Effects of Arginine on Proliferation and Differentiation of Fish Muscle Cells and Their Mechanisms: Postprint

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Abstract

Arginine (Arg) serves as a functional amino acid and is also an essential amino acid for fish. Arg and some of its metabolites can influence the proliferation and differentiation of fish muscle cells by regulating the secretion of certain endocrine hormones [such as growth hormone (GH), insulin-like growth factors (IGFs), etc.], thereby affecting the expression of related genes and proteins during myogenesis. This paper provides a brief review of the effects of Arg on fish muscle cell proliferation and differentiation and their underlying mechanisms.

Full Text

Preamble

Effects of Arginine on Proliferation and Differentiation of Fish Myoblasts and Its Mechanisms

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Abstract: Arginine (Arg) is a functional amino acid and an essential amino acid for fish. Arg and its metabolites can influence the expression of genes and proteins involved in myogenesis by regulating the secretion of certain endocrine hormones [such as growth hormone (GH), insulin-like growth factors (IGFs), etc.], thereby affecting the proliferation and differentiation of fish myoblasts. This article provides a brief review of the effects of Arg on fish myoblast proliferation and differentiation and its underlying mechanisms.

Keywords: arginine; fish; myoblast; proliferation; differentiation

Fish skeletal muscle accounts for 30%–80% of total body mass [1], and body weight gain in fish is largely associated with increased skeletal muscle mass. Skeletal muscle is composed of post-mitotic multinucleated muscle fibers. Undifferentiated mononuclear myoblasts proliferate, exit the cell cycle, express specific proteins, fuse to form multinucleated myotubes, and eventually develop into mature muscle fibers. Therefore, myoblast proliferation and differentiation are critically important for skeletal muscle growth and development [2]. Arginine (Arg) is abundant in fish tissues and has endocrine-promoting effects [3-4]. Studies have shown that Arg and its metabolites [nitric oxide (NO), creatine] can directly or indirectly promote the secretion of growth hormone (GH), insulin-like growth factors (IGFs), and insulin (INS) in animals, regulate the expression of genes and proteins involved in myogenesis, and thereby influence myoblast proliferation and differentiation. This review summarizes the effects of Arg on fish myoblast proliferation and differentiation and its mechanisms, providing a reference for further research on Arg' s regulation of fish muscle growth.

1 Effects of Arg on Fish Myoblast Proliferation and Differentiation

Myoblasts proliferate through mitosis of spindle-shaped mononuclear stem cells located between the sarcolemma and basement membrane, ultimately resulting in increased myoblast numbers. Proliferating cell nuclear antigen (PCNA) is an indicator of cell proliferation status. Treatment of Atlantic salmon myoblasts with 2.87 mmol/L L-Arg significantly increased the percentage of PCNA-positive cells [5], indicating that Arg can promote fish myoblast proliferation. The fusion and differentiation of fish myoblasts lead to muscle fiber hypertrophy. Neu et al. [6] found that dietary L-Arg supplementation significantly increased the number of white muscle fibers with diameters greater than 20 m in juvenile tilapia, suggesting that Arg can stimulate muscle fiber hypertrophy and promote fish myoblast differentiation.

Cellular total nuclear density, which reflects cell proliferation, refers to the number of nuclei observed per unit area after hematoxylin-eosin (HE) staining. The fusion coefficient represents the percentage of nuclei in myotubes relative to the total nuclei in the culture medium, while surface myotube density refers to the ratio of myotube area to culture area; both parameters reflect the degree of myoblast fusion and differentiation. Studies have shown that treatment of mouse myoblasts (C2C12 cells) with 1 mmol/L L-Arg increased total nuclear density by 27%, while cell fusion coefficient and surface myotube density increased by 77% and 72%, respectively [7]. These results demonstrate that Arg can stimulate C2C12 cell proliferation and differentiation, promoting myotube formation.

2.1.1 NO Pathway

Arg is a precursor for NO synthesis in vivo, and fish can synthesize NO from Arg via nitric oxide synthase (NOS). Dietary L-Arg supplementation dose-dependently increased serum NO content in gibel carp [8]. NOS is the key enzyme in NO synthesis, and its activity reflects the capacity of tissues to produce NO. Studies have shown that dietary supplementation with 2.57% L-Arg increased serum total NOS activity by 30.9% in juvenile cobia [9], and liver total NOS activity in gibel carp increased with dietary L-Arg levels [8]. These findings indicate that Arg can enhance NOS activity and promote NO production.

