Effect of *Haemoproteus columbae* infection on the hemogram of the Pigeons (*Columba livia domestica*)

Amir Dehghani Samani¹ · Khodadad Pirali Kheirabadi² · Abdonnaser Mohebbi¹

Received: 29 October 2013 / Accepted: 19 June 2015 / Published online: 28 July 2015
© Indian Society for Parasitology 2015

Abstract It has been reported that parasites affect the health and productivity of birds, initiate excessive preening which interrupts feeding, as the birds spend much time preening rather than being involved in other essential life activities. *Haemoproteus columbae* occurs in pigeons widely in tropical and subtropical regions. The present investigation was concentrated on the effect of *H. columbae* infection on the hemogram and changes in the number of white blood cells in infected pigeons. Blood samples taken from 100 pigeons in Shahrekord. Blood smears stained and observed for *H. columbae* infection. Packed cell volume (PCV) and total white blood cells (WBC) count and differential WBC count determinate for every smears. This study show that PCV increased in infected pigeons by *H. columbae* than non infected pigeons. There were increasing in the number of total WBC in the infected pigeons by *H. columbae* than non infected pigeons. Also there were significant differences between the number of lymphocytes and eosinophils in the infected pigeons by *H. columbae* than non infected pigeons. This study and same studies show the part of the effects of blood parasites on the hemogram, but there were many questions about the effects of blood parasites such as *H. columbae* on the other parameters of host's blood, so we advise to focus on the other hematological parameters in the future studies.

Keywords *Haemoproteus columbae* · Hemogram · Pigeons · White blood cell

Introduction

In recent years, pigeons and doves has colonized suburban and urban areas of world. It's interaction with man and other domestic and wild birds, portends it as a potential carrier of zoonotic parasites. Parasites are regarded as the basic causes of retardation in growth, lowered vitality and poor conditions of the birds. They can affect bird health directly by causing irritation, discomfort, tissue damage, blood loss, toxicosis, allergies and dermatitis which in turn reduce the quality and quantity of meat and egg production (Ruff 1999).

Haemoproteus species are commonly occurring avian haemosporidian parasites (Bennett 1987). *Haemoproteus columbae* occurs in pigeons widely in tropical and subtropical regions (Springer 1972). The natural hosts of this parasite include domestic pigeons (*Columba livia domestica*), many species of wild pigeons, mourning doves (*Zenaidura macroura*), turtle doves and other wild bird species (Soulsby 1986). The gamonts of *H. columbae* develop from tiny forms to elongated, crescent-shaped forms, which partially encircle the nucleus of the host cell. The host cell's nucleus may be displaced but not to the edge of cell. The mature gamonts of *H. columbae* occupy the host cell completely. They distort it and push the host cell's nucleus to one side. The vector of *H. columbae* is a hippoboscids fly that named *Pseudolynchia canariensis*. Both *H. columbae* and *P. canariensis* are widely distributed in the world, particularly in warm and temperate climates (Soulsby 1982).

It has been reported that parasites affect the health and productivity of birds, initiate excessive preening which

✉ Khodadad Pirali Kheirabadi
pirali-k@vet.sku.ac.ir; khpirali@yahoo.com

¹ Department of Clinical Sciences, Faculty of Veterinary Medicine, Shahrekord University, 115 Shahrekord, Iran

² Department of Pathobiology, Faculty of Veterinary Medicine, Shahrekord University, 115 Shahrekord, Iran

interrupts feeding, as the birds spend much time preening rather than being involved in other essential life activities (Clayton et al. 1999). But there were few studies about the hematological effects of blood parasites such as *H. columbae*.

The present investigation was concentrated on the effect of *H. columbae* infection on the hemogram and changes in the number of white blood cells in infected pigeons. The aim of this study was to presentation the effect of *H. columbae* on the hemogram of pigeons as the model for the blood parasite in humans and other animals.

Materials and methods

Birds for experiment

100 pigeons, aged between 6 and 18 months, served as the subjects of the experiment. They were purchased from Shahre Kord (latitude, 32°19'32"N and longitude, 50°51'52"E), in southwest of Iran.

Blood sampling

Blood sampling was through wing venepuncture, using 23 gauge sterile hypodermic needles and syringes. About two milliliters of blood were collected on each pigeon. Bloods were collected into labeled Bijou bottles, containing ethylene diamine tetra acetic acid (EDTA) at 2 mg for each ml blood as anticoagulant.

