Effects of Feeding Ice Fish and Feed on the Flavor of Chinese Crab

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Abstract— *Eriocheir sinensis is an important aquaculture animal in China. In order to compare the effects of feeding chilled fish and feed on the flavor of Chinese mitten crab, this experiment compared the volatile flavor substances, sensory evaluation and the differences of amino acids (AA), fatty acids (FA) and nucleotides. As a result, the sweet taste, fresh taste and grass flavor of Chinese chelate crab in the feed group were significantly higher than those in the ice fish group (P<0.05). The fishy smell of the feed group was significantly lower than that of the ice fish group (P<0.05). Amino acids in feed group and chilled fish group were not significantly different. Only 5'-adenosyl monophosphate (AMP) was found to be significantly different between the two groups (P<0.05), and the AMP content in feed group was significantly higher than that in ice fish group. The fatty acid composition of feed group and chilled fish group varied greatly. compared with the chilled fish group, saturated fatty acids (SFAs) and monounsaturated fatty acids (MUFAs) in feed group decreased significantly (P<0.05), while high unsaturated fatty acids (PUFAs) increased significantly (P<0.05). Gas chromatography-mass spectrometry (GC-MS) was used to study volatile small molecules in muscle difference, compared with the ice fish group, the content of aldehydes in the feed group increased significantly (P<0.05), and the content of ketones and nitrogen compounds decreased significantly (P<0.05). The enzyme (lipoxygenase) that catalyzes the formation of aldehydes from polyunsaturated fatty acids was further analyzed. Compared with the ice fish group, the expression of LOX 5 genes and proteins and LOX enzyme activity in the feed group were significantly increased (P<0.05).*

Keywords— Chinese mitten crab, Eriocheir sinensis, amino acid, fatty acid, flavor.

I. INTRODUCTION

Chinese mitten crab *(Eriocheir sinensis)* is favored by consumers because of its rich nutritional value and unique flavor, and is one of the important economically farmed crabs in China [1,2]. In 2015, the total output of river crabs in China reached about 823,000 tons, most of which came from pond culture [3]. For a long time, traditional feeds such as chilled miscellaneous fish, corn, wheat, soybeans and cakes have been mainly used for breeding river crabs in ponds. However, these traditional feeds have some shortcomings such as unstable sources, unbalanced nutrient composition, low feed utilization rate and easy to cause water quality deterioration, which easily lead to the disease of river crabs and unstable quality of adult crabs [4-6]. Traditional breeding mode has become one of the important factors restricting the sustainable development of Chinese crab breeding industry [7,8]. Although the artificial compound feed has been gradually applied to the production of river crabs with the continuous improvement of the culture concept and technical level, in the late stage of culture, especially after the reproductive molting of river crabs, farmers still generally use a large number of Iced fish for fattening [9]. At present, there are two kinds of fattening feed on the market: puffed feed has good stability in water, less anti-nutritional factors in raw materials, which is beneficial to digestion and absorption of aquatic animals, and has the advantages of high utilization rate of raw materials, less pollution, safety and hygiene, etc. The processing of hard pellet feed has low requirements on equipment and relatively low manufacturing cost, and can also avoid oxidation loss of vitamins and fatty acids caused by puffing processing technology [10]. At present, Chinese feed manufacturers mostly adopt the processing technology of hard pellet feed. There are few reports on the effect of feed on the flavor of river crabs. In this experiment, hard pellet feed and Iced fish were used to feed river crabs, and their effects on the flavor of river crabs were compared to provide scientific basis and practical reference for the cultivation and quality control of river crabs.

II. MATERIALS AND METHODS

2.1 Experimental design and sample collection

The *Eriocheir sinensis* purchased from a local farm in Pukou District, Nanjing. During the experiment, the crabs were fed with ice fish (purchased locally) or artificial compound feed (Jiangsu Haipurui Feed Co., Ltd.) every day. The main nutritional components are shown in Table 1, and the experimental crabs were given two weeks to adapt to the experimental conditions. 80 crabs (25.33±0.79g) were evenly distributed to 8 cement ponds (1.5×1.5×0.5m, length: width: height). These crabs were divided into two groups, with four repetitions in each group: one group was fed with ice fish, and the other group was fed with compound feed for 12 weeks. During the experiment, the water temperature was controlled at 24±2^{o}C, pH 8.5-8.6, and dissolved oxygen was more than 5 mg/L.

