Short Communications

Pakistan J. Zool., vol. 44 (5), pp. 1439-1442, 2012.

A Preliminary Report on the Diversity of Spiders (Arachnida: Araneae) in the Cholistan Desert, Pakistan

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Abstract.- Spiders (Arachnida.: Araneae) were collected by hand picking and shaking plants methods from different habitats of the Cholistan Desert. These samples contained 10 families, 32 genera and 62 species. The Philodromidae was found to have greatest diversity of species (30%) followed by Lycosidae (26%) and Gnaphosidae (18%) respectively. Studies on species sex ratios studies indicated that generally female spiders were more abundant than males and immature spiders during the entire study. The present study provides preliminary data on the diversity of spiders which can be useful for the researchers working on diversity, taxonomy and conservation of spiders. It was concluded that the Arachnida is a diverse class of Arthopoda in the Cholistan Desert. However, it is suggested that more intensive studies are needed to fully document the arachnid diversity in this region.

Key words: Araneae, Cholistan Desert, sex ratio.

The Cholistan Desert comprises of a large area of 26000 km² in the South Eastern portion of Punjab Province. It is located between latitude $27^{\circ}42$ and $29^{\circ}45$ north and longitude $69^{\circ}52$ and $72^{\circ}24$ east (FAO, 1993; Akbar *et al.*, 1996). This desert is characterized by extremely hot summers ($\pm 51^{\circ}$ C) and cold winters ($\pm 0^{\circ}$ C). The relative humidity is low in summer but high during winter (Fig.1). The flora of the desert mostly consists of drought resistant plants. The most common grasses such as *Cenchrus, Lasiurus, Cymbopogon* and *Penicum* and shrub and tree species (*Calligonum, Haloxylon, Prosapis, Zizyphus and Acacia*) have profound effects on habitat structure and fauna (Arshad and Rao, 1994). A number of researchers have been exploring the ecology and taxonomy of desert spiders all over the world (Ward and Lubin, 1993: Hanschel, 1994: Lubin and Hanschel, 1996: Brandit, 1998), and India (Tikader, 1974, 1980, 1982; Tikader and Biswas, 1981). Information on the desert spiders of Pakistan is very scarce. Ghafoor and Beg (2002) and Mukhtar (2004) have provided important taxonomic information on species from various non desert locations in Punjab Province, Pakistan. Considering the small amount of data available on the taxonomy of Pakistan desert spiders, this study was initiated to provide bench mark data on the diversity of desert spiders of the Cholistan Desert which can be utilized by future researchers working on the taxonomy, distribution and conservation of desert spiders.



Fig. 1. Monthly average meteorological data for five years (2001 – 2005) including the study years (2001 – 2003) from Cholistan Desert. Source: Regional Meteorological Station Bahawalpur.

Materials and methods

Specimens were collected from January 2001 to December 2003. The samples were collected from various localities around Bahawalpur, its out skirts and the desert area of Bughdad - ul - jadeed Campus, The Islamia University of Bahawalpur.

Arachnids were collected using hand picking and shaking plants methods. Collected samples were stored in separate vials containing preservative solution (50 ml 70% ethanol, 1 ml glycerin, and 5ml glacial aectic acid). Samples were stored at a cool place in the laboratory until studied. Specimens were identified to the species level with the aid of various standard taxonomic keys provided by Pocock, (1900), Tikadar (1980) and Tikadar and

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Biswas (1981). The specimens collected were enumerated and sexed. Some specimens could not be identified to the species level because of unresoleved taxonomic problems. Immature spiders are generally considered difficult to the species level. Some specimen may represent new species.

Results

A total of 10 families, 32 genera and 62 spider species were recoded from the specimens collected during this study. The Philodromidae had the highest diversity (19 species) while Therididae, Zodaridae and Tetragnathidae each had but one species. The Lycosidae, Graphosidae, Araneidae, Thomisidae and Erasidae were represented 16, 11, 4, 4 and 2 species, respectively (Table I).

The relative diversity (%) among families is given in Figure 2. The Philodromidae has the highest diversity (30% of spider species collected), followed by the Lycosidae (26%) and Gnaphosidae (18%), respectively. The Araneidae and Thomisidae had 6% of the total diversity, while the rest of the families had 5% or less.



Fig. 2. Relative diversity of spider families recorded during in three study years

The sex ratios for the examined spiders (Fig. 3) showed that the maximum percentage of females to males occurred during 2001 and 2003 while the minimum occurred in 2002. The number of male spiders generally remained very low compared to that of female spiders; however, relatively more male spiders were collected during 2002. The lowest number of immature spiders was observed in 2003 and the highest in 2002.



Fig. 3. Sex ratio of spiders collected by hand picking method during study period (2001 -2003)

Discussion

Little is known about the ecology and diversity of arachnids from Pakistan's desert areas, particularly the Cholistan Desert. Our results show that 10 families with 62 species were encountered. A relatively high diversity of spiders can be expected in the desert areas considering our findings and those of Sivaperuman and Rathore (2004) who observed 13 families and 28 species in the Desert National Park of Rajusthan India. The diversity of spiders in deserts appears to be strongly influenced by habitat conditions and vegetation structure as we collected a higher number of spiders from the bushy habitats than in tree habitats (Bonte and Maelfait, 2001; Sorensen, 2003). These data would thus suggest that only selected taxa are able to adapt successfully to the extreme climatic conditions and limited habitat diversity (Pearce et al., 2003). During our study it was also noted that in spite of very harsh environmental conditions in the desert some spider were visible throughout year.

The strong dominance of Philodromideae in this study may be attributed to the vegetation composition (bushy and grassy). Abundance of individual species in different habitats may be affected by the vegetation types (Pearce *et al.*, 2003). The hand picking and plant shaking methods are more useful than the pit fall method in the desert areas due to arid conditions causing the preserving solution to evaporate too soon to capture the specimens.

