Into the Pigmentation of Fish: A Physiological Perspective

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Abstract: ***The nature's aim in imparting versatile colours to the fishes is to facilitate their survival and as a mode of communication. Colourations in fishes are highly correlated to their behaviour and habitat as morphological patterns always indicate the physiological functions. An evaluation of the colouration patterns together with the behavioural and physiological adaptation in the natural habitat can interpret the evolutionary behaviour of the particular species. The basic purpose of colouration is to transmit information within species, between species for sexual as well as non-sexual causes. Besides, the peculiar colour patterns in some fishes will help in performing mimicry and camouflage for their life and survival. Thus, the studies on pigmentation will give insights into the physiology and behaviour. Therefore, it is imperative to conduct more studies on the regulation and evolutionary emergence of pigmentation pattern in different fish species. This will also help us to emulate and modulate coloration in fishes, contributing to the improvement of ornamental value of fishes.***

Keywords: fish, pigmentation, physiology, perspective

1. Introduction

Fishes, the most diverse group of vertebrates exhibit innumerable kind of morphological features including different pigmentation patterns. The nature's aim in imparting versatile colours to the fishes is to facilitate their survival and as a mode of communication. Colourations in fishes are highly correlated to their behaviour and habitat as morphological patterns always indicate the physiological functions. Many fishes are exploited from natural environment or raised in confined environment only because of their colouration, which lead to the development of one of the most sought after pet industry called ornamental fisheries. An evaluation of the colouration patterns together with the behavioural and physiological adaptation in the natural habitat can interpret the evolutionary behaviour of the particular species. Each pattern and each colour could provide basic behavioural data about that fish species and give an idea about its mode of communication. The basic purpose of colouration is to transmit information within species, between species for sexual as well as non-sexual causes. Besides, the peculiar colour patterns in some fishes will help in performing mimicry and camouflage for their life and survival. Pigmentation patterns in fish have specialized functions which determine the animal behaviour. The functions may vary like imparting ornamental value, helping for camouflage /mimicry (cryptic), giving warning coloration (aposematic) etc. The changes in the colouration patterns are regulated physiologically and behaviourally. Thus, the studies on pigmentation will also give insights into the physiology and behaviour. Here we appraise the physiological background behind the pigmentation of fish.

2. Chromatophores

Chromatophores are pigment containing cells, which are involved in colouration by regulating the distribution of pigments inside them. In fishes, chromatophores are found in the dermis; in response to various stimuli. The pigments in these cells are transported to or from the centre of the cell during changes in colour. The changes in the distribution of pigment permit the animal to display variations in colour.

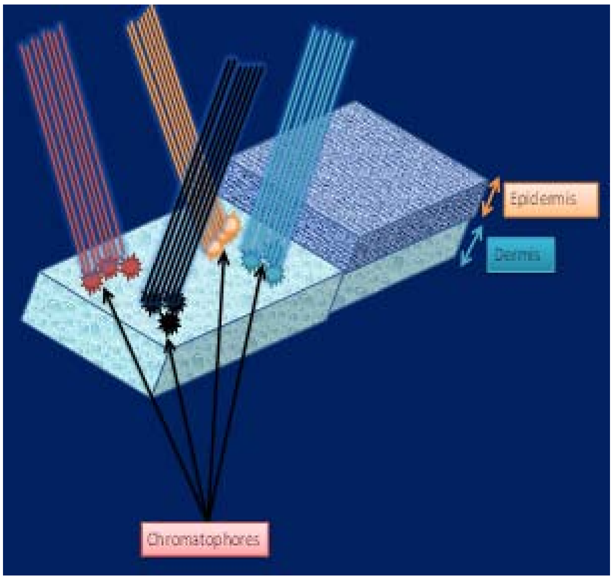

Figure 1: The distribution of Chromatophores in the fish skin

3. Origin of Chromatophores

All fish chromatophores except that of retinal epithelium have originated from the embryonic neural crest. Neural crest is a set of cells that forms at the junction between nascent Central Nervous System and epidermis, i. e the cells placed between neural and non-neural tissues. The migration and formation of xanthophores, melanophores and iridophores are clearly understood. Neural crest migration pathways of melanophores include both lateral and medial pathways. Lateral pathway is the path beneath the ectoderm and medial pathway is through the somites and along the length of the body. Xanthophores are restricted to the lateral pathway while iridophores are restricted to the medial pathway. Nevertheless, pigment cells migrate in an unpigmented state, referred as chromoblasts. In adult zebrafish, the pattern of alternating blue and golden stripes displayed is composed of three kinds of pigment cells - black melanophores, yellow xanthophores and silvery/blue iridophores. Larval xanthophores start to proliferate at the onset of metamorphosis and give rise to adult xanthophores before the arrival of stem-cell-derived iridophores and melanophores. Xanthophores compact to densely cover the iridophores forming the interstripe, and acquire a loose stellate shape over the melanophores in the stripes.