NO is an important intracellular signaling molecule. When muscle tissue is damaged, muscle fibers release NO, which activates quiescent satellite cells and mediates myoblast proliferation and differentiation [10]. N-nitro-L-arginine methylester (L-NAME) is an NOS inhibitor that suppresses NOS activity and blocks NO production. Long et al. [7] co-treated C2C12 cells with 0.1 mmol/L L-NAME and 1 mmol/L L-Arg, resulting in decreases in total nuclear density, cell fusion coefficient, and surface myotube density by 15.8%, 90.5%, and 92.9%, respectively. This demonstrates that L-NAME can inhibit Arg' s effects on C2C12 cell proliferation and differentiation, and that blocking NO synthesis suppresses Arg' s promoting effect on C2C12 cell proliferation and differentiation. Therefore, Arg may promote myoblast proliferation and differentiation by upregulating NO synthesis.

Paired box 7 (pax7) is an essential upstream regulatory molecule for myoblast formation that regulates myoblast proliferation. Myogenic differentiation antigen (MyoD) is a gene associated with myoblast formation that is expressed during early myogenesis and participates in myoblast proliferation. Myogenin (MyoG) is expressed at the terminal stage of differentiation, controls the initiation of myoblast fusion, and is an essential factor for myotube and muscle fiber formation. Studies have found that co-treatment of chicken breast muscle cells with 0.05 mmol/L L-Arg and L-NAME significantly reduced pax7, MyoD, and MyoG mRNA expression [11]. This suggests that L-NAME can downregulate Arg' s promoting effect on the transcription of pax7, MyoD, and MyoG genes, thereby inhibiting myoblast proliferation and differentiation. Therefore, Arg may promote myoblast proliferation and differentiation by promoting NO production and upregulating the transcription of pax7, MyoD, and MyoG genes. However, whether Arg mediates fish myoblast proliferation and differentiation through the NO pathway remains to be investigated.

Interestingly, 2.5 mmol/L L-Arg can inhibit myotube atrophy caused by growth factor and nutrient deficiency, but co-treatment of C2C12 myotubes with 2.5 mmol/L L-Arg and 10 mmol/L L-NAME showed no significant change in myotube diameter compared to Arg treatment alone [12]. This indicates that L-Arg can also mediate myoblast fusion independently of NO. Therefore, besides the NO pathway, L-Arg may mediate myoblast proliferation and differentiation

through other pathways. However, no related reports exist in fish, and further research is needed.

2.1.2 Polyamine Pathway

Arg can be metabolized to produce ornithine, which is then converted to putrescine by ornithine decarboxylase (ODC). Putrescine is further metabolized to produce spermidine and spermine. These three compounds are collectively known as polyamines. Polyamines are important bioactive substances that participate in protein synthesis, cell proliferation and differentiation, and regulate gene expression. Muscle satellite cells can proliferate and self-renew after muscle injury. Mouse studies have shown that polyamine levels increase 2-fold after muscle incision [13], suggesting that polyamines may be involved in the proliferation of satellite cells following muscle injury. Actin polymerization forms polymeric filaments, which further assemble into microfilaments that participate in myofibril formation. Treatment with 0.5 mmol/L spermine and spermidine achieved 90% actin polymerization in rabbit muscle cells [14]. Erwin et al. [15] found that L6 myoblast differentiation was accompanied by increased putrescine and spermidine levels, and that putrescine could alleviate the inhibitory effect of an irreversible ODC inhibitor on L6 myoblast differentiation. These results indicate that polyamines are involved in the myoblast differentiation process.

Tu et al. [8] found that hepatic arginase activity in gibel carp increased dose-dependently with dietary L-Arg levels. This suggests that Arg can enhance arginase activity and promote the formation of polyamine precursors. Therefore, Arg may participate in regulating fish myoblast proliferation and differentiation by promoting polyamine generation. Intracellular ^{14}C-thymidine deoxynucleotide content can reflect cell proliferation activity. GC7 (N1-guanyl-1,7-diaminoheptane) is an inhibitor of deoxyhypusine synthase that blocks eukaryotic translation initiation factor 5A (eIF5A)-mediated translation initiation activated by spermidine. Studies have shown that 25 mol/L GC7 significantly reduced ^{14}C-thymidine deoxynucleotide content in C2C12 cells, and quantitative PCR (QPCR) detected a highly significant decrease in the expression of the specific protein myomesin 1 (Myom1), indicating that GC7 significantly inhibited C2C12 cell proliferation and differentiation [16]. Therefore, spermidine can enhance eIF5A activity, upregulate muscle cell protein synthesis, and promote myoblast proliferation and differentiation. However, the mechanism of polyamine action remains uncertain, and whether Arg affects fish myoblast proliferation and differentiation through polyamines requires further investigation.

