

## Comparative analysis of muscle nutrient in two types of hybrid bream and native bream



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### ABSTRACT

Distant hybridization can integrate parental advantages to produce new varieties, which may also improve the fish meat quality. Fish is considered to be one of the healthiest meats because it can provide rich protein sources and high levels of omega-3 long-chain polyunsaturated fatty acids. Two types of hybrid bream (BBTB and BTBB) were obtained by the two rounds of backcrossing with an established hybrid lineage of blunt snout bream (*Megalobrama amblycephala*, BSB, ♀) × topmouth culter (*Culter alburnus*, TC, ♂). In the present study, the muscle nutrients, including moisture, fat, protein and ash contents, as well as amino acid and fatty acid compositions, in BBTB, BTBB and their parent BSB with the age of four months were investigated and analyzed by biochemical methods. The results showed that the moisture content of BTBB (76.6%) was lower than that of BSB (77.4%) and BBTB (77.3%), while the protein content of BBTB (17.1%) and BTBB (17.6%) was higher than that of BSB (16.3%). Seventeen amino acids were detected in the muscles from three kinds of fish, among which total amino acids, essential amino acids and delicious amino acids in BTBB and BBTB were all to different degrees higher than those of BSB. Further analysis of fatty acids revealed that BTBB and BBTB exhibited higher levels of unsaturated fatty acids (UFAs) content and the sum content of eicosapentaenoic acid (EPA, C20:5 n-3) and docosahexaenoic acid (DHA, C22:6 n-3) in muscle than BSB. Taken together, the results showed the advantages of BBTB and BTBB in nutritional value and taste, which provided theoretical support for their production application.

### 1. Introduction

Freshwater fish are rich in high-quality protein, unsaturated fatty acids (UFAs), vitamins and essential minerals, and also contain large amounts of essential amino acids (EAAs), especially lysine (Lys), which is low in cereals [1]. The EAA pattern of fish protein is very close to human needs, and its digestion and absorption rate is as high as 95% [2], which is an ideal source of animal protein. Therefore, fish protein can be used to supplement the amino acids species and improve the overall protein quality in a mixed diet [1]. Fat is an important part of the human diet, but excessive intakes of fat or unbalanced fat types have negative health effects [3]. Excessive intakes of saturated fatty acids (SFAs) can lead to fat accumulation and inflammation, and intakes of polyunsaturated fatty

acids (PUFAs) can control the oxidation of SFAs and the synthesis of monounsaturated fatty acids (MUFAs), thereby reducing liver fat content [4–6]. The fat of freshwater fish contains a low ratio of n-6/n-3 PUFAs [7], which can improve people's nutrition structure and reduce the risk of cardiovascular disease [8–10]. Fish is also considered to be one of the healthiest meats because it can provide rich protein sources and high levels of omega-3 long-chain PUFAs [11–13]. Some problems are currently restricting the fishery development, such as the reduced natural fish resources, limited water resources and aquaculture area. It is necessary to develop fish with the advantages of fast growth, high-quality meat and strong stress resistance.

Distant hybridization, or interspecific hybridization, refers to the cross between different species or higher-ranking taxa, which can

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integrate parental advantages and obtain the hybrid offspring showing heterosis in growth rate, meat quality, stress resistance, appearance characteristics and survival rate [14–16]. It is widely accepted that distant hybridization cannot produce a fertile lineage because of the existence of reproductive isolation in interspecific hybridization [16]. However, through long-term systematic research, several excellent fish lineages, such as allodiploid fish lineages [17–19], allotetraploid fish lineages [20], homodiploid fish lineages [21,22] and autotetraploid fish lineages [23,24] were established by distant hybridization. Using these fertile lineages to mate with related diploid fish, high-quality hybrid fishes with multiple advantages have been developed. For example, the allotetraploid hybrid lineage was obtained from the hybridization of female *Carassius auratus* red var. and male *Cyprinus carpio* L. [20,25,26]. Subsequently, allotriploid crucian carp was produced from the hybridization of female *C. auratus* red var. and male allotetraploid hybrids, which were sterile and exhibited fast growth, high-quality meat and strong stress resistance [27–29].