Packed cell volume (PCV)

The PCV values were determined by the microhaematocrit method of Benjamin (1985). Blood samples containing EDTA were aspirated into a set of plain capillary tubes. The tubes were sealed and spun on Hettich centrifuge (Hawksley and Sons Limited, England) at 5000 rpm for 5 min. After spinning, the PCV was read as a percentage directly from Graphic Reader (Hawksley and Sons Limited, England).

Preparing the blood smears

A small amount of blood (~50 µl) used for preparing the smears, smears dried by heating and then fixed in absolute methanol for 5 min and finally stained with giemsa stain 10 % for 15 min.

Diagnose of *Haemoproteus columbae* infection in blood smears

Stained blood smears observed for *H. columbae* carefully. 24 pigeons infected by *H. columbae*. Gamonts of *H.*

columbae developed from tiny forms to elongated, crescent-shaped forms, which partially encircled the nucleus of the erythrocytes.

Total WBC count

The total number of WBC per each µl blood of pigeons determinate via crossing the total number of counted WBC in 10 microscopic fields (by lens 40×) of every stained blood smears to 200 (Fudge 2000).

Differential WBC count

The differential number of every groups of WBC per each µl blood of pigeons determinate via crossing the total WBC count of every stained blood smears to differential percentages of each groups of WBC for these smear. For determination of differential percentages of each groups of WBC, firstly the number of each groups of WBC differed in 200 counted WBC in many different microscopic fields (by lens 100×) of every pigeons blood smears with emersion oil, then differential percentages of each groups of WBC calculated via division of counted number of each groups to 2 (Fudge 2000).

Statistical analysis

The data were expressed as the Mean ± SEM. Groups were compared using one-way ANOVA for repeated measurements. Tukey test was used for post hoc analysis. A value of ($P < 0.05$) was considered significant.

Results

Although the blood parasites damaged the erythrocytes and reduce PCV due to these damages, but PCV in infected pigeons by *H. columbae* increased than the non infected pigeons because of the increasing in the total number of WBC in the infected pigeons blood. PCV determinate 46.07 ± 2.04 % in infected pigeons and 44.38 ± 3.21 % in non infected pigeons. Although the PCV increased in infected pigeons than non infected pigeons but there were no significant difference ($P < 0.05$) between the PCV in the infected and non infected pigeons. Figure 1 show the PCV in both groups.

Total number of WBC determinated 30283.33 ± 102.84 WBC/µl blood of infected pigeons and 27564.54 ± 89.33 WBC/µl blood of non infected pigeons. There were significant difference ($P < 0.05$) between the number of total WBC in the infected and non infected pigeons. Figure 2 show the number of total WBC in both groups.

Fig. 1 PCV in infected and non infected pigeons. There were no significant difference ($P < 0.05$) between the PCV in the infected and non infected pigeons

Fig. 2 Total WBC/μl blood of infected and non infected pigeons. There were significant difference ($P < 0.05$) between the Total WBC in the infected and non infected pigeons

Number of Heterophils determinate 11443.94 ± 23.24 WBC/μl blood of infected pigeons and 10868.48 ± 41.24 WBC/μl blood of non infected pigeons, Although the increasing in the number of heterophils in infected pigeons than non infected but there were no significant difference ($P < 0.05$) between the number of heterophils in the both groups. Number of lymphocytes determinate 12884.35 ± 60.04 WBC/μl blood of infected pigeons and 16094.61 ± 24.68 WBC/μl blood of non infected pigeons, there were significant ($P < 0.05$) reducing in the number of lymphocytes in infected pigeons than non infected pigeons. Number of monocytes determinate 2446.86 ± 21.02 WBC/μl blood of infected pigeons and 2808.78 ± 12.07 WBC/μl blood of non infected pigeons, Although the increasing in the number of monocytes in infected pigeons than non infected but there were no significant difference ($P < 0.05$) between the number of monocytes in the both groups. Number of eosinophils determinate

1474.78 ± 10.54 WBC/μl blood of infected pigeons and 449.29 ± 12.91 WBC/μl blood of non infected pigeons, there were significant ($P < 0.05$) increasing in the number of eosinophils in infected pigeons than non infected pigeons. Number of basophils determinate 351.28 ± 9.09 WBC/μl blood of infected pigeons and 228.78 ± 8.16 WBC/μl blood of non infected pigeons, Although the increasing in the number of basophils in infected pigeons than non infected but there were no significant difference ($P < 0.05$) between the number of basophils in the both groups. Table 1 and Fig. 3 show the data about differential WBC count in both groups.

