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RFLP-PCR Analysis of Growth Hormone and Myostatin Gene Polymorphism in *Pangasius hypophthalmus* and *Clarias gariepinus*: Implications for Growth Performance in Aquaculture

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ABSTRACT

Growth-related traits, such as body weight and length, are influenced by key genes, including the Growth Hormone (GH) gene, which promotes somatic growth, and the Myostatin (MSTN) gene, a negative regulator of muscle development. This study investigated the genetic polymorphisms of the Growth Hormone (GH) and Myostatin (MSTN) genes in two economically important aquaculture species, *Pangasius hypophthalmus* (striped catfish) and *Clarias gariepinus* (African catfish), using restriction fragment length polymorphism (RFLP) analysis combined with polymerase chain reaction (PCR). A total of 300 fish (150 per species) were sampled from commercial farms in Southwest Nigeria, and genomic DNA was extracted from fin tissue. PCR amplification of the GH and MSTN genes was followed by RFLP digestion using *HaeIII* and *EcoRI* enzymes, respectively. Genotypic and allelic frequencies were calculated, and growth performance (body weight and total length) was analyzed across genotypes. The results revealed distinct polymorphic patterns in both genes, with the heterozygous genotypes (GH: AB; MSTN: CD) being the most prevalent in both species. In *P. hypophthalmus*, the AB genotype for the GH gene occurred at a frequency of 50%, while in *C. gariepinus*, the AB genotype was observed at 40%. For the MSTN gene, the CD genotype was dominant in both species, with frequencies of 60% in *P. hypophthalmus* and 70% in *C. gariepinus*. Growth performance analysis demonstrated that heterozygous individuals (GH: AB; MSTN: CD) exhibited significantly higher mean body weight and total length compared to homozygous counterparts ($P < 0.05$). For example, *P. hypophthalmus* individuals with the AB genotype had a mean body weight of 335 ± 20 g and total length of 23.0 ± 1.8 cm, outperforming AA and BB genotypes. Similarly, *C. gariepinus* individuals with the CD genotype showed superior growth (425 ± 30 g, 27.0 ± 2.3 cm) compared to CC and DD genotypes. These findings highlight the potential of GH and MSTN polymorphisms as molecular markers for selective breeding programs. The observed heterozygote advantage suggests that maintaining genetic diversity in breeding populations may enhance growth efficiency and productivity in aquaculture.

Keywords: Growth Hormone (GH), Myostatin (MSTN), *Pangasius hypophthalmus*, *Clarias gariepinus*

INTRODUCTION

Recent advancements in molecular genetics have significantly enhanced aquaculture breeding programs by enabling the precise selection of desirable traits such as growth rate, feed efficiency, and muscle development (Robinson *et al.*, 2020). Among the key genetic regulators of fish growth are the Growth Hormone (GH) gene, which stimulates somatic growth, and the Myostatin (MSTN) gene, a negative regulator of muscle development (Gao *et al.*, 2021; Li *et al.*, 2022). Polymorphisms in these genes have been shown to significantly influence growth performance, making them critical targets for genetic

improvement in aquaculture (Wang *et al.*, 2023). Molecular methods like restriction fragment length polymorphism (RFLP) analysis, when paired with polymerase chain reaction (PCR), offer a quick and dependable way to assess genetic variation and uncover links between genotypes and growth characteristics (Zhang *et al.*, 2021).

Pangasius hypophthalmus (striped catfish) and *Clarias gariepinus* (African catfish) are two economically important species in global aquaculture, valued for their rapid growth rates and adaptability to diverse environmental conditions (FAO, 2022). In Africa, *C. gariepinus* is particularly significant due to its hardiness and suitability for small-scale and commercial aquaculture systems (Akinwande *et al.*, 2021). However, significant variations in growth performance exist between these species, highlighting the need for a deeper understanding of the genetic factors underlying these differences. Identifying genetic markers associated with growth traits can facilitate the development of targeted breeding strategies, ultimately enhancing productivity and sustainability in aquaculture (Nguyen *et al.*, 2023; Adeleke *et al.*, 2022). This study focuses on characterizing polymorphisms in the GH and MSTN genes of *P. hypophthalmus* and *C. gariepinus* using RFLP-PCR analysis. By examining the genetic variability within these key regulatory genes, the study aims to provide insights into the molecular mechanisms driving growth performance in these species.

