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Study on the Quality and Related Volatile Compounds of Beijing You- Chicken at Different Temperatures

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ABSTRACT

Background: Beijing You-chicken with a unique flavor and rich nutrition is an excellent local chicken breed in China. Clarifying the change of volatile odor during the storage of chicken is helpful to assist in judging the freshness of chicken.

Methods: The total viable count (TVC), total volatile basic nitrogen (TVB-N), lightness value L* and thiobarbituric acid reactive rubstances (TBARS) of chicken breast were measured at 4°C and 25°C, respectively. And the electronic nose and gas chromatography mass spectrometry (GC-MS) technology were used to determine the changes of volatile compounds during storage.

Result: Significant changes of the volatile compounds were observed on first day at 25°C and the 8th day at 4°C of storage, which agreed with the results evaluated by detection of physicochemical indicators. The results of this study could serve as a theoretical basis for future research on detecting changes in chicken freshness by using the electronic nose and GC-MS technology.

Key words: Chicken, Cold storage, Electronic nose, Gas chromatography mass spectrometry (GC-MS), Volatile compounds.

INTRODUCTION

As a chicken breed of both meat and egg, Beijing You-chicken has the advantages of unique appearance, delicious meat and rich in fat breed (Liu *et al.* 2001). Clarifying the change of volatile odor during the storage of chicken is helpful to assist in judging the freshness of chicken and has important guiding significance for the storage and transportation. However, there are few studies on changes of freshness and volatile compounds during storage based on electronic noses and GC-MS on fresh chicken.

This work aims to study the changes and relationship between freshness and volatile compounds of Beijing You-chicken with electronic nose technology and GC-MS technology at 4°C and 25°C. This is a preliminary exploration of evaluating the freshness of fresh chicken through electronic nose and GC-MS.

MATERIALS AND METHODS

Materials and reagents

Beijing You-chicken (weight 1567±134 g), 400 days old, all female, provided by the breeder farm of Beijing Academy of Agriculture and Forestry Sciences; Colony count test piece, produced by 3M petrifilm; Sodium chloride, boric acid, magnesium oxide, hydrochloric acid, methyl red, methine blue and ethanol are all analytically pure, purchased from Sinopharm Chemical Reagent Co., Ltd.

Instruments and equipment

KDY-9820 semi-automatic Kjeldahl nitrogen analyzer (China Tongrunyuan Company); 3k15 refrigerated centrifuge (German sigma company); FOX4000 electronic nose (France Alpha Mos company); PAL3-7890B-7000D gas chromatography-mass spectrometer (USA Agilent Corporation).

Method

Beijing You-chickens of 400-day-old with the same batch and no significant difference in quality were selected and slaughtered. A total of 38 samples were taken. The chicken breasts were taken and the fat and connective tissue were removed. All experiments, the measurements were performed every 8 hours at 25°C and every two days at 4°C with 8 times in parallel.

GB5009.228-2016 (China Standard Press, 2016) was referenced to determine TVB-N; GB4789.2-2016 (China Standards Press, 2016) was referenced to determine the TVC; the method of Wang *et al.* (2020) was referenced for the determination of TBARS; The method of proposed by Liu *et al.* (2018) was used for determining luminance value L *; sensory evaluation was referenced to the method of Xu *et al.* (2015), the method of electronic nose proposed by

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Shi *et al.* (2018) was referenced, the parameters of sensors were shown in Table 1.

Determination of GC-MS: the model of extraction head was 50/30 μm DVB/CAR/PDMS and SPME extraction needle was aged at the GC inlet for 30 min at the aging temperature of 250°C. Five grammes of thawed chicken and 1.5 mL of saturated sodium chloride solution were taken into a 20 ml extraction tube, then sealed and mixed with the thoroughly. The temperature of the heating box was set to 52°C, the heating time was 60 min, the adsorption time was 30 min and the desorption time is 5 min.