2.1.3 Creatine Pathway

Arg generates creatine under the action of amidinotransferase. Creatine can potentially treat muscle diseases such as muscular dystrophy [17-18]. Treatment of C2C12 cells with 5 mmol/L creatine significantly increased myotube diameter [17] and fusion index by 40% [19]. Myogenic regulatory factor 4 (MRF4) is an important myogenic regulatory factor that controls myotube differentiation,

and its deficiency leads to muscle dysplasia. Treatment with 5 mmol/L creatine increased MyoD mRNA expression in C2C12 cells by 1.3-fold, while MyoG and MRF4 mRNA expression increased by 56% and 233%, respectively [17]. Additionally, creatine can enhance the synthesis of muscle-specific proteins such as myosin heavy chain in muscle cells [17]. These results suggest that Arg can promote C2C12 cell proliferation and differentiation. Therefore, Arg may promote myoblast proliferation and differentiation by upregulating the transcription of myogenic regulatory factors and the synthesis of related muscle-specific proteins through creatine. However, research on the effects of creatine on fish myoblast proliferation and differentiation is currently lacking.

The p38 mitogen-activated protein kinase (MAPK) signaling pathway is a cascade in the MAPK family that participates in regulating myoblast proliferation and differentiation. Creatine can accelerate C2C12 cell differentiation through the p38 pathway [19]. p38 has four isoforms (α, β, γ, δ), and experiments have demonstrated that p38α has the greatest impact on myoblast differentiation. p38α-deficient mouse myoblasts exhibit excessive proliferation, with a highly significant increase in mononuclear myoblasts and a highly significant decrease in myotube number [20]. This suggests that p38α may inhibit proliferation and promote differentiation in mouse myoblasts. Currently, the pathway through which Arg regulates p38 MAPK gene expression remains unclear. Elisabeth et al. [21] reported that Arg can upregulate p38 MAPK gene expression in Atlantic salmon liver, indicating that Arg can regulate p38 MAPK signaling pathway activity in fish liver. Whether Arg regulates fish myoblast proliferation and differentiation by upregulating muscle p38 MAPK gene expression through creatine remains to be investigated.

2.2 Arg May Indirectly Affect Fish Myoblast Proliferation and Differentiation by Regulating Endocrine Hormone Secretion

GH, IGFs, and INS are growth-related hormones in animals that can directly act on tissue cells, enhance metabolism, and promote cell growth and development.

2.2.1 GH

GH is produced by the pituitary gland and can stimulate fish myoblast proliferation and differentiation [22]. Studies have shown that dietary Arg supplementation significantly increased GH mRNA expression in the pituitary and serum GH levels in largemouth bass [3]. Therefore, Arg may affect myoblast proliferation and differentiation by promoting pituitary GH synthesis and secretion. However, L-Arg had no significant effect on serum GH levels in channel catfish [23], possibly due to differences in fish species and size. Arg increased pituitary GH mRNA expression and serum GH levels in gibel carp weighing 51.6 g but had no significant effect on fish weighing 147.8 g [8].

Fish skeletal muscle formation is regulated by several myogenic regulatory fac-

tors, with MyoD and Myf5 expressed during myoblast proliferation, while MyoG and MRF4 are expressed during differentiation. GH treatment of sea bream myoblasts significantly increased MyoD2 and Myf5 mRNA expression [24]. MyoG was overexpressed in muscle of GH-transgenic zebrafish [25]. Intraperitoneal injection of recombinant bovine GH in rainbow trout significantly increased myosin heavy chain protein expression in white muscle [26]. These studies indicate that GH can promote fish myoblast proliferation and differentiation by regulating the transcription levels of Myf5, MyoD, MyoG, and myosin heavy chain. Therefore, Arg may also promote fish myoblast proliferation and differentiation by regulating GH secretion to upregulate myogenic regulatory factor expression and related protein synthesis. Additionally, Uretsky et al. [27] demonstrated that NO can promote GH secretion in goldfish, suggesting that Arg may also mediate this process through NO.

