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Apparent Digestibility of Nutrients in Six Protein Ingredients by Juvenile Huso dauricus (Postprint)

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Abstract

To investigate the digestive capacity of juvenile Huso dauricus for different protein ingredients, experimental diets were formulated using 0.4% titanium dioxide (TiO) as an indicator by mixing six protein ingredients—fish meal, poultry meal, meat and bone meal, feather meal, canola meal, and corn gluten meal —with a basal diet at a 3:7 ratio, and the apparent digestibility of dry matter, crude protein, crude lipid, gross energy, and total amino acids of these six protein ingredients for juvenile Huso dauricus was determined. A total of 420 juvenile Huso dauricus with an initial body weight of (66.79±2.18) g were selected and randomly divided into 7 groups with 3 replicates per group and 20 fish per replicate. After being fed the basal diet for 2 weeks, the fish were fed the experimental diets; feces were collected using a scoop net after 1 week of feeding the experimental diets for a total of 10 days. The results showed that the apparent digestibility of dry matter, crude protein, crude lipid, gross energy, and amino acids in the six protein ingredients for juvenile Huso dauricus ranged from 54.79%-88.07%, 73.62%-89.47%, 99.80%-100.74%, 66.64%-89.24%, and 51.27%-98.62%, respectively. Among the six protein ingredients, the apparent digestibility of crude lipid was above 99% for all ingredients, while the apparent digestibility of dry matter, crude protein, and gross energy was higher for fish meal and poultry meal, moderate for corn gluten meal and canola meal, and lower for meat and bone meal and feather meal. Specifically, fish meal had the highest crude protein apparent digestibility at 89.47%, which was significantly higher than that of other protein ingredients ($P<0.05$); poultry meal ranked second at 81.14%, showing no significant difference from meat and bone meal, canola meal, and corn gluten meal ($P>0.05$); feather meal had the lowest crude protein apparent digestibility at 73.62%, which showed no significant difference from meat and bone meal ($P>0.05$) but was significantly different from other protein ingredients ($P<0.05$). The apparent digestibility of total amino acids in

each protein ingredient showed a basically consistent trend with that of crude protein. Therefore, for juvenile Huso dauricus diets, fish meal is the optimal protein source, while poultry meal, corn gluten meal, and canola meal can also serve as quality protein sources; however, the inclusion levels of feather meal and meat and bone meal should be limited when used as protein sources.

Full Text

Nutrient Apparent Digestibility in Six Protein Ingredients for Juvenile Kaluga (Huso dauricus)

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Abstract

To evaluate the digestive capacity of juvenile kaluga (Huso dauricus) for different protein ingredients, test diets were formulated by replacing 30% of a basal diet with each of six protein sources: fish meal, chicken meal, meat and bone meal, feather meal, canola meal, and corn gluten meal, using 0.4% titanium dioxide (TiO) as an inert marker. The apparent digestibility coefficients (ADCs) of dry matter, crude protein, crude lipid, gross energy, and total amino acids were determined. A total of 420 juvenile kaluga with an initial body weight of (66.79 ± 2.18) g were randomly allocated into seven groups with three replicates each (20 fish per replicate). After a two-week acclimation period feeding the basal diet, fish were fed the test diets for one week, followed by a 10-day fecal collection period using a fine-mesh net. The results showed that ADCs for dry matter, crude protein, crude lipid, gross energy, and total amino acids ranged from 54.79% to 88.07%, 73.62% to 89.47%, 99.80% to 100.74%, 66.64% to 89.24%, and 51.27% to 98.62%, respectively. Among the six protein ingredients, crude lipid ADCs exceeded 99% across all treatments. Fish meal and chicken meal exhibited higher ADCs for dry matter, crude protein, and gross energy, followed by corn gluten meal and canola meal, while feather meal and meat and bone meal showed lower values. Fish meal achieved the highest crude protein ADC at 89.47%, significantly greater than all other ingredients ($P < 0.05$). Chicken meal ranked second at 81.14%, which did not differ significantly from meat and bone meal, canola meal, or corn gluten meal ($P > 0.05$). Feather meal had the lowest crude protein ADC at 73.62%, which was not significantly different from meat and bone meal ($P > 0.05$) but differed significantly from the other ingredients ($P < 0.05$). The ADCs for total amino acids followed a similar trend to those for crude protein. These findings indicate that fish meal is the optimal protein source for juvenile kaluga diets, while chicken meal, corn gluten meal, and canola meal can serve as quality protein sources. However, the inclusion levels of feather meal and meat and bone meal should be carefully controlled when used as dietary protein sources.

