International Journal of Science and Research (IJSR)
ISSN (Online): 2319-7064
Index Copernicus Value (2015): 78.96 | Impact Factor (2015): 6.391

Haematological and Serum Biochemical Analyses of Wild Male Blackrats, *Rattusrattus* (Linnaeus, 1758)

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Abstract: ***The present study focuses to evaluate the haematological and biochemical profile of wild black rats. In this study, the estimated values of Rattusrattus with respect to age were compared. A total of 40 black wild rats (20 each from youngs and adults) were collected. Blood samples were then collected from the caudal vein of anaesthetized rats.Haematological parameters such as haemoglobin (Hb), total erythrocyte count (TEC), total leucocyte count(TLC), packed cell volume (PCV), mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) and the serum biochemical parameters like protein, albumin, globulin, glucose and cholesterol were measured and each parameter was expressed as mean±SE in both young and adult rats and compared according to the age group using t-test. In haematological findings, all parameters namely RBC, Hb, MCV (p<0.001) and MCHC, PCV (p<0.01) and WBC (p<0.5) reveal significant difference between young and adult rats. But, no significant difference was observed in MCH.The biochemical parameters such as total protein, cholesterol, glucose (p< 0.001), and globulin (p<0.01) reveal significant difference where ass no significant difference was recorded in albumin between youngs and adults. The findings would be helpful to establish a baseline value and to draw conclusive remarks as to the health status of Rattusrattus.***

Keywords: *Rattusrattus*, blood, haematological parameters, serum, biochemical parameters

1. Introduction

Rats are mammals of the order Rodentia because of the presence of long incisors used for continuous gnawing.Rodents are widely distributed in all continentsexcept Antarctica. These are considered to be a suitable mammalian model organism for experiments. The order Rodentia includes a number of long-lived species namely naked mole-rats, beavers, porcupines, and some squirrels with average lifespan exceeding 20 years [1].The black rat (*Rattusrattus*) is a common longtailed rodent of the genus *Rattus* in the sub-family Murinae for which these are commonly known as murine rodents. Haematological and biochemical test are not widely applied for the diagnosis of mammalian medicine but these tests could be a suitable diagnostic tool for monitoring physiological and pathological changes in rats.Different factors such as seasons[2],stressors [3],age and sex [4],and rat species [5] affect the haematological parameters. The serum biochemical analyses provide information about internal organs, electrolytes, proteins, and nutritional and metabolic parameters [6]. One of the difficulties in assessing the state of health of natural rodent population has been the deficiency of reliable references of the normal condition. The studies of haematology provide a valuable diagnostic tool in evaluating human health. Moreover, recently attention has been given to the biochemical characterization of blood of rats as an internal index. Due to handling and hypoxic stress, fluctuations were detected in the concentration of cortisol, glucose, cholesterol, and other basic components in response [6].This is useful for the diagnosis of many diseases caused due to pathogens[7].This study reports the baseline reference values of haematological and biochemical parameters of the normal healthywild black rats.The analysis could help in understanding the better health condition of maleblackratsin natural habitat. Since information on the detailed haematological investigation of rats are inadequate, the present investigation reports on the study of haematological and biochemical profile of wild black rats.

2. Materials and Methods

For haematology, the black rats were collected by wooden and wire net trapping in evening time from the backyard of the residential complex of the Utkal University campus, Bhubaneswar, Odisha during the period July 2016 to December 2016. Next day the animals were transported to the laboratoryand the analyses were carried out. A total of 20 adult and 20 young specimens having mean sv length of young 26.16 ± 0.70cm, and weight 60.76 ± 2.34gand sv length of adult 32.65 ± 0.75cm and weight 106.38 ± 5.36were taken for the present investigation.Trappedrats were kept at room temperature and were fed with paddy, grain and biscuits. These were allowed to acclimatize to captive condition prior to experimentationand were carefully handled to minimize the stress. Then the rats were anaesthetized by using chloroform in a jar. After that, 2 ml of whole blood was drawn from the caudal vein by putting on the tray in the morning hours to avoid diurnal variation.Collected blood was transferred from the syringe into an anticoagulant, namely ethylenediaminetetraacetic acid (EDTA) containing vials, for haematological studies. For biochemical studies, some amount of blood was kept in an Eppendorf tube for clotting. After clotting, the supernatant serum was pipetted into labelled vials and were stored at 8° C until analyzed. After the collection of sample,rats were released at the place from where they were trapped after being normal.