MATERIALS AND METHODS

Fish Sampling and Tissue Collection

A total of 300 catfish, comprising 150 *Pangasius hypophthalmus* and 150 *Clarias gariepinus*, were sampled from three commercial fish farms in Southwest Nigeria between March 2024 and May 2024. Farms were selected based on production scale and accessibility. Fish were randomly selected and measured for body weight (g) and total length (cm) using a digital weighing scale and an ichthyometer, respectively. Fin clips (~0.5 cm^2) were aseptically excised using sterilized surgical scissors, placed in 1.5 mL microcentrifuge tubes containing 95% ethanol, and stored at –20°C until DNA extraction. All procedures adhered to ethical guidelines for animal research, with approval from Tai Solarin University of Education Animal Care and Use Research Ethics Committee (Approval No. TASUED-ACUREC/2024/045).

Genomic DNA Extraction

Genomic DNA was extracted from approximately 20 mg of fin tissue using a modified phenol-chloroform method (Sambrook & Russell, 2001). Briefly, tissue samples were chopped and lysed in 500 µL lysis buffer (50 mM Tris-HCl, 100 mM EDTA, 100 mM NaCl, 1% SDS) with 20 µL proteinase K, then incubated at 55°C for 3 hours. After digestion, phase separation was performed by adding 500 µL phenol:chloroform:isoamyl alcohol (25:24:1), followed by centrifugation at 12,000 × g for 10 minutes. The aqueous phase was transferred to a new tube, and DNA was precipitated with 1/10 volume of 3 M sodium acetate (pH 5.2) and 2.5 volumes of cold absolute ethanol. The DNA pellet was washed with 70% ethanol, air-dried, and resuspended in 50 µL TE buffer (10 mM Tris-HCl, 1 mM EDTA, pH 8.0). DNA concentration and purity were determined using a NanoDrop spectrophotometer (Thermo Fisher Scientific), and integrity was checked by 1% agarose gel electrophoresis.

Polymerase Chain Reaction (PCR) Amplification

The growth hormone (GH) and myostatin (MSTN) genes were amplified by PCR using specific primers (Table 1). The PCR reactions were carried out in a final volume of 25 µL, containing 50 ng of genomic DNA, 0.2 µM of each primer, 200 µM dNTPs, 1.5 mM $MgCl_2$, 1× PCR buffer, and 1 unit of Taq DNA polymerase (Thermo Fisher Scientific). The PCR cycling conditions were as follows: **For the GH gene**: initial denaturation at 95°C for 5 minutes, followed by 35 cycles of denaturation at 95°C for 30 seconds, annealing at 58°C for 30 seconds, extension at 72°C for 45 seconds, and a final extension at 72°C for 5 minutes.

For the MSTN gene: the same conditions were used, except with an annealing temperature of 56°C. PCR products were visualized on 1.5% agarose gels stained with ethidium bromide and documented using a GelDoc imaging system (Bio-Rad).

Restriction Fragment Length Polymorphism (RFLP) Analysis

RFLP digestion was performed using HaeIII for the GH gene and EcoRI for the MSTN gene. Each 20 µL digestion reaction contained 10 µL of PCR product, 2 µL of the respective restriction enzyme buffer, 0.5 µL of restriction enzyme (10 U), and sterile nuclease-free water. Samples were incubated at 37°C for 2 hours. The digested products were separated on 3% agarose gels in 1× TBE buffer at 100 V for 1 hour and visualized under UV light. The expected fragment sizes for each allele are detailed in Table 1.

Statistical Analysis

Genotypic and allelic frequencies were calculated for both genes. Growth performance data (body weight and total length) were analyzed using one-way analysis of variance (ANOVA) in SPSS v22 (IBM Corp.), with genotype as the independent variable. Normality was checked using the Shapiro-Wilk test, and homogeneity of variance was

verified with Levene's test. Post hoc comparisons were performed using Tukey's HSD test when significant differences were detected. Statistical significance was considered at $P < 0.05$.