Muscle nutritional component is one of the basic contents of fish nutrition research, and the detection and analysis of the basic nutrition components of economic fish are helpful to understanding their nutritional value. At present, the muscle nutritional components of many economic cyprinid fishes have been studied [30–33], and focus primarily on the general nutritional components, fatty acid and amino acid composition of muscle. Blunt snout bream (*Megalobrama amblycephala*, BSB), belonging to *Megalobrama*, Cultrinae, Cyprinidae, is an herbivorous freshwater fish native to China with great commercial value [34]. Topmouth culter (*Culter alburnus*, TC), belonging to the *Culter*, Cultrinae, Cyprinidae, is a carnivore freshwater fish cultured widely in East Asia with high-quality meat [25]. In previous studies, the fertile hybrid lineage (BTF<sub>1</sub>-BTF<sub>2</sub>) was obtained from the distant hybridization of female BSB and male TC [19,35]. Subsequently, the backcross offspring (BTB) was produced from the hybridization of female BTF<sub>1</sub> and male BSB [30]. Currently, a new type of hybrid bream (BBTB) was obtained by the backcrossing female BSB to male BTB [36], and another new type of hybrid bream (BTBB) was obtained by the backcrossing female BTB to male BSB [37]. Moreover, the biological characteristics of BBTB, BTBB and their parents (BSB, TC and BTB), such as appearance characteristics, gonadal development, chromosome number, DNA content, muscle composition, and molecular genetic composition, have been studied systematically. These results indicated that BBTB and BTBB were different from their original parents (BSB and TC) in terms of appearance characteristics and molecular composition, showing hybrid characteristics and hybrid heterosis. The appearances of BBTB and BTBB were similar to that of BSB [36,37]. However, the analysis of muscle nutritional components of BBTB and BTBB has not been reported.

This study aimed to study and analyze the muscle nutrition composition of two types of hybrid bream (BBTB and BTBB) and their parent BSB through a series of biochemical methods and to reveal the muscle nutrition value characteristics of BBTB and BTBB. This study would provide theoretical support for the application of BBTB and BTBB in production, and provide useful scientific data for fish hybrid breeding.

## 2. Materials and methods

### 2.1. Ethics statement

All experimental protocols and animal care procedures were approved by the Animal Care Committee of Hunan Normal University. Approval was acquired for the Administration of Affairs Concerning Animal Experimentation Guidelines from the Science and Technology Bureau of China.

### 2.2. Sample collection and preparation

In September 2021, eighteen individuals including six BSB, six BBTB and six BTBB (4-month-old individuals) were randomly selected from the

State Key Laboratory of Developmental Biology of Freshwater Fish located at Hunan Normal University, Changsha, China. To avoid experimental errors caused by the environmental conditions and forage, all individual fish was cultured in an open pool of 0.067-ha with artificial feeding under the same conditions (dissolved oxygen content, photoperiod, water temperature, forage, pH) prior to muscle collection. The individuals were euthanized with 100 mg/L MS-222 (Sigma-Aldrich, St Louis, MO, USA) before being dissected to minimize suffering. Subsequently, both sides of dorsal white muscle with the same position were excised surgically from fish, immediately frozen in liquid nitrogen and then stored at  $-80^{\circ}\text{C}$  refrigerator. Six fish from the same species were mixed as one biological replicate for nutritional value detection. The morphological characteristics data about fish samples, including whole length, weight and condition factor, were recorded at the time of the experiment (Table 1).

### 2.3. Proximate composition analyses

The proximate composition analyses in muscle were performed according to the procedure of the Association of Official Analytical Chemists [38]. Moisture content was determined by oven drying until a constant weight at  $105^{\circ}\text{C}$  for 24 h. Fat content was determined by Soxhlet extraction method using a Soxtec System HT (Soxtec System HT6, Tecator, Sweden). Protein content was determined by Kjeldahl method using an Auto Kjeldahl System (FOSS KT260, Switzerland). Ash content was determined by combustion in an electric muffle furnace at  $550^{\circ}\text{C}$  for 4 h [39]. Carbohydrates were determined by subtracting the mass fractions of fat, protein and ash. The total energy value for each sample was determined by the content of protein, fat and total carbohydrates multiplied by the corresponding energy value, with the different components of each 100 g of muscle multiplied by the energy values of 16.75, 37.68 and 16.75 kJ, respectively [30,36,37].

