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HEMATOLOGICAL PARAMETERS AS A TOOL TO STUDY CAPTIVE HOUSE SPARROWS (*PASSER DOMESTICUS*)

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ABSTRACT

During the era of extinction, bringing organisms to captivity is an effective method for conservation of declining populations. Keeping house sparrow in captivity was an effort to understand whether captivity affects physiology and behaviors of this species or not. In this study we focused on hematological parameters to study the physiology of *Passer domesticus*. The birds were kept in captivity in summer and winter for a couple of months and tested for hematological parameters. Platelets and MCH remained lower in captive birds, while WBCs remained higher during winter. During summer, maximum parameters remained similar to wild ones. Summer captivity was more successful than winter. Stress indicated by H/L ratio was higher in both seasons. Sparrows didn't breed in captivity. The group behavior in winter probably to keep them warm, was observed. The current is the first report for the first-time in-house sparrow.

1. INTRODUCTION

The house sparrow is an anthrodependent bird, known for its strong association with human (Ravinet *et al.,* 2018). It developed a commensal relation with human beings (Kauserud *et al.,* 2007). It is highly adaptive bird as it can be found breeding in hot places like deserts in southern California or living in cold areas of Arctic Circle. (Hanson *et al.,* 2020). Despite of its historical success in chasing human beings unfortunately, its population is declining since 1980 (Murgui and Macias 2010). It is categorized as Least concerned in IUCN list (Anonymous 2018). Due to its extensive decline in the Europe and especially in Britain where about 60% of the sparrow decline was reported in about 30 years. It was entered in the list of species of European conservation concern (Baillie *et al.,* 2010), SPEC category 3, Birdlife International 2006). After that its decline reported in many parts of world. In Pakistan the decline has been observed by ecologists, wildlife biologists and common people throughout the country but we are lacking the documented data. Only study from Pakistan in district Sargodha, Punjab the sparrow was observed less in urban areas as compared to rural areas (Mustafa *et al.,* 2015). This reveals that we may experience severe population decline in future. So, we need to study the ecology and physiology of sparrow in details to save this species in future.

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Conservation physiology is a new emerging field that uses physiological tools for conservation (Cook *et al.,* 2013). Blood biochemistry and clinical hematology is used as the physiological indicator in captive programs for conservation as well as to study animals in wild (Khan *et al.,* 2011). Animals are brought into captivity from natural environment for research or conservation purpose (Fischer and Romero 2019). The world is going through the sixth episode of extinction (Rahbek 1993) it is said that in the next decades the 50% of species will be eradicated (Ceballos *et al.,* 2017). Captivity is good for endangered species conservation but can change the behaviors and physiology of animals (Jandt *et al.,* 2015, Buehler *et al.,* 2008, Ewenson *et al.,* 2001, Kuhlman and Martin 2010). It varies from species to species, some species can adapt to the captivity environment and some cannot and undergoes physiological changes like many organisms might be using different immune responses in captive environments (Mason 2010, Morgan and Tromborg 2007). The work on Sparrow's physiology and behaviors in captivity will be valuable addition in literature. This study focused on whether the physiological changes take place during captivity. This is the first study of this kind in our knowledge. In Pakistan no such study has done before in Sparrow.

The objective of this research is to study the effect of captivity on the behavior and physiology (hematological physiology is being tested in this study) of House Sparrow.

2. MATERIALS AND METHODS

The study was approved by the ethics committee of University of Arid Agriculture Rawalpindi. All efforts were taken to minimize pain and discomfort to the animal while conducting these experiments

House Sparrow (both male and female) were captured in December 2021. These were kept in two cages (length 18.5-inch, width 12-inch, height 14.5 inch). Jugular vein was punctured for blood sampling by using a 1cc syringe. Blood was taken from 13 Sparrows in February 2022 (after 2 months of captivity). Again, sparrows were captured in April 2022 and kept in cage (length 4ft, width 2.5ft, height 5 ft.). Blood samples were taken at the end of June 2022 from 10 Sparrows (after 3 months of captivity). The blood parameters tested included RBC counts, WBCs counts, platelet counts, hemoglobin concetration, hematocrit, MCV, MCH, MCHC and differential leukocyte counts (Heterophill, lymphocytes, monocytes, eosinophils and basophils). The blood tests were performed on a hematology machine Urate Hematology Analyzer veterinary version model 3000 plus Germany. The wild group was captured in each season and released after taking blood samples. It is used to compare hematological parameters with captive birds. Independent t-test is used in this study. Value of $p < 0.05$ is significant.

