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Title: Replacement of Fish Meal with Mixed Rendered Animal Protein in Practical Diets for Siberian Sturgeon (*Acipense baeri* Brandt)

By: Dr. Min XUE

University / Location:

Min Xue, Ph.D
The Lab of Aquaculture Nutrition and Feed
Feed Research Institute, the Chinese Academy of Agricultural Sciences,
12# Zhongguancun South Street,
Beijing,100081
P.R.CHINA
Tel/Fax:8610-68919753; Email: xuemin@caas.net.cn

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Objectives:

1) To evaluate a higher mixed rendered animal protein utilization level (75-100%) in diet for Siberian sturgeon on the growth performance, economical returns and somatotropic axis responsiveness to fish meal replacement.
2) Develop least-cost feed formulae for Siberian sturgeon.

Replacement of Fish Meal with Mixed Rendered Animal Protein in Practical Diets for Siberian Sturgeon (*Acipenser baerii* Brandt)

Min Xue[1*] Hongjian SHENG[1] Haiyan LIU[1] Xiufeng WU[1] Liya GUO[1] Weixiang ZHAO[2], Sergio Nates [3] Yu YU[4]

1. Feed Research Institute, the Chinese Academy of Agricultural Sciences, Beijing; P. R. China
2. Beijing Enhalor Biotechology Co. Ltd. Beijing, P. R. China
3. Fats and Proteins Research Foundation, Inc, USA
4. National Renderers Assocciation, Int. Hongkong

Running head: Mixed Rendered Animal Protein Utilization for Sturgeon

[*]Corresponding author: Tel/Fax: 86-10-68919753; e-mail: xuemin@caas.net.cn

ABSTRCT

The effect of rendered animal protein blend (APB, including meat & bone meal, MBM: 35%; poultry by-product meal, PBM: 40%; hydrolysed feather meal, HFM: 5%; spray dried blood meal, BM: 20%) as a partial or total replacement of fishmeal (FM) was studied in juvenile Siberia sturgeon (*Acipenser baerii* Brandt) in an eight-week growth trial. Six extruded experimental diets were formulated. 48.3% and 40% of fishmeal (FM) were used in control diets with crude protein at 40% and 36%, respectively. 75% or 100% of FM will be replaced by MP in other 4 diets, in which lysine, methionine, and threonine will be balanced under ideal protein concept by crystallized amino acid（Lys-H_2SO_4, 65 %；DL-Met. 98% and L-Thr. 98%）, respectively. Accordingly the six diets were named as FM40, APB75-40, APBT-40, FM36, ABP75-36, ABPT-36, respectively. 0.1% of Yttrium oxide (Y_2O_3) as inner marker for digestibility determination was designed for nitrogen and phosphorus excretion. Fish body composition and serum somatotrpic axis hormone GH and IGF-I were determined.

Substitution of 75% or 100% fishmeal with MP, either at dietary 40% CP or at dietary 36% CP, did not result in the decrease of the growth performance, feed efficiency, nitrogen retention and phosphorus retention ($P>0.05$), and did not affect the body or liver composition in Siberian sturgeon as well ($P>0.05$). As to the effects of dietary protein, Siberian sturgeon fed diets with the optimal protein level (40% CP) or with the sub-optimal protein level (36% CP) also showed the same growth performance, nitrogen retention and phosphorus retention, but the feed intake in 36% CP was higher than that in 40% CP, and the feed efficiency in 36% CP was lower than that in 40% CP($P<0.05$). The results indicated that MP can be utilized by juvenile Siberia sturgeon up to 378.20g kg^{-1} to replace 100% of fishmeal protein under ideal amino acid concept with dietary 36% CP in this study.

Key words: Siberian sturgeon (*Acipense baerii* Brandt); ideal protein concept; Mixed territorial animal protein; Fishmeal.

1. Introduction

Sturgeon has mainly been farmed for caviar production, but in the last decade there has been increased interest in farming for flesh production. Because of the need to expand production by introduce of new species, sturgeon farming has been increased rapidly in China. From 1998 to 2002, production of sturgeon had increased by 50 times whereas the market price dropped by 10 times in China, and it is believed that China has become the largest sturgeon aquaculture country in the world from 2000 (Wei and Yang, 2003; Wei, et al., 2004). However, there have been only a few studies on Siberian sturgeon feeding and nutrition undertaken by French teams (Médale, et al., 1995), but seldom concern on fish meal and fish oil substitution. Gisbert and Williot (2002) reported a fully detailed review on Siberian sturgeon larval rearing. Papers on Siberian sturgeon feeding have been reported by Daborwski, et al. (1985) and later by Médale, et al. (1995). Among the other species of sturgeon, white sturgeon (*Acipenser transmontanus*) feeding and nutrition had been investigated to some extent (Hung, 1991). However, there is no standard practical growth out diet for sturgeon yet. Most sturgeon farmers use existing commercially available diets, particularly those of high energy salmonid diets with 40-45% crude protein content and 18-22MJ/kg gross energy with or without modification. Because the price of sturgeon is much higher than salmonid in China, higher feed cost can be accepted by farmer. Generally 40-50% FM used to be utilized in commercial sturgeon feed before 2006.