Keywords: juvenile kaluga (Huso dauricus); protein ingredient; nutrient; apparent digestibility

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Introduction

Kaluga (Huso dauricus) is one of two species in the genus Huso within the family Acipenseridae and one of two indigenous sturgeon species in the Heilongjiang River basin of China. The hybrid "Sturgeon Dragon No. 1,"produced by crossing female kaluga with male Amur sturgeon (Acipenser schrenckii), represents a major cultured sturgeon variety in China. While several studies have investigated the dietary requirements of sturgeons for protein, lipid, carbohydrate, vitamins, and minerals, these efforts remain insufficient to meet industry demands. Current commercial sturgeon feed formulations primarily reference those developed for eels and soft-shelled turtles, lacking essential fundamental research, and complete nutritional profiles are unavailable for any sturgeon species. Consequently, research on sturgeon nutrition and feed development has become increasingly urgent.

Sturgeons are carnivorous fish requiring dietary crude protein levels around 40% for grow-out, with even higher levels for broodstock and larvae. Identifying suitable animal and plant protein sources is therefore critical. Determining apparent digestibility coefficients (ADCs) of nutrients in feed ingredients is a key method for evaluating nutritional value and formulating balanced, cost-effective aquafeeds. This approach is essential for optimizing ingredient inclusion rates, improving feed utilization efficiency, reducing environmental pollution from aquaculture operations, and minimizing production costs. Previous studies on protein ingredient digestibility in sturgeons include reports on hybrid sturgeon (A. gueldenstaedti × A. baerii) by Li et al., Siberian sturgeon (A. baerii) by Liu et al., and Caspian great sturgeon (Huso huso) subadults by Safari et al. However, no studies have examined nutrient digestibility in juvenile kaluga. Therefore, this study investigated the ADCs of nutrients in six protein sources for juvenile kaluga to provide theoretical support for developing environmentally sustainable sturgeon feeds.

1.1 Test Protein Ingredients and Experimental Diets

Six protein ingredients were selected: fish meal, chicken meal, meat and bone meal, feather meal, canola meal, and corn gluten meal. Fish meal (Peru), feather meal (origin unspecified), canola meal (Canada), and corn gluten meal (Henan) were obtained from Wuhan DaBeiNong Aquatic Technology Co., Ltd., while chicken meal (USA) and meat and bone meal (Australia) were sourced from Zhejiang Xinglongma Feed Co., Ltd. Their nutritional levels and amino acid compositions are presented in Table 1 .

A basal diet was formulated with 0.4% TiO as an inert marker, and its composition and nutritional levels are shown in Table 2 . All ingredients were ground to pass through a 60-mesh sieve, mixed thoroughly using a stepwise dilution method, and the basal diet was prepared. Test diets were formulated by replacing 30% of the basal diet with each protein ingredient. Specifically, 70% of the TiO -supplemented basal diet was combined with 30% of the test ingredient, mixed thoroughly, and approximately 20% water was added before extrusion into 3 mm diameter pellets using a small meat grinder. The pellets were air-dried and stored at -20 °C until use.