### 2.4. Determination of fatty acids

The fatty acids were extracted effectively from muscle samples using methanol-chloroform method, and then chloroform was volatilized by nitrogen filling. Fatty acid methyl esters (FAMES) were prepared according to the procedure as described previously [40]. FAMES were subsequently analyzed on a gas chromatography mass spectrometry (GC-MS, Thermo Trace 1310 ISQ, Thermo Fisher Scientific, MA, USA), equipped with a chromatographic column (TG-5MS,  $30\text{ m} \times 0.25\text{ mm} \times 0.25\text{ }\mu\text{m}$ ). The identification of fatty acids in the samples was determined accurately based on retention time and mass spectra of unknowns with blend standards (Sigma, China) run under the same conditions. All samples were measured in triplicate, and the calculation result was expressed as the relative content of each fatty acid (% of total fatty acid).

### 2.5. Determination of amino acids

The amino acids were detected by Agilent 1200 Series High Performance Liquid Chromatograph (Agilent Technologies, CA, USA) according to the manufacturer's protocol. The content of tryptophan was

**Table 1**  
Sample information.

Species	Weight (g)		Body length (cm)		Condition factor (%)	
	Range	Mean ± S.E.M.	Range	Mean ± S.E.M.	Range	Mean ± S.E.M.
BSB	29.3–39.7	32.08 ± 3.93	11.4–13.0	11.80 ± 0.62	1.81–2.01	1.95 ± 0.07
BBTB	32.4–44.6	40.02 ± 4.26	11.3–12.4	12.00 ± 0.43	2.15–2.42	2.31 ± 0.10
BTBB	25.0–49.1	37.70 ± 8.20	11.4–13.5	12.42 ± 0.78	1.69–2.24	1.95 ± 0.21

analyzed by alkaline hydrolysis of each muscle sample, and other amino acids were determined by acid hydrolysis. All measurements were performed in triplicate, and the results were expressed as g/100 g on a dry weight basis.

### 3. Results

#### 3.1. Proximate composition

The phenotypes of two types of hybrid bream (BBTB and BTBB) and BSB were illustrated in Fig. 1. The proximate composition of muscle of BBTB, BTBB and BSB was shown in Table 2. The moisture content of BTBB was 76.6%, which was lower than that of BSB (77.4%) and BBTB (77.3%). The protein content of BBTB and BTBB was 17.1% and 17.6%, respectively, which was higher than that of BSB (16.3%). The ash content of BSB and BBTB was the same (1.4%), but lower than that of BTBB (1.6%). The carbohydrate content of BBTB and BTBB was 78.3% and 77.4%, respectively, which was lower than that of BSB (79.0%). Besides, the fat and energy value contents of BSB (3.3% and 1720.62 KJ, respectively) were similar to that of BBTB (3.2% and 1718.53 KJ, respectively) and BTBB (3.4% and 1719.36 KJ, respectively). Taken together, the aforementioned data indicated that, in terms of muscle nutrient composition, two types of hybrid bream (BBTB and BTBB) showed higher protein content, lower moisture and carbohydrate contents than BSB.

#### 3.2. Fatty acid composition

The muscle fatty acid profiles of two types of hybrid bream (BBTB and BTBB) and BSB were shown in Table 3. A total of seventeen fatty acids, including four SFAs and thirteen UFAs (including five MUFAs and eight PUFAs), were detected and identified. With respect to the SFAs, palmitic acid (PA, C16:0) was dominant, accounting for 15.41%–16.26% of the total fatty acids. Stearic acid (SA, C18:0) was also abundant. UFAs were the main fatty acids in the muscle of three fishes, accounting for 76.46%–77.73% of the total fatty acids. Besides, the proportion of UFAs in BTBB muscle was higher than that of BBTB, and was much higher than that of BSB. MUFAs and PUFAs in three fishes accounted for 52.58%–53.31% of the total fatty acids and 23.15%–25.14% of the total fatty acids, respectively. The proportion of PUFAs in BTBB muscle was higher than that of BBTB, and was much higher than that of BSB (Fig. 2). The main MUFA was oleic acid (OA, C18:1 n-9), accounting for 47.52%–48.46% of the total fatty acids, followed by palmitoleic acid (PA, C16:1). The proportion of OA in BSB muscle was higher than that of the other two fishes, followed by BTBB and BBTB in decreasing amounts. However, the PA content in BSB muscle was lower than that of the other two fishes. Among the PUFAs, linoleic acid (LA, C18:2 n-6) was dominant, accounting for 18.55%–19.55% of the total fatty acids. The proportion of LA in BBTB and BTBB muscle was higher than that of BSB. The  $\alpha$ -linolenic acid (ALA, C18:3 n-3), docosahexaenoic acid (DHA, C22:6 n-3) and dihomo- $\gamma$ -linolenic acid (C20:3 n-6) concentration in BSB muscle were lower than those of BBTB and BTBB. Moreover, BSB had a lower concentration of n-3 PUFAs than that of BBTB and BTBB. The sum content of eicosapentaenoic acid (EPA, C20:5 n-3) and DHA of BSB muscle were lower than those of BBTB and BTBB. The proportion of n-6 PUFAs in BBTB and BTBB muscle was higher than that of BSB (Fig. 3). The ratio of n-3/n-6 PUFAs in BBTB and BTBB muscle was slightly higher than that of BSB. On the whole, the

**Table 2**

Proximate composition of BBTB, BTBB and BSB.