GH binding to its receptor activates Janus kinase 2 (JAK2), which subsequently phosphorylates signal transducers and activators of transcription (STATs) that bind to DNA and regulate gene expression [28]. Whether Arg can upregulate Myf5, MyoD, and MyoG gene expression and promote fish myoblast proliferation and differentiation through the GH/JAK2/STATs pathway requires further investigation.

2.2.2 IGFs

The growth-promoting effects of GH can be mediated through IGFs. IGFs act on cells through autocrine and paracrine pathways [26] and play important roles in cell proliferation and differentiation. IGFs include two subtypes, IGF-I and IGF-II, both of which can promote sea bream myoblast proliferation and differentiation, although IGF-II has a stronger effect on proliferation than IGF-I [24], while IGF-I has a stronger effect on differentiation than IGF-II [29]. Current research on Arg' s effects on IGFs in fish has focused primarily on IGF-I. Tu et al. [8] and Chen et al. [3] demonstrated that dietary L-Arg supplementation significantly increased serum IGF-I levels and hepatic IGF-I mRNA expression in gibel carp and largemouth bass. However, Pohlenz et al. [23] reported that Arg had no significant effect on serum IGF-I levels in channel catfish, while hepatic IGF-I mRNA expression decreased significantly with increasing dietary L-Arg levels. These results suggest that Arg' s effects on IGFs vary among different fish species.

Jiménez-Amilburu et al. [29] found that recombinant human IGF-II treatment significantly upregulated MyoD2 mRNA expression in sea bream muscle cells, suggesting that IGF-II may promote sea bream myoblast proliferation by upregulating MyoD2 expression. Treatment with 100 nmol/L recombinant human IGF-I increased MyoG mRNA expression in sea bream myoblasts by 2-3 fold [29], indicating that IGFs can promote sea bream myoblast proliferation and differentiation by regulating MyoD2 and MyoG expression. Therefore, Arg may promote fish myoblast differentiation by upregulating IGF synthesis and secretion to increase expression of related myogenic regulatory factors. Additionally,

Arg can produce creatine during metabolism. Louis et al. [17] reported that 5 mmol/L creatine increased IGF-I mRNA expression in C2C12 cells by 133%. Therefore, Arg may upregulate IGF-I transcription through creatine to enhance myoblast proliferation and differentiation, while also suggesting that creatine' s promoting effect on C2C12 cell proliferation and differentiation may be achieved through IGF-I. No related research has been reported in fish.

Notably, Sotiropoulos et al. [30] suggested that GH may promote mouse myoblast fusion independently of IGF-I upregulation, meaning GH can directly act on myoblast fusion. Bower et al. [31] found that amino acids alone can stimulate increased IGF gene expression, indicating that IGFs' effects on myoblasts may not require GH participation, and that Arg may directly act on fish myoblasts to promote IGF synthesis. However, this hypothesis remains to be verified.

2.2.3 INS

INS is a peptide hormone with a similar peptide structure to IGF-I, and both belong to the insulin superfamily [32]. Erwin et al. [15] demonstrated that 1 mol/L insulin doubled the number of nuclei in L6 myotubes, indicating that insulin can stimulate L6 myoblast fusion and differentiation. Arg significantly increased serum insulin levels in rainbow trout [4] (intraperitoneal injection) and barfin flounder [33] (intramuscular injection). Therefore, Arg may promote myoblast differentiation by increasing insulin levels in fish. Polyamines participate in regulating myoblast proliferation and differentiation, and studies have shown that insulin can increase polyamine levels in L6 myoblasts [15], suggesting that Arg may promote myoblast proliferation and differentiation by increasing insulin secretion to induce polyamine generation. The mechanism by which Arg promotes insulin release in fish remains unclear. The brockmann body (BB) is a component of the endocrine pancreas in some teleost fish, containing pancreatic A, B, and D cells [4,34]. Researchers have speculated through in vitro studies that Arg may directly act on the BB [4,34] to increase insulin secretion. Therefore, Arg may promote myoblast differentiation by regulating insulin levels in fish, but the specific mechanism requires further investigation.

