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Short-term effects on body condition and size of immunocompetent organs in the hooded crow

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ABSTRACT

Only recently have there been attempts to assess the short-term variations of indices of body conditions and immunocompetence in free-living populations, and to evaluate the presence of significant fluctuations of these indices between years. We compared body conditions and size of immunocompetent organs (spleen and bursa of Fabricius) in wild populations of hooded crows in NW Italy during three years. Differences in population density among sites did not influence any of the variables, while a great difference was found between adult and subadult birds. In the study period, adults did not show any year-related variation of mass, structural size, lean mass (as assessed by TOBEC electroconductivity index), and spleen mass. Instead subadults of both sexes showed differences among years in mass; subadult females had the lowest values of bursa and spleen mass in the year when mass was smallest. The results show that in natural populations between-year variation of body conditions may exist. Subadults are probably most sensitive to adversities producing short-term variations in body conditions.

KEY WORDS: Hooded crow - Body condition - Spleen - Bursa of Fabricius - Age-related variation - Population density.

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INTRODUCTION

It is known that intraspecific competition can affect individual fitness. Density-dependent effects in birds have been widely reported and experimentally demonstrated in natural populations (Newton, 1998), while a number of studies has focused on the long-term effects of density on demographic reproductive parameters. In general, high density negatively affects immigration and survival of juveniles or adults, fecundity, and other parameters related to life history (Bendell *et al.*, 1972; Zwikel, 1972; Ekman *et al.*, 1981; Coulson *et al.*, 1982; Verhulst, 1992). However, few studies of birds have compared the short-term effects on body condition parameters in areas with different densities or have assessed whether those parameters show year-to-year variations in nature. In this kind of study, population composition should also be considered, since sex and age differences in body parameters have been reported for birds (Alisauskas & Ankney, 1987).

In the present study, we assessed body conditions in populations of hooded crow, *Corvus corone cornix* (Linnaeus, 1758), characterized by different densities. Moreover, we measured the mass of two immunocompetent organs (bursa of Fabricius and spleen), whose sizes have been assumed to be related to immune defence (Møller *et al.*, 1998).

The hooded crow is a generalist bird that may exploit a diversity of agricultural crops, and its ability to use a variety of food resources is a major reason for its success in Europe. The species occupies most habitats and is absent only from dense forests (Hagemeijer & Blair, 1997). It has developed a close association with human activity (Houston, 1977) and is a common urban scavenger (Coombs, 1978). In almost half the European countries, this species has increased its range since the mid-1970s (Cramp & Perrins, 1993), which reflects the crow's ability to adapt to environmental changes, particularly habitat fragmentation (Andrén, 1992). The hooded crow is particularly abundant in the agricultural plains of northern Italy, where nest density is generally higher than three per square kilometre. Since a high density of hooded crows can have negative effects on agriculture, game birds (Erikstad *et al.*, 1982; Tapper *et al.*, 1996), passerines (Loman, 1979), and waterfowls (Broyer *et al.*, 1995), population management (including culling) has been carried out (Slagsvold, 1978; Parker, 1985).

The present study was performed in association with a major culling operation conducted by faunal services in NW Italy who made available a large quantity of specimens. Our aim was to evaluate whether (a) density affects body condition and the mass of immunocompetent organs, and (b) if immunocompetent organ mass and body condition are subject to variations related to the year, locality, sex, and age.

MATERIALS AND METHODS

The study was carried out at eight sites in the province of Alessandria, NW Italy, in the spring of 1997, 1998, and 1999. We analysed 494 individuals: 410 from high density areas (Castellazzo, Novi, Castelceriolo, and Sezzadio) and 84 from low density ones (Montecastello, Marazzi, Cerrina, Solero). These individuals were a sample drawn from the approximately 11,000 hooded crows captured since 1996 in Larsen traps as part of the provincial administration's agricultural pest management program. No birds were sacrificed merely to collect scientific data. The personnel of the provincial administration were authorized to cull crows by the Istituto Nazionale Fauna Selvatica. Sex was determined by gonadal examination. Age was identified by the different upper mandible and plumage colours: subadults have unmoulted feathers and a pink mandible, while adults have uniformly moulted feathers and a dark grey upper mandible (Svensson, 1992). Captures were made in April and May, when some recently fledged individuals can be found in the area. However, in the present study recently fledged young were not considered, and all subadults were individuals born in the previous year. Measurements were taken on two occasions. In the field a few hours after capture, we measured (*in vivo*) each individual's mass and lean mass index with a TOBEC apparatus. Post mortem, each bird's body was brought to the lab, where morphological variables and internal organs were measured and body fat was extracted by Soxhlet. The following data were obtained.