1.2 Experimental Fish and Culture Conditions

Kaluga fertilized eggs were obtained from naturally spawning wild broodstock in the Fuyuan section of the Heilongjiang River, transported to the Taihu Base of the Yangtze River Fisheries Research Institute in Jingzhou for incubation and rearing. Juvenile fish meeting the experimental size requirements were selected and randomly distributed into 21 flow-through tanks (105 cm diameter, 0.43 m^3 volume) at 20 fish per tank. A total of 420 healthy juvenile kaluga with uniform size and initial body weight of (66.79 ± 2.18) g were allocated into seven groups with three replicates each. After stocking, fish were fed the basal diet twice daily (08:00 and 15:00) to apparent satiation for two weeks, followed by the test diets for one week. Fecal collection began one week after initiating the test diets and continued for 10 days. The experiment was conducted under natural photoperiod with filtered groundwater. Water temperature ranged from 18.2 to 20.0 °C, dissolved oxygen concentration was   5 mg/L, and pH was 7.8–8.2.

1.3 Fecal Collection

Residual feed and feces were siphoned from tanks 0.5 h after each morning and afternoon feeding. Intact, well-formed feces were collected with a fine-mesh net between 10:00–13:00 and 17:00–18:00 daily and placed in labeled petri dishes, then frozen at -20 °C. After sufficient quantities were collected, samples were freeze-dried for approximately 72 h, ground, and stored at -40 °C until analysis.

1.4 Index Determination and Calculation

Crude protein, crude lipid, and ash contents in diets and feces were determined using the Kjeldahl method (GB/T 5009.5-2003), Soxhlet extraction (GB/T 5009.6-2003), and gravimetric analysis after incineration (GB/T 5009.4-2003), respectively. Gross energy was measured directly using an oxygen bomb calorimeter (Parr-6200). Amino acid content was analyzed using an amino acid analyzer (Agilent-1260) according to GB/T 5009.124-2016. TiO content was determined using the method described in reference [11], and dietary phosphorus content was measured by colorimetry after acid digestion (GB/T 5009.87-2016).

The ADCs of crude protein, crude lipid, amino acids, and gross energy in diets (including basal and test diets) were calculated using Equation (1), while dry matter ADC was calculated using Equation (2):

$$\text{ADC}_d(\%) = \left[1 - \left(\frac{P_f}{P_d}\right) \times \left(\frac{R_d}{R_f}\right)\right] \times 100$$

$$\text{ADC}_{dm}(\%) = \left[1 - \left(\frac{R_d}{R_f}\right)\right] \times 100$$

where ADCd is the ADC of crude protein, crude lipid, amino acids, or gross energy (%); ADCdm is the ADC of dry matter (%); Pf is the content (mass fraction, %) of crude protein, crude lipid, or amino acids, or gross energy (MJ/kg) in feces; Pd is the corresponding content in test diets; Rd is the TiO content (mass fraction, %) in test diets; and Rf is the TiO content (mass fraction, %) in feces.

The ADCs of nutrients in the six test ingredients were calculated using Equation (2):

$$\text{ADC}_i = \text{ADC}_t + \left[(\text{ADC}_t - \text{ADC}_r) \times \frac{(0.7 \times E_r)}{(0.3 \times E_i)}\right]$$

where ADCi is the ADC (%) of dry matter, crude protein, crude lipid, amino acids, or gross energy in the test ingredient; ADCt is the ADC (%) of the test diet containing 30% test ingredient, calculated using Equation (2); ADCt is the ADC (%) of the basal diet, calculated using Equation (1); Er is the content (mass fraction, %) of dry matter, crude protein, crude lipid, amino acids, or gross energy in the basal diet; and Ei is the corresponding content in the test ingredient.

1.5 Data Processing and Analysis

Results are expressed as mean ± standard deviation. All data were analyzed using SPSS 22.0 software. One-way ANOVA followed by Duncan' s multiple comparison test was applied, with P < 0.05 indicating significant differences among groups.

Results

The ADCs of dry matter, crude protein, crude lipid, and gross energy in the six protein ingredients for juvenile kaluga are presented in Table 3 . Dry matter ADCs ranged from 54.79% to 88.07%, with fish meal showing the highest value, followed by chicken meal (>80%), both significantly higher than meat and bone meal and canola meal ($P < 0.05$). Meat and bone meal had the lowest dry matter ADC, significantly different from all other ingredients ($P < 0.05$). Feather meal had a dry matter ADC of 72.35%, which did not differ significantly from chicken meal, canola meal, or corn gluten meal ($P > 0.05$).

