considered. The cooking process can affect the pancake's aroma and taste by different mechanisms, such as Maillard reaction, lipid oxidation, protein degradation, etc.[25] Although Chinese pancake as a traditional whole-grain-based staple is very popular in China, research on its flavor profile is rarely reported.

Therefore, in the present study, the flavor profile identification of seven different Chinese pancakes consumed in China was carried out by smart instruments such as e-nose and e-tongue combined with principal component analysis (PCA) and discriminant factor analysis (DFA). Furthermore, the composition of flavor substances in Chinese pancakes was determined by SPME-GC-MS, ion chromatograph, and amino acid analyzer. The results presented in this work could be beneficial for providing a comprehensive method for flavor profile identification of traditional whole-grain-based staples such as Chinese pancakes.

2 Materials and methods

2.1 Chinese pancake samples

Seven Chinese pancake samples used for analysis were obtained from different cities of Shandong province, such as Tai'an, Gaomi, Qingzhou, Jinan, Shenxian, Linqu, and Yishui. The main ingredients of the seven traditional Chinese pancakes were as follows: Tai'an pancake: foxtail millet flour 65%, yellow maize flour 20%, soybean flour 15%; Gaomi pancake: yellow maize flour 40%, foxtail millet flour 35%, japonica rice flour 20%, soybean 5%; Qingzhou pancake: foxtail millet flour 80%, soybean flour 20%; Jinan pancake: foxtail millet flour 70%, soybean flour 30%; Shenxian pancake: foxtail millet flour 40%, yellow maize flour 35%, soybean flour 25%; Linqu pancake: foxtail millet flour 50%, yellow maize flour 40%, soybean flour 10%; Yishui pancake: foxtail millet flour 45%, yellow maize flour 35%, soybean flour 20%. All the samples were kept in vacuum packaging and stored at −25°C before analysis.

2.2 E-nose analysis

The flavor volatiles of seven different Chinese pancakes were detected using PEN3 electronic nose (Airsense, Germany) with a sensor array consisting of 10 metal oxide sensors. Static head space was generated before analysis in a 15 mL headspace vial using 1.0 g of each sample, which was equilibrated at 60°C for 20 min after sealing. At ambient temperature (25±2)°C, the carrier air gas was transferred to the sensors chamber at a flow rate of 300 mL/min. The sample was detected by sampling the headspace under sealed conditions, with a cleaning time for sensors of 180 s and a sampling time of 60 s. The E-tongue response data were analyzed using the principal component analysis (PCA) method. All measurements were obtained in triplicate.

2.3 E-tongue analysis

An Astree electronic tongue system (Alpha M.O.S, France) which was equipped with an automatic sampler unit and an array of seven chemical sensors, including AHS (sourness), NMS (umami), CTS (saltiness), ANS (sweetness), SCS (bitterness), PKS, and CPS (comprehensive taste) and one Ag/AgCl reference electrode, was applied for taste measurements of Chinese pancake samples. The sourness, saltiness, sweetness, bitterness, and umami of different Chinese pancakes were determined using average responding values of seven E-tongue sensors. Before signal collection of sensors, the E-tongue system was conditioned and calibrated in accordance with instructions. Four grams of each pancake sample was immersed in 50 mL ultrapure water to extract and equilibrate at 60°C for 20 min and then centrifuged for 10 min at 12 000×g. Then, 30 mL of filtrate was diluted with ultrapure water to 100 mL as a test fluid. The test fluid was transferred to a 100 mL beaker. Data acquisition was carried out for ultrapure water and the test fluid alternately, each test fluid was measured for 120 s, and ultrapure water was used to clean the sensors before each subsequent measurement to ensure that stable potentials were obtained. The experiment was implemented at room temperature. The E-tongue response data were analyzed using the discriminant factor analysis (DFA) method. All analysis was conducted in triplicate.

