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HAEMOPROTEUS COLUMBAE INFECTION OF FERAL PIGEONS IN SINGAPORE AND ISRAEL

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ABSTRACT. – Infection characteristics of *Haemoproteus columbae* in feral pigeons (*Columba livia*) in Singapore are compared with those in Israeli pigeons. Parasitaemia and recruitment rates of gametocytes were higher in Singapore pigeons than in those from Israel. Infection prevalence and levels of parasitaemia and recruitment did not vary among months in tropical Singapore but they did in Mediterranean Israel, with January having the lowest levels. In March and May samples from Singapore and in October samples from Israel pigeons with lower body mass had significantly higher parasitaemia. This trend could not be statistically validated in the remaining months' samples. Individual variation in body mass is suggested to be correlated with dominance position which modulates access to food, rather than with age. Parasite recruitment did not correlate significantly with body mass either in Singapore or in Israel. The prevalence of louse flies (*Pseudolynchia canariensis*) and abundance on individual pigeons was about the same in Singapore and Israeli samples.

KEY WORDS. – *Haemoproteus columbae*, *Columbia livia*, Singapore, Israel, seasonality, prevalence, parasitaemia, recruitment.

INTRODUCTION

Feral pigeons (*Columba livia* Gmel) accompany human settlements worldwide. Several species of parasites occur in pigeons throughout their global range, in among these are the hemosporidid, *Haemoproteus columbae* Kruse and its vector the pupiparan louse fly *Pseudolynchia canariensis* (Marquart) (Zwart, 1986; Bennet & Peirce, 1990). There is limited quantitative information on the geographic variation in hematozoan infections (see Merila et al., 1995; Sol et al., 2000).

Host-parasite relationships were suggested to differ in tropical from non-tropical areas (Moller, 1998). We compared the characteristics of *H. columbae* infection in feral pigeon populations of semi-urban locations in a relatively non-seasonal tropical climate of Singapore and in a seasonal Mediterranean climate in Israel. In tropical India, a year-round abundance of parasitaemia in feral pigeons has been reported (Mandal, 1991). Vector abundance was claimed to be the major factor influencing the spatial variation in prevalence of *H. columbae* in pigeons (Sol et al., 2000). In higher latitudes, louse flies were reported to disappear in winter (Klei & De Giusti, 1975). Temperature requirements of the vector, the louse fly, may be critical in this context. In temperate zones the fly activity is interrupted as temperatures decline to 13°C (Klei & De Giusti, 1975). A winter decline in parasitaemia might be expected in feral pigeons from northern latitudes; consequently to the decline in the vector abundance, however, this could not be confirmed from the available literature.

Specifically, in both areas, we attempted to answer the following questions 1) what is the prevalence, the intensity (parasitaemia) and recruitment of the *H. columbae* infections, 2) does the prevalence, parasitaemia and recruitment of *H. columbae* vary among months, 3) does parasitaemia and recruitment of *H. columbae* infections vary in relation to host body mass, and 4) does the abundance of the vector fly affect parasitaemia and recruitment.

MATERIALS AND METHODS

The study sites were in semi-urban areas in Singapore, and comparable locations in Israel. In Singapore (103°50'E, 1°20'N), 47 pigeons were caught between 1 August and 17 September 1996 and 79 between 12 March and 30 June 1998. Pigeons were caught using alpha chlorhalose baits (Quek et al., 1999). In Israel, 97 pigeons were caught by nets between

Table 1. *Haemoproteus columbae* infection characteristics and mean body masses of feral Pigeons from Singapore. ± indicates standard error.

	August	September	March	April	May	June
No. of pigeons sampled	26	21	17	28	18	16
% infected	100	95	100	93	100	94
Mean parasitaemia	13.5 ±7.72	11.7 ±5.85	8.0 ±2.76	20.4 ±9.44	28.5 ±10.08	29.1 ±10.35
Mean parasite recruitment	2.47 ±0.07	2.57 ±0.07	2.53 ±0.06	2.73 ±0.08	2.39 ±0.09	2.31 ±0.10
Mean body mass (g)	246.73 ±6.78	257.19 ±6.96	252.65 ±6.46	273.21 ±7.67	239.08 ±9.22	231.94 ±10.23

15 November 1998 and 31 October 1999 from inside of roof ledges in suburban locations south of Tel Aviv (34°50'E, 32°00'N). In Singapore all pigeons were fully fledged. In Israel, the sample included 6 squabs.