However, the price of fishmeal was up to 1600 US$/ton in 2006 and it was forecasted to be higher in the future due to higher freight costs and due to the Peruvian government decision to slow down the fish catch in order to replenish anchovy supplies (Goettl, 2003). Therefore, replacement of fishmeal with less expensive protein sources would be beneficial in reducing feed costs. Rendered animal protein ingredients, such as poultry by-product meal (PBM), meat and bone meal (MBM), blood meal (BM) and hydrolysed feather meal (HFM) have been used successfully in feeds for various fish species, such as Chinook salmon (Fowler, 1991), rainbow trout (Bureau, et al., 2000), red drum (Kureshy, et al., 2000), Australian silver perch (Allan, et al., 2000) and hybrid tilapia (Xue, et al., 2003).

The present study was conducted to assess the effect of combination of PBM, MBM and HFM and BM as a FM substitution on growth, feed utilization, nitrogen and phosphorus excretion, body composition and physiological responses in Siberian sturgeon practical feed under ideal protein concept.

2. Materials and methods

2.1 Diets

A growth trial was conducted to examine the utilization of a mixed-protein (MP), which combined with 4 rendered animal protein, poultry by-product meal (PBM), meat and bone meal (MBM), hydrolysed feather meal (HFM) (supplied by National Renderers Association, Ltd. Hongkong) and spray dried blood meal (BM, local production) at the ratio of 35:40:5:20 (Table 1), which held similar protein and amino acids profile to Peru fishmeal (FM).

Six extruded experimental diets were formulated. 48.3% and 40% of fishmeal (FM) were used in control diets with crude protein at 40% and 36%, respectively. 75% or 100% of FM will be replaced by MP in other 4 diets, in which lysine, methionine, and threonine will be balanced under ideal protein concept by crystallized amino acid（Lys-H2SO4, 65 %；DL-Met. 98% and L-Thr. 98%）, respectively. Accordingly the six diets were named as FM40, MP75-40, MPT-40, FM36, MPT-40, MPT-36. 0.1% of Yttrium oxide (Y2O3) as inner marker for digestibility determination was designed for nitrogen and phosphorus excretion. The diet formulation and chemical composition are shown in Table 1, and the amino acids composition of the experimental diets are presented in Table 2. The diets were made into pellets under the extrusion condition as: feeding section (90 /5s), compression section (150 /5s) and metering section (60 /4s) using a single-screwed extruder (EXT50A, YANGGONG MACHINE, Beijing, China). Analysed amino acids compositions of diets were showed in table 2.

2.2 Animals and husbandry

Siberian sturgeons (*Acipenser baerii* Brandt) were obtained from the Beijing Fishery Institute, Beijing, and maintained in the laboratory for 2 weeks prior to the experiment. The fish were maintained in conical fibreglass tanks (water depth: 80 cm; volume: $0.25m^3$) in a recirculating system during the acclimation and experimental period. Water temperature was 20-22 ℃., pH=7.5 and NH_4-N<0.5mg l^{-1}. Aeration was supplied to each tank 24h per day, and photoperiod was 12D:12L using eight 40W fluorescent light.

Initial body weight (IBW) of juvenile Siberian sturgeon was 38.98±0.02 g. Four tanks were randomly assigned to each diet group in the 8 weeks growth experiment. At the start of the experiment, fish were not fed for 1d, and then 20 fishes were batch weighed and stocked into each tank. During the feeding period, fish were fed the experimental diets to apparent satiation 3 times a day at 9:00, 15:00 and 21:00, respectively. One h later for each feeding, uneaten feed was removed, dried to constant weight at 70℃ and reweighed. Leaching loss in the uneaten diet was estimated by leaving five samples of each diet in tanks without fish for 1h, recovering, drying and reweighing. Feaces of each tank were collected after 2 weeks. All the fish of each tank were batch weighed at each 2 weeks for regulating the feeding rate. Throughout the 8 week experiment, mortalities were recorded daily. At the end of experiment, carcass, fillet, liver and serum of fish from each tank were prepared for later analysis.

