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Genetic Analysis of Tilapia Strainsin Different Continents: Tracing Population Structureand Domestication History

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Abstract Tilapia has excellent adaptability, rapid growth and high nutritional value, and has become an important freshwater breeding species. However, intensive breeding and artificial breeding have led to the gradual loss of genetic diversity of tilapia species. This study takes tilapia strains from different continents as the research object, and displays the genetic structure and domestication history of tilapia populations through genetic analysis. Research background shows that African native populations retain high genetic diversity, while in introduced areas such as Asia and the Americas, there are obvious genetic differentiation and characteristic differences due to artificial breeding and environmental pressure. The research hopes to clarify the global diffusion path and domestication process of tilapia, find out the genetic variants and genetic markers related to economic traits during domestication, provide a scientific basis for the sustainable management and protection of tilapia genetic resources, and explore the potential of future intercontinental cooperative research, thereby promoting the long-term development of tilapia breeding technology.

Keywords Tilapia; Genetic diversity; Population structure; Domestication history; Genetic drift

1 Introduction

Tilapia is highly adaptable, grows rapidly and is rich in nutrition. It is the main target of the United Nations Food Organization’s promotion of freshwater aquaculture to the world and is also a preferred variety of freshwater aquaculture that China has prioritized development. Lyapia is native to freshwater waters in Africa and then gradually expands to Asia, America and other regions, and forms multiple localized varieties worldwide. Its wide distribution is due to its excellent biological characteristics and is closely related to long-term domestication and breeding choices in humans.

In the aquaculture industry, the genetic resources of tilapia directly affect farming efficiency and variety improvement. However, with the advancement of intensive farming and the intensification of environmental pressure, the genetic diversity of tilapia is at risk of loss. Intensive artificial breeding may lead to a genetic bottleneck effect, while the introduction of foreign genes may change the genetic structure of local lines. Therefore, evaluating the genetic diversity of tilapia, tracing the domestication process, and digging out key genes that affect economic traits have become the core topics of current research.

This study will sort out the genetic structure and domestication history of tilapia strains in different continents, and analyze the process of tilapia spreading around the world. Combined with genetics, we hope to reveal the genetic changes of tilapia and provide a scientific basis for the management and protection of genetic resources.

2 Global Population Structure of Tilapia

2.1 Genetic diversity of African native populations

Africa is the origin of tilapia, where tilapia populations show rich genetic diversity. There is a clear geographical isolation effect in African tilapia, and the genetic structure of tilapia populations in East, North and West Africa are different. There are studies that report the population genetic structure and genetic diversity of red-bellied tilapia in three different Egyptian aquatic environments: salty (Lake Idku), ocean (Al-Max Bay) and freshwater (Lake Nasser). Habitat differences, environmental factors and harvest pressure are the main characteristics of the sampling site. Three mitochondrial DNA markers were used to evaluate population structural differences between these three populations (Soliman et al., 2017), and the results showed that they had differences in population structure and genetic diversity. The research team of Jiang Bingjie selected Nile tilapia (*Oreochromis niloticus*), Oreochromis (*O. aureus*), Mozambique tilapia (*O. mossambicus*), and GIFT as the research subjects. The neutral test Tajima's D value showed that the population size expanded after experiencing bottleneck effects and/or purification selection, and *Oreochromis* had the lowest genetic diversity (Jiang et al., 2019).

2.2 Genetic characteristics of tilapia strains in Asia

Tilapia has experienced extensive introduction and breeding in Asia. The tilapia breeding industry in Asia is huge in scale, diverse in varieties, and its genetic characteristics are affected by various factors such as the source of introduction, breeding methods and breeding strategies. Research experts surveyed important breeding strains from the Philippines and Africa, and used SNP array data and Poolseq SNP to characterize the population structure of these samples (Barría et al., 2023). They observed the largest gap between Asian and African populations, and the mixing degree of Asian populations was higher than that of African populations, and determined that the SNP array data could successfully parse the relationship between different Nile tilapia populations. Poolseq data identified genomic regions with high differentiation levels between GIFTw and other populations. Gene ontology terms associated with mesodermal development are significantly abundant in genes located in these regions. In pairwise comparisons between GIFTw and all other populations, there was a genetic difference in one region on chromosome Oni06.

