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饲料中豆粕替代鱼粉蛋白对异育银鲫生长、代谢及免疫功能的影响

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摘要:本实验评价了饲料中豆粕替代鱼粉蛋白后对异育银鲫的生长、饲料利用、氮代谢和鱼体免疫力等的影响。实验设计 4种等氮等能的饲料,每种 3个重复,分别以豆粕替代饲料中鱼粉蛋白的 0(对照, D1)、20% (D2)、80% (D3)和 100% (D4)。实验在半循环水养殖系统持续 16周,鱼的初重约 2.32g,实验期间水温 23—30℃。结果表明,随着饲料中豆粕含量的升高,摄食率显著升高 ($p<0.05$),特定生长率、饲料转化效率、蛋白沉积率和能量沉积率显著降低 ($p<0.05$);蛋白表观消化率显著升高,干物质和能量表观消化率则显著降低 ($p<0.05$);总氮摄入量、表观氮摄入量、粪氮排出量、非粪氮排泄量、总氮沉积率均随着饲料中豆粕含量的升高呈显著降低的趋势 ($p<0.05$),生产每千克鱼的氮排放量则随着饲料中豆粕含量的升高显著升高 ($p<0.05$);血清葡萄糖和甘油三酯的含量显著升高,而胆固醇的含量显著降低 ($p<0.05$);血清的溶菌酶显著降低,超氧化物歧化酶逐渐升高 ($p<0.05$)。

关键词:豆粕; 鱼粉;异育银鲫

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鱼粉的蛋白含量高,各种氨基酸平衡性好,是水产饲料中最主要的蛋白源,但由于资源短缺,其价格一直居高不下。目前,主要可以从以下两方面减少鱼粉在饲料中的用量:一是利用蛋白质节约效应,即以脂肪或碳水化合物等非蛋白物质部分替代用于供能的蛋白质[1,2];另一方面是利用廉价的蛋白源。在过去的几十年里,很多研究报道饲料中鱼粉蛋白可以被一些价格低廉的蛋白源部分或完全替代[3,4]。其中,与动物蛋白源相比,植物蛋白源的来源更加广泛。已有的研究结果显示,水产饲料中 30%—50%的鱼粉可以被植物蛋白替代[5]。

豆粕是植物蛋白中来源最广泛的饲料原料,蛋白含量比较高,已经在水产饲料中广为应用。但是豆粕中的胰蛋白酶抑制因子和血球凝集素等抗营养因子以及氨基酸的不平衡性和可利用性限制其在鱼类饲料中的用量。一些研究报道豆粕或大豆产物可以部分或完全替代饲料中的鱼粉[6,7]。也有研究表明,全脂大豆或浸提豆粕会抑制生长,降低饲料效率[8]。豆粕中的抗营养因子会损伤肠道[9],影响肠道内的微生态环境[10],抑制消化酶的活性[11],影响营养物质的消化吸收。本实验的目的是探讨饲料中豆粕替代不同水平的鱼粉后对异育银鲫的生长、饲料利用、消化率、氮代谢和鱼体免疫力的影响。

1 材料与方法

1.1 养殖系统 实验在 12个塑料水族箱 (60cm × 40 cm ×50 cm,水体积: 120L)组成的循环水系统中进行,每个缸的水流速为 10L/min。每天测一次水温,每周测一次水体溶氧和氨氮。实验期间,水温在 23—30℃范围内波动;氨氮浓度小于 0.5mg/L;溶氧高于 5mg/L;余氯小于 0.05 mg/L; pH约为 6.4;光照周期控制为 12L/12D,光亮周期为 08:00到 20:00。

1.2 实验饲料与实验设计 实验以豆粕和鱼粉为蛋白源,豆粕是经溶剂浸出法制得,购自武汉当地市场;鱼粉是美国进口的白鱼粉。实验设计四种等氮(蛋白水平为 38%)等能 (能值为 18 kJ/g)的饲料,以豆粕蛋白分别替代 0(对照, D1)、20% (D2)、80% (D3)和 100% (D4)的鱼粉蛋白,每种饲料 3个重复。饲料中添加 1% Cr_2O_3作为表观消化率测定的指示剂。饲料配方如表 1所示。饲料原料过四十目后充分混匀,经饲料机制成直径为 2mm的颗粒, 60℃烘干后,置于冰箱中备用。

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表 1 饲料配方和化学组成(% 干物质)

Tab. 1 Formulation and chemical composition of the experimental diets (% dry matter)