The mammalian target of rapamycin (mTOR) regulates protein transcription and translation, serves as an important hub for regulating cell growth and the cell cycle, and is a key functional factor in skeletal muscle [35]. IGF-I and INS can activate mTOR in fish myoblasts through the phosphatidylinositol 3-kinase/protein kinase B (PI3K/Akt) signaling cascade [36]. Ham et al. [12] found that rapamycin (an mTOR inhibitor) significantly inhibited myotube diameter increase and blocked Arg' s effects on C2C12 myotubes. This indicates that mTOR activation is a necessary step for Arg to promote myoblast fusion and differentiation. Arg can upregulate mTOR mRNA expression in muscle of gibel carp [8] and Jian carp [37]. Therefore, Arg may promote fish myoblast differentiation by directly activating mTOR [38] or indirectly by regulating growth factor levels (IGFs/INS, etc.) to activate mTOR through the

PI3K/Akt signaling pathway [36]. Wu et al. [35] suggested that mTOR may promote differentiation of goat skeletal muscle satellite cells by reducing STAT1 nuclear transport and decreasing the opportunity for STAT1 to bind to MyoD1, thereby promoting MyoD' s regulation of MyoG expression. Whether Arg can regulate fish myoblast proliferation and differentiation through similar pathways by activating mTOR in fish myoblasts remains to be investigated.

2.3 Arg May Affect Fish Myoblast Proliferation and Differentiation by Regulating Myostatin (MSTN)

MSTN, also known as growth differentiation factor-8, is a member of the transforming growth factor superfamily. MSTN is a negative regulator of mammalian skeletal muscle development and growth, expressed primarily in skeletal muscle, and can inhibit myoblast proliferation and differentiation [39]. However, current fish studies have found that 50 nmol/L recombinant mouse MSTN inhibited the increase in rainbow trout myoblast numbers while upregulating MyoG mRNA expression and myosin heavy chain levels [40]. This suggests that recombinant mouse MSTN may inhibit proliferation but promote differentiation in rainbow trout myoblasts [40]. However, 100 nmol/L recombinant human MSTN had no significant effect on rainbow trout myoblast differentiation. These results indicate that different MSTN types and doses produce different effects on rainbow trout myoblasts, and MSTN' s role in rainbow trout myoblast differentiation remains controversial. Notably, different MSTN subtypes may have different physiological functions in fish [40].

In avian studies, treatment of chicken breast muscle cells with 0.01 mmol/L L-Arg significantly reduced MTT values and increased MSTN mRNA expression [11], suggesting that Arg may inhibit chicken breast muscle cell proliferation activity by upregulating MSTN gene transcription. However, research on Arg' s effects on fish myoblast proliferation and differentiation through MSTN regulation is currently lacking.

In summary, Arg and its metabolites NO, polyamines, and creatine promote mammalian myoblast proliferation and differentiation, but whether they mediate fish myoblast proliferation and differentiation requires further investigation. Whether Arg can directly or indirectly activate p38 MAPK and mTOR signaling pathways to affect fish myoblast proliferation and differentiation by regulating the transcription and translation of myogenic regulatory factors and synthesis of muscle-specific proteins needs further study. Whether and how Arg may affect fish myoblast proliferation and differentiation by regulating MSTN also requires in-depth research.

References

[1] BONE Q. 6-locomotor muscle[J]. Fish Physiology, 1978, 7: 361–424.
[2] ZIMMERMAN A M, LOWERY M S. Hyperplastic development and hypertrophic growth of muscle fibers in the white seabass (Atractoscion nobilis)[J].

