

# The role of phenylalanine in the fish pigmentation process: a review

Marcos Paiva Scardua  · Shelby M. Walker · Antonio Cesar Godoy · Claucia A. Honorato · Daclely H. Neu

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**Abstract** In the global pet market, fish are the number one selling pets in the world. Considering a market that moves billions of dollars annually and where the body color of these animals is one of the main factors that attracts consumers to this niche, it is important to understand the pigmentation process and what can influence the metabolism of these animals, which directly affects their color. This understanding will allow for the appropriate manipulation of diets without compromising animal welfare, ultimately resulting in more attractive specimens for the market. Biological pigments, namely melanin, carotenoids, pteridine, and purines, are not only responsible for the color of fish, but also influence health, disease resistance, well-being, social interaction, growth, and reproduction. Providing these pigments in the diet is critical because some are not biosynthesized by fish. For example, pheomelanin and carotenoids are not synthesized by fish, whereas compounds such as pteridine, eumelanin, and purines are synthesized. During the synthesis phase, certain amino acids, especially phenylalanine, play a key role in the metabolic pathways. Thus, this work describes the role of phenylalanine in the metabolic pathways of these pigments, characterizes the types of pigments, highlights their importance for animals, and explains the mechanisms of action and control involved in the pigmentation process.

**Keywords** Feed supplementation · Pigments · Metabolites · Nutrition · Technological additives · Ornamental fish

## Introduction

Aquaculture aims to produce biomass to feed an ever-growing population (FAO 2021). However, ornamental aquaculture takes a different approach, focusing on individual traits that determine selling prices, with color being an important factor in this market (Luo et al. 2021; McLean 2021; Vissio et al. 2021; Lau et al. 2023).

Fish color is the result of a complex relationship between several biological pigments (melanin, pteridine, carotenoids, and purines) that are either ingested and metabolically converted or biosynthesized. The control mechanisms for fish skin pigmentation integrates internal factors such as: genetics, life cycle, pigment deposition ability, and body size, as well as external factors such as: feed, environmental conditions, and pigment sources (Sathyaruban et al. 2021; Vissio et al. 2021).

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Marcos Paiva Scardua (✉) · Antonio Cesar Godoy · Claucia A. Honorato · Daclely H. Neu  
Faculdade de Ciências Agrárias, Universidade Federal da Grande Dourados, Rodovia Dourados - Itahum, km 12, Dourados, 79804-970, Mato Grosso do Sul, Brazil  
e-mail: marcos.scardua@ifce.edu.br

Shelby M. Walker  
School of Fisheries, Aquaculture and Aquatic Science, Auburn University, 203 Swingle Hall, Auburn, 36849, Alabama, USA

Antonio Cesar Godoy  
Departamento de Pesquisa em Recursos Naturais, Instituto Federal de Educação do Paraná, 475 Cívica Avenue, Assis Chateaubriand, 85935-000, Paraná, Brazil

The color patterns and hues of a given fish species exhibit variability due to factors such as age, sex, health, stress, and reproductive stage (Vissio et al. 2021). Consequently, these colorations serve as a central visual mechanism for social communication. However, the maintenance of such coloration comes at a metabolic cost, as some of these pigments are derived from the diet, including those synthesized endogenously. This phenomenon underscores the importance of coloration as a crucial social communication mechanism, as its manifestation provides insight into the biological capabilities of each individual (Gempel and Visconti 2020).

This importance is mainly due to the multi-functionality of these pigments in fish. Beyond their role in coloration, these pigments exhibit antioxidant properties, contribute to anti-stress and anti-inflammatory responses, provide cellular photoprotection, enhance reproductive performance, influence egg and larval quality, act as provitamin A, play a role in the immune system, influence growth, and modulate lipid metabolism. Collectively, these diverse functions contribute to the overall well-being of the organism (Bell et al. 2019; Nakano and Wiegertjes 2020; Ciji and Akhtar 2021; Svitačová et al. 2023; Wassef et al. 2023). Consequently, the utilization of these pigments in the context of pigmentation becomes a resource-intensive endeavor due to the myriad metabolic pathways involved.

