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大口黑鲈配合饲料中 3 种动物蛋白源的不同添加比例对其生长性能、肠道健康及蛋白质代谢的影响*

江坤生[1,2#] 陈 芳[1#] 张关荣[1] 黄泽葵[1] 关俊锋[1]
麻永财[1] 陈宇航[2] 谢帝芝[1] 李远友[1①]

(1. 华南农业大学海洋学院 广东 广州 510642；2. 广东德宁水产科技有限公司 广东 佛山 528399)

摘要 目前，大口黑鲈(*Micropterus salmoides*)配合饲料对鱼粉的依赖性大，而鱼粉价格不断上涨，导致其饲料成本居高不下，严重制约了其养殖业的健康发展。为降低饲料成本，本研究以鱼粉、鸡肉粉和猪肉粉为饲料动物蛋白源，制作 7 种配合饲料(D1~D7)，其鱼粉/鸡肉粉/猪肉粉的添加百分比分别为 45.0/22.6/0、37.1/22.6/8.0、28.8/22.6/16.0、45.0/14.5/8.0、45.0/5.3/16.0、41.6/18.0/8.0 和 37.0/13.8/16.0。采用上述饲料投喂大口黑鲈幼鱼(初始体重约为 55 g) 60 d，评估饲料动物蛋白源组合对鱼生长性能、组织生化指标、肌肉质构特性以及肝脏蛋白质代谢和肠道炎性因子相关基因表达的影响。结果显示，相比于其他饲料投喂组，D3 组鱼的终末体重、增重率、特定生长率显著提高，饲料系数显著降低(P<0.05)；D3 组全鱼粗蛋白质显著高于 D5 组，其粗脂肪水平显著低于 D4 和 D6 组(P<0.05)。在组织生理生化指标方面，D3 组鱼血清总氨基酸含量显著高于 D1 和 D4 组(P<0.05)，而其谷草转氨酶活性显著低于 D5 组(P<0.05)；D3 组鱼肝脏总蛋白含量显著高于 D7 组(P<0.05)。在肌肉品质方面，D3 组肌肉硬度和胶着性以及咀嚼性分别显著低于 D4 组和 D6 组(P<0.05)。此外，D3 组肠抗炎基因 *il-10*，肝脏蛋白质合成基因 *tor*、*s6k1*、*akt*、*pi3k* mRNA 表达水平上调，显著高于 D7 组(P<0.05)；而肠促炎因子 *il-1β*、*il-6* 和肝脏翻译抑制因子 *4ebp-1* mRNA 表达水平下调，显著低于 D1 组(P<0.05)。上述结果表明，饲料中添加 28.8%鱼粉、16.0%猪肉粉和 22.6%鸡肉粉对大口黑鲈的促生长效果最优，且有利于提高肝脏蛋白质合成，维护肠道健康。研究结果可为降低大口黑鲈配合饲料对鱼粉的依赖提供技术支撑。

关键词 大口黑鲈；动物蛋白源；生长性能；蛋白质合成；肠道健康

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同等贡献作者：江坤生，E-mail: 2316080858@qq.com；陈 芳，E-mail: chenfang51@163.com

① 通信作者：李远友，教授，E-mail: yyli16@scau.edu.cn

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鱼粉的蛋白质、必需脂肪酸、矿物质、维生素含量高，必需氨基酸种类齐全、比例平衡，适口性和消化吸收率好，是传统水产配合饲料主要的蛋白源(Tacon *et al*, 2008)。近年来，水产养殖规模不断扩大，资源有限的鱼粉供不应求，价格不断攀高，大大增加了水产养殖的成本投入，限制了水产养殖业的可持续发展。因此，高效低鱼粉蛋白源的开发已成为当前水产饲料研究中的热点问题。植物蛋白来源广泛、供应稳定、价格较低，是良好的鱼粉替代物，但因抗营养因子含量较高、氨基酸不平衡、适口性差等原因，过量添加易对养殖鱼类(特别是肉食性鱼类)的生长和健康产生不良影响(肖金星等, 2011)。相比于植物蛋白，陆生动物蛋白源的抗营养因子少、蛋白含量高，且含有对鱼体健康有益的功能性因子(吴瑞, 2018)。其中，鸡肉粉(含 65%~73%蛋白，亮氨酸、赖氨酸及维生素含量丰富)和猪肉粉(含 45%~60%蛋白，脯氨酸和甘氨酸含量高)在水产配合饲料中应用最为广泛，是重要的鱼粉替代源(郭小瑞等, 2022)。

大口黑鲈(*Micropterus salmoides*)俗称加州鲈，具有生长速度快、肉多刺少、营养丰富等优点，年均养殖量高达 15 万 t，为我国第“五”大家鱼(赵鹏飞等, 2016)。作为肉食性鱼类，大口黑鲈对饲料鱼粉的依赖性较高，其商品饲料中鱼粉添加水平高达 50% (李永娟等, 2016)。而鱼粉价格的不断上涨，导致大口黑鲈养殖成本增加，亟需寻找合适的替代蛋白源，以降低其饲料鱼粉添加量，从而降低饲料成本。因此，本研究通过分析 7 组不同比例鱼粉、猪肉粉、鸡肉粉的动物蛋白源对大口黑鲈生长性能、肠道健康及肝脏蛋白质代谢的影响，评估猪肉粉和鸡肉粉替代鱼粉的效果，为开发高效低鱼粉配合饲料提供依据。

1 材料与方法

1.1 实验饲料

本研究以不同比例的鱼粉、鸡肉粉、猪肉粉为动物蛋白源，豆油为脂肪源配制 7 种等氮(粗蛋白 55.0%)等脂(粗脂肪 13.0%)的配合饲料(D1~D7)。其鱼粉/鸡肉粉/猪肉粉的添加百分比分别为 45.0/22.6/0 (对照组)、37.1/22.6/8.0、28.8/22.6/16.0、45.0/14.5/8.0、45.0/5.3/16.0、41.6/18.0/8.0 和 37.0/13.8/16.0，饲料配方及营养组成见表 1。各种饲料原料进行粉碎后，将粉碎的饲料原料混匀，加入水再次混匀，接着在膨化机(XBF-62, 鑫贝发)的作用下制成膨化饲料。烘干后外喷油(XBF-65, 鑫贝发)制成饲料。制好的饲料阴干后，于–20 ℃保存。

1.2 实验设计

实验鱼购于佛山当地养殖户，并置于广东德宁水产科技有限公司杏坛基地大网箱(5 m×5 m×2 m)中暂养。暂养期间，用容川商品饲料投喂至实验规格，使其适应养殖环境。养殖实验开始前，禁食 24 h，挑选大小一致的健康幼鱼(初始体重约为 55 g)随机分配至 35 个实验网箱(1.5 m×1.5 m×1.8 m)中，每组 5 个重复。每个网箱 100 尾鱼，养殖周期为 60 d。养殖期间每天投喂 2 次(06:30 和 17:45)，水温为 27.4~32.3 ℃，氨氮含量为 0.1~0.2 mg/L。同时，每天观察鱼的摄食、活动情况，如有异常及时处理。若有死鱼及时捞出并记录重量。

1.3 样品采集

养殖实验结束后，禁食 24 h。对每个网箱中的鱼逐一称重，并统计其重量。从每个网箱随机选取 3 尾鱼，放入含有 0.01% 2-苯氧乙醇麻醉剂的水中，采用尾部静脉取血，并分离其肌肉、肝脏、肠道等组织。另外，每个网箱随机选取 3 尾鱼，测定其内脏重、肝脏重和体长，用于计算肝体比、脏体比及肥满度。最后，从每个网箱取 2 尾鱼分别用于测定肌肉质构特性和全鱼营养成分。