2.4 SPME-GC-MS analysis

In this study, an SPME device (50 mm DVB/ PDMS) (Supelco) was activated and then the SPME fiber was inserted in a headspace glass vial with an accurately weighed Chinese pancake sample (2 g) to extract and absorb the volatile compound at 50°C for 60 min while shaking at regular intervals (stop for 2 s after every 20 s). After the extraction, the volatile compound was desorbed within 5 min at 250°C into GC inlet with an autosampler for the subsequent GC-MS (Shimadzu, Japan) analysis. An HP-5MS capillary column (30.00 m×0.25 mm×0.25 μm) was used for separation on the capillary gas chromatography system. The flow rate of the carrier helium gas was 1.0 mL/min. Initially, the temperature of gas chromatograph oven was maintained at 35°C for 5 min, then increased to 50°C at a rate of 5.0°C/min and held for 5 min, and then increased to 220°C at a rate of 5.5°C/min and finally held for 5 min. The temperature of the mass spectrometry ion source was 220°C, and the parameter of the mass spectrometry detector was adjusted in electron impact mode with an ionizing voltage of 70 eV and a scan range from 35 to 550 m/z. Extracted volatile compounds were identified by comparing the linear retention index obtained with the mass spectrometry spectrum data library research and retaining components with a similarity of more than 85%. All analysis was conducted in triplicate.

2.5 Determination of relative odor activity value

The overall aroma of specific foods usually was determined by a few key odor compounds. While some other substances may play a supplementary role, most compounds have no significant influence on the overall aroma. In order to evaluate the contribution of each odor compound to the overall aroma, the odor activity value (OAV) was measured as the ratio of the concentration of each compound to its sensory threshold in water. Relative odor activity value (ROAV) was used to quantify the contributions of detected odor compounds to the overall aroma, and to ascertain the key odor compounds. The ROAV of the compound which has the greatest contribution to the overall aroma was taken as 100, the ROAV of other components can be calculated by the following equation:

$$\mathrm{ROAV}_i = \frac{Cr_i}{Cr_{\max}} \cdot \frac{T_{\max}}{T_i} \times 100 \quad (1)$$

where, Cr and T respectively mean relative concentration and published odor sensory threshold of the compounds; i and max refer to the detected odor compound and the compound with maximum odor activity value.

2.6 Determination of small molecule carbohydrate content

The contents of sucrose, glucose, maltose, and fructose in Chinese pancakes were determined by an ion chromatographic method. 500 mg of each example was distributed uniformly in 9 mL of pure water, equilibrated at 60°C for 20 min, and centrifuged for 10 min with 12 000×g and then filtered. The filtrate was diluted 1000 times and 5 mL solution was used for determination. The chromatographic conditions were as follows. For analyte separation, a Carbo Pac^{TM} PA20 (150.0 mm×3.0 mm) anion exchange column was used with a mobile phase of 250 mmol/L NaOH at a flow rate of 0.5 mL/min. The column temperature was initially maintained at 35°C with the 10 μL of sample injection. The total analysis time was 45 min, gradient elution conditions were carried out respectively with water and NaOH of 250 mmol/L, from 0 min to 20 min, an eluent composed of 94% of water and 6% of NaOH; from 20 min to 35 min, 45% of water and 55% of NaOH; from 35 min to 45 min, 94% of water and 6% of NaOH. Acquisition of the detection signal was conducted by a pulsed ampere detector. All analyses were done in triplicate. Comparing peak areas with the standard, the identity and concentration of the small molecule carbohydrates were determined.

2.7 Determination of amino acid content

In order to analyze the amino acid contents of Chinese pancake samples, an amino acid analyzer (L-8900, Hitachi, Japan) was used. Before determination, 100 mg of each sample was hydrolyzed using 10 mL of 6 mol/L HCl solution in the hydrolysis pipe. The acid hydrolysis was conducted at 35°C for 24 h after 1 min of nitrogen blowing to eliminate the air in the hydrolysis pipe. All samples after hydrolysis were cooled to room temperature and diluted to 50 mL with pure water, and filtered with a filter membrane. 1 mm of filtrate was evaporated by nitrogen and redissolved in 1.0 mL of 0.2 mol/L HCl solution, then passed through a 0.22 μm membrane filter. The filtrates were used for detection with an auto-sampler. Mixed standard amino acids were analyzed prior to sampling. Comparing the peak profiles of samples with standard amino acid, amino acid contents were quantified. All analysis was conducted in triplicate.