PCR Amplification and RFLP Profiles

Successful amplification of the target gene fragments was confirmed by gel electrophoresis. Digestion with HaeIII and EcoRI produced clear banding patterns, allowing unambiguous assignment of genotypes.

Table 1: PCR Primers and RFLP Enzymes for GH and MSTN Genes

Gene	Primer (Forward)	Primer (Reverse)	Annealing Temp.	Restriction Enzyme	Expected Fragment Sizes (bp)
Growth Hormone	5'-AGTCTGACTCCAGTGGCTTC-3'	5'-CTTGTGATGGTGAGCTGGTG-3'	58°C	HaeIII	Allele A: 150 & 200; Allele B: intact 350
Myostatin	5'-TGACTCGATGACCTGACTTC-3'	5'-CACAGTCTGGCTAGCTTGGT-3'	56°C	EcoRI	Allele C: 180 & 170; Allele D: intact 350

RESULTS

Genotype and Allele Frequencies for the GH Gene

Table 2 shows the distribution of GH gene genotypes in *Pangasianodon hypophthalmus* and *Clarias gariepinus*. In *P. hypophthalmus*, the AB genotype is the most prevalent (50%), followed by AA (40%) and BB (10%). The frequency of allele A is 0.65, while allele B has a frequency of 0.35. In *C. gariepinus*, the AB genotype is also the most frequent (40%), with the AA and BB genotypes both occurring at 30%. Allele A and B each have a frequency of 0.50.

Table 2: Genotype and Allele Frequencies for the GH Gene

Species	Genotype	n	Frequency (%)	Allele A Frequency	Allele B Frequency
P. hypophthalmus	AA	20	40.0	0.65	0.35
	AB	25	50.0		
	BB	5	10.0		
C. gariepinus	AA	15	30.0	0.50	0.50
	AB	20	40.0		
	BB	15	30.0		

Genotype and Allele Frequencies for the MSTN Gene

Table 3 shows the genotype and allele frequencies for the MSTN gene in both fish species. In *P. hypophthalmus*, the CD genotype is the most frequent (60%), while CC and DD occur at 20% each. The allele C and D frequencies are equal at 0.50. For *C. gariepinus*, CD is the most common (70%), followed by DD (20%) and CC (10%). The allele C frequency is 0.45, while allele D is 0.55.

Table 3: Genotype and Allele Frequencies for the MSTN Gene

Species	Genotype	n	Frequency (%)	Allele C Frequency	Allele D Frequency
P. hypophthalmus	CC	10	20.0	0.50	0.50
	CD	30	60.0		
	DD	10	20.0		
C. gariepinus	CC	5	10.0	0.45	0.55
	CD	35	70.0		
	DD	10	20.0		

Growth Performance by GH Genotype

Table 4, presents body weight and total length across GH gene genotypes. In both species, fish with the AB genotype exhibit the highest mean body weight and length, followed by AA, while BB individuals have the lowest values.

Table 4: Growth Performance by GH Genotype

Species	Genotype	Body Weight (g) Mean ± SE	Total Length (cm) Mean ± SE
P. hypophthalmus	AA	310 ± 15	22.0 ± 1.5
	AB	335 ± 20	23.0 ± 1.8
	BB	295 ± 18	21.5 ± 1.2
C. gariepinus	AA	400 ± 25	26.0 ± 2.0
	AB	420 ± 30	27.0 ± 2.2
	BB	390 ± 22	25.5 ± 1.8

Growth Performance by MSTN Genotype

Table 5, displays the growth performance of different MSTN genotypes. Similar to the GH gene, individuals with the heterozygous CD genotype show the highest mean body weight and total length, while DD individuals have the lowest values.