		Vear 2001	Vear 2002	Vear 2003	Total	
Families	Species	F M L (E M L 4		spec	imens
		$\mathbf{F} = \mathbf{N} \mathbf{I} = \mathbf{I} \mathbf{M} \mathbf{I}$	$\mathbf{F} = \mathbf{M} = \mathbf{I}\mathbf{M}\mathbf{t}$	$\mathbf{F} - \mathbf{M} - \mathbf{Imt}$	N0.	%
Lycosidae	Evippa shivajii (Tikader & Malhotra, 1980)		56 - 14 - 0	0 - 0 - 0	70	2.33
-	Flanona puellula (Simon 1898)	0 - 0 - 0	42 - 14 - 0	0 - 0 - 0	56	1.86
	Evippa sohani (Dyal 1935)	28 - 0 - 0	28 - 0 - 0	0 - 0 - 0	56	1.86
	Aractosa mulani (Dyal 1935)	0 - 0 - 0	28 - 0 - 0	0 - 0 - 0	28	0.93
	Hippasa madrasptana (Gravely 1924)	0 - 0 - 0	14 - 0 - 0	0 - 0 - 0	14	0.47
	Hippasa partita (G. P. Cambridge 1876)	0 - 0 - 28	42 - 0 - 0	0 - 0 - 0	70	2.33
	Ocyale atlanta (Sudouin 1826)	0 - 0 - 0	28 - 0 - 0	0 - 0 - 0	28	0.93
	Lycosa poondensis (G. P. Cambridge 18/6)	0 - 0 - 0	14 - 0 - 0	0 - 0 - 0	14	0.47
	Evippa Rajastnanicus (Tikader & Malifotra, 1980) Evippa praelonginas (O. P. Cambridge 1870)	14 - 0 - 0 154 0 14	0 - 0 - 0 210 0 14	0 - 0 - 0 420 0 28	14 840	0.47
	Evippa praetongipes (O. F. – Cambridge, 1870) Evippa rubigiposa (Simon, 1885)	134 - 0 - 14 546 0 0	210 = 0 = 14 0 0 252	420 - 0 - 28	812	27.93
	Evippa Fullginosa (Sinton, 1885)	0 = 01	9 - 0 - 8	0 - 14 - 0 1 - 0 - 1	20	0.67
	Evippa Sp.1 Evippa Sp.2	0 - 0 - 0	1 - 1 - 0	1 = 0 = 1 0 = 0 = 0	20	0.07
	Evippa Sp.2 Evippa Sp.3	0 - 0 - 3	0 - 0 - 1	1 - 0 - 0	5	0.17
	Hippasa Sp.	0 - 0 - 0	0 - 0 - 0	1 - 0 - 0	1	0.03
	Evippa Sp.4	0 - 0 - 0	0 - 0 - 0	1 - 0 - 0	1	0.03
Araneidae	Cyrtophora feae (Threll 1887)	42 - 0 - 0	0 - 0 - 0	0 - 0 - 0	42	1 40
Tunicidae	Araneus bitubercula (Walckenaer 1802)	$\frac{42}{28} = 0 = 0$	28 - 0 - 0	0 - 0 - 0	56	1.46
	Neoscona theis (Walckenaer 1841)	20 0 0	28 - 14 - 0	0 - 0 - 0	42	1.40
	Neoscona Sp.	1 - 0 - 0	0 - 0 - 0	0 - 0 - 0	1	0.03
Tetragnathidae	Leucauge Sp	1 - 0 - 1	0 - 0 - 0	0 - 0 - 0	2	0.06
Eresidae	Stegodyphus Sp.	0 - 0 - 0	1 - 0 - 0	1 - 0 - 0	2	0.06
Theridiidae	Latrodectus Sp.	0 - 0 - 0	0 - 0 - 0	2 - 0 - 0	2	0.06
Zodaridae	Asceua Sp.	0 - 0 - 0	1 - 1 - 0	0 - 1 - 0	3	0.10
Clubionidae	Clubiona pashabhai (Litsinger 1992)	0 - 0 - 0	42 - 0 - 14	0 - 0 - 0	56	1.86
	Clubiona filicata (O. P Cambridge, 1874)	42 - 0 - 0	0 - 0 - 0	0 - 0 - 0	42	1.40
	Castianeir tinae (Patel & Patel 1974)	0 - 0 - 0	28 - 0 - 0	0 - 0 - 0	28	0.93
Gnaphosidae	Gnaphosa poonaensis (Tikader 1973)	56 - 0 - 112	0 - 0 - 0		168	5.59
1	Cllilepis rajasthanicus (Tikader & Gaybe 1977)	42 - 0 - 14	0 - 0 - 14	70 - 0 - 14	154	5.12
	Scotophinus maind (Simon 1905)	0 - 14 - 0	0 - 0 - 0	28 - 0 - 0	42	1.40
	Sosticus sundar (Chamvberlin 1922)	14 - 0 - 0	0 - 0 - 0	0 - 0 - 0	14	0.47
	Eilica platnigki (Tikader & Gaybe 1976)	14 - 0 - 0	0 - 0 - 0	0 - 0 - 0	14	0.47
	Scotophinus Sp.1	1 - 0 - 1	0 - 0 - 0	0 - 0 - 0	2	0.06
	Poecilochroa Sp.	3 - 0 - 1	0 - 0 - 0	0 - 0 - 0	4	0.13
	Callilepis Sp.1	1 - 0 - 1	0 - 0 - 0	0 - 0 - 0	2	0.06
	Callilepis Sp.2	1 - 0 - 1	0 - 0 - 0	0 - 0 - 0	12	0.03
	Callilaria Sp.5	2 - 0 - 2	2 - 0 - 2	2 - 0 - 2	12	0.40
	Callepis Sp.4	0 - 0 - 0 0 0 1	0 - 0 - 0	1 - 0 - 0 1 0 0	1	0.05
	Drassodas Sp.2	0 = 0 = 1 0 = 0 = 0	0 = 0 = 0 0 = 0 = 0	1 = 0 = 0 1 = 0 = 0	1	0.00
	Gnaphosa Sp.	1 - 1 - 0	0 - 0 - 0 0 - 0 - 0	1 = 0 = 0 0 = 1 = 0	3	0.05
	Scotophaeus Sp.2	0 - 0 - 0	0 - 0 - 0	0 - 1 - 0	1	0.03
Thomisidae	Amentila raanaa (Basu 1964)	14 0 0	0 0 0	0 0 0	14	0.47
THOMISIUAE	Diege kapuri (Thorell 1869	0 - 0 - 0	14 - 0 - 0	0 - 0 - 0	14	0.47
	Xysticus Sp	3 - 0 - 0	0 - 0 - 0	0 - 0 - 0	3	0.10
	Ebo emo (Tikader 1970)	0 - 1 - 3	1 - 0 - 1	5 - 0 - 1	12	0.40
	Ebo somathaii (Tikader 1965)	1 - 0 - 0	0 - 0 - 0	0 - 0 - 0	1	0.03
Philobromidae	Tibellus pashanensis (Tikader 1960)	14 - 0 - 0	14 - 0 - 0	0 - 0 - 0	28	0.93
1 million omiliae	Thantus dhakuricus (Tikader 1960)	42 - 0 - 0	0 - 0 - 0	0 - 0 - 0	42	1.40
	Philodromus decoratus (Tikader 1962)	28 - 0 - 0	0 - 0 - 0	0 - 0 - 0	28	0.93

Table I. –	Number and abundance of	arachnid species	recorded from	different habitats	s during the stud	y period	(January
	2001 – December 2003).						