Fish type	Moisture (g/100 g)	Fat (g/100 g)	Protein (g/100 g)	Ash (g/100 g)	Carbohydrates (g/100 g)	Energy value (kJ/100 g)
BSB	77.4	3.3	16.3	1.4	79.0	1720.62
BBTB	77.3	3.2	17.1	1.4	78.3	1718.53
BTBB	76.6	3.4	17.6	1.6	77.4	1719.36

Data were the mixed determination results of six samples.

**Table 3**

Fatty acids composition (g/100 g total fatty acids) of BBTB, BTBB and BSB.

Fatty acids	BSB	BBTB	BTBB
C14:0	0.58	0.59	0.59
C15:0	–	0.12	–
C16:0	15.54	15.41	16.26
C18:0	7.42	6.64	5.42
$\sum$ SFAs	23.54	22.76	22.27
C16:1	2.03	2.23	2.53
C18:1 n-9c	48.46	47.52	47.59
C20:1	1.41	1.53	1.32
C22:1 n-9	1.25	1.18	1.03
C24:1	0.16	0.12	0.12
$\sum$ MUFAs	53.31	52.58	52.59
C18:2 n-6c (LA)	18.55	19.18	19.55
C18:3 n-6 (GLA)	–	0.25	0.25
C18:3 n-3 (ALA)	1.49	1.89	1.74
C20:2	0.40	0.46	0.40
C20:3 n-6	0.89	0.95	1.02
C20:4 n-6 (ARA)	1.09	1.05	1.20
C20:5 n-3 (EPA)	–	–	0.13
C22:6 n-3 (DHA)	0.73	0.88	0.85
$\sum$ PUFAs	23.15	24.66	25.14
$\sum$ n-3 PUFA	2.22	2.77	2.72
$\sum$ n-6 PUFA	20.53	21.43	22.02
$\sum$ n-9 MUFA	49.71	48.70	48.62
$\sum$ (EPA + DHA)	0.73	0.88	0.98
$\sum$ (EPA + DHA)/ $\sum$ PUFA	0.03	0.04	0.04
$\sum$ n-3 PUFA/ $\sum$ n-6 PUFA	0.11	0.13	0.12
$\sum$ PUFA/ $\sum$ SFA	0.98	1.08	1.13
$\sum$ UFAs	76.46	77.24	77.73

Data were the mixed determination results of six samples. - represented not detected.

$\sum$ n-3 PUFA: n-3 polyunsaturated fatty acids.

$\sum$ n-6 PUFA: n-6 polyunsaturated fatty acids.

$\sum$ n-9 MUFA: n-9 monounsaturated fatty acids.

$\sum$ (EPA + DHA): eicosapentaenoic acid and docosahexaenoic acid.

$\sum$ (EPA + DHA)/ $\sum$ PUFA: the ratio of eicosapentaenoic acid and docosahexaenoic acid to polyunsaturated fatty acids.

$\sum$ n-3 PUFA/ $\sum$ n-6 PUFA: the ratio of n-3 polyunsaturated fatty acids to n-6 polyunsaturated fatty acids.

single fatty acid content of two types of hybrid bream (BBTB and BTBB) was basically higher than that of BSB.

#### 3.3. Amino acid composition

The muscle of hydrolyzed amino acids composition of two types of hybrid bream (BBTB and BTBB) and BSB were summarized in Table 4. A total of seventeen amino acids, including eight EAAs, two semi-essential



**Fig. 1.** Appearance of BBTB, BTBB and BSB. A. Appearance of BSB. B. Appearance of BBTB. C. Appearance of BTBB. Scale bar, 1 cm.