Table 5: Growth Performance by MSTN Genotype

Species	Genotype	Body Weight (g) Mean ± SE	Total Length (cm) Mean ± SE
P. hypophthalmus	CC	315 ± 15	22.5 ± 1.6
	CD	330 ± 20	23.0 ± 1.7
	DD	300 ± 18	21.8 ± 1.5
C. gariepinus	CC	410 ± 28	26.2 ± 2.1
	CD	425 ± 30	27.0 ± 2.3
	DD	395 ± 24	25.8 ± 1.9

Table 6: ANOVA and Post Hoc Test Results for Growth Performance across GH and MSTN Genotypes

Species	Gene	Genotype	n	Body Weight (g) Mean ± SD	Total Length (cm) Mean ± SD	F-value	P-value	Significant Difference (Tukey's HSD)
P. hypophthalmus	GH	AA	20	280 ± 25	20.1 ± 1.5	14.62	0.000***	AB > AA, BB
		AB	25	335 ± 20	23.0 ± 1.8			
		BB	5	290 ± 30	21.0 ± 2.0			
	MSTN	CC	15	295 ± 22	21.5 ± 1.7	10.45	0.001**	CD > CC, DD
		CD	30	340 ± 18	23.5 ± 1.5			
		DD	5	310 ± 26	22.1 ± 2.2			
C. gariepinus	GH	AA	15	370 ± 28	25.2 ± 2.0	13.87	0.000***	AB > AA, BB
		AB	20	420 ± 25	27.5 ± 2.3			
		BB	15	375 ± 30	25.5 ± 2.5			
	MSTN	CC	10	390 ± 20	25.6 ± 1.9	11.25	0.001**	CD > CC, DD
		CD	30	425 ± 30	27.0 ± 2.3			
		DD	10	400 ± 22	25.8 ± 2.1			

****P < 0.001; *P < 0.01; P < 0.05; n = number of individuals per genotype; Tukey's HSD test shows heterozygous genotypes (AB, CD) consistently had significantly higher growth performance than homozygous genotypes.*

Table 6, presents the results of the analysis of variance (ANOVA) and post hoc Tukey's HSD tests assessing the effects of GH and MSTN gene polymorphisms on growth traits, body weight and total length in *Pangasius hypophthalmus* and *Clarias gariepinus*. Significant differences ($P < 0.01$) were observed among the genotypes for both genes in each species. In both fish species, heterozygous genotypes (AB for GH and CD for MSTN) exhibited significantly higher mean body weight and total length compared to their respective homozygous counterparts (AA, BB for GH; CC, DD for MSTN). For instance, in *P. hypophthalmus*, individuals with the AB

genotype for GH had a mean body weight of 335g and a total length of 23.0 cm, which was significantly higher than those with AA or BB genotypes. A similar trend was observed in *C. gariepinus*, where AB (GH) and CD (MSTN) genotypes showed superior growth traits. These findings suggest that heterozygosity at the GH and MSTN loci may confer a growth advantage, indicating potential markers for selective breeding programs in aquaculture.

DISCUSSION OF THE FINDINGS

This study investigated the genotype and allele frequencies of the Growth Hormone (GH) and Myostatin (MSTN) genes in *Pangasianodon hypophthalmus* and *Clarias gariepinus*, revealing significant associations between specific genotypes and growth performance. These findings contribute to the growing body of knowledge on the genetic basis of growth traits in aquaculture species and provide valuable insights for selective breeding programs.
The GH gene exhibited a predominance of the heterozygous AB genotype in *P. hypophthalmus* (50%), while *C. gariepinus* displayed a more balanced distribution of genotypes (AA: 30%, AB: 40%, BB: 30%). This variation likely reflects species-specific evolutionary pressures and breeding practices. For instance, the higher frequency of Allele A in *P. hypophthalmus* (0.65) compared to *C. gariepinus* (0.50) suggests a possible fixation of beneficial alleles in the former, possibly due to intensive selection for growth traits in farmed populations (Liu *et al.*, 2020). Similar patterns have been observed in Nile tilapia (*Oreochromis niloticus*), where allelic shifts were linked to improved growth performance (Maluwa & Gjerde, 2006).

For the MSTN gene, the heterozygous CD genotype was the most prevalent in both species, with a notably higher frequency in *C. gariepinus* (70%) than in *P. hypophthalmus* (60%). This suggests a potential selective advantage for the heterozygous genotype, which may provide an optimal balance of MSTN gene expression for muscle development (Biga *et al.*, 2015). The slightly higher frequency of Allele D in *C. gariepinus* (0.55) compared to *P. hypophthalmus* (0.50) further supports the role of selective breeding in shaping genetic composition. These findings align with studies on channel catfish (*Ictalurus punctatus*), where MSTN polymorphisms were associated with enhanced growth performance in hybrid breeding programs (Danzmann *et al.*, 2016).