Singapore has a tropical climate with high temperature and humidity all year round. The daily temperatures range from minimum 23-26 to maximum 31-34°C with average annual rainfall of 2375 mm (*Meteorological Service - Singapore, P.O.B. 8, Changi Airport, Singapore 9181*). The Tel Aviv area has a coastal Mediterranean climate, with minimum-maximum mean monthly temperatures ranging from 7 to 17°C during December-February and from 20 to 31°C during June-August, with a rainfall of about 600 mm during the cold season (*Meteorological Service - Israel P.O.B. 25, Beit Dagan 50250*).

Louse flies were collected by placing each bird in a bag containing chloroform fumes with only its head out of the bag. Anesthetised louse flies were removed by feather ruffling. The birds were then weighed (in the field by using 500 g Pesola™ precision scale) and a small amount of blood was collected into heparin capillary by puncturing the brachial vein. Thin blood smears were made and air-dried, fixed with absolute methanol and stained in 10% Giemsa in pH 7.4 buffered phosphate (for 1 hour).

Blood smears were differentially counted for juvenile (non-differentiable trophozoites) and mature stages (sexually differentiatiable halteridia) of *H. columbae*. Level of parasitaemia was reported as the number of parasites (mature and juvenile stages) per 1000 erythrocytes (p/1000), and was established following counts made from 50 microscopic fields (of approximately 20.000 erythrocytes). Mean parasitaemia was calculated from all sampled birds (e.g. = abundance sensu Bush et al., 1997). The recruitment refers to the number of juvenile stages per 1000 erythrocytes. Data were statistically analyzed using SAS version 6.2. The used tests are mentioned in the text.

As a preliminary analysis showed close correlation (Kendall's rank-order correlation coefficient $T = 0.72$, $n = 34$, $p = 0.0001$) between body mass (= weight) and body condition (= body mass /length of tarsus in mm), data processing proceeded, using body mass as a correlating parameter.

RESULTS

Body masses of pigeons in Singapore (Kruskal-Wallis ANOVA, KW = 13.15, df = 5, $P = 0.02$) and in Israel (KW = 10.73, df = 3, $P = 0.01$) differed among months (Tables 1 and 2). In Singapore, the lightest and heaviest pigeons were recorded in June and April, respectively (Multiple-comparison tests, $P < 0.05$) vs. Israel, where the lightest and heaviest pigeons were recorded in April and October, respectively (Multiple-comparison tests, $P < 0.05$). Body masses of pigeons in April, the only month in which both areas were sampled, did not differ between Singapore and Israel (Mann-Whitney U-test, $Z = 1.36$, df = 28,50, $P = 0.16$).

Haemoproteus columbae was the only blood parasite recorded. The monthly prevalence of infected pigeons ranged from 94 to 100% in Singapore (Table 1) and from 65 to 100% in Israel (Table 2). Thus, overall prevalence of infection was higher in Singapore (96.8%) than in Israel (76.3%) ($\chi^2 = 21.51$, df = 1, $P = 0.0001$). A larger number of birds were infected in April in Singapore (92.9%) than in Israel (72.0%) ($\chi^2 = 4.79$, df = 1, $P = 0.03$). In any given month (except November in Israel - with n only = 5) prevalence in Singapore was higher, including the warmer months in Israel.

The mean parasitaemia did not vary among months in Singapore (Table 1, KW = 4.06, df = 3, $P = 0.25$), but varied significantly in Israel (Table 2, KW = 14.53, df = 3, $P = 0.002$). The lowest and highest mean parasitaemia in Israel was in January and October, respectively (Multiple-comparisons tests, $P < 0.05$). In April, the mean parasitaemia was still higher in Singapore than in Israel (Tables 1 and 2, $Z = 2.85$, df = 28,50, $P = 0.004$).