Continued

Families	Species	Year 2001	Year 2002	Year 2003	T spec	otal imens
	-	$\mathbf{F} - \mathbf{M} - \mathbf{Imt}$	F – M – Imt	F – M – Imt	No.	%
Philodro	mus Sp.1	9 - 0 - 0	0 - 0 - 0	5 - 0 - 5	19	0.63
Philodro	mus Sp.2	0 - 0 - 0	0 - 0 - 0	4 - 0 - 6	10	0.33
Philodro	mus Sp.3	5 - 0 - 2	0 - 0 - 0	16 - 0 - 0	23	0.76
Philodro	mus Sp.4	1 - 0 - 0	2 - 0 - 0	1 - 0 - 0	4	0.13
Philodro	mus Sp.5	0 - 0 - 0	0 - 0 - 1	3 - 0 - 0	4	0.13
Philodro	mus Sp.6	17 - 0 - 1	7 - 2 - 4	0 - 0 - 0	31	1.03
Philodro	mus Sp.7	2 - 0 - 2	0 - 0 - 4	1 - 2 - 4	15	0.50
Philodro	mus Sp.8	0 - 0 - 0	1 - 0 - 1	1 - 0 - 1	4	0.13
Philodro	mus Sp.9	0 - 0 - 0	2 - 0 - 1	0 - 0 - 1	4	0.13
Philodro	mus Sp.10	0 - 0 - 0	0 - 0 - 0	1 - 0 - 0	1	0.03
Philodro	mus Sp.11	0 - 0 - 0	1 - 0 - 0	1 - 0 - 0	2	0.06
Philodro	mus Sp.12	1 - 0 - 0	1 - 0 - 0	1 - 1 - 1	5	0.17
Philodro	mus Sp.13	0 - 0 - 0	0 - 0 - 0	1 - 0 - 2	3	0.10
Philodro	mus Sp.14	1 - 0 - 0	0 - 0 - 0	0 - 0 - 0	1	0.03
Tibellus	Sp.1	1 - 0 - 0	4 - 0 - 1	0 - 0 - 0	6	0.20
Tibellus	Sp.2	0 - 0 - 0	0 - 0 - 0	1 - 0 - 4	5	0.17

Imt, immature; F, female; M, male.

Females were found more frequently than males which may be attributed to the time of collection during our survey. Most of our collections were limited to early mornings or during the mild and cool seasons of the year. It has been reported that desert insects are more active during the above mentioned times. (Pearce et al., 2003). During summer most of the spider webs were observed either on the undersurface of the plant leaves or beneath a bush on the ground where shade and moisture was available. Another reason for this sex ratio may be the breeding behavior of the spiders as it has been reported that breeding of spiders under the desert conditions is more common in cooler, milder conditions. The seasonal timing of the surveys may also explain the high number of immature specimens encountered (Foelix, 1982; Walker and Rypstra, 2002).

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(Received 13 July 2010, revised 9 March 2012)

Pakistan J. Zool., vol. 44(5), pp. 1440-1446, 2012.

New Record of Some Freshwater Seed Shrimps (Ostracoda: Podocopida) from Lakes of Sindh, Pakistan

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> Abstract.- Five species of seed shrimps belonging to five genera *Cypris subglobosa*, *Eucypris virens*, *Dolerocypris sinensis*, *Herpetocypris fontinalis* and *Cypridopsis obesa* have been recorded for the first time from Pakistan. All these belong to family Cyprididae, except *C. obesa*, which has been placed in family Cypridopsidae (Pennak, 2001). The specimens were collected from open water and weedy areas of Keenjhar lake, Manchar lake and Chotiari lake of Sindh.

Key words: Crustacea, lacustrine, zooplankton, ostracodes.

Ostracods are referred to as "seed shrimps" because of their shape and small size. The body is enclosed in a bivalve, calcified shell (carapace). They are found in marine, brackish and freshwaters. The order Ostracoda is divided into three suborders (i) Myodocopida (ii) Platycopida and (iii) Podocopida. First two suborders are represented in marine and members of the third are found in freshwater (Pennak, 2001). About 5000 species of Ostracodes are known from various habitats (Oliver *et al.*, 2000). Members of family Cyprididae are cosmopolitan in distribution (Bronshtein, 1988).

Ostracod fauna has been extensively studied from different areas of the world, such as USA (Henderson, 1990), USSR (Bronshtein, 1988), Europe (Meisch, 2000), Australia (McKenzie, 1971), south-east Asia (Victor and Fernando, 1982), India (Klie, 1927; Victor and Fernando, 1979, Batish, 1978, 1981) and Turkey (Dole–Ozelo *et al.*, 2001). Before 1947, Gurney (1920) described few species of seed shrimps from western Balochistan. Arora (1931) studied ostracodes from different areas of Lahore and recorded 10 species. Similarly Arora (1935) described six species from Karachi area. Kazmi (2002) also listed these species. Later on twelve more species were recorded from Lahore (Mahoon and Sultana, 1977). One species, *Stenocypris fontinalis* was reported in plankton samples from Deg Nalla near Lahore (Chowdhry *et al.*, 1986). As there is paucity of data on ostracods of Pakistan, present study was initiated.

Three lakes of Sindh, Keenjhar (Distt: Thatta), Manchhar (Distt: Dadu) and Chotiari (Distt: Sanghar) were surveyed for the occurrence of seed shrimps during 2005-2006. Random sampling was carried-out by using No. 25 plankton net (mesh size 55μ m). Some specimens were also collected from algal mates in shallow, weedy marginal areas. The samples were preserved in 5% formalin solution for later studies in the laboratory. Identification was conceded with the help of taxonomic keys (Ward and Whipple, 1976; Bronshtein, 1988; Pennak, 2001). Photographs were taken with a digital camera DCM 35 (350 K Pixels, USB 1.0, mounted on the trinocular microscope (Nikon, Eclips E-200 and Swift 300-D).

The following ostracods were recorded.

1. Cypris subglobosa (Sowerby, 1840)

Shell somewhat globular in dorsal view, length 1.19 mm, height 0.7mm, width 0.62mm, anterior and posterior margins of shell rounded, posterior portion denticulate. Surface sculptured with deep, rounded pits. Shell blackish-green in colour (Fig. 1A). Antenule and antennae natotary, with long plumose setae thoracic leg II with a long terminal spine. Collected from Chotiari lake.

2. Eucypris virens (Jurine, 1820)

Shell somewhat bean shaped, dorsal margin arched with maximum width in the middle, ventral margin straight. Length 1.97mm and height 1.06mm. Anterior margin of shell more denticulate

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(Fig.1B) then posterior margin. Shell surface covered with short setae. Antennae 1 notatory with long plumose setae. Setae of antennae II short. Furca with three spines, about 12 times as long as wide, terminal spine length 2/3 of stem. This species was collected from weedy area of Keenjhar lake during April in association with rotifers and cladocerans.

3. Dolerocypris simensis (Sars, 1903)

Shell elongated, slightly convex dorsally, ventral margin straight. Length 2.02mm, width 0.72mm. Anterior margin more rounded then posterior. Anterior margin bears marginal bristles. Shell fairly transparent, yellowish brown in colour, a yellowish green spot extends across the shell. Six, elongated muscle attachment scars visible in the centre of shell (Fig. 1C). This species was collected from turbid waters of shallow area of Manchar lake during July.



Fig. 1. A, Cypris subglobosa; B, Eucypris virens; C, Dolerocypris simensis; D, Herpetocyperis fontinalis; E, Cypridopsis obesa.

