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Seasonal variations in chromatophore index in fish *Puntius sophore* from Jammu water bodies, Jammu and Kashmir (India)

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Abstract
The paper deals with the seasonal changes in colour/chromatophores in *Puntius sophore.* In the present studies chromatophore index has been calculated throughout different seasons for different size groups from the scales of the fish *Puntius sophore* and observations are recorded. Table 1 shows the average chromatophore index in all size groups in the month from June to August, the chromatophore index for A, B and C size groups in the month from June to August (Monsoon) is 49.33±3.21 to 51.04±2.64; 54.43±1.96 to 60.13±1.34 & 59.16±1.52 to 62.39±1.42 respectively. September onwards, the chromatophore index follows a decline trend, the values being 27.49±1.15 to 38.53±2.01; 28.36±2.03 to 47.23±1.03 & 31.46±2.03 to 48.61±1.33 for A, B & C size groups respectively. This indicates that chromatophores are concentrated in the monsoon and in post monsoon the chromatophore index decreases i.e. the chromatophores are dispersed in the post-monsoon. Colour changes result from dispersion and/or concentration of pigment granule with in epithelial chromatphores. In muddy water the chromatophore index is higher than that of the clean water. The results clearly show that during the monsoon period when there is a plenty of floods and water becomes muddy in the streams and rivers, the chromatophores begin to concentrate in the scales of the fish and density of chromatophores increases and develop dark black colours so as to become visible.

Keywords: *Puntius sophore,* chromatophore index, colour, concentrated, dispersed

Introduction

The fisheries of Khulna district consist of inland open water fisheries and fresh water aquaculture. Like other parts fisheries sector of the country, particularly fisheries of Khulna district is a major source of nutrition, income, employment and livelihood support of the local people.

Of all the vertebrates, fish have the greatest number of color producing cells. The countless colors and patterns expressed by fishes are principally determined through genetic mechanism. It is therefore quite understable why ichthyologists continue to use color dress to distinguish smaller taxonomic categories such as species and subspecies (Lagler *et al.,* 1962 [29] and Malek, 200) [30]. Usually, fish coloration is derived from a number of sources. A background coloration comes from underlying body tissues, bodily fluids and sometimes, even gut content. Overlying hues are produced by specialised color cells called chromatophores or less often by bioluminescent structures called photophores (Lagler *et al.* 1962) [29]. Chromatophores produce either structural or pigmentry coloration (Malek, 2008) [30]. Seven distinct sorts of chromatophores can be found in fish-irridiophores, leucophores, melanophores, xanthophores, erythrophores, *cyanophora* and erythro iridophores (Hickman *et al.* 1995 [28]; Goda *et al.* 2013) [27]. Bagnara and Hadley, 1973 [26]; Aspengren *et al*., 2009 [2] reported rapid color change on the scale of fish, termed physiological color change, is typically accomplished via mobilization of pigments within specialized cells called chromatophores. Although physiological color change has been observed in insects (Hinton and Jarman, 1973) [17], arachnids (Insausti and Casas, 2008) [18], crustaceans (Brown and Sandeen, 1948) [5], and cephalopods (Florey, 1969; Hanlon and Messenger, 1988; Messenger, 2001) [14, 16, 20], the structures and mechanisms of invertebrate color change are markedly different than those of vertebrates (Umbers *et al*., 2014) [25].

Many animals undergo physiological color changes in order to adapt to their environment and enhance their chances of survival and reproduction. These changes depend on the arrangement of pigments in dermal chromatophores. Melanophores are specific types of chromatophores studied to understand pigment (melanosome) transfer. Nascimento *et al*., 2003 reported when melanosomes aggregate towards the cell center, paling occurs. Melanosome dispersal throughout the cytoplasm results in darkening.

Present studies on seasonal variations in chromatophore index in fish *Puntius sophore* were conducted and chromatophore index was generated in different size groups throughout the year.