The observed relationship between GH and MSTN genotypes and growth performance is consistent with prior research on growth-related genes in aquaculture species. In both *P. hypophthalmus* and *C. gariepinus*, the heterozygous genotypes (GH: AB; MSTN: CD) exhibited significantly higher body weight and total length compared to homozygous counterparts. This pattern suggests an overdominance or heterozygote advantage effect, a well-documented phenomenon in livestock and aquaculture genetics (Rahman *et al.*, 2019).

For the GH gene, *P. hypophthalmus* individuals with the AB genotype had an average body weight of 335 ± 20 g and total length of 23.0 ± 1.8 cm, outperforming AA (310 ± 15 g, 22.0 ± 1.5 cm) and BB (295 ± 18 g, 21.5 ± 1.2 cm) genotypes. A similar trend was observed in *C. gariepinus*, where AB individuals displayed superior growth (420 ± 30 g, 27.0 ± 2.2 cm) compared to AA (400 ± 25 g, 26.0 ± 2.0 cm) and BB (390 ± 22 g, 25.5 ± 1.8 cm) genotypes. These findings align with research on GH polymorphisms in other fish species, where heterozygous variants were linked to enhanced somatic growth and metabolic efficiency (Pillay *et al.*, 2021; Adeleke *et al.*, 2022). Moreover, GH plays a crucial role in regulating growth by stimulating the production of insulin-like growth factors (IGFs), which influence cell proliferation and muscle differentiation (Reinecke, 2010). The ability to identify and select for favorable GH alleles allows for accelerated growth in commercial strains, especially when used in genomic selection schemes (Vallejo *et al.*, 2017).

Similarly, for the MSTN gene, heterozygous CD individuals outperformed homozygous CC and DD individuals in both species. In *P. hypophthalmus*, the CD genotype was associated with a mean body weight of 330 ± 20 g and a total length of 23.0 ± 1.7 cm, compared to CC (315 ± 15 g, 22.5 ± 1.6 cm) and DD (300 ± 18 g, 21.8 ± 1.5 cm). *C. gariepinus* followed a similar pattern, where CD fish had the highest body weight (425 ± 30 g) and length (27.0 ± 2.3 cm). This supports previous studies that identified MSTN polymorphisms as crucial regulators of muscle development, with heterozygous individuals often displaying increased muscle fiber density and growth efficiency (Gabillard *et al.*, 2013; Li *et al.*, 2022). Myostatin, being a negative regulator of muscle growth, has been targeted in several aquaculture breeding programs. Loss-of-function or reduced-expression variants of MSTN have been shown to enhance muscle mass in species like zebrafish, rainbow trout, and tilapia (Lee *et al.*, 2016; Wang *et al.*, 2018). Such evidence emphasizes the gene's importance as a functional candidate for genetic improvement.

The results of this study highlight the potential utility of GH and MSTN polymorphisms as molecular markers for selective breeding. The heterozygous advantage observed in both genes suggests that breeding programs could focus on maintaining genetic diversity rather than selecting for homozygosity. Studies on genetically improved farmed tilapia (GIFT) and other aquaculture species have demonstrated that maintaining genetic variability in

growth-related loci enhances production efficiency and adaptability (Eknath & Hulata, 2009). The use of marker-assisted selection (MAS) incorporating GH and MSTN markers can significantly increase selection accuracy and shorten breeding cycles, thereby reducing production costs and improving profitability in commercial aquaculture (Houston *et al.*, 2020).

In the African context, where *C. gariepinus* is a key species for small-scale and commercial aquaculture, these findings are particularly relevant. By incorporating GH and MSTN markers into breeding programs, African aquaculture stakeholders can enhance growth performance and productivity, contributing to food security and economic development (Akinwande *et al.*, 2021; Adeleke *et al.*, 2022). Furthermore, integrating molecular genetics into traditional breeding methods has the potential to address challenges of low productivity, inconsistent performance, and genetic bottlenecks faced by hatchery systems in developing countries (FAO, 2020).

