On Individual, Sex and Age Differentiation of Indian House Crow (*Corvus splendens*) Call: A Preliminary Study in Potohar, Pakistan

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ON INDIVIDUAL, SEX AND AGE DIFFERENTIATION OF INDIAN HOUSE CROW (*CORVUS SPLENDENS*) CALL: A PRELIMINARY STUDY IN POTOHAR, PAKISTAN

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ABSTRACT

Considering importance of acoustics studies in population biology, 500 calls of the Indian House Crow (*Corvus splendens*) were recorded in morning - mid-afternoon hours (January-February, 2009) from 23 sites of urban areas of Potahar (Punjab, Pakistan). Calls were recorded using Sony CFS 1030 S sound records (sampling rate = 48 KHz) and edited using Sound Analysis Pro (Version 1.02). software using FFT method rate 50%, data window 9.27 ms, advanced window 1.36 ms. Through editing of calls, we selected 60 (37 ♂♂, 17 ♀♀, 6 Juvenile ♂♂) good quality spectrograms for detailed analysis. Spectrograms were characterized by rapid frequency modulations using 6 (call pitch, mean pitch goodness, mean frequency of the calls, frequency of modulations, mean amplitude modulation, mean wiener entropy) acoustic parameters. Significance of difference was analysed using Multivariate and Discriminate Function Analysis. Calls could be assigned to correct individual in 10.8% males, 21.0% females, and 42.9% juveniles, which was significantly higher than percentage of correct classification per chance. Calls could be attributes to correct sex in 88.5% and to correct age group in 80.6% of cases.

Keywords: acoustic parameters, male, female, juvenile.

INTRODUCTION

Acoustic signals play an important role in bird communication (Catchpole and Slater, 1995). These signals can be classified into songs and calls. Calls, unlike songs which are produced mainly during breeding season, are shorter and simpler elements and produced all the year round.

Calls play a role in social contacts and cohesion amongst flock family members, species and individual recognition, parent-offspring interactions, in escaping from a threat/danger (Catchpole and Slater, 1995; Kroodsma, 1996). Bioacoustics has in recent years developed rapidly as a tool to study communicative mechanisms in birds. However, studies on crow vocalisation are sparse (Chamberlain *et al.*, 1967, 1968, Chamberlain and Cornwell, 1971; Yorzinski *et al.*, 2006; Yorzinski and Vehrencamp, 2009), and probably none on Indian Common or House

Crow (*Corvus splendens*). Such studies can be useful in understanding species behaviour and developing automatic acoustic monitoring of the species (Bardeli *et al.*, 2009). Bioacoustics has a role in taxonomy (Martens *et al.*, 2000), and audiospectrographic analyses of typical calls of North American ((*C. brachyrrhynchus*) and Mexican (*C. imparatus*) Crows allowed Davis (1958) to propose a species status for Sinaloa (Mexico) Crow (*C. sinaloae*).

Chamberlain (1967) studied the sounds of Indian Common Crow with reference to environmental and behavioural contexts. Crow's call was used to disperse crow roosts (Delwiche *et al.*, 2007; Frings and Frings, 1957). Low auditory frequency cutoff (5.6 KHz) in Hooded Crow (*C. cornix*) suggested adaption to long-range communication useful in acoustic dispersal (Jensen and Klokke, 2006). Indian House Crow (IHC; *Corvus splendens*; family Corvidae, order Passeriformes). IHC is widely distributed in Pakistan, from lower coastal areas of Balochistan

to northern mountains up to 600 m above sea level. Bird calls throughout the year and are often very noisy and vocal even during the hottest summer mid-days. The commonest call is a high pitched nasal Kaan-Kaan or Quah-Quah (Roberts, 1992). The present report presents the results of the study instituted to: 1). describe common call of Indian House Crow, and 2). investigate potential individual, sex and age related differences in these calls. Such differences have a role in social interactions and reproductive behaviour of this species; and can potentially be used in studies on population biology using computer assisted automatic call recognition and classification.

MATERIALS AND METHODS

Field Recordings

We recorded calls of 500 (275 male, 145 female, 80 juvenile or subadult) IHC from early morning to mid-afternoon at 23 study sites located in peri-urban areas of Potahar (33° 24-39 N; 72° 19-29 E; Punjab, Pakistan) during January-February, 2009 using a Sony CFS 1030S sound recorder (sampling rate: 48 kHz).

The birds/group of birds were filmed at the same time using a Panasonic digital video camera (model NV-GS60GC-S), which were used for age (juvenile: bright red inside of inside the mouth) and sex (females having smaller and thinner black border around eyes) identification. We subjected 60 (adult males 37, adult females 17, juveniles 6) good quality and well identified recordings for present acoustic analysis.