成分 Ingredients	D1	D2	D3	D4
替代水平 Replacement level (%)	0	20	80	100
鱼粉 Fishmeal	53.06	42.45	21.23	0.00
豆粕 Soybean meal	0.00	14.11	42.33	70.55
玉米淀粉 Corn starch	31.42	26.18	15.71	15.24
鱼油 Fish oil	3.00	3.80	5.50	7.20
维生素预混物 Vitamin premix[1]	0.45	0.45	0.45	0.45
氯化胆碱 Choline chloride	0.11	0.11	0.11	0.11
矿物盐 Mineral premix[2]	5.00	5.00	5.00	5.00
三氧化二铬 Chromic oxide	1.00	1.00	1.00	1.00
羧甲基纤维素 Carboxymethyl cellulose	2.00	2.00	2.00	2.00
纤维素 Cellulose	3.96	4.90	6.67	8.45
生化成分 Chemical composition				
干物质 Dry matter	95.69	95.56	95.58	96.20
粗蛋白 Crude protein	37.32	37.46	37.53	38.05
粗脂肪 Crude lipid	7.17	7.00	7.09	7.90
灰分 Ash	13.22	12.44	11.13	12.56
总能 Gross energy (kJ/g)	18.00	17.60	18.30	17.92
Lys/CP(%)	7.76	7.53	7.08	6.62
Met/CP (%)	4.55	4.09	3.19	2.28

注: [1]维生素预混物(mg/kg饲料):维生素 B_1, 20;维生素 B_2, 20;维生素 B_6, 20;维生素 B_{12}, 0.020;叶酸, 5;泛酸钙, 50;肌醇, 100;烟酸, 100;生物素, 0.11;淀粉, 3226;维生素 C, 100;维生素 A, 110;维生素 D, 20;维生素 E, 50;维生素 K, 10;胆碱, 550; [2]无机盐预混物(mg/kg饲料):氯化钠, 500;硫酸镁, 4575.0;磷酸二氢钠, 12500.0;磷酸二氢钾, 16000.0;磷酸二氢钙, 6850.0;硫酸亚铁, 1250.0;乳酸钙, 1750.0;硫酸锌, 111.0;硫酸锰, 61.4;硫酸铜, 15.5;硫酸钴, 0.5;碘化钾, 1.5;淀粉, 6385.1

Note: [1]Vitamin premix (mg/kg diet): thiamin, 20; riboflavin 20; pyridoxine, 20; cyanocobalamine, 0.020; folic acid, 5; calcium patotheniate, 50; inositol, 100; niacin, 100; biotin, 0.11; Starch, 3226; ascorbic acid, 100; Vitamin A, 110; Vitamin D, 20; Vitamin E, 50; Vitamin K, 10; Choline chloride, 550; [2]Mineral premix (mg/kg diet): NaCl, 500; $MgSO_4 \cdot 7H_2O$, 4575.0; $NaH_2PO_4 \cdot 2H_2O$, 12500.0; KH_2PO_4, 16000.0; $Ca(H_2PO_4)_2 \cdot H_2O$, 6850.0; $FeSO_4$, 1250.0; $C_6H_{10}CaO_6 \cdot 5H_2O$, 1750.0; $ZnSO_4 \cdot 7H_2O$, 111.0; $MnSO_4 \cdot 4H_2O$, 61.4; $CuSO_4 \cdot 5H_2O$, 15.5; $CoSO_4 \cdot 6H_2O$, 0.5; KI, 1.5; Starch, 6385.1

1.3 实验鱼与饲养实验 异育银鲫为中国科学院水生生物研究所关桥渔场的当年鱼苗。实验鱼在室内水泥池中暂养两周后转入实验系统用四种实验饲料的混合料暂养 4周,每天 09:00和 15:00饱食投喂两次。实验开始时,实验鱼饥饿 24h,选取体质健康、规格均匀的个体(平均尾重约 2.32g)随机放入 12个鱼缸中,每缸 40尾。生长实验持续 16周。实验期间,每天 09:00和 15:00饱食投喂两次,投喂后 1h收集残饵, 60℃烘干称重用以计算摄食量;每天搜集新鲜完整的粪样, 60℃烘干后置冰箱保存留待分析测试。实验结束后,在所有的实验鱼缸中分别投放对应的已称重的饲料, 1h后回收饲料, 60℃烘干称重计算饲料的溶失率,校正摄食量。

1.4 实验取样 实验开始时,取 50尾鱼称重后 -20℃保存用作体成分分析。在实验结束时,将实验鱼饥饿、称重、取样,每缸取 3尾鱼分析生化成分;每缸取 6尾鱼,尾静脉取血,以 3500 r/min的转速离心 15min,取上层的血清备用。