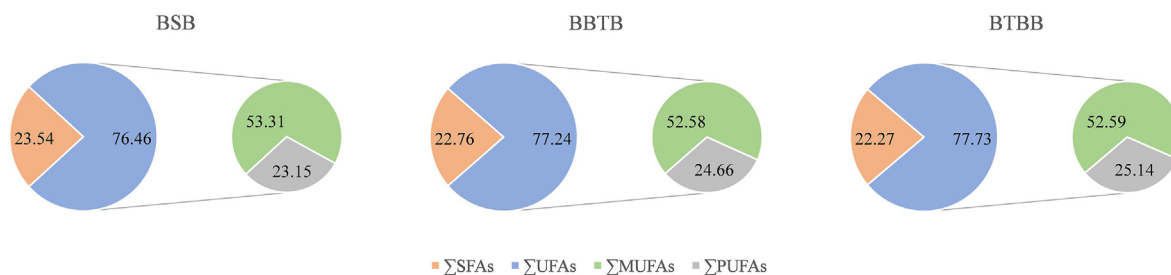


Fig. 2. Fatty acid composition of BBTB, BTBB and BSB.

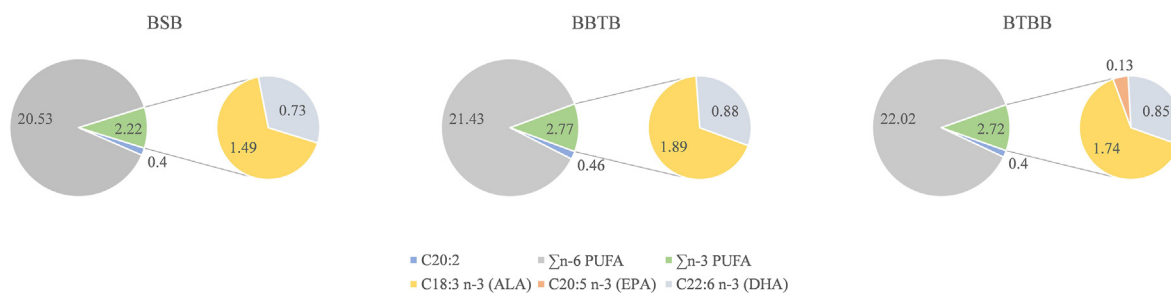


Fig. 3. PUFAs composition of BBTB, BTBB and BSB.

Table 4

Amino acid composition (g/100 g protein) of BBTB, BTBB and BSB.

Amino acid	BSB	BBTB	BTBB
Aspartic acid (Asp) <sup>#</sup>	1.58	1.74	1.79
Glutamic acid (Glu) <sup>#</sup>	2.61	2.69	2.75
Glycine (Gly) <sup>#</sup>	0.86	0.79	0.78
Alanine (Ala) <sup>#</sup>	0.85	0.82	0.82
Threonine (Thr) <sup>*</sup>	0.79	0.72	0.72
Valine (Val) <sup>*</sup>	0.80	0.77	0.81
Methionine (Met) <sup>*</sup>	0.47	0.48	0.49
Isoleucine (Ile) <sup>*</sup>	0.79	0.88	0.92
Leucine (Leu) <sup>*</sup>	1.43	1.36	1.42
Phenylalanine (Phe) <sup>*</sup>	0.76	0.71	0.74
Lysine (Lys) <sup>*</sup>	1.43	1.60	1.62
Tryptophan (Trp) <sup>*</sup>	0.15	0.14	0.15
Serine (Ser)	0.66	0.68	0.65
Histidine (His) <sup>**</sup>	0.40	0.49	0.54
Arginine (Arg) <sup>**</sup>	0.91	0.97	0.99
Proline (Pro)	0.58	0.57	0.56
Tyrosine (Tyr)	0.59	0.58	0.60
DAA	5.90	6.04	6.14
EAA	6.62	6.66	6.87
TAA	15.66	15.99	16.35
NEAA	7.73	7.87	7.95
DAA/TAA (%)	37.68	37.77	37.55
EAA/TAA (%)	42.27	41.65	42.02
EAA/NEAA (%)	85.64	84.63	86.42

Data were the mixed determination results of six samples. \* indicated essential amino acids; \*\* indicated semi-essential amino acids; # indicated delicious amino acids.

DAA: delicious amino acids.

EAA: essential amino acids.

NEAA: non-essential amino acids.