1.4 组织生理生化指标测定

血清中的白蛋白(ALB)、尿素氮(BUN)、总氨基酸(TAA)、谷丙转氨酶(ALT)、谷草转氨酶(AST)、总蛋白(TP)及血氨(SA)，肝脏中的谷草转氨酶、谷丙转氨酶、总蛋白均采用南京建成生物工程研究所的试剂盒测定，具体测定步骤见各试剂盒说明书。

1.5 肌肉质构特性测定

肌肉质构特性使用质构仪(Universal TA 型，上海腾拔仪器科技有限公司)进行测定，具体测定步骤参考 Ma 等(2020)：取下实验鱼的背肌，在 TPA 模式下，使用 TA 25/1000 探头，测试参数：测试前后速度为 2.00 mm/s，测试时速度为 1.00 mm/s，压缩比为 75%，前后 2 次压缩时间间隔为 2 s。

1.6 常规成分测定

肌肉和全鱼的常规成分测定方法参考 Li 等(2017)和 Li 等(2008)的方法。水分、粗脂肪、粗蛋白及灰分分别采用常压干燥法、索氏抽提法、凯氏定氮法和马弗炉灼烧法测定。

表 1 饲料组成及其营养成分/%

Tab.1 Composition and nutrient levels of experimental diets/%

饲料组成 Dietary composition	饲料处理组 Dietary treatment						
	D1	D2	D3	D4	D5	D6	D7
秘鲁鱼粉 Peruvian fish meal	45.0	37.1	28.8	45.0	45.0	41.6	37.0
猪肉粉 Porcine meat meal		8	16	8	16	8	16
鸡肉粉 Poultry by-product meal	22.6	22.6	22.6	14.5	5.3	18.0	13.8
去皮豆粕 Peeled soybean meal	4.6	4.6	4.9	4.7	5.5	4.7	5.3
棉籽蛋白 Cottonseed protein	5	5	5	5	5	5	5
大豆油 Soybean oil	6.7	5.7	4.9	6.0	5.6	5.9	5.2
谷朊粉 Gluten powder	2	2	2	2	2	2	2
木薯粉 Tapioca flour	11	11	11	11	11	11	11
大口黑鲈预混料 Largemouth bass premix[1]	1	1	1	1	1	1	1
磷酸二氢钙 $Ca(H_2PO_4)_2$	1.5	2.0	2.5	2.0	2.5	2.0	2.5
氯化胆碱 Choline chloride	0.6	0.6	0.6	0.6	0.6	0.6	0.6
L-赖氨酸盐酸盐 L-lysine hydrochloride		0.2	0.5	0.2	0.3	0.2	0.4
L-苏氨酸 L-threonine		0.1	0.1		0.1		0.1
DL-蛋氨酸 DL-methionine		0.1	0.1		0.1		0.1
总计 Total	100	100	100	100	100	100	100
饲料营养成分 Nutrient composition of diets							
水分 Moisture	5.11	5.49	5.67	5.92	4.71	6.03	5.71
粗蛋白 Crude protein	54.99	55.39	55.73	54.72	55.38	55.63	54.94
粗脂肪 Crude lipid	13.28	13.21	12.90	13.48	13.80	13.82	13.24
灰分 Ash	12.85	13.90	14.10	13.56	13.29	12.54	12.35
饲料氨基酸成分 Amino acid composition of diets							
天冬氨酸 Asp	4.36	4.56	4.54	4.44	4.35	4.01	3.96
苏氨酸 Thr	2.08	2.11	2.09	2.10	2.04	2.16	2.22
丝氨酸 Ser	2.09	2.15	2.15	2.18	2.05	2.09	2.10
谷氨酸 Glu	7.41	7.62	7.63	7.58	7.34	7.19	7.12
甘氨酸 Gly	4.10	3.71	4.14	4.74	4.03	3.68	3.91
丙氨酸 Ala	3.48	3.49	3.58	3.70	3.84	3.98	4.05
缬氨酸 Val	2.48	2.60	2.56	2.57	2.48	2.37	2.34
蛋氨酸 Met	1.11	1.16	1.17	1.08	1.17	1.05	1.05
异亮氨酸 Ile	2.04	2.24	2.15	2.00	2.03	1.99	1.90
亮氨酸 Leu	3.65	3.88	3.81	3.65	3.65	3.74	3.65
酪氨酸 Tyr	1.59	1.78	1.75	1.56	1.54	1.87	1.84
苯丙氨酸 Phe	2.14	2.32	2.30	2.09	2.16	2.24	2.18
赖氨酸 Lys	3.56	3.72	3.84	3.66	3.66	3.33	3.38
组氨酸 His	1.40	1.43	1.46	1.39	1.42	1.78	1.21
精氨酸 Arg	3.35	3.36	3.49	3.61	3.23	3.05	3.09
脯氨酸 Pro	2.75	2.59	2.75	3.13	2.42	3.12	3.30
必需氨基酸总和 EAA[2]	21.82	22.82	22.87	22.13	21.83	21.71	20.99

注：1. 每千克大口黑鲈预混料中含有：V_A 100 万 IU，V_{D3} 25 万 IU，DL-α-生育酚乙酸酯 8.0 g，V_{K3} 2.0 g，V_{B1} 2.0 g，V_{B2} 1.6 g，V_{B6} 1.6 g，V_{B12} 5.0 mg，D-泛酸钙 7.0 g，烟酰胺 12.0 g，叶酸 0.4 g，D-生物素 16 mg，肌醇 24.0 g，L-抗坏血酸-2-磷酸酯 62.0 g，Zn 1.5 g，Mn 1.25 g，Cu 0.7 g，Fe 12 g，Co 0.15 g，I 0.1 g，Se 0.015 g，Mg 3.5 g，K 6.0 g，预混料由广东德宁水产科技有限公司提供。

2. 必需氨基酸总和包括 Lys、Phe、Met、Thr、Ile、Leu、His、Arg 和 Val。

Note: 1. Largemouth bass premix per kg contains: V_A 1 million IU, V_{D3} 250 thousand IU, DL-alpha-tocopherol acetate 8.0 g, V_{K3} 2.0 g, V_{B1} 2.0 g, V_{B2} 1.6 g, V_{B6} 1.6 g, V_{B12} 5.0 mg, D-calcium pantothenate 7.0 g, niacinamide 12.0 g, folate 0.4 g, D-biotin 16 mg, inositol 24.0 g, L-ascorbate-2-phosphate 62.0 g, Zn 1.5 g, Mn 1.25 g, Cu 0.7 g, Fe 12 g, Co 0.15 g, I 0.1 g, Se 0.015 g, Mg 3.5 g, K 6.0 g. The premix is provided by Guangdong Daynew Aquatic Sci-Tech Co., Ltd.