2.3 Chemical analysis:

Dry matter, crude protein, lipid, ash, energy and amino acids were analyzed for experimental diets and fish samples. Fibers were analyzed for diets. Crude protein, amino acids were analysed for protein ingredients. Plasma somatotropic axis hormone GH and IGF-I, hepatic activity of amino acid catabolising enzymes (alanine aminotransferase (ALT), sapartate aminotransferase (AST)), serum total protein (TP), glucose (GLU), triglyceride (TG) and Total cholesterol (TC) were determined for blood samples.

Dry matter was analyzed by drying the samples to constant weight at 105℃. Crude protein was determined by combustion using the Kjeldahl method (AOAC, 1997) and Crude protein content estimated by multiplying nitrogen by 6.25. Crude lipid by acid hydrolysis with a Sotex System HT 1047 Hydrolyzing Unit (Tecator Application Note 92/87), followed by Soxhlet extraction using a Soxtex system 1043, ash was analysed by combustion in a muffle furnace at 550℃ for 16h, crude fiber was determined using a Fibrotec System 1020. The amino acids of ingredients, diets and fish muscle were determined by HPLC (Agilent, 1100). Plasma GH and IGF-I were determined by a RIA(Radioimmunoassay) kit produced by Diagnostic System Laboratories USA. Serum ALT, AST, TP, Glu, TG, UREA, TC were determined by Beckman coulter CX4 Pro. using commercial kits (Randox AL 7930; Randox AS 7938; Marker 3400359; Marker 3400360; Labo D-79343; Marker 3400362), Serum lysozyme, SOD and MDA were analyzed with commercial kits (Nanjing Jiangcheng Biotech. Co.). Duplicate analyses were conducted for each sample.

2.4. Statistical analysis

Data were reported as mean values ± standard error of mean (S.E.). Homogeneity of variance was confirmed and comparison between means was by two-way ANOVA. Tukey’s procedure was used for multiple comparisons. Differences were regarded as significant when P <0.05. All the statistical analyses were performed by STATISTICA 6.0.

3. Results

3.1. Growth performance

The growth performance and feed efficiency values are presented in Table 4. In this study, fish growth (final body weight, FBW; weight gain, WG and specific growth rate, SGR; protein efficiency ratio, PER; net protein retention, NPR) was not significantly affected by the MP and diet protein level (P>0.05). The feeding rate, FR and feed conversion rate, FCR were significantly affected by the CP levels but not by MP. FR and FCR in 40% CP groups were higher than those in 36% CP groups (P<0.05). No significant difference was observed for condition factor (CF), hepatosomatic index (HSI) and viscerasomatic index (VSI) among all groups (P>0.05).

Apparent digestibility coefficients

Seen from Table 5, the Apparent digestibility coefficients of dry matter (ADCd) were significantly affected by CP and MP, the ADCd in 40% protein group was higher than that in 36% protein group. MP has significantly decreased the ADC of dry matter(ADCd), crude protein (ADCp) and gross phosphorus (ADCp), while the ADCd and ADCp were not significantly affected by CP level in this study.

Nitrogen and phosphorus excretion

Table 6 showed, the nitrogen intake, apparent N intake and nitrogen in faeces were significantly affected by dietary CP level (P<0.05), these values in 36% CP groups were lower than those in 40% CP groups. MP substitution has significantly increased the nitrogen in faeces. The phosphorus intake, apparent P intake and phosphorus in faeces in 36% CP groups were lower than those in 40% CP groups. MP substitution has significantly decreased the phosphorus intake and apparent P intake.

No significant differences were observed on the nitrogen retention, NRR and phosphorous retention, PRR among all the groups. So, the gross nitrogen and phosphorus excretion rates were not affected by the dietary CP level and MP level.

Body,muscle and liver compositions

Table 7 showed that there were no significant differences on the whole body composition (crude protein, crude lipid, crude ash, moisture, gross energy and phosphorus) in Siberian sturgeon among all the experimental groups(P>0.05). The moisture and the crude lipid contents in muscle were significantly affected by MP whenever under the 40% crude protein or the 36% crude protein (P<0.05), the moisture contents of muscle were significantly increased in MPT group, and the crude lipid contents of muscle were significantly decreased in MPT groups (Table 8). No significant difference was observed in the liver composition (P>0.05).

Physiological responses

The results of activity of hepatic activity of amino acid catabolising enzymes (ALT, AST) and serum glucose (GLU), total protein (TP), triglyceride (TG) ,total cholesterol (TC), urea ammonia (UREA), lysozyme, superroxide dismutase, SOD activity and malondialdehyde, MDA contents were showed in Table 9. The plasma GH and IGF-I contents were also presented in Table 9.

