Impact of Feeding Habits on Population Ecology and Breeding Biology of Grey Francolin (Francolinus pondicerianus) in District Bhakkar, Punjab, Pakistan

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ARK performed the experiment, data analysis, wrote the manuscript. AAK conceived and designed the study and wrote the manuscript. JI and AMK, conceived and designed the study, edited manuscript. ZH, HMA and US, formal data analysis, edited the manuscript.

Key words
Grey francolin, Dispersion index, Fledging success, Arthropods, Ecological threats

ABSTRACT

Human-mediated wildlife reshuffling has raised concerns over the conservation of insects and predatory birds. Globally the population of grey francolin Francolinus pondicerianus is the Least Concern in the Red List of Endangered Species of Fauna and Flora of the International Union for Conservation of Nature (IUCN). However, its population has been significantly reduced in the desert ecosystem of Bhakkar (Punjab) Pakistan. The study was conducted to study the population ecology and breeding biology in relation to feeding habits of grey francolin in 16 different stands in Bhakkar, Punjab, Pakistan. The line transect data suggested grey francolin presence in 15 out of 16 stands (93.75%). An average density of grey francolins 13.01±6.39/km² varying between 0.30±0.23 and 1.97±0.65/ km² was recorded in different stands. Sex ratio of grey francolins (1.41:1) was skewed towards males. The current data on overall dispersion index of 2.1±0.43 per km² in different calendar months reflected a clumped dispersion. Sixty crops of adult birds were analyzed for food contents in different seasons. Leaves, seeds and arthropods were highly consumed food contents in all seasons. Descriptive data analysis indicated a statistically significant value of $p < 0.05$ between arthropod abundance and volume of arthropods in the diet of both adults and juveniles. Chi-square test ($\chi^2$) reflected a positive relationship between adult crop contents and different calendar months and stands. The data regarding breeding biology exhibited mean incubation period, clutch size and egg weight of 16.67±1.45, 4.0±0.58, 9.56±0.33 in study (stand 12) and 13.60±0.46, 6.4±0.46, 11.44±0.58 in control area (stand 16) respectively. There was a positive trend of threat factors with density and dispersion index ($p < 0.05$). The results indicated that, seasonal variations affected the feeding habits of grey francolin, which caused fluctuation in density and dispersion index of this species. Future studies are also warranted to evaluate the impact of injudicious use of pesticides on predatory performance of these birds against a wide range of arthropods including grasshoppers, crickets and locusts.

INTRODUCTION

Grey francolin, Francolinus pondicerianus (Gmelin, 1789) is widely associated with drier parts of the Indus plains and penetrates Thar desert in Sindh, and Dera Ghazi Khan, Cholistan and Thal deserts, Kala Chitta Range, Salt Range and Potwar plateau in Punjab, the bird also occurs in suitable habitats occurring in Balochistan, Khyber Pakhtunkhwa and Azad Kashmir. Grey francolin, a medium sized bird is the prime game and cage bird of India and Pakistan (Roberts, 1991; Rasmussen and Anderton, 2012). Indiscriminate hunting, poaching, habitat destruction, severe cold, draught, agricultural pesticides and agricultural intensification were the most common threats causing the decline of grey francolin populations (Potts, 1997; Richard et al., 2002; Bro et al., 2004; Liao et al., 2007). Grey francolins are omnivorous (Fulher et al., 2000; Sande et al., 2007; Hussain et al., 2012). Mahmood et al. (2010) reported that average population density of grey francolin was 6/km² (0.06/ha) in Lehri National Park, Jhelum, although this habitat is different from the habitat in Bhakkar. Hussain et al. (2012) estimated per hectare...
population density of grey francolins as 1.59 ± 0.39 per hectare (159 per km²) in irrigated tract and in forest tracts it was 0.87 ± 0.14 per hectare (87 per km²) birds. The grey francolin indicated a declining trend in the past, and was an indicator species for farmland ecosystems suggesting wildlife populations not always getting benefit from different agricultural schemes (Kleijn et al., 2001; Kleijn and Sutherland, 2003).

In agro-ecosystem of Pothwar Plateau, the average breeding performance of grey francolin was observed as 7.0±0.36 (clutch size), 6.00±0.36 days (egg laying period), 20.6±0.50 days (incubation period) and 5.33±1.22 (hatching success) (Hussain et al., 2012). The analysis of 140 crops showed that the species had a higher dependence on arthropods during summer and on plants during winter. They observed that 10 crop contents had arthropods which constituted 28% of food of grey francolin which preferred a wider base of food contents, advantageous for survival under all the odds without lowering their populations seriously (Khan et al., 2015). The frequency distribution of food types within crops of grey-winged francolins (Scleroptila atra) in Losotho indicated that arthropods constituted 7.3% of food items by volume (Little et al., 1992). Keeping in view the declining patterns of this phasianid species, the present study was conducted in different areas of District Bhakkar during June 2018 to May 2019, wherefrom so far, no data were available. The present study provides information on population ecology and breeding biology of grey francolin, a valuable asset of the Thal desert ecosystem of District Bhakkar, with reference to feeding habits.