2.8 Statistical analysis

A comparison of the mean values was carried out by Tukey's test using the statistical package SPSS 17.0. Principal component analysis (PCA) and discriminant factor analysis (DFA) were performed using detection system instrument software.

3 Results and discussion

3.1 Aroma profile based on electric nose analysis

The E-nose is an innovative instrument in various areas of food safety assessment for food freshness detection, quality determination, and adulteration detection by analyzing volatile substances in foods[26]. Principal component analysis (PCA) is an effective analytical method used to reduce high-dimensional information to low-dimensional spaces built on the dissimilarities between samples. The two-dimensional PCA plot results of the electronic nose response signals of seven Chinese pancakes are shown in Figure 1a. Principal components PC1 and PC2 explained 75.74% and 23.2% of the total variance respectively. Cumulatively, these two PCs accounted for 98.94% of the total variance and clearly expressed the most information on dissimilarities between samples. As shown in Figure 1a, different colored clusters represented the data acquisition points of specific varieties of Chinese pancakes, and the distance between clusters indicates the difference between samples. The pancake samples were clustered in the positive axis of PC1 and PC2, which were correlated to the sensors of W1C (sensitive to aromatic compounds), W5S (sensitive to nitrogen oxides), W3C (sensitive to ammonia and aromatic compounds), W6S (sensitive to hydrogen), W5C (Sensitive to hydrocarbons, aromatic compounds), W1S (sensitive to methane in the environment, with broad range), W1W (sensitive to sulfur compounds), W2S (sensitive to ethanol, some aromatic compounds), W2W (sensitive to ethanol, some aromatic compounds), and W3S (sensitive to methane and some high concentration compounds). The Linqu pancake samples occupied a relatively independent space in the principal component space. Moreover, it had low values for PC1, and no overlay with other samples, indicating that it differed significantly from other Chinese pancakes in terms of its flavor compound composition. However, there were overlaps in the flavor characteristics of the pancake samples from Shenxian, Yishui, Gaomi, T'ai'an, Qingzhou, and Jinan.

The sensory spider plot of the average responding values of 10 electronic nose sensors to flavor compounds of each sample is presented in Figure 1b. The responding values of sensors W1C (S1), W5S (S2), W3C (S3), W1W (S7), and W2W (S9) exceeded one in all of the samples, indicating that aromatic, nitrogen oxides, terpene, and aromatic compound contents were higher in Chinese pancakes. Significant differences were found in the responding values of sensors S2 (W5S) and S7 (W1W), and then sensors S2 (W5S) and S7 (W1W), indicated that they contribute more than other sensors to the evaluation of flavor intensities in Chinese pancake samples.

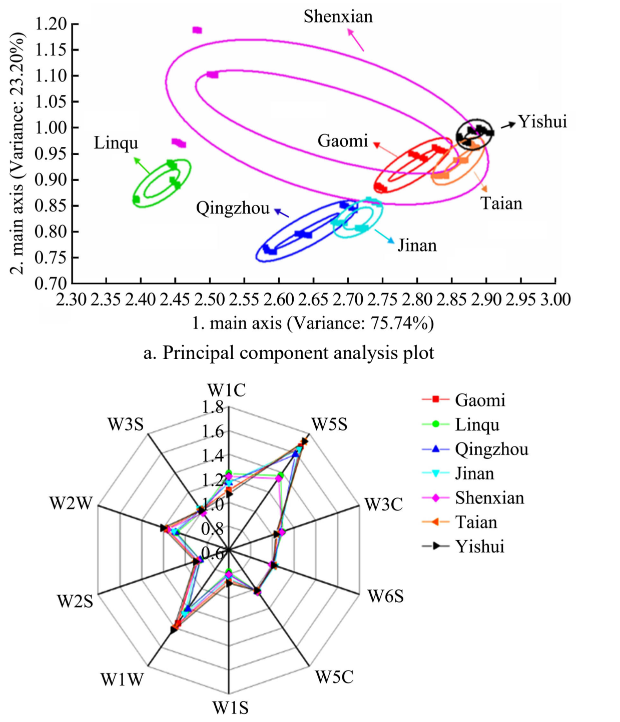


Figure 1 Principal component analysis plot and sensory spider plot based on data acquired with the electronic nose from various Chinese pancakes