TAA: total amino acids.

amino acids and seven non-essential amino acids (NEAAs), were detected and identified. By comparing the measured amino acids content of BBTB, BTBB and BSB, it was found that the individual amino acid content of BBTB and BTBB was basically higher than that of BSB. The total amino acids (TAAs) content in BBTB and BTBB muscle was higher than that of BSB (Fig. 4). Glutamic acid (Glu) was the most abundant amino acid in the three fishes, followed by aspartic acid (Asp), Lys, leucine (Leu) and

arginine (Arg) in decreasing amounts, and tryptophan (Trp) was the lowest amongst amino acid. The EAAs content in BBTB and BTBB muscle was slightly higher than that of BSB (Fig. 4). The ratio of EAA to TAA (EAA/TAA) in three fishes ranged from 41.65% to 42.27%, and the ratio of EAA to NEAA (EAA/NEAA) ranged from 84.63% to 86.42%. According to the ideal FAO/WHO model, the EAA/TAA and EAA/NEAA ratios of high-quality protein should be around 40% and higher than 60%, respectively [41]. The muscle amino acid composition of the three fishes was all higher than the FAO/WHO evaluation standard, which were high-quality protein sources. Asp, Glu, glycine (Gly) and alanine (Ala) were umami and sweet amino acids. Glu was the most abundant delicious amino acid (DAA), followed by Asp, Ala and Gly in decreasing amounts. Both Asp and Glu concentrations in BBTB and BTBB muscle were higher than those of BSB, while Ala and Gly concentrations in BBTB and BTBB muscle were lower than those of BSB. The content of these four DAAs in BBTB and BTBB muscle was slightly higher than that of BSB (Fig. 4). However, there was little difference in the ratio of DAA to TAA (DAA/TAA) among the three fishes.

#### 4. Discussion

As an important source of human protein intake, muscle nutrition is one of the important indicators for evaluating the quality and production performance of fish. The edible part of fish is mainly the muscle of the trunk. Protein, fat, moisture and ash are the basic components of muscle, and their relative contents can not only be used as nutritional indicators to scientifically evaluate the nutritional value of fish, but also to guide breeding and evaluation of germplasm standards.

The nutritional value of fish mainly depends on the protein and fat content. Fish is mainly composed of water, and the moisture content of fish muscle ranges from 70% to 85%. In the present study, the moisture content of BTBB was lower than that of BSB and BBTB. Previous studies have reported the muscle nutrients of common commercial fishes such as grass carp (*Ctenopharyngodon idella*, GC), mandarin fish (*Siniperca chuatsi*, MF), silver carp (*Hypophthalmichthys molitrix*, SC), bighead carp (*Aristichthys nobilis*, BC) and mirror carp (*Cyprinus carpio* var. *specularis*, MC) [42,43]. Compared with the moisture content of two types of hybrid bream (BBTB and BTBB) and the aforementioned fishes, the results showed that the moisture content of BBTB and BTBB was also lower than



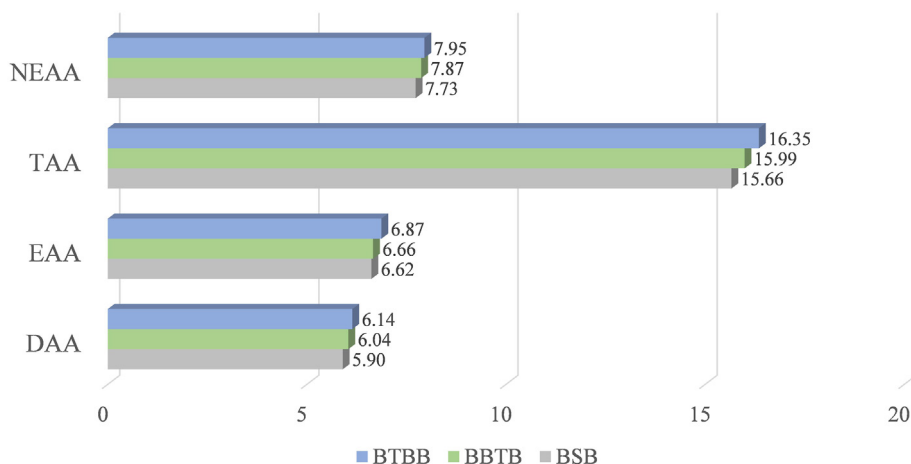


Fig. 4. Comparison of NEAA, TAA, EAA and DAA of BBTB, BTBB and BSB.

that of GC, MF, SC and BC (Table 5). The protein content of BBTB and BTBB was higher than that of BSB, and also much higher than that of GC, SC and MC (Table 1, Table 5). The protein content of BBTB and BTBB was higher than that of GC, MF, SC and BC (Table 5). The carbohydrate content of BTBB was lower than that of GC, BC and MC, and BBTB was lower than that of GC and MC (Table 5). The ash content of BBTB and BTBB was lower than that of SC (Table 5). The energy value content of BBTB and BTBB was higher than that of GC, MF, SC and BC (Table 5). Therefore, BBTB and BTBB had lower moisture and carbohydrate contents but higher protein and energy value contents, indicating that they were new high-quality fishes with distinct characteristics from BSB and other related fish. These characteristics of BBTB and BTBB will be beneficial to their application in production, and also provide new materials for nutritionists and scientific researchers to improve the nutritional value of fish.