4. Herpetocyperis fontinalis (Bronshtein, 1928)

Shell elongated, surface covered with small setae. Length 1.49 mm, width 0.49 mm, height 0.54 mm. Anterior portion of shell broadly rounded, while posterior portion distinctly narrow (Fig. 1D). Dorsal margin of shell gently convex, while ventral margin slightly concave in the middle. Color of shell olive green with violet spots. Natatory Setae of antenna II not very long but reach the tip of terminal portion. This specie was collected from middle area of shallow Manchar lake (z=3m) During March, June and December.

5. Cypridopsis obesa (Brady and Robertson, 1877)

Shell roughly sub-globular, anterior side broadly rounded while posterior end truncated. Dorsal side of shell distinctly convex in the anterior half of the body, ventral margin slightly concave in the middle. Surface of shell covered with fine setae. Length 0.67 mm, width 0.4 mm and height 0.34 mm. Color of shell yellowish green. Furca rudimentary with a long flagellum (Fig. 1E). This specie was collected from Keenjhar and Chotiari lakes during summer season.

Discussion

Cosmopolitanism is the prominent feature of distribution of freshwater ostracods. Victor and Fernando (1982) while discussing the distribution in south-east Asia have shown that 14 genera of family Cyprididae were cosmopolitan and one genus each had the affinities of Palaeartic-Oriental, Australian-Ethiopian region. Five genera and species of seed shrimps recorded from Pakistan in the present study are cosmopolitan. Various modes of dispersal of resistant eggs of ostracods have been suggested, such as migratory birds (De Deckker, 1977). Wind (Meckenzee, 1971), fish (Kornicker and Solen, 1971) and rice seed transport (Victor and Fernando, 1982). In Oriental region the dispersal of ostracodes due to rice seed transport has been indicated by recorded of similar rice-field Ostracodes in wide zoogeographical areas (Neal, 1977; Victor and Fernando, 1979; Batish, 1978). Cypris subglobosa reported from Pakistan is true cosmopolitan specie. It has been reported from USA

(Fergusan, 1964), India (Victor and Fernando, 1979), Sri Lanka (Neal, 1977), Indonesia (Victor and Fernando, 1979), Japan (Okubo, 1974), China (Yunfang, 1995) and Russia (Bronshtein, 1988). C. subglobasa was originally described from fossil record of India (Sowerby, 1840). Baird (1859) asked an interesting question that how a specie which has been described as fossil can be found in living condition, when he studied the collection of some pools in Nagpur (India). The fossil C. subglobasa continued to be referred as such for more than 150 years. This was a typical case of incorrect identification. Whatley et al. (2003) studied the original fossils (described by Sowerby, 1840) in British Museum of Natural History, London and concluded that these fossils be renamed as Peraperacypretta subglobasa, so the confusion between a fossil specie and a living specie has been finally resolved.

Eucypris virens was first described from Western Europe (Jurine, 1820). It has been recorded from North America, Russia, Greenland, Iran (Brroushtein, 1988), China (Yunfang, 1995) and Sicily southern Italy (Pieri *et al.*, 2006). *Dolerocypris sinensis* was first recorded from China (Sars, 1903). This species has also been recorded from spring pools and rice fields of Samarkand, Russia (Broushtein, 1988). It has recently been described from various parts of China (Yunfang, 1995).

Herpetocypris fontinalis was first described from Russia in spring pools, where there was dense growth of Fontenalis sp. Two other species of this genus H. chevrenxi and H. reptans (also recorded from Russia) are known to be distributed in western Europe, north Africa, and south Africa (Bronshtein, 1988). H. chevreuxi and H. brevicaudatus have been recently recorded from various water bodies of (Sicily) southern Italy (Pieri et al., 2006). Cypridopsis obesa was first recorded by Brady and Roberts in 1870. Species of genus Cypridopsis are known from Australian and Oriental region. C. obesa has also been recorded from England, Germany, Switzerland and Norway (Bronshtien, 1988). Two species of this genus C. vidua and C. elonga have been recently recorded from (Sicily) southern Italy (Pieri et al., 2006). Gurney (1920) reported six species from western Balochistan,

while Arora (1931, 1935) recorded 10 species from Lahore city and six species from Karachi area respectively. Later on, one species *Stenocypris fontinalis* was reported from Lahore area (Mahoon and Sultana, 1977; Chowdhry *et al.*, 1986). As two species are common from Sindh and Balochistan, in all 21 species have already been reported from Pakistan. The present new record of five species from Pakistan (Lower Sindh) is an addition to already known freshwater ostracod fauna of Pakistan.

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(Received 8 February 2008, revised 31 January 2011)

Pakistan J. Zool., vol. 44(5), pp. 1446-1449, 2012.

Assessment of Boron in Water, Sediment and Fish Tissues of Porsuk Stream, Turkey

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> Abstract.- In this study, boron concentrations has been estimated between 2010 and 2011 in water, sediment and some cyprinid species from thirteen stations (five of them were on the Porsuk Dam Lake) of Porsuk Stream, a tributary of Sakarya River. According to our data, in general boron accumulations in

Porsuk Stream follows as sediment > fish tissues > water, respectively. The highest boron concentrations of abiotic components detected in Porsuk Stream were recorded as 2.37 mg L^{-1} in station 9 for water and as 71.8 mg kg⁻¹ in station 3 for sediment in summer season. Also the highest boron concentrations of biotic components detected in Porsuk Stream was recorded as 24.8 mg kg⁻¹ in gill tissues of *Carassius gibelio* collected from station 9.

Keywords: Boron, Porsuk Stream, Water, Sediment, Fish tissues.

Boron occurs naturally in rocks, some soils and coal. The borate content of surface water can be significantly increased as a result of wastewater discharges, because borate compounds are ingredients of domestic washing agents. Boron is an essential element for organisms, but can be toxic for aquatic and terrestrial organisms especially when accumulated in high concentrations. More than 60% of boron resources at the earth are found in Turkey (WHO, 1998; Özen *et al.*, 2009).

Fishes are widely used as bioindicator organisms in aquatic environment. Much of the element variability in fish tissues have been attributed to variability of age, life cycle and feeding habits of species (Canbek *et al.*, 2007; Özan and Kir, 2008; Uysal *et al.*, 2008).

Porsuk Stream that is one of the most important tributaries of Sakarya River, is an economically important area of the northwestern of the Central Anatolia and Porsuk Dam Lake is known as a highly polluted freshwater reservoir in Kütahya/Turkey due to industrialization and urbanization. The surroundings of this lake are also very rich in boron minerals and thermal springs. The aim of this paper was to determine the boron concentrations in water, sediment and fish tissues of Porsuk Stream (including Porsuk Dam Lake).