In the metabolic processes of these pigments, *de novo* synthesis of carotenoids is observed exclusively in plants, fungi and bacteria. The synthesis of other pigments, on the other hand, is typically found in different animal species, and certain amino acids play a crucial role in these processes. For example, in melanogenesis (the formation of melanin) and pheomelanin production, the involvement of phenylalanine, tyrosine, cysteine, and glutathione has been documented (Li et al. 2008; Bilandija et al. 2013). In the pteridine pathway, glutamine/glutamate, tryptophan, phenylalanine, and tyrosine are major contributors (Ziegler 2003; Leclercq et al. 2009). However, it is important to note that the synthesis of carotenoids, specifically via the mevalonate pathway, is not addressed in this work.

Although *de novo* synthesis of carotenoids does not occur in fish, there is compelling evidence indicating metabolic pathways through which fish can transform assimilated carotenoids from the diet. This phenomenon was observed in goldfish, as described by Hata and Hata (1972), where astaxanthin was formed from zeaxanthin through 4-keto-zeaxanthin. Similar findings were reported in another cyprinid, *Pseudogobio esocinus*, as documented by Tsushima and Matsuno (1999). Further insights into carotenoid metabolism in fish were provided by Bijun et al. (2024), where the study reported on genes intricately involved in carotenoid metabolism in fish, focusing on key aspects such as the uptake and transport of carotenoids into tissues (SCARB and TTC39B genes), carotenoid degradation (BCO2 gene), conversion of yellow dietary carotenoids into red keto carotenoids (CYP2J19 gene), and the coding for proteins that bind to carotenoids (CRCN and GSTA2 genes). Additionally, the study presented evidence supporting a cooperative relationship between lipid metabolism and carotenoid transport and deposition, given the hydrophobic nature of carotenoids.

Regardless of whether the pigment is synthesized in the fish, aquaculture production systems typically have limited availability and access to natural diets compared to organisms developing in their natural habitat. Natural diets, which are rich in biological pigments (Sathyaruban et al. 2021), are a valuable source of these pigments. Therefore, access to pigments in aquaculture depends on supplementation of diets or live feed by producers. Of the four pigments (carotenoids, purines, melanin and pteridine) associated with fish pigmentation, three are endogenously synthesized by fish. Among these three, phenylalanine, an essential amino acid and a precursor of tyrosine, is involved in the metabolic pathways of two pigments (melanin and pteridine) (Ziegler 2003; Ito and Wakamatsu 2008; Leclercq et al. 2009; Gempel and Visconti 2020). This observation has sparked interest in exploring its potential inclusion in commercial diets for ornamental fish to assess its efficacy in enhancing the animal pigmentation process.

This review will elucidate the relationship between certain amino acids, particularly phenylalanine and tyrosine, and the metabolic pathways governing specific biological pigments responsible for fish coloration. The discussion will include the different types of biological pigments, the cells involved in pigment deposition in fish, the regulatory processes that influence pigmentation control, and the metabolic pathways that orchestrate the synthesis of biological pigments responsible for the observed coloration. Attention will also be given to the metabolic pathways associated with pigments in which phenylalanine plays a critical role.



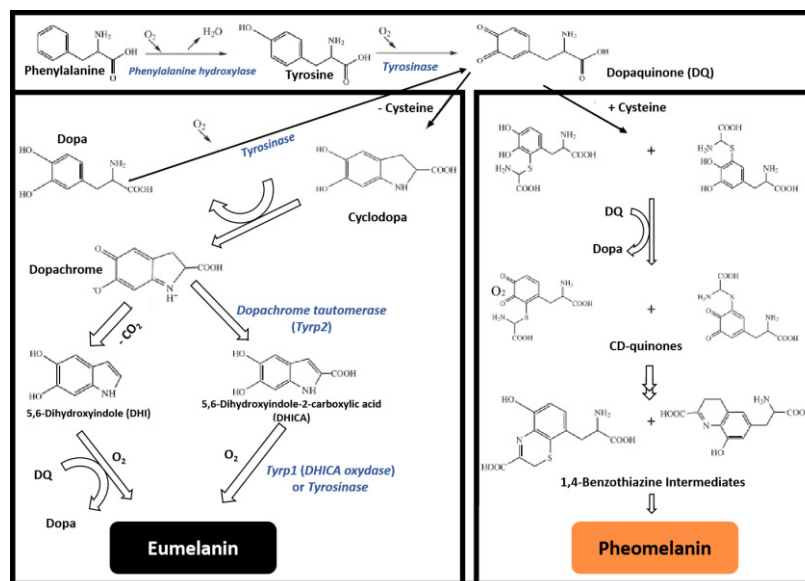
## Phenylalanine, pigments and regulation mechanisms