TABLE 1
COMPARISON OF NUTRITIONAL COMPONENTS BETWEEN COMPOUND FEED AND ICED FISH

Projects	Compound Feed	Iced Fish
Protein	42.89±0.12^{b}	64.55±0.72^{a}
Total fat	8.09±0.56^{b}	13.98±1.52^{a}
Crude ash	12.96±0.04^{b}	14.98±0.45^{a}
Moisture	9.35±0.18^{b}	77.51±0.82^{a}

After the experimental breeding stage, the crab was put on ice for 10 minutes to reduce its vitality. Each crab was weighed separately, the length and width of the shell were measured, and then killed. The muscles of single crab were dissected on ice, washed thoroughly with 0.89g/L NaCl, frozen in liquid nitrogen immediately after treatment and stored at -80^{o}C for subsequent analysis.

2.2 Sensory analysis

We invited 30 ordinary diners to do sensory analysis. Diners should not eat, smoke or drink within 1 hour before the evaluation. The *Eriocheir sinensis* used for sensory analysis was washed and steamed for 20 minutes without adding any spices or flavoring agents. In order to prevent crabs of different genders from interfering with the taste, the female crab and the male crab are separated during cooking. The cooked crab is divided into the following parts: breastplate, leg, body muscle and hepatopancreas, which are placed in numbered dishes for diners to evaluate. After tasting a sample, the participating diners need to gargle with purified water before evaluating the next sample.

2.3 Analysis of free amino acids

Use 3 to 5mL of 80% ethanol per 0.5g of dry tissue. Then the tissue was homogenized in an ice bath with a ceramic homogenizer for 5 minutes. The homogenate was centrifuged at 3000r/min for 15 minutes, and then the clear supernatant was centrifuged again, which was repeated 3 times. After taking supernatant and vacuum drying to remove ethanol, the residue was dissolved in 8 ml of 6 mol/L HCl and placed in a 40mL hydrolysis tube. Then, the hydrolysis tube was vacuumized and filled with nitrogen at 110^{o}C for 24 hours. After hydrolysis, 1mL of hydrolysate was taken out and dried by vacuum evaporation at 50^{o}C to remove HCl. The hydrolysate was dissolved in 5ml of 0.02m HCl, and 50μL of supernatant was used for amino acid analysis by Biochrom 30 automatic amino acid analyzer (Cambridge, United Kingdom). Set the detection wavelength to UV 570 and 440nm (for Pro). All analyses were performed in triplicate. The characteristics and quantity of amino acids were determined by comparing with the retention time and peak area of each amino acid standard.

2.4 Fatty acid composition analysis

The fatty acid was analyzed by Morrison and Smith(1964) [11], which was methyl esterified with 14% boron trifluoride (BF3) methanol solution to produce fatty acid methyl esters, FAME). The fatty acid composition of fat source, liver and muscle was determined by gas chromatography [12]. The gas chromatograph model used is agilent 6890 (agilent technologies, Santa Clara, ca, USA), and the column model is Omegawax 320(30m×0.32mm; Supelco, Billefonte, PA, USA).

2.5 Statistical analysis

The experimental data are expressed by (mean standard error). After considering the normality of distribution and homogeneity of variance, SPSS 19.0 software is used to analyze all the data by independent sample T-test. Let the significance level be 0.05.

III. RESULTS AND ANALYSIS

3.1 Sensory evaluation

Sensory evaluation is a traditional experimental method often used to describe the smell and taste of crab muscles [13]. Each sample was evaluated in this test, and the results are shown in Figure 1. It can be found that the scores of bitter taste and salty taste of ice fish group and feed group are similar, while the scores of sweet taste and savory taste of feed group are higher than those of ice fish group. Ice fish group and feed group showed similar scores in meat flavor and fat odor, while compared with ice fish group, feed group could experience higher grass flavor and lower fishy smell.