Gas chromatographic conditions: the chromatographic column of HP-5MS UI (30 m×0.250 mm, 0.25 μm) was used, the high-purity helium was used as the carrier gas and the sample was spitless. Heating program: hold at 40°C for 5 minutes and raised to 130°C with 2°C/min, without holding, then raised to 240°C with 8°C/min, hold on for 3 minutes. Mass spectrometry conditions: EI ion source, ionization energy was 70 eV, ion source temperature was 230°C, interface temperature was 230°C.

All experiments were carried out in 2021 at Information Technology Research Center, Beijing Academy of Agriculture and Forestry Sciences.

RESULTS AND DISCUSSION

Changes of TVB-N at 4°C and 25°C

As shown in Fig 1, the content of TVB-N (15.86 mg /100g) exceeded the referenced limit of 15 mg/100g (China Standard Press GB16869-2005, 2005) after 32 h at the storage of 25°C, which meant that the chicken was corrupted. The increase rate of TVB-N at 4°C was slower than that at 25°C. After 8 days of storage, the TVB-N content exceeded the limit, which was consistent with the results of Zhang *et al.* (2019).

Table 1: Response characteristics of FOX4000 E-nose sensors.

Sensors	Response
LY2 / LG	Fluoride, chlorine, nitrogen oxide, ozone
LY2 / G	Ammonia, amine, carbon monoxide, carbon monoxide
LY2 / AA	Ethanol
LY2 / GH	Ammonia, amine
LY2 / gCTL	Hydrogen sulfide
LY2 / gCT	Propane/Butane
T30 / 1	Organic solvent compound
P10 / 1	Hydrocarbon
P10 / 2	Methane
P40 / 1	Fluoride
T70 / 2	Aromatic compounds
PA / 2	Alcohol, ammonia, amine
P30 / 1	Combustible gas
P40 / 2	Chlorine
P30 / 2	Combustible gas
T40 / 2	Chlorine
T40 / 1	Fluoride
TA / 2	Alcohol, general air pollutant

Changes of TVC at 4°C and 25°C

In Fig 2, the initial value of TVC of chicken was 3.80 lg (CFU)/g. The TVC increased exponentially at 25°C and reached up to 6.12 lg (CFU)/g after 24 h, exceeding the referenced limit of 6.0 lg (CFU)/g (China Standard Press GB16869-2005, 2005). The TVC stored at 4°C increased slowly in the early stage, while increased gradually in the later stage. After 8 days of storage, the TVC was 6.43 lg (CFU)/g, which exceeded the limit. The results were similar to the research of Zhang *et al.* (2019). Compared with 25°C, the growth rate of colony at 4°C slowed down significantly, thus the shelf life was significantly prolonging.

Changes of TBARS of at 4°C and 25°C

As shown in Fig 3, at 25°C, the value of TBARS increased rapidly, indicating that the degree of oxidation increased sharply. After 32 hours the value of TBARS was 0.72 mg/

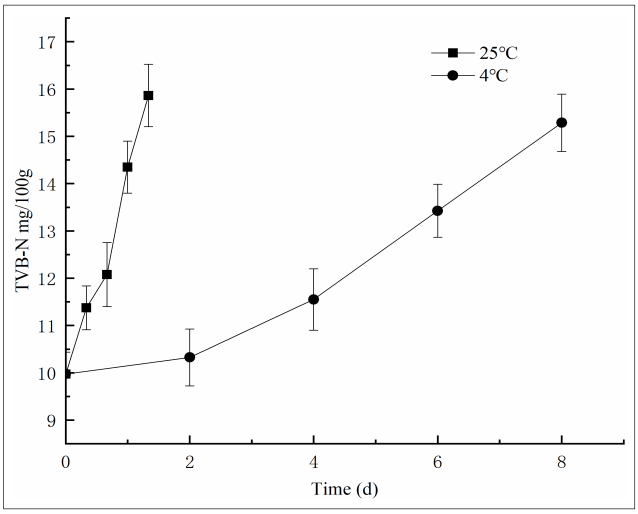

Fig 1: Changes of TVB-N content of chicken stored at 4°C and 25°C.

