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## **ORIGINAL ARTICLE**

## Haematological Assessment of Indian Roofed Turtle (*Pangshura tecta*) in River Chambal

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#### ABSTRACT

Author has selected three experimental stations in river Chambal subsidiary of Yamuna from Dholpur to Morena, author took blood samples from turtle Panshura from three different locations in river and investigated 16 haematological parameters such as No. of RBC ( $\times 10^{6}/\mu l$ ) 0.45-0.88, Haematocrit (%) 20-26, WBC,s (×10<sup>3</sup>/µl) 11.5-14.5, Heterophils (%) 30.5-48.5, Lymphocyte (%) 30-40, Monocytes (%) 2-4, Eosinophills (%) 10-14, Basophils (%) 00-2, Glucose (mmol/L) 5-11.5, Uric Acid (mmol/L) 0.022-0.01, Hb (%) 4.5-8.5, Protein (g/dl) 3.1-8.5, MCV (fL) 21.8-30.4, MCH (pg) 0.33-0.54, Thrombocytes (%) 8-12, Azurophils (%) 2-6. Entire study was conducted for year 2015. Quarterly samples were collected and brought to laboratory and all parameters were estimated there. Except couple of parameters most of those ones were in the limit prescribed as well as were corroborated with expected values. After applying chi square test over variants at site-A this concludes that a difference was determined between observed and expected values while critical value was bit less than chi-square (24.99<29.12); at site B. This concludes that a no difference was determined between observed and expected values while critical value was more than chi-square (24.99>15.55); at site C. This concludes that a no difference was determined between observed and expected values while critical value was more than chi-square (24.99>14.95). It was conclude in this research output that even after dirtiness of river as per literature of researches findings available although water analysis was not done by author, no anyone blood inclusion of three sites seated turtles was affected. It was verified and proved by statistically in this paper.

Key Words: thrombocytes, chi square test, dirtiness, blood inclusion

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#### **INTRODUCTION**

Bulk of surface water in comparison to ground one is dirty (polluted) and the condition is getting most worsen by excess anthropogenic activities and promptly industrialization and growth of secondary sector. Untreated domestic sewage, industrial effluents and other source s byproducts on the rampage in to inland water bodies specially rivers have arrived in danger. Deteriorating water quality, intrusion of toxicants is making water unfit for all life forms from invertebrates to vertebrates including man in hydrosphere ecosystems. The harmful lethal chemicals affect severely morphology, anatomy and physiology including haematology of the all aquatic organisms.

Moreover Haemotoxins change blood components characteristics both qualitatively and quantitatively thus nosy the normal functions [1], and leading to disorders and water born ailments [2]. Animal fecundity, survival graph, reproduction severely are affected when water gets spoiled. The requisite for adding knowledge about blood parameters is

how far significant especially for domestic animals has now become inevitable, [3] due to its prominence in the diagnosis of many diseases [4].

The many species of freshwater turtle, listed in Schedule I under the Wildlife (Protection) Act, 1972 (amended in 1991), is under serious threat mainly due to high level pollution in their habitat. Local people always try to hunt such species for food. This paper deals with the study of haematological status of roofed turtle (*Pangshura tecta*) in relative polluted water. The haematological profile can provide a base line data comparing with such animal's status towards water quality itself and haematology of other aquatic fauna as well in polluted waters with various degrees. Big advantage of conducting such type study about haematological profile of freshwater species especially threaten ones like turtles, crocodiles in polluted environments is to suggest appropriate conservation measures to government and other conservating agencies anywhere.

The present study is an attempt to collect and add information on haematology to the existing knowledge of blood parameters of other turtles from fresh water. This study also explores the commercial potentiality of turtle farming.

In physiological milieu hematology has been inordinate concern for the scientists as it is very imperative getting the results and details richness and nature of wild and modern aquatic species in aquatic lotic ecosystems which can be used as diagnostic tool. A complete blood picture of several parameters let us know latest and gather information about normal and abnormal health of the animal. If any disturbance or ups and down seen in these parameters by environmental and man-made factors, it becomes necessary for the academicians to explore cause and effect of such changes in the living environment and their impact over physiology of aquatic animals. In this connection author did experiments on hematology of Panshura in river Chambal at stretch in between Dholpur to Morena and tried to find out the how far variations in blood parameters can affect physiology of turtle in study.

The study was conducted at three different Ghats of River Chambal between Dholpur and Morena, located between Latitude 26°41' 34.30"N to 26°29' 56.11"N and Longitude 77°52' 46.85"E to 77°59' 43.22"E. At the experimental points or stations where town sewage water and other nallahs carrying effluents from whatever small industries and tanneries join the mainstream of river. Three turtles from three pre-opted points weighing 0.800 kg., 1.00 kg and 1.200 kg got achieved from local fisherman of the area.