Morphological variables, internal organs, and body composition

We measured: (i) body mass (0.1 g accuracy); (ii) wing length, with the flattened wing outstretched perpendicular to the body; (iii) tail length; (iv) tarsus length; (v) bill length. Tarsus and bill measurements were made to the nearest 0.1 mm with callipers, while the other two variables were measured to the nearest 0.5 mm with a flat ruler. The four body measurement (ii to v) were closely correlated; thus they were compacted in an index of structural size calculated as the first factor from a PCA. Factor 1 accounted 59.3 % of the variance in the original variables and was highly related to them (loadings: wing = 0.814, tail = 0.793, tarsus = 0.737, bill = 0.733). To obtain an index of body condition while controlling for structural size, the body condition of each individual was expressed as the residuals of body mass on Factor 1.

The abdominal cavity of dead birds was examined to extract internal organs, with (i) spleen mass; (ii) bursa of Fabricius mass; (iii) gonad mass being measured. All measurements were made with a precision balance to 0.1 mg accuracy.

In 201 live birds, the body composition was evaluated non-invasively by measurement of electrical conductivity with the TOBEC (Total Body Electrical Conductivity) system. This device estimates lean body mass, since the contribution of lipid tissue to conductivity is negligible (Walsberg, 1988; Castro *et al.*, 1990). Each individual was placed in the detection chamber (152 mm diameter) of an SA-3000 Small Animal Body Composition Analyzer (EM-SCAN Inc., Springfield, Illinois) for 5-6 sec and the electrical conductivity index was recorded: TOBEC index = (S - E) / R, where S = measurement with the specimen, E = empty measurement, and R = reference number (detailed instructions in the EM-SCAN operator's manual). For each individual, the measurement was repeated three times and the average value was employed in statistical analyses. A smaller sample of 36 individuals was used to determine body composition by Soxhlet after their TOBEC indices had been measured in vivo. The plucked carcasses were opened and dried to constant mass at 60° C in a vacuum oven. This process lasted 5 to 6 days and was followed by Soxhlet extraction of the whole dried carcass for 12 h in petrol ether. Total lean mass was calculated by summing the mass of the lean dried carcass and the water lost after initial drying (Scott *et al.*, 1991).

Population density and climatic variables

In the three study years, the line transects method was utilized to assess the abundance of hooded crows in the area. Eight transects, on average 14.6 km long (range 6-19 km), were positioned along existing roads and we drove past them in a slowly moving car. Both sides of the line were observed and when one or more crows were seen we stopped moving and measured their perpendicular distance to the transect with a telemeter. Transect surveys were carried out four times each winter, in the months of December, January, February, and March. Data were processed with the DISTANCE software (Laake *et al.*, 1996) to calculate crow densities. From the resulting values, the localities where birds were captured were divided into two categories:

- high-density sites (Castellazzo, Novi, Castelceriolo, and Sezzadio): the density in all years and all months was higher than 19.8 ind./km^2, the mean value in the study years being 27.4;
- low-density sites (Montecastello, Marazzi, Cerrina, Solero): the density was always lower than 5.9 ind./km^2, the mean value in the study years being 5.7.

At the sites to which sample birds belonged, culling activity was substantial (in the four high-density sites, about 900 birds were culled per year, while 200 were culled each year in the low-density ones). However, there was only slight variation of density during the study period; thus, no sites switched from the high- to low-density category or vice versa.

Meteorological data were obtained from the station of Casale (Alessandria). Climatic conditions were compared in different years by analysing the temperature and rainfall values during April and May, the months in which the hooded crows were captured.