Phenylalanine, an aromatic amino acid, falls into the category of glucoketogenic amino acids, meaning it has the ability to produce both ketone bodies and glucose (Nelson et al. 2021). In addition, according to Li et al. (2008), it has the status of an essential amino acid for fish, requiring dietary intake. Phenylalanine plays a central role in several metabolic pathways, contributing to the synthesis of essential compounds, including cholesterol, steroids, carotenoids, and intermediates in the citric acid cycle (fumarate and acetoacetyl-CoA). It also contributes to the production of ketone bodies and facilitates the synthesis of tyrosine through the cofactor tetrahydrobiopterin and the enzyme phenylalanine hydroxylase. The cooperation between phenylalanine and tyrosine extends to their involvement in the formation of several commercially important natural products, such as lignin, tannin, cinnamate, and morphine. In addition, this tandem plays a crucial role in the synthesis of essential neurotransmitters, including dopamine, epinephrine, and norepinephrine (Nelson et al. 2021).

The importance of phenylalanine in fish diets extends beyond its role in pigmentation to include vital physiological processes that actively contribute to fish growth (Khan and Abidi 2007; Rodrigues et al. 2019; Sayed and Ahmed 2023). Its importance is closely linked to various aspects of production performance, protein digestibility, immunity and overall survival. This is particularly evident because phenylalanine metabolism is closely intertwined with that of tyrosine, serving as a precursor for important hormones and neurotransmitters, including thyroxine, tri-iodo-thyronine, epinephrine, dopamine, and melanin, among others. Phenylalanine is absorbed by intestinal cells and subsequently converted to tyrosine primarily in the liver and kidney (Li et al. 2008). While phenylalanine can be converted to tyrosine, a balanced diet rich in both amino acids is critical for optimal fish development. This balance is essential due to their key roles in physiological functions, overall metabolism, and protein synthesis (Ahmed 2022). Sayed and Ahmed (2023) indicate that the best dietary inclusion ratio of phenylalanine and tyrosine (60:40) provides the best growth for catfish fry (*Heteropneustes fossilis*).

In the synthesis of biological pigments in fish, phenylalanine plays a critical role in the formation of melanin. This process involves the conversion of tyrosine to L-DOPA and its subsequent conversion to L-dopaquinone, which ultimately leads to the production of one of the melanin types (Fig. 1). The synthesis of pteridine in fish is closely linked to melanogenesis, as highlighted by a metabolic pathway (Leclercq et al. 2009). This pathway involves the synthesis of tetrahydrobiopterin (4Hbiopterin), a cofactor essential for the conversion of phenylalanine to tyrosine. In addition, tetrahydrobiopterin is responsible for the synthesis of several forms of pteridines (Fig. 2).

In nature, several groups of biological pigments contribute to the vibrant colors observed in fish: mela-

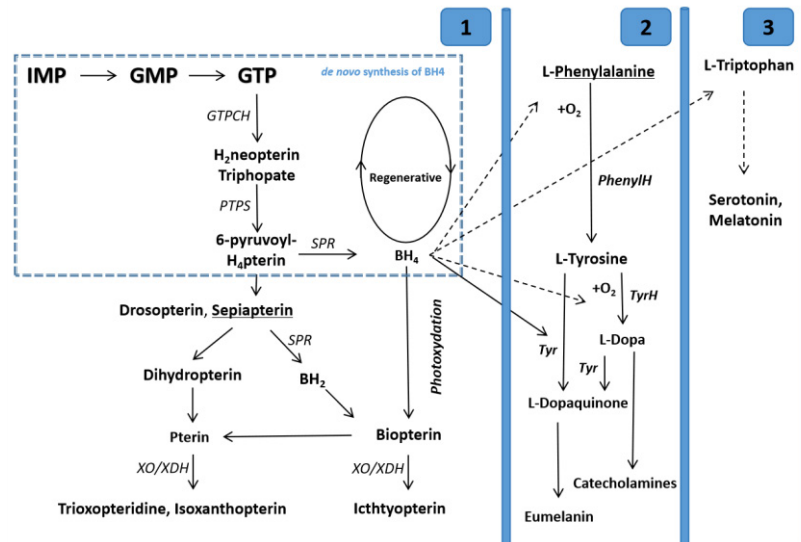


**Fig. 1** Melanogenesis, production of eumelanin and pheomelanin. Source: Adapted from Ito and Wakamatsu (2008). \*Typr1= Tyrosinase 1 and Typr2 = Tyrosinase 2