## MATERIALS AND METHODS

About 6 grown matured *Panshura tecta* have been collected from the three of stations A, B & C in Chambal River of Dholpur-Morena stretch and have been used for experiments according to plan of study. They were kept separately in moderate sized terrarium with a weighty down dish at one end for incoming water and so long as with eco-friendly in nature. The floor of all terraria was filled with a steady slope of sandy mud and pebbles.

Before introduction the turtles in the terraria, they got washed cleanly carefully to remove dirtiness. It was obvious that turtles acclimatized after some time naturally. What is more turtles were fed once in 2 days. The blood was taken from 3 turtle out of 6 on random basis through sterilized with 22 SWO hypodermic needles having a pit in plastron above the heart (cardiocentesis) with no dissection. This blood put into refined vials with right anti-coagulant.

For the hematological parameteric analysis, the hematocrit was determined with a microcentrifuge. Hemoglobin was assessed using Drab-kin's solution or by Sahli's haematometer. The RBC and leukocytes counts and the red blood cell index were estimated by methods followed of Garcia-Navarro and Pachaly [5], in which a Neubauer chamber and light microscopy used. The DLC and the thrombocyte were estimated using blood smears, rapid Panoptic staining, and light microscopic analysis. Moreover other methods were deployed for Erythrocyte count, haemoglobin, haematocrit value, plasma

protein, blood sugar, blood uric acid and other physiological parameters as per the methods provided by Kolmer *et al.* [6].

S.No.	Haematological Parameter	Site A	Site B	Site C
		(Mean)	(Mean)	(Mean)
1.	No. of RBC (×10 <sup>6</sup> /µl)	0.58	0.88	0.45
2.	Haematocrit (%)	26	24	20
3.	WBC,s (×10³/µl)	14.5	11.5	12.5
4.	Heterophils (%)	48.5	38.5	30.5
5.	Lymphocyte (%)	40	34	30
6.	Monocytes (%)	2	2	4
7.	Eosinophills (%)	10	14	11.5
8.	Basophils (%)	2	1	00
9.	Glucose (mmol/L)	11.5	6.5	5
10.	Uric Acid (mmol/L)	0.022	0.075	0.01
11.	Hb (%)	8.5	4.5	5.5
12.	Protein (g/dl)	4.5	8.5	3.1
13.	MCV (fL)	30.4	27.4	21.8
14.	MCH (pg)	0.54	0.74	0.33
15.	Thrombocytes (%)	12	20	8
16.	Azurophils (%)	6	2	3

Table 1: Haematoloy of Panshura at three Stations A, B & C in Chambal River in year 2015



Fig. 1: Site A



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Fig. 4: Comparative values of 16 parameters at all three sites

## **OBSERVATIONS**

16 parameters were taken under this study from different sites A, B and C at Dholpur-Morena stretch spread over 20 km. Samples were collected quarterly i.e., total four times in a year from January 2015 to December 2015. All values estimated as mean values. Findings were presented in the tabulated form.

Range of parameters was from No. of RBC (×10<sup>6</sup>/µl) 0.45-0.88, Haematocrit (%) 20-26, WBC,s (×10<sup>3</sup>/µl) 11.5-14.5, Heterophils (%) 30.5-48.5, Lymphocyte (%) 30-40, Monocytes (%) 2-4, Eosinophills (%) 10-14, Basophils (%) 00-2, Glucose (mmol/L) 5-11.5, Uric Acid (mmol/L) 0.022-0.01, Hb (%) 4.5-8.5, Protein (g/dl) 3.1-8.5, MCV (fL) 21.8-30.4, MCH (pg) 0.33-0.54, Thrombocytes (%) 8-12, to Azurophils (%) 2-6.

# **RESULT AND DISCUSSION**

These freshwater turtles play magnificent role in rivers by feeding other aquatic animals like insects, snails, crabs, organic matter and dead biotic portions which ultimately impede water pollution of water bodies. Beside this they act as scavenger in removing waste material from rivers. Many scientists around the globe conducted different studies on several aspects of turtles ecological and biological variants, like Mertens [7], Bennette *et al.* [8], Fugler [9], Dass [10, 11], Bhupathi and Vijayan [12, 13], Rashid and Swingland [14], Nicholas [15], Hossain [16], Sandra and Daniela [17].

But in Indian perspective the research work on turtles ecology, demography, cytology and Genetics, fecundity and reproduction, aging and survival, evolution has not been done in appropriate proportion so far although few scientist like Smith [18], Pritchard [19], Das [20], Moll [21, 22] have completed their study on taxonomy and distribution upon the turtles.

Work on turtle haematology was done by many scientists but very little statistically verification or significance was done by researchers but author in this work by chi square formulation proved his research significantly.

As continuous watch and analysis on and of water from different water bodies especially rivers by hydrobiologist shows ruined water quality trend as composition of water getting changed negatively on account of daily anthropological needs.

The haematological values run with the water quality of rivers. Additionally changes in the water quality in turn influence blood composition. Changes in RBC, WBC, Platelets, organic and inorganic ions in plasma may give rise to imbalance in the homeostasis of the water animal.