MP replacement and CP levels did not affect the serum SOD, MDA, AST, ALT, ALP, UREA, GLU and plasma GH, IGF-I (P>0.05). The serum lysozyme activities were significantly decreased by MP substitution. Results showed irregularly effects on serum TP, TG and TCHO.

4. Discussion

Siberian sturgeon (*Acipense baeri* Brandt) may live for up to sixty years and the maximum weight recorded is 210kg, but usually weigh approximately 65kg (Anon, 2000). As one of the oldest fish species lives, its original spawning area has declined by up to 40% in some areas due to dam construction and increased abnormal oogenesis has been observed, probably due to chemical and nuclear water pollution. All production of Acipenseriformes species, including caviar, fertilized eggs and live fish were listed in CITES-list (Anon, 2000). Sturgeon aquaculture can be used as a tool not only for economic development to meet the market demand, but also for restocking and for the preservation of the gene pool of endangered sturgeon species (Chebanov, et al., 2002). In the last decade, sturgeon has not only been farmed for caviar, there flesh production has also been increased rapidly.

There have been a few studies on Siberian sturgeon feeding and nutrition undertaken by French teams (Médale, et al., 1995), but seldom concerned fish meal and fish oil substitution. Rónyai, et al. (2002) reported that Siberian sturgeon at least require 28% animal protein from fishmeal and/or meat meal. There were no significant difference on growth performance of fish when fed diet, in which 50% FM was replaced by full-fat soybean meal. According to the results of Palmegiano et al (2005), Siberian sturgeon grew better when fed diet with 50% and 75% FM replaced by *Spirulina*, a freshwater microalgae. Although *Spirulina* is considered to be a good source of protein and energy (Harel, et al., 2002), it is more expensive than FM. Therefore, it is almost impossible to be used for commercial least cost feed formula in some extent. Ng et al. (1996) reported that white sturgeon grew poorly when fed a diet with its intact protein substituted by crystalline amino acids (AA). Similar results were reported on common carp, *Cyprinus carpio* (Murai, et al., 1989) and channel catfish, *Ictalurus punctatus* (Robinson, et al., 1984).

Substitution of fishmeal with less expensive protein sources would be beneficial in reducing feed costs. Least cost formulation for high-energy diets should be based on meeting the optimal essential amino acids requirements by assorted various protein sources and/or supply with crystalline amino acids. The limitations for most alternative proteins were showed on lower digestibility (Luo et al., 2006), poorer amino acids profiles and palatability (El-Syaed 1990; Xue et al., 2004) etc. Generally, animal protein showed better palatability than vegetable protein for carnivorous fish species, and fish got higher growth performance when fed terrestrial animal protein used diets (Wang, et al., 2006, Guo, et al., 2007). Rendered by-products from slaughter houses, such as MBM, PBM, BM and HFM have been successfully used in feeds for various fish species. (Fowler, 1991; Bureau et al., 2000;

Kureshy et al., 1997; Allan et al., 2000 ; Millamena, 2002; Xue et al., 2003). Combination of various animal or plant ingredients, such as PBM and soybean meal (Quartararo et al., 2000), MBM and BM (Milliamena, 2002), soybean, cotton seed, sunflower and linseed meals (El-Saidy and Gaber, 2003), PBM, MBM and SBM (Goda, et al., 2007) and PM, MBM and BM (Guo, et al., 2007), has been demonstrated to be nutritionally adequate for many fish species.

In the present study, substitution of 75% or 100% fishmeal with combination of PBM, MBM, HFM and BM, in which the amino acids were balanced by commercial crystallized amino acid whether under dietary 40% CP or under dietary 36% CP, did not result in the decrease of the growth performance, feed efficiency, nitrogen retention and phosphorus retention, and did not affect the body, liver compositon and almost physiological indexes in Siberian sturgeon as well. As to the effects of dietary protein, Siberian sturgeon fed diets with the optimal protein level (40% CP) or the sub-optimal protein level (36% CP) showed the same growth performance, the nitrogen retention and phosphorus retention, but the FR in 36% CP was higher than that in 40% CP, and the feed efficiency in 36% CP was lower than that in 40% CP. This suggested that the dietary sub-optimal protein (36% CP) under ideal protein concept could satisfy the protein requirement of Siberian sturgeon by increasing the feeding rate, and the MP could be utilized by juvenile Siberia sturgeon up to 378.20g kg^{-1} to replace 100% of fishmeal protein under ideal amino acid concept with dietary 36%CP in this study.