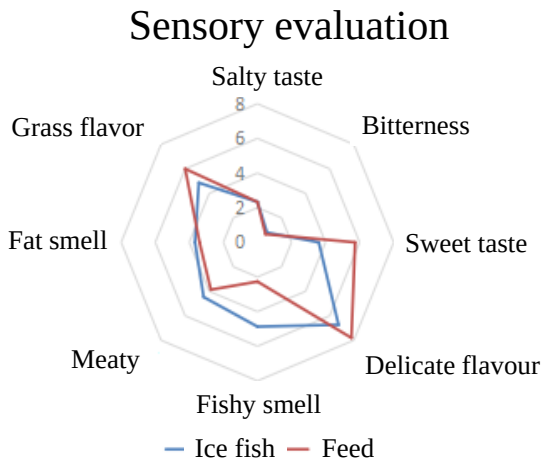

FIGURE 1: Radar image of sensory evaluation of *Eriocheir sinensis*

3.2 Comparison of fatty acid composition

The FAs composition in muscle of *Eriocheir sinensis* fed with compound feed and Iced fish for 8 months is shown in Table 2. In order to further understand the changes of FAs composition in crab muscle, we also detected FAs composition in Iced fish and feed, and the results are shown in Table 3.

According to the analysis of fatty acid saturation, compared with the ice fish group, the muscle of crab fed with feed group contains lower proportion of SFA and MUFA (Table 3), while the proportion of PUFA in feed group increases significantly ($P<0.05$). In this study, there was no significant difference in EPA content between ice fish group and feed group [14]. Compared with the ice fish group, the content of ALA and AA diets increased significantly ($P<0.05$), while the content of DHA decreased significantly ($P<0.05$). As shown in Table 1, the content of PUFA in compound feed (51%) is 14 percentage points higher than that of Iced fish (37%).

In terms of FAs composition, the feed group showed higher PUFA and lower SFA and MUFA than the ice fish group.

TABLE 2
FATTY ACID COMPOSITION OF ICED FISH AND COMPOUND FEED AND FATTY ACID COMPOSITION% OF *ERIOCHEIR SINENSIS* AFTER 8 MONTHS

Fatty acid	Iced fish	Compound Feed	Eight months(Iced fish)	Eight months(compound feed)
C12:0	0.110	0.044	0.0647±0.005	0.053±0.007
C14:0	5.602	1.449	1.171±0.117	0.542±0.068*
C15:0	0.728	0.159	0.375±0.017	0.257±0.010*
C16:0	24.804	16.238	16.741±0.271	15.320±0.183*
C17:0	0.695	0.238	0.759±0.044	0.619±0.023*
C18:0	4.791	4.326	8.320±0.371	8.619±0.224
C20:0	0.533	0.371	0.190±0.016	0.114±0.005*
C22:0	0.143	0.394	0.072±0.006	0.088±0.013
C16:1	5.816	1.936	3.435±0.499	2.790±0.452
C18:1	11.080	22.506	21.980±0.499	22.502±0.363
C20:1	3.385	0.724	1.925±0.17	0.817±0.049*
C22:1	4.785	0.448	0.702±0.088	0.169±0.008*
C18:2	1.028	40.095	6.804±1.074	13.631±0.491*
C20:2	0.163	0.092	1.417±0.033	2.018±0.095*
C18:3n6	0.130	0.061	0.083±0.003	0.090±0.010
C18:3n3	0.463	4.912	0.580±0.178	0.991±0.099
C20:3	0.040	0.041	0.032±0.005	0.042±0.005
C22:3	0.141	0.051	0.885±0.083	0.700±0.073
C20:4	1.255	0.381	3.249±0.364	3.916±0.103
C22:4	0.477	0.084	0.210±0.029	0.243±0.011
C20:5	10.658	2.410	14.329±0.565	12.988±0.673
C22:5	1.012	0.299	0.435±0.037	0.530±0.011*
C22:6	22.167	2.746	16.24±0.696	12.963±0.703*
SFA	43.220	25.153	27.691±0.330	25.613±0.073*
MUFA	19.249	23.677	28.042±0.181	26.282±0.731*
PUFA	37.533	51.170	44.277±0.273	48.118±0.733*