2. Essential amino acids (EAA) is the sum of Lys, Phe, Met, Thr, Ile, Leu, His, Arg and Val.

1.7 肠道炎症基因、肝脏蛋白质代谢基因 mRNA 表达量测定

根据鱼体生长性能和组织蛋白含量，挑选出对照组(D1 组)、生长最优组(D3 组)和生长最差组(D7 组)，比较分析 3 组鱼体肝脏蛋白质合成、肠炎症因子相关基因表达水平，以探讨摄食 D3 饲料的鱼具有优良生长性能的潜在机制。肠和肝脏组织总 RNA 采用 Simply P Total RNA extraction kit (Bioflux)提取，具体步骤详见说明书。提取的组织总 RNA 用 1%的琼脂糖凝胶电泳检查 RNA 的完整性。用核酸分析仪测定 RNA 浓度。之后采用反转录试剂盒 PrimeSript™ RT regent kit (TaKaRa)将组织总 RNA 反转录 cDNA。

炎症因子、蛋白质合成代谢相关基因以及 *β-actin* 引物序列均根据 NCBI 中相关基因并采用 Premier 5.0 设计特异性引物(表 2)。采用实时荧光定量 PCR 仪(BIO-RAD, 美国)和 Thunderbird® SYBR qPCR Mix 试剂盒(TOYOBO)进行荧光定量 PCR，反应程序采用两步法：95 ℃预变性 30 s，95 ℃变性 5 s，60 ℃延伸 30 s，共 40 个循环。数据采用 $2^{-\Delta\Delta CT}$ 法进行处理。

表 2 实时荧光定量 PCR 引物

Tab.2 Primer sequences for real-time quantitative PCR

基因 Gene	正向引物 Forward primer	反向引物 Reverse primer	基因序列号 Gene serial number
β-actin	ATCGTCCACCGCAAATGCTT	TGGTGTGGTTGTTTTGCACAG	XM_038695351
il-1β	AGCCGTCATTGAACATGGGA	GAAACATCAGGGGGTGACCA	XM_038733429
tnfα	TACAGCCAAGCGTCCTTCAG	GGACCAGCGCTGAACAGTAT	XM_038733429
il-6	CGCGCAATTTGCCGATGATA	CGTTGTTGCTGGTTGCATGA	XM_038732985
il-10	CCACCAGAATGACTCCTCGG	TGGTTGTTGCACATGGGACT	XM_038696252
tgf-β1	CTTTACTACGTGGGCAGGCA	ATAGGTTTGAGGCGAGGCAG	XM_038693206
tor	CCAAAGACGTGCTGTTCACC	GAGCCTTCAGAAACCTGCGA	XM_038702468
pi3k	GGATGAGACACAGAAGATGCGA	CCTCAGGTTTCCCAGTTGGT	XM_038715187
akt	TCTGGAGCATGTCTGCCAAT	TTTTTGCCAAAGGACAGCCG	XM_038701564
4ebp1	ATATCCGATACAGGCGCGTT	TGGTCTTCTGGCAGTCAGTG	XM_038703877
s6k1	GATTCTTGTGTGCGCCGTTT	ACCTACGGAGAAAGCAACCG	XM_038694705

1.8 计算公式

实验鱼的增重率、特定生长率、饲料系数、成活率、肥满度、肝体比、脏体比和日摄食率的计算公式：

增重率(WGR, %)=(终末鱼均重–初始鱼均重)/初始体重×100

特定生长率(SGR, %/d)=[ln(终末鱼均重)–ln(初始鱼均重)]/养殖天数×100

饲料系数(FCR)=摄入的饲料总重/(终末鱼总重–初始鱼总重)

成活率(SR, %)=终末鱼体数量/初始鱼体数量×100

肥满度(CF)=鱼体重量/(鱼体长)^3×100

肝体比(HSI, %)=肝脏重量/鱼体重量×100

脏体比(VSI, %)=内脏重量/鱼体重量×100

日摄食率(FR, %)=摄入的饲料总量/[(初始鱼体重+终末鱼体重)/2]/养殖天数×100

1.9 数据处理

采用 SPSS 25.0 软件对数据进行方差分析，并用 Tukey test 进行多重比较，以 $P<0.05$ 作为显著性差异。所有数据均用平均值±标准误(Mean±SE)表示。

2 结果

2.1 不同饲料投喂组大口黑鲈生长性能及全鱼常规成分

不同饲料投喂组鱼的生长性能及全鱼常规成分如表 3 所示。在生长性能方面，D1~D7 组中，D3 组鱼的末重、增重率、特定生长率最大，显著高于其他组($P<0.05$)，其饲料系数最小，显著低于其他组($P<0.05$)。各组鱼的肥满度、肝体比、脏体比、成活率均无显著性差异($P>0.05$)。在全鱼常规成分方面，各组间的水分、灰分无显著性差异($P>0.05$)。D3 组全鱼粗蛋白含量显著高于 D5 组($P<0.05$)，与其他组无显著差异($P>0.05$)。D3 组全鱼粗脂肪含量显著低于 D4 和 D6 组($P<0.05$)，与其他组无显著差异($P>0.05$)。

2.2 不同饲料投喂组大口黑鲈组织生化指标

不同饲料投喂组鱼组织生化指标见表 4。各组鱼的血清尿素氮、谷丙转氨酶活性和血氨含量，以及肝脏谷丙转氨酶活性均无显著性差异($P>0.05$)。其中，

表 3 不同饲料投喂组大口黑鲈的生长性能及全鱼常规成分

Tab.3 Growth performance and whole-body composition of *Micropterus salmoides* fed with different diets

项目 Items	饲料处理组 Dietary treatment						
	D1	D2	D3	D4	D5	D6	D7
初始体重 IBW/g	55.10±0.92	56.06±0.04	55.30±0.15	55.25±0.13	55.86±0.28	56.16±0.15	55.70±0.31
终末体重 FBW/g	167.61±2.75^{b}	169.58±0.96^{b}	180.76±2.19^{a}	171.45±2.26^{b}	168.48±1.55^{b}	169.57±0.95^{b}	165.42±1.71^{b}
增重率 WGR/%	204.22±1.44^{bc}	202.50±1.69^{bc}	226.86±3.69^{a}	210.32±4.19^{b}	201.60±2.04^{bc}	201.94±1.93^{bc}	196.97±1.46^{c}
特定生长率 SGR/(%/d)	1.87±0.01^{bc}	1.85±0.01^{bc}	1.97±0.02^{a}	1.89±0.02^{b}	1.84±0.01^{bc}	1.84±0.01^{bc}	1.82±0.01^{c}
饲料系数 FCR	0.98±0.01^{ab}	0.96±0.01^{b}	0.91±0.01^{c}	0.97±0.01^{b}	1.00±0.01^{ab}	0.98±0.01^{b}	1.01±0.01^{a}
日摄食率 FR	1.65±0.01^{ab}	1.60±0.01^{b}	1.60±0.02^{b}	1.64±0.01^{ab}	1.64±0.01^{ab}	1.62±0.01^{b}	1.67±0.03^{a}
肥满度 CF	2.50±0.07	2.49±0.03	2.43±0.02	2.61±0.06	2.52±0.04	2.35±0.14	2.52±0.08
肝体比 HSI/%	2.28±0.16	2.06±0.11	1.93±0.01	2.10±0.02	2.28±0.13	1.82±0.13	2.30±0.10
脏体比 VSI/%	8.22±0.15	7.79±0.18	7.51±0.12	8.12±0.17	8.17±0.40	7.68±0.14	7.94±0.15
成活率 SR/%	95.20±1.39	96.20±0.58	95.33±2.19	95.75±0.63	95.00±1.08	94.80±1.32	95.00±0.58
常规成分 Composition/%WT							
水分 Moisture	68.41±0.54	68.43±0.21	69.14±0.35	68.23±0.42	68.50±0.20	68.44±0.11	68.46±0.27
粗蛋白 Crude protein	17.57±0.10^{ab}	17.54±0.05^{ab}	17.78±0.09^{a}	17.20±0.29^{ab}	17.06±0.06^{b}	17.30±0.11^{ab}	17.36±0.12^{ab}
粗脂肪 Crude lipid	9.53±0.23^{bc}	9.26±0.19^{bc}	9.15±0.18^{c}	10.29±0.27^{ab}	9.63±0.32^{abc}	10.73±0.14^{a}	9.94±0.17^{abc}
灰分 Ash	3.81±0.06	3.87±0.06	3.97±0.22	3.94±0.21	4.09±0.12	4.06±0.25	3.95±0.11

注：同一行中不同小写字母表示相互间有显著差异(P<0.05)，下同。

Note: Values in the same row without common superscript letter means significant differences (P<0.05), the same as below.