2.3 Genetic structural characteristics of tilapia population on the American continent

The tilapia population on the American continent mainly originates from African original species. After long-term trans-ocean introduction, settlement and breeding processes, it has formed unique genetic structural characteristics. Studies have shown that multiple introduction events, interspecies hybridization, and environmental adaptation pressures have jointly shaped the genetic diversity of these populations. In terms of genetic differentiation, tilapia populations in major distribution areas such as Brazil, Mexico and the United States show significant geogenetic structures. This differentiation is not only reflected in mitochondrial DNA sequence variation, but also in the allelic frequency distribution of microsatellite sites (Nyingi et al., 2009). It is worth noting that artificial selection stress and environmental adaptation jointly drive the genetic variation of these populations, where farmed populations show obvious selection signals, while wild populations retain more original genetic characteristics (Barluenga et al., 2006). This mechanism of genetic structure formation provides an important case for understanding species adaptive evolution.

3 Genetic Clues of the History of Tilapia Domestication

3.1 Genetic evidence of origin of tilapia domestication

In recent years, through genetic analysis of wild and farmed tilapia populations in various regions of Africa, people have gradually unveiled the mystery of the origin of tilapia domestication. Studies based on mitochondrial DNA and nuclear genome analysis show that the Nile River Basin and the East African lake area are likely to be the main sites for the initial domestication of tilapia (Nayfa et al., 2020). Wu Feng from Sun Yat-sen University studied the genetic and molecular markers of tilapia. Using random amplified polymorphic DNA and mitochondrial gene sequence analysis technology, the genetic variation of nuclear genes and extranuclear genes was detected respectively. In the UPGMA and NJ phylogeny trees constructed by mitochondrial genes, red tilapia and Honalong tilapia first gathered together, then formed sister branches with the *Mozambique matriline*, and then clustered with the *Nile tilapia matriline*; *Oriya tilapia* is located at the bottom of the phylogeny tree.

3.2 Effects of early domestication events on population genetic structure

The bottleneck effect during early domestication significantly reduced the genetic diversity of some populations. After genetic analysis of *Oreochromis niloticus*, it was found that the allelic richness and heterozygosity of breeding populations were significantly lower than that of wild populations, which was related to the fact that a few individuals were selected as breeding populations during domestication. Anthropogenic selection stress leads to rapid changes in gene frequencies associated with economic traits, and insulin-like growth factor (IGF) genes associated with growth rate show significant selection signals in breeding populations (Cádiz et al., 2020). The breeding populations in different regions show obvious regional differences in genetic structure. Brazil's tilapia farming populations exhibit unique haplotype distributions on disease resistance-related genes, which may be due to local disease stress-driven selection results. Mexican breeding populations exhibit higher allelic frequencies on low temperature-tolerant-associated genes (heat shock protein HSP70), which is in line with its high-altitude breeding environment. These cases show that genetic selection during domestication directly shapes the functional genetic structure of the population.

3.3 Genetic differentiation characteristics of domesticated strains in different continents

As tilapia gradually expanded to all parts of the world, domesticated strains from different continents showed unique genetic characteristics. After long-term artificial breeding, tilapia in Asia has formed multiple strains that adapt to different breeding modes. For example, GIFT (Genetic Improvement of Farmed Tilapia) shows faster growth rate and higher survival rate. Countries in the Americas have set different breeding goals based on their own breeding environment and market needs. In Brazil, breeding efforts focus on the improvement of growth rate and feed conversion rate. Through whole-genome resequencing, the researchers found that Wnt signaling pathway, gonadotropin-releasing hormone receptor and integrin signaling pathways are positively selected in improved lines, indicating that these pathways are closely related to growth and reproductive performance. In order to improve the comprehensive performance of tilapia, some countries in the Americas have carried out hybridization tests between different strains. Comparison of salt tolerance performance of the positive and negative progeny of the orange *Mozambique tilapia* and the *Honalong tilapia*. The difference in salt tolerance of the positive and negative progeny of the orange Mozambique tilapia and the *Honalong tilapia* was compared through the acute salt tolerance experiment. The results showed that the 96 h semi-lethal salinity (MLS-96) of the orthogonal offspring (Mohalo) and the anti-progeny (Mohalo) were 29.80±3.03‰ and 29.38±4.48‰, respectively. There was no significant difference between the two. The two hybrid offspring had obvious hybrid advantages and their salt tolerance was higher than that of the two parents.