2.1. Haematological analysis

All the haematological parameters were determined by using the standard techniques. The haematological parameters

include, haemoglobin (Hb), total erythrocyte count (TEC/RBC), total leukocyte count (TLC/WBC) and packed cell volume (PCV).Haemoglobin concentration (Hb) was measured by Sahli's acid hematin method[13]. Red blood cell count and total leucocytes count were carried out with a Haemocytometer method. Packed cell volume (PCV) was determined by means of a microhaematocrit method [14].Erythrocyte indices such as mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), and mean corpuscular haemoglobin concentration (MCHC) were calculated as per following formulae.

MCV=PCV/ Erythrocytes count ×10
MCH= Haemoglobin/ Erythrocytes count ×10
MCHC= Haemoglobin /PCV ×100

2.2. Biochemical analyses

Only limited data on the clinical biochemistry of mice are available from studies [8-14].The serum biochemical analyses provide information about internal organs, electrolytes, proteins, and, metabolic parameters[15]. Biochemical test has not been widely applied for the diagnosis of mammalian health disorder but these tests could be a suitable diagnostic tool for monitoring physiological and pathological changes inmammals. Parameters on blood biochemistry can also be used to detect the health status of the animals.The serum biochemical parameters like protein, albumin, globulin, glucose, and cholesterol were estimated using standard kit (Coral Clinical System, US Nagar, Uttarakhand, India).Haematological and Biochemical parameters were expressed as mean±SE in both young and adult rats and compared according to the age using t-test. All these statistical analyses were performed using the Microsoft Office Excel 2013.

3. Results

The present work focuses on the changes in value of blood with respect to the differentage group of the *rats* (Table 1)

Table 1: Mean ±SE Haematological parameters results of the young and adult rats(n=10 per age group)

Sl no	*Hematological parameters*	*Age of rats*	
		Young	*Adult*
1	RBC (10^6 mm^{-3})	4.39 ± 0.12***	3.65 ± 0.15***
2	WBC (10^3 mm^{-3})	4.61 ± 0.24*	3.59 ± 0.43*
3	Hb (g/dl)	10.41 ± 0.17***	8.59 ± 0.29***
4	PCV (%)	31.64 ± 0.48**	29.1 ± 0.92**
5	MCV (fl)	72.64 ± 2.48***	37.16 ± 3.21***
6	MCH (pg)	23.91 ± 0.86^{NS}	24.84 ± 1.82^{NS}
7	MCHC (%)	32.90 ± 0.31**	29.37 ± 1.08**

*Shows significance ($p<0.01$) between samples.
**Shows significance ($p<0.05$) between samples.
***Shows significance ($p<0.001$) between samples.
NS shows not significant ($p>0.05$) between samples.

The total erythrocyte count and haemoglobinshow highly significant difference ($p<0.001$) between youngs and adults. They alsoshow significant difference ($p<0.01$) in case of PCV and MCHC. WBCs show significant difference ($p<0.05$) and MCH does not show any significant difference with respect to age in rats.The biochemical parameters have been reported to be higher in youngs than adults and are significantly different (Table 2).