Discussion

In Queensland (Adlard et al. 2004), Colombia (Rodriguez and Matta 2001), Bulgaria (Shurulikov and Golemsky 2002) and the United States (Garvin et al. 1993), the prevalence rate for *Hemoterosus spp.* ranged from 20 to 32 %. The prevalence of blood parasite in pigeons and birds in Japan (Murata 2002), Costa Rica (Valkiunas et al. 2004) and Alaska (Deviche et al. 2001) was shown to be lower than 10 %. *H. columbae* infects pigeons which are associated with human settlements. Throughout the world prevalence of *H. columbae* in feral pigeons in different geographical area varies from 14 to 100 % (Paperna 2002). Yunus and Arsalan reported 74 % (73/98) of pigeons collected from a local zoo were infected with blood parasites (Yunus and Arsalan, 2001). Moreover, 105 *Columba livia* in Galapagos Island were investigated and 89 % were infected with *Hemoterosus spp.* (Luis et al. 2004). Dranzoa examined 34 pigeons and the survey of ectoparasites revealed that *P. canariensis* was the most prevalent parasite (100 %) (Dranzoa et al. 1999).

Many studies had been reported that the blood parameters most commonly associated with health of individuals and serological and hematological values could be an important source of information and could provide or

Table 1 Differential WBC count in both groups

	Infected pigeons	Non infected pigeons
Heterophils	$11443.94 \pm 23.24a$	$10868.48 \pm 41.24a$
Lymphocytes	$12884.35 \pm 60.04a$	$16094.61 \pm 24.68b$
Monocytes	$2446.86 \pm 21.02a$	$2808.78 \pm 12.07a$
Eosinophils	$1474.78 \pm 10.54a$	$449.29 \pm 12.91b$
Basophils	$351.28 \pm 9.09a$	$228.78 \pm 8.16b$

Different lower cases (a–b) show the significant difference between columns of every rows. Values of ($P < 0.05$) were considered significant

Fig. 3 Differential WBC count in both groups. There were no significant difference ($P < 0.05$) between the number of heterophils, monocytes and basophils in the both groups and there were significant difference ($P < 0.05$) between the number of lymphocytes and eosinophils

support an objective assessment of the health status especially the values of PCV, WBC. and Hb concentration (Toro et al. 1999; Schulz et al. 2000; Norte et al. 2008; Lashev et al. 2009).

Saeed et al. (2009) reported significant decrease in the values of PCV and Hb concentration in infected chicken with cestodes compared with non infected. Ullah et al. (2005), Al-Saffar and Al-Mawla (2008) and Saeed et al. (2009) reported that the heavy infection with parasites (lice, cestodes and other parasites) can lead to blood loss anemia in birds, PCV is quickest and most practical method for evaluation the red cell mass of birds.

The mean number of total WBC was increased with increasing of intensity of infection. These results reflect that infection with these parasites appeared to be associated with hematological changes and this may reflect immune response in the birds (Muhsin 2008).

Leucocytosis and heterophilia had been reported in parasitic diseases in birds (Al-Saffar and Al-Mawla 2008; Muhsin 2008). Eosinophilia was predominant and very characteristic feature seen in birds infected with parasite specially moderate and heavy infection. Al-Saffar and Al-Mawla (2008) had reported that leucocytosis refer to an absolute increase in total number of while blood cells in circulation caused by inflammation, resulting from parasitic infections the present study also show the same results.

This study show that PCV increased in infected pigeons by *H. columbae* than non infected pigeons but there were no significant difference ($P < 0.05$) between the PCV in the infected and non infected pigeons. Results obtained from this study show that there were significant difference ($P < 0.05$) between the number of total WBC in the infected pigeons by *H. columbae* than non infected pigeons. Also this study show that there were no significant

difference ($P < 0.05$) between the differential number of heterophils, monocytes and basophils in the infected pigeons by *H. columbae* and non infected pigeons and there were significant difference ($P < 0.05$) between the number of lymphocytes and eosinophils in the infected pigeons by *H. columbae* than non infected pigeons. There were many questions about the effects of blood parasites such as *H. columbae* on the hemogram and other parameters of infected groups. So we should focus on the other hematological parameters in the next studies.