4 Effects of Domestication on the Tilapia Genome

4.1 Changes in functional genes of tilapia under artificial selection

Artificial selection drives significant changes in tilapia functional genes. For example, growth rate-related genes show significant frequency increases in long-term breeding. Taking Nile tilapia as an example, through genome-wide association analysis (GWAS), researchers found that the expression levels of the growth hormone-related gene GH and the insulin-like growth factor IGF-1 are significantly improved (Khaw et al., 2008). The increase in these gene expression directly promotes the rapid growth of fish and greatly improves the breeding benefits. Disease resistance traits have also been widely concerned. By selecting streptococcal disease resistance genes, strains with strong disease resistance are selected. Actual cases show that immune-related genes such as TLR, MHC, and other genomic regions of certain lines are significantly enriched, indicating that these genes are significantly enhanced under the pressure of artificial selection. Changes in these functional gene frequency reflect the directional effects humans exerted on the tilapia genome during domestication (Figure 1) (Mwanja et al., 2010).

4.2 Analysis of genome structure variation during domestication

During the domestication of tilapia, the genome structure showed obvious mutations. Changes at the chromosome level are the most representative, such as chromosome inversion, translocation and repeat sequence amplification. Taking the genome sequencing study of Asian farmed tilapia as an example, through high-throughput sequencing and comparative genomic analysis, it was found that artificial domesticated lines had a large number of structural mutations in specific chromosomal segments. These variant regions often contain important functional genes, which directly affect the economic traits of tilapia. Further research shows that structural variation plays a key role in improving the adaptability of tilapia to high-density farming environments. Inverted variation in specific chromosomal regions makes it easier for certain strains to adapt to hypoxic environments and improves overall aquaculture efficiency (Nayfa et al., 2020). The universality and influence of this structural variation reveals the important role of genomic structural variation in the domestication of tilapia.

Figure 1 Representatives of Nile tilapia populations from EA illustrating digitized landmarks used in geometric morphometrics (Adopted from Mwanja et al., 2010)

Image caption: A = Albert, E = Edward, G = George, KC=Kazinga Channel, RN = River Nile, T = Turkana, Ky = Kyoga, VK=Victoria Kakyanga, M = Mulehe, Ka = Kayumbu, Rf = Rwitabingi farm, Sf = Sindi farm and Bf = Bagena farm (Adopted from Mwanja et al., 2010)

4.3 Traces of domestication of genes related to important economic traits

Economic trait genes leave clear traces during the domestication of tilapia. These genes are often closely related to growth rate, meat quality, reproductive performance, and environmental tolerance. Some tilapia species are good candidates for saltwater and seawater farming because they can grow at high salinity. In the article "Status of conventional and molecular breeding of salinity-tolerant tilapia", *Mozambique tilapia*, golden tilapia, maolipapia, uropia, galipapia and zili tilapia are the most salt-resistant (Yue et al., 2023). Hybrids from salt-tolerant tilapia species can tolerate a certain degree of salinity. They have been used in aquaculture production in saltwater and all seawater. Conventional breeding has been applied to improve the growth rate of salt-resistant tilapia. However, they grow at a lower rate than freshwater *Oreochromis niloticus*. Recently, many genomic resources and tools have been developed for salt-resistant tilapia. Quantitative trait loci (QTL) mapping and genome-wide association studies (GWAS) for important economic traits have been applied in molecular breeding of excellent salt-tolerant tilapia lines.

5 Cross-Regional Introduction and Genetic Drift of Tilapia

5.1 Genetic drift phenomenon in the process of transcontinental introduction of tilapia

Since its introduction to Africa and its growth, tilapia has experienced many complex transmission paths. This long-distance migration and breeding process is inevitably accompanied by genetic drift phenomenon. Taking the introduction of tilapia from Africa to Asia as an example, the initial introduction was limited, and many of the originally abundant genotypes gradually decreased or even disappeared when these fish were breeding in new environments. At the same time, some of the rarer genes may increase due to accidental events. In the 1950s, Taiwan introduced tilapia from Africa. After decades of breeding, the rare genotypes in some primitive African populations have become mainstream among Asian breeding populations (Yustiati et al., 2020). This random change in genotype frequency causes significant differences between the introduced population and the origin. This phenomenon of genetic drift has also been widely observed not only in Asia, but also in tilapia farming groups in the Americas.