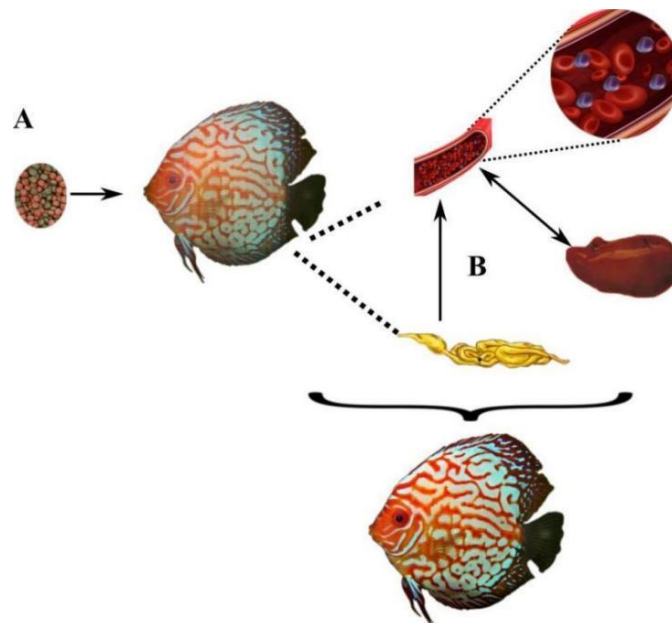


nin, pteridine, carotenoids, and purines. While fish can synthesize melanin, pteridine, and purines, *de novo* synthesis of carotenoids does not occur (Johnson and Fuller 2014; Gempel and Visconti 2020; Luo et al. 2021; Yang et al. 2021; Vissio et al. 2021). Instead, some fish species exhibit the conversion of certain carotenoids into others, although these are initially ingested through their diet (Sathyaruban et al. 2021). As a result, carotenoids are naturally incorporated into fish through dietary intake in the wild or in controlled environments such as aquariums or hatcheries where they are provided in the diet.

According to Sefc et al. (2014), pigmentation in fish is a metabolically demanding and costly process that relies heavily on both biosynthesis and dietary absorption of pigments. This process serves as a reflection of an individual's ability to adapt to environmental conditions, thereby signaling the health status of each animal (Gempel and Visconti 2020). Once pigments are ingested through the fish's diet (Fig. 3), they are emulsified by digestive enzymes and solubilized into micelles. These micelles are then taken up by



**Fig. 2** Synthesis of pteridine and relationships with melanogenesis and melatonin production. Adapted from Leclercq et al. (2009). IMP: Inosine monophosphate; GMP: guanosine monophosphate; GTP: guanosine triphosphate; PTPS: 6-pyruvyl tetrahydropterin synthetase; SPR: Sepiapterin reductase; BH<sub>2</sub>: Dihydrobiopterin; XO: Xanthine oxidase; XDH: Xanthine dehydrogenase; PhenylH: Phenylalanine hydroxylase; TyrH: Tyrosine hydroxylase; Tyr: Tyrosinase; TyrpH: Tryptophan hydroxylase; NAS: N acetylserotonin.



**Fig. 3** Scheme of absorption and destination of pigments in fish. A: Ingestion of feeds containing pigments. B: In the intestine, absorption occurs in enterocytes. Carotenoids are transported by lipoproteins through the bloodstream to lymphatic vessels where they are destined for the liver, muscles, gonads, eyes and skin.



enterocytes in the intestine. Chylomicrons, formed from the absorbed pigments, are then transported from the intestinal mucosa to the bloodstream via lipoproteins and subsequently taken up by lymphatic vessels (Murota 2020). These chylomicrons are distributed to various tissues, including liver, muscle, skin, scales (Granneman et al. 2017; von Lintig et al. 2020; Fang et al. 2022; Yao et al. 2023), eyes (Salis et al. 2019; Svitačová et al. 2023). During the reproductive period, carotenoids may also be targeted to the gonads, ovaries and testes (Mookkan et al. 2022).