Except for the reasons of high feeding frequency and amino acids balance, an extrusion processing was important for the high performance in this study. Even though pelleting feed is still the main products for most aquaculture species, commercial feed for sturgeon generally are cooked high-energy diet processed advanced extruder in market of China. Extrusion processing will improve the performance of alternative used feed by increasing digestibility and palatability (Cai, et al. 2005). In the present study, experimental diets were processed by a commercial extruder and starch gelatinization of all diets were higher than 80%, which quality is similar to the commercial feed in market. This provides evidence that Siberian sturgeon can accept terrestrial animal protein well and got satisfied growth performance when they ingest enough digestible nutrients, and a higher replacing level or total replacement can be applied in Siberian sturgeon diet.

The changes in the dietary IAA (indispensable amino acids) profile and DAA (dispensable amino acids) content were able to induce some state of liver GH resistance in conjunction with reduced growth rates (Gómez-Requeni et al., 2003), and the activity of the GH-liver axis was significantly affected by dietary protein sources (Gómez-Requeni et al., 2004).The GH-liver axis provides an integrated signal for growth and nutrient partitioning (Beckman and Dickhoff, 1998). Insulin-like growth factor-I (IGF-I) is involved in the GH negative feedback loop (Weil et al., 1999) and consistent changes in plasma GH levels occur in response to a shift in ration size and dietary protein/energy ratio (Pérez-Sánchez et al., 1999; Company et al., 1999). Accordingly, another FM substitution by fermented feather meal (FFM) experiment was conducted on Siberian sturgeon, except for the same parameters, plasma IGF-I of each group was determined and the results showed that Siberian sturgeon might possess function of adjusting the feed intake to get enough digestible amino acids and energy for growth when they were fed less quality protein sources. In this study, plasma level of IGF-I of fish fed MP was similar to that of FM group. We suppose that Siberian sturgeon might adjust the somatotropic axis responsiveness for compensation growth by adjusting feed intake when fed on alternative protein used diets.

Shi, et al (2006) compared plasma biochemistry of Amur sturgeon (*A. schrenckii*) and Chinese sturgeon (*A.sinensis*). The results showed that almost all parameters of Amur sturgeon were higher than those of Chinese sturgeon. The haematological index of Siberian sturgeon is similar to that of Amur sturgeon in the present study. Activity of hepatic enzymes of amino acid catabolism such as ALT and AST of Siberian sturgeon were relatively higher than most of teleostean (McDonald and Milligan, 1992; Svobodová, et al., 2006). Measurement of biochemical and physiological parameters is a commonly used diagnostic tool in aquatic toxicology and biomonitoring (McDonald and Milligan, 1992; Soimasuo, et al., 1995; Harikrishnan, et al., 2003). In the present study, dietary CP and MP did not significantly affected the biochemical and physiological parameters of the Siberian sturgeon. The ranges of the normal values of the key biochemical parameters are still undefined for different species in different aquaculture conditions. The normal values should be quantified for different species under different conditions, which will be important for diagnostication for fish health.

As to the body composition, the muscle lipid was significantly decreased by the mixed rendered animal protein. This was also proved in our previous study. The reason needs further research in the future.

In conclusion, the MP can be utilized by juvenile Siberia sturgeon up to 378.20g kg^{-1} to replace 100% of fishmeal protein under ideal amino acid concept with dietary 36%CP in this study.

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Table 1. Ingredients of the mixed protein (MP)

Ingredients	Ratio
.Meat and bone meal (beef)	35%
Poultry meal	40%
Feather meal(hydrolysed, ring dried)	5%
Blood meal(SDBM,Shunyi,Beijing)	20%

Table 2. Formulation and proximate composition of experimental diets ($g.kg^{-1}$, wet basis)

Ingredients	FM-40	MP75-40	MPT-40	FM-36	MP75-36	MPT-36
Fishmeal	483.00	121.00	0.00	400.00	100.00	0.00
Beer yeast	50.00	50.00	50.00	50.00	50.00	50.00
Squid meal	50.00	50.00	50.00	40.00	40.00	40.00
Mixed protein	0.00	342.00	451.00	0.00	283.00	378.20
Wheat flour	250.00	250.00	250.00	280.00	280.00	280.00
Wheat shorts	80.00	96.20	105.40	150.00	161.70	166.00
Soybean meal	0.00	0.00	0.00	0.00	0.00	0.00
Wheat gluten meal	0.00	0.00	0.00	0.00	0.00	0.00
Lecithin	20.00	20.00	20.00	20.00	20.00	20.00
Choline chloride	4.00	4.00	4.00	4.00	4.00	4.00
$Ca(H_2PO4)_2$	6.00	6.00	6.00	6.00	6.00	6.00
Premix	10.00	10.00	10.00	10.00	10.00	10.00
Lysine(65%)	0.00	3.80	4.60	0.00	2.50	3.80
Methionine	0.00	2.00	3.00	0.00	2.00	2.50
Threonine,	0.00	2.00	3.00	0.00	1.80	2.50
Fish oil	20.00	20.00	23.00	20.00	20.00	20.00
Soybean oil	27.00	23.00	20.00	20.00	19.00	17.00
Proximate analysis						
Moisture	90.53	87.83	91.03	90.79	90.47	95.27
Crude protein	406.29	412.80	407.53	367.51	368.63	369.26
Crude lipid	114.75	119.24	115.09	107.75	110.93	97.02
Gross energy (MJ/kg)	18.42	18.81	19.27	18.70	18.85	18.76
Total phosphorus	15.85	15.65	14.60	14.36	13.65	13.22
Ash	107.68	98.37	96.43	95.66	89.45	86.21
Y_2O_3	1.00	1.00	0.99	1.00	0.99	1.00