Fish flesh is an important source of protein in human diet [40]. Protein is the first nutritional element of the human body and plays a contributory role in dietary nutrition. However, it cannot be directly utilized in the human body, but can be exploited after decomposition into small amino acid molecules [31]. The nutritional value of protein is not only reflected in the content but also depends on the quality, that is, the composition and content of amino acids, especially the type, composition and content [44]. In meat research, the EAA pattern of fish protein is very similar to that of human needs and has high nutritional value. In the present study, seventeen amino acids were detected in the muscle of BSB, BBTB and BTBB. The individual amino acid content in two types of hybrid bream (BBTB and BTBB) was basically higher than that of BSB. The EAA/TAA and EAA/NEAA ratios of the three fishes were 41.65%–42.27% and 84.63%–86.42%, respectively, which were all higher than the ideal FAO/WHO model (EAA/TAA around 40%, EAA/NEAA >60%) [41]. Therefore, they both meet the FAO/WHO evaluation standard, with a balanced proportion and high nutritional value. Whether animal protein is delicious or not to a certain extent depend on the composition and content of DAAs. The DAAs mainly include Glu, Asp, Ala and Gly, among which Glu and Asp are umami amino acids, and Glu has the strongest umami taste. Ala and Gly can stimulate the palate to produce sweetness and neutralize bitter and salty amino acids [45]. The DAAs content of BTBB (6.14 g/100 g) and BBTB (6.04 g/100 g) was higher than that of BSB (5.90 g/100 g). Glu is of great significance in human metabolism and is the primary amino acid in the biochemical metabolism of brain tissue, which can help to improve memory. In the present study, Glu content in BTBB muscle ranked first among the amino acids measured (2.75 g/100 g), and was also higher than that of BBTB (2.69 g/100 g) and BSB (2.61 g/100 g). Lys is one of the essential amino acids in the human body, which can promote human development, enhance immune function and improve the function of the central nervous system [40,46]. Lys is a

Table 5

The comparison of proximate composition of BBTB, BTBB and other common commercial fish species.

Fish type	Moisture (g/100 g)	Fat (g/100 g)	Protein (g/100 g)	Ash (g/100 g)	Carbohydrates (g/100 g)	Energy value (kJ/100 g)
BBTB	77.30	3.20	17.10	1.40	78.30	1718.53
BTBB	76.60	3.40	17.60	1.60	77.40	1719.36
GC	82.71	0.45	15.10	1.17	83.28	1664.82
MF	79.03	1.50	16.75	2.67	79.08	1661.67
SC	78.79	2.20	15.71	3.42	78.67	1663.76
BC	80.18	0.74	16.75	2.08	80.23	1655.65
MC	78.24	6.09	13.11	1.11	79.69	1783.87

Data were the mixed determination results.

common limiting amino acid because it is the most deficient amino acid in nearly all wheat and corn diets [47]. Long-term consumption of cereal alone can inevitably cause Lys deficiency in the human body, leading to diseases, such as loss of appetite, metabolic disorders and reduction activity of various enzymes in the body. Lys content of BTBB (1.62 g/100 g) and BBTB (1.60 g/100 g) was higher than that of BSB (1.43 g/100 g). The aforementioned research results indicated that BBTB and BTBB had obvious advantages in terms of nutritional value and taste, and were economic fish worthy of promotion.