表 4 不同饲料投喂组大口黑鲈的血清和肝脏生化指标

Tab.4 Serum and liver biochemical parameters of *Micropterus salmoides* fed with different diets

项目 Items	饲料处理组 Dietary treatment						
	D1	D2	D3	D4	D5	D6	D7
血清 Serum							
白蛋白 ALB /(g/L)	10.86±0.35^{ab}	12.52±0.61^{a}	11.62±0.29^{ab}	9.70±0.07^{b}	12.77±0.45^{a}	11.91±0.45^{a}	12.15±0.55^{a}
尿素氮 BUN /(mmol/L)	5.36±0.30	5.70±0.43	5.99±0.03	4.76±0.20	5.56±0.30	4.71±0.24	5.56±0.21
总氨基酸 T-AA /(μmol/mL)	85.39±7.61^{b}	93.98±7.56^{ab}	117.93±9.52^{a}	82.77±4.44^{b}	103.01±3.54^{ab}	97.14±4.86^{ab}	100.75±5.59^{ab}
谷丙转氨酶 ALT (U/L)	4.95±0.35	4.33±0.50	4.91±0.93	5.71±0.92	4.62±0.86	5.09±0.86	4.20±0.45
谷草转氨酶 AST /(U/L)	4.04±1.11^{ab}	3.90±0.50^{ab}	2.81±0.48^{b}	3.47±0.15^{b}	6.48±0.45^{a}	3.88±0.28^{b}	3.37±0.81^{b}
总蛋白 TP /(g/L)	36.46±0.70^{b}	39.88±1.50^{ab}	41.53±1.21^{ab}	39.84±2.29^{ab}	41.29±0.70^{ab}	43.76±0.69^{a}	39.48±1.65^{ab}
血氨 SA /(μmol/L)	1 876.89± 162.81	1 520.80± 169.86	1 360.51± 107.12	1 432.52± 94.65	1 632.30± 70.77	1 377.70± 116.41	1 511.50± 156.09
肝脏 Liver							
谷丙转氨酶 ALT /(U/g prot)	22.67±2.45	24.11±2.66	24.86±0.68	28.12±3.74	27.40±0.87	23.23±0.96	26.52±1.55
谷草转氨酶 AST /(U/g prot)	19.69±1.41^{ab}	19.24±0.48^{ab}	20.92±1.73^{ab}	25.31±1.08^{a}	17.10±0.73^{b}	20.01±1.97^{ab}	19.14±1.72^{ab}
总蛋白 TP /(g/L)	5.28±0.10^{ab}	5.17±0.13^{ab}	5.89±0.29^{a}	4.85±0.58^{ab}	5.01±0.08^{ab}	5.68±0.20^{ab}	4.62±0.41^{b}

D3 组鱼的血清血氨含量最低。D3 组的血清总蛋白含量、肝脏谷丙转氨酶与其他组间无显著差异(P>0.05)。D3 组鱼的血清总氨基酸含量显著高于 D1 组和 D4 组(P<0.05)，肝脏总蛋白含量显著高于 D7 组(P<0.05)，与其他组无显著性差异(P>0.05)；而 D3 组鱼血清的谷草转氨酶最低，显著低于 D5 组(P<0.05)，与其他组无显著性差异(P>0.05)。

2.3 不同饲料投喂组大口黑鲈肌肉常规成分和质构特性

不同饲料投喂组鱼肌肉质构特性和常规成分如表 5 所示。在肌肉质构特性方面，各组间的粘性、弹性和黏聚性均无显著差异(P>0.05)。D3 组肌肉的硬度和胶着性显著低于 D4 组(P<0.05)，与其他组无显著性差异(P>0.05)。D3 组肌肉的咀嚼性显著低于 D6 组(P<0.05)。D3 组鱼的肌肉回复性显著小于 D1 组和 D7 组(P<0.05)，与其他组无显著差异(P>0.05)。在肌肉常规成分方面，各组间水分、粗蛋白、粗脂肪及灰分含量均无显著差异(P>0.05)，其中，D3 组鱼的肌肉粗蛋白含量最高，灰分含量最低。

2.4 不同饲料投喂组大口黑鲈肠炎症因子和肝脏蛋白合成代谢相关基因表达水平

不同饲料投喂组鱼肠道炎症因子相关基因表达水平见图 1A。在 D1、D3 和 D7 组中，D3 组肠道促炎因子基因 *il-1β*、*tnfα* 和 *il-6* mRNA 表达水平最低，其中，*il-1β* 和 *il-6* mRNA 表达水平显著低于 D1 组(P<0.05)。D3 组肠道抗炎因子基因 *il-10* 和 *tgf-β* mRNA 表达水平最高，其中，*il-10* mRNA 表达水平显著高于 D7 组(P<0.05)。

不同饲料投喂组鱼肝脏蛋白质代谢相关基因表达水平见图 1B。在 D1、D3 和 D7 组中，D3 组蛋白代谢基因 *tor*、*s6k1*、*akt* 和 *pi3k* mRNA 表达水平最高，显著高于 D7 组(P<0.05)，与 D1 组无显著差异(P>0.05)。D3 组翻译抑制因子 *4ebp-1* 表达水平显著低于 D1 组(P<0.05)，与 D7 组无显著差异(P>0.05)。

表 5 不同饲料投喂组大口黑鲈的肌肉常规成分和质构特性

Tab.5 Muscle texture and composition of *Micropterus salmoides* fed with different diets

项目 Items	饲料处理组 Dietary treatment						
	D1	D2	D3	D4	D5	D6	D7
常规成分 Composition/(% wet weight basis)							
水分 Moisture	76.76±0.65	75.94±1.43	77.21±0.71	74.80±1.48	76.98±0.14	76.92±0.48	76.93±0.23
粗蛋白 Crude protein	19.47±0.23	19.90±0.50	20.28±0.41	19.64±0.36	19.97±0.11	19.34±0.20	19.88±0.15
粗脂肪 Crude lipid	3.28±0.27	4.07±0.12	3.34±0.47	3.76±0.31	3.86±0.17	3.52±0.17	3.35±0.45
灰分 Ash	1.35±0.09	1.27±0.04	0.98±0.08	1.37±0.16	1.24±0.11	1.05±0.04	1.19±0.11
质构特性 Muscle texture							
硬度 Hardness/gf	82.39±3.12^{ab}	69.25±4.41^{b}	68.45±3.66^{b}	96.06±3.83^{a}	85.17±5.68^{ab}	71.38±4.47^{b}	83.06±3.01^{ab}
粘性 Adhesiveness/(gf-mm)	0.46±0.06	0.54±0.03	0.68±0.08	0.71±0.09	0.54±0.07	0.55±0.03	0.68±0.04
弹性 Springiness/mm	0.56±0.01	0.57±0.02	0.56±0.02	0.59±0.01	0.57±0.02	0.58±0.01	0.57±0.01
咀嚼性 Chewiness/gf	30.46±1.57^{ab}	27.93±2.47^{b}	28.18±1.63^{b}	36.06±2.38^{ab}	37.79±3.34^{ab}	41.68±3.89^{a}	31.38±2.25^{sb}
胶着性 Gumminess/gf	51.17±2.58^{ab}	47.34±3.57^{b}	47.93±2.54^{b}	65.77±2.80^{a}	58.91±5.64^{ab}	54.67±4.46^{ab}	50.52±2.23^{ab}
黏聚性 Cohesiveness	0.64±0.01	0.64±0.01	0.63±0.01	0.65±0.01	0.63±0.01	0.64±0.01	0.65±0.01
回复性 Resilience	0.82±0.02^{a}	0.77±0.02^{ab}	0.73±0.01^{b}	0.77±0.01^{ab}	0.81±0.02^{ab}	0.81±0.02^{ab}	0.83±0.02^{a}