Note: * It shows significant difference between ice fish group and feed group (P<0.05).
SFA: saturated fatty acid; MUFA: monounsaturated fatty acid; PUFA: Polyunsaturated Fatty Acids

TABLE 3
FLAVOR NUCLEOTIDES AND EQUIVALENT FLAVOR CONCENTRATION EUC (n=3) IN CHINESE MITTEN CRAB MEAT AFTER FEEDING ICED FISH AND FEED FOR 8 MONTHS

Tasty nucleotide	Iced fish	Compound Feed
GMP (mg/100g)	41.62±2.05	36.98±1.65
IMP (mg/100g)	394.95±46.69	530.19±62.55
AMP (mg/100g)	253.56±61.28	617.84±81.41*
EUC (g MSG/100g)	163.8±0.95	215.9±1.20*

Note: "*" shows significant difference between ice fish group and feed group (P<0.05).
Imp: 5'-inosine monophosphate; Gmp: 5'-guanosine monophosphate; AMP: 5'-adenosine monophosphate; Ump: 5'-uridine monophosphate.

3.3 Comparison of free amino acids and nucleotides

We detected 17 kinds of amino acids in crabs, and the results are shown in Table 2. There is no significant difference in the content of 17 AAs between ice fish group and feed group (Table 4). Besides AA, we also detected three nucleotides (AMP, IMP, GMP), and the results are shown in Table 3 [15]. AMP concentration in feed group was significantly higher than that in ice fish group (P<0.05), but there was no significant difference in IMP and GMP concentration between the two groups.

TABLE 4
AMINO ACID COMPOSITION OF *ERIOCHEIR SINENSIS* AFTER FEEDING ICE FISH AND FEED FOR 8 MONTHS

Amino acid	Taste	Iced fish	Compound Feed
Asp	Fresh	1.654±0.123	1.501±0.077
Glu	Fresh	2.498±0.121	2.304±0.115
Ser	Sweet	0.704±0.036	0.653±0.032
Thr	Sweet	0.774±0.057	0.677±0.032
Gly	Sweet	0.935±0.097	0.907±0.054
Ala	Sweet	1.089±0.021	1.066±0.044
Cys	Sweet	0.178±0.009	0.169±0.006
Val	Bitter	0.776±0.058	0.707±0.034
Met	Bitter	0.597±0.075	0.532±0.059
Ile	Bitter	0.722±0.067	0.683±0.055
Leu	N.A.	1.248±0.081	1.161±0.068
Tyr	N.A.	0.731±0.069	0.67±0.028
Phe	N.A.	0.828±0.065	0.736±0.054
Lys	N.A.	1.198±0.063	1.046±0.08
His	N.A.	0.471±0.057	0.368±0.029
Arg	N.A.	1.761±0.1	1.589±0.107
Pro	N.A.	0.623±0.066	0.606±0.024
Total		16.788±0.9	15.376±0.836

Note: N.A.: No.