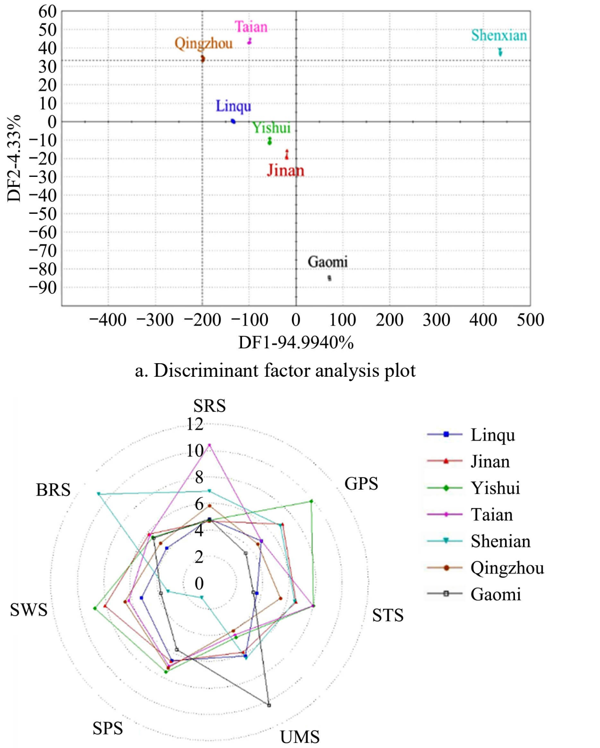


Figure 2 Discriminant factor analysis plot and sensory spider plot based on data acquired with electronic tongue from various Chinese pancakes

3.2 Taste profile based on electric tongue analysis

The scores and loadings of all pancake samples were projected onto the DF1-DF2 plane, and discriminant factors DF1 and DF2 were found to represent 94.99% and 4.33% of the total variance, respectively. Cumulatively, the cumulative contribution rate of the first and second principal components reaches 99.32% as shown in Figure 2a. As observed in the plot, the 7 Chinese pancakes were well classified by the discriminant factor analysis, and clear separation was observed for all pancake samples, with no overlap. The Gaomi and Shenxian pancakes were located on DF1 with positive scores, and could clearly be distinguished from the other samples.

The sensory spider plot of the average responding values of 7 E-tongue sensors (SRS, STS, UMS, SWS, BRS, SPS, GPS) to taste compounds of each sample are presented in Figure 2b. The responding values of sensors SRS, STS, UMS, SWS, and BRS represent sour, salty, umami, sweet, and bitter tastes, respectively. The other SPS and GPS are compound sensors, which could detect sweet and bitter tastes. As shown in Figure 2b, the differences in the responding values of E-tongue sensors were significant among the 7 Chinese pancakes, especially the responding values of BRS, SRS, UMS, and GPS sensors. It indicated that using the E-tongue cloud distinguish the taste profile of different Chinese pancake.

3.2 Composition of aroma compounds and key odorants in Chinese pancake

The volatile compounds identified in Chinese pancake samples are shown in Table 1. Overall, thirty-three flavor compounds were identified, including 6 aldehydes, 5 alcohols, 7 alkanes, 6 acids, 3 esters, 1 ketone, 3 aromatics, and 2 phenols. Flavor compounds identified in Chinese pancakes are significantly different among samples. Aldehydes were detected in all samples, but the composition of aldehydes in different pancakes was quite different with the highest relative percentage in the Qingzhou pancake. Aldehydes are generated mainly from lipid oxidation, and a small part is produced by fermentation and Maillard reaction[27,28]. Moreover, lipid oxidation can be influenced by the fatty composition and the addition of fatty in the ingredients. In baked products, lipid oxidation mainly leads to aldehydes such as (*E*)-2-octenal, hexanal, nonanal, and so on. The alcohols were identified in Qingzhou, Shenxian, and Gaomi pancake samples, whereas not found in Linqu, Tai'an, Jinan, and Yishui pancakes. Alkanes were identified in most of the samples except for Linqu and Jinan pancakes. The relative percentage of acids in Jinan pancake samples was the highest, and there was no acid found in the Gaomi pancake. Esters were only detected in Linqu and Tai'an pancakes and not found in the other pancake samples. The relative percentage of ketones in Tai'an pancake was the highest followed by Shenxian and Qingzhou pancakes. Aromatics were identified in most of the samples except for the Yishui pancake. The phenols only detected in only Qingzhou and Shenxian pancakes.