3 讨论

大量研究表明，水产配合饲料的蛋白源及比例不同，其促生长和维护健康的效果不一样，其原因可能与鱼粉的替代量、蛋白源的种类、氨基酸组成、适口性等有关(郝甜甜等, 2022; 黎恒基等, 2022; Ma *et al*, 2020; Mohammadi *et al*, 2020; Khalil *et al*, 2021; Hang *et al*, 2022)。在杂交石斑鱼(*E. lanceolatus*♂ × *E. fuscoguttatus*♀)相关研究中发现，相比于鱼粉对照组(55%鱼粉)，饲料中鸡肉粉替代 40%~60%的鱼粉具有相同的促生长效果，而当饲料中鱼粉降至 11%、鸡肉粉升至 44%，鱼体的增重率和特定生长率被显著抑制(Wang *et al*, 2020)。曹晓莉等(2020)研究发现，鸡肉粉替代鱼粉比例不超过 45%(基础饲料鱼粉为 55%)对黄鳝(*Monopterus albus*)的生长无负面影响，而替代 60%饲料鱼粉则显著抑制了其生长性能。相似的是，Huang 等(2022)在卵形鲳鲹(*Trachinotus ovatus*)中发现，猪肉粉替代饲料中 30%鱼粉(基础饲料鱼粉为 30%)对生长

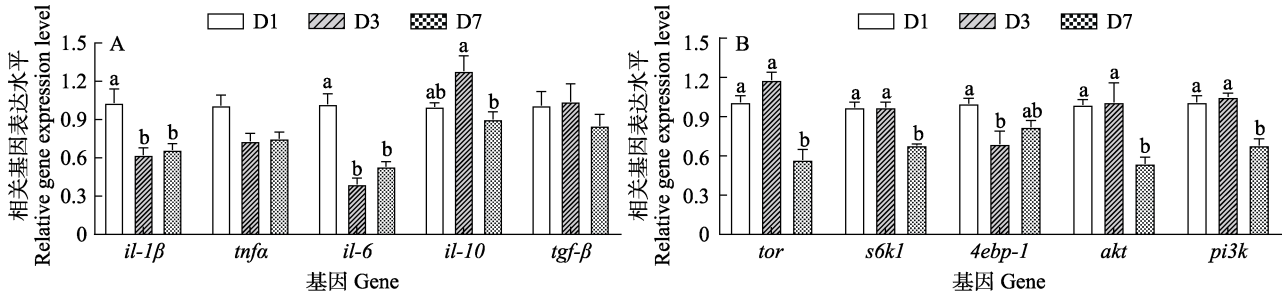

图 1 不同饲料投喂组大口黑鲈肠炎性因子(A)和肝脏蛋白合成代谢相关基因(B)的表达水平

Fig.1 Relative mRNA expression of intestine inflammatory factor (A) and liver protein synthesis (B) in the *Micropterus salmoides* fed with different diets

不同小写字母表示组间有显著差异(P<0.05)。

Values without sharing a common letter means significant differences (P<0.05).

性能无负面影响,而当替代比例超过 30%则显著降低了鱼的增重率。本研究发现,D3 组大口黑鲈增重率、特定生长率显著高于其他组,且饲料系数显著低于其他组,说明 28.8%鱼粉/16.0%猪肉粉/22.6%鸡肉粉的饲料动物蛋白源组合更有利于大口黑鲈生长。

鱼体的生长是其体蛋白合成的体现。血清总氨基酸、血氨及总蛋白含量是评估鱼体蛋白质合成的重要指标。部分氨基酸在鱼体内被氧化分解,其主要产物是氨。研究表明,血氨含量与机体氨基酸分解代谢成正相关性,而氨基酸水平与机体蛋白质合成正相关(林淑琴等, 2014; Wang *et al*, 2020)。肝脏的谷草转氨酶、谷丙转氨酶活性及总蛋白含量也与机体的蛋白质合成正相关(石英等, 2009)。在暗纹东方鲀(*Takifugu obscurus*)的研究中,发现饲料中鸡肉粉替代 15%~60%的鱼粉(对照组鱼粉为 45%)不会显著影响鱼体血清总蛋白含量(崔锡帅, 2022)。Li 等(2019)对大口黑鲈的研究发现,相比于对照饲料组(51%鱼粉),采用鸡血浆粉替代饲料中 10%~30%鱼粉,对血清总蛋白含量无显著影响。对西伯利亚鲟(*Acipenser baerii* Brandt)的相关研究发现,饲料中添加 5%的鸡肉粉不会影响其肝脏谷丙转氨酶活性,对蛋白质代谢无显著影响(Zhu *et al*, 2011)。本研究中,D3 组血清的血氨含量较低,而血清总氨基酸和肝脏总蛋白含量较高,表明该组饲料有利于大口黑鲈的蛋白合成。

研究表明,mTOR 能够感知细胞内的营养素变化,从而调控下游 4E 结合蛋白 1 (4EBP-1)、核糖体蛋白 S6 激酶(S6K1)等靶蛋白,在蛋白质合成代谢中有重要作用(Jing *et al*, 2016; Kim *et al*, 2003; Ma *et al*, 2009)。同时,在蛋白质合成代谢中,PI3K/AKT 信号通路也发挥着关键作用,PI3K/AKT 信号通路可通过激活 mTOR 信号通路、影响蛋白质翻译起始,进而影响蛋白质合成(Rommel *et al*, 2001; Terada *et al*, 1994)。对青斑(*Cromileptes altivelis*)的研究发现,与对照组(69.18%鱼粉)相比,饲料中用复合蛋白(鸡肉粉、血红蛋白粉和大豆蛋白)替代 21%~63%的鱼粉不会显著影响其肝脏 *tor* 和 *s6k1* 的 mRNA 表达水平(Geng *et al*, 2022)。Irm 等(2020)在黑鲷(*Acanthoparus schlegelii*)中发现,饲料中鸡肉粉替代 30%的鱼粉(对照组饲料鱼粉含量为 40%)会显著上调其肝脏 *tor*、*s6k1*、*pi3k*、*akt* 的 mRNA 表达水平,促进蛋白质合成。Li 等(2021)报道,相比 45%鱼粉+17%植物蛋白组,饲料中添加 35%鱼粉、5.4%水解虾蛋白和 23.4%植物蛋白显著上调了大口黑鲈肝脏 *tor* 的 mRNA 表达水平,同时显著下调了肝脏 *4e-bp1* 的 mRNA 表达水平。本研究中,D3 组肝脏 *tor*、*s6k1*、*pi3k* 及 *akt* 的 mRNA 表达水平显著高于 D7 组,*4ebp-1* 表达水平显著低于 D1 组,表明 D3 组饲料可通过激活肝脏 TOR 信号通路,提高机体蛋白质合成代谢表达水平,从而提高大口黑鲈的生长性能。