The results ANOVA and Post Hoc Test Results for Growth Performance across GH and MSTN Genotypes clearly demonstrate that polymorphisms in the GH and MSTN genes significantly influence growth performance in both *Pangasius hypophthalmus* and *Clarias gariepinus*, with heterozygous genotypes (AB for GH and CD for MSTN) exhibiting superior body weight and total length. This pattern is consistent with previous findings in aquaculture genetics, where heterozygosity often confers phenotypic advantages due to increased genetic diversity and hybrid vigor. For example, Falco *et al.* (2020) reported that GH gene polymorphisms in *Oreochromis niloticus* were associated with enhanced growth rates, especially in heterozygous individuals, making such loci valuable in selective breeding programs. Similarly, a study by Dong *et al.* (2021) on *Cyprinus carpio* found that MSTN gene variants significantly affected muscle development, and fish with heterozygous MSTN genotypes exhibited better growth traits compared to homozygous types. These findings align with the current study's observation that AB and CD genotypes in *P. hypophthalmus* and *C. gariepinus* had significantly higher body weights (e.g., 335 g and 420 g, respectively) and longer total lengths. The implication of these results is significant for aquaculture: incorporating GH and MSTN polymorphism screening in breeding strategies can optimize fish growth and productivity. As highlighted by Martínez *et al.,* (2022), genetic markers like GH and MSTN can improve selection accuracy and accelerate genetic gains in aquaculture species. Therefore, the consistent growth superiority of heterozygotes across species in this study supports the use of marker-assisted selection to enhance performance traits in cultured fish populations. With the ongoing advances in genomics and CRISPR-based gene editing, these polymorphic loci may eventually serve not just for selection but for direct trait manipulation, ushering in a new era of precision breeding in aquaculture (Zheng *et al.*, 2022).

CONCLUSION

This study confirms the existence of GH and MSTN polymorphisms in *P. hypophthalmus* and *C. gariepinus*, as well as their significant relationship with growth performance. The observed heterozygous advantage indicates that preserving genetic diversity in breeding populations could improve growth efficiency. These results are consistent with global research in aquaculture genetics and lay the groundwork for future breeding strategies focused on enhancing fish growth performance using molecular marker-assisted selection.

REFERENCES

Adeleke, B., Robertson-Andersson, D., & Moodley, G. (2022). Genetic improvement of African catfish (*Clarias gariepinus*) for sustainable aquaculture in sub-Saharan Africa. *Aquaculture Reports*, 25, 101-112.

Akinwande, A. A., Fagbenro, O. A., & Adebayo, O. T. (2021). Advances in the breeding and genetics of African catfish (*Clarias gariepinus*): A review. *Aquaculture Research*, 52(6), 2375-2388.

Biga, P. R., Schelling, G. T., Hardy, R. W., Cain, K. D., Overturf, K., & Ott, T. L. (2015). The effects of growth hormone transgenesis on muscle growth and gene expression in rainbow trout (*Oncorhynchus mykiss*). *Comparative Biochemistry and Physiology Part D: Genomics & Proteomics*, 14, 22-35.

Danzmann, R. G., Li, S., & Rexroad, C. E. (2016). Myostatin profiles were compared between fast and slow-growing rainbow trout across seasonal gradients . *Aquaculture Research, 47*(1), 123–137. https://doi.org/10.1111/are.12976

Dong, Z., Zhang, Y., & Jiang, J. (2021). Effects of Myostatin gene polymorphisms on growth and carcass traits in common carp *(Cyprinus carpio). Aquaculture Reports, 19*, 100620. https://doi.org/10.1016/j.aqrep.2020.100620

Eknath, A. E., & Hulata, G. (2009). Use and exchange of genetic resources of Nile tilapia (*Oreochromis niloticus*). *Reviews in Aquaculture*, 1(4), 197–213

Falco, M., Marquez, M., & Hernández, D. (2020). Association of growth hormone gene polymorphisms with growth traits in Nile tilapia (*Oreochromis niloticus). Aquaculture Research, 51*(1), 135–143. https://doi.org/10.1111/are.14304

FAO. (2020). *The state of world fisheries and aquaculture 2020: Sustainability in action*. Food and Agriculture

Organization of the United Nations. https://www.fao.org/documents/card/en/c/ca9229en

FAO. (2022). *The State of World Fisheries and Aquaculture 2022*. Rome.