Journal of Experimental Zoology, 1999, 284(3): 299–308.
[3] CHEN N, JIN L, ZHOU H, et al. Effects of dietary arginine levels and carbohydrate-to-lipid ratios on mRNA expression of growth-related hormones in largemouth bass, Micropterus salmoides[J]. General and Comparative Endocrinology, 2012, 179(1): 121–127.
[4] MOMMSEN T P, MOON T W, PLISETSKAYA E M. Effects of arginine on pancreatic hormones and hepatic metabolism in rainbow trout[J]. Physiological and Biochemical Zoology: Ecological and Evolutionary Approaches, 2001, 74(5): 668–678.
[5] LATIF M S. Influence of nutritional factors on muscle development and texture of Atlantic salmon (Salmo salar L.): in vivo and in vitro studies[D]. Master Thesis. Ås, Norway: Norwegian University of Life Sciences, 2010.
[6] NEU D, BOSCOLO W, ZAMINHAN M, et al. Growth performance, biochemical responses, and skeletal muscle development of juvenile Nile tilapia, Oreochromis niloticus, fed increasing levels of arginine[J]. Journal of the World Aquaculture Society, 2016, 47(2): 248–259.
[7] LONG J H D, LIRA V A, SOLTOW Q A, et al. Arginine supplementation induces myoblast fusion via augmentation of nitric oxide production[J]. Journal of Muscle Research & Cell Motility, 2006, 27(8): 577–584.
[8] TU Y Q, XIE S Q, HAN D, et al. Dietary arginine requirement for gibel carp (Carassis auratus gibelio var. CAS) reduces with fish size from 50 g to 150 g associated with modulation of genes involved in TOR signaling pathway[J]. Aquaculture, 2015, 449: 37–47.
[9] REN M C, AI Q H, MAI K S. Dietary arginine requirement of juvenile cobia (Rachycentron canadum)[J]. Aquaculture Research, 2014, 45(2): 225–233.
[10] ANDERSON J E. A role for nitric oxide in muscle repair: nitric oxide-mediated activation of muscle satellite cells[J]. Molecular Biology of the Cell, 2000, 11(5): 1859–1874.
[11] LI Y, WANG Y, WILLEMS E, et al. In ovo L-arginine supplementation stimulates myoblast differentiation but negatively affects muscle development of broiler chicken after hatching[J]. Journal of Animal Physiology and Animal Nutrition, 2016, 100(1): 167–177.
[12] HAM D J, CALDOW M K, LYNCH G S, et al. Arginine protects muscle cells from wasting in vitro in an mTORC1-dependent and NO-independent manner[J]. Amino Acids, 2014, 46(12): 2643–2652.
[13] KAMINSKA A M, STERN L Z, RUSSELL D H. Polyamine metabolism in muscle: differential response to tenotomy and denervation in the soleus and gastrocnemius muscle of adult rats[J]. Experimental Neurology, 1982, 78(2): 331–339.
[14] LARQUÉ E, SABATER-MOLINA M, ZAMORA S. Biological significance of dietary polyamines[J]. Nutrition, 2007, 23(1): 87–95.
[15] ERWIN B G, EWTON D Z, FLORINI J R, et al. Polyamine depletion inhibits the differentiation of L6 myoblast cells[J]. Biochemical and Biophysical Research Communications, 1983, 114(3): 944–949.
[16] LUCHESSI A D, CAMBIAGHI T D, HIRABARA S M, et al. Involvement of eukaryotic translation initiation factor 5A (eIF5A) in skeletal muscle stem

cell differentiation[J]. Journal of Cellular Physiology, 2009, 218(3): 480–489.
[17] LOUIS M, VAN BENEDEN R, DEHOUX M, et al. Creatine increases IGF-I and myogenic regulatory factor mRNA in C2C12 cells[J]. FEBS Letters, 2004, 557(1): 243–247.
[18] TARNOPOLSKY M, MARTIN J. Creatine monohydrate increases strength in patients with neuromuscular disease[J]. Neurology, 1999, 52(4): 854.
[19] DELDICQUE L, THEISEN D, BERTRAND L, et al. Creatine enhances differentiation of myogenic C2C12 cells by activating both p38 and Akt/PKB pathways[J]. American Journal of Physiology: Cell Physiology, 2007, 293(4): C1263-C1271.
[20] PERDIGUERO E, RUIZ-BONILLA V, GRESH L, et al. Genetic analysis of p38 MAP kinases in myogenesis: fundamental role of p38α in abrogating myoblast proliferation[J]. The EMBO Journal, 2007, 26(5): 1245–1256.
[21] ELISABETH H, ESPE M, ANDERSEN S M, et al. A co-culture approach shows that polyamine turnover is affected during inflammation in Atlantic salmon immune and liver cells and that arginine and LPS exerts opposite effects on p38MAPK signaling[J]. Fish & Shellfish Immunology, 2014, 37(2): 286–298.
[22] BIGA P R, MEYER J. Growth hormone differentially regulates growth and growth-related gene expression in closely related fish species[J]. Comparative Biochemistry and Physiology Part A: Molecular & Integrative Physiology, 2009, 154(4): 465–473.
[23] POHLENZ C, BUENTELLO A, MILLER T, et al. Effects of dietary arginine on endocrine growth factors of channel catfish, Ictalurus punctatus[J]. Comparative Biochemistry and Physiology Part A: Molecular & Integrative Physiology, 2013, 166(2): 215–221.
[24] RIUS-FRANCINO M, ACERETE L, JIMÉNEZ-AMILBURU V, et al. Differential effects on proliferation of GH and IGFs in sea bream (Sparus aurata) cultured myocytes[J]. General and Comparative Endocrinology, 2011, 172(1): 44–49.
[25] KURADOMI R Y, FIGUEIREDO M A, LANES C F C, et al. GH overexpression causes muscle hypertrophy independent of local IGF-I in a zebrafish transgenic model[J]. Transgenic Research, 2011, 20(3): 513–521.
[26] BIGA P R, CAIN K D, HARDY R W, et al. Growth hormone differentially regulates muscle myostatin1 and-2 and increases circulating cortisol in rainbow trout (Oncorhynchus mykiss)[J]. General and Comparative Endocrinology, 2004, 138(1): 32–41.
[27] URETSKY A D, CHANG J P. Evidence that nitric oxide is involved in the regulation of growth hormone secretion in goldfish[J]. General and Comparative Endocrinology, 2000, 118(3): 461–470.
[28] GRACIELA P P, HUO J S, JESSICA S. Growth-hormone signal transduction[J]. Journal of Pediatric Endocrinology & Metabolism, 2002, 15(6): 771–786.
[29] JIMÉNEZ-AMILBURU V, SALMERÓN C, CODINA M, et al. Insulin-like growth factors effects on the expression of myogenic regulatory factors in gilthead sea bream muscle cells[J]. General and Comparative Endocrinology,