Materials and Methods

The fish *Puntius sophore* were collected from a stream at Gho-Manhasan located at a distance of 20 km North West of Jammu city. Captured live fishes were acclimatized in the laboratory in plastic troughs at room temperature for about 6-7 days before the start of experiment. These fishes were fed on live feed and artificial diet.

Procedure for chromatophore analysis: The fish *Puntius sophore* of different size groups were used as the experimental animals.

The scales of fish were removed with the help of fine forceps and these were immediately transferred into the glass petridishes containing 0.7% NaCl. Thereafter the material was now on the slide to study the chromatophore under stereo microscope.

Digital photography

Standardised digital photography of chromatophores was carried out to study the chromatophore index in fish.

Chromatophore index calculation

Chromatophores of different colors were observed in different stage ranges from 1st to 5th stage in fish. A comparative study of chromatophores was carried out on monthly basis by counting the chromatophores of fish under the stereo microscope and chromatophore index was calculated by using the formula:

$$C.I = C.S \times C.NO$$

C.I. = Chromatophore Index. C.S. = Chromatophore Stage. C.No. = Chromatophore Number.

Results and Discussion

Fishes are able to change their colouration or shade in response to numerous environmental changes and can exhibit a great variety of pigment colours (Fingerman, 1970 and Fuji, 1993). This ability to match body colour with their background provides a very useful mechanism for avoiding predators. The ability to change colour to match environmental changes (physiological colour change) is brought about by pigment movement within the different chromatophore present at different sites of fishes, so as to visualise as to how fishes go for colour change according to background adaptation. In Present study chromatophore index has been calculated year round for different size groups from the scales of the fish *Puntius sophore* and observations are presented in Table 1 & 2.

Average chromatophore index as evident from the table 1 is highest in all the three size groups A, B and C in the month from June to August (Monsoon), the values being 49.33±3.21 to 51.04±2.64; 54.43±1.96 to 60.13±1.34 & 59.16±1.52 to 62.39±1.42 respectively. September onwards, the chromatophore index follows a decline trend, the values being 27.49±1.15 to 38.53±2.01; 28.36±2.03 to 47.23±1.03 & 31.46±2.03 to 48.61±1.33 for A, B & C size groups respectively (Table1).

This indicates that chromatophores are concentrated in the monsoon and in post monsoon the chromatophore index decreases i.e. the chromatophores are dispersed in the post-monsoon (Figure 2 and 3). Colour changes result from dispersion and/or concentration of pigment granule with in epithelial chromatphores. In muddy water the chromatophore index is higher than that of the clean water. The discovery of hormonal involvement in the control of pigment movement in crustacean chromatophores was reported by Koller, 1927 [19] and Perkins, 1928. [24] Fingerman (1955) [6] had made an extensive contribution towards the elucidation of the regulation of crustacean pigmentary effectors especially in area such as localization and differentiation of pigment-effector hormone. Similar contributions were made by Fingerman (1963, 1966,1969,1970,1985 and 1988) [7-12] and Figerman *et al.* (1994) [13] on pigment concentration and dispersion.

In the present study, the perusal of the Table 1 & 2 & Figure 1 to 13 reveals the chromatophore index for all the three size groups, chromatophore index was calculated monthwise during the year and results observed revealed that chromatophore index of A, B & C size groups of *Puntius sophore* in the pre-monsoon (March to May) to be 45.86±1.52 to 48.13±1.00; 52.33±2.51 to 54.43±1.96 & 56.96±1.73 to 59.16±1.52 respectively and in winter (December to February) is slightly lower for all size groups, A, B & C size groups viz., 38.53±2.01 to 41.39±2.30; 47.23±1.03 to 50.03±1.73 & 48.61±1.73 to 55.36±1.86 than the monsoon period (June to August) (49.56±2.08 to 51.04±2.64), (54.43±1.56 to 60.13±1.34) & (61.06±1.73 to 69.39±1.42) respectively & post-monsoon (September to November) (27.49±1.15 to 37.53±2.30), (28.36±2.03 to 46.66±2.51), (31.46±2.03 to 46.67±2.51) for A,B and C size groups respectively.