Both PCA and regression analyses were performed with SYSTAT (Wilkinson, 1992). In the hooded crow, males are significantly larger than females and adults are larger than subadults. Therefore, all statistical analyses were conducted separately for each sex and age class.

RESULTS

The influence of population density on hooded crow body condition was evaluated only in 1997, due to a lack of samples in low-density areas during 1998 and 1999. There were no differences between high- and low-density areas for captured adults and subadults of either sex (Table I, Fig. 1).

Adults of neither sex varied during the three study years in body mass, mass of internal organs, and structural size. This constancy was observed at both of the most sampled sites, i.e. Castellazzo (Table II) and Novi (Table not reported; all P > 0.10).

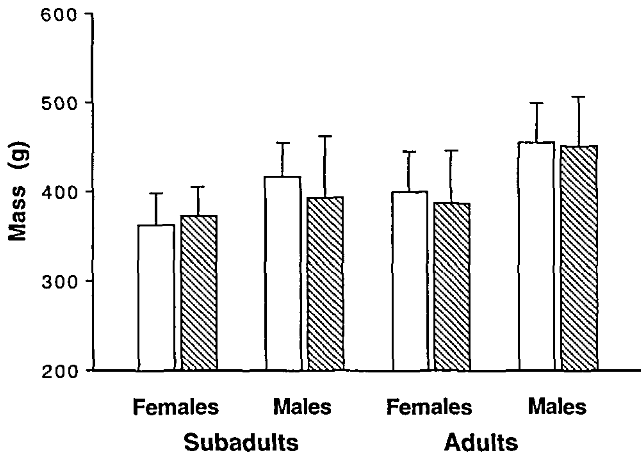

Fig. 1 - Body mass in high- (white bars) and low-density (hatched bars) areas in 1997 (mean ± SD).

TABLE I - *Comparison of mass, body size, and immunocompetent organs in areas with low and high density of crows in 1997.*

Parameter	Difference high/low density	
	F	P
Adult females (high: n = 26; low: n = 12)		
Mass	0.51	0.48
Size (PCA factor 1)	1.56	0.22
Body fat index	0.19	0.67
Spleen	0.60	0.45
Adult males (high: n = 36; low: n = 19)		
Mass	0.21	0.64
Size (PCA factor 1)	0.37	0.55
Body fat index	0.31	0.58
Spleen	0.60	0.44
Subadult females (high: n = 58; low: n = 24)		
Mass	1.35	0.25
Size (PCA factor 1)	2.32	0.13
Body fat index	0.68	0.41
Spleen	0.11	0.74
Bursa	1.14	0.29
Subadult males (high: n = 38; low: n = 29)		
Mass	3.24	0.08
Size (PCA factor 1)	2.10	0.15
Body fat index	0.50	0.48
Spleen	1.41	0.24
Bursa	1.29	0.26

Subadults of neither sex varied in structural size during the three years (Tables II and III), but they varied significantly in their mass (Fig. 2). At Castellazzo, the female and male subadult masses were lower in 1998 (post-hoc Tukey tests: 1997 vs 1998, $P < 0.05$; 1997 vs 1999, NS; 1998 vs 1999, $P < 0.05$), while at Novi the masses were lower in 1999 (post-hoc Tukey tests: 1997 vs 1998, NS; 1997 vs 1999, $P < 0.05$; 1998 vs 1999, $P < 0.05$).

In subadult males, there were no year-related variations in internal organ sizes at Castellazzo (Table II), nor at Novi (Table III). In subadult females, there were no year-related variations at Novi, while at Castellazzo the mass of the spleen and bursa differed significantly during the three years (Table II). Spleen mass was lower in 1998, when body mass was at its lowest levels (Fig. 3). In 1998, bursa mass was also lower than in 1997, but size did not increase in 1999.

Mean temperature during the capture period (from April to May) did not differ between years ($F_{2,180}$ = 1.28, P = 0.28) nor was there a difference in the observed rainfall in the three years ($F_{2,180}$ = 1.48, P = 0.23).