Such haematological changes like reduction in oxygen transportation efficiency, low Hb, thrombocyte clumping, high leucocytes, leukopenia, haematourea, hyperglycemia etc. may lead to pathological and diseased conditions [23, 24].

Presences of heavy metals in water bodies affect blood morphology as raising acidity of blood. Researches on other turtles indicated the pH of blood should be in basic range [23, 4]. Thus blood picture at any point of time from at any point of river can reflect the degree of pollution and regular checking is indispensible to determine the extent of lethality to be counter by aquatic species. The information from such research output may be used to plan a strategy in order to conserve and survive of the endangered species.

C.E.S. Zago et al. [25] investigated blood picture as Hemoglobin polymorphism and hematological profile of Geoffroy's side-necked turtle (*Phrynops geoffroanus*, Testudines) in the northwestern region of Sao Paulo State, Brazil and enumerated results as mean hematocrit values were 24.0% for males and 22.5% for females. For total hemoglobin, they found mean values of 7.22 g/dL in males and 6.66 g/dL in females. For thrombocytes, they found mean of 12.0% for females and 16.5% for males while RBC count showed mean of 7.4 x  $10^{5}/\mu$ L for males and 5.9 x  $10^{5}/\mu$ L for females. While in present study No. of RBC (×10<sup>6</sup>/ $\mu$ l) 0.45-0.88. The number of TLC was 2775 x 10<sup>3</sup>/ $\mu$ L for males and 2725 x  $10^{3}/\mu$ L for females. There was a predominance of heterophils, eosinophils, and monocytes in both sexes. No significant differences were found between males and females for hematological parameters. They compared hematological results with literature data for other Chelonia. Range of parameters was similar to what is known for fresh water turtles even like in present study on Panshura of Chambal River. In present study all parameters estimated in the limitations corroborated the findings of Zago et al. The parametric test used to compare averages was the Student t-test, while in present study chi square test was applied.

Study conducted by Moon and Foerster, [26], on *Chrysemys picta, Chelonia agassizzi, Dermochelys coriacea*, and study of Wood and Ebanks, [27] on *Chelonia mydas* and found Hematocrit (%) range 19-45 and Hb range was 6.6-12.5 (g/dL). Hutchison and Szarski [28] mentioned that geographic location may influence hematological values, particularly red blood cell count. We made the same observation in our study when we compared the results obtained for this species to those obtained by Brites and Rantin [29] in another region in Brazil. Solanki [4] conducted study on Yamuna River and estimated haematological parameters which shown similarity with present work.

The average white blood cell count was similar to that of some turtle species, such as *Chelonia agassizii* and *Chelonia mydas*, and was higher than those values found for *Chrysemys picta* and *Lepidochelys kempi* (Moon and Foerster,) [26]. Different values were found for the different types of leukocytes, which is likely a reflection of environmental variations and physiological adaptations.

In present investigation all values of 16 haematological parameters were mean values from 12 months study in year 2015.

At site-A statistically calculation was done and observed that a difference was determined between observed and expected values while critical value was bit less than Chi square (24.99<29.12) In this case since  $H_0$  was rejected hence at site-A statistically little difference was noted down which proved that limnological parameters (not done in present study) on the basis of other work done might have had little effect on hematology of Panshura, but effect is nil practically.

At site-B statistically calculation was done and observed that no difference was determined between observed and expected values while critical value was more than Chi square (24.99>15.55). In this case since  $H_0$  was accepted hence at site "B" statistically no difference was noted down which proved that limnological parameters had no effect at all on hematology of Panshura.

At site-C statistically calculation was done and observed that no difference was determined between observed and expected values while critical value was more than Chi square (24.99>14.95). In this case since  $H_0$  was accepted hence at site-C statistically no difference was noted down which proved that limnological parameters had no effect on hematology of Panshura.

#### CONCLUSION

At all experimental stations Haematolgy of Panshura, a roofed turtle was unaffected on account of dirtiness and pollutional conditions of river Chambal, although water quality or pollutional parameters were not analyzed in present study. In line of plenty of researches done earlier on different rivers across India and abroad it was concluded that water quality must had affected flora and fauna of aquatic bodies. River Chambal flows in three states of India and subsidiary of river Yamuna. Both rivers are considered polluted like many others rivers in India. Many works have been done with respect to water quality and relation with aquatic fauna. Very little information is available in research area over hematology of turtle as did by present author. At site-A very little statically difference was observed due to moderate variations in parameters, what was the reason behind it, not discussed in research while rest of two sites-B and C no statically difference was observed as little variations are seen in parameters. Moreover irrespective of any point of river, if turtle had taken, no significant difference would have observed. Even by disturbances in hematological variants, it is yet to study the Morpho-pathological manifestation in turtles. Turtles even now have longest life span ever among aquatic fauna.

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