The results clearly show that during the monsoon period when there is plenty of floods and water becomes muddy in the streams and rivers, chromatophores concentrate in the scales of the fish and density of chromatophores increases and develop dark black colouration. This explains why chromatophore index is higher in the monsoon period. The chromatophore index in C size groups is higher than other two groups. The larger size group *Puntius sophore* exhibit more chromatophores than the smaller size group fishes. During the other period of the year in which water is transparent and clear the chromatophore get dispersed and this decreases the chromatophore index. The chromatophores, chiefly melaonophore predominates during the monsoon period as compared to other groups of chromatophores viz; erythrophores, xanthophores, iridiophore and are responsible for overall dark colouration of the fishes. ANOVA demonstrate that chromatophore index differs significantly at 1% level of significance ($P<0.01$) during the year and show significant seasonal differences in chromatophore indices for all the size groups.

The results of the present study do agree with the observations given by Birtz and Piennar, (1992) [4]; Appelbaum and Kamler, (2000) [1] while studying pigmentation in fish species they studied. Mona *et al.* (2011) [22] have reported similar results of chromatophores in the Arctic under-ice amphipod *Apherusa glacialis.* Miner *et al.* (2000) [21] and Auerswald *et* *al.* (2008) [3] have reported that coloration in fishes is different during different seasons which may be in response to background colouration and light intensity and simply support present viewpoint that chromatophore index being highest during monsoon period is due to muddy water and chromatophore index is lowest during winter season when water is more clear in streams and rivers.

Conclusion

Presently chromatophore index has been calculated throughout different seasons for different size groups from the scales of the fish *Puntius sophore*. Average chromatophore index has been found to be highest in all the three size groups A, B and C in the months from June to August (Monsoon) and lowest during the months of September to December (Post-Monsoon and winter). The results clearly showed that during the monsoon period when there is plenty of floods and water becomes muddy in the streams and rivers, chromatophores concentrate in the scales of the fish and develop dark black colours. This explains why chromatophore index is higher in the monsoon period.

Table 1: Seasonal differences in chromatophore index in *Puntius sophore* of A, B & C size groups (A: 2.5-3.5 cm; B: 3.6-4.5 cm & C: 4.6-5.5 cm)

Months/Fish	A (2.5-3.5 cm)	B (3.6-4.5 cm)	C (4.6-5.5cm)
September	27.49±1.15	28.36±2.03	31.46±2.03
October	36.66±1.15	38.23±2.03	40.13±2.01
November	37.03±2.30	46.66±2.51	46.67±2.51
December	38.53±2.01	47.23±1.03	48.61±1.33
January	40.61±3.05	50.33±1.46	51.87±1.52
February	41.39±2.30	50.03±1.73	55.36±1.86
March	45.86±1.52	51.03±2.08	56.96±1.73
April	46.36±0.57	52.33±2.51	58.63±1.52
May	48.13±1.00	52.16±2.08	59.72±2.08
June	49.33±3.21	54.43±1.96	59.16±1.52
July	49.56±2.08	58.23±1.56	61.06±1.73
August	51.04±2.64	60.13±1.34	62.39±1.42

Table 2: Seasonal differences in chromatophore index in *Puntius sophore* of A, B & C size groups (A: 2.5-3.5 cm; B: 3.6-4.5 cm & C: 4.6-5.5 cm)

Season	Size group		
	A (2.5-3.5 cm)	B (3.6-4.5 cm)	C (4.6-5.5cm)
Post-monsoon (Sept-Nov.)	35.66	41.63	42.83
Winter (Dec-Feb.)	39.43	50.03	54.31
Premonsoon (March- May)	47.33	51.63	59.63
Monsoon (June –July)	50.76	59.37	61.76