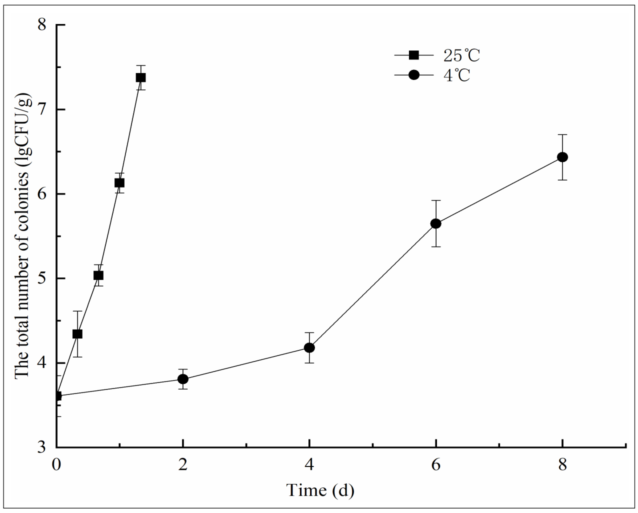

Fig 2: Changes in the total number of colonies of chicken stored at 4°C and 25°C.

100 g, which was close to the suggested limit of chicken (1.00 mg/100 g) (Zhou *et al.,* 2020). The growth rate of TBARS was relevant more slowly at 4°C than at 25°C. The value of TBARS was 0.85 mg/100g at the 8th d of storage, which was close to the limit of 1 mg/100 g.

Change of L* at 4°C and 25°C

The research has been proposed that the lightness value of chickens over 53 is light-colored appearance (Haghighi *et al.,* 2021). According to this classification, all samples in the experiment have light-colored appearances. The L* showed a trend of increasing firstly and then decreasing as shown in Fig 4. The L* reached to the maximum values of 65.1 and 67.2 at 8th hours at 25°C and at 2ed d at 4°C from the initial value of 63.8, respectively and then decreased below the initial value, which was similar to previous research (Xu *et al.* 2015).

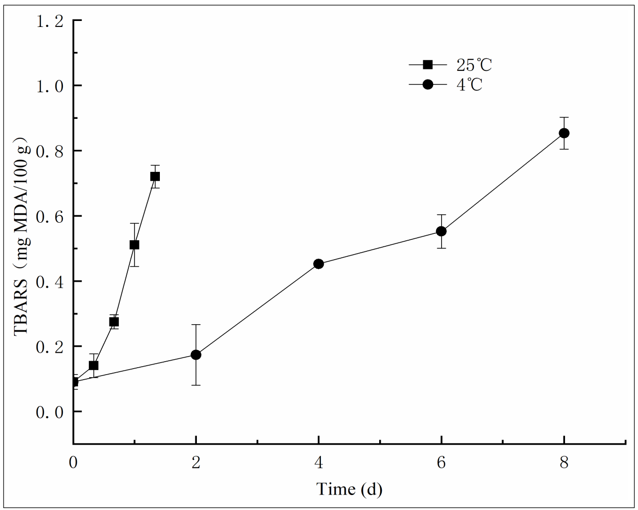

Fig 3: Changes of TBARS of chicken stored at 4°C and 25°C.

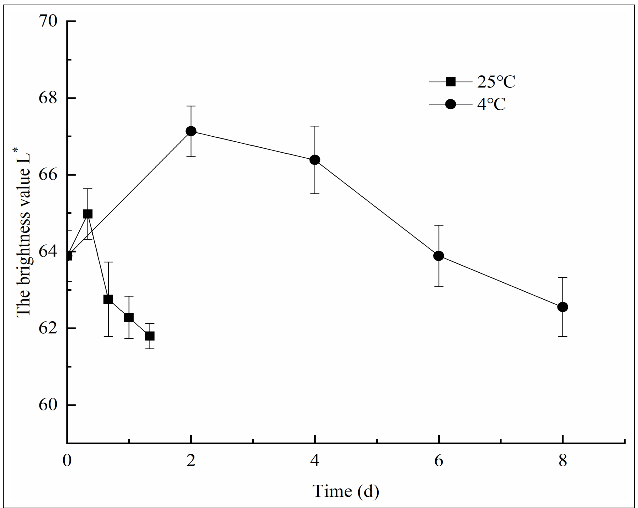

Fig 4: Changes of L* of chicken stored at 4°C and 25°C.