Acoustic Analysis

Spectrograms of the recordings were edited using Sound Analysis Pro (version 1.02) software with the following settings: FFT method, Ratio 50%, data window 9.27 ms, advanced window 1.36 ms. We selected a total of 17 females, 37 males and 6 juvenile males good quality recordings (low level of background noise) for subsequent analyses ($N=7$ calls per

individual). Crow calls are harsh vocalisations characterized by a rapid frequency modulation (Figure 1). To investigate whether these calls are individualized and whether differences exist between calls of two sexes and between adult and juvenile calls, six acoustic parameters were measured automatically by Sound Analysis Pro software from the spectrogram of the calls (frequency vs. time): mean fundamental frequency along the call (Pitch), mean pitch goodness (PitchGood), mean frequency of the call (MeanFr), mean frequency modulation (FM), mean amplitude modulation (AM), and mean Wiener entropy (Entropy).

Figure 1: Spectrograms (above) and oscillograms (below) of an adult female call, an adult male call and a juvenile call (Seewave v.1.5.4, R package version 2.9.0. (Sueur *et al.*, 2009): FFT length, 512; overlap, 95%; resolution, 94 Hz, Hamming window).

Pitch goodness quantifies the periodicity of the spectrum, harmonic sounds having high values of pitch goodness and broadband noise and pure tones having low values of pitch goodness. Wiener entropy measures the width and uniformity of the power spectrum, broadband noise having high entropy values and pure tones and harmonic sounds having low entropy values. Frequency modulation is the time-related change of signal frequency and is measured as the angular component of squared time and frequency

derivatives of a sound, sounds whose frequency is not changing in time having FM values close to 0 degrees and those with rapidly changing frequency have values closer to 90 degrees. Amplitude modulation is the time-related change of signal amplitude, sounds whose amplitude is not changing in time having low AM values and those with rapidly changing amplitude having high AM values.

Statistical Analyses

We quantified the individual distinctiveness of calls by performing a Multivariate Analysis of Variance (MANOVA) and a Discriminant Function Analyses (DFA) on each category of calls (adult female, adult male and juvenile male). We first used MANOVA to confirm statistical differences in the parameters measured across individuals. We then used a DFA to quantify the extent to which individuals can be classified on the basis of their calls and which parameters account most for this classification (Johnson and Wichern, 1992). We calculated success rate of individual discrimination (correct classification) using a leave-one-out cross validation, in which the call to be classified is not used in the creation of the classification function (Lachenbruch and Mickey, 1968).

Percentages of correct classification by chance were calculated by applying a randomization procedure. The expected level of correct assignment was averaged from DFAs performed on 1000 randomized permutations of the data set (Solow, 1990; McGarigal et al., 2000). As sample sizes varied between different categories of calls (adult female: $N = 17$; adult male: $N = 37$; juvenile male: $N = 6$), we carried out a second DFA for adult male calls and adult female calls with 6 randomly chosen individuals to allow comparisons between call categories.

We investigated sex differences and differences between adult and juvenile males using a DFA and Generalized Linear Mixed Models (GLMMs) fitted with residual maximum

likelihood estimation (RELM, lme function; Venable and Ripley, 2002).

First, we carried out a DFA on the mean values per individual to calculate percentages of correct classifications of mean call values to their category (female vs. male, or adult male vs. juvenile male), and to examine which parameters account most for the difference between sexes and for the difference between adult and juvenile. Then, for each acoustic parameter measured, statistical comparison of male and female vocalisations and of adult male and juvenile male vocalizations were performed using GLMMs with individual identity fitted as a random term to control for repeated measurements of the same individual ($N = 7$ calls per individuals). Statistical analyses were carried out using R v.2.9.0 (R development core team 2009). All tests were two tailed and factors were considered to have a statistically significant influence if $P < 0.05$. All means are given with SEs.

RESULTS

Individual Differences

There was a significant difference among individuals for the acoustic parameters measured (MANOVA: female, $F_{96,612} = 3.19$; male, $F_{216,1332} = 1.98$; juvenile, $F_{30,175} = 3.04$; $P < 0.0001$ for all call categories). The first two discriminant functions (DF1 and DF2) generated by the DFAs carried out on each category of calls are presented in Table 1. DF1 and DF2 accounted together for $77.47 \pm 8.21\%$ ($N = 3$ DFAs). They were correlated ($|r| \geq 0.5$) with Pitch, MeanFr, FM and AM for male calls, Pitch, FM, PitchGood and MeanFr for female calls, and PitchGood, FM and MeanFr for juvenile calls.

Cross-validated DFA classified 21.0% of female calls to the correct female ($N = 17$ individuals), which was higher than the percentage of correct classification by chance (5.9%; permutation test: 1000 permutations, $P < 0.001$). Male calls were assigned to the correct

male in 10.8% of cases ($N = 37$ individuals), which was still higher than the percentage of correct classification by chance (2.7%; permutation test: 1000 permutations, $P < 0.001$). Calls of 42.9% of the juvenile were assigned to the correct juvenile ($N = 6$ individuals), which was also higher than the percentage of correct classification by chance (17.0%; permutation test: 1,000 permutations, $P < 0.001$, Figure 2).