5.2 Analysis of gene exchange between foreign populations and local populations

The phenomenon of genetic exchange between foreign populations and native populations is equally common in tilapia farming. This communication is usually manifested as introducing populations to escape into the wild environment and then crossing with local wild populations. Florida, USA is a typical case. After a large number of breeding foreign tilapia escaped, they had extensive genetic communication with the local original fish population. The consequence of this communication is that the genetic structure of the native population has been significantly changed. Specific studies have shown that this hybridization will lead to reduced adaptability of native populations and even the disappearance of original unique genes (Angienda et al., 2011). Some studies also believe that this genetic communication may enhance the competitiveness of hybrid offspring in new environments to a certain extent. Therefore, how to reasonably control and manage these gene exchanges has become the common concern for aquaculture and ecological protection.

5.3 The challenge of genetic drift on the protection of tilapia germplasm resources

Tilapia is native to Africa and the Middle East. Due to its fast growth rate, strong reproductive ability, low requirements for bait, strong adaptability and disease resistance, tilapia has been introduced to most parts of the world in recent decades, and its breeding output accounts for 11% of the global freshwater aquaculture production. Everything is a double-edged sword. A series of suitable characteristics such as fast growth and reproduction and strong environmental adaptability have also promoted its diffusion and invasion in natural waters, making it a “typical invasive species” (Figure 2). Currently, most tropical and subtropical areas around the world are threatened by tilapia invasion. In terms of resource volume, the tilapia resources account for 11.81% of the total catch in the main rivers in Guangdong, China. In some river sections, the tilapia resources even exceed the sum of other fish. It gradually reduces the genetic diversity of breeding populations, and even causes the disappearance of specific excellent genotypes; It also allows genes of foreign populations to be integrated into local wild populations, threatening the purity and characteristics of local populations (Tibihika et al., 2018). The spread and invasion of tilapia not only affects the stability of fishery resources and fishermen's income, but also poses a serious threat to biodiversity and the structure and function of aquatic biological systems (Mwanja et al., 2010).

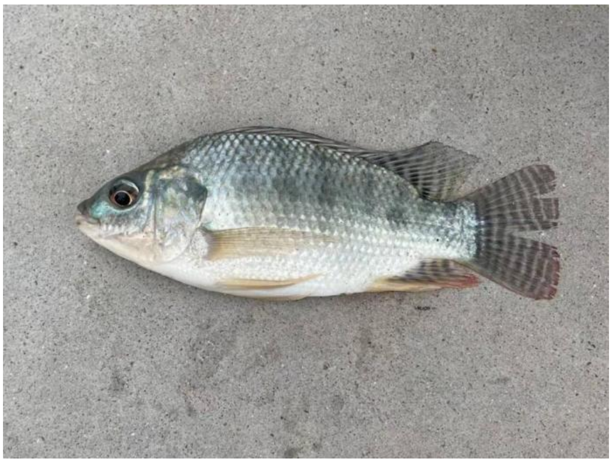

Figure 2 Tilapia in natural waters

6 Genetic Improvement Strategies for Tilapia Strains in Different Continents

6.1 Protection and utilization of native African strains

Africa, as the origin of tilapia, has abundant wild germplasm resources. In order to protect the genetic diversity of tilapia, African countries have taken various measures. Establishing a germplasm resource library is one of them, ensuring that their genetic resources are not lost by collecting and preserving wild tilapia populations in different regions. African scientists actively study the characteristics of native tilapia, especially its advantages in disease resistance and environmental adaptability. Through breeding technologies such as breeding and hybridization, these excellent characteristics are introduced into breeding varieties to improve breeding benefits. At the same time, promote ecological breeding models, use local tilapia to adapt to the local environment, reduce the impact on the environment, and promote sustainable development.

6.2 Genetic strategies and effectiveness of tilapia breeding in Asia

Asia is one of the main regions for tilapia farming, especially in countries such as China, Vietnam and the Philippines. To meet market demand, these countries have implemented several genetic improvement programs. The most well-known is the GIFT project launched in the 1990s, aiming to improve the growth rate and disease resistance of tilapia through selective breeding. The researchers collected basic groups from eight countries in Asia and Africa, and after multiple generations of breeding, they successfully cultivated strains with an increase in growth rate of 85%. These improved varieties have been promoted in 16 countries including Asia, Africa and Latin America, which has significantly improved the local breeding benefits. In addition, Asia is also actively adopting genomics technology to accelerate the genetic improvement process of tilapia. Genome-wide association analysis (GWAS) and genome selection reveal to scientists genetic variations related to economic traits, promoting genetic gain in breeding programs.