Mean parasite recruitment (abundance of juvenile gametocytes invading circulating erythrocytes) did not vary among months in Singapore (Table 1, KW = 4.39, df = 3, $P = 0.22$) but did vary among months in Israel (Table 2, KW

Table 2. *Haemoproteus columbae* infection characteristics in feral Pigeons from Israel. ± indicates standard error.

	November	January	April	October
No. of pigeons sampled	5	20	50	22
% infected	100	65	72	91
Mean parasitaemia	1.3±0.96	0.3±0.12	5.41±1.61	9.3±3.63
Mean parasite Recruitment	0.4±0.31	0.04±0.02	2.32±0.33	2.4±1.40
Mean body mass (g)	264.7±14.31	284.6±26.14	245.8±64.3	284.2±30.82

= 9.94, df = 3, P = 0.02). In Israel, mean parasite recruitment was the lowest and highest in January and October, respectively (Multiple-comparison tests, P < 0.05). In April, mean parasite recruitment was higher in Singapore than in Israel (Z = 2.52, df = 28,50, P = 0.01).

In Singapore, in March (r_s = -0.603. df = 15, P = 0.02) and May (r_s = -0.829, df = 17, P = 0.0001) heavier individuals had lower parasitaemia (Fig. 1, a and b). In Israel, heavier individuals had lower parasitaemia only in October (r_s = -0.428, df = 22, P = 0.05) (Fig. 1c); in all other months in both areas, this correlation was not statistically significant. All six squabs examined in Israel were negative for *Haemoproteus*. In both Singapore and Israel, parasite recruitment did not correlate with body mass in any of the months (P ≥ 0.08).

The proportion of individuals with and without louse flies did not differ both in Singapore (χ^2 = 0.37, df = 2, P = 0.83) and Israel (χ^2 = 1.74, df = 2, P = 0.41). The prevalence of louse flies in Singapore varied between 46 and 56% (Table 3) and between 35 and 50% in Israel (Table 4). The proportion of individuals with and without flies did not differ between Singapore and Israel both when sampling months were pooled (χ^2 = 0.36, df = 1, P = 0.54) and when only April was compared (χ^2 = 0.23, df = 1, P = 0.63). The mean number of louse flies did not differ among months both in Singapore (KW = 3.56, df = 2, P = 0.16) and in Israel (KW = 2.85, df = 2, P = 0.23; Tables 3 and 4). The mean number of louse flies did not differ between Singapore and Israel when sampling months were pooled (KW = 0.49, df = 1, P = 0.47) and when only April was compared (KW = 0.40, df = 1, P = 0.52).

DISCUSSION

Haemoproteus columbae occurs in pigeons associated with human settlements throughout the world (Wenyon, 1926; Bennett & Peirce, 1990). The prevalence of *H. columbae* in feral pigeons in different geographical locations varies from 14 to 100% (Garnham, 1966; Markus & Oosthnizen, 1972; Earle & Little, 1993; Sol et al., 2000). We found a relatively high prevalence and levels of parasitaemia of *H. columbae* in pigeons from Singapore and Israel. However, both the prevalence and the intensity of the parasitation was higher in Singapore than in Israel.

Although parallel data for January, the coldest and the least infected in Israel are lucking from Singapore, there, January is as hot and humid as the rest of the year's months. In Singapore there was little monthly variation prevalence and differences in mean levels of parasitaemia. Statistically not significant. Similarly, mean parasite recruitment (abundance of juvenile blood stages) varied only slightly throughout the study period. In Israel, on the other hand, levels of prevalence, parasitaemia and also the occurrences of juvenile stages varied from a low value in January (winter) to high levels in April to October. Recruitment figures during April and October were as high as in Singapore samples. These results clearly emphasize differences in the course of parasitation between Singapore's tropical and Israeli Mediterranean seasonal climates.