Based on the qualitative analysis of volatile compounds and the peak area ratio of volatile flavor compounds, the key flavor compounds in Chinese pancakes were determined with the relative aroma activity value (ROAV). Typically, only components with a ROAV equal to or greater than 1 contribute individually to the overall aroma and can be defined as key odorants; components with ROAVs between 0.1 and 1 can be considered to have an important influence on the overall aroma. As shown in Table 2, the ROAV among all the Chinese pancake samples was significantly different. Aldehydes with a ROAV greater than 1 were the most significant compounds contributing to the overall aroma. Aldehydes have a low odor threshold and greatly contribute to the overall flavor of the pancakes. (E,E)-2,4-Nonadienal was mainly present in Qingzhou, Jinan, and Yishui pancakes, which may originate from oxidative degradation of methyl and ethyl esters of linoleic acid in foxtail millet flour[29]. Hexanal was mainly present in Shenxian and Gaomi pancakes, which were supposed to originate as a reaction product from the oxidation of fatty acid components in yellow maize flour[30]. The nonanal contents in the Linqu pancake were higher than the other samples, which could be related to the degradation of unsaturated fatty acids in soybean flour[31]. The key odorant in Qingzhou pancake, (E,E)-2,4-nonadienal, had the highest ROAV, followed by (E,E)-2,4-decadienal and (E)-2-nonenal. The (E,E)-2,4-decadienal has the odor of fat, green, and potato[25]. In Linqu pancake, the compound with high relative odor activity values (ROAV≥1) was nonanal. In Shenxian pancake, the compound with the relative odor activity value (ROAV≥1) was hexanal, naphthalen, and 4-ethyl-2-methoxy-phenol. Compounds such as hexanal, phenylethyl alcohol, and naphthalene had a ROAV≥1 in Gaomi pancake, and thus most likely were the principal contributors to the aroma. Both aldehydes and aromatics, including benzeneacetaldehyde and naphthalene, were found to be important contributors to the characteristic aroma of Tai'an pancake. (E,E)-2,4-Nonadienal had the highest ROAV in Jinan pancake, followed by benzeneacetaldehyde and nonanal. Moreover, the key odorants of Yishui pancake were (E,E)-2,4-nonadienal and (E,E)-2,4-decadienal. Aldehydes play a positive role in overall aroma, which can make food aroma more mellow, showing greasy, nutty, and grass aroma[32-34]. Alkanes, acids, esters, and ketones compounds had a relatively low ROAV, which could be due to their high sensory thresholds.

3.5 Composition of small molecule carbohydrates and amino acids in Chinese pancake

Small molecule carbohydrates could promote Maillard reaction and produce more volatile flavor compounds. In addition, these small molecule carbohydrates also play a major role in sweet taste. Small molecule carbohydrates can also react with amino acids in the food system to produce flavor substances. The contents of glucose, sucrose, fructose, and maltose in Chinese pancake samples are shown in Figure 3. Overall, the contents of those small molecule carbohydrates in Chinese pancakes varied greatly, with glucose ranging from 0.91 to 4.21 mg/mL, sucrose ranging from 0.03 to 0.11 mg/mL, fructose ranging from 0.04 to 0.31 mg/mL, and maltose ranging from 0.59 to 5.12 mg/mL. The contents of sucrose and fructose in all pancakes were much lower than glucose and maltose. Analysis of individual small molecule carbohydrates revealed lower maltose but higher glucose levels in Gaomi, Shenxian, and Tai'an pancakes, whereas lower glucose but higher maltose levels in Linqu, Qingzhou, Jinan, and Yishui pancakes.