MATERIALS AND METHODS

**Study area and sampling design**

District Bhakkar (31.6° N 71.0° E, 149m (522 feet) above mean sea level) is part of Thal desert, Punjab, Pakistan. Thal sand dunes and xeric vegetation are the main features of the desert landscape. Bhakkar is divided into four tehsils: Kalurkot, Darya Khan, Bhakkar and Mankera. Average annual temperature is 24.6 °C while mean annual precipitation is 213 mm.

A detailed survey was carried out during June 2018 to May 2019 in different areas of District Bhakkar. Population of grey francolin was estimated by using line transect method as explained by Burnham et al. (1980). In order to find out the potential population of grey francolin, a few preliminary surveys were conducted which provided a base to divide the study area into 16 stands (1. Taragran wala 2. Ghulamaan 3. Rakh Ghulamaan 4. Chak 19 ML 5. Rakh Mahota 6. Rakh Dalaaan 7. Joyia 8. Jhamat Shumali 9. Dajal 10. 51 TDA 11. 57 TDA 12. 61 TDA 13. Rakh Karlu Wala 14. Rakh Mankera 15. Rakh Kapahi and 16. Rakh Mahni), with possible dense population of grey francolin. Although habitat conditions in these 16 stands might be different from one another, they were selected based on similar cropping pattern in the study area. One stand is situated along the Indus belt (stand 9) while others are in the plains region.

A permanent fixed transect line (1.5 km) was established in each of 16 stands, trying to maintain a straight line passing diagonally through stand area covering all possible microhabitat variations. The grey francolin was searched by direct (sighting) and indirect (calls, faecal pellets) methods. Each transect line was walked by two workers at a moderate speed of 2-3 km/hour in the morning (0600 to 0930 h) and in the evening (1600 to 1830 h) from June 2018 to May 2019. The data were collected fortnightly from each stand. Four stands (transects) were walked in one day in each tehsil and walked by different workers, completing the exercise in four days every fortnight. Each line transect covered an average band of about 40 m on each side as record suggested an approximate distance of sighted birds from the transect line, hence a uniform transect width of 80 m (0.08 km) was fixed for analysis of fine transect data. Data on age (young and adult) and sex (male and female based on spurs on legs of males) of each sighted bird were recorded.

**Collection and analysis of food contents**

Thirty crops were obtained from grey francolins collected randomly from local hunters after fulfilling formalities of the respective Department from different stands in the study area during different seasons (spring from February-April, n= 9; summer from May-July, n= 8; autumn from August-October, n= 6; winter from November-January, n= 7). Sixty crops were obtained randomly from adults during different seasons in spring (n= 15), summer (n= 15), autumn (n= 15) and winter (n= 15). The weight of each crop was measured and the crop packed separately in plastic bags. Each crop was preserved in 7% formalin solution and was dissected in the laboratory for removal of food contents. The crops were weighed for food contents. The identifiable food contents were separated and categorized to the lowest possible taxonomic ranks following the work of Faruqi et al. (1960). Some crop contents could not be identified and were designated as unidentified plants or animals. Arthropods were collected randomly from different stands by quadrat sampling method. A quadrate of 0.25 m² size was used fortnightly each month at a rate of one minute for arthropod collection. All samples were measured in volume (ml) by water displacement method.
Threat survey and data analysis

Besides direct field observations, questionnaires were used to determine threats to grey francolin population. Random sampling methodology was applied during the current study. Detailed structured open and close-ended questionnaires were used to get information on various threat factors in different stands of the study area. The questionnaires included information on the use of pesticides, hunting, habitat destruction and other factors. Questionnaires (160) were filled from local hunters for the study from different stands (10 from each). On the basis of this information stands were designated as affected areas (stand 12) and control areas (stand 16) in order to evaluate impact of threat factors on population ecology, breeding biology (clutch size, incubation period, hatching success, fledging success, egg weight and egg width) and feeding habits. The stand 12 was considered an affected area due to low population of grey francolin and habitat destruction while stand 16 was taken as control area due to high grey francolin population and habitat intact. Prior to data collection, an informed consent was taken from the participants for their willingness to share the information.