Parasite gametocytes are recruited into the blood from tissue meronts (schizonts), either pre-erythrocytic, which develope from sporozoites inoculated by louse flies (Aragao, 1908; Garnham, 1966), or from merozoites of chronic exoerythrocytic stages (Coatney, 1933). Sol et al. (2000) demonstrated a correlation between prevalence of infection in pigeons and abundance of the vector. Parasitaemia levels in pigeons, however, did not co-vary with abundance of vectors. Estimates of fly population size, however, are of limited reliability, since they are highly mobile, move among birds, and can escape during capture.

In Singapore louse flies were abundant in all sampling periods. In Israel, they only moderately declined in abundance in January (when ambient temperatures are 7 to 17°C). Fahmy et al. (1977) in Egypt found louse fly numbers to be significantly higher in summer: their population increased during summer (>25°C ambient temperatures) and declined in winter (at ambient temperatures below 20°C). In tropical India, local environmental constraints (such as extreme humidity) were suggested to lower louse fly population density (decline in yield, or delay in development of pupal stages) with consequent decline in transmission and infestation of pigeons (Mandal, 1991).

The prevalence of the louse fly alone, which is apparently

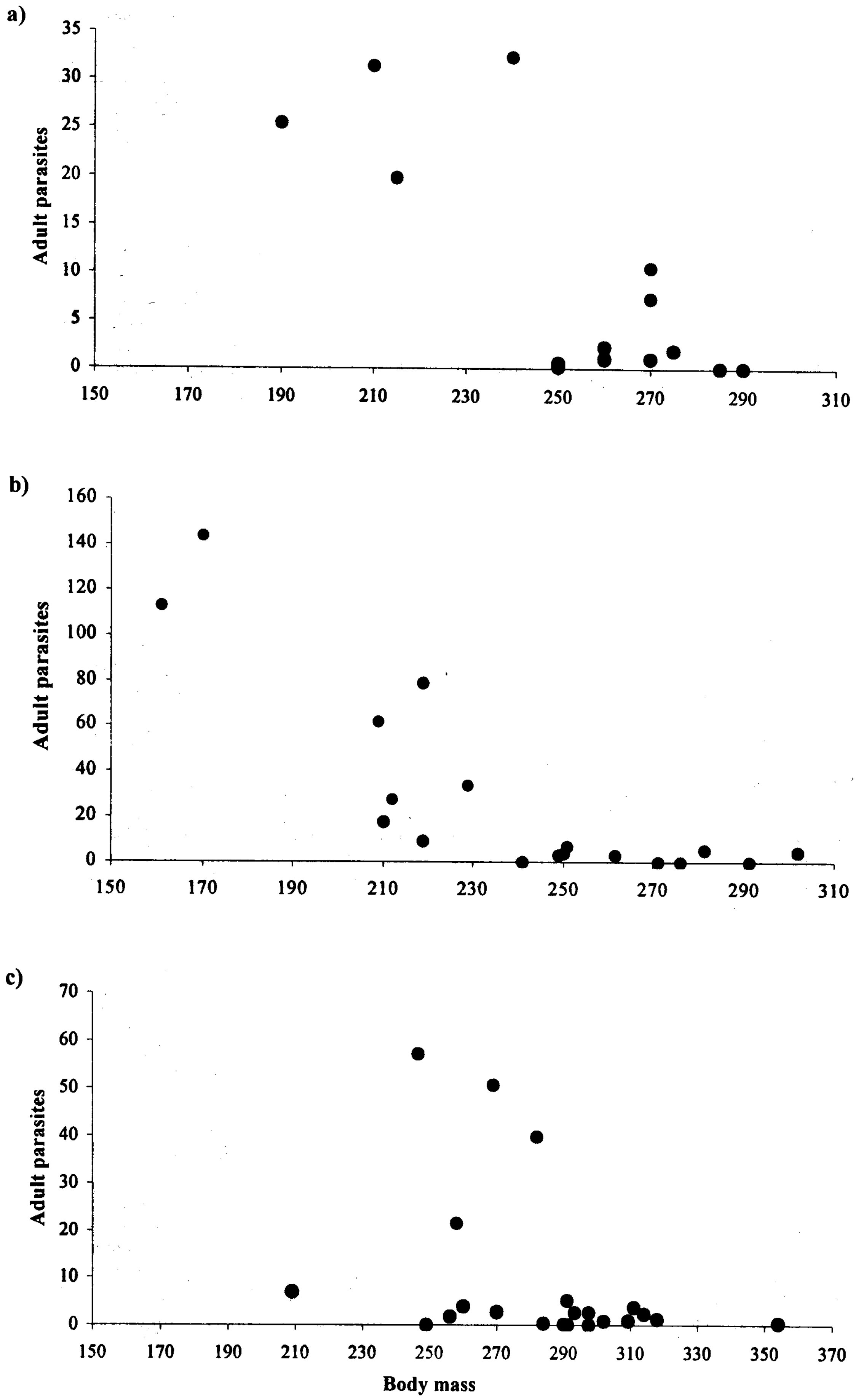

Fig. 1. Plotting of parasite count in blood (parasites/1000 erythrocytes) against body mass of individual pigeons: a) March data, Singapore; b) May data, Singapore; and c) October data, Israel.

Table 3. Monthly prevalence of louse fly in Singapore and Israel. ± indicates standard error.

	No. of pigeons sampled	% pigeon host to flies	Mean no. flies per pigeon
Singapore			
Apr.	28	46	0.5±0.12
May.	18	55	1.6±0.51
Jun.	16	56	1.2±0.32
Israel			
Nov.	5	40	2.2±1.02
Jan.	20	35	0.6±0.23
Apr.	44	50	0.9±0.20

about the same in Singapore and Israel, cannot explain the higher prevalence, parasitaemia and recruitment of *H. columbae* in Singapore compared with Israel. We found that recruitment through relapses played an important role in maintaining chronic infections. Follow up of blood infection in pigeons retained in captivity, in Singapore as well as in Israel, demonstrated repeated relapses, despite the absence of vector flies. Pigeons, kept fly-free, maintained infection for over 3-4 months. Large exoerythrocytic