Fatty acids in two types of hybrid bream (BBTB and BTBB) and BSB muscle were abundant, and a total of seventeen fatty acids including four SFAs, five MUFAs and eight PUFAs were detected. In the present study, the major SFA was PA and other predominant SFAs were SA in these three fishes. SFAs are generally thought to be “bad” fat, because overconsumption of SFAs in the diet is associated with lipotoxicity-related disorders, including obesity, diabetes, cancer, cardiovascular diseases and nonalcoholic fatty liver diseases [48]. The UFAs content of BTBB (77.73%) was higher than that of BBTB (77.24%), and was much higher than that of BSB (76.46%). The nutritional value of lipids largely depended on the type and quantity of fatty acids, especially UFAs content [49]. The most abundant MUFA was OA in these three fishes, in agreement with that reported previously [30]. These values were consistent with approximately 40%–50% OA in fatty acids in animal fat. OA is considered to possess modulatory effects in a wide range of physiological functions, while some studies also suggest a significant role in cancer, inflammatory and autoimmune diseases, in addition to its ability to promote wound healing [50]. According to a nutritional classification, fatty acids that are indispensable for development and health but cannot be synthesized by humans are classified as essential fatty acids, while those that can be produced by humans are considered as non-essential

fatty acids. Therefore, LA and ALA are PUFAs classified as essential fatty acids, while MUFAs are considered as non-essential fatty acids [50]. The LA content of BTBB (19.55%) and BBTB (19.18%) was both higher than that of BSB (18.55%). LA is valued for its ability to lower blood cholesterol levels and prevent atherosclerosis [51]. Fat is one of the important substances that produce aroma when fish is cooked. The high content of PUFAs can significantly enhance the aroma while reflecting the juiciness of the muscles. A growing body of evidence indicates that PUFAs are a class of active substances with special physiological functions, which could reduce cholesterol and triglycerides in the blood, thereby preventing hypertension and atherosclerosis, but also can reduce blood viscosity, improve blood microcirculation, and prevent cardiovascular diseases [52]. The PUFAs content of BTBB (25.14%) was higher than that of BBTB (24.66%), and was significantly higher than that of BSB (23.15%). The PUFA/SFA ratio in BSB (0.98), BBTB (1.08) and BTBB (1.13) was both much higher than the FAO/WHO evaluation standard (PUFA/SFA >0.45). Generally, n-3 PUFAs are bound up with human health, which serve an essential role in reducing cardiovascular and cerebrovascular diseases, inflammation, and enhancing the activity of anticancer drugs [53]. The n-3 PUFAs content of BBTB (2.77%) and BTBB (2.72%) was higher than that of BSB (2.22%). Representatives of n-3 PUFAs are EPA and DHA. They have exceedingly beneficial characteristics for the prevention of human cardiovascular and cerebrovascular diseases [49,50]. EPA and DHA are mainly provided by aquatic products to the human diet. In nature, only aquatic organisms such as red algae, brown algae and diatom can synthesize EPA and DHA, which are introduced into the food chain after being eaten by aquatic animals [54]. The sum content of EPA and DHA of BBTB (0.98%) was the highest, followed by BTBB (0.88%) and BSB (0.73%) in decreasing amounts. Therefore, these results demonstrated that BTBB had obvious advantages in terms of UFAs and the sum content of EPA and DHA compared with BBTB and BSB.

Fish is a low-cost but high nutritional value source of animal protein that can serve as an important source to meet the human body's protein demands, resulting in significant economic advantages. New varieties of high-quality freshwater fish with high protein and low moisture are the goal of modern fish breeding. Hybridization is an effective genetic improvement method that combines the genomes of different species, resulting in genotypic and phenotypic changes of hybrid offspring [14, 15,26]. In the present study, the muscle quality of BBTB and BTBB was improved by hybridization, which was consistent with previous studies [30,31,36,37]. The combination of heterologous genomes of BSB and TC can lead to changes in the phenotype of BBTB and BTBB, including changes in the UFAs and DAAs contents.

In summary, compared to native bream, two types of hybrid bream (BBTB and BTBB) had the characteristics of high protein, low moisture, abundant amino acid species, balanced proportion, high content of EAAs and UFAs, which had high edible value and exhibited certain hybrid heterosis. As two new types of freshwater aquaculture fish, BBTB and BTBB not only enrich the freshwater aquaculture species, but also have important significance for the research of fish genetics breeding and fish nutrition value.

#### Author contributions

Min Tao and Shaojun Liu conceived and designed the research. Siyu Fan, Zhifeng Zhou, Chang Wu, Dingbin Gong and Ming Wen contributed to sampling. Shengnan Li and Xiangqiong Yang analyzed the data. Shengnan Li wrote the manuscript. Min Tao, Rong Zhou and Yuequn Wang revised the manuscript. All authors read and approved the final manuscript.

#### Declaration of competing interest

The authors declare that they have no competing interests.

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