Materials and methods

Study area and sampling methods

The selected stations on the Porsuk Stream are shown on the map (Fig. 1). Sediment and water samples were collected seasonally from 13 stations by using Ekman grab and suitable containers. Five of them were on the Porsuk Dam Lake (4.1, 4.2, 4.3, 4.4, 4.5 stations). Significant differences of boron

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accumulations could not be detected from selected stations on the Porsuk Dam Lake so the average values of five stations were only given in the present study. Fish samples were collected seasonally (2010-2011) from four stations by using power of 220 V Honda generator and from Porsuk Dam Lake by fishing net (*Cyprinus carpio* Linnaeus, 1758; *Squalius pursakensis* Hankó, 1925; *Capoeta baliki* Turan, Kottelat, Ekmekçi ve Imamoglu 2006; *Capoeta sieboldii* Steindachner 1864; *Carassius gibelio* Bloch 1782; *Rutilus rutilus* Linnaeus 1758, *Barbus tauricus* Kessler, 1877).



Fig. 1. Porsuk Stream Basin and selected stations.

Chemical analysis

Water samples (1 liter) were taken at each sampling point and their pH adjusted at 2 with 2 ml of HNO₃. Sediment and fish samples were dried for 3 h at 105°C. Each sample (0.25 g) was placed in Pyrex reactors of a CEM Mars Xpress 5 microwave digestion unit. HClO₄:HNO₃ acids of 1:3 proportions were added in the reactor. Samples were mineralized at 200°C for 30 min. Afterwards, the samples were filtered and the volumes made up to 100 ml with ultra-pure distilled water. Element levels in samples were determined by ICP-OES (Varian 720 ES). The element analyses were recorded as means of triplicate measurements (ASTM, 1985; APHA, 1992; EPA, 1998, 2001).

Statistical analysis

Cluster analysis was done by using the Minitab 15 program and significant differences were determined by using SPSS 17.

Results and discussion

Boron values of water and sediment samples of Porsuk Stream are given in Table I. The highest B concentrations in water was found at station 9 in summer season (2.37 mg L⁻¹), the lowest B concentration were found at station 2 in winter season (0.024 mg L⁻¹). Boron concentration was not detected in summer season for station 7. The highest B concentration of sediment was determined in station 3 in summer season (73.2 mg kg⁻¹). The lowest B concentration of sediment was determined at station 6 in autumn season (2.9 mg kg⁻¹).

Table I.-Boron concentrations in water (Mg L⁻¹) and
sediment of Porsuk stream, a tributary of
Sakarya River.

<i>a.</i>	Seasons						
Stations	Spring	Summer	Autumn	Winter			
Water							
1	0.09 ± 0.00	0.0 ± 0.002	0.04 ± 0.004	0.04 ± 0.00			
2	0.08 ± 0.00	0.10 ± 0.005	0.03 ± 0.002	0.02 ± 0.00			
3	0.15 ± 0.00	0.26 ± 0.003	0.23 ± 0.04	0.06 ± 0.005			
4	0.15 ± 0.00	0.20 ± 0.0007	0.10 ± 0.004	0.07 ± 0.003			
5	0.21±0.00	0.16 ± 0.002	0.07 ± 0.008	0.07 ± 0.001			
6	0.21 ± 0.00	0.27 ± 0.004	0.07 ± 0.001	0.07 ± 0.008			
7	0.21 ± 0.00	0.23 ± 0.001	0.07 ± 0.003	0.07 ± 0.001			
8	0.22 ± 0.0	0.23±0.003	0.13±0.02	0.08 ± 0.007			
9	0.57 ± 0.00	2.37 ± 0.009	0.91 ± 0.005	0.46 ± 0.010			
Sediment							
1	12.1±2.3	48±9.6	28±10.2	24.4 ± 4.8			
2	54.8±0.6	11.4 ± 4.1	21.1±10.0	19.3±6.4			
3	21.8±0.5	73.2±1.1	71.8±8.5	65.3±4.0			
4	15.7±6.4	12.7±5.7	19.9±4.6	18.5 ± 4.0			
5	9.40±0.17	10.7±4.8	28.5±5.3	18.6 ± 3.2			
6	21.66±0.5	3.0±1.3	15.1±5.4	20.6 ± 4.4			
7	23.13±1.5	Nd	2.9±0.6	24.6±5.3			
8	13.86±0.7	7.2±3.3	12.8 ± 5.7	10.6±3.6			
9	31.13±0.1	4.9±2.2	29.8±1.0	27.33±4.9			

Mean \pm Standard Error; nd: not detected.

Based on boron concentration of water (Fig. 2A), Porsuk Stream was categorized class I (< 1 mg L^{-1}) according to Turkish Environmental Legislation for all stations except for station 9. In summer season water of station 9 was of class IV category (SKKY, 2004).

Cluster analysis was used to detect similar groups (Fig. 2) according to boron contents of water and sediment. According to water, three clusters were determined. Cluster 1 contained the stations 1 and 2, cluster 2 comprised the stations 3, 4, 5, 6, 7 and 8, cluster 3 contained the station of 9. The

Fish	Tissue			Stations		
		1	2	4	5	9
S. pursacensis	Muscle	15.1 ± 6.0	6.7±0.2	-	6.3 ± 1.1	14.7 ± 2.3
	Gill	4.5 ± 1.5	7.52 ± 5.3	-	5.8 ± 1.3	19.9 ± 6.5
	Liver	19.4 ± 1.0	12.3.±9.8	-	1.0 ± 0.0	3.6 ± 0.8
C. sieboldii	Muscle	12.0 ± 7.1	16.7±3.4	-	-	23.9 ± 0.7
	Gill	16.8 ± 4.8	24.2±5.6	-	-	23.3 ± 0.4
	Liver	11.1 ± 2.8	12.4±5.1	-	-	-
C. baliki	Muscle	3.1 ± 1.6	-	-	-	-
	Gill	15.7 ± 6.2	-	-	-	-
	Liver	nd	-	-	-	-
B. tauricus	Muscle	0.2 ± 0.1	nd	-	-	-
	Gill	1.8 ± 0.6	3.4±1.7	-	-	-
	Liver	0.4 ± 0.2	nd	-	-	-
C. gibelio	Muscle	-	-	15.3 ± 3.6	-	14.7 ± 2.8
0	Gill	-	-	15.7 ± 6.4	-	24.8 ± 3.5
	Liver	-	-	16.2 ± 4.3	-	16.1 ± 4.3
C. carpio	Muscle	-	-	23.3 ± 3.3	-	23.6 ± 1.5
	Gill	-	-	17.6 ± 4.7	-	22.1 ± 0.6
	Liver	-	-	18.7 ± 2.9	-	
R. rutilus	Muscle	_	_	3.1 ± 0.5	-	-
	Gill	-	_	2.4 ± 0.4	-	-
	Sin			2.1 = 0.1		

Table II.-Mean boron values of fish tissues (mg kg⁻¹).

-: enough sample could not be obtained

station 9 was located in the flows of Sakarya River and reflects all the discharges of Porsuk Stream. Canbek *et al.* (2007) reported that heavy metals were gradually increasing in Porsuk Stream.