3.4 Comparison of volatile compounds

The volatile compounds in meat were detected by SPME-GC-MS, and the results are shown in table 5. A total of 55 volatile compounds (2549.32±12ug/kg in ice fish group; 2392.62±4.85ug/kg in feed group, including 16 aldehydes (878.98±5.75ug/kg in ice fish group; The feed group was 962.26±8.52μg/kg/kg) and 6 kinds of ketones (the ice fish group was 104.11 4.72 ug/kg; 72.44±3.77ug/kg in the feed group) and 6 kinds of alcohols (149.31±6.12ug/kg in the ice fish group; 163.47 3.2 ug/kg in the feed group) and 6 aromatic hydrocarbons (119.45 1.05 ug/kg in the ice fish group; 120.61±1.97ug/kg in the feed group), 5 compounds containing n (599.65±18.03ug/kg in the ice fish group; The feed group is 371.26±16.17ug/kg), and 10 kinds of hydrocarbons (the ice fish group is 587.4±11.17ug/kg; The feed group is 592.13±0.3ug/kg) and the other 6 species (ice fish group is 110.41±3.85ug/kg; The feed group was 110.45±4.95ug/kg). Compared with the ice fish group, the contents of total volatile compounds and aldehydes in the feed group increased significantly ($P<0.05$), while the contents of ketones and nitrogen compounds decreased significantly ($P<0.05$). Odor activity value (OAV) is an index to evaluate whether each volatile compound reaches the taste concentration, which can be calculated by dividing the concentration of the compound by the odor threshold of the compound, and if the value is greater than 1, the compound presents taste [16]. Nineteen AAC were selected from all 55 volatile compounds in each group, including 14 aldehydes, 1 alcohol, 1 N-containing compound, 1 hydrocarbon and 2 other compounds.

TABLE 5
CONCENTRATION OF VOLATILE COMPOUNDS IN *ERIOCHEIR SINENSIS* AFTER FEEDING ICE FISH AND FEED FOR 8 MONTHS

Compound	LRI	Threshold (ng/g)	Authenticate	Iced fish	Compound feed	ACCs
Aldehydes						
2-Methylbutanal	665	1	MS, RI	7.41±1.45	7.28±1.17	Y
Pentanal	699	9	MS, RI	31.07±2	27.27±1.24	Y
2-Methyl-2-butenal	740	458.9	MS	52.7±2.04	63.48±1.62*	N
Hexanal	800	2.8	MS, RI	14.52±0.58	67.02±4.95**	Y
4-Heptenal	901	4.2	MS, RI	4.23±0.51	7.02±0.37*	Y
Heptanal	903	2.8	MS, RI	25.02±0.56	18.62±1.13*	Y
Benzaldehyde	971	41.7	MS, RI	222.08±1.47	229.53±2.83*	Y
Octanal	1002	0.587	MS, RI	154.74±2.69	179.69±4.36*	Y
2,4-Heptadienal	1019	15.4	MS, RI	19.44±1.05	17.07±1.72	Y

