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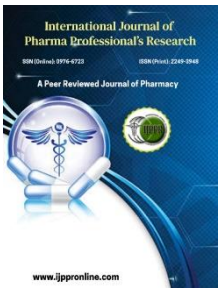

SCIENCE BEHIND THE COLOR CHANGES IN TILAPIA DURING STRESS

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ABSTRACT:
Tilapia is the common name applied to three genera of fish in the family Cichlidae. the genera include Oreochromis, Sarotherodon and Tilapia. Tilapia has become the third most important fish in aquaculture after carp and salmon. The nature's aim in imparting versatile colours to the fishes is to facilitate their survival and as a mode of communication. Colouration in fishes are highly correlated to their behaviour and habitat as morphological patterns always indicate the physiological functions. Some fishes may change its colour due to environmental conditions. Due to the growth of Tilapia production in aquaculture field, indicates how tilapia is important for world's total production and economy.

Introduction:

In the 1960s and 1970s tilapia culture was aimed at the production of food for local consumption, utilizing primarily extensive or semi-intensive culture methods with minimal amount of fertilizer and foods. In the US states of Florida and Texas, Tilapia were originally introduced to curtail invasive plants. In effort to meet the growing demand for tilapia, humans have farmed these fish in countries around the world. The accidental and deliberate introductions of tilapia into the south and southeast Asian freshwater lakes have inspired outdoor aquaculture projects on various countries with tropical climates including Honduras, Papua New Guinea, the Philippines and Indonesia.

Global Tilapia production grew by 3.3 percent in 2020 to top 6 million tonnes for the first time, despite the impact of Covid-19. In China, the largest producers and exporter of tilapia, harvest dropped around 3% in 2020 from 2019. Asia's total contribution to global tilapia production stood at some 66% in 2020, or 4.55 million tonnes. Production in the Middle East Africa and Latin America, meanwhile increased marginally in 2020 as sector development continued.

Tilapia is the common name applied to three genera of fish in the family Cichlidae. The genera include *Oreochromis*, *Sarotherodon* and *Tilapia*. Tilapia has become the third most important fish in aquaculture after carp and salmon. The nature's aim in imparting versatile colours to the fishes is to facilitate their survival and as a mode of communication. Colouration in fishes are highly correlated to their behaviour and habitat as morphological patterns always indicate the physiological functions. Some fishes may change its colour due to environmental conditions. Due to the growth of Tilapia production in aquaculture field, indicates how tilapia is important for world's total production and economy. So, the present article is aimed to review the colour changes in the tilapia during stress condition in aquaculture prospects.

PHYSIOLOGICAL COLOUR CHANGES IN TILAPIA:

Individual kept in captive conditions have been observed to change colour due to stressful condition. Rapid change in colour is important for rapid adjustments to environmental conditions and flexibility of the situation of the environment. Some fishes can change in minutes this is due to neurohumoral regulation of fish in Chromatophores. The instant colour change in fish is called Physiological color change and refers to the synchronous movement of pigment granules within the chromatophores. These pigment granules within the cells are called Melanin granules. Chromatophore consists of Black Melanophore which contains Melanin, Red erythrocytes contain carotenoids, yellow xanthophores which contain pteridine and rare blue cyanophores contain cyanbichrome.

Melanophore are specialized cells derives from the neural crest that contain membrane bound vesicles called melanosomes. Melanosomes are filled with melanin, a dark, non-fluorescent pigment that plays a principal role in physiological color changes in tilapia.

During stress response a fast increase in catecholamine levels to increase oxygen uptake and transport via the blood as well as mobilization of energy substrates. After the brain has processed a stressful condition, the hypothalamus sends a signal to the chromaffin cells in the head kidney via sympathetic nervous pathways to release the catecholamines. This increase in catecholamine level results in the activation of the Hypothalamic sympathetic chromaffin cells (HSC axis).

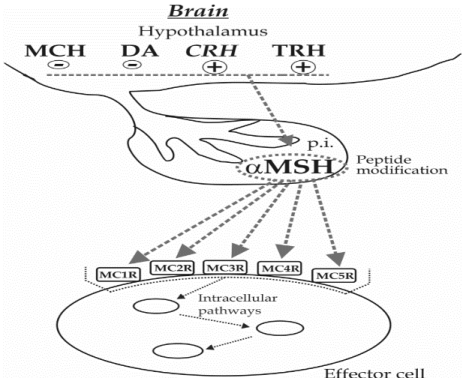
The second axis is also activated, the hypothalamic pituitary interrnal (HPI axis) which increase the release of cortisol from the interrnal cells of the head kidney within a few minutes after exposure to acute stressors. The release of cortisol is primarily under control of ACTH. The increase in plasma αMSH levels upon exposure to stressors would indicate a physiological role for αMSH in the response to those stressors. However αMSH has also been reported to have corticotrophic potencies in tilapia, in combination with β-endrophin (released by hypothalamus and pituitary gland in response to pain or stress). These peptides are derived from the precursor molecules Proopiomelanocortin (POMC). Upon synthesis, this peptide is differentially cleaved into ATCH and opioid β-endrophin(in the corticotropic cells in the pars distalis of the pituitary gland) or in to αMSH (in the melanophore cells of the pars intermedia of the pituitary gland). N-acetylated β-endrophin (the non-opioid form of the peptide) and the two other MSH forms (β MSH and γ MSH) also derived from the POMC precursor hormone.