Significance of difference between male: female sex ratio for calendar months, and adult crop contents in different months and from different stands was tested at 0.05 level by using Chi square test ($\chi^2$). The formula, number of sighted birds divided by transect area (transect length x average transect width, 1.5 km x 0.08 km= 0.12 km$^2$) was used to calculate the density of bird population (per km$^2$) in each stand. Independent samples t-test and One-way-ANOVA at significance level of 0.05 were analyzed by using SPSS 16 software package. Pearson coefficient of linear correlation was calculated to determine association of densities of different calendar months with different variables. Pearson coefficient of linear correlation and Regression model was used to determine association of arthropod abundance in different calendar months. Dispersion index, which is the scattering of grey francolin from the transect line, was calculated by dividing group variance by group mean for each category (Odum, 1971). The values of index <1 suggested a uniform, =1 random and >1 clumped population.

RESULTS AND DISCUSSION

Human-mediated wildlife reshuffling has raised concerns over the conservation of biodiversity. Globally the population of grey francolin is categorized as “Least Concern” in the Red List of threatened species (IUCN 2019-20). In investigated desert areas, the grey francolin presented a high density of 1.97±0.65/km$^2$ (Fig. 1) and 1.76±0.62/km$^2$ in stand 16 (Table 1). Pearson coefficient of linear correlation and Regression model suggested a direct link of seasonal factors with average densities of grey francolin and arthropod abundance (individuals/m$^2$) ($p < 0.05$) (not shown in tables). t-test and One-Way-ANOVA results indicated that there was no significant difference between morning and evening samples of grey francolin (data not shown). Chi square test ($\chi^2$) indicated that there was no significant difference between different samples of males and females of grey francolin during different calendar months. Stand data suggested that grey francolin had an average of 0.14 young/adult and 0.33 young/female ratios (data not shown). Grey francolin maintained a fairly clumped dispersion during different calendar months and showed an overall dispersion index of 2.1±0.43 (Fig. 2). Pearson coefficient of linear correlation between dispersion index and precipitation indicated values of $p > 0.05$, $t = 0.65$ (Fig. 2). Dispersion indices, for morning and evening samples for different calendar months were not much different ($p > 0.05$, df =21, $t = 0.71$; $p > 0.05$, df = 02, F = 0.90). The morning and evening samples in different stands revealed a clumped dispersion (mornings =15.61±3.5, evenings = 14.40±2.78). t-test ($p > 0.05$, df = 30, $t = 0.25$) and One-Way-ANOVA ($p > 0.05$, df = 1, F = 0.25) indicated that there was no significant difference between morning and evening samples.

![Fig. 1. Adult grey francolin abundance and population density (mean±SE/ km$^2$) in District Bhakkar (Punjab) Pakistan during different calendar months.](image-url)
Fig. 2. Density (mean±SE/ km²), dispersion index (variance/mean) of grey francolin and Arthropod populations in District Bhakkar (Punjab) Pakistan affected by precipitation during different calendar months.

Table 1. Population density (mean±S.E./ km²) of grey francolin in different stands in District Bhakkar (Punjab) Pakistan. Number of transects (n=384) and transect area (46.08 km²) remained constant in all stands.

| Stands | Number of birds sighted | Density per km² |
|--------|--------------------------|-----------------|
| 1      | 27                       | 0.59±0.33       |
| 2      | 13                       | 0.28±0.24       |
| 3      | 21                       | 0.46±0.35       |
| 4      | 14                       | 0.30±0.23       |
| 5      | 18                       | 0.39±0.27       |
| 6      | 63                       | 1.37±0.43       |
| 7      | 72                       | 1.56±0.51       |
| 8      | 57                       | 1.24±0.70       |
| 9      | 0                        | 0               |
| 10     | 39                       | 0.85±0.54       |
| 11     | 19                       | 0.41±0.38       |
| 12     | 12                       | 0.26±0.24       |
| 13     | 49                       | 1.06±0.59       |
| 14     | 75                       | 1.63±0.52       |
| 15     | 39                       | 0.85±0.44       |
| 16     | 81                       | 1.76±0.62       |
| Overall| 599                      | 13.01±6.39      |