Table 1 Identification and peak area percentage of volatile flavor compounds in Chinese pancakes

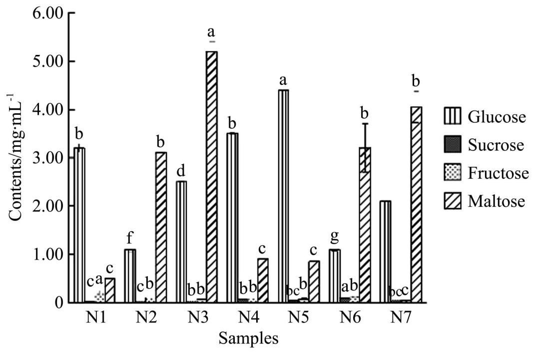
	Volatile compounds	Retention time/min	Qingzhou	Linqu	Shenxian	Gaomi	Tai'an	Jinan	Yishui
Aldehydes	Hexanal	18.657	3.66		1.63	2.01			0.49
	2,4-Nonadienal, (E,E)-	25.255	4.79					2.56	1.20
	Nonanal	29.819		3.83				6.08	
	2-Nonenal, (E)-	33.000	0.74					0.62	
	Benzeneacetaldehyde	34.386					1.02		
	(E,E)-2,4-decadienal	38.650	1.12						0.27
Alcohols	2-Nonen-1-ol	29.848	5.09						
	2,3-Butanediol, [R-(R*,R*)]-	32.150			0.94				
	2-Furanmethanol	34.000				0.39			
	Phenylethyl Alcohol	39.492				3.85			
	2-Propanol,1,1'-[(1-methyl-1,2-ethanediyl)bis(oxy)]bis-	42.907	1.69						
Alkanes	Dodecane, 2,6,11-trimethyl-	24.500				1.59			
	Nonane, 5-buty	24.516					7.72		
	Dodecane, 4-methyl-	24.567							1.77
	Undecane, 4,7-dimethyl-	24.820				3.98			
	Nonane, 2,6-dimethyl-	24.830	5.96						
	Eicosane	34.389			0.99				
	Oxime-, methoxy-phenyl-	36.059			2.46				
Acids	Acetic acid	28.976		0.61	4.32		9.11	0.57	1.10
	Hexanoic acid	37.925	4.53		2.21			2.12	
	Undecanoic acid	42.028	0.77						
	Octanoic acid	42.031						0.20	0.38
	2H-Pyran-2-one, 3-acetyl-4-hydroxy-6-methyl-	44.497		47.37				30.93	1.29
	n-Decanoic acid	45.912	0.65				1.17	0.48	
Esters	Butanedioic acid, dimethyl ester	33.088					2.09		
	Pentanedioic acid, dimethyl ester	35.474					2.54		
	1,2-Benzenedicarboxylic acid, bis (2-methylpropyl) ester	43.908		1.08					
Ketone	4H-Pyran-4-one,2,3-dihydro-3,5-dihydroxy-6-methyl-	45.354	1.03		2.69		2.98		
Aromatics	Naphthalene	37.161	0.35		0.29	2.19	0.23	0.17	
	Naphthalene, 1-methyl-	39.602			1.66				
	Azulene	37.160		1.13					
Phenols	Phenol,4-ethyl-2-methoxy-	41.823			0.96				
	2-Methoxy-4-vinylphenol	44.492	0.96						

Note: Compounds were identified by comparison with reference substances from the MS NIST library and present in at least three replicates.

Table 2 Relative odor activity values (ROAV) of volatile flavor compounds detected in Chinese pancakes