Table 2: Mean ±SE Biochemical parameters results of the young and adult rats (n=10 per age group)

Sl no	*Biochemical parameters*	*Age of rats*	
		Young	*Adult*
1	Total protein (g/dl)	5.14 ± 0.14***	4.04 ± 0.13***
2	Cholesterol (mg/dl)	67.22 ± 0.41***	61.13 ± 0.26***
3	Glucose (mg/dl)	160.66 ± 0.38***	175.02 ± 0.37***
4	Albumin (g/dl)	3.58 ± 0.23^{NS}	3.13 ± 0.18^{NS}
5	Globulin (g/dl)	1.40 ± 0.10**	1.11 ± 0.05**

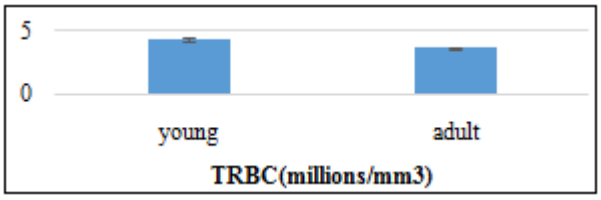

Figure 1: Comparision of RBC of young and adult rats

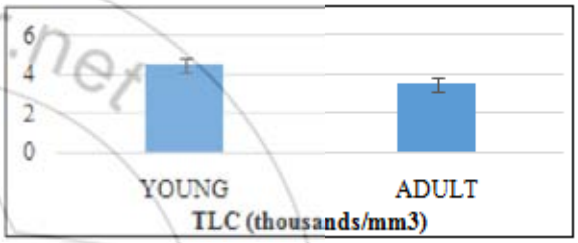

Figure 2: Comparison of WBC of young and adult rats

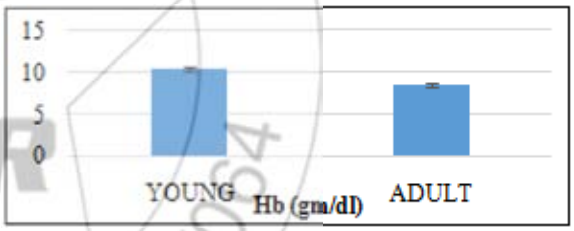

Figure 3: Comparison of Hb of young and adult of rats

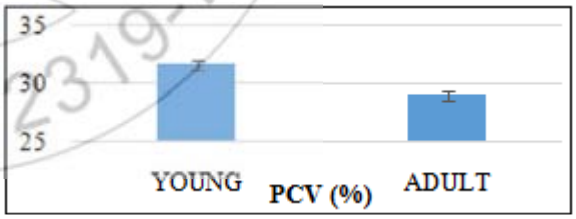

Figure 4: Comparison of PCV of young and adult rats

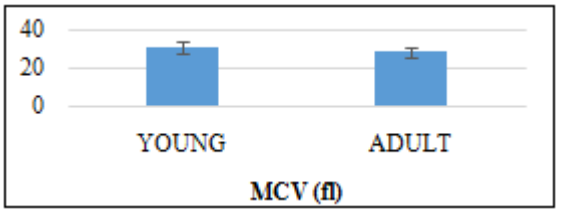

Figure 5: Comparison of MCV of young and adult rats

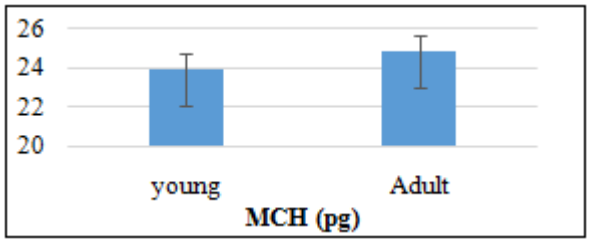

Figure 6: Comparison of MCH of young and adult rats

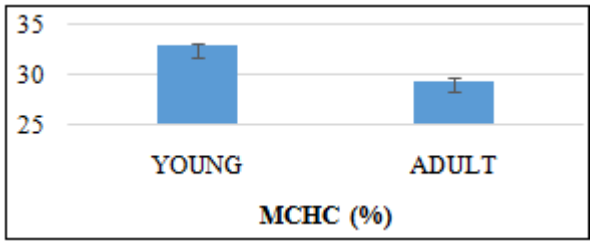

Figure 7: Comparison of MCH of young and adult rats

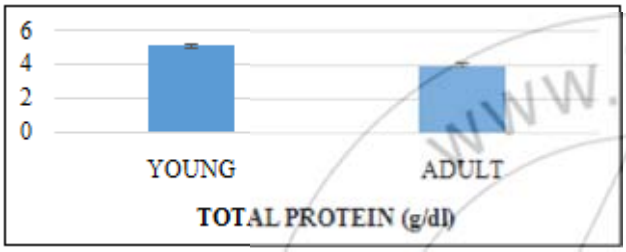

Figure 8: Comparison of total protein of young and adult rats.