Benzeneacetaldehyde	1030	4	MS	22.25±1.09	21.5±0.5	Y
Nonanal	1102	1.1	MS, RI	137.05±5.49	153.38±6.09	Y
2, 6-Nonadienal	1116	0.15	MS, RI	14.84±1.55	21.59±0.9*	Y
Decanal	1220	0.1	MS	114.4±7.1	94.41±2.73	Y
Undecanal	1315	5	MS, RI	15.65±1.42	10.76±0.93*	Y
2,4-Decadienal	1329	0.07	MS, RI	24.95±0.74	23.46±1.3	Y
Hexadecanal	1820	N.A.	MS, RI	18.62±0.67	20.17±0.45	N.J.
Subtotal (16)				878.98±5.75	962.26±8.52**	
Ketones						
Acetone	<500	14500	MS	34.11±1.24	23.74±1.5*	N
2-Butanone	589	35400	MS, RI	8.49±0.74	5.86±1.28	N
2-Octanone	994	50.2	MS, RI	36.39±2.91	21.14±1.12*	N
2-Nonanone	1091	38.9	MS, RI	17.3±0.69	13.88±0.57*	N
3,5-Octadien-2-one	1102	150	MS	4.87±0.2	4.85±0.67	N
2-Decanone	1190	7.94	MS, RI	2.95±0.15	2.97±0.97	N
Subtotal (6)				104.11±4.72	72.44±3.77*	
Alcohols						
1-Penten-3-ol	682	358.1	MS, RI	22.76±1.7	22.46±0.7	N
1-Pentanol	675	150.2	MS, RI	1.71±0.64	1.75±0.19	N
1-Octen-3-ol	978	1.5	MS, RI	86.86±2.93	101.65±1.74*	Y
1-Heptanol	981	N.A.	MS	4.52±0.8	4.93±0.5	N.J.
2,4-Undecadienol	1071	N.A.	MS, RI	4.69±0.47	5.86±0.66	N.J
Cedrol	1792	N.A.	MS	28.78±3.77	26.82±1.54	N.J
Subtotal (6)				149.31±6.12	163.47±3.2	
N-containing compounds						
Trimethylamine	<500	2.4	MS, RI	540.33±18.34	320.4±15.69*	Y
Pyridine	775	2100	MS, RI	13.08±0.81	8.83±0.28*	N
2-Ethylpyridine	909	57	MS, RI	23.31±0.89	21.42±0.49	N
2,5-Dimethylpyrazine	916	1700	MS, RI	2.91±0.65	2.41±0.52	N
2,3,5-Trimethylpyrazine	1005	350.12	MS, RI	20.02±1.09	18.2±0.62	N
Subtotal (5)				599.65±18.03	371.26±16.17*	
Aromatics						
Benzene	668	1500	MS, RI	72.85±1.17	74.49±2.56	N
Toluene	770	1550	MS, RI	19.78±0.99	15.8±1.27	N
Ethylbenzene	865	2205.25	MS, RI	8.56±0.69	8.9±0.98	N
P-Xylene	873	450.23	MS, RI	10.66±0.85	11.28±1.13	N
Xylene	879	N.A.	MS, RI	5.25±0.58	7.62±0.8	N.J
Naphthalene	1215	60	MS, RI	2.34±0.23	2.51±0.07	N
Subtotal (6)				119.45±1.05	120.61±1.97	
Hydrocarbons						
2,4-Dimethyl-heptane	840	N.A.	MS, RI	21.72±0.87	21.41±1.13	N.J
Limonene	1038	10	MS, RI	117.4±3.77	117.25±4.28	Y
Undecane	1102	1170	MS, RI	12.43±0.3	14.44±0.46*	N
Dodecane	1201	2040	MS, RI	22.42±0.48	20.7±0.1*	N
Tridecane	1297	2140	MS, RI	5.96±0.64	5.1±0.63	N
Tetradecane	1398	N.A.	MS, RI	1.72±0.1	1.9±0.07	N.J.
Pentadecane	1500	N.A.	MS, RI	25.35±2.03	23.35±2.03	N.J
Hexadecane	1602	N.A.	MS, RI	35.95±2.6	34.34±3.39	N.J

2,6,10,14-Tetramethylpentadecane	1702	N.A.	MS, RI	4.03±0.71	6.73±0.3	N.J
Eicosane	1999	N.A.	MS, RI	340.41±13.01	346.91±3.56	N.J
Subtotal (10)				587.4±11.17	592.13±0.3	
Other				0±0	0±0	
2-Acetylthiazole	1205	10	MS, RI	3.2±0.3	3.87±0.24	N
2-Ethylfuran	700	2.3	MS, RI	5.85±0.27	5.05±0.41	Y
2-Pentylfuran	992	5.8	MS, RI	57.55±3.26	56.68±2.82	Y
Iodomethane	586	N.A.	MS, RI	24.81±0.44	23.99±1.14	N
Phthalic acid, butyl tetradecyl ester	1570	N.A.	MS	13.85±0.77	15.41±1.1	N.J
Hexadecanoic acid, methyl ester	1917	N.A.	MS	5.14±0.42	5.44±0.71	N.J
Subtotal (6)				110.41±3.85	110.45±4.95	
Total				2549.32±12	2392.62±4.85*	

Note: *P<0.05, **P<0.01.

IV. DISCUSSION

4.1 Sensory changes

After sensory evaluation, it can be clearly found that the bitter taste and salty taste of crabs in ice fish group and feed group are almost unchanged, while the delicious taste and sweet taste of Chinese mitten crab fed with feed are more prominent. Shi Jing et al. reported similar results, that is, Chinese mitten crab fed with compound feed had a higher taste and sweetness score than that fed with ice fish. The scores of meat smell and fat smell of the two groups were similar, while the experimental members tasted lower fishy smell and higher grass smell in the crabs of the feed group.