Cross-validated DFAs carried out on six randomly chosen individuals classified 45.2% of female calls to the correct female and 26.2% of male calls to the correct male (Figure 2). Both these percentages were higher than the percentage of correct classification by chance (17.0%; permutation test: 1,000 permutations, $P < 0.001$ for both females and males). These results indicate that calls of females, males and juveniles are slightly individualized. Parameters accounting for this individuality vary between call categories, except for the frequency modulation and the mean frequency, which seem to be important for call individuality in all categories. Male calls seem to be poorly individualized compared to female and juvenile male calls (Figure 2).

Figure 2: Discriminant function scores of adult female, adult male and juvenile calls. Same shapes represent calls of same individual ($N = 6$ individuals, 7 calls per individual for each call category). Percentages of correct classification (CC) for

each category of calls are indicated. CC for male calls are lower than CC for female and juvenile calls.

Sex Variations

The first discrimination function generated by DFA carried out on female and male (Table 1) suggest that mean call values per individual was correlated ($|r| \geq 0.5$) with all parameters measured except for entropy ($|r| = -0.13$).

Mean call values were correctly attributed to their category (female or male) in 88.5% of the cases ($N = 54$ individuals), which was higher than the percentage of correct classification by chance (50.2%; permutation test: 1000 permutations, $P < 0.001$). GLMMs indicated sex differences for all parameters, except entropy (Table 2; Figure 3).

Pitch and MeanFr were higher whereas PitchGood, FM and AM were lower in females compared to males (Figure 3). Thus, crow calls differ between males and females, with female calls being higher-pitched and harsher (PitchGood), with slower frequency and amplitude modulation.

Table 2: Generalized linear mixed model output for differences between adult female and adult male calls and between adult and juvenile calls.

Parameter	Female/Male		Adult/Juvenile	
	$F_{1,51}$	P	$F_{1,40}$	P
Pitch	17.08	<0.001	3.56	0.07
PitchGood	62.61	<0.001	1.96	0.17
MeanFr	34.05	<0.001	15.02	<0.001
FM	106.50	<0.001	9.80	<0.01
AM	38.47	<0.001	10.77	<0.01
Entropy	1.42	0.24	16.92	<0.001
SylDur	423.59	<0.001	180.95	<0.001

Figure 3: Mean $\pm$ SEM values of the acoustic parameter measured for female calls (17), male calls (37) and juvenile calls (6) (GLMM: ** $P < 0.01$; * $P < 0.001$).**

Age Differences

First discrimination function generated by the DFA carried out on adults and juveniles (Table 1) indicated that mean call values per individual was correlated ($|r| \geq 0.5$), except for entropy ($|r| = 0.66$), and FM ($|r| = 0.40$). Mean call values were correctly attributed to their category (male or juvenile) in 80.6% of cases ($N = 43$ individuals), which was higher than the percentage of correct classification by chance (50.0%; permutation test: 1,000 permutations, $P < 0.001$). GLMMs indicate sex differences for all parameters except Pitch and PitchGood (Table 2; Figure 3).

MeanFr, FM, AM and entropy were higher in male calls compared to juvenile calls (Figure 3). Thus, crow calls differ between adult and juvenile males, with adult calls having higher frequencies, with more rapid frequency and amplitude modulations, and being harsher (entropy).

DISCUSSION

This was a preliminary study on the species and required standardizations. For the

purpose we restricted our studies to a specific area and to a limited part of the year, so that seasonal, geographical or ecotype differences do not complicate the results. We also selected 60 good quality recordings out of a total of 500 initially available, where call was clear, represented a single individual and confidence existed on sex and age of the individual. We could not separate sexes in juveniles, and hence present analysis was only on mixed population of juveniles.

Further studies are required on a broader selection of samples of calls coming from different localities, seasons and ecotypes, so that these could be used in the future population biology studies with some degree of confidence. With further standardizations acoustic analysis can potentially be a good alternative to call count techniques when analysing IHC population structure (size, sex and age structure) for exploited previously in songbirds (Rempel et al., 2005; Bardeli et al., 2009).

Present analysis has used calls recorded in wild, suggesting possibility of future use of techniques for studies on different population biology parameters of the wild population of IHC. This also suggests potentials of exploitation of the technique in other bird species. The results of this study with further refinement can also be used in analysis of mating, movement, anti-predatory and social behaviour in this species. We investigated individual, sex and age differences in IHC vocalisations. We found that IHC calls are slightly individualized, yet there are strong differences between sexes and age. We could correctly separate two sexes in 88.5% and adults from juveniles in 80.6% of the cases. Indeed, female calls are higher in frequency and harsher, compared to lower frequency and amplitude modulation in male calls. Adult male calls are harsher, higher in frequency, with more rapid frequency and amplitude modulations compared to juvenile calls. Further refinement is though required for better identification of individual call, yet present result suggest that crow calls contain individual, sex and age cues in their

vocalisations. These call distinction allow individuals to individually recognize their conspecifics, and to classify them as male or female or juvenile, which may be important for social interactions.

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