	Volatile compounds	Threshold /mg·kg^{-1}	Qingzhou	Linqu	Shenxian	Gaomi	Tai'an	Jinan	Yishui
Aldehydes	Hexanal	0.007500	0.610000		100.000000	100.000000			0.330000
	(E,E)-2,4-Nonadienal	0.000060	100.000000					100.000000	100.000000
	Nonanal	0.003500		100.000000				4.070000	
	(E)-2-Nonenal	0.000065	14.260000					22.360000	
	Benzeneacetaldehyde	0.009000					100.000000		
	(E,E)-2,4-decadienal	0.000050	28.060000						27.000000
Alcohols	2-Nonen-1-ol	--	--						
	2,3-Butanediol, [R-(R*,R*)]-	400.000000			< 0.010000				
	2-Furanmethanol	1.000000				0.150000			
	Phenylethyl Alcohol	0.045000				31.920000			
	2-Propanol,1,1'-[(1-methyl-1,2-ethanediyl)bis(oxy)]bis-	--	--						
Alkanes	Dodecane, 2,6,11-trimethyl-	--				--			
	Nonane, 5-buty	--					--		
	Dodecane, 4-methyl-	--							--
	Undecane, 4,7-dimethyl-	--				--			
	Nonane, 2,6-dimethyl-	--	--						
	Eicosane	--			--				
	Oxime-, methoxy-phenyl-	--			--				
Acids	Acetic acid	22.000000		<0.010000	0.090000		0.370000	<0.010000	<0.010000
	Hexanoic acid	80.000000	<0.010000		0.010000			<0.010000	
	Undecanoic acid	1.000000	<0.010000						
	Octanoic acid	0.800000						<0.010000	<0.010000
	2H-Pyran-2-one, 3-acetyl-4-hydroxy-6-methyl-	--		--				--	--
	n-Decanoic acid	120.000000	<0.010000				0.010000	<0.010000	
Esters	Butanedioic acid, dimethyl ester	3360.000000					<0.010000		
	Pentanedioic acid, dimethyl ester						--		
	1,2-Benzenedicarboxylic acid, bis(2-methylpropyl) ester			--					
Ketones	4H-Pyran-4-one,2,3-dihydro-3,5-dihydroxy-6-methyl-	200.000000	<0.010000		0.010000		0.010000		
Aromatics	Naphthalene	0.050000	0.010000		2.670000	16.340000	4.060000	0.010000	
	Naphthalene, 1-methyl-	1.400000			0.550000				
	Azulene			--					
Phenols	4-ethyl-2-methoxy-Phenol	0.010000			44.170000				
	2-Methoxy-4-vinylphenol	0.005000	0.240000						

Note: Compounds were identified by comparison with reference substances from the MS NIST library and present in at least three replicates.



Note: N1: Gaomi pancake; N2: Linqu pancake; N3: Qingzhou pancake; N4: Shenxian pancake; N5: Tai'an pancake; N6: Jinan pancake; N7: Yishui pancake. Data followed by different letters within a column are significantly different (p <0.05).

Figure 3 Small molecule carbohydrate contents in various Chinese pancakes

Proteolysis is associated with the formation of flavors[35]. The products of protein hydrolysis such as amino acids and peptides are very important precursors in the Maillard reaction for flavor formation[36]. Furthermore, amino acids contribute directly to the sensory perception of sweet, salty, acid, bitter, and umami. The concentrations of seventeen amino acids grouped according to their flavor characteristics were determined and are listed in Table 3. Those umami taste amino acids in the Chinese pancake samples ranged from 1.57 to 2.65 g/100 g. It was found that the amount of sweet amino acids was substantially lower, ranging from 1.65 to 2.55 g/100 g. The amount of bitter amino acids ranged from 1.09 to 1.94 g/100 g, relatively lower than both umami taste and sweet taste amino acids. In total, seventeen amino acids were quantified and recorded. Qingzhou pancake had a relatively high amino acid content, which distinguished it from the other samples.

Table 3 Amino acids contents of Chinese pancakes (g/100 g)