1.5 样品分析 实验饲料和鱼体的干物质、粗蛋白、粗脂肪、灰分、总能及粪样的粗蛋白和总能的测定参照文献[12]的方法进行测定:干物质在 105℃烘干至恒重,通过失重法测定;粗蛋白采用 FOSS定氮仪(2300 Kjeltec Analyzer Unit)测定;粗脂肪采用索氏抽提仪(Soxtec system HT6, Tecator, Hoganas, Sweden)进行测定;灰分在马福炉中 550℃燃烧 3h,失重法测定;能值通过氧弹测热仪(Philipson microbomb calorimeter, Gentry Instruments Inc., Aiken, USA)测定。饲料和粪样中 Cr_2O_3的测定参照 Bolin 等[13]的方法。

1.6 血清生理指标的测定 血清葡萄糖采用葡萄糖氧化酶法[14];甘油三酯采用酶法[15];血清的胆固醇采用胆固醇氧化酶法[16];超氧化物歧化酶(SOD)活性的测定参照 McCord & Fridovich[17]的方法测定;血清溶菌酶采用比浊法[18]。

1.7 数据统计分析 用统计软件 Statistica 6.0进行统计分析。实验结果经过一元方差分析(One-way ANOVA)后,进行 Duncan's多重比较各实验组间差异的显著性, $p < 0.05$表示差异显著。

2 结　果

2.1 生长性能

实验结果如表 2所示,饲料中豆粕替代不同水平的鱼粉蛋白后,显著影响异育银鲫的摄食率、特定生长率、饲料转化效率、蛋白沉积率和能量沉积率($p < 0.05$)。随着饲料中豆粕含量的增多,异育银鲫的摄食率显著升高($p < 0.05$), D4的摄食率显著高于其他处理组($p < 0.05$);异育银鲫的特定生长率显著降低($p < 0.05$), D4的特定生长率显著低于其他处理组($p < 0.05$);饲料转化效率、蛋白沉积率均与特定生长率的变化趋势相同;随着饲料中豆粕含量的增多,能量沉积率显著降低($p < 0.05$), D1

和 D2的能量沉积率无差异 ($p > 0.05$),但 D1和 D2的能量沉积率要显著高于 D3和 D4 ($p < 0.05$)。

表 2 饲料中豆粕替代鱼粉蛋白对异育银鲫生长和饲料利用的影响 (平均值 ±标准误) *

Tab. 2 Effects of soybean meal replacing dietary fishmeal protein on growth and feed utilization in gibel carp (means ±SE) *

饲料 Diet	D1	D2	D3	D4
初始体重 *IBW*[1]	2.32 ±0.00	2.33 ±0.00	2.32 ±0.00	2.32 ±0.00
终末体重 *FBW*[2]	54.95 ±0.76^d	40.66 ±1.17^c	29.40 ±0.16^b	20.46 ±0.41^a
摄食率 *FR*[3]	1.98 ±0.02^a	2.10 ±0.01^b	2.37 ±0.02^c	2.68 ±0.04^d
特定生长率 *SGR*[4]	2.80 ±0.01^d	2.53 ±0.02^c	2.24 ±0.00^b	1.93 ±0.02^a
饲料转化效率 *FCE*[5]	82.02 ±0.70^d	75.08 ±0.48^c	63.83 ±0.57^b	52.51 ±0.86^a
蛋白沉积率 *PRE*[6]	36.11 ±0.49^c	33.14 ±0.13^c	27.19 ±0.84^b	22.70 ±0.63^a
能量沉积率 *ERE*[7]	33.91 ±0.55^c	33.52 ±0.44^c	26.22 ±0.94^b	21.44 ±0.38^a

注: *表中同行数值后不同上标英文字母表示差异显著 ($p < 0.05$). Means in the same row with different superscripts are significantly different ($p < 0.05$); [1]初始体重 *IBW* (g): Initial body weight; [2]终末体重 *FBW* (g): Final body weight; [3]摄食率 *FR* (%体重/天) = 100 ×干物质摄食量 / [天数 ×(初始体重 +终末体重)/2] Feeding rate (% *BW* /day) = 100 ×dry feed intake / [days ×(*FBW* + *IBW*)/2]; [4]特定生长率 *SGR* (%/d) = 100 ×[ln(末重) - ln(初重)]/天数 Specific growth rate (%/d) = 100 ×[ln(final weight) - ln(initial weight)]/days; [5]饲料效率 *FCE* (%) = (100 ×鱼体总增重)/总干物质摄食量 Feed conversion efficiency (%) = (100 ×fresh body weight gain)/dry feed intake; [6]蛋白质沉积率 *PRE* (%) = (100 ×鱼体蛋白质沉积量)/蛋白质摄入量 Protein retention efficiency (%) = (100 ×ptotein retained in fish body)/ptotein intake; [7]能量沉积率 *ERE* (%) = (100 ×鱼体能量沉积量)/能量摄入量 Energy retention efficiency (%) = (100 ×energy retained in fish body)/energy intake