Crude protein ADCs exceeded 73% for all ingredients, with fish meal achieving the highest value at 89.47%, significantly greater than all other ingredients ($P < 0.05$). Chicken meal ranked second at 81.14%, showing no significant difference from meat and bone meal, canola meal, or corn gluten meal ($P > 0.05$). Feather meal had the lowest crude protein ADC at 73.62%, which was not significantly different from meat and bone meal ($P > 0.05$) but differed significantly from the other ingredients ($P < 0.05$).

Gross energy ADCs ranged from 66.64% to 103.18%, with fish meal and chicken meal showing the highest values (>89%), significantly greater than meat and bone meal and feather meal ($P < 0.05$). Feather meal had the lowest gross energy ADC at 66.64%, significantly lower than fish meal, chicken meal, and corn gluten meal ($P < 0.05$), but not significantly different from meat and bone meal or canola meal ($P > 0.05$). Crude lipid ADCs exceeded 99% for all ingredients, with no significant differences among them ($P > 0.05$); canola meal showed the highest value at 100.74%.

The amino acid ADCs in the six protein ingredients are shown in Table 4 . Amino acid ADCs followed similar trends to crude protein ADCs. Fish meal exhibited the highest total amino acid ADC at 98.62%, significantly different from all other ingredients ($P < 0.05$). Feather meal had the lowest total amino acid ADC at 51.27%, significantly different from all other ingredients ($P < 0.05$). Chicken meal, canola meal, and corn gluten meal showed intermediate values, with no significant differences among them ($P > 0.05$), but all were significantly higher than meat and bone meal ($P < 0.05$).

3.1 Selection of External Indicator and Calculation Method

This study adopted the method of Cho et al. [12] to formulate test diets by replacing 30% of the basal diet with test ingredients. However, the calculation method of Cho et al. [12] did not account for nutrient contributions from both the basal diet and test ingredients. Therefore, we employed the improved calculation method developed by Bureau et al. [13-14] based on Cho' s approach [Equation (2)], which reduces errors caused by nutrient differences between basal and test diets, thereby improving accuracy.

Indirect determination of nutrient utilization in feed ingredients based on marker concentrations in feces and diets is a common method for studying feed digestibility. Common indicators include TiO , yttrium oxide, titanium dioxide, and acid-insoluble ash. TiO offers several advantages: chemical stability, insolubility in water and dilute acids, safety at intake levels of 2–3 g/d, complete excretion in feces (>95% recovery), low recovery variability, no alteration of feed color, non-carcinogenicity, legal approval for use, high detection sensitivity, and low required supplementation (0.2%-0.5%) [15-16]. Consequently, TiO was selected as the indicator in this study to ensure data accuracy.

3.2 Nutrient Apparent Digestibility of Six Protein Ingredients for Juvenile Kaluga

Dry matter digestibility reflects overall feed utilization efficiency in fish and is influenced by crude fiber and ash contents as well as nutrient (protein and lipid) digestibility. Dry matter ADCs for the six protein ingredients ranged from 54.79% to 88.07%, with fish meal and chicken meal showing higher values, feather meal, canola meal, and corn gluten meal showing intermediate values, and meat and bone meal showing the lowest value (54.79%). Previous studies reported similarly low dry matter ADCs for meat and bone meal in hybrid sturgeon [8] and Siberian sturgeon [9], consistent with our results, likely due to high ash content in meat and bone meal. Kitagima et al. [17] reported low dry matter ADCs for high-ash fish meal and shrimp byproduct meal in channel catfish (Ictalurus punctatus), with similar findings reported for hybrid tilapia [18], rainbow trout [13], and Amur catfish [19].