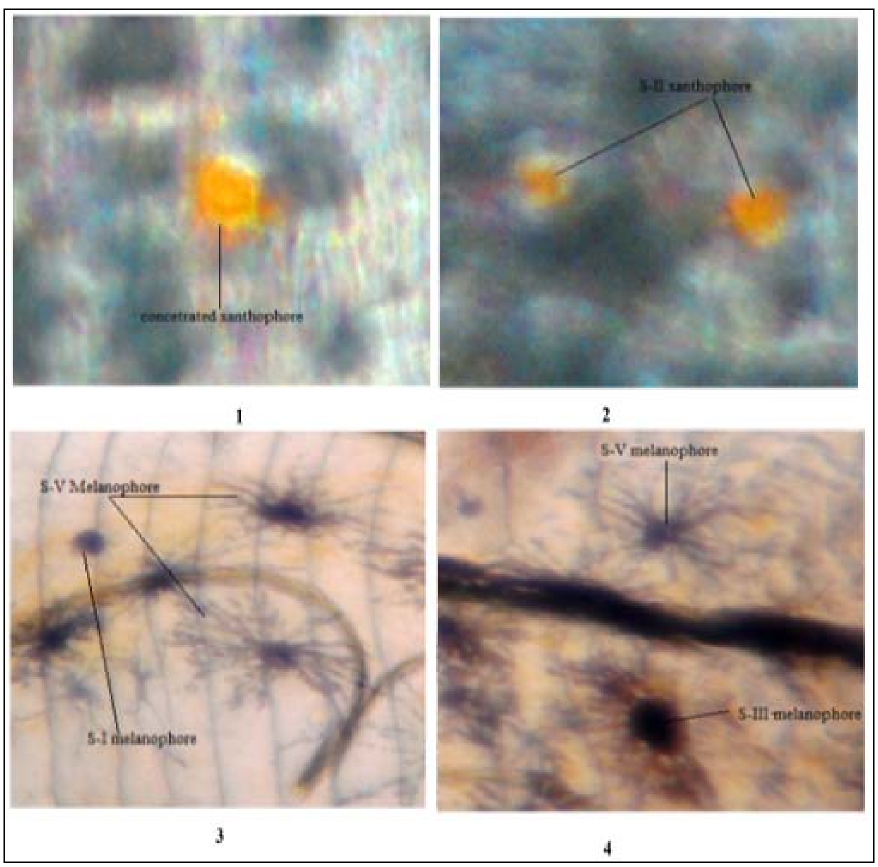
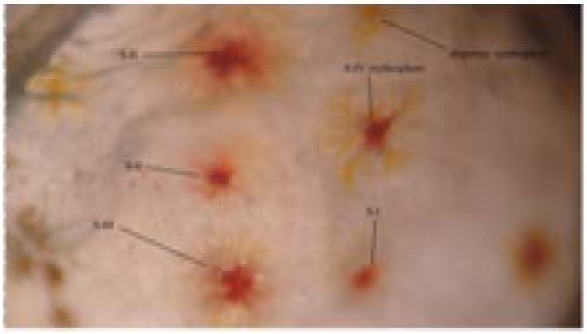


Fig 1: Photograph of scale of *Puntius sophore* showing cocentrated xanthophore of s-II.
Fig 2: Photograph of scale of *Puntius sophore* showing cocentrated xanthophore of s- I & II.
Fig 3: Photograph of scale of *Puntius sophore* showing cocentrated and dispersed melanophores of s-I and V.
Fig 4: Photograph of scale of *Puntius sophore* showing cocentrated and dispersed melanophores of s-III and V durinh monsoon season.
S-I = Highly concentrated chromatophore S-II = Concentrated chromatophore S-III = Concentrated chromatophore S-IV = Dispersed chromatophore S-V = Highly dispersed chromatoph