2.2 表观消化率

如表 3所示,随着饲料中豆粕含量的升高,异育银鲫的干物质和能量表观消化率显著降低 ($p < 0.05$),但是 D1和 D2之间差异不显著 ($p > 0.05$);蛋白表观消化率随着饲料中豆粕含量的升高显著升高 ($p < 0.05$),蛋白表观消化率在 D3和 D4之间差异不显著 ($p > 0.05$)。

表 3 异育银鲫对实验饲料的表观消化率 (平均值 ±标准误) *

Tab. 3 Apparent digestibility coefficient (ADC) of experimental diets in gibel carp (means ±SE) *

饲料 Diet	干物质表观消化率 *ADCd*[1]	蛋白表观消化率 *ADCp*[2]	能量表观消化率 *ADCe*[3]
D1	69.04 ±0.46^{bc}	82.83 ±0.48^a	79.88 ±0.51^{bc}
D2	70.30 ±1.29^c	86.69 ±1.70^b	81.51 ±1.39^c
D3	67.52 ±0.17^b	89.97 ±0.12^c	78.13 ±0.54^b
D4	56.70 ±0.31^a	91.86 ±0.57^c	68.24 ±0.19^a

注: * 表中同列数值后不同上标英文字母表示差异显著 ($p < 0.05$). Means in the same column with different superscripts are significantly different ($p < 0.05$); [1]干物质表观消化率 *ADCd* (%) = 100 ×[1 - (饲料 Cr_2O_3含量 /粪样 Cr_2O_3含量)] *ADC* of dry matter (%) = 100 ×[1 - (Cr_2O_3 in the diet/ Cr_2O_3 in the faeces)]; [2]蛋白质表观消化率 *ADCp* (%) = 100 ×[1 - (饲料 Cr_2O_3含量 /粪样 Cr_2O_3含量) ×(粪样中粗蛋白含量 /饲料中粗蛋白含量)] *ADC* of protein (%) = 100 ×[1 - (Cr_2O_3 in the diet/ Cr_2O_3 in the faeces) ×(crude protein in the faeces / crude protein in the diet)]; [3]能量表观消化率 *ADCe* (%) = 100 ×[1 - (饲料 Cr_2O_3含量 /粪样 Cr_2O_3含量) ×(粪样中粗能量含量 /饲料中能量含量)] *ADC* of energy (%) = 100 ×[1 - (Cr_2O_3 in the diet/ Cr_2O_3 in the faeces) ×(energy in the faeces / energy in the diet)]

2.3 氮代谢

由表 4可见,豆粕替代饲料中的鱼粉显著影响异育银鲫的氮代谢。总氮摄入量、表观氮摄入量、粪氮排出量、非粪氮排泄量、总氮沉积率均随着饲料中豆粕含量的升高呈显著降低的趋势 ($p < 0.05$),生产每千克鱼的氮排放量则随着饲料中豆粕含量的升高显著升高 ($p < 0.05$)。

2.4 血液生化指标

随着饲料中豆粕含量的升高,各处理的血清葡萄糖和甘油三酯的含量均显著升高 ($p < 0.05$);血清的胆固醇水平和溶菌酶活性显著降低 ($p < 0.05$);超氧化物歧化酶的活性显著升高 (表 5)。

2.5 鱼体生化成分

表 6的实验结果表明,随着异育银鲫饲料中豆粕含量的升高,鱼体的粗蛋白含量不受影响 ($p > 0.05$);各处理组鱼体的干物质含量显著降低 ($p < 0.05$),D1和 D2之间无差异,D2鱼体干物质含量显著高于 D3和 D4 ($p < 0.05$);鱼体粗脂肪含量先显著升高后显著降低 ($p < 0.05$),D2与 D3的鱼体粗脂肪含量无差异,但 D2显著高于 D1和 D4;各处理组鱼体的灰分含量随着饲料中豆粕含量的升高而显著降低 ($p < 0.05$);鱼体的能值也随着饲料中豆粕含量的升高而降低,D2鱼体的能值显著高于 D4 ($p < 0.05$)。