Sensory score of chickens stored at 4°C and 25°C

As shown in Fig 5, the sensory score was consistent with the changes in other physical and chemical indicators of chicken stored at 4°C and 25°C. The sensory score of chicken decreased more rapidly at 25°C than at 4°C. After 16 hours of storage, the color of chicken was yellowish, the indentation could not be recovered after pressing and the

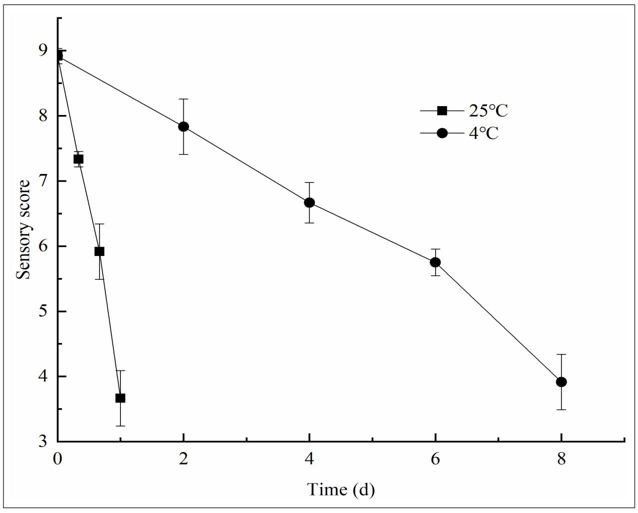

Fig 5: Sensory score changes of chicken stored at 4°C and 25°C.

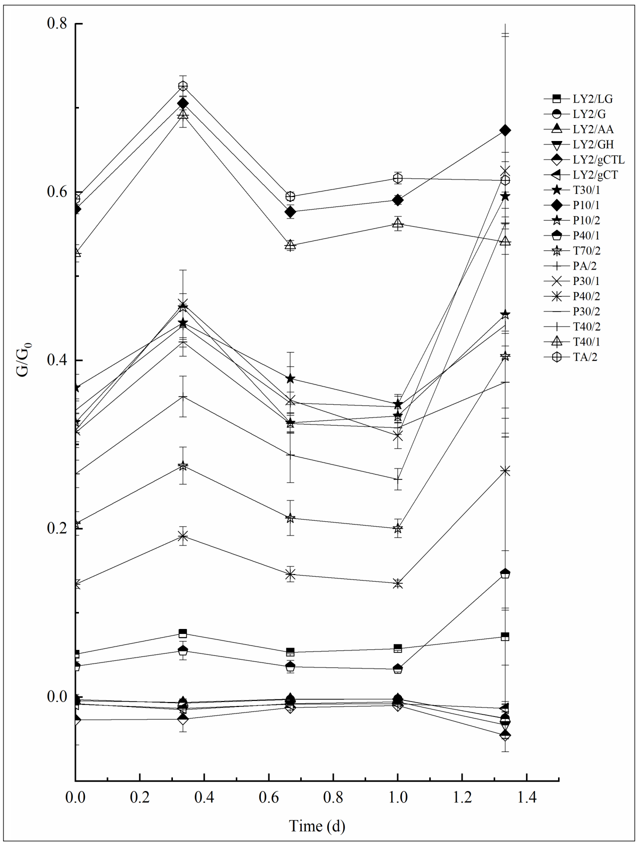

Fig 6: Electronic nose response signal of chicken stored at 25°C.

water effluent was severe. After 24 hours of storage, the chicken was dark yellow in color, indentation obviously after pressing and obvious peculiar smell accompanied by water effluent severely was observed.