The analysis of seasonal samples revealed significant increase in body mass, crop mass and crop volume of adult males while young females elucidated a positive increase than males with slight but non-significant variation, respectively (Table II). The rigorous consumption of leaves, seeds and arthropods was observed in all seasons and stands for adult grey francolin (Figs. 3 and 4) while arthropods (0.11±0.01) were highly preferable food for young ones (Fig. 5, p < 0.05). t-test results elucidated a positive relationship between seasonal fluctuations and feeding habits of grey francolin (p < 0.05) (data not shown). Chi square test (χ²) explained linear relationship between different food items of leaves, fruits, grit, other invertebrates, arthropods and other animals in adults during different months and stands (Table V). A statistically positive trend was observed between feeding habits and density and dispersion index of adult and juvenile grey francolins. t-test and One-Way-ANOVA results clearly reflected a highly significant association of crop mass and crop volume with body mass, density and dispersion index (p < 0.05) (Table III). p < 0.05 (t-test) elucidated a direct

Fig. 3. Seasonal distribution of proportion of food items by volume (ml) within crops of grey francolin adults in District Bhakkar (Punjab) Pakistan.

Fig. 4. Stand wise distribution of proportion of food items by volume (ml) within crops of grey francolin adults in District Bhakkar (Punjab) Pakistan.
Table II. A comparison of sample sizes between male and female grey francolin in District Bhakkar (Punjab) Pakistan during four seasons of the year 2018.

| Grey francolins | Season | Spring | Summer | Autumn | Winter |
|-----------------|--------|--------|--------|--------|--------|
|                 | Body mass (g) |        |        |        |        |
| Adult           | 443.4±2.9 M | 442.8±2.2 M | 436.8±1.69 M | 435.4±3.1 M |
|                 | 440.7±3.7 F | 439.5±2.9 F | 436.3±1.14 F | 432.2±2.8 F |
| Juvenile        | 346.78±5.13M | 359.5±1.22 M | 325.1±5.76M | 330.35±3.51M |
|                 | 344.98±4.65 F | 332.43±0.95 F | 328.3±1.33 F | 322.3±4.78 F |
|                 | Crop mass (g) |        |        |        |        |
| Adult           | 5.09±0.1 M | 5.06±0.16 M | 4.64±0.14 M | 4.59±0.28 M |
|                 | 4.99±0.13 F | 5.01±0.06 F | 4.71±0.13 F | 3.51±0.61 F |
| Juvenile        | 4.35±0.16 M | 4.35±0.09 M | 3.40±0.10 M | 3.87±0.16 M |
|                 | 4.38±0.15 F | 4.40±0.11 F | 4.0±0.15 F | 3.76±0.21 F |
|                 | Crop volume (ml) |        |        |        |        |
| Adult           | 3.78±0.05 M | 3.82±0.02 M | 3.64±0.05 M | 3.51±0.17 M |
|                 | 3.67±0.06 F | 3.78±0.03 F | 3.67±0.05 F | 3.73±0.02 F |
| Juvenile        | 3.34±0.14 M | 3.54±0.06 M | 2.69±0.13 M | 2.78±0.14 M |
|                 | 3.50±0.13 F | 3.77±0.02 F | 2.92±0.13 F | 2.68±0.18 F |

Table III. The statistical analysis of density and dispersion index of grey francolin in relation to threat factors in different stands in District Bhakkar (Punjab) Pakistan.

| Density | Pesticides | Shooting | Habitat destruction | Others | t-value  | p-value |
|---------|------------|----------|---------------------|--------|----------|---------|
| Total respondents (N=160) | (N=60) | (N=48) | (N=27) | (N=25) |          |         |
| t-value | 10.249     | 8.834    | 3.018               | 2.845  |          |         |
| p-value | 0.000**    | 0.000**  | 0.005*              | 0.008* |          |         |

Dispersion index

| t-value | 5.463     | 4.556    | 1.170               | 0.577  |          |         |
| p-value | 0.000**   | 0.000**  | 0.251               | 0.568  |          |         |

Significant, *; Highly significant, **; $Agriculture practices by farmers and fuel wood collection.

Table IV. Chi square test ($\chi^2$) for relationship of different food items of adult grey francolin during different months and stands in District Bhakkar (Punjab) Pakistan.

| Food items | Month wise data | Stand wise data |
|------------|-----------------|-----------------|
|            | p-value | p-value |
| Leaves     | 0.046* | 0.130 |
| Buds       | 0.020* | 0.189 |
| Seeds      | 0.087  | 0.000** |
| Grit       | 0.009** | 0.000** |
| Fruits     | 0.006** | 0.000** |
| Unidentified plants | 0.012* | 0.082 |
| Other invertebrates | 0.035* | 0.000** |
| Arthropods | 0.033* | 0.000** |
| Unidentified animals | 0.027* | 0.000** |

Significant*; Highly significant**

Fig. 5. Seasonal distribution of proportion of food items by volume (ml) within crops of grey francolin juveniles (< 1 year old) in District Bhakkar (Punjab) Pakistan.