表 4　饲料中豆粕替代鱼粉蛋白对异育银鲫氮代谢的影响 (平均值 ±标准误) *

Tab. 4　Effects of soybean meal replacing dietary fishmeal protein on nitrogen metabolism (means ±SE) *

饲料 Diet	D1	D2	D3	D4
总氮摄入量 *TNI*[1]	3.83 ±0.06^{d}	3.06 ±0.07^{c}	2.55 ±0.04^{b}	2.10 ±0.04^{a}
表观氮摄入量 *ANI*[2]	3.17 ±0.04^{d}	2.66 ±0.12^{c}	2.29 ±0.03^{b}	1.93 ±0.04^{a}
粪氮排出量 *FNO*[3]	0.66 ±0.03^{c}	0.40 ±0.04^{b}	0.26 ±0.00^{a}	0.17 ±0.01^{a}
非粪氮排泄量 *NFE*[4]	1.73 ±0.03^{b}	1.59 ±0.10^{ab}	1.54 ±0.04^{a}	1.40 ±0.02^{a}
总氮沉积率 *NE*[5]	36.11 ±0.49^{c}	33.14 ±0.13^{c}	27.19 ±0.84^{b}	22.70 ±0.63^{a}
氮排放量 *NL*[6]	46.52 ±0.72^{a}	53.38 ±0.32^{b}	68.52 ±1.39^{c}	89.68 ±2.09^{d}

注: * 表中同列数值后不同上标英文字母表示差异显著 ($p < 0.05$). Means in the same column with different superscripts are significantly different ($p < 0.05$); [1]总氮摄入量 *TNI* (g/fish) = 干物质摄入量 ×饲料中氮含量 Total nitrogen intake (g/fish) = dry matter intake ×nitrogen concentration in the diet; [2]表观氮摄入量 *ANI* (g/fish) = 总氮摄入量 ×氮的消化率 (%) / 100 Apparent nitrogen intake (g/fish) = total nitrogen intake ×apparent digestibility of nitrogen (%) / 100; [3]粪氮排出量 *FNO* (g/fish) = 总氮摄入量 ×(100 - 氮的消化率 (%)) / 100 Faecal nitrogen output (g/fish) = total nitrogen intake ×(100 - apparent digestibility of nitrogen (%)) / 100; [4]非粪氮排泄量 *NFE* (g/fish) = 表观氮摄入量 - 鱼体总氮含量 Non-faecal nitrogen excretion (g/fish) = apparent nitrogen intake - nitrogen concentration in fish body; [5]氮的沉积率 *NE* (%) = 氮储积量 /氮的摄食量 ×100 Nitrogen retention (%) = nitrogen deposited / nitrogen fed ×100; [6]氮排放量 *NL* (g/kg fish production) = (氮的摄食量 (g) - 氮的储积量 (g)) /湿重增重量 (kg) Nitrogen load (g/kg) = (nitrogen consumed (g) - nitrogen retained (g)) /weight gain (kg)

表 5　饲料中豆粕替代鱼粉对异育银鲫血清生化指标的影响 (平均值 ±标准误) *

Tab. 5　Effects of dietary soybean meal replacing fishmeal on serum parameters in gibel carp (means ±SE) *

饲料 Diet	D1	D2	D3	D4
葡萄糖 Glucose (mmol/L)	2.86 ±0.08^{a}	2.79 ±0.35^{a}	4.94 ±0.50^{b}	8.57 ±0.61^{c}
甘油三酯 Triacylglycerol (mmol/L)	2.05 ±0.04^{a}	3.26 ±0.41^{b}	4.88 ±0.21^{c}	8.17 ±0.36^{d}
胆固醇 Cholesterol (mg/dl)	423.99 ±21.38^{b}	459.08 ±39.24^{b}	285.71 ±9.12^{a}	286.42 ±8.56^{a}
溶菌酶 Lysozyme (μg/L)	4.33 ±0.10^{c}	4.05 ±0.07^{c}	3.53 ±0.21^{b}	3.08 ±0.09^{a}
超氧化物歧化酶 Superoxide dismutase (U/mL)	335.81 ±4.09^{a}	328.80 ±11.79^{a}	326.80 ±22.71^{a}	440.33 ±15.79^{b}

注: *表中同行数值后不同上标英文字母表示差异显著 ($p < 0.05$). Means in the same row with different superscripts are significantly different ($p < 0.05$)