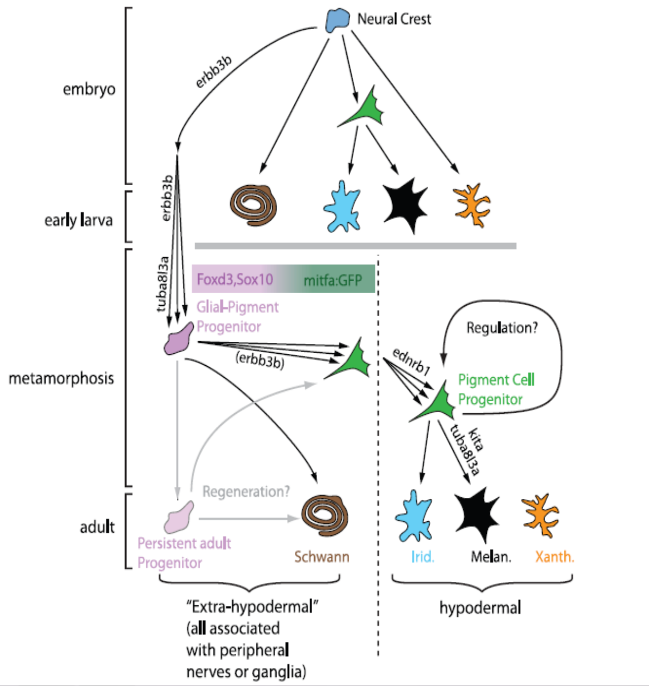

Figure 2: Model for neural origins of metamorphic melanocytes and iridophores (Kelsh R. N. and Barsh G. S., 2011)

Melanophores

Melanophores are also called melanocytes which includes the most important type of chromatophores contains black or dark brown pigment granules (melanosomes). Melanosomes are membrane bound organelles that synthesize and store melanin. The pigment, melanin is synthesised from the amino acid, L-Tyrosine. Many bony fishes have melanophores within a dermal layer on the scales. It is present in both mammals and fishes.

Iridophores

Most of the chromatophores (melanophores, erythrophores, xanthophores and cyanophores) are specialized in producing, storing and translocating their numerous pigment particles. However, Iridophores use refractive guanine platelets to create a silvery appearance or to generate structural colours. e.g.; most blue hues in fishes. It can vary the configuration of platelet-units to alter hue or brightness. These pigment cells can reflect light using plates of crystalline chemochromes made of guanine.

Leucophores

The leucophores, are found in some fish, particularly in the tapetum lucidum. They also utilize crystalline purines (often guanine) to reflect light as that of iridophores and have more organized crystals that reduce diffraction. In a source of white light, they produce a white shine. In fish, the distinction between iridophores and leucophores is not always obvious. In general, iridophores are considered to generate iridescent or metallic colours, while leucophores produce reflective white hues.

Erythrophores

These are the chromatophores which mainly contain red/ orange carotenoids. The ultrastructure of the erythrophores consists of ellipsoidal electron-lucent vesicles that had limiting membranes and inner lamellae which resembles endoplasmic reticulum. The pigment material of erythrophores is composed of two different groups of compounds: orange coloured, organic solvent miscible compounds and yellow coloured water miscible pteridines. The combined morphological and biochemical approaches revealed that pteridine pigments of erythrophores are located characteristically in pigment granules and are the primary yellow pigments of these organelles.

Cyanophores

These are the blue chromatophores and the organelles are termed as cyanosomes. Even though most of the vibrant blue colours in animals and plants are created by structural coloration rather than by pigments, some types of fishes such as mandarin fish, *Synchiropus splendidus* possess cells named cyanophores which carry vesicles of a cyanbiochrome of unknown chemical structure.