饲料蛋白源和氨基酸的营养失衡易导致鱼体产生氧化应激(于晓彤, 2016; Ji *et al*, 2022)。而机体氧化应激会导致炎症的发生。在调节炎症反应过程中,TNF-α、IL-1β 和 IL-6 等促炎因子,引发炎症反应;而 IL-10、TGF-β 等抗炎因子能缓解炎症,对机体具有保护作用(Liang *et al*, 2018)。Gaudioso 等(2021)在虹鳟(*Oncorhynchus mykiss*)的研究中发现,饲料中添加 17.8%或 36.0%鸡肉粉不会影响其肠道 *il-1β*、*il-10* 及 *tgf-β* 的 mRNA 表达水平。在大口黑鲈的相关研究中,饲料中添加 36.5%的蛋白水解物会显著提高鱼肠道抗炎基因 *il-10* 和 *tgf-β* 的表达水平(Sheng *et al*, 2022)。Li 等(2022)则发现,与对照组(59.14%鱼粉)相比,棉籽浓缩蛋白替代 100%的鱼粉会上调大口黑鲈肠道促炎因子的表达,下调鱼肠道抗炎因子的表达。本研究发现,D3 组鱼肠 *il-1β*、*il-6* mRNA 表达

水平下调，而 *il-10* mRNA 表达水平上调。说明 D3 组饲料可降低其炎症反应，维护肠道健康。以上结果说明，饲料中的不同蛋白源的不同组成比例会对鱼类肠道炎症反应产生不同的结果，可能与蛋白源的种类及添加比例、鱼的种类等有关。本研究表明，D3 组饲料能够降低大口黑鲈肠道炎症反应，改善其健康，从而促进鱼的生长。

鱼类的肌肉品质会受到内外多种因素的影响，如品质、规格、遗传、养殖条件以及饲料组成等(de Paula *et al*, 2014; Gisbert *et al*, 2016)。质构特性是评价肌肉品质的重要指标。研究表明，肌肉硬度与其嫩度存在一定的负相关(鉏晓艳等, 2021)。本研究发现，相比其他饲料组，D3 组大口黑鲈肌肉硬度最小，其水分含量最大,这表明 D3 组饲料有利于提高其肌肉嫩度。程小飞等(2020)发现饲料中添加猪肉骨粉不会显著影响芙蓉鲤鲫(*Cyprinus capio* Furong ♀ × *Carassius auratus* red var. ♂)肌肉的硬度。在尖吻鲈(*Lates calcarifer*)中的相关研究则发现，饲料添加鸡肉粉对其肌肉硬度无显著影响(Chaklader *et al*, 2022)。Chaklader 等(2021)也在尖吻鲈的研究中发现，用鸡肉粉、黑水虻粉、金枪鱼水解产物替代全部鱼粉(基础饲料鱼粉含量为 72.6%)不会影响其肌肉硬度。本研究表明，D3 组饲料能降低肌肉硬度，从而提高大口黑鲈的鱼肉嫩度，具体机理机制有待进一步实验验证。

4 结论

在本研究实验条件下，饲料中添加 28.8%鱼粉、16.0%猪肉粉及 22.6%鸡肉粉有利于降低其炎症反应(维护肠道健康)，提升肝脏蛋白合成代谢，进而促进鱼体生长。本研究结果可为降低大口黑鲈饲料对鱼粉的依赖、研发高效低鱼粉配合饲料提供技术支撑。

参 考 文 献

CAO X L, ZHONG L, DAI Z Y, *et al*. Effects of fish meal replacement by pet grate poultry by-product meal on growth performance, intestinal digestive enzyme activities and serum biochemical indexes of rice filed eel (*Monopterus albus*). Chinese Journal of Animal Nutrition, 2020, 32(5): 2352–2360 [曹晓莉, 钟蕾, 戴振炎, 等. 宠物级鸡肉粉替代鱼粉对黄鳝生长性能、肠道消化酶活性及血清生化指标的影响. 动物营养学报, 2020, 32(5): 2352–2360]

CHAKLADER M R, CHUNG W H, HOWIESON J, *et al*. A combination of *Hermetia illucens* reared on fish waste and poultry by-product meal improves sensory and physicochemical quality of farmed barramundi filets. Frontiers in Nutrition, 2022, 8: 788064

CHAKLADER M R, HOWIESON J, SIDDIK M A B, *et al*. Supplementation of tuna hydrolysate and insect larvae improves fishmeal replacement efficacy of poultry by-product in *Lates calcarifer* (Bloch, 1790) juveniles. Scientific Reports, 2021, 11: 49971

CHENG X F, JIANG G M, XIANG J, *et al*. Effects of dietary fishmeal replacement with meat and bone meal on the growth performance, blood physiological and biochemical indices, muscle chemical composition and texture characteristics in juvenile furong crucian carp (Furong carp♀ × Red crucian carp♂). Acta Hydrobiologica Sinica, 2020, 44(1): 85–94 [程小飞, 蒋国民, 向劲, 等. 饲料中猪肉骨粉替代鱼粉对芙蓉鲤鲫生长性能、血液生理生化、肌肉组成及质构特性的影响. 水生生物学报, 2020, 44(1): 85–94]

CHU X Y, LI P, LI H L, *et al*. Correlation analysis between moisture and texture in muscles of *Micropterus salmoides* during fasting temporary cultivation. Science and Technology of Food Industry, 2021, 42(13): 45–50 [鉏晓艳, 李湃, 李海蓝, 等. 加州鲈禁食暂养过程中肌肉水分与质构特性相关性分析. 食品工业科技, 2021, 42(13): 45–50]

CUI X S. Effects of fishmeal replacement by poultry by-product meal, black soldier fly larvae meal and *Clostridium autoethanogenum* protein on growth performance, protein metabolism and related gene expression for juvenile obscure. Master´s Thesis of Shanghai Ocean University, 2022 [崔锡帅. 鸡肉粉、黑水虻幼虫粉和乙醇梭菌蛋白替代鱼粉对暗纹东方鲀生长性能、蛋白代谢及相关基因表达的影响. 上海海洋大学硕士研究生学位论文, 2022]

DE PAULA T G, ALVES DE ALMEIDA F L, CARANI F R, *et al*. Rearing temperature induces changes in muscle growth and gene expression in juvenile pacu (*Piaractus mesopotamicus*). Comparative Biochemistry and Physiology B-Biochemistry and Molecular Biology, 2014, 169: 31–37

GAUDIOSO G, MARZORATI G, FACCENDA F, *et al*. Processed animal proteins from insect and poultry by-products in a fish meal-free diet for rainbow trout: Impact on intestinal microbiota and inflammatory markers. International Journal of Molecular Sciences, 2021, 22: 545411

GENG L N, ZHANG J F, MU W, *et al*. Replacing fishmeal protein with blended alternatives alters growth, feed utilization, protein deposition and gut micromorphology of humpback grouper, *Cromileptes altivelis*. Animal Feed Science and Technology, 2022, 292: 115434

GISBERT E, MOZANZADEH M T, KOTZAMANIS Y, *et al*. Weaning wild flathead grey mullet (*Mugil cephalus*) fry with diets with different levels of fish meal substitution. Aquaculture, 2016, 462: 92–100

GUO X R, DAI J H, ZHANG J Z, *et al*. Research progress on chicken meal and enzymatic hydrolysates of chicken

skeleton in aquatic feeds. Modern Journal of Animal Husbandry and Veterinary Medicine, 2022(2): 80–83 [郭小瑞, 戴济鸿, 张俊智, 等. 水产饵料中应用鸡肉粉及酶解鸡浆的研究进展. 现代畜牧兽医, 2022(2):80–83]

HANG Y, FU Y, JIN C X, *et al*. Effects of supplemental amino acids and bile acid in a completely replaced fish meal by enzymatically hydrolysed soybean meal diet on growth performance, liver health and fillet quality of rainbow trout (*Oncorhynchus mykiss*). Aquaculture Research, 2022, 53(9): 3297–3308