表 6　饲料中豆粕替代鱼粉对鱼体生化组成的影响 (占鲜重 %) (平均值 ±标准误) *

Tab. 6　The effect of dietary soybean meal replacing fishmeal on body composition in gibel carp (means ±SE) *

饲料 Diet	干物质 Dry matter (%)	粗蛋白 Crude protein (%)	粗脂肪 Crude lipid (%)	灰分 Ash (%)	总能 Energy (kJ/g)
初始 Start	22.06 ±0.72	15.02 ±0.54	3.81 ±0.15	3.32 ±0.12	4.76 ±0.16
D1	29.93 ±0.35^{bc}	16.37 ±0.12	8.93 ±0.03^{b}	3.08 ±0.05^{c}	7.33 ±0.09^{ab}
D2	31.00 ±0.35^{c}	16.45 ±0.13	9.59 ±0.19^{c}	2.85 ±0.03^{b}	7.68 ±0.08^{b}
D3	29.37 ±0.55^{ab}	15.90 ±0.32	9.30 ±0.27^{bc}	2.64 ±0.04^{a}	7.30 ±0.19^{ab}
D4	28.28 ±0.43^{a}	16.29 ±0.25	8.27 ±0.15^{a}	2.59 ±0.09^{a}	7.03 ±0.09^{a}

注: *平均值后面的字母不同表示差异显著 ($p<0.05$). Means with different superscripts show significant difference ($p<0.05$)

3　讨　论

3.1　豆粕替代鱼粉对异育银鲫生长与饲料利用的影响

在本实验中,随着饲料中豆粕替代鱼粉蛋白水平的升高,异育银鲫的特定生长率和饲料转化效率显著降低,这与豆粕在斑点叉尾鮰 (*Ictalurus punctatus*)[19]、长鳍叉尾鮰 (*Ictalurus furcatus*)[20]中应用结果是一致的。而豆粕在南方鲇 (*Silurus meridionalis*)饲料中的研究表明,当大豆蛋白替代鱼粉蛋白 39%时,南方鲇的特定生长率和饲料转化效率显著高于对照组,当替代水平超过 52%时,其特定生长率和饲料转化效率显著低于对照和其他各处理组[21]。也有的研究表明,当饲料中的豆粕含量低于 30%,

欧鳗的特定生长率和饲料转化效率与对照组之间没有差异,当超过这个范围,其特定生长率和饲料转化效率显著降低[22]。由此可见,不同的鱼对豆粕的利用情况是不同的。

异育银鲫是杂食性鱼类,鱼粉蛋白需求量为38%[23]。畜禽副产品粉替代异育银鲫饲料中的鱼粉蛋白的最适水平为66.5%[24],肉骨粉能够替代异育银鲫饲料中20%的鱼粉蛋白而不影响其生长[25]。已有的研究表明畜禽副产品粉和肉骨粉中赖氨酸、蛋氨酸不足是影响异育银鲫生长的主要因素[26]。赖氨酸和蛋氨酸是鱼类生长的最重要的限制性氨基酸,通过添加这两种氨基酸可以有效改善鱼类生长[27]。在本实验中,随着饲料中豆粕含量的升高,饲料中蛋氨酸和赖氨酸的含量呈降低的趋势,这可能是影响其生长的因素。

饲料的适口性是影响鱼类摄食的主要因素之一。豆粕中的抗营养因子影响饲料的适口性,进而影响鱼类摄食。在红拟石首鱼(*Sciaenops ocellatus*)[28]、合齿鲷(*Symphysodon aequifasciata* HECKEL)[29]的研究中,鱼的摄食率随着饲料中豆粕含量的增多显著降低。而对埃及鳎(*Solea aegyptiaca*)[30]的研究结果则表明鱼类的摄食率不受饲料中豆粕含量的影响。还有研究表明鱼类的摄食率会随着饲料中豆粕含量的增多而升高,如豆粕在鮸状黄姑鱼(*Nibea miichthioides*)[31]中的应用。豆粕替代鱼粉在虹鳟(*Oncorhynchus mykiss*)[32]、大西洋鲑(*Salmo salary*)[33]中的研究表明,在实验开始阶段鱼摄食率较低,不能满足营养需求,经过一段时间的调节,鱼可以适应高含量的豆粕饲料,并通过增加摄食量来满足生长所需要的能量需求。本实验中,随着饲料中豆粕含量的升高,异育银鲫的摄食率显著升高,这说明含豆粕饲料的适口性不是影响异育银鲫利用豆粕的原因。