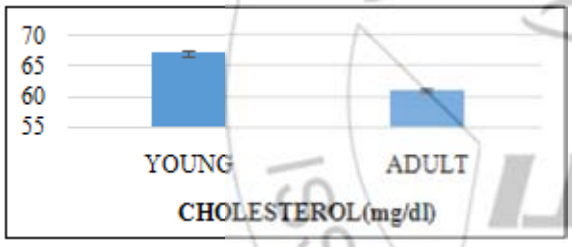

Figure 9: Comparison of cholesterol of young and adult rats

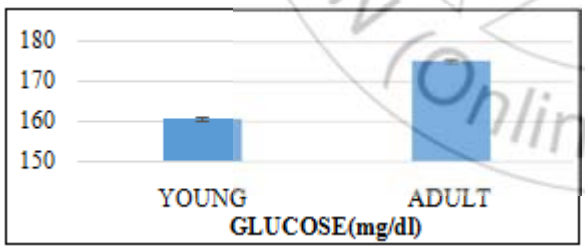

Figure 10: Comparison of glucose of young and adult rats

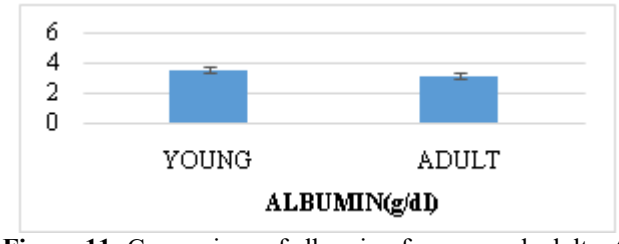

Figure 11: Comparison of albumin of young and adult rats

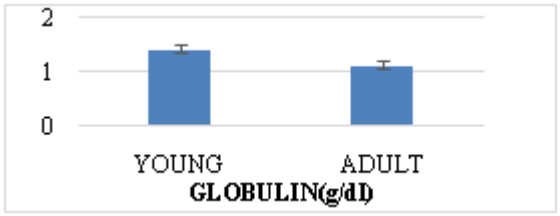

Figure 12: Comparison of globulin of young and adult rats

4. Discussion

Haematological analyses of rats are important as these are associated with the health of the animals.They adapt tosomewhat adverse conditions by changing their physiological activities. Changes in haematological parameters are often used to determine various conditions of the body and to stresses due to environmental, nutritional and/or pathological factors [16].Findings of reference value for wild black rat will help to establish and identify the causes of disease in wild black rats. Red blood cells play role in the transport of oxygen and carbon dioxide in the body [17]. The RBC count for young and adult rats is 4.39 $\pm$0.12x10 ^{6} /mm^{3} and 3.65$\pm$0.15x10^{6}/mm^{3} respectively.Present study shows significantly higher value of RBC between youngs and adults, which may be because of their agility and variation of age group.Red blood cells (erythrocytes) serve as a carrier of haemoglobin. The haemoglobin (Hb) that reacts with oxygen is carried in the blood to form oxyhaemoglobin during respiration [18,19]. The Hb for youngs is noted to be 10.41$\pm$0.17 g/dl and that of adult is 8.59$\pm$0.29 g/dl of blood respectively. Highly significant difference ($p<0.001$) in the haemoglobinis found between youngs and adults and this deviation may be due tothe variation in age group.