2013, 188: 151-158.
[30] SOTIROPOULOS A, OHANNA M, KEDZIA C, et al. Growth hormone promotes skeletal muscle cell fusion independent of insulin-like growth factor 1 up-regulation[J]. Proceedings of the National Academy of Sciences of the United States of America, 2006, 103(19): 7315-7320.
[31] BOWER N I, JOHNSTON I A. Transcriptional regulation of the IGF signaling pathway by amino acids and insulin-like growth factors during myogenesis in Atlantic salmon[J]. PLoS One, 2010, 5(6): e11100.
[32] BAÑOS N, MOON T W, CASTEJÓN C, et al. Insulin and insulin-like growth factor-I (IGF-I) binding in fish red muscle: regulation by high insulin levels[J]. Regulatory Peptides, 1997, 68(3): 181-187.
[33] ANDOH T. Stress inhibits insulin release induced by feeding and arginine injection in barfin flounder Verasper moseri[J]. Fisheries Science, 2014, 80(2): 311-316.
[34] RONNER P, SCARPA A. Secretagogues for pancreatic hormone release in the channel catfish (Ictalurus punctatus)[J]. General and Comparative Endocrinology, 1987, 65(3): 354-362.
[35] 吴海青. mTOR 信号通路对山羊骨骼肌卫星细胞增殖及分化的影响 [D]. 博士学位论文. 呼和浩特: 内蒙古大学, 2015.
[36] VÉLEZ E J, LUTFI E, JIMÉNEZ-AMILBURU V, et al. IGF-I and amino acids effects through TOR signaling on proliferation and differentiation of gilt-head sea bream cultured myocytes[J]. General and Comparative Endocrinology, 2014, 205: 296-304.
[37] CHEN G, FENG L, KUANG S, et al. Effect of dietary arginine on growth, intestinal enzyme activities and gene expression in muscle, hepatopancreas and intestine of juvenile Jian carp (Cyprinus carpio var. Jian)[J]. British Journal of Nutrition, 2012, 108(2): 195-207.
[38] SEILIEZ I, GABILLARD J C, SKIBA-CASSY S, et al. An in vivo and in vitro assessment of TOR signaling cascade in rainbow trout (Oncorhynchus mykiss)[J]. American Journal of Physiology: Regulatory, Integrative and Comparative Physiology, 2008, 295(1): R329-R335.
[39] 黄志清. Myostatin 负调控成肌细胞增殖和分化的信号转导机制研究 [D]. 博士学位论文. 雅安: 四川农业大学, 2008.
[40] GARIKIPATI D K, RODGERS B D. Myostatin inhibits myosatellite cell proliferation and consequently activates differentiation: evidence for endocrine-regulated transcript processing[J]. Journal of Endocrinology, 2012, 215(1): 177-187.

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