Gabillard, J. C., Weil, C., Rescan, P. Y., Navarro, I., Gutierrez, J., & Le Bail, P. Y. (2013). Regulation of protein metabolism by insulin and GH in rainbow trout cells. *General and Comparative Endocrinology*, 183, 1-9.

Gao, Y., Li, S., & Wang, X. (2021). The role of myostatin in muscle growth and development in fish: A review. *Aquaculture Reports*, 20, 100712.

Houston, R. D., Bean, T. P., Macqueen, D. J., Gundappa, M. K., Jin, Y. H., Jenkins, T. L.& Primmer, C. R. (2020). Harnessing genomics to fast-track genetic improvement in aquaculture. *Nature Reviews Genetics, 21*(7), 389–409. https://doi.org/10.1038/s41576-020-0227-y

Lee, S. J., & McPherron, A. C. (2011). Regulation of myostatin activity and muscle growth. *Annual Review of Cell and Developmental Biology, 27*, 133–153. https://doi.org/10.1146/annurev-cellbio-092910-154002

Li, J., Zhang, H., & Chen, J. (2022). Genetic regulation of growth hormone in fish: Implications for aquaculture. *Reviews in Aquaculture*, 14(2), 789-805.

Maluwa, A. O., & Gjerde, B. (2006). Genetic evaluation of four strains of *Oreochromis shiranus* for harvest body weight in a diallel cross. *Aquaculture, 259*(1–4), 28–37. https://doi.org/10.1016/j.aquaculture.2006.06.003

Martínez, P., Robledo, D., & Houston, R. D. (2022). Applications of genomics in aquaculture breeding and conservation. *Fish and Fisheries, 23*(2), 333–349. https://doi.org/10.1111/faf.12623

Nguyen, T. T. T., Nguyen, H. Q., & Tran, M. T. (2023). Advances in genetic improvement of aquaculture species in Asia: Progress and challenges. *Aquaculture*, 567, 739-751.

Pillay, T. V. R., Kutty, M. N., & Pillay, T. (2021). *Aquaculture: Principles and Practices*. Wiley-Blackwell.

Rahman, M. A., Uddin, M. N., & Shahjahan, M. (2019). Genetic improvement of fish for aquaculture: A review. *Aquaculture International*, 27(2), 351-369.

Reinecke, M. (2010). Influence of the somatotropic axis on the immune system of teleost fish. *Frontiers in Endocrinology, 1*, 15. https://doi.org/10.3389/fendo.2010.00015

Robinson, N., Baranski, M., & Gjedrem, T. (2020). The role of genomics in aquaculture breeding programs. *Aquaculture Genomics*, 12(3), 45-60.

Vallejo, R. L., Leeds, T. D., Gao, G., Parsons, J. E., Martin, K. E., Evenhuis, J. P., ... & Wiens, G. D. (2017). Genomic selection models for improving disease resistance in rainbow trout. *Frontiers in Genetics, 8*, 122. https://doi.org/10.3389/fgene.2017.00122

Wang, Y., Liu, Z., & Xu, P. (2023). Polymorphisms in growth-related genes and their association with growth performance in aquaculture species. *Aquaculture Research*, 54(1), 1-15.

Wang, Y., Zhang, Z., Liu, J., Huang, X., Li, Y., Zhou, G., & Gui, J. (2018). Targeted disruption of the myostatin gene in yellow catfish (*Pelteobagrus fulvidraco*) using CRISPR/Cas9. *Scientific Reports, 8*, 9276. https://doi.org/10.1038/s41598-018-27649-w

Zhang, X., Li, Y., & Wang, J. (2021). Application of RFLP-PCR in genetic diversity studies of aquaculture species. *Journal of Fish Biology*, 98(4), 1023-1035.

Zheng, X., Zhang, Y., Wang, Y., & Wang, G. (2022). CRISPR/Cas9-mediated gene editing in aquaculture: Progress and prospects. *Reviews in Aquaculture, 15*(1), 187–202. https://doi.org/10.1111/raq.12699