Amino acids		Tai'an	Gaomi	Qingzhou	Jinan	Shenxian	Linqu	Yishui
Umami taste amino acids	Asp	0.49±0.01	0.59±0.03	0.73±0.01	0.52±0.01	0.54±0.01	0.55±0.02	0.52±0.01
	Glu	1.24±0.06	1.01±0.04	1.92±0.03	1.43±0.01	1.33±0.02	1.25±0.06	1.05±0.01
	Sub-total	1.73	1.60	2.65	1.95	1.86	1.79	1.57
Sweet taste amino acids	Thr	0.28±0.00	0.27±0.00	0.37±0.00	0.30±0.00	0.31±0.00	0.31±0.02	0.28±0.01
	Ser	0.33±0.00	0.28±0.01	0.48±0.01	0.32±0.01	0.31±0.02	0.33±0.01	0.30±0.00
	Pro	0.61±0.04	0.48±0.05	0.74±0.03	0.71±0.01	0.74±0.01	0.67±0.01	0.57±0.01
	Gly	0.25±0.01	0.24±0.01	0.34±0.01	0.28±0.00	0.28±0.00	0.28±0.02	0.21±0.00
	Ala	0.50±0.03	0.39±0.01	0.63±0.01	0.50±0.01	0.54±0.00	0.5±0.02	0.40±0.00
	Sub-total	1.96	1.65	2.55	2.11	2.18	2.07	1.76
Bitter taste amino acids	Val	0.09±0.00	0.05±0.02	0.14±0.01	0.08±0.01	0.10±0.03	0.06±0.01	0.07±0.00
	Ile	0.77±0.04	0.51±0.02	0.92±0.01	0.75±0.01	0.85±0.00	0.77±0.04	0.63±0.01
	Leu	0.19±0.01	0.19±0.01	0.26±0.01	0.18±0.02	0.20±0.01	0.17±0.01	0.14±0.00
	Tyr	0.32±0.00	0.29±0.01	0.46±0.00	0.34±0.01	0.37±0.01	0.33±0.02	0.28±0.01
	Phe	0.08±0.01	0.07±0.00	0.17±0.00	0.13±0.01	0.08±0.00	0.14±0.02	0.11±0.01
	Sub-total	1.43	1.09	1.94	1.47	1.59	1.46	1.22
Others	Try	0.28±0.01	0.28±0.02	0.39±0.01	0.32±0.01	0.32±0.00	0.32±0.01	0.26±0.01
	Met	0.17±0.00	0.18±0.00	0.31±0.00	0.22±0.01	0.21±0.01	0.21±0.00	0.18±0.01
	Lys	0.19±0.01	0.15±0.02	0.24±0.00	0.20±0.01	0.22±0.01	0.20±0.01	0.17±0.01
	His	0.27±0.01	0.35±0.01	0.45±0.00	0.33±0.01	0.29±0.00	0.34±0.02	0.28±0.00
	Arg	0.03±0.01	0.03±0.01	0.03±0.01	0.03±0.01	0.07±0.01	0.04±0.01	0.04±0.01
	Sub-total	0.93	0.98	1.42	1.08	1.10	1.11	0.92
Total		6.01	5.29	8.52	6.56	6.66	6.39	5.42

4 Conclusions

In this study, the flavor profiles of seven Chinese pancakes consumed in China, including aroma compounds and taste compounds, were analyzed. All the Chinese pancake samples could be distinguished by E-nose and E-tongue with principal component analysis (PCA) and discriminant factor analysis (DFA), respectively. Overall, thirty-three flavor compounds were identified in Chinese pancakes, including aldehydes, alcohols, alkanes, acids, and aromatics. The ROAVs indicated that among the flavor components, aldehydes with a ROAV of more than 1 were the most significant contributors to the overall aroma of most Chinese pancakes. In particular, the aroma compounds that might result from lipid oxidation, such as nonanal, hexanal, (*E*)-2-nonenal, and (E,E)-2,4-decadienal significantly contributed to the overall aroma of Chinese pancakes. The contents of sucrose and fructose in all pancakes were much lower than glucose and maltose. Analysis of individual small molecule carbohydrates revealed lower maltose but higher glucose levels in Gaomi, Shenxian, and Tai'an pancakes, whereas lower glucose but higher maltose levels in Linqu, Qingzhou, Jinan, and Yishui pancakes. Of all flavor-related amino acids, those contributing to umami and sweet taste were the most abundant in all of the Chinese pancake samples. Qingzhou pancake had relatively higher amino acid content, which distinguished it from the other samples. These results indicated the aroma and taste profiles of Chinese pancakes differed significantly in terms of their flavor compound composition. Therefore, smart instruments like E-nose and E-tongue is expected to be a comprehensive method for flavor profile identification of traditional whole-grain-based staple such as Chinese pancake.

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