The release of αMSH in fish is under multifactorial hypothalamic control. MCH and Dopamine (DA) have an inhibitory effect on the release of αMSH in several fishes, while thyrotropin releasing hormone (TRH) and corticotropin releasing hormone (CRH) – stimulate the release of αMSH from the pituitary gland. CRH is the main hypothalamic messenger that stimulates the release of adrenocorticotropic hormone (ACTH) from the pituitary pars distalis which promotes the secretion of cortisol by the interrenal tissue. This hypothalamic-pituitary-interrenal axis is activated in the response to an acute stressor. CRH-stimulated release of αMSH in tilapia. the release of αMSH was stimulated by thyrotropin releasing hormone (TRH) in a concentration-dependent fashion. The EC50 for TRH of 7.4 x 10-10 M is compatible with a physiological function of TRH in this fish. TRH has also been reported to stimulate the release of αMSH. However, in tilapia stimulation of αMSH release from isolated pars intermedia is characterised by an EC50 of 1.6 x10-7 M. the high αMSH levels that arise from this response may also promote skin darkening. In cichlids, DA increases locomotor activity via a D1 receptor in telencephalic circuits. This locomotor activity may serve to find the most suitable or safe habitat. Mok et al. (1998) suggest that DA ergic neurons that mediate responses to fearful stimuli may therefore be activated during stress. DA has also been reported to have a direct effect on the release of αMSH (Hagan et al., 1996; Lindley et al., 1990; Omeljanuk et al., 1989). MCH directly inhibits the release of αMSH by the pituitary gland at picomolar concentrations and this is in line with the reported picomolar concentrations of this hormone in blood plasma of fish (Green et al., 1991). The release of αMSH was inhibited at all concentrations tested. Indeed, MCH has previously been identified as an important colour changing agent through its aggregating effect on chromatophores (e.g. Burton et al., 2000; Fujii, 2000; Nery et al., 1997; Oshima et al., 2001) but it is also recognized as a factor that influences the hypothalamus-pituitary-interrenal (HPI-) axis. higher MCH levels combined with lower cortisol levels (Green et al., 1991). In tilapia, it was shown that concentrations of MCH below 10 µM inhibit αMSH release from the pituitary gland (Gröneveld et al., 1995).

After synthesis of αMSH in fish by the melanotrope cells, acetylation of the peptide can occur prior to the release, αMSH can be found in 3 acetylation states, des-acetylated MSH (no acetyl group), mono or di-acetylated αMSH (one or two acetyl groups). In this particularly di-acetylated αMSH could stimulate the release of cortisol from the interrenial tissue in tilapia, a function that may be potentiated by simultaneous stimulation with β-endorphin.

Recent research has identified 5 types of G-protein-coupled 7- transmembrane receptors that respond to POMC-derived peptides. These melanocortin receptors (MCRs), designated MC1R to MC5R, were first identified in mammals but have now also been identified in fish and show remarkable amino acid sequence homology between mammals, birds and fish. The control of pigmentation of the skin by αMSH is regulated via the melanocortin (MC) 1-receptor that is localised in the membrane of melanocytes. This receptor was first designated the αMSH receptor, as binding of αMSH induced a darkening of the skin. The pathways involved in the functions of the pleiotropic hormone αMSH. Upon synthesis in the Pars intermedia (p i), des acetyl αMSH can be acetylated once (to yield mono acetyl αMSH) or twice (diacetyl αMSH). The release of αMSH can be stimulated by TRH and CRH and inhibited by MCH and DA. αMSH

is transported via the blood to reach an effector cell (e.g. the melanophore, interrenal cells). αMSH can bind to any of the five MC receptors (perhaps excluding the MC2R). The affinity and the expression of these receptors may be altered during stress. Finally, within the cell the signalling pathway may become sensitised, for example in melanophores during adaptation to a black background, or desensitised.