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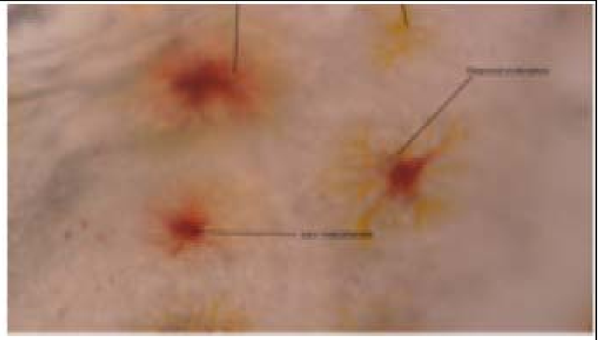

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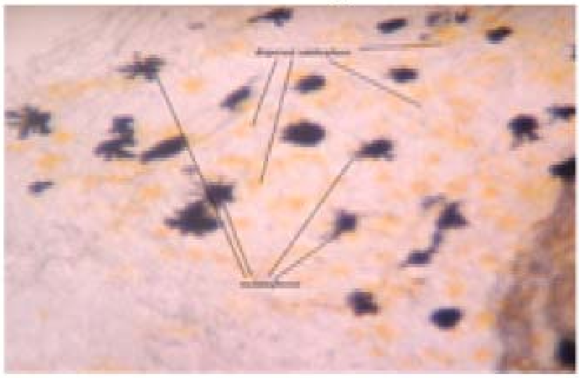

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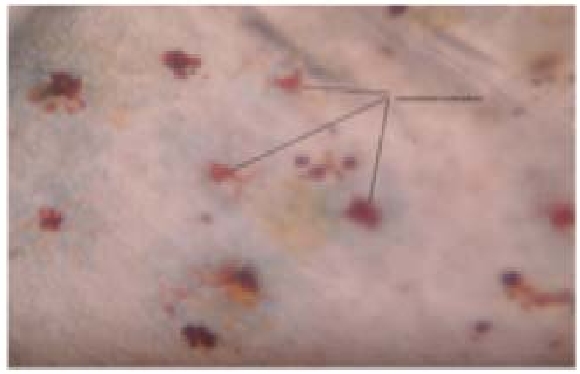

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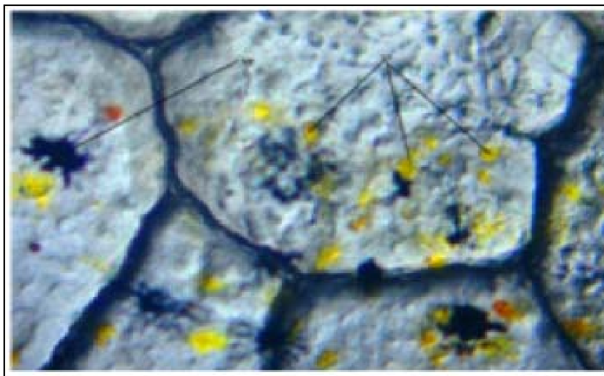
Fig 5: Photograph of scale of *Puntius sophore* showing cocentrated and dispersed erythrophores of stage-I to V.
Fig 6: Photograph of scale of *Puntius sophore* showing cocentrated and dispersed erythrophores and xanthophores of s-I -V.
Fig 7: Photograph of scale of *Puntius sophore* showing dispersed xanthophores and concentrated melanophores.
Fig 8: Photograph of scale of *Puntius sophore* showing cocentrated erythrophores of s-I -III.