Understanding the absorption and fate of pigments in animals is critical to understanding their presence, forms, behavior, and the colors they impart. Melanin, pteridine, carotenoids, and purines are the primary pigments found in animals. Melanin manifests in two forms: eumelanin, responsible for black and brown tones, and pheomelanin, responsible for reddish and yellowish tones (although the latter is not typically found in fish) (Irion and Nusslein Volhard 2019). Nevertheless, Zhu et al. (2016) suggest the possible occurrence of pheomelanin synthesis in red tilapia, which warrants further investigation. Extending the findings of Colanesi et al. (2012); Gempel and Visconti (2020); Sathyaruban et al. (2021); Vissio et al. (2021), it is noted that pteridines play a role in imparting reddish and yellowish colors, while carotenoids contribute to a diverse spectrum of colors. Also confirmed by Leclercq et al. (2009); Gempel and Visconti (2020), purines, when present in the form of transparent crystals, possess the ability to reflect incident light, thereby yielding a spectrum of colors including blue, silver, gold, or even white tones.

In fish, biological pigments are housed in specialized cells known as chromatophores, which are located primarily in the dermis and epidermis. These specialized cells are generated during embryonic development by the differentiation of neural crest cells (Parichy 2006; Kottler et al. 2013; Lau et al. 2023). The cytoplasm of chromatophores contains organelles called chromatosomes, which are responsible for the accumulation of these pigments and serve as the site of their biosynthesis (Gempel and Visconti 2020). Notably, certain cells, such as erythrophores and xanthophores, have the capacity to store multiple types of pigments, as exemplified by their ability to contain both pteridine and carotenoid pigments (Ligon and McCartney 2016; Luo et al. 2021).

In contrast to birds and mammals, which have only one type of chromatophore, fish have six different types, according to Sathyaruban et al. (2021); Wu et al. (2022). These include melanophores (black and brown), erythrophores (red), xanthophores (yellow), cyanophores (blue), leucophores (white and silver), and iridophores (iridescent). Melanophores, erythrophores, xanthophores, and cyanophores are characterized by their ability to absorb light, while iridophores and leucophores reflect light. Chromatophores that reflect light contain purines (Ligon and McCartney 2016; Luo et al. 2021) and uric acid (Luo et al. 2021). Notably, leucophores are less abundant and have only been described in a few fish species (Kimura et al. 2014).

As elucidated by Yang et al. (2021), the development of fish coloration involves two main types: pigmentary coloration and structural coloration. Pigmentary coloration results from the absorption of light by intracellular pigments such as melanophores, erythrocytes, and xanthophores. On the other hand, structural coloration is formed by the reflection of light by purine crystals present in the skin or scales of fish, particularly within iridophores. Chromatophores play a key role in the dynamic display of color, responding to stimuli by either aggregating or dispersing the pigment granules within them, thereby altering the externalized color pattern. Dispersed pigments contribute to a darker skin appearance, whereas aggregated pigments create a lighter pattern (Ligon and McCartney 2016; Gempel and Visconti 2020). As classified by Gempel and Visconti (2020), the direct response of chromatophores to ambient light, independent of the nervous or hormonal systems, is considered the primary physiological response to color change. This response involves the aggregation or dispersion of pigments within fish chromatophores. In contrast, a secondary response occurs when the endocrine or nervous system is involved in the regulation of color change.

According to Bertolesi et al. (2019); Gempel and Visconti (2020); Sathyaruban et al. (2021); Vissio et al. (2021), factors that influence or are responsible for changes in fish color can be categorized as external (biotic and abiotic) or internal (genetics, life cycle, cell type, and physiological). External factors, such as the response to incident light, can rapidly induce color changes, potentially acting as a defense mechanism against predators (Iwashita et al. 2006; Gempel and Visconti 2020). Internally, genetic factors, cell type, and the nervous or endocrine system (Iwashita et al. 2006; Bertolesi et al. 2019; Gempel and Visconti 2020; Vissio et al. 2021), including hormones such as pigment concentrating hormone (MCH) and pigment dispersing hormone (MSH) (Bertolesi et al. 2019), play a role in mediating these color changes.