6.3 New trends in genetic improvement of tilapia population in America

In North and South America, the genetic improvement of tilapia populations has flourished. Recently, Brazilian tilapia producer Brazilian Fish announced that it plans to accelerate the genetic improvement of tilapia through genome editing technology, and to improve the breeding efficiency of tilapia, improve the production performance and fish meat output of tilapia, and enhance the disease resistance of farmed fish. Some cities in North America have jointly developed the tilapia gene editing system FLT 01, which is not subject to genetically modified regulations, according to the provisions of the National Advisory Committee on Agricultural Biotechnology (CONABIA). This tilapia production line achieves more sustainable production by improving fillet yield, growth and feed conversion efficiency, allowing tilapia to reach market weight in a shorter time while also consuming less feed than traditional varieties (Yang et al., 2022).

7 Future Research Prospects

7.1 New technologies and methods for tilapia genetic resources management

With the development of biotechnology, tilapia genetic resource management is ushering in new technologies and methods. Genome editing technologies, such as the emergence of CRISPR/Cas9, provide precise tools for directional improvement of the economic traits of tilapia. This technology can quickly and efficiently edit target genes, making it more accurate and effective to improve the characteristics of disease resistance, growth rate and environmental adaptability. The rise of genome-wide selection technology has also brought new possibilities for tilapia breeding. By analyzing genome-wide data, breeders can more accurately evaluate and predict strain performance, thereby accelerating the variety improvement process. In addition, the application of emerging means such as blockchain technology and big data analysis will also greatly improve the management efficiency of tilapia genetic resource information and the ability to share data.

7.2 Potential and suggestions for cross-continental population cooperative research

The tilapia populations in different regions around the world have their own characteristics, and intercontinental cooperative research is of great significance to the sustainable development of the global tilapia industry. Strengthening international cooperation can promote the sharing and exchange of excellent genetic resources and help solve the bottlenecks of genetic resources in specific regions. Through intercontinental research, researchers can better understand the adaptation mechanisms of different populations under different environmental conditions. This collaboration can also promote international exchange of technologies and methods, such as complementary advantages between the rich genetic resources of Africa’s origin and advanced breeding technologies in Asia and the Americas. It is recommended that a more effective international cooperation network should be established in the future, unified genetic resource evaluation standards should be formulated, and talent and technology exchanges should be strengthened to provide a solid scientific foundation for global tilapia breeding.

7.3 Long-term development goals of genetic breeding of tilapia

The long-term development goals of genetic breeding of tilapia should focus on three main aspects: continuous optimization of varieties, improvement of environmental adaptability and improvement of breeding benefits: it is necessary to ensure the protection of genetic diversity and prevent excessive selection from leading to the singleization of the gene bank; the long-term goals should also focus on the improvement of the comprehensive traits of varieties, such as simultaneously improving growth rate, disease resistance, nutritional value and tolerance to extreme environments; while achieving genetic breeding goals, we should also pay attention to the impact of breeding on the ecological environment to avoid ecological risks caused by blind pursuit of economic interests. The future genetic breeding strategy should be more comprehensive and sustainable, and the healthy development of the tilapia industry should be achieved through technological innovation and reasonable planning.

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Conflict of Interest Disclosure

The author confirms that the study was conducted without any commercial or financial relationships and could be interpreted as a potential conflict of interest.