Table 3. Amino acids composition of the experimental diets (% of total amino acids)

	FM-40	MP75-40	MPT-40	FM-36	MP75-36	MPT-36
Indispensable amino acids,IAA						
Lysine,Lys	7.73	6.84	6.70	7.48	6.75	6.31
Threonine, Thr.	4.18	4.01	4.13	4.09	3.97	4.02
Leucine,Leu	8.12	8.18	8.63	8.01	8.39	8.47
Isoleucine,Ile	4.75	3.53	3.40	4.62	3.91	3.54
Arginine,Arg	6.11	6.45	6.01	6.06	6.06	6.54
Phenylalanine,Phe	4.83	5.04	5.12	5.00	5.05	5.15
Histidine,His	3.66	3.50	3.59	3.33	3.14	3.14
Methionine,Met	2.59	2.55	2.33	2.53	2.63	2.27
Valine,Val	5.54	5.91	5.84	5.59	6.00	5.95
Dispensable amino acids, DAA						
Asparagine, Asp	9.51	8.88	8.85	9.21	8.78	8.81
Serine, Ser	4.44	4.60	4.75	4.53	4.63	4.67
Glutamine, Glu	16.17	14.88	14.66	16.87	15.53	15.15
Glycine, Gly	6.24	7.79	8.20	6.09	7.56	7.93
Alanine, Ala	6.42	6.67	6.70	6.30	6.51	6.51
Cysteine,Cys	1.28	1.54	1.50	1.38	1.58	1.56
Proline,Pro	4.60	5.77	6.06	4.74	5.70	6.29
Tyrosine,Tyr	3.84	3.84	3.51	4.15	3.79	3.71
Total	100.00	100.00	100.00	100.00	100.00	100.00

Table 4. Effects of MP on the growth performance and feed efficiency in Siberian sturgeon fed with diets containing two levels of protein（Means ± S.E.）

	CP(%)	FM	MP75	MPT	CP	MP	Interaction
IBW(g)	40	39.01±0.03	38.95±0.02	39.03±0.03			
	36	38.94±0.02	39.00±0.07	38.98±0.01			
FBW(g)	40	104.26±4.12	105.81±5.25	99.46±4.87	ns	ns	ns
	36	104.82±4.48	91.86±6.15	98.18±3.97			
WGR(%)	40	267.28±10.77	267.86±16.24	253.64±11.25	ns	ns	ns
	36	266.79±12.72	234.46±15.47	251.92±10.28			
SGR(%)	40	2.32±0.05	2.34±0.08	2.26±0.06	ns	ns	ns
	36	2.33±0.05	2.16±0.09	2.24±0.05			
FR(%)	40	2.63±0.05	2.60±0.06	2.63±0.03	**	ns	ns
	36	2.81±0.07	2.72±0.03	2.73±0.03			
FCR	40	1.29±0.03	1.28±0.03	1.30±0.03	**	ns	ns
	36	1.38±0.06	1.42±0.04	1.38±0.04			
PER(%)	40	191.02±5.33	189.96±4.85	185.99±3.39	ns	ns	ns
	36	198.05±8.71	191.48±5.00	196.74±5.42			
NPR	40	27.36±0.58	26.30±1.59	25.26±0.82	ns	ns	ns
	36	27.05±1.46	26.48±0.26	26.19±0.87			
CF	40	0.53±0.02	0.51±0.02	0.52±0.01	ns	ns	ns
	36	0.53±0.01	0.52±0.02	0.53±0.03			
HSI	40	2.73±0.30^{b}	2.45±0.34^{ab}	2.24±0.20^{ab}	ns	ns	ns
	36	2.51±0.21^{ab}	2.28±0.22^{ab}	1.88±0.11^{a}			
VSI	40	6.52±0.36	6.42±0.32	6.25±0.12	ns	ns	ns
	36	6.93±0.52	6.13±0.44	6.57±0.28			

[1] *: $P<0.05$; **: $P<0.01$; ns: no significant difference, $P>0.05$

[2] WGR: Weight gain rate (%) = 100 × (FBW-IBW)/IBW

[3] SGR,Specific growth rate (% day) = 100 × [ln(FBW)-ln(IBW)]/feeding days, where IBW is initial body weight.