HAO T T, WANG J Y, PAN J M, *et al*. Effects of partial replacement of fish meal with ribose protein on growth performance, intestinal digestive enzyme activities, specific immune indices and physiological and biological indices of juvenile turbot (*Scophthalmus maximus* L.). Progress in Fishery Sciences, 2022, 43(1): 86–96 [郝甜甜, 王际英, 潘金敏, 等. 核糖蛋白替代鱼粉对大菱鲆幼鱼生长性能、肠道消化酶活性、免疫能力及血清生化指标影响. 渔业科学进展, 2022, 43(1): 86–96]

HUANG J B, ZHOU C P, XU F, *et al*. Effects of partial replacement of fish meal with porcine meat meal on growth performance, antioxidant status, intestinal morphology, gut microflora and immune response of juvenile golden pompano (*Trachinotus ovatus*). Aquaculture, 2022, 561: 738646

IRM M, TAJ S, JIN M, *et al*. Effects of replacement of fish meal by poultry by-product meal on growth performance and gene expression involved in protein metabolism for juvenile black sea bream (*Acanthoparus schlegelii*). Aquaculture, 2020, 528: 735544

JI Y, YANG Y, SUN S Q, *et al*. Insights into diet-associated oxidative pathomechanisms in inflammatory bowel disease and protective effects of functional amino acids. Nutrition Reviews, 2022, 81(1): 95–113

JING Y Y, CAI X C, XU Y Q, *et al*. Alpha-lipoic acids promote the protein synthesis of C2C12 myotubes by the TLR2/PI3K signaling pathway. Journal of Agricultural and Food Chemistry, 2016, 64(8): 1720–1729

KHALIL H S, MOMOH T, AL-KENAWY D, *et al*. Nitrogen retention, nutrient digestibility and growth efficiency of Nile tilapia (*Oreochromis niloticus*) fed dietary lysine and reared in fertilized ponds. Aquaculture Nutrition, 2021, 27(6): 2320–2332

KIM D H, SARBASSOV D D, ALI S M, *et al*. GbetaL, a positive regulator of the rapamycin-sensitive pathway required for the nutrient-sensitive interaction between raptor and mTOR. Molecular Cell, 2003, 11(4): 895–904

LI H J, XU C, SU Z L, *et al*. Replacing fishmeal with fish protein hydrolysate and terrestrial complex protein in the compound feed of *Trachinotus ovatus*. Progress in Fishery Sciences, 2022, 43(5): 205–216 [黎恒基, 徐超, 苏泽亮, 等. 卵形鲳鲹配合饲料中酶解鱼浆蛋白和陆生复合蛋白替代鱼粉的研究. 渔业科学进展, 2022, 43(5): 205–216]

LI L K, LIU X J, WANG Y, *et al*. Effects of alternate feeding between fish meal and novel protein diets on the intestinal health of juvenile largemouth bass (*Micropterus salmoides*). Aquaculture Reports, 2022, 23: 101023

LI M M, ZHAI S S, XIE Q, *et al*. Effects of dietary n-6:n-3 pufa ratios on lipid levels and fatty acid profile of cherry valley ducks at 15–42 days of age. Journal of Agricultural and Food Chemistry, 2017, 65(46): 9995–10002

LI S L, DAI M, QIU H J, *et al*. Effects of fishmeal replacement with composite mixture of shrimp hydrolysate and plant proteins on growth performance, feed utilization, and target of rapamycin pathway in largemouth bass, *Micropterus salmoides*. Aquaculture, 2021, 533: 736185

LI S L, DING G T, WANG A, *et al*. Replacement of fishmeal by chicken plasma powder in diets for largemouth bass (*Micropterus salmoides*): Effects on growth performance, feed utilization and health status. Aquaculture Nutrition, 2019, 25(6): 1431–1439

LI Y J, HUO Y W, DU Y, *et al*. Research progress on nutritional biology and formula feed of Largemouth bass (*Micropterus salmoides*). Aquaculture Nutrition, 2016(23): 39–44 [李永娟, 霍雅文, 杜瑜, 等. 大口黑鲈营养生物学及配合饲料研究进展. 饲料研究, 2016(23): 39–44]

LI Y Y, HU C B, ZHENG Y J, *et al*. The effects of dietary fatty acids on liver fatty acid composition and delta(6)-desaturase expression differ with ambient salinities in *Siganus canaliculatus*. Comparative Biochemistry and Physiology B-Biochemistry and Molecular Biology, 2008, 151(2): 183–190

LIANG H L, MOKRANI A, JI K, *et al*. Effects of dietary arginine on intestinal antioxidant status and immunity involved in Nrf2 and NF-kappaB signaling pathway in juvenile blunt snout bream, *Megalobrama amblycephala*. Fish and Shellfish Immunology, 2018, 82: 243–249

LIN S Q, CHENG L, ZHANG W B, *et al*. Effect of dietary protein content on the protein retention and nitrogenous metabolism of large yellow croaker. Periodical of Ocean University of China (Natural Science), 2014, 44(5): 32–39 [林淑琴, 成琳, 张文兵, 等. 饲料蛋白质水平对大黄鱼体内蛋白质的沉积和代谢反应的影响. 中国海洋大学学报(自然科学版), 2014, 44(5): 32–39]

MA X M, BLENIS J. Molecular mechanisms of mtor-mediated translational control. Nature Reviews Molecular Cell Biology, 2009, 10(5): 307–318

MA Y C, LI M M, XIE D Z, *et al*. Fishmeal can be replaced with a high proportion of terrestrial protein in the diet of the carnivorous marine teleost (*Trachinotus ovatus*). Aquaculture, 2020, 519: 734910

MOHAMMADI M, IMANI A, FARHANGI M, *et al*. Replacement of fishmeal with processed canola meal in

diets for juvenile nile tilapia (*Oreochromis niloticus*): Growth performance, mucosal innate immunity, hepatic oxidative status, liver and intestine histology. Aquaculture, 2020, 518: 734824

ROMMEL C, BODINE S C, CLARKE B A, *et al*. Mediation of IGF-1-induced skeletal myotube hypertrophy by PI(3)K/Akt/mTOR and PI(3)K/Akt/GSK3 pathways. Nature Cell Biology, 2001, 3(11): 1009–1013

SHENG Z Y, TURCHINI G M, XU J M, *et al*. Functional properties of protein hydrolysates on growth, digestive enzyme activities, protein metabolism, and intestinal health of larval largemouth bass (*Micropterus salmoides*). Frontiers in Immunology, 2022, 13: 913024

SHI Y, LENG X J, LI X Q. Effect of dietary protein levels on growth performance, whole body composition and intestinal digestive enzyme activities of *Cichlasoma* sp. juvenile. Acta Hydrobiologica Sinica, 2009, 33(5): 874–880 [石英, 冷向军, 李小勤, 等. 饲料蛋白水平对血鹦鹉幼鱼生长、体组成和肠道蛋白消化酶活性的影响. 水生生物学报, 2009, 33(5): 874–880]

TACON A G J, METIAN M. Global overview on the use of fish meal and fish oil in industrially compounded aquafeeds: Trends and future prospects. Aquaculture, 2008, 285(1/2/3/4): 146–158

TERADA N, PATEL H R, TAKASE K, *et al*. Rapamycin selectively inhibits translation of mrnas encoding elongation factors and ribosomal proteins. Proceedings of the National Academy of Sciences of the United States of America, 1994, 91(24): 11477–11481