Xanthophores

These chromatophores impart a yellow to golden yellow colouration. Xanthophores appear to have supplemental biochemical pathways enabling them to accumulate yellow pigment as compared to most other chromatophores that generate pteridines from guanosine triphosphate.

Table1: Types of chromatophores in fish

Chromatophores	*Color*	*Principle of coloration*
Melanophore	Black	Absorption
Xanthophore	Yellow/orange	Absorption/reflection
Erythrophore	Red	Absorption/reflection
Cyanophore	Blue	Absorption/reflection
Blue iridophores	Blue	Thin film interference
Silver iridophores	Silver	Refraction
White iridophores	White	Scattering

Source: Wucherer *et al.*, 2012

Types of color change

There are 2 types of color changes evident in fishes such as physiological/neural color changes and morphological color changes. Physiological/neural color changes are short term and environment induced color changes involving dispersion or aggregation of pigments. These types of color changes are mediated by neural and hormonal mechanisms. Conversely morphological color changes are long term changes with change in life stages involving variation in the number of chromatophores. They are genetically controlled and specific to the species.

Mechanism of color change

The pigment granules are transported along microtubules at rates of about 1µsec either towards or away from the cell centre. When pigment granules are aggregated at the centre of the cell, major portion of the cell becomes unpigmented and appears lightly coloured, known as *blanching*. When pigment granules are dispersed throughout the cell, the cell is uniformly pigmented and appears darkly coloured, called colouration.

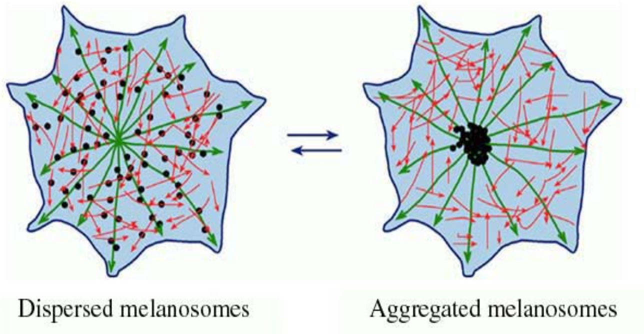

Figure 3: Movement of Melanosomes in melanocytes (*Gelfand and S. Rogers, 1999)*

4. Colouration in ornamental fishes

The fishes with colourful and diverse pigmentation patterns are called ornamental fishes and they are one of the most sought after pets of the world. However, colours and even its absence can have ornamental value e.g. Albino fish. Albino varieties of ornamental fishes have less melanin pigment or they lack melanin. Albinism in animals is considered as a pigmentation abnormality; however it is beneficial for ornamental fishes. The manipulation of colour in ornamental fishes has always attracted researchers. The methods used for the improvement in colouration are genetic engineering and nutritional supplementation of pigments.

(a) Gold fish *(Carassius auratus)* (b) Yellow platy *(Xiphophorus maculatus)* (c) Koi carp *(Cyprinus carpio)* (d) Kissing gourami *(Helostoma temminckii)* (e) Denison's barb (*Puntius denisonii)* (f) Albino butterfly discus *(Symphysodon aequifasciatus)*

(Courtesy: Aquarium, Central Institute of Fisheries Education, Mumbai-400061)

Special functions of colouration in fish

1. ***Camouflage***- a resembling color to the background/environment to escape from the predators
2. ***Conspicuous colouration***- a different colouration from the background so as to attract the mate
3. ***Obliterative shading***- also called counter shading, dorsal dark coloration and ventral light coloration, make the outline of the fish inconspicuous so as to escape from the aerial predators
4. ***Disruptive coloration***- a disruptive pattern, which breaks the solid outline of the body to confuse the predators

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Figure 4: Special coloration patterns in fish (a) Camouflage grouper (b) Conspicuous coloration of lion fish (c) Counter shading in shark (d) Disruptive coloration in Jackknife fish

5. Regulation of Coloration

The regulation and mechanisms of coloration and pigment translocation has been well studied in a number of different species, including amphibians and teleost fish. It has been established that the process is regulated by hormonal or neuronal means or both.