MECHANISM:

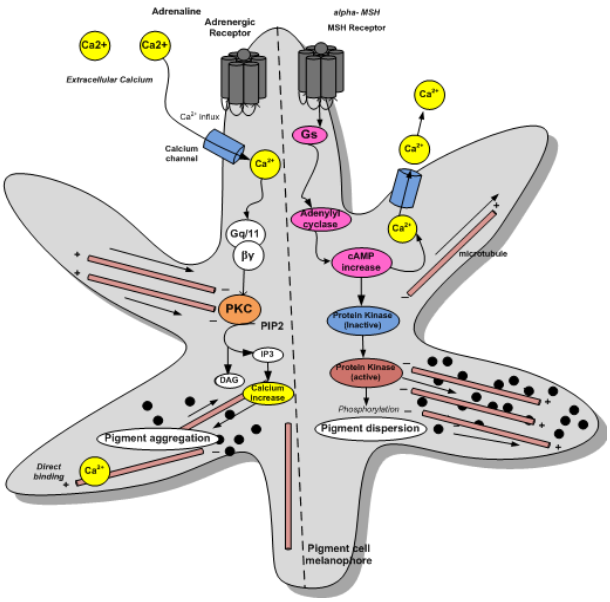
During stress, the hypothalamus-pituitary-interrenal axis is activated (Wendelaar Bonga, 1997). Besides adrenocorticotropic hormone (ACTH), the pituitary gland releases αMSH that induces cortisol release from the interrenal tissue, as has been demonstrated for the tilapia, *Oreochromis mossambicus* (Lamers et al., 1992). Therefore, αMSH is considered the main hormone causing dispersion of melanin granules in melanophores in tilapia and MCH has opposite effects and causes pallor of fish. It is released from the hypothalamus and for a number of species it was shown to inhibit αMSH release. Moreover, MCH exerts a direct effect on cortisol release in fish. Elevated plasma cortisol level is generally used as a main indicator for stress and activation of HPI axis. MCH – inhibit the release of αMSH and cortisol level.

αMSH induces skin darkening by causing the stellar shaped pigment cells (melanophores) to disperse their black pigment (melanin) granules within the cytoplasmic process of the cell. Control level of cortisol release is through negative feedback of the hormone at the levels of HPI axis.

The pigment granules are aggregated at the center of the cell, major portion of the cell becomes unpigmented and appears lightly colored known as blanching. When the pigment granules are dispersed throughout the cell, the cell is uniformly pigmented and near darkely coloured called colouration.

Melanocortins are hormones produced, by the cleavage of POMC include ACTH, MSH, lipotropin and endorphin. The most important one is MSH. 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 c AMP or its analogue, dibutyryl c AMP also can produce same response through MSH activation. It is known that increase in c AMP initiates the influx of Ca^{2+} from the cytoplasm to the endoplasmic reticulum or mitochondria. Whereas the Melanosome dispersion progress in the absence of Ca^{2+} and presence of Ca^{2+}.

The crucial role of Ca^{2+} and MSH in the bidirectional movement of melanophores. Extracellular stimuli (either hormonal: adrenaline or stress response) could result in inward movement of Ca^{2+} from the extracellular matrix. This is followed by activation of Phospholipase c (PLC) promoting 1) inositol triphosphate (IP3) – dependent release of Ca^{2+} from intracellular stored. 2) Diacylglycerol (DAG) – sensitive activation of protein kinase c(PKC), which facilit$_{ates}$ Ca^{2+} entry through voltage dependent calcium channels, resulting into pigment aggregation.



Stimulation of α MSH receptor due to stress condition, couples to GSα proteins stimulates adenylyl cyclase and increase intracellular CAMP resulting into pigment dispersion by activating a calcium pump that drives the expulsion of calcium pump that drives the expulsion of calcium and inactivate protein kinase and activate protein out from the cells resulting kinase by phosphorylation into pigment dispersion.

In case of pigment cell, the action of MCH is enhanced by the absence of intracellular Ca^{2+} which in the case of smooth muscle be an apparent counterpart triggered by β adregenic receptors (NE or Noradrenaline). The actual process of how melanosomes translocate from the concentrated to dispersed state and vice versa is not completely unfurled. However, this mechanism is highly co-ordinate to the colour change in tilapia.

RECENT TRENDS:

Tilapia species are suitable for culture in the heated effluents of power stations. Growth and reproduction may be normal or near normal in saline conditions. Tilapia may have a secondary role as sport fish or in clearing excessive plant growth in power stations.

Impact of different colors of Artificial light on pigmentation and growth performance of Hybrid Red Tilapia (*Oreochromis mosambicus* X *Oreochromis hornorium*) reared in saline well water. Effect of light intensity and Photoperiod on the growth and stress response of juvenile Nile Tilapia (*Oreochromis niloticus*) in RAS. Effect of environmental factors related to stress and change in body pattern of Tilapia. Effect of colour production in Tilapia with cross breeding with flowerhorn.

CONCLUSION:

In this thesis, the function of the hormone αMSH (α-melanophore stimulating hormone) in the skin colour control and in response have been noted in tilapia. Acute stress can induce a rise in plasma αMSH levels and chronic low water pH can increase plasma αMSH in tilapia. Melanophores do show a dispersing response after stimulation with three isoforms of αMSH. The mechanism of colour change is seen in this thesis and the hormones responsible to physiological colour change in stress condition in tilapia. Therefore, αMSH is considered the main hormone causing dispersion of melanin granules in melanophores in tilapia and MCH has opposite effects and causes pallor of fish which results in the physiological colour change in tilapia.

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