Table V. Measurements of egg weight (g), egg width (mm) and breeding success of adult grey francolin in study and control area (Mean±S.E) in District Bhakkar.

| Parameters | Stand 12 | Stand 16 | t-test | p-value |
|------------|----------|----------|--------|---------|
| Clutch size | 4.0±0.58 | 6.40±0.46 | -3.000 | 0.024*  |
| Incubation period | 16.7±1.45 | 13.6±0.46 | 2.462 | 0.049*  |
| Hatched eggs | 2.33±0.33 | 5.4±0.34 | -5.231 | 0.002** |
| Fledging success | 1.67±0.33 | 4.2±0.33 | -4.564 | 0.004** |
| Egg weight | 9.57±0.33 | 11.44±0.58 | -2.095 | 0.081  |
| Egg width | 29.9±0.21 | 31.1±0.40 | -1.985 | 0.094  |

Significant*; Highly significant**
link of density and dispersion index with different threat factors (Table IV) and arthropod abundance with volume of arthropods in diet of adults in different seasons with slight variations in juveniles (data not shown). A significant variation in clutch size, hatched eggs and fledging success was observed in stand 12 ($p < 0.05$, t-test).

Population density of grey francolin ($13.01 \pm 6.39$ /km$^2$) was found to be higher in the morning (mean 29.25) due to foraging than in the evening (mean 20.67). These results matched with the findings of Rotella and Ratti (1988). Maximum population density of grey francolins ($1.97$ birds / km$^2$) was in April. Population density of grey francolin in Lal Suhanra National Park, Bahawalpur, Pakistan was $8.40$ birds / km$^2$ (Khan, 2010). The lowest density $0.30 \pm 0.23$ was reported in December 2018. It might be due to low temperature, drought, hunting, use of pesticides and local migrations for new food sources (Odum, 1971; Khan et al., 2015). Population recruitments expressed seasonal changes in age structure when chicks were more than two months old after September. It was difficult to differentiate young ones from adults under field conditions. Male: Female sex ratio was skewed towards males in grey francolins (1.41: 1). It was due to severe shooting and temperature variations (Khan et al., 2015). The present findings of dispersion index of grey francolin (2.1$\pm$0.43) showed a clumped dispersion. Jansen et al. (2001) studied different levels of clumping of redwing francolin ($F$. levaillantii) in South Africa. Odum (1971) suggested that dispersion index had survival value for bird species. Uniform dispersion indicated a decreased intraspecific competition and a group protection from environmental hazards.

Leaves contributed $0.094 \pm 0.006$ km$^2$ of average volume of crop contents while arthropods expressed $0.161 \pm 0.022$ km$^2$ for adult grey francolin during different calendar months. Little (1992) indicated high proportion of arthropods by volume in crops of adult grey francolin in all seasons. Leaves, seeds and arthropods were highly consumed food diversity by Phasianid species. Tsachalidis et al. (2007) and Paralikidis et al. (2010) confirmed such findings. Phasianid species number was affected by human agricultural practices which modified the diet of these species (Chamberlain et al., 2000).

The data regarding breeding biology exhibited mean hatching success and fledging success of 2.33$\pm$0.33, 1.67$\pm$0.33 per clutch respectively in the study area (stand 12), and $5.4 \pm 0.36$, $4.2 \pm 0.33$ per clutch in control area (stand 16) respectively. In agro ecosystem of Pothwar Plateau, the average breeding performance of grey francolin was observed as $5.1 \pm 0.65$ (hatching success) and $4.3 \pm 0.66$ (fledging success) in forest habitat; $5.5 \pm 2.10$ (hatching success) and $5.0 \pm 1.95$ (fledging success) in cultivated habitat (Hussain et al., 2012). Since we lack previous published work in Bhakkar region, the above cited study might be appropriate for comparison.

**CONCLUSION**

Seasonal variations affected the feeding habits of grey francolin, which resulted in fluctuations in density and dispersion index of this species. Illegal hunting, habitat destruction and use of pesticides are the main threats to this valuable asset of Thal desert ecosystem. In grey francolin, there was a positive trend of crop mass and crop volume with body mass. Future studies are also warranted to evaluate their predatory performance against a wide range of arthropods and especially grasshoppers, crickets and locusts. It is recommended to conserve foraging areas and furthermore promote the judicious use of pesticides and control illegal hunting to save these game and cage birds that help the agriculturist by consuming crop pests.

**Statement of conflict of interest**

The authors have declared no conflict of interest.

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