4.2 Fatty acid changes

The fatty acids of Chinese mitten crab under two feeding modes have great changes. Compared with the ice fish group, the muscle of Chinese mitten crab fed with feed contains a lower proportion of SFA ($P<0.05$). It may be that SFA in compound feed is lower than that in Iced fish. Many studies in recent years have confirmed an undisputed view that foods rich in SFA will have negative effects on human health [17]. Therefore, from the perspective of healthy diet, artificial compound feed can improve the nutritional value of *Eriocheir sinensis*. Confusingly, the proportion of MUFA in compound feed is higher than that of Iced fish, while the proportion of MUFA in feed group is lower than that of Iced fish group. Further analysis shows that the higher proportion of MUFAs in compound feed is mainly due to the fact that the proportion of oleic acid in compound feed (22%) is twice that of Iced fish (11%). Oleic acid (OA, C18: 1) can be converted into linoleic acid (LA, C18: 2) catalyzed by desaturase. The LA content in the feed group was twice as high as that in the ice fish group (6.80%), which indicated that Chinese mitten crab was more inclined to deposit OA-transformed LA in muscle in the form of PUFA instead of MUFA, which was the main reason why Chinese mitten crab was rich in polyunsaturated fatty acids. The impact of MUFA on human health is still controversial, and there is no unified view at present. However, in clinical investigation, it is found that the proportion of MUFAs is better than 30, otherwise it will induce cardiovascular diseases. As we all know, PUFA has many benefits to human health, including preventing chronic diseases and promoting brain development. Compared with ice fish group, the proportion of PUFA in feed group increased significantly ($P<0.05$). This may be related to the difference of PUFA content between oleic acid synthesis and diet. As shown in Table 3, the content of PUFA in compound feed (51%) is 14 percentage points higher than that of Iced fish (37%). Among various polyunsaturated fatty acids, there are four main types of fatty acids, which are very important for maintaining cell morphology and preventing chronic diseases [18]. (ν)-linolenic acid (ALA, 18: 3n-3) is the precursor of n-3- family, eicosapentaenoic acid (EPA, 20: 5n-3) or docosahexaenoic acid (DHA, 22: 6n-3), while linoleic acid (LA, 18: 2n-6) is arachidonic acid AA and DHA are the main components of membrane phospholipids, and the phospholipids of central nervous system membrane are mainly long-chain PUFA. In this study, there was no significant difference in EPA content between ice fish group and feed group. Compared with the ice fish group, the content of ALA and AA diets increased significantly ($P<0.05$), while DHA content decreased significantly ($P<0.05$).

4.3 Changes of free amino acids and nucleotides

It is reported that the content of amino acids (AAs) has a strong correlation with the taste of crabs. In particular, some flavor amino acids (glutamic acid, aspartic acid, glycine, serine, alanine and proline) contribute greatly to the sweetness and umami taste of crabs. There was no significant difference in the content of 17 AAs between ice fish group and feed group. The AA composition of aquatic animals is closely related to the protein sources in the diet [19]. At present, fishmeal is the main and necessary protein source in the compound feed of *Eriocheir sinensis*. Fish meal is mainly processed from Iced fish, so it has similar AA composition to Iced fish. In this study, the consistent AA composition between the ice fish group and the feed group may be attributed to fish meal being selected as the main protein source for preparing feed. As far as amino acids are concerned, the compound feed developed by our laboratory has no significant effect on muscle taste.

Besides AA, disodium salt of 5'- nucleotide has great influence on the taste of Chinese mitten crab. Among the three nucleotides, AMP is considered to have the greatest influence on the flavor of crab muscle, because AMP is detected as the highest content in all tissues of *Eriocheir sinensis*. From the results of three disodium salts of 5'- nucleotides (AMP, IMP and GMP), it can be seen that AMP concentration in feed group is significantly higher than that in ice fish group ($P<0.05$), but there is no significant difference in IMP and GMP concentration between the two groups. The significant increase of AMP content may be related to the higher PUFA content in compound feed. Many studies have confirmed that PUFA can activate AMPK, which indicates that AMP content increases [20]. Nucleotides have a unique synergistic effect between AA to synergistically increase the flavor. Monosodium glutamate equivalent (EDC) is the flavor intensity given by the mixture of amino acids and 5'- nucleotides, which is considered as a very useful tool to evaluate the flavor of food. As shown in Table 3. Compared with the ice fish group, the EUC of the feed group increased significantly ($P<0.05$), which was consistent with the analysis of sensory evaluation of higher umami score in the feed group. From the point of view of free amino acids and nucleotides, the compound feed used in our laboratory improved the delicious taste of Chinese mitten crab, which was mainly due to the increase of AMP content [21].