Fish coloration plays a crucial role in camouflage, and its effectiveness is optimized when it is matched to a specific background color. This mechanism has been conserved throughout fish evolution, with the genes responsible for pigment aggregation or dispersion and pigment synthesis remaining intact. Consequently, when fish are exposed to a light background, the pigments within the melanophores aggregate, resulting in a lighter color that enhances camouflage with the environment (PMCH and PMCHL genes). Conversely, when faced with a dark background, pigments disperse within melanophores, resulting in a darker color that reduces contrast with the environment and facilitates better camouflage (POMC gene) (Bertolesi et al. 2019). McLean (2021) also notes variation in fish color response to changes in tank bottom color, and Padhi et al. (2022) found that the black coloured tank improved body pigmentation, survival and growth of *Dawkinsia filamentosa*. Song et al. (2023) suggest that environmental adaptation of the eyes of the fish (*Plectropomus leopardus*) may be responsible for variation in skin color through several pathways, including stress protein synthesis, phenylalanine metabolism, and energy homeostasis.

Sefc et al. (2014) observed different responses depending on body region, chromatophore type, fish species, and even between populations of the same species. This highlights the challenge of elucidating the specific regulatory mechanisms at play in each fish species. Fish coloration is of great importance as a fundamental component of social and visual communication, providing insights into individual health, reproductive potential, dominance (Johnson and Fuller 2014; Sefc et al. 2014; Vissio et al. 2021), and welfare (Svitačová et al. 2024). Understanding the regulatory mechanisms of pigmentation not only contributes to the welfare of captive animals, but also helps to promote and maintain vibrant coloration.

## Melanine

Melanin pigmentation serves a dual purpose in fish, providing protection against predation through the classic light ventral and dark dorsal coloration pattern (Irion and Nusslein Volhard 2019). In addition, melanin acts as a photoprotector, providing defense against harmful UV radiation (Mueller and Neuhauss 2014; Lau et al. 2023). The synthesis of melanin, known as melanogenesis, involves the formation of pheomelanin and eumelanin (Fig. 1). This complex process takes place within melanosomes, specialized organelles embedded in melanophores located in the dermis and epidermis of fish. It is generally believed that only eumelanin is present in fish (Leclercq et al. 2009; Kottler et al. 2013; Irion and Nusslein Volhard 2019).

Phenylalanine and tyrosine play fundamental roles in the melanogenic pathway. Within the cytosol of the melanophore, L-phenylalanine is converted to L-tyrosine by the enzyme phenylalanine hydroxylase, which requires oxygen as a cofactor, specifically tetrahydrobiopteridine. L-tyrosine is then activated by tyrosinase and converted to dihydroxyphenyl-L-alanine (L-DOPA) within the melanosome membrane. From this point, there are two possible pathways: conjugation of dopaquinone with cysteine or glutathione, leading to the formation of pheomelanin through the action of cesteinil-DOPA or glutathione-DOPA. Alternatively, it can be converted to dopachrome, which serves as a substrate for tyrosinase 2 (Tyrp2). Tyrp2 catalyzes the formation of 5,6-dihydroxyindole-2-carboxylic acid, which is subsequently oxidized by tyrosinase 1 (Tyrp1) to eumelanin (Ito and Wakamatsu 2008; Gempel and Visconti, 2020).

Meredith and Sarna (2006) emphasize the lack of precision in determining the macromolecular structure of eumelanin, with only a general acceptance that eumelanin consists of macromolecules of 5,6-dihydroxyindole (DHI) and 5,6-dihydroxyindole-2-carboxylic acid (DHICA). The structure is considered amorphous and eumelanins are insoluble in solvents (Xiao et al. 2020). Despite recent studies, uncertainties remain regarding the exact composition of eumelanin (Cao et al. 2021). However, due to the diverse functions of these melanin compounds, including photoprotection, antioxidant, antimicrobial, radioactive absorption, anticancer properties, heavy metal absorption, thermal regulation, healing, anti-inflammatory effects, and coloration, they are biodegradable and highly valued for potential applications in medicine and the food industry. The synthesis of synthetic melanin compounds is of great interest and importance (Meredith and Sarna 2006; Xiao et al. 2020; Cao et al. 2021; Ghattavi et al. 2022).