3. RESULTS AND DISCUSSION

According to present study, during winter WBC counts were significantly (p= 0.00) high in captive sparrows. Heterophil were found significantly (p=0.00) higher in captive birds and lymphocytes were significantly (p= 0.00) higher in wild birds. This made heterophil to lymphocyte (H/L) ratio to be higher in captive birds. H/L ratio was significantly (0.00) higher in captive birds. Heterophil and Lymphocyte are negatively correlated with each other. Monocyte (p=0.006), eosinophil (p=0.011) and basopohil (p= 0.029) were significantly higher in captive sparrows. High WBC counts and H/L ratio indicates high level of stress in captive sparrows. Our winter captive sparrows had significantly (p= 0.018) higher platelets. Negative correlation was found between WBCs and Platelets. The wild birds showed positive correlation between WBCs and Platelets. MCH was found significantly (p= 0.026) greater in wild birds

In current study during summer season, no significant (p= 0.271) difference in WBCs was found between wild and captivity. However, H/L ratio was significantly (p= 0.00) high in captive birds. Heterophil was significantly (p= 0.00) greater in captivity and lymphocyte was significantly (p= 0.001) greater in wild. We found negative correlation between Heterophil and Lymphocyte.

During our study hematological parameters of summer captivity and winter wild (taken as control group) were compared. Then these results were compared with wild ones.

In the present research, we studied the seasonal variations of hematological parameters in captive sparrows and compared those results with wild sparrows (taken as control group). Platelets were significantly (p= 0.00) in summer in captive sparrows that was similar to wild birds (p= 0.001). In captive birds WBCs and Platelets have negative correlation during both seasons. Hematocrit value was found significantly greater in summer in both captive (p= 0.022) and wild (p= 0.001) birds. WBC counts were significantly (p= 0.00) high in winter in captive sparrows. However, WBCs in wild sparrows were significantly (p= 0.023) higher in summer season. Heterophil was significantly greater in summer in both wild (p= 0.00) and captivity (p= 0.00). Lymphocyte count was significantly high in winter in wild (p= 0.00) and captivity (0.00). This made H/L ratio to be greater in summer. Greater stress calculated in summer season in both wild and captivity. H/L ratio was significantly greater in summer in wild (p= 0.00) and captive (p= 0.00) sparrows. No significant (p= 0.123) difference in captive sparrows between RBCs of summer and winter.

During captivity House Sparrow made no association with the food provider or people around them. A dying member of the population (or flock in each cage) was not given attention and they tried to kill that member by knocking it by their beaks and by mounting on it. It was also observed that during night if some of the bird died in their roosting place, they keep siting on it. They didn't bother for any member died in their vicinity.

During winter, in low temperature they showed group behavior. They group up at night and shared warmth with each other, sit side by side in a circle. One of them enters in the center of the circle then after some time it comes off and the other member enters in the center. This behavior may have evolved to keep them warm and maintain heat in the winter. The same kind of behavior was documented in a BBC documentary for penguins to survive in very low temperature. Molting was observed throughout the winter season. During winter captivity high mortality rate (50%) was recorded. They seemed to be very relaxed and apparently no signs of stress were observed. Higher level of stress was found by our hematological results.

During the captivity it was noticed that they prefer to live on height so may be due to behavioral problem they can't survive in that small cage (the small cage was used to maintain temperature and keep them warm). They roost on the basket hanged on one side of cage wall at night.

During summer captivity the birds readily acclimatized to the cage environment, they start using available nesting sites. They were provided with straws, grass and stuff for making nests in the provided nesting sites (nest boxes). They used very little of that but no observations were recorded. No signs of courtship behavior or mating was observed. They didn't breed during three-month captivity period.

Mortality rate was low in summer as compared to winter. In the first day of captivity (that is very important for captive birds) 4 birds died (1 female and 3 male). Three birds (2 males and 1 female) died in 2 months. Some birds especially male House Sparrows had problem in flying and they were sitting on the cage floor for the whole day even during night. This caused their death. Some of them recovered from illness by giving antibiotics but 3 of them died. In the last month of captivity 2 male and 1 female House Sparrow died. Out of these 3 birds 2 were died of unknown reason found dead in their nest box.