Responses of electronic nose at 4°C and 25°C

As shown in Fig 6, the main volatile compounds produced of the chicken spoilage were hydrocarbons (P10/1), fluorides (T40/1) and ethanol (TA/2) at 25°C. It was proposed that the peculiar smell of chicken breasts came from reducing gases such as ethanol (Tomson *et al.*, 2016). At 25°C, it was observed that the signal of each sensor was increased in the 0-8 h, then decreased in 8-16 h. After 24 hours of storage, the aromatic compounds (T70/2), combustible gases (P30/1, P30/2), methane (P10/2), fluoride (P40/1) and other volatile compounds increased rapidly and butane (LY2/gCT), ammonia (LY2/GH, LY2/G), ethanol (TA/2) and other compounds decreased slowly. According to the changes of the sensor responses of the chicken stored at 25°C, it was found that the inflection point was appeared at the 24th h, at which the volatile compounds changed significantly. This was consistent with the shelf life of chicken evaluated by TVC. As shown in Fig 7, the main volatile compounds of chicken stored at 4°C were also hydrocarbons (P10/1), fluorides (T40/1) and ethanol (TA/2). The changes trends of electronic nose responses were during 0-6 d of storage was not obviously, which was consistent with the previous research (Tang *et al.*, 2020). On the 8th day of storage, aromatic compounds (T70/2), combustible gases (P30/1, P30/2), methane (P10/2) and fluoride (P40/1) and other volatile compounds increased rapidly, while, propane, butane (LY2/gCT), ammonia (LY2/GH, LY2/G), ethanol (TA/2) decreased slowly. It can be seen that the 6th day of storage was the turning point of the change of volatile compounds produced during the spoilage process of chicken. The volatile compounds changed drastically after 8 days of storage, which coincided with the shelf life of chickens evaluated by the TVC and TVB-N.

GC-MS detection results of chicken stored at 4°C and 25°C

As shown in Table 2, 22 compounds were detected in Beijing You-chicken stored at 4°C, including 7 aldehydes, accounting for about 40%; 7 alcohols, accounting for about 20% and 2 ketones, accounting for about 10%. Among them, the change trend of the main compounds of hexanal and 1-octen-3-ol was similar, it reached the lowest point at the 2ed d and the 6th d of storage, respectively, then gradually increased in the late storage period. The content of hexanal was higher than of threshold. As a compound with strong

Table 2: Changes of volatile compounds of chicken stored at 4°C.

Retention time (min)	Compounds	Relative content (%)				
		0d	2d	4d	6d	8d
	Alcohols					
13.846	1-octen-3-ol	17.08±1.12	13.91±1.34	16.04±0.98	9.54±0.89	10.16±1.23
5.822	1-pentanol	1.16±0.23	0.72±0.11	0.95±0.20	0.34±0.09	1.28±0.34
17.333	2-octen-1-ol	0.98±0.34	0.91±0.33	1.02±0.10	ND	ND
15.935	4-ethylcyclohexanol	0.57±0.23	ND	0.43±0.23	ND	ND
9.506	1-hexanol	ND	0.34±0.11	ND	ND	ND
13.509	1-heptanol	ND	ND	0.44±0.05	ND	ND
17.447	3-decyn-2-ol	ND	0.57±0.23	0.74±0.32	ND	ND
	Aldehydes					
17.459	cis-4-decenal	0.45±0.11	ND	ND	ND	ND
12.952	benzaldehyde	0.653±0.22	0.36±0.03	ND	ND	0.42±0.06
14.756	octanal	1.29±0.54	0.72±0.19	1.79±0.16	ND	ND
18.694	nonanal	3.48±0.26	3.96±0.11	4.81±0.37	ND	1.63±1.23
2.134	pentanal	1.05±0.21	1.44±0.12	0.91±0.33	0.50±0.20	0.75±0.15
6.803	hexanal	35.55±1.53	33.59±0.89	37.90±2.33	28.74±1.10	36.06±3.22
10.689	Heptanal	1.95±0.45	1.23±0.23	2.11±0.45	ND	0.80±0.56
	Ketones					
14.062	2,3-octanedione	9.77±0.32	10.21±0.21	11.14±0.11	10.02±0.04	7.21±2.21
10.291	2-heptanone	ND	ND	0.69±0.21	ND	ND
	Heterocyclic compounds					
14.288	Furan,2-pentyl-	1.16±0.15	1.62±0.23	1.77±0.12		0.55±0.22
9.313	p-xylene	ND	ND	0.42±0.09	0.41±0.12	1.26±1.04
10.154	styrene	ND	ND	ND	ND	0.39±0.12
3.885	toluene	ND	ND	ND	0.45±0.11	0.37±0.05
9.324	benzene	ND	ND	ND	1.07±0.11	0.43±0.21
9.51	o-xylene	ND	ND	ND	ND	0.39±0.07