Energy digestibility reflects the overall availability of dietary protein, lipid, and carbohydrate. Gross energy ADCs showed similar trends to dry matter ADCs. Fish meal (with a measured ADC >100%) and chicken meal exhibited higher gross energy ADCs, followed by canola meal and corn gluten meal, while meat and bone meal and feather meal showed lower values. The lower dry matter and gross energy ADCs for canola meal and corn gluten meal compared to fish meal align with findings in Amur catfish [19], large yellow croaker [20], and Caspian great sturgeon [10], likely because plant proteins contain higher proportions of cellulose, which fish cannot digest due to lack of cellulolytic enzymes [21], and high cellulose content (>8%) may reduce dry matter and energy digestibility [22]. Dry matter and gross energy ADCs for feather meal (72.35% and 66.64%, respectively) were comparable to values reported for hybrid sturgeon [8], Siberian sturgeon [9], and blunt snout bream [23].

Crude lipid ADCs exceeded 99% for all ingredients, indicating strong lipid utilization capacity in juvenile kaluga. These results are consistent with reports of 95.7% and 103.4% lipid ADCs for expanded feather meal and corn gluten meal in blunt snout bream [23], 94.07% and 91.01% for fish meal and meat and bone meal in Amur catfish [19], and 98.1% for fish meal in Caspian great sturgeon subadults [10]. However, lipid ADCs for corn gluten meal and canola meal were higher in juvenile kaluga than in Caspian great sturgeon subadults [10] and

Amur catfish [19].

Protein quality is the primary factor affecting fish nutrition, and protein digestibility is a crucial indicator of ingredient utilization [24]. Crude protein ADCs exceeded 73% for all ingredients, with fish meal and chicken meal achieving 89.47% and 81.14%, respectively, higher than canola meal (79.52%) and corn gluten meal (79.52%). This may be attributed to: (1) imbalanced essential amino acid profiles in plant proteins [25], and (2) antinutritional factors in plant proteins that impair digestion and absorption [26-27]. The crude protein ADCs of approximately 80% for canola meal and corn gluten meal indicate good digestibility in juvenile kaluga. Sullivan et al. [28] suggested that although plant protein ingredients generally have lower crude protein content than animal sources, many can be effectively utilized by carnivorous and omnivorous fish. Therefore, canola meal and corn gluten meal can serve as alternative protein sources in juvenile kaluga diets. The crude protein ADC for feather meal (73.62%) was lower than that reported for Siberian sturgeon (88.2%) [9], similar to Caspian great sturgeon (75%) [10], but higher than hybrid sturgeon (50.54%) [8]. The crude protein ADC for meat and bone meal (76.24%) was lower than values reported for hybrid sturgeon (84.95%) [8] and Siberian sturgeon (83.9%) [9], but similar to another report for hybrid sturgeon (77.54%) [8]. These discrepancies may result from genetic differences in digestive capacity among sturgeon species [8] and variations in ingredient processing methods and sources, which can affect nutrient composition and digestibility.

Amino acid ADCs showed similar trends to crude protein ADCs across ingredients. Additionally, substantial differences were observed for the same amino acid among different ingredients, and among different amino acids within the same ingredient, consistent with previous reports [23,29]. Total amino acid ADCs exceeded 80% for four of the six ingredients (excluding feather meal and meat and bone meal), indicating effective utilization of most amino acids. The lower total amino acid ADCs for feather meal and meat and bone meal (<70%) may be related to ingredient source, freshness, nutritional composition, and processing methods.

Conclusion

In summary, juvenile kaluga exhibited the highest ADCs for dry matter, crude protein, crude lipid, gross energy, total amino acids, and essential amino acids in fish meal, followed by chicken meal, canola meal, and corn gluten meal, with feather meal and meat and bone meal showing the lowest values. These results demonstrate that chicken meal, canola meal, and corn gluten meal can serve as quality animal and plant protein sources for juvenile kaluga. Due to variable ingredient quality and processing methods affecting nutrient composition, the inclusion levels of feather meal and meat and bone meal should be carefully controlled and consideration given to freshness, source, and nutritional composition when formulating diets for juvenile kaluga.

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