In the current study during winter high WBC counts and H/L ratio indicates high level of stress in captive birds. Kilgas *et al.,* (2006) studied Great tit's (a passerine bird) local survival. They reported birds having less H/L ratio had greater survival. Thus, in our study to ensure maximum survival in H/L ratio should be reduced in captivity. During winter molt House sparrows decreased their WBCs. It is a tradeoff between molting and immunity (Nava *et al.,* 2001). Captivity caused stress as indicated by H/L ratio. Stressful environment disturbs molting causing malformation in feathers. According to literature male birds with malformation in feather are less likely to be selected by female for mating (Wood 1950). It decreases male sparrows' fitness in captivity. Instead of WBCs other parameters remained similar in our summer captivity that indicates good sign towards the success of captivity. Summer captivity was more successful than winter as indicated by results of hematological parameters. Our captive sparrows had

higher platelets. Negative correlation was found between WBCs and Platelets. Platelets play important role in homeostasis and immune responses (Weyrich *et al.,* 2003). No difference in WBCs wild and captive sparrows during summer season is a good sign towards stability and normal life in captivity. However, H/L ratio was greater in summer captive sparrows that still indicates stress in captive sparrows. Comparing results of captivity to wild (control) sparrows during summer season gave similar values for haematological parameters. The results indicate that summer captivity was more successful than winter.

Greater value of heterophil to lymphocyte ratio can be the possible reason for death in captivity. The House Sparrow were found very sensitive towards low temperature during winter. Low temperature was also a big reason for the death. Factor of temperature during winter season in House Sparrow population decline is very important. It is decreasing its survival. As it is a common observation that the house sparrow is no more making nests in our homes due to modern infrastructure, and changed life style, we don't want to share our living space. For instance, we will not like to have a house sparrow nest in our living room or kitchen. when I was collecting house sparrow, they were found only in the old houses made of mud (in villages) recently used by some rural people for keeping their livestock. But that type of nesting sites is vanishing now because we rarely found such mud houses. House sparrow prefers to nest in cavities in human settlements and consider trees and shrubs as the alternative nest sites if they don't find place to nest. The open places on tress nests are dangerous for the sparrows and they are more vulnerable to predation and nest destruction due to any calamity (Summer Smith). During summer season mortality rate was greater in male. It may suggest less investment of immunity in males as studied by Moller *et al.,* (1996) that males invest more in sexual characters than immunity. They founded small bursa of fabricius in high male having sexual traits. But our WBCs results didn't support this argument.

While studying seasonal variations in captive House Sparrows and comparing with the wild sparrow (taken as control group) no significant difference in maximum parameters were found. This was a good sign as the blood parameters are shaped by ecological factors and life history traits (Soulsbury *et al.,* 2021). This indicates that birds responded to seasonal variations during captivity. They were doing same as in wild natural habitat. Hematocrit value was found greater in summer in both captive and wild sparrows. The blood plasma may decrease during summer season that increases hematocrit value in summer.

Khan *et al.,* (2011) studied differential leukocyte counts in blue rock pigeon *Columba livia* they concluded that the risk of infections in captive pigeons decreased. According to their results of WBCs were significantly lower in captivity. This makes Khan *et al.,* (2011) contradictory to present study findings. In our study House Sparrows' WBCs values were higher in captivity during winter. During summer WBCs remained same in wild and captivity. Elevated levels of WBCs in captivity may be due to infection or high stress level indicated by H/L ratio.

Based on our findings no difference in captive sparrows between RBCs of summer and winter. In wild RBCs were greater in summer. The RBCs and HCT have positive correlation. It shows low blood plasma volume due to shortage of water. During captivity RBCs remained same in winter and summer. Equal number of RBCs shows plasma level remained same. Hematocrit in summer and winter captivity have positive correlation. While in wild it has negative correlation. Captivity may possibly reduce water shortage problem. Hematocrit and RBC counts are plastic traits. Their values indicate normal oxygen transportation in body (Polo *et al.,* 1992). But hematocrit remained higher in summer in captive birds. This suggests decrease of plasma level in summer as compared to winter in captivity. This may indicate that plasma level in summer captive sparrows is greater than wild birds. Some scientists consider Hemoglobin (Hb) concentration to be the best parameter to know health of birds (Minias 2015, Milenkaya *et al.,* 2013). Taking in view this argument, it can be predicted that captive birds are in good health. As there is no significant difference in Hb in wild and captive birds.