Three clusters were determined according to boron contents of sediment. Cluster 1 contained the stations 4, 5, 6, 7, 8 and 9, cluster 2 comprised the stations 1 and 2, cluster 3 comprised station 3. The station 3 was located in exit of Kütahya and the agricultural, domestic and industrial wastes of Kütahya were discharged to the region.

Mean concentrations (mg.kg⁻¹) of B in different tissues (muscle, gill and liver) of fishes are given in Table II. Uysal (2011) reported that the levels of B in different cyprinid species in Porsuk Dam Lake and B accumulations in all tissues of species were below the detection limits of ICP-OES. In this study B accumulation in fish of Porsuk Dam Lake were found significantly higher than in other studies. The highest B concentration was found in gill of *C. gibelio* collected from station 9 (24.8 mg kg⁻¹). The lowest B concentration was found in muscle of *B. tauricus* (0.2 mg kg⁻¹) collected from station 1. In general, gill tissues of fishes accumulate boron higher than other tissues. Gills are the first target organs to be exposed to resuspended with sediment, so gills can be significant sites of interaction with metal ions. On the other hands, the liver has a significant role in basic metabolism (Ural *et al.*, 2011; Fernandes *et al.*, 2007).

Results of this study could not have been compared with the levels of Turkish Food Codex Standard (TGK, 2002), because there is no information about maximum permissible boron limits in fish tissues in the Turkish Standards. Fishes, living in the polluted waters may accumulate trace metals via their food chains. The accumulation of trace elements are strongly related to the feeding habitat and life style of species (Emiroğlu *et al.*, 2010; Ural *et al.*, 2011). Emiroğlu *et al.* (2010) reported higher boron concentration of water, sediment and different organisms in Seydisuyu compared to our data. Seydisuyu is also one of important tributaries of Sakarya River and is significantly affected by Kırka-Boron Works.



Fig. 2. Dendogram showing clustering of stations according to surface water (A) and sediment (B) of Porsuk stream

Conclusions

The results of our study indicate that the boron discharges of system affects the biotic components more than abiotics and cause significant bioaccumulation in fishes. Therefore, if this bioaccumulation continues unchecked, the biotic components of ecosystem including human around the basin are likely to be severally affected in a short time.

Acknowledgement

This work was supported by TUBITAK

(Project No: 109Y394). All authors would like to thank TUBITAK.

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(Received 24 March 2012, revised 23 July 2012)

Pakistan J. Zool., vol. 44(5), pp. 1450-1452, 2012.

A Study of Morphological Variations in Populations of *Euphlyctis cyanophlyctis* (Schneider, 1799) (Anura: Ranidae) From District Jamshoro, Sindh

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> Abstract.- In total twenty three adult Euphlyctis cyanophlyctis specimens of (Schneider, 1799) (Anura: Ranidae) were collected from three different but adjoining areas of District Jamshoro viz., village Sain Dino Mallah, K.B. Feeder canal and village Faiz Mohammad. The collected specimens were identified using authentic literature and variations were determined by international taxonomic characteristics. E. cyanophlyctis collected from the adjacent areas were observed to be morphologically variable from each other. The most important variable characters were dorsal body color, body length and body weight. This species is being reported for the first time from the studied area.

Key words: Skittering frog, *Euphlyctis cyanophlyctis*, morphology, variation.

Euphlyctis cyanophlyctis was first discovered and reported by Schneider in 1799. It is commonly called the Skittering frog because of its peculiar unique habit of skittering over the water surface (Das and Dutta, 1998). The skittering frog is widely distributed throughout the Pakistan. It extends from Thailand to Nepal, India, Sri Lanka, Iran and Afghanistan (Suresh and Katti, 2002). E. cyanophlyctis is highly aquatic and littoral. It inhabits different types of habitats including water pools, plains and sub-mountainous areas (Khan, 1997). The body length is about 50-60 mm from snout to vent; throat is smooth; vocal sacs are light brown; male has vocal slits under the lower jaw; dorsal side of the body is light grey, olive green or light brown, some times black with numerous scattered small smooth tubercles and irregular black spots; ventrum is white and smooth, immaculate or with dark speckling or reticulation (Khan and Tasnim, 1989). Head is moderate in size; snout is scarcely pointed; canthus rostralis indistinct; tympanum is distinct, about two third the size of the eye; interorbital space narrower than the upper eyelid; fingers are slender and pointed or slightly swollen at the tips, the first not extending beyond second; toes of hind limbs completely webbed; inner metatarsal tubercle is long, conical like a rudimentary toe (Boulenger, 1890).

The aim of this study was to investigate the intraspecific variations among *E. cyanophlyctis* populations.

Materials and methods

District Jamshoro is located on the right bank of Indus River. It covers the geographical area of 11,517 km² and embraces large number of *E. cyanophlyctis*.

Nine adult specimens of skittering frog were collected from stagnant water pond of Village Sain Dino Mallah during May 2011, eight adult specimens were collected from K.B. Feeder canal during June 2011 and six adult specimens were collected from village Faiz Muhammad during July 2011 by using an Arial Net. The specimens were identified using taxonomic literature (Das and Dutta, 1998; Dubois and Ohler, 1995; Khan and Mufti, 1995; Ford and Cannatella, 1993; Khan and Ahmed, 1987; Balletto et al., 1985). The morphometrics of collected specimens was determined by using international taxonomic characteristics. Body weight of specimens was sought in grams by using electronic balance. Where as Metric ruler and Divider were used for various length measurements in millimeters including body (snout to vent), fore limbs, hind limbs, eye diameter and tympanum diameter.

The male and female skittering frogs were identified by tympanum diameter: Larger than eye in the male frog, and equal or smaller than eye in females. In male body weight was lighter than female, and the body length was shorter than female

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Results and discussion

District Jamshoro is highly variable in soil texture and environmental conditions. It is rich in water reservoirs including river tributaries, ditches, ponds, canals and swampy field areas that contain large number of different frog species including Skittering frog.

E. cyanophlyctis has previously been reported by several researchers of Pakistan and other regions (Nauwelaerts *et al.*, 2004; Das and Dutta, 1998; Khan, 1987; Boulenger, 1890), but none of them have reported any variations within different populations of *E. cyanophlyctis* of same region.

The skittering frogs collected from Village Sain Dino Mallah had dorsal side light brown in color, entirely plane without any dark patch or mark. Thighs had very rare patches (Fig. 1). They were determined to be heaviest in body weight and largest in body length (Table I) in comparison with other skittering frogs collected from other areas.

Skittering frogs collected from village Sain Dino Mallah differs from others (Nauwelaerts *et al.*, 2004; Khan, 1987) in having no slim body with dark spots or patches on dorsal side.

The male and female specimens collected from K.B. Feeder Canal observed to be grey in dorsal body color with dark patches on all the dorsal side of the body (Fig. 1). Body weight and body length determined to be less than specimens collected from village Sain Dino Mallah (Table I).

All the specimens of this *E. cyanophlyctis* collected from Village Faiz Muhammad were light yellow dorsally with dark patches (Fig. 1). The lightest weight and shortest length of the body was also recorded from the specimens of this area (Table I).