DISCUSSION

The density of hooded crows in the different areas had no significant influence on the morpho-physiological parameters and immunocompetent organs considered. The important changes in population condition were related to the different years and age classes: while in adults the measured parameters did not change among localities and years, there were significant variations in subadults.

Relationships between density and immunocompetence have been reported in poultry and laboratory animals, and recently in mammals in natural conditions (Saino *et al.*, 2000; Sinclair & Lochmiller, 2000). However no similar relationships have been investigated in wild birds. We did not find any effect of density on body size or immunocompetent organs in the hooded crow. The absence of density-dependent effects is not surprising: although various effects of density have been reported, in most cases density-dependence has been difficult to detect in natural populations (Lack, 1954, 1966). Indeed, comparisons of individuals living in high- and low-density areas could be complicated by possible differences in environmental quality of the zones, i.e. sites with high crow densities might also have a high-quality environment, and this could mask the effects of density on body parameters. It should also be considered that Larsen traps may be selective in relation to the territorial versus floater status of the crows, and this effect might also vary between areas with different population densities. Another possible complication is that the culling, performed both in high- and low-density areas, could have changed the density values during the study years. Although the culling activity lasted many years and was substantial, there was only slight density variation during the study period. It can be supposed that a large influx of birds from adjacent areas, not subjected to culling, may have replaced culled individuals. Thus, culling was not successful in

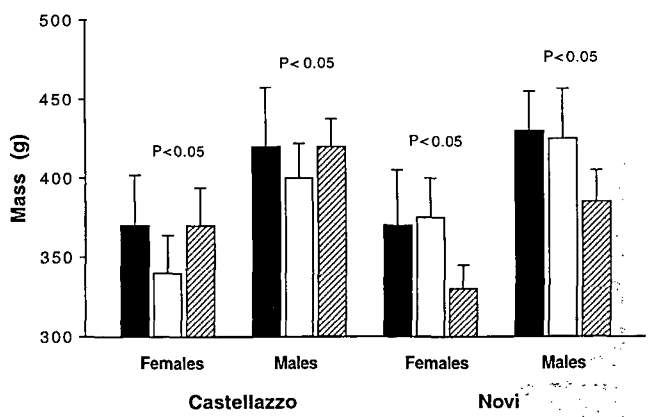

Fig. 2 - Body mass of subadults at Castellazzo and Novi during the three years (black bars = 1997; white = 1998; hatched = 1999).

TABLE II - *Hooded crows at Castellazzo. Comparison of mean values of body size, lean mass, fat index and internal organs over three years (1997 to 1999).*

Parameter	Difference high/low density	
	F	P
Adult females		
(1997: n = 18; 1998: n = 7; 1999: n = 5)		
Mass (g)	0.56	0.58
Size (PCA factor 1)	0.13	0.88
Lean mass (TOBEC)	0.05	0.95
Body fat index	0.02	0.97
Spleen (g)	2.21	0.13
Adult males		
(1997: n = 30; 1998: n = 9; 1999: n = 16)		
Mass (g)	0.73	0.49
Size (PCA factor 1)	0.72	0.49
Lean mass (TOBEC)	0.45	0.64
Body fat index	0.66	0.42
Spleen (g) 1.77 0.18		
Subadult females		
(1997: n = 45; 1998: n = 18; 1999: n = 53)		
Size (PCA factor 1)	1.20	0.30
Lean mass (TOBEC)	2.58	0.08
Body fat index	0.79	0.38
Spleen (g)	2.98	0.05*
Bursa (g)	3.86	0.03*
Subadult males		
(1997: n = 32; 1998: n = 12; 1999: n = 13)		
Size (PCA factor 1)	0.72	0.49
Lean mass (TOBEC)	2.40	0.09
Body fat index	0.66	0.42
Spleen (g)	1.22	0.30
Bursa (g)	0.19	0.83

reducing density. Indeed, apart from some systematic long-term removal experiments, in several cases there have been no clear-cut effects of removal on life-history parameters, nor on density, mainly because of immigration, which can maintain population density (Feare, 1991; Newton, 1998).