During the three months' summer captivity period House Sparrow didn't breed. In spite of House Sparrows' strong association with humans it is not clear that why this bird is very difficult to be bred in

captivity (Lombardo and Thorpe 2009). According to Love *et al.,* (2017) captive House sparrows have elevated levels of cortisone that indicates stress that have long lasting effects. The birds cannot acclimatize to the captivity even after 9 weeks or more. The human presence around them causes the birds natural response of flushing (in case of House sparrow 2 to 3 steps distance from bird causes flee of bird). In captivity they don't learn to decrease this response. They didn't consider captivity as safe and remain alert, as in state of emergency prepared them for any kind of unusual situation. Stress along with flush response in the captivity might have kept them continuously stressed. However, apparently the birds seemed to be very relaxed in captivity. This causes the energy investment to immediate survival, then in reproduction and breeding. As the breeding and reproduction are very energy demanding activities (Angelier *et al.,* 2009). Lombardo and Thorpe 2009 studied and concluded that higher corticosterone levels caused a decrease in testosterone and sperm production. They linked this testosterone decline with color of beak. They reported change of beak color from black to horn-colored. Same kind of color change was observed in captive male sparrows in present study. The clutch size determination is a phenotypic plastic trait in House sparrow. It allows the female to lay number of eggs depending upon the availability of food recourses (Perrins and Moss 1975). The birds require proper diet especially insect's larvae to feed their young ones. The absence of the proper food required for hatchlings especially insects' availability in the captivity may be a reason for not breeding or laying eggs during captivity. First reason is more valid because the food availability effects clutch size after mating takes place. Courtship behavior or mating during experiment was not observed or it might be possible that they mated or performed courtship behavior in our absence.

4. CONCLUSION

The study concludes that captivity of house sparrows in summer months is more successful than the winter. However, the animal did not breed in captivity. Additionally, group behavior was more evident in winter to keep the body warm.

5. CONFLICT OF INTEREST

All authors have declared that there is no conflict of interest regarding the publication of this article.

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Table 1. Table showing hematological parameters of House sparrows in Winter captivity. LT

S. No	Hematological Parameters	p- value	Control Sparrows	Captive Sparrows
1	WBC counts/ uL	0.00	41493.3±9712	68307.6±12052
2	RBC counts M/uL	0.469	2.95±0.28	3.07±0.55
3	Platelets count /uL	0.018	44733.3±8697	35230.7±11263
4	Hemoglobin g/dL	0.465	14.8±1.59	14.32±2.17
5	Hematocrit %	0.567	36.76±4.64	37.7±3.98
6	MCV (fL)	0.420	124.7±5.37	123±5.5
7	MCH (pg)	0.026	51.3±3.25	48.6±2.62
8	MCHC g/dL	0.027	45.84±2.85	48.6±3.41
9	Heterophil %	0.00	7.53±1.95	12.8±2.4
10	Lymphocyte %	0.00	87.3±2.52	79.8±2.15
11	Monocyte %	0.006	2.53±0.63	3.4±0.87
12	Eosinophil %	0.011	2.06±0.59	2.9±0.95
13	Basophil %	0.029	0.46±0.51	0.92±0.49
14	H/L ratio	0.00	0.079±0.03	0.161±0.03

Table 2. Table showing hematological parameters of House sparrows in Winter captivity. LT

S. No	Hematological Parameters	p- value	Control Sparrows	Captive Sparrows
1	WBC counts/ uL	0.271	52087.5±12948	46840±7427
2	RBC counts M/uL	0.737	3.37±0.40	3.38±0.28
3	Platelets count /uL	0.659	53666±13838	55900±8211
4	Hemoglobin g/dL	0.263	14.32±2.26	15.21±0.95
5	Hematocrit %	0.199	43.9±5	41.48±3.08
6	MCV (fL)	0.573	112.3±8.15	115.08±13.02
7	MCH (pg)	0.114	43±3.43	45.7±4.01
8	MCHC g/dL	0.099	47.8±3.48	49.9±2.92
9	Heterophil %	0.00	11.75±3.10	17.7±2.2
10	Lymphocyte %	0.001	79±2.9	73.8±3.11
11	Monocyte %	0.702	4±0.95	3.9±0.87
12	Eosinophil %	0.335	4.5±1.08	4±1.05
13	Basophil %	0.907	0.75±0.62	0.7±0.48
14	H/L ratio	0.00	0.149±0.045	0.24±0.03

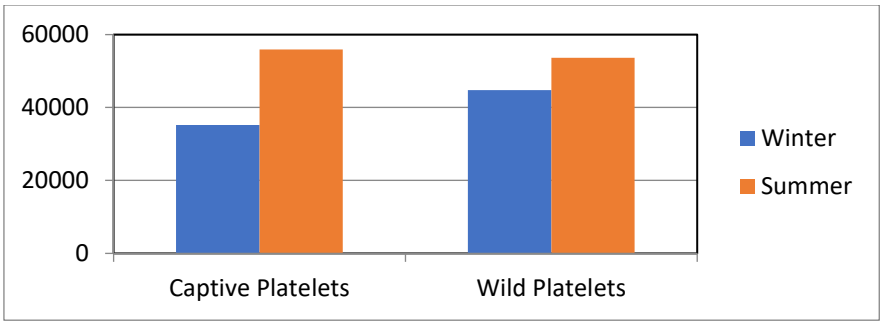

Figure 1. Graphical presentation of seasonal variations of Platelets counts in captive sparrows comparing with control.