The major functions of the white blood cells are to fight against infections, defend the body by phagocytosis against invasion by foreign organisms, and to produce or at least transport and distribute antibodies in immune response[20].The result of WBC found in this study for youngs and adults is 4.61$\pm$0.24x10^{3}/mm^{3} 3.59$\pm$0.43x10^{3}/mm^{3}respectively. Higher WBCs count found in youngs compared to adults may be due to difference in age group or certain infection due to bacteria or parasites. The value of PCVis found to be31.64$\pm$0.48% and29.1$\pm$0.92% between youngs and adults, and this value is higher in case of youngs which is possibly because of relatively higher RBC counts in youngs. Another reason could be the difference in the age group. The value ofMCV reflects the size of red blood cells by expressing the volume occupied by a single red blood cell. The value of MCV for youngs and adults is 72.64$\pm$2.48% and 37.16$\pm$3.21% respectively which indicates a significantly higher value of MCV in youngs compared to adults. The value of MCH is reported to be higher in youngs (23.91$\pm$0.86 pg) than adults(24.84$\pm$1.82 pg) but the difference is insignificant.The higher value of MCH in young than adults indicates higher likelihood of occurrence of macrocytic anemia in youngs than adults [21]. The value of MCHC was observed to be higher in youngs than adults, which is 32.90$\pm$0.31% and 29.37$\pm$1.08% respectively.High level of MCHC in youngs indicates more Hb in a unit of RBCs [21].In the present study, all the haematological parameters reflect higher value

in youngs than adults which may be due to the difference in age group or agile activities of youngs.

The results of the biochemical profile of *Rattusrattus* are used as a tool to assess the health status of rats(Table 2). These parameters may change with respect to age, species, sexual maturity, and health status of rats.Serum protein is the protein component of the blood which is vulnerable to increase with starvation or physiological stress. In the present case, the concentration of plasma protein is higher in youngs (5.14±0.14 mg/dl) than adults(4.04±0.13 mg/dl). Plasma protein is an index of the health [22] as well as indicator of nutritional status [23]. Albumin in the blood performs the transportation of lipids [24] and helps in the general metabolism of organisms. The rise in albumin concentration in animals is due to the loss through urine or faeces or through break down resulteddue to impaired synthesis. In this study, the level of albumin is more in youngs(3.58±0.23 g/dl) than adults(3.13±0.18 g/dl) which may be due to variation in the age group.Globulin is known to be associated with innate response in organisms.Generally, glucose is continuously required as a source of energyby all cells of the body and must be maintained at adequate level in the plasma [25].The study further reveals the higher value of glucose in adults(160.66±0.38 mg/dl) than youngs (175.02±0.37 mg/dl) which is believed due to variation in the age group .The liver plays a major role in cholesterol homeostasis[26] by regulating plasma lipoprotein metabolism and lipid output in bile [27]. The cholesterol concentration in this study is found to be 67.22±0.41 mg/dl and 61.13±0.26 mg/dl between youngs and adults. Except albumin, all other biochemical parameters (Table 2) such as protein, glucose, globulin and cholesterol differ significantly ($p<0.001$) between the youngs and adults (Figs.8 to12).The value of all biochemical parameters has been found to be higher in youngs than adults except the glucose.This may be due to the difference in age group. The value ofglobulin is recorded to be higher in youngs (1.40±0.10 mg/dl) than adults(1.11±0.05mg/dl) but the difference is insignificant. Blood biochemical parameters with significant variations ($P<0.001$) were observed for the protein, glucose and cholesterol between young and adult rats.

5. Conclusion

The present findings reveal differences in blood profile of wild black rats *Rattusrattus*, due to variation in the age group. The results of the present investigation serve as a baseline value of haematological and biochemical parameters from which the condition of health of the rats can be known through significantly differential value of the haematological parameters.

6. Acknowledgments

The authors are thankful to the Head, PG Department of Zoology, Utkal University, Bhubaneswar 751 004,Odisha, India for providing facilities to carry out the investigation. First author is thankful to Department of Science and Technology (DST), Govt. of India for the financial support vide letter no DST/INSPIRE/2015/IF150460 dated 24/08/15.

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