References

- Adlard RD, Peirce MA, Lederer R (2004) Blood parasites of birds from southeast Queensland. *Ornithology* 104:191–196
- Al-Saffar TM, Al-Mawla ED (2008) Some hematological changes in chickens infected with ectoparasites in Mosul. *Iraqi J Vet Sci* 22(2):95–100
- Benjamin MM (1985) Outline of veterinary pathology, 3rd edn. Kalyani Publishers, New Delhi
- Bennett GF (1987) In: Burr EW (ed) Companion bird medicine, Iowa State University Press, Ames, pp 120–128
- Clayton DH, Lee PLM, Tompkins DM et al (1999) Reciprocal natural selection on host-parasite phenotypes. *Am Nat* 154:261–270
- Deviche P, Greiner EC, Manteca X (2001) Interspecific variability of prevalence in blood parasites of adult passerine birds during the breeding seasons in Alaska. *J Wildl Dis* 37:28–35
- Dranzoa C, Ocaido M, Katete P (1999) The ecto- gastro-intestinal and haemo-parasites of live pigeons (*Columba livia*) in Kampala, Uganda. *Avian Pathol* 28:119–124
- Fudge AM (2000) Laboratory medicine avian and exotic pets. W.B. Saunders, Philadelphia, pp 9–15
- Garvin MC, Remsen JV, Bishop F (1993) Hematozoa from passeriform birds in Louisiana. *J Parasitol* 79:318–321
- Lashev L, Hubenov H, Nikolov Y, Laheva V, Mihailov R (2009) Comparison of some hematological parameters between three bird species from the Columbidae family. *Vet Arch* 79(4):409–414
- Luis RP, Alarcon DS, Merkel J, Miller RE, Parker PG (2004) Survey for *Haemoproteus* spp., *Trichomonas galinae*, *Chlamydophila psittaci* and *Salmonella* spp. in Galapagos Islands Columbiformes. *J Zoo Wildl Med* 35:60–64
- Muhsin SJ (2008) Epidemiological and parasitological study of *Ascaridia galli* in chickens in Holy Najaf governorate. Ph.D. thesis, College of Education for girls, University of Kufa, pp 109–127
- Murata K (2002) Prevalence of blood parasites in Japanese wild birds. *J Vet Med Sci* 64:785–790
- Norte AC, Sheldon B, Sousa JP, Ramos JA (2008) Repeatability and method dependent variation of blood parameters in wild caught Great Tits *Parus major*. *Acta Ornithol* 43(1):66–75
- Paperna I (2002) *Haemoproteus Cloumbae* infection of feral pigeons in Singapore and Israel. *Raffles B Zool* 50:281–286
- Rodriguez OA, Matta NE (2001) Blood parasites of birds from eastern plains of Colombia. *Mem Inst Oswaldo Cruz* 96:1173–1176
- Ruff MD (1999) Important parasites in poultry production systems. *Vet Parasitol* 84:337–347
- Saeed AE, Abdelkarim MEI, Ahmed BM, Ibrahim KE, Hafiz ISA, Suliman MI, Mohammed OSA (2009) Anticestodal activity and toxicity of some paziquantal analogues. *J. Cell Anim Biol* 9:165–170

- Schulz JH, Bermudez AJ, Tomlinson JL, Firman JD, He Z (2000) Blood plasma chemistries from wild mourning doves held in captivity. *J Wildl Dis* 36(3):541–545
- Shurulikov P, Golemsky V (2002) Haemoproteids (Haemosporidia: Haemoportefidae) of wild birds in Bulgaria. *Acta Protozool* 41:359–374
- Soulsby EL (1982) Helminths, arthropods and protozoa of domesticated animals, 7th edn. Bailliere Tindall, London, pp 56–80
- Soulsby EL (1986) Genus haemoproteus kruse in helminths, arthropods and protozoa of domesticated animals, 7th edn. Bailliere Tindall, London, pp 700–706
- Springer WT (1972) Other blood and tissue protozoa. In: Hofstad MS (ed) Diseases of poultry, 6th edn. Iowa State University Press, Ames, pp 727–740
- Toro H, Saucedo C, Borie C, Gough RE, Alcaino H (1999) Health status of free living pigeon in the city of Santiago. *Avian Pathol* 28:619–623
- Ullah A, Khan MS, Khan JA, Perrez K, Avais M, Maqbool A (2005) Effect of dilution and route of ivermectin on lice infested domestic pigeons. *Int J Agric Biol* 7(5):855–856
- Valkiunas G, Iezhova TA, Brooks DR (2004) Additional observations on blood parasites in Costa Rica. *J Wildl Dis* 40:555–561
- Yunus G, Arsalan M (2001) Blood parasites of wild pigeons in Ankara District. *Turk J Vet Anim Sci* 25:169–172