The specimens collected from K.B. Feeder canal and village Faiz Muhammad exhibit variations from skittering frogs of other localities (Khan, 1987; Boulenger, 1890) in having dorsal side of body without tubercles, rugose and distinct rows of pores.

No yellow or white irregular longitudinal stripes found on the posterior side of thighs of any specimens of *E. cyanophlyctis* of District Jamshoro as some workers found this character in skittering frogs of other regions (Das and Dutta, 1998). Skittering frogs of closely adjacent areas show high variations in morphology as stated above however



Fig. 1. *Euphlyctis cyanophlyctis* collected from Village Sain Dino Mallah (A), K.B. Feeder (B), and Village Faiz Mohammad (C).

S. no	Sex	Body weight (g)	Body length (mm)	Fore limbs length (mm)	Hind limbs length (mm)	Eye diameter (mm)	Tympanum diameter (mm)
Sain Dino Ma 5 4	llah Male Female	35.4±0.89 38.5±1.73	52.6±2.40 58.25±2.36	22.4±1.81 24.25±0.95	30.2±1.64 34±1.15	7.4±0.54 7.5±0.57	8.8±0.44 7.25±0.5
K.B. Feeder c 3 5	anal Male Female	32±1.73 35.4±0.89	47±1.73 49.6±0.89	18.66±0.57 19.8±0.44	25.66±0.57 27.4±0.89	7.33±0.57 7.6±0.54	$\begin{array}{c} 8.66 \pm 0.57 \\ 6.8 \pm 0.83 \end{array}$
Village Faiz N 4 2	Iohammad Male Female	21.5±1 24.5±0.70	34±3.36 41±1.41	16.25±1.25 18.5±0.70	25.25±0.5 26.5±0.70	6.25±0.5 6.5±0.70	7.5±0.57 5.5±0.70

 Table I. Morphometrics of *Euphlyctis cyanophlyctis* population recorded from village Sain Dino Mallah, K.B. Feeder canal, and village Faiz Mohammad.

light intensity and atmospheric temperature of these areas are not very different from each other but difference in chemical quality of water reservoirs inhabited by different populations, difference in the food quantity and age groups may be the cause of such great variation.

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(Received 10 May 2012, revised 3 August 2012)

Pakistan J. Zool., vol. 44(5), pp. 1452-1457, 2012.

Wildlife Protection Along the Karakorum Highway in Khunjerab National Park

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Abstract.- The Karakorum Highway (KKH) which connects Pakistan and China passes through Khunjerab National Park in Pakistan. The park has extremely rich wildlife diversity. The potential adverse impacts of KKH improvement project on wildlife were analyzed with field surveys, interviews and secondary data for the period from 2009 to 2011. Protective measures were developed and used to guide highway construction. Study results indicated that 147 wildlife species exist along the KKH. Twenty-four of these have international protective value. The most obvious impact of the KKH improvement project on wildlife was habitat loss. Eleven locations of Himalayan Ibex (Capra ibex sibirica) safe passages along the KKH were identified and protected with a number of innovative construction measures and practices that proved effective.

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Key words: Biodiversity conservation; road ecology; Himalayan Ibex; Pakistan

 ${f T}$ he Pakistan-China Karakorum Highway (KKH) is an international highway connecting Kashgar (historical city located in western China) with Islamabad (capital of Pakistan). It is the only overland route between Pakistan and China, built along the historic silk route. The highway was constructed under a mutual construction program between Chinese and Pakistani Governments during 1966 to 1978. The KKH passes through some of the most famous highest mountain ranges in the world (Himalavas, Hindu Kush, and Karakorum), and is generally known as the "Eighth Wonder of the World." In October 2005, a devastating earthquake occurred, causing severe destruction along the KKH. In February 2006, Pakistan and China signed a Memorandum of Understanding which initiated the improvement of the highway between Raikot Bridge and Khunjerab Pass during first phase of project (Tao et al., 2010).

The section of the KKH from K753+800 to K811+343 (kilometer markers) bisects Khunjerab National Park (KNP). The KNP was built in 1975 with the primary objective of protecting the threatened species Marco Polo sheep (*Ovis ammon polii*) and its natural habitat. Other protected species found in the KNP include: the snow leopard (*Uncia uncia*) and the brown bear (*Ursus arctos*). These species of wildlife make the KNP one of the most important centers for biodiversity in Pakistan (Qureshi *et al.*, 2011).

The impact of highway construction on wildlife and the need to protect wildlife are becoming critical issues for zoologists throughout the world (Forman and Alexander, 1998). The primary impacts to wildlife include: road mortality (Bujoczek *et al.*, 2011), the road effect-zone (Forman and Deblinger, 2000), and habitat loss and degradation (Parris and Schneider, 2009). To date, no research on the impacts of highway construction on wildlife has been done in Pakistan. This study is focusing for the first time on methods to protect wildlife during highway construction in Pakistan.

Materials and methods

Before KKH improvement project initiation,

traffic volume of K753+800 to K811+343 was about 723 vehicles/day (2002, 24h), the width of road was 6.5m and speed of vehicles was about 20km/h. After KKH improvement project completion, traffic volume will arrive at 1363 vehicles/day in 2015, the width of road will be 8.5m and speed of vehicles will reach 30km/h.

We drove at low speed (10-20km/h) along the KKH in the KNP(about 58km) during daylight and used line transect methods for recording all wildlife species visually identified using binoculars and a high powered telescope. From June-September 2009-2011, we investigated at least once each year, 3 days for each time, from 10am to 6.00pm every day. Width of the transect is about 200m. Meantime we searched for any wildlife fatality on the highway. If a dead animal was observed on the highway, we stopped to record the species, number of individuals, and also took photos. The study was restricted to amphibians, reptiles, birds and mammals. The data were systematically recorded on standard sheets designed for this study.

Along KKH in KNP the plant coverage is very low, only on the bottom of valley there are some spares plant communities distributed along KKH, including *Myricaria elegans, Ephedera intermedia, and Salix* spp. All of these plant species are palatable and favorite for many protective species, such as Marco Polo sheep, Ibex (Qureshi *et al.*, 2011). The specific kilometer markers of start point and end point of plant community along KKH were distinguished through design drawing provided by China Road and Bridge Corporation.

Concerning the protective significance of plant community along these road sections, we worked with highway engineers together to provide creative adjusted designs of ecological drainage ditch. Meantime, we provided the construction regimes of ecological drainage ditch.

In addition, we interviewed the policeman responsible for security matters in this area. The policeman is stationed along the KKH in the KNP and is responsible for inspecting and protecting the biodiversity, year-round, except during periods of heavy snowfall. The interviewed question mainly concentrated on species, number, time, location of wildlife emerged on roadside, crossing highway, road kill by vehicles etc. Since the natural habitat and wildlife species were similar between the KNP and a number of natural reserves in Xinjiang Uygur Autonomous Region of China, we consulted wildlife biologists from the Xinjiang Uygur Autonomous Region and conducted a review of pertinent Chinese scientific publications.