chicken fat aroma, hexanal was one of the main sources of the odor contribution of chicken during the storage (Timsorn *et al.* 2016; Tang *et al.* 2020). Alcohols such as 2-octenol, hexanol, heptanol and 3-decyn-1-ol were not detected at 6-8 days, which was consistent with the results measured by the electronic nose of decreased alcohol response at the late storage period. In addition, aromatic compounds such as para-xylene, styrene, toluene, benzene, o-xylene at 8th day, which were not only agree with the results measured by the electronic nose of the increased responses of aromatic compounds in the late storage period, but also agree with the shelf life evaluated by the TVC and TVB-N.

In Table 3, 20 compounds were detected in Beijing You-chicken at 25°C, including 8 aldehydes, accounting for about 40%, 7 alcohols, accounting for about 20% and 2 ketones, accounting for about 10%. The content of main compounds of hexanal and 1-octen-3-ol have the similar trends, which reaching the lowest point at 8 h and 32 h, then reaching the highest point at the 16th h and gradually decreasing subsequently. Compared with 4°C, the change trend of volatile compounds in chickens at 25°C was more significant, which may be due to high temperature can promote the oxidation process of fat, causing the main volatile compounds to reach the highest point rapidly and then continuous declined. Hexanal accounted for the largest proportion due to it was derived from o-6 unsaturated fatty acids, which were one of the main unsaturated fatty acids in terrestrial animal tissues (Wettasinghe *et al.,* 2001).

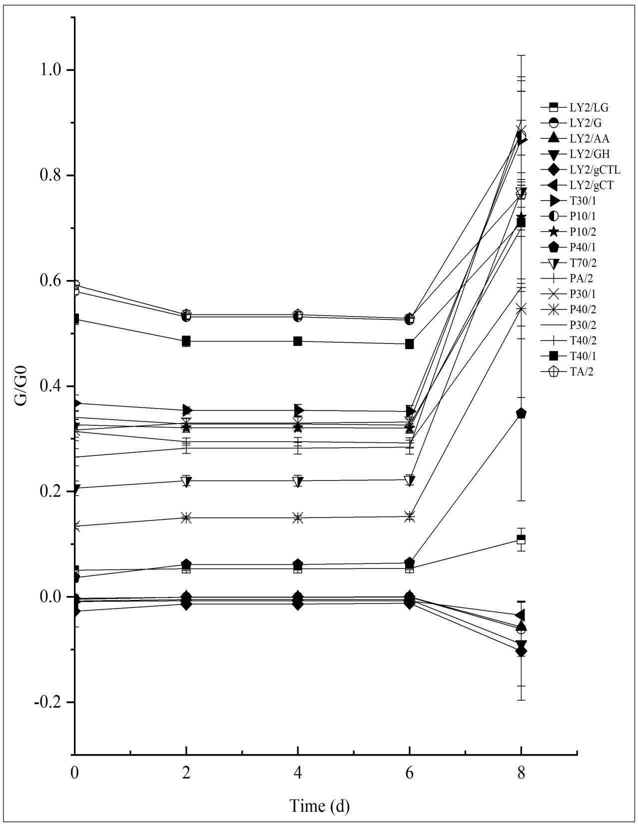

Fig 7: Electronic nose response signal of chicken stored at 4°C.