3.2 豆粕替代鱼粉对异育银鲫消化率和氮代谢的影响

消化率是影响鱼类利用豆粕的重要因素。在黄尾鰤(*Seriola lalandi* Valenciennes)[34]的研究中,饲料中豆粕含量不会影响其蛋白表观消化率。在合齿鲷[29]的研究中,蛋白的表观消化率呈下降的趋势。在本研究中随着饲料中豆粕含量的升高,异育银鲫的蛋白表观消化率显著升高,说明异育银鲫可以吸收豆粕中的蛋白。从粪氮和非粪氮排出量的数据分析,豆粕替代饲料中的鱼粉后会显著降低鱼体氮排放,但是生产每千克鱼的氮排放量却随着饲料中豆粕含量的升高显著升高。氮沉积率与氮排放成负相关。这表明鱼体吸收的蛋白质不能有效的用于生长。这可能是由于豆粕的中赖氨酸和蛋氨酸不足影响鱼类生长时蛋白质的合成,多余的氨基酸经脱氨基作用,含氮的部分以氨、尿素和三甲胺的形式排出体外[35]。也可能是因为饲料中的必需氨基酸与非必需氨基酸的比例不合适影响了鱼体蛋白的合成,饲料中的必需氨基酸与非必需氨基酸的比例必须有一个适宜的比例,才能使蛋白质最大程度地用于生长[36]。如罗非鱼饲料中必需氨基酸和非必需氨基酸比例为40∶60其蛋白沉积率最高,氮排放量最低[37]。此外,豆粕中的胰蛋白酶抑制因子、植酸等抗营养因子都会影响鱼类利用饲料中的蛋白[5]。

随着饲料中豆粕的含量的升高,异育银鲫的干物质和能量表观消化率呈降低的趋势。本实验的结果与豆粕蛋白在虹鳟饲料中的研究结果一致,作者认为豆粕中的不易消化的碳水化合物是影响消化率的因素[38]。由于鱼类肠黏液中缺乏α半乳糖苷酶,豆粕中低聚糖很难被鱼类消化[39]。Refstie, *et al.*[40]发现豆粕中的多糖会干扰大西洋鲑对脂肪的吸收。

3.3 豆粕替代鱼粉对异育银鲫鱼体成分的影响

豆粕替代饲料中的鱼粉蛋白后,在不同的实验中对鱼体生化成分的影响并不一致。豆粕替代饲料中的鱼粉蛋白后,对虹鳟[41]、大西洋鲑[42]的鱼体蛋白、脂肪、水分和灰分均无显著影响。在本实验中,异育银鲫鱼体的蛋白含量不受影响,而干物质、灰分、脂肪含量均受到影响。饲料中豆粕含量越高,异育银鲫的生长越差,鱼体的干物质、灰分和脂肪含量越低。

3.4 豆粕替代鱼粉对异育银鲫血清免疫力的影响

在本实验中,饲料中豆粕含量越高,血清中的葡萄糖和甘油三酯的含量越高,异育银鲫生长越差,这表明豆粕通过影响鱼体的糖脂代谢进而影响鱼的生长。异育银鲫血清的胆固醇随着饲料中豆粕含量的升高显著的降低。当植物蛋白源替代饲料中鱼粉后,虹鳟[38]和舌齿鲈(*Dicentrarchus labrax*)[43]血浆中的胆固醇也呈现降低的趋势。研究表明,植物产物会降低陆生动物体内的胆固醇含量,主要是由植物产物中的类雌激素异黄酮所致[44]。

溶菌酶广泛存在于动物、植物和微生物的各种组织、体液及分泌物中,是生物体内重要的非特异性免疫因子之一,在抵抗外来病原入侵中起重要作用。溶菌酶是一种水解酶,具有抗菌活性,通过破坏细胞

壁使细菌裂解[45]。其活性的高低是衡量机体免疫状态的指标之一。张松用肉骨粉和豆粕替代异育银鲫饲料中的鱼粉后,血清的溶菌酶活性明显降低[46]。在本研究中,随着饲料中豆粕含量的升高,异育银鲫血清溶菌酶活性显著降低,这意味着高含量的豆粕替代饲料中的鱼粉蛋白会降低异育银鲫的免疫力。

超氧化物歧化酶是一种生物活性蛋白质,清除超氧阴离子自由基,保护细胞免受损伤[17]。本实验的研究结果显示,豆粕替代饲料中的鱼粉后,异育银鲫的血清中超氧化物岐化酶的活性显著的升高,这表明鱼体处于氧化应激的状态。已有研究表明,大豆异黄酮可以显著提高小鼠的抗氧化活性[47];豆类作物中的类黄酮经体外实验证明也具有抗氧化活性[48]。豆粕中含有多种抗营养因子,但这些因子究竟是通过何种途径影响鱼类健康的,目前尚不清楚,有待深入研究。