WANG Z, QIAN X Q, XIE S Q, *et al*. Changes of growth performance and plasma biochemical parameters of hybrid grouper (*Epinephelus lanceolatus* male × *Epinephelus fuscoguttatus* female) in response to substitution of dietary fishmeal with poultry by-product meal. Aquaculture Reports, 2020, 18: 100516

WU R. Effects of replacement of dietary fishmeal by maggot meal on growth performance, digestion and intestinal healthy in Japanese seabass (*Lateolabrax japonicus*). Master´s Thesis of Jimei University, 2018 [吴瑞. 复合蛋白 KT67 替代鱼粉对花鲈生长、消化和肠道健康的影响. 集美大学硕士研究生学位论文, 2018]

XIAO J X, PENG X, SHAO Q J. Research progress of development and utilization of protein resources in aquatic feed. China Feed, 2011(5): 30–34 [肖金星, 彭翔, 邵庆均. 水产饲料蛋白源开发与利用的研究进展. 中国饲料, 2011(5): 30–34]

YU X T. Effects of dietary protein sources on feeding response, immunology response and flesh quality of grass carp (*Ctenopharyngodon idellus*). Master´s Thesis of Chinese Academy of Agricultural Sciences, 2016 [于晓彤. 不同蛋白源对草鱼摄食反应、免疫应答及肉质的影响研究. 中国农业科学院硕士研究生学位论文, 2016]

ZHAO P F, PENG X H, CHEN Y J, *et al*. Effects of dietary fermented mulberry leaves on growth performance, metabolism and antioxidant ability of juvenile *Micropterus salmoides*. Freshwater Fisheries, 2016, 46(6): 86–91 [赵鹏飞, 彭祥和, 陈拥军, 等. 高脂或低蛋白日粮中添加发酵桑叶对大口黑鲈生长、代谢与抗氧化能力的影响. 淡水渔业, 2016, 46(6): 86–91]

ZHU H, GONG G, WANG J, *et al*. Replacement of fish meal with blend of rendered animal protein in diets for siberian sturgeon (*Acipenser baerii* Brandt), results in performance equal to fish meal fed fish. Aquaculture Nutrition, 2011, 17(2): E389–E395

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Effects of Dietary Animal Protein Source Composition on the Growth Performance, Intestinal Health, and Protein Metabolism of Largemouth Bass (*Micropterus salmoides*)

JIANG Kunsheng[1,2#], CHEN Fang[1#], ZHANG Guanrong[1], HUANG Zekui[1], GUAN Junfeng[1], MA Yongcai[1], CHEN Yuhang[2], XIE Dizhi[1], LI Yuanyou[1①]

(1. *College of Marine Sciences, South China Agricultural University, Guangzhou* 510642*, China;* 2. *Guangdong Daynew Aquatic Sci-Tech Co., Ltd., Foshan* 528399*, China*)

Abstract Terrestrial animal protein sources contain less antinutritional factors, high protein content, and functional factors, which are beneficial to fish health. Among them, poultry byproduct meal (containing 65%–73% protein rich in vitamins) and porcine meat meal (containing 45%–60% protein and high contents of proline and glycine) are the most widely used meals in aquatic compound feeds, and are important fish meal replacement sources. As a carnivorous fish species, largemouth bass (*Micropterus salmoides*) is highly dependent on dietary fish meal, and the level of fish meal added in its commercial feeds is up to 50%. However, the rising price of fish meal increases the farming cost of *M. salmoides*. Therefore, it is necessary to identify a suitable alternative protein source to reduce the amount of dietary fish meal and the feed cost. Therefore, seven compound feeds (D1–D7) were prepared in this study. The added ratios of fish meal/poultry byproduct meal/porcine meat meal were as follows: 45.0/22.6/0, 37.1/22.6/8.0, 28.8/22.6/16.0, 45.0/14.5/8.0, 45.0/5.3/16.0, 41.6/18.0/8.0, and 37.0/13.8/16.0. Juvenile *M. salmoides* (initial body weight ~55 g) were fed the above diets for 60 days with five replicates in each group. The effects of the animal protein source combination on the growth performance, tissue biochemical indices, muscle texture characteristics, liver protein metabolism, and intestinal inflammatory factor-related gene expression were evaluated. The water temperature during the feeding trial was 27.4–32.3 ℃ and the ammonia nitrogen concentration was 0.1–0.2 mg/L. After the feeding experiment, three fish were randomly selected from each cage to collect the serum, liver, intestinal tract, muscle, and other samples, which were then stored at –80 ℃. In addition, three fish were randomly selected from each cage to determine their morphological indices. At the same time, two fish were selected from each cage to determine the muscle texture characteristics and the whole fish proximate composition. Physiological and biochemical indices of serum and liver tissues, albumin, urea nitrogen (BUN), total amino acid (T-AA), alanine aminotransferase (ALT), aspartate aminotransferase, total protein (TP), and blood ammonia (SA) levels), were determined using commercial kits, and the texture characteristics of muscle were determined by using a texture analyzer. The moisture, crude fat, crude protein, and ash contents of whole fish and muscle were determined by atmospheric drying, Soxhlet extraction, Kjeldahl nitrogen determination, and Muffle furnace incineration, respectively. Real-time quantitative PCR was used to determine the expression levels of genes related to liver protein metabolism and the intestinal inflammatory response. All test data were expressed as the mean±standard error, and multiple comparisons were made by the Tukey test, with $P<0.05$ indicating a significant difference. The results showed that, compared with other groups, the final body weight, weight gain rate, and specific growth rate of fish in the D3 group were significantly higher, and the feed conversion ratio was significantly lower ($P<0.05$). There were no

① Corresponding author: LI Yuanyou, E-mail: yyli16@scau.edu.cn

significant differences in the condition factor, hepatosomatic index, viscerosomatic index, and survival rate among all groups (P>0.05). The whole-body crude protein content in the D3 group was significantly higher than that in the D1 group, and the crude lipid level in the D3 group was significantly lower than that in the D6 group (P<0.05). In terms of tissue physiological and biochemical indices, there were no significant differences in the activities of BUN and ALT in serum, SA content, and ALT activity in the liver among all groups (P>0.05). The serum T-AA content of fish in the D3 group was significantly higher than that in the D1 and D4 groups (P < 0.05), but the AST activity in the D3 group was significantly lower than that in the D5 group (P<0.05). The liver TP content in the D3 group was significantly higher than that in the D7 group (P<0.05). There were no significant differences in the serum TP and liver ALT contents in the D3 group compared with those of the other groups (P>0.05). In terms of muscle quality, the muscle hardness, adhesion, and mastication in the D3 group were significantly lower than those in the D4 and D6 groups, respectively (P<0.05). There were no significant differences in the muscle adhesiveness, elasticity, cohesiveness, moisture, crude protein content, crude lipid content, and ash content among all groups (P>0.05). In addition, the mRNA expression levels of intestinal *il-10* and liver *tor*, *s6k1*, *akt*, and *pi3k* in the D3 group were upregulated, and were significantly higher than those in the D7 group (P<0.05). The mRNA expression levels of *il-1β* and *il-6* in the intestines and *4ebp-1* in the liver of the D3 group were significantly lower than those of the D1 group (P<0.05). These results indicated that combined use of 28.8% fish meal, 16.0% porcine meat meal, and 22.6% poultry byproduct meal had the best growth promotion effect on *M. salmoides*, and was able to improve liver protein synthesis and maintain intestinal health. The results of this study provided technical support for reducing the dependence of *M. salmoides* compound feed on fish meal.

Key words *Micropterus salmoides*; Animal protein source; Growth performance; Protein synthesis; Intestinal health