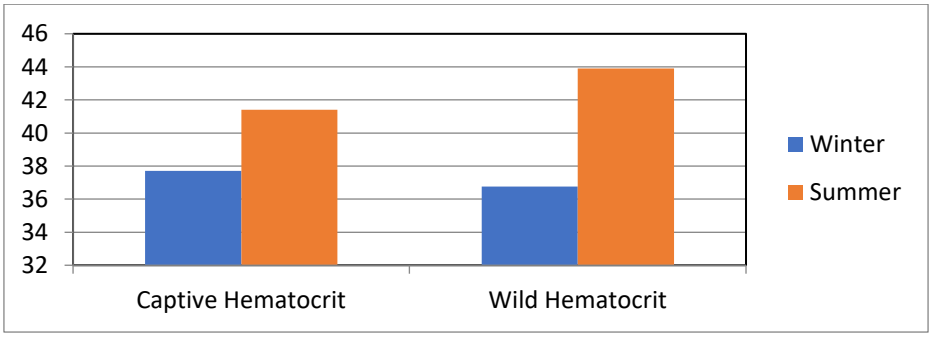

Figure 2. Graphical presentation of seasonal variations of hematocrit in captive sparrows comparing with control.

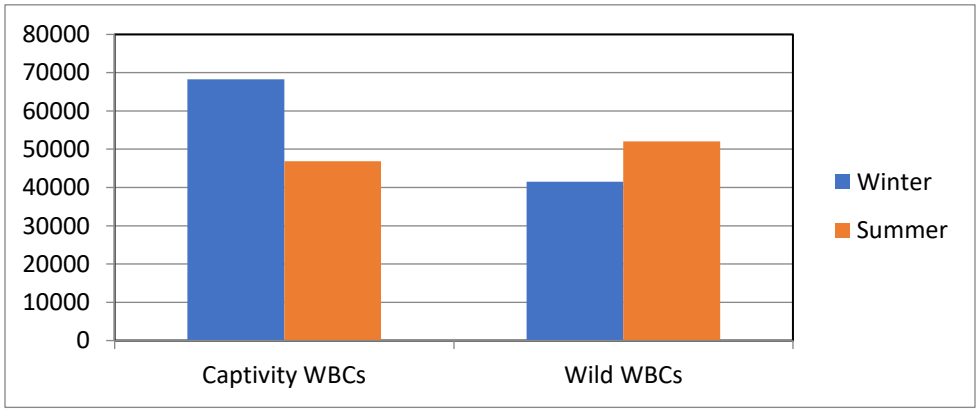

Figure 3. Graphical presentation of seasonal variations of White blood cells (WBCs) in captive sparrows comparing with control

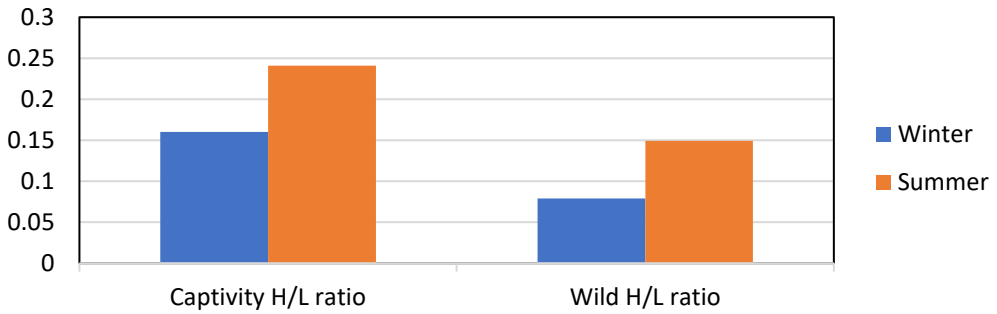

Figure4. Graphical presentation of seasonal variations of Heterophil to Lymphocyte (H/L) ratio in captive sparrows comparing with control.