Table 3: Changes of volatile compounds of chicken stored at 25°C.

Retention time (min)	Compounds	Relative content (%)				
		0 h	8 h	16 h	24 h	32 h
	Alcohols					
13.846	1-octen-3-ol	17.08±1.23	15.63±0.89	18.23±2.27	14.17±2.01	12.03±1.06
5.822	1-pentanol	1.16±0.23	1.15±0.21	1.04±0.09	1.16±0.13	1.48±0.09
17.333	2-octen-1-ol	0.98±0.21	0	1.28±0.11	0	0
15.935	4-ethylcyclohexanol	0.57±0.11	0.69±0.02	0.73±0.05	0.50±0.07	0
9.506	1-hexanol	ND	0	0.42±0.21	0	6.34±2.19
13.509	1-heptanol	ND	0	1.66±0.36	1.48±0.22	0.82±0.12
17.326	2-octen-1-ol	ND	ND	ND	0.82±0.21	ND
	Aldehydes					
17.459	cis-4-decenal	0.45±0.04	0.50±0.02	0.51±0.03	0.39±0.07	0
12.952	benzaldehyde	0.65±0.21	0.64±0.01	0.63±0.02	0.59±0.02	0
14.756	octanal	1.29±0.23	1.89±0.34	1.24±0.29	0.72±0.17	1.50±0.41
18.694	nonanal	3.48±0.21	4.37±0.34	3.66±0.91	2.73±0.21	3.88±0.21
2.134	pentanal	1.05±0.02	0.95±0.03	0.97±0.01	0.77±0.22	0.75±0.22
6.803	hexanal	35.55±3.99	30.75±1.67	42.15±3.44	37.16±2.46	30.71±2.73
10.689	Heptanal	1.95±0.55	2.55±0.45	0	0	1.94±0.34
16.913	2-octenal	ND	0.40±0.07	ND	ND	ND
	Ketones					
14.062	2,3-octanedione	9.77±0.34	9.15±0.21	8.45±0.12	10.26±0.89	6.87±1.02
	Heterocyclic compound					
14.288	Furan,2-pentyl-	1.16±0.12	1ÿ05±0.08	1.95±0.12	1.40±014	0.45±0.21
9.313	p-xylene	ND	0	0	0.39±0.12	0
9.324	benzene	ND	ND	ND	0.56±0.11	1.02±0.36
9.51	o-xylene	ND	ND	ND	0.15±0.06	0.42±0.10

Hexanal was also the main source of the odor contribution rather than the other kinds of compounds though they were in higher content (Qi *et al.*, 2021). Alcohol compounds such as 2-octenol and 4-ethylcyclohexanol were not detected after 8 days of storage, which were consistent with the resulte measured by the electronic nose of decreased change of the alcohol signal at the late storage. Aromatic compounds such as para-xylene, benzene, o-xylene and so on were detected after storage of 24 hours, which were not only consistent with the inflection points of the enhanced response of aromatic compounds in the late storage measured by the electronic nose, but also were consistent with the shelf life evaluated by the TVC.

CONCLUSION

During the storage, the microorganisms in the chicken continuously increased, the protein was decomposed constantly and the fat was oxidized constantly, causing the continuous changes of volatile compounds and the changing regularity has a high correlation with the physiochemical indicators. In our work, the electronic nose and GC-MS technology were used to determine the changing regularity of volatile compounds during storage effectively. The hexanal, 1-octen-3-ol and 2,3-octanedione are the main odor contribution sources of chicken. At 25°C and 4°C, the alcohols including 1-octene-3-ol, 2-octen-1-ol and 4-ethylcyclohexanol decreased significantly, while aromatic compounds involved benzene, o-xylene and p-xylene increased at the 24th h and 8th d, respectively. In addition, the changes of alcohols and aromatic compounds were consistent with the shelf life evaluated by the TVC and TVB-N. This research provided basis for freshness detection of the chicken by GC-MS and electronic nose.

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Conflicts of interest

The authors declare no conflict of interest.

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