1) Neuronal Regulation

In bony fishes the pigmentation is under the control of sympathetic nervous system whose signals are communicated by chemicals released by neurons. Some chromatophores have 2 types of innervations, adrenergic and cholinergic. Norepinephrine signaling induces the aggregation of pigments, causing blanching, while acetyl choline induces dispersion of pigments, triggering coloration. It is synthesized from dopamine by dopamine β-hydroxylase in the secretory granules of the medullary chromaffin cells. It is released from the adrenal medulla into the blood as a hormone, and is also a neurotransmitter in the central nervous system and sympathetic nervous system, where it is released from noradrenergic neurons in the locus coeruleus. The actions of norepinephrine are carried out via the binding to adrenergic receptors. Norepinephrine signaling induces the aggregation of pigments, causing blanching, while acetyl choline induces dispersion of pigments, triggering coloration.

2) Endocrine Regulation

It mainly includes hormones called melanocortins, which are melanocyte stimulating hormone (MSH), melanocyte concentrating hormone (MCH) and adrenocorticotropic hormone (ACTH). Estradiol and testosterone also affect coloration by way of controlling sexual dichromatism. Melatonin also plays an important role in endocrine regulation of pigmentation. Somatolactin is involved in proliferation and aggregation of chromatophores.The chromatophores respond to circulating hormones (e.g., epinephrine, melanophore stimulating hormone, melatonin) and neurotransmitters (e.g., norepinephrine-SNS) via identified cell-surface receptors. Norepinephrine and epinephrine produce pigment aggregation and paling of the skin (blanching reaction). Pigment cell signaling is mediated through cell surface alpha adrenergic receptors, which respond to catecholamines. Pigment dispersion is activated by an increase in cAMP levels while aggregation occurs when cAMP levels are reduced. In addition, fish integument contains some specialised regulatory factors called MSF (melanocyte stimulating factor) and MIF (melanocyte inhibitory factor).

a) Melanocortins

Melanocortins are hormones produced by the cleavage of POMC (Pro-opio melanocortin) include ACTH, MSH, lipotropin and endorphin. The most important one is MSH (melanocyte stimulating hormone). This peptide hormone from pars intermedia directly acts on melanocytes and stimulates an increase in the tyrosinase activity, melanin content and intracellular levels of cyclic AMP. Instead of cyclic AMP or its analogue, dibutyryl cyclic AMP also can produce same responses through MSH activation.

b) MCH

MCH (melanocyte concentrating hormone) is a cyclic neuropeptide synthesized as a preprohormone in the hypothalamus of all vertebrates. MCH signalling system may also involve in regulating food intake in fishes. This is antagonistic to MSH, which act through cell surface receptors and causes blanching instead of colouration.

c) MSF and MIF

MSF and MIF are intrinsic pigment cell regulatory factors in the integument of fish, which are involved in the differentiation of chromoblasts. It is observed that the inhibitory activity being stronger in the ventral surface. The light-coloured ventral surface of the catfish and other poikilotherms may result from the presence of higher levels of MIF than MSF. On the other hand, the stimulatory activity is more on the dorsal surface of the body in comparison with the inhibitory activity.

3) UV Stimulated Color Change

The ultraviolet light has a positive influence on pigmentation. The UV light not only impacts the amount of pigments produced, but also the density of the pigments or how spread out in the cell they are, which also affects the coloration of the fish. This has been proved in several colored fishes including gold fish. When fish are kept outdoors, they will receive ultraviolet light from the sun. UVA and UVB are both high-energy light, and can be harmful to organisms. Pigments can provide some protection from this. Thus ultraviolet light stimulates the chromatophores to produce more pigment. In contrast to the natural sunlight, artificial lights in aquariums usually only provide a limited spectrum of light and generally do not produce ultraviolet light. Thus, in course of time chromatophores exposed to artificial light will produce less pigment and the fish will look paler, while those exposed to sunlight appears in vibrant color. It is recently reported that some of the fishes e.g. Damselfish use ultraviolet rays for colour generation. The presence and expression of genes encoding UV-sensitive opsins also suggests the importance of UV communication in a number of fish species.

6. Conclusion

There are very few studies aiming to unravel the mechanisms underlying the evolution and expression of pigment-based colour patterns in fish. The diversity of chromatophores in fish itself indicates the importance of pigmentation in its physiological functions and survival. Therefore, it is imperative to conduct more studies on the regulation and evolutionary emergence of pigmentation pattern in different fish species. This will also help us to emulate and modulate colouration in fishes, contributing to the improvement of ornamental value of fishes.

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