Albinism is a genetic disorder associated with the absence of melanin pigmentation in animals. Fontes et al. (2023) found abnormal pigmentation in marine fish in the natural environment and speculate that these mutations may be associated with population decline and isolation of remote populations, but in certain environments, such as caves, where pigmentation provides no advantage and represents an unnecessary energy expenditure, some fish have evolved with partial or total loss of pigmentation, resulting in true



albinos (Bilandija et al. 2013). In their study of albino fish of the species *Astyanax mexicanus*, Bilandija et al. (2013) focused on the mutation of the OCA2 gene, which is responsible for oculocutaneous albinism, specifically type 2 albinism. The researchers observed that the mutation in this gene affects the initial phase of melanin synthesis by blocking the conversion of L-tyrosine to L-DOPA. Interestingly, even with the addition of L-tyrosine, melanin synthesis does not occur; however, the addition of L-DOPA leads to successful synthesis.

The cited study by Bilandija et al. (2013) emphasized that the unused L-tyrosine serves an alternative purpose by connecting to the catecholamine pathway, which is responsible for the production of neurotransmitters such as dopamine, epinephrine, and norepinephrine. This underscores the metabolic pathway taken by L-tyrosine, which produces products that may be more useful to albino fish in their environment than the production of pigments. Although rare, the albino phenotype is highly sought after in the ornamental fish industry, primarily due to the high prices that these individuals can command (Wang et al. 2022). The generation of albino mutants can be achieved by inhibiting the central genes involved in melanogenesis, but there are few studies exploring this aspect, especially given the vast diversity of species available in the ornamental market (Bian et al. 2021; Zhang et al. 2023).

### Purins

Purines are nitrogenous organic compounds that form nucleotides, of which guanine and adenine are the best known. Initially, purines are biosynthesized as nucleotides, starting from inosine monophosphate (IMP) to guanosine monophosphate (GMP) and then to guanosine triphosphate (GTP). However, the free bases adenine, guanine, xanthine, and hypoxanthine are by-products of purine nucleotide catabolism, primarily for the excretion of nitrogenous waste (Leclercq et al. 2009). The major reflective components present in teleost cells (leucophores and iridophores) are primarily compounds generated during purine metabolism, including guanine, adenine, xanthine, and hypoxanthine. Within this metabolic process, xanthine oxidase and xanthine dehydrogenase play a key role in limiting the rate of purine catabolism by catalyzing the oxidation of hypoxanthine to xanthine and xanthine to uric acid (Leclercq et al. 2009).

Similar to other pigments localized in specific chromatophores, purines accumulate in iridophores and leucophores, which are located in the skin and scales of teleosts (Yang et al. 2021). In addition, uric acid, a product of purine metabolism, is also present in these cells (Luo et al. 2021). Purines accumulate as transparent crystals that reflect incident light and produce blue, silver, gold, or white iridescence (Gempel and Visconti 2020). Notably, there is an intriguing interplay between purine, pteridine, and eumelanin through metabolic pathways. GTP serves as a common substrate in purine synthesis and is converted by guanosine cyclohydrolase (GTPCH), which through subsequent reactions leads to the formation of a pteridine, 4Hbi-pteridine (BH4). BH4 is essential for the formation of various forms of pteridines and is also somewhat related to the formation of eumelanin (Fig. 2).

### Pteridine

As described by Leclercq et al. (2009), pteridines are a group of heterocyclic compounds with pyrimidine and pyrazine rings, classified as pterins or flavins. Many natural pteridines are readily oxidized, sensitive to light, and poorly soluble in water. Within this group, certain pteridines are colorless but fluoresce under UV light, including bipterin and xanthopterin, while others have distinct colors, such as erythropterin and drosopterin (red), leucopterin and isoxanthopterin (white), and sepiapterin and xanthopterin (yellow). Pteridine biosynthesis results in the production of yellow to reddish pigments within pterinosomes located in xanthophores and erythroptores (Braasch et al. 2008).

Pteridines have multiple functions, including immune response, antioxidant activities, neurotransmitter synthesis, and pigmentation. In particular, 7-hydroxybiopterin, extracted from the skin of the cyprinid *Devario malabaricus*, has been identified as an alarm pheromone. It is released after skin lesions and induces a fear response in conspecifics (Leclercq et al. 2009). In addition, 7-hydroxybiopterin is associated with melanogenesis through its role as a cofactor in the conversion of phenylalanine to tyrosine. This process occurs in the melanogenesis pathway, leading to the formation of dopaquinone and ultimately resulting in eumelanin synthesis.