4 结　论

与优质白鱼粉相比,饲料中豆粕的使用显著影响异育银鲫的生长和饲料利用。适口性不是影响异育银鲫利用豆粕的因素,饲料中的氨基酸不平衡是影响其生长的关键因素,也是导致鱼体的氮排放量增加的因素。随着饲料中豆粕含量的升高,蛋白表观消化率显著升高,干物质和能量表观消化率则显著降低;鱼体的免疫力和糖脂代谢均受到影响。

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EFFECT OF DIETARY REPLACEMENT OF FISHMEAL PROTEIN BY SOYBEAN MEAL PROTEIN ON GROWTH PERFORMANCE, METABOLISM AND IMMUNITY OF GIBEL CARP (CARASSIUS AURATUS GIBELIO)

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Abstract: A 16-weeks growth was conducted to evaluate the effect of replacement of dietary fishmeal by soybean meal on growth performance, feed utilization, nitrogen metabolism and immunity in gibel carp. Four isonitrogenous and isocaloric diets were formulated. Each diet was fed to triplicate groups of fish with the initial weight at about 2.32 g. Soybean meal was used to replace 0 (Control, D1), 20% (D2), 80% (D3) and 100% (D4) of dietary fishmeal protein. The fish was reared in a semi-recirculating system. During the experiment, water temperature was 23—30℃, photoperiod was 12D∶12L with the light period from 08:00 to 20:00, dissolved oxygen was above 5 mg/L, ammonia-N (NH_4^+-N plus NH_3-N) was less than 0.5 mg/L, pH was about 6.4. Fish were fed to satiation twice daily (9:00 and 15:00). At the beginning of the experiment, healthy fish (initial body weight about 2.32 g) were batch weighed after 24h feed deprivation and randomly distributed into the 12 tanks (40 fish per tank). The tanks were randomly assigned the four diets. Fifty fish were taken from the remaining fish and frozen for initial fish body chemical analysis. During the experiment, an excess amount of feed was fed to fish and uneaten feed were collected after 1h in each feeding, dried at 60℃ and reweighed. Leaching rate of uneaten feed in tanks was estimated by placing weighed feeds into a tank without fish for 1h and then recovering, drying and reweighing. The average leaching rate was used to calibrate the amount of uneaten feed. Faeces were collected after uneaten feed collection at the start of the experiment and through all the experiment period after 7d. To minimize nutrient leaching in faeces, only fresh and intact faeces were collected. Faeces were dried at 70℃ for digestibility determination. At the end of the trial, the fish were starved for 1d and batch weighed. Fish were killed by a blow on the head and blood samples were collected (24h after last feeding) from the caudal vein of six fish from each tank at the end of the feeding trial by using heparinized syringes. Blood was centrifuged at 3500 r/min for 15 min, plasma-separated and stored at -80 ℃. The remaining fish in each tank were taken for final fish body composition analysis. The results showed that feeding rate (FR), specific growth rate (SGR), feed conversion efficiency (FCE), protein retention efficiency (PRE) and energy retention efficiency (ERE) decreased significantly ($p < 0.05$). Apparent digestibility coefficient of protein (ADCp) significantly increased ($p < 0.05$), but ADC of dry matter and energy decreased ($p < 0.05$). Total nitrogen intake (TNI), apparent nitrogen intake (ANI), faecal nitrogen output (FNO), non-faecal nitrogen excretion (NFE) and nitrogen retention (NE) significantly decreased with the increasing dietary soybean meal ($p < 0.05$), but nitrogen load (NL) significantly increased ($p < 0.05$). Levels of plasma glucose, triacylglycerol and superoxide dismutase activities increased significantly with the increasing dietary soybean meal ($p < 0.05$), but plasma cholesterol concentration and lysozyme activities decreased ($p < 0.05$). In conclusion, the results from this study showed adverse effects of inclusion of the soybean meal in diets on growth performance, feed utilization, nitrogen metabolism and immunity in gibel carp. The palatability was not negatively affected when soybean meal was included in the diets. Unbalanced amino acid composition of soybean meal diets seem to be the main reason to influence growth performance and nitrogen load of gibel carp.

Key words: Soybean meal; Fishmeal; Gibel carp (*Carassius auratus gibelio*)