4.4 Changes of Volatile Compounds

Comparing the volatile small molecules of the two groups, it can be found that the contents of total volatile compounds and aldehydes increased significantly ($P<0.05$), while the contents of ketones and nitrogen-containing compounds decreased significantly ($P<0.05$). Compared with other volatile compounds, aldehydes are considered to contribute the most to crab flavor because of their higher content and lower threshold [22]. In this study, the total content of aldehydes in feed group was significantly higher than that in ice fish group ($P<0.05$). The contents of 7 aldehydes (2- methyl -2- butenal, hexanal, 4- hexenal, benzaldehyde, nonanal, octanal aldehyde and 2,6- nonadialdehyde) in the feed group were significantly higher than those in the ice fish group ($P<0.05$). The higher content of these aldehydes may be caused by the feed group of *Eriocheir sinensis* eating compound feed rich in PUFA. PUFA in compound feed (51.170%) was higher than that in wild fish (37.533%). Meanwhile, the contents of two aldehydes (Heptanal and Undecanal) in the feed group were significantly lower than those in the ice fish group ($P<0.05$).

Besides aldehydes, ACCs also includes 2- ethyl furan, 2-pentylfuran, limonene, 1- octene -3- alcohol and trimethylamine. 2- ethyl furan and 2-pentylfuran, which belong to furan, also contribute greatly to the taste of crabs, and have similar performances in several crabs. Limonene, the only hydrocarbon in AAC, is common in *Eriocheir sinensis* [23]. In this study, compared with feed group, the content of trimethylamine in ice fish group increased significantly ($P<0.05$), which was consistent with the sensory evaluation of ice fish group with higher fishy smell score. The higher content of trimethylamine in ice fish group may be due to the consumption of ice fish by Chinese mitten crab, so crabs contain more fish compounds than feed group. It is reported that only one alcohol, 1- octene -3- ol, exists in *Eriocheir sinensis*. In this study, the content of 1- octene -3- ol in feed group was higher than that in ice fish group. Aldehydes can be converted into corresponding alcohols by alcohol dehydrogenase. The higher content of 1- octene -3- ol may be related to the increase of aldehyde [24-25].

Based on the experimental data, we speculated that the difference of aldehyde was the main reason for the flavor change between the ice fish group and the feed group. Previous studies have confirmed that aldehydes produced by PUFA catalyzed by lipoxygenase play an important role in forming unique aroma in fruits and vegetables.

V. CONCLUSION

It was found that the Chinese mitten crab in the feed group had higher sweetness, umami taste and grass fragrance, while the fishy smell was lower than that in the ice fish group. There was no significant difference in AA components between the two groups, but the PUFA ratio of Chinese mitten crab fed with feed increased significantly, while the ratio of SFA and MUFA

decreased significantly. In addition, AMP and EDU increased significantly in the feed group. Aldehydes are one of the important factors that can produce unique flavor of Chinese mitten crab. Aldehydes increase significantly after feeding Chinese mitten crab with compound feed, which may be related to the activity of lipoxygenase. Feeding feed can completely replace the traditional Iced fish. Feeding feed is easy to operate and store, and Chinese mitten crab has more flavor.

ACKNOWLEDGMENT

This research was financially supported by "Qing Lan Project of Jiangsu Province" (〔2018〕12)and "311 Talents Project in Taizhou City" (2017Ⅲ−804), College Students' Innovative Entrepreneurial Training Plan Program (202012806019Y).

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