[4] FCR: feed conversion rate = feed consumption/fish weight gain

[5] FR, feeding rate = 100 × feed consumption/((IBW + FBW)/2)/feeding days

[6] NPR, net protein retention = 100 * (protein gain/protein comsumption)

[7] PER, protein efficiency ratio = body wet weight gain (g)/ dry protein intake (g)

[8] CF: Condition factor=100* (live weight,g)/body length^{3},cm

[9] HSI: Hepatosomatic index (%)=100%*liver weight/fish weight

[10] VSI: Viscerasomatic index (%) =100* (viscera weight)/body weight

Table 5. Apparent digestibility coefficients of dry matter (ADCd) , crude protein (ADCp) and gross phosphorus (ADCp) of 6 diets for Siberian sturgeon（Means ± S.E）

	CP(%)	FM	MP75	MPT	CP	MP	Interaction
ADCd	40	81.16±0.25	77.05±0.52	75.04±0.14	*	*	*
	36	76.32±0.47	73.65±0.45	74.61±0.06			
ADCp	40	90.49±0.28	88.69±0.38	88.67±0.29	ns	**	ns
	36	90.46±0.2	88.92±0.16	88.75±0.26			
ADCp	40	62.90±0.85	59.79±1.21	56.39±0.53	ns	**	ns
	36	60.58±0.47	58.94±0.70	57.41±0.77			

*: $P<0.05$; **: $P<0.01$; ns: no significant difference, $P>0.05$

Table 6. Effect of test diets on indexes of nitrogen and phosphorus discharge (g/tank)（Means ± S.E）

N/P indexes	CP(%)	FM	MP75	MPT	CP	MP	Interaction
Nitrogen intake	40	174.60±4.23	175.27±6.20	170.12 ± 5.10	**	ns	ns
	36	167.77±0.95	152.52±6.47	158.60 ± 2.86			
Apparent N intake	40	158.00±4.02	155.48±5.91	150.88±4.87	**	ns	ns
	36	151.77±1.11	135.64±5.92	140.79±3.10			
Nitrogen in faeces	40	16.60±0.51	19.79±0.67	19.24±0.46	**	**	ns
	36	16.01±0.40	16.87±0.57	17.81±0.35			
NRR (%)	40	27.36±0.58	26.30±1.59	25.26±0.82	ns	ns	ns
	36	27.05±1.46	26.48±0.26	26.19±0.87			
Phosphorus intake	40	42.56±1.03	41.54±1.46	38.10±1.14	**	**	ns
	36	40.97±0.23	35.30±1.50	35.48±0.64			
Apparent P intake	40	26.77±0.71	24.84±1.05	21.47±0.57	**	**	ns
	36	24.82±0.27	20.78±0.68	20.38±0.64			
P in faeces	40	15.80±0.56	16.70±0.76	16.62±0.62	*	ns	ns
	36	16.15±0.19	14.52±0.84	15.09±0.07			
PRR (%)	40	22.29±0.69	20.46±1.30	23.64±0.24	ns	ns	ns
	36	21.26±1.87	25.14±2.00	23.13±1.95			

[1] *: *P*<0.05; **: *P*<0.01; ns: no significant difference, *P*>0.05

[2] Apparent N intake (ANI)= Nitrogen intake×Nitrogen digestibility

[3] N in faeces(FN)= Nitrogen intake×(1- Nitrogen digestibility)

[4] NRR, net nitrogen retention rate = 100 * (nitrogen gain/ nitrogen comsumption)

[5] PRR, net phosphorus retention rate = 100 * (phosphorus gain/ phosphorus comsumption)

Table7. Effects of experimental diets on the body composition of Siberian sturgeon （Means ± S.E）