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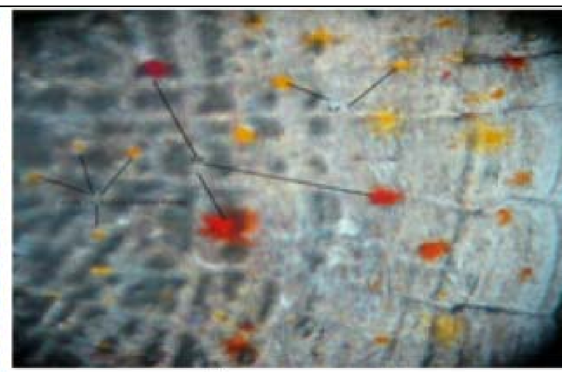

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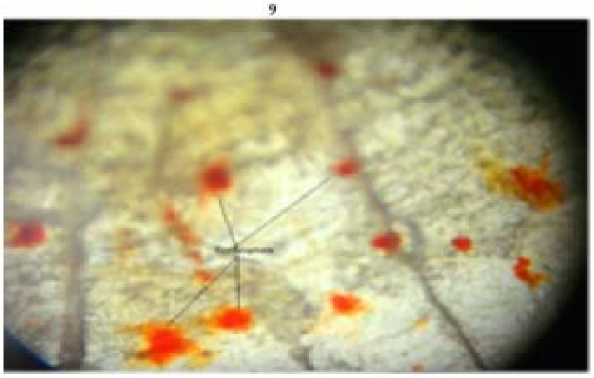

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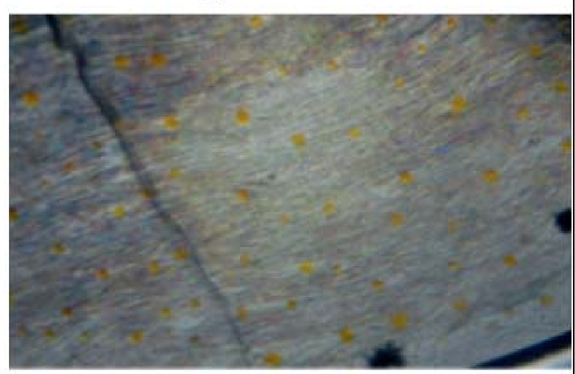

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Fig 9: Photograph of scale of *Puntius sophore* showing cocentrated xanthophores of s-I -III.
Fig 10: Photograph of scale of *Puntius sophore* showing cocentrated erythrophores and xanthophores of s-I -III.
Fig 11: Photograph of scale of *Puntius sophore* showing cocentrated erythrophores and of s-I -III.
Fig 12: Photograph of scale of *Puntius sophore* showing cocentrated xanthophores of s-I -III.

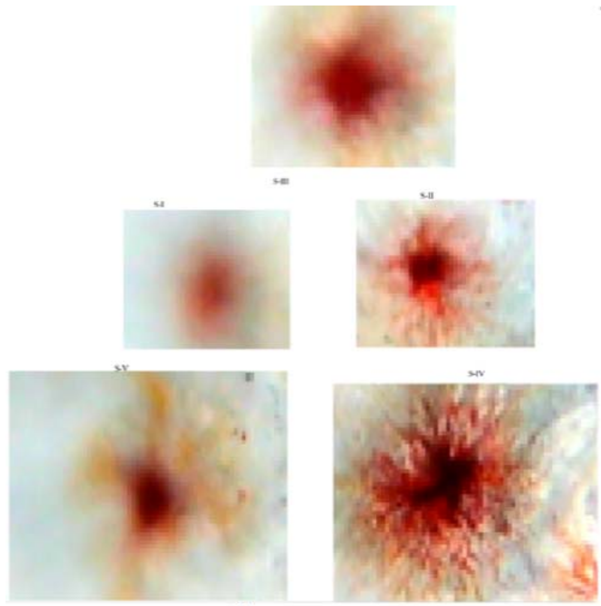

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Fig 13: Photograph of scale of *Puntius sophore* showing chromatophore index of Stage-I to V.
S-I = Highly Concentrated S-II = Concentrated chromatophore S-III = Concentrated chromatophore
S-IV = Dispersed chromatophore S-V =Highly dispersed chromatophore

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