An important finding in our study was that there were year-to-year fluctuations of some parameters (body, spleen, and bursa mass) in subadults, while adults did not show changes among localities and years for any of the variables. It should be emphasized that the differences in subadult mass during the three study years did not follow a consistent trend. This age-class had a lower mean value in 1998 at Castellazzo and in 1999 at Novi. In this study, neither density nor a general climatic factor (temperature or rainfall) explained the fluctuation of body mass in subadults at the two most sampled sites. Both localities had high density and since they are near to one another, the temperature and rainfall did not vary significantly.

Why there were fluctuations only in subadults could be explained by their lesser ability to face environmental harshness. It has been shown that juvenile birds have a higher mortality than adults (Lack, 1966; Martin, 1995). Adults are larger than subadults, and larger individuals have a higher probability of breeding (Alisauskas, 1987) or obtaining a territory. In crows, Richner (1989) found that smaller individuals remain non-territorial: the social hierarchy of crows, in which juveniles are subordinate, can make it more difficult for them to find food. Finally, food capture efficiency increases with age in corvids (East, 1988). In summary, the subadult component of the population is probably most sensitive to adversities and is more likely to show short-term variations in body parameters.

Differences in subadult female mass among years were related to spleen and, at least in two years, to bursa mass. The avian spleen and the bursa of Fabricius are very important immune defence organs (John, 1994) involved in the production of antibodies designed to combat a range of different pathogens and parasites. Sex and age may affect the size of immune defence organs (Møller & Erritzoe, 1998), which undergo seasonal fluctuations in size (John, 1994). However, the possible existence of year-to-year variation has not been investigated in natural populations.

In our study, subadult females had the smallest sizes of immunocompetent organs in the year when mass was smallest. By assuming that a larger bursa of Fabricius or spleen can produce a better immune defense than a smaller organ (Møller *et al.*, 1998), thus in 1998 subadult females probably suffered more critical health conditions. A similar conclusion was reached in the same hooded crow population assessing the health status with blood analyses (Acquarone *et al.*, submitted).

In conclusion, the absence of an effect of density on body condition suggests that it might be appreciable only in extreme conditions (Newton, 1998). Our and

TABLE III - *Subadult hooded crows at Novi. Comparison of mean values of body size, lean mass, fat index and internal organs over three years (1997 to 1999).*

Parameter	Difference high/low density	
	F	P
Subadult females		
(1997: n = 8; 1998: n = 25; 1999: n = 29)		
Size (PCA factor 1)	0.32	0.73
Body fat index	1.61	0.21
Spleen (g)	1.05	0.30
Bursa (g)	0.53	0.62
Subadult males		
(1997: n = 6; 1998: n = 11; 1999: n = 30)		
Size (PCA factor 1)	0.56	0.58
Body fat index	0.46	0.50
Spleen (g)	1.48	0.16
Bursa (g)	0.52	0.62

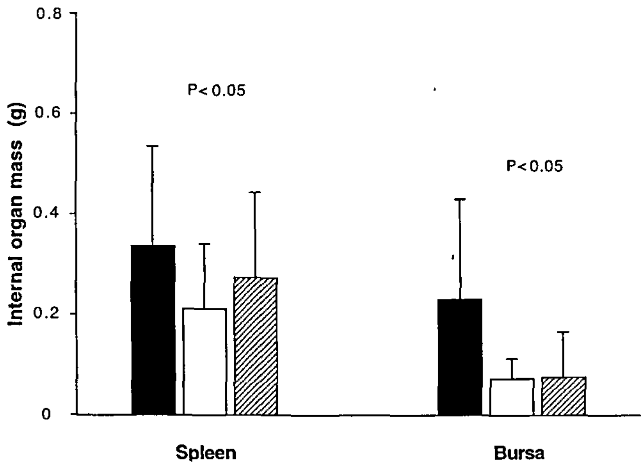

Fig. 3 - Comparison of spleen and bursa size in subadult females at Castellazzo during the three years (black bars = 1997; white = 1998; hatched = 1999).

other studies (Bosch *et al.*, 2000) show that sometimes differences in body conditions due to year-to-year variations can occur. Furthermore, our results suggest that when body and health conditions are compared, it is important to consider the differences between age classes.

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