	CP(%)	FM	MP75	MPT	CP	MP	Interaction
CP(%)	40	13.68±0.33	13.68±0.07	13.24±0.33	ns	ns	ns
	36	13.43±0.11	13.39±0.17	13.02±0.13			
Ash(%)	40	2.68±0.10	2.45±0.04	2.57±0.05	ns	ns	ns
	36	2.44±0.10	2.74±0.21	2.43±0.08			
Moisture(%)	40	74.66±0.90	75.70±0.60	76.17±0.74	ns	ns	ns
	36	75.27±0.70	76.92±0.77	75.98±1.14			
Phosphorus(%)	40	0.46±0.02	0.43±0.01	0.46±0.00	ns	ns	ns
	36	0.44±0.02	0.48±0.03	0.43±0.02			
Gross energy (MJ/kg)	40	6.54±0.38	6.13±0.21	5.84±0.21	ns	ns	ns
	36	6.29±0.30	5.55±0.30	6.10±0.44			
Crude lipid(%)	40	8.31±1.03	7.39±0.60	7.13±0.36	ns	ns	ns
	36	8.31±0.88	6.19±0.70	7.76±1.04			

*: $P<0.05$; **: $P<0.01$; ns: no significant difference, $P>0.05$

Table 8. Effects of experimental diets on the muscle and liver compositions in Siberian sturgeon（Means ± S.E）

		CP(%)	FM	MP75	MPT	CP	IP	Interaction
Muscle	CP(%)	40	16.45±0.08	16.33±0.10	16.25±0.23	ns	ns	ns
		36	16.18±0.15	16.34±0.22	15.74±0.31			
	Moisture(%)	40	74.03±1.14a	74.80±0.91a	76.55±0.81b	ns	*	ns
		36	75.04±0.58a	75.11±0.45a	77.42±0.66b			
	Crude lipid(%)	40	8.63±1.19a	8.23±0.84a	6.40±0.85b	ns	*	ns
		36	7.56±0.54a	7.64±0.55a	5.76±0.45b			
Liver	Moisture(%)	40	53.59±2.70	57.34±2.29	55.85±1.65	ns	ns	ns
		36	55.66±1.70	53.77±1.53	58.32±2.21			
	Crude lipid(%)	40	31.48±2.72	27.99±1.73	30.06±1.73	ns	ns	ns
		36	28.47±2.04	32.12±1.96	27.57±2.65			

*: *P*<0.05; **: *P*<0.01; ns: no significant difference, *P*>0.05

Table 9. Effects of experimental diets on the physiological responses in Siberian sturgeon （Means ± S.E）

	CP(%)	FM	MP75	MPT	CP	MP	Interac
LZM（U/ml ）	40	100.08±13.82	57.23±11.27	75.99±10.62	ns	*	ns
	36	84.90±21.52	57.00±5.39	60.49±3.56			
SOD（U/ml)	40	134.22±13.00	124.65±13.23	133.45±15.61	ns	ns	ns
	36	127.15±4.64	136.50±7.88	110.94±7.02			
MDA（nmol/ml)	40	16.80±3.23	34.32±16.51	22.64±11.51	ns	ns	ns
	36	16.70±3.39	23.43±7.58	11.96±1.91			
hGH(ng/ml)	40	0.79±0.25	0.77±0.15	0.74±0.13	ns	ns	ns
	36	0.74±0.21	0.73±0.12	0.72±0.23			
IGF-I(ng/ml)	40	15.12±2.34	6.67±2.16	14.30±3.03	ns	ns	ns
	36	9.63±2.77	11.22±3.09	15.65±6.36			
AST(U/L)	40	228.25±16.94	218.25±21.81	219.00±9.37	ns	ns	ns
	36	221.00±25.94	170.00±7.22	212.25±22.56			
ALT(U/L)	40	5.50±1.94	6.75±2.43	5.25±0.48	ns	ns	ns
	36	7.25±2.01	6.75±2.43	6.25±2.59			
TP(g/L)	40	15.25±0.75	14.25±1.25	13.50±0.50	ns	**	**
	36	18.25±0.95	10.50±0.65	14.00±0.91			
ALP(U/L)	40	181.25±34.44	159.25±11.65	157.75±10.40	ns	ns	ns
	36	199.00±15.11	129.75±5.34	158.50±22.19			
TG(mmol/L)	40	4.52±0.74	8.25±1.06	4.33±0.40	ns	*	**
	36	5.17±0.39	4.46±0.29	4.35±0.50			
TC(mmol/L)	40	3.11±0.15	3.53±0.37	2.68±0.06	*	ns	**
	36	2.74±0.09	2.30±0.04	2.94±0.28			
UREA(mmol/L)	40	0.28±0.03	0.25±0.03	0.33±0.09	ns	ns	ns
	36	0.38±0.03	0.33±0.09	0.30±0.04			
GLU(mmol/L)	40	3.93±0.21	3.73±0.30	3.88±0.13	ns	ns	ns
	36	4.38±0.45	3.40±0.27	4.18±0.20			

*: $P<0.05$; **: $P<0.01$; ns: no significant difference, $P>0.05$