meronts were found in lungs and spleen of necropsied pigeons from these groups (Paperna, Sodhi & Smallridge, unpublished data). The magnitude and length of the relapse appears to be conditioned by immune responses of the bird (Coatney, 1933). Ahmed & Mohamed (1978), following up course of infection among experimentally infected young pigeons, found considerable variation in immune responses among birds, some developed immunity and others were repeatedly re-infected.

Body condition was found to correlate closely with body mass. For convenience, the latter parameter was preferred for data analysis. We found that lower weight individuals had higher parasitaemia in March and May in Singapore and in October in Israel. However, we found no relationship between the body mass and recruitment. The reasons for having a negative correlation between body mass and parasitaemia, and during some months and not during other months are difficult to explain. Sol et al. (2000) reports that older individuals had a higher probability of being parasitized, whereas younger birds when infected suffered higher parasitaemia. Richardson & Kendall (1963) similarly reported acute infections in young birds. Feral pigeons breed throughout the year in both areas. The few examined squabs, and prior fledging juveniles (in Israel) were all uninfected. Some samples might have been comprised of larger proportions of juvenile birds, which may have induced during these months significant correlation between body mass and parasitaemia.

From our experience, body mass does not seem to be a reliable measure for age, weight (~270 g) of fledged squabs was above the pigeon's sample mean (~250 g). Observed extreme individual variation in body mass, however could have been related to the hierarchy (dominance position) of the bird in the flock, which modulates access to food (Bertram, 1978). Lower rank birds with least access to food will apparently demonstrate lower body mass. Alternative options for reduced body weight are birds in the midst of their reproductive activity. Such birds in poor body condition, presumably, could have been with a lower immunological capacity to modulate the parasitaemia (Coatney, 1933; Ahmed & Mohammed, 1978); or as stipulated by Sheldon & Verhust (1996), cost of reproduction trades-off with the costly parasite defences.

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