References

Angienda P.O., Lee H.J., Elmer K.R., Abila R., Waindi E.N., and Meyer A., 2011, Genetic structure and gene flow in an endangered native tilapia fish (*Oreochromis esculentus*) compared to invasive Nile tilapia (*Oreochromis niloticus*) in Yala swamp, East Africa, Conservation Genetics, 12: 243-255.
https://doi.org/10.1007/s10592-010-0136-2

Barluenga M., Stölting K.N., Salzburger W., Muschick M., and Meyer A., 2006, Sympatric speciation in Nicaraguan crater lake cichlid fish, Nature, 439(7077): 719-723.
https://doi.org/10.1038/nature04325

Barría A., Peñaloza C., Papadopoulou A., Mahmuddin M., Doeschl-Wilson A., Benzie J.A.H., Houston R.D., and Wiener P., 2023, Genetic differentiation following recent domestication events: a study of farmed Nile tilapia (*Oreochromis niloticus*) populations, Evolutionary Applications, 16(6): 1220-1235.
https://doi.org/10.1111/eva.13560

Cádiz M.I., Lopez M.E., Diaz-Dominguez D., Cáceres G., Yoshida G.M., Gomez-Uchida D., and Yáñez J.M., 2020, Whole genome re-sequencing reveals recent signatures of selection in three strains of farmed Nile tilapia (*Oreochromis niloticus*), Scientific Reports, 10(1): 11514.
https://doi.org/10.1038/s41598-020-68064-5

Yue G.H., Ma K.Y., and Xia J.H., 2023, Status of conventional and molecular breeding of salinity-tolerant tilapia, Aquacture, 16(1): 271-286.
https://doi.org/10.1111/raq.12838

Jiang B.J., Fu J.J., Zhu W.B., Wang L.M., Fang M., and Dong Z.J., 2019, Genetic variation analysis of seven tilapia populations based on *COI* gene, Journal of Shanghai Ocean University, 28(6): 827-834.

Khaw H.L., Ponzoni R.W., and Danting M.J.C., 2008, Estimation of genetic change in the *GIFT* strain of Nile tilapia (*Oreochromis niloticus*) by comparing contemporary progeny produced by males born in 1991 or in 2003, Aquaculture, 275(1-4): 64-69.
https://doi.org/10.1016/j.aquaculture.2008.01.022

Mwanja W.W., Kaufman L., and Fuerst P.A., 2010, Comparison of the genetic and ecological diversity of the native to the introduced tilapiines (Pisces: Cichlidae), and their population structures in the Lake Victoria region, East Africa, Aquatic Ecosystem Health & Management, 13(4): 442-450.
https://doi.org/10.1080/14634988.2010.527268

Nayfa M.G., Jones D.B., Benzie J.A.H., Jerry D.R., and Zenger K.R., 2020, Comparing genomic signatures of selection between the Abbassa strain and eight wild populations of Nile tilapia (*Oreochromis niloticus*) in Egypt, Frontiers in Genetics, 11: 567969.
https://doi.org/10.3389/fgene.2020.567969

Nyingi D.W., Vos L.D., Aman R., and Agnèse J.F., 2009, Genetic characterization of an unknown and endangered native population of the Nile tilapia *Oreochromis niloticus* (Linnaeus, 1758) (Cichlidae; Teleostei) in the Loboi Swamp (Kenya), Aquaculture, 297(1-4): 57-63.
https://doi.org/10.1016/j.aquaculture.2009.09.017

Soliman T., Aly W., Fahim R.M., Berumen M.L., Jenke-Kodama H., and Bernardi G., 2017, Comparative population genetic structure of redbelly tilapia (Coptodon zillii (Gervais, 1848)) from three different aquatic habitats in Egypt, Ecology and Evolution, 7(24): 11092-11099.
https://doi.org/10.1002/ece3.3586

Tibihika P.D., Waidbacher H., Masembe C., Curto M., Sabatino S., Alemayehu E., Meulenbroek P., Akoll P., and Meimberg H., 2018, Anthropogenic impacts on the contextual morphological diversification and adaptation of Nile tilapia (*Oreochromis niloticus* L., 1758) in East Africa, Environmental Biology of Fishes, 101: 363-381.
https://doi.org/10.1007/s10641-017-0704-0

Yang Z.T., Yu Y.P., Tay Y.X., and Yue G.H., 2022, Genome editing and its applications in genetic improvement in aquaculture, Reviews in Aquaculture, 14(1): 178-191.
https://doi.org/10.1111/raq.12591

Yustiati A., Hutagalung R.M., Andriani Y., and Lili W., 2020, Genetic analysis of tilapia (*Oreochromis niloticus*) nirwana strain cultured in aceh besar and wanayasa by using random amplified polymorphic dna method, Asian Journal of Biochemistry, Genetics and Molecular Biology, 4(4): 1-9.
https://doi.org/10.9734/ajbgmb/2020/v4i430111