In the study by Ziegler (2003), the regulation and biosynthesis of pteridines during embryonic development of *Danio rerio* was meticulously described. As elucidated by Leclercq et al. (2009), this intricate pathway begins with the action of guanosine triphosphate hydrolase (GTP-CH) on GTP, resulting in the formation of H<sub>2</sub>neopterin triphosphate. This initiates a metabolic sequence leading to the synthesis of drosopterin, sepiapterin, and 4Hbiopteridin. Subsequently, sepiapterin and 4Hbiopteridin play key roles in the generation of a variety of pteridines, as shown in Fig. 2.

### Carotenoids

In aquaculture, carotenoids, in particular astaxanthin, stand out as essential pigments in commercial fish feeds. In Atlantic salmon farming, dietary astaxanthin supplementation can represent a significant portion of production costs, underscoring its importance in achieving desirable pigmentation levels (Sanches 2021). While animals do not synthesize carotenoids, certain fish species exhibit transformations of these pigments that show preferences in absorption efficiency. For example, cyprinids convert zeaxanthin to astaxanthin, while trout convert astaxanthin to zeaxanthin. Interestingly, some species such as salmon and *Pagrus major* do not convert carotenoids, but their absorption efficiency varies; salmonids absorb canthaxanthin and astaxanthin more efficiently than lutein and zeaxanthin, while channel catfish show the opposite pattern, absorbing lutein and zeaxanthin more efficiently than astaxanthin (Sathyaruban et al. 2021).

Although carotenoid synthesis is primarily dependent on plants, fungi, and bacteria, recent research, such as that by Du et al. (2021), has shed light on the genetic regulation of carotenoid transport in chromatophores of common carp (*Cyprinus carpio*). More than 40 genes have been identified as critical for fish pigmentation (Fadeev et al. 2016; Irion and Nusslein Volhard 2019; Salis et al. 2019; Du et al. 2021; Mohammed et al. 2021; Fang et al. 2022; Tang et al. 2022; Poon et al. 2023; Bijun et al. 2024), indicating the complexity of the underlying mechanisms. These findings not only contribute to our understanding of fish pigmentation, but also open avenues for potential selective breeding programs.

Ornamental fish have a high commercial value and their color directly implies this aspect (Luo et al. 2021; Vissio et al. 2021; Lau et al. 2023), as the buyer buys them for their beauty. Based on the data collected in this review, in addition to the direct practical application of promoting sales, it is possible that breeders can incorporate pigments in feed, through industrial amino acids or natural products, in appropriate doses that favor the triggering of the metabolic pathway, providing the action of pigments that promote the shine, color and health of the fish. This will contribute to the development of the feed production sector, which will be able to offer differentiated and higher quality products to the market.

Skin pigmentation is related to aspects of behavior and inter- and intraspecific social interaction of fish (Eaton et al 2016; Svitačová et al. 2024), therefore the production and maintenance of coloration can facilitate the conduct of studies that generate more appropriate management protocols for the breeding and maintenance of these animals. Furthermore, the production of more attractive specimens in captivity helps to reduce extractivism in the natural environment when it is carried out inappropriately, thus reducing the environmental impact of this practice on both the target species and the surrounding ecosystem, thus contributing to the conservation of natural stocks, biodiversity and sustainability of the activity.

The ornamental fish market, driven by dynamic consumer and producer demands, is constantly seeking advances in pigmentation mechanisms to produce new and improved varieties. This compilation aims to provide insight into the regulatory aspects of fish pigmentation, with a particular focus on the role of phenylalanine.

### Conclusions

Numerous amino acids play critical roles in the metabolic pathways governing fish pigmentation, with phenylalanine and tyrosine contributing prominently to coloration through the synthesis of pigments such as melanin and pteridine. While carotenoid pigments dominate commercial fish feed formulations, it is noteworthy that melanin, pteridines and purines are also of paramount importance in both inter- and intraspecific social interactions, as well as in providing protection against predation and ensuring overall animal health. Therefore, supplementing the diet not only with pigments but also with amino acids that facilitate the maintenance or synthesis of these pigments is emerging as a strategic approach in ornamental fish production.





**Competing interests** The authors declare that there is no conflict of interest.

**Authors' contributions** Antonio Cesar Godoy - Conceptualization; Methodology; Writing; General review. Claucia A. Honorato - Conceptualization; Methodology; General review. Dacley H. Neu - Conceptualization; Methodology; Writing; General review. Marcos Paiva Scardua - Writing; General review. Shelby Maura Walker - Writing; General review

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