Results

According to our surveys and preliminary classification results: there are 22 orders \Box 51 families, 105 genera and 147 species of wild animals distributed along the KKH. These include: 8 species of reptiles, 103 species of birds and 36 species of mammals. Notably, 24 of these species are listed as international precious, endangered, protective significance (Table I).

 Table I. Valuable/Significant and endangered wildlife species along KKH in KNP.

English name	Scientific name	IUCN	CITES
Black kite	Milvus korschun(migrans)	VU	II
Sparrow-hawk	Accipiter nisus		II
Crested goshawk	Accipiter trivirgatus		II
Golden eagle	Aquila chrysaetos		II
European black vulture	Aegypius monachus		II
Griffon vulture	Gyps fulvus		II
Himalayan griffon	Gyps himalayensis		п
Bearded vulture	Gypaetus barbatus		II
Marsh harrier	Circus aeruginosus		II
Sakar falcon	Falco charrua		п
Lesser kestrel	Falco naumanni		п
Peragrine falcon	Falco paragripus		T
Common kestrel	Falco tinnunculus		n i
Common Restrer	Tuco innuncuius		п
Tibetan snowcock	Tetraogallus tibetanus		I
Great eagle-owl	Bubo bubo		II
Indian wolf	Canis lupus	VU	II
Dhole	Cuon alpinus		II
Brown bear	Ursus arctos		I
Pallas'cat	Felis manul	LR	II
Lynx	Lynx lynx		II
Snow leopard	Úncia uncia	EN	Ι
Kiang	Equus kiang	DD	11
Marco polo sheep	Ovis ammon polii	VU	I&II
Blue sheep	Pseudois navaur	LR	
······································			

IUCN, EN, Endangered; VU, Vulnerable; LR, Lower Risk; DD,Data Deficient.

CITES: Appendix I - I, Appendix II - II

We did not observe the road mortality along the KKH. During our field survey, raptors were seen

flying in the sky and stopping near the KKH. Himalayan Ibex were found feeding on vegetation along KKH and drinking water in the Khunjerab River. From our observations, it appears wildlife survive in close proximity to the KKH.

It is estimated that at least 10,600 m^2 habitat will be lost to road improvement construction. We selected 11 locations of preferred habitat/safe passages for Himalaya Ibex along the KKH as key protective areas (Table II). These locations were improved for Himalaya Ibex use by modifying the highway cross-section designs and optimizing construction regimes (Figs. 1, 2). For the perfection of cross-section designs, we selected K757+550-750, K759+500-K760Dehee Valley, K766+200-600, K776-K777 to adjust original designs without influencing the safety and drainage function of road, means improved from concrete rectangular ditch to pateriform ecological ditch (Fig. 1); for the optimizing construction regimes, five processes were provided: 1) defining the area of permanently occupied habitat and right-of-way of pateriform ecological ditch; 2) clearing the vegetation of permanently occupied habitat, while stripping humus soil 10cm of surface layer to deposit the shady environment: 3) adopting the method of artificial clearance in right-of-way of pateriform ecological ditch, making endeavor to keep original vegetation will not be disturbed and retain in the ditch, meantime stripping humus soil 10cm of surface layer to deposit the shady environment; 4) constructing the pateriform ecological ditch as soon as possible, and backfilling the humus soil; 5)watering the pateriform ecological ditch three times to keep the moist micro-habitat.



Fig. 1. Sketch map of modifying the highway cross-section designs, concrete rectangular ditch (up, original design), pateriform ecological ditch (down, improvement design)



Fig 2. Roadside vegetation was protected entirely by modifying the highway cross-section designs and optimizing construction regimes (kilometer markers K757+550 along KKH in KNP, a is before KKH improvement project, b is after)

Discussion

Many studies have found wildlife species richness is high along roadsides (Way, 1977; Wang *et al.*, 2011). In Australia, the giant green network is composed of roadside natural vegetation and has become the key habitat to protect wildlife (Bennett, 1991). Many protective species exist on KKH roadside. Consequently, it is vital to protect biodiversity during construction activities on the KKH.

The KKH is located at high altitude and is subject to snow and low temperatures. The highway grade is low and the road width is narrow. The design speed of the highway is only 30km/h. Our field records indicate traffic volume is less than 600 vehicle/day. Wildlife found along the KKH is fast moving and respond quickly to oncoming motor vehicles. As a consequence of the highway geometrics and wildlife characteristics, there appears to be no motor vehicle related mortality on the highway at this time.

The brown bear, an important protected species in the KNP, was reported in the Khunjerab Pass (Nawaz, 2007). The brown bear population was reported to have declined quickly because of noise generated by heavy vehicles operating along the KKH in the KNP (Shafiq and Ali, 1998). However, according to our field inspection, we believe the noise effect is rather limited, due to low traffic volume and low traffic speed. As the Khunjerab Pass remained opened only from May 1 to October 31; we suspect the decline of brown bear population was not entirely caused by the noise of heavy vehicles, and that there must be other factors involved. Behavioral responses to roads will have the greatest impact on species and will increase the barrier effect of a road (Eigenbrod et al., 2009). Wildlife movement across roads can be correlated with species-specific behavior characteristics, traffic flow, and environment (Forman et al., 2003). Due to the extreme conditions of the road and the arid environment along the KKH, we did not conduct quantitative research on road effect-zone for wildlife in KNP. In the future, through our primary research specific species will be selected to quantify the road effect-zone.

At this time, the impact of KKH construction on wildlife is expected to be limited to habitat loss due to increases in the highway footprint. While the population of Himalaya Ibex appears to be growing (Shafiq and Ali, 1998), the Himalaya Ibex remains the primary food source for large carnivores, such as the snow leopard (Uncia uncia) and wolf (Canis lupus). Consequently, to ensure the long-term survival of large carnivores and Marco Polo sheep in the KHP, it is vital to protect the habitat of Himalaya Ibex. Marco Polo sheep also inhabit this habitat due to vast amount of Myricaria elegans found there. Research has found Myricaria elegans is an important food source for herbivores in the area, including Marco Polo sheep (Qureshi et al., 2011). The population of Marco Polo sheep was

Kilometer markers	GPS	Altitude (m)	Width of passage (m)	Important Habitat plant (vegetation coverage %)	No. of Ibex crossing KKH (by estimate)
K753+800-1300	N36°49′06.2" E74°57′49 9"	3174	500	Myricaria elegans (35%)	30-40
K755+30-800	N36°49'37.6" E 74°58'22.0"	3200	500	Ephedra intermedia (35%)	15-20
K756+100-400	N36°50′31.8" E74°59′03.4"	3240	300	Ephedra intermedia (35%)	10-15
K757+550-750	N36°51′14.4" E74°59′13.6"	3278	200	Ephedra intermedia (15%)	20-25
K759+500-K760Dehee Valley	N36°51′44.2" E74°59′59.0"	3314	600	Salix spp. Populus afghanica, Myricaria elegans (80%)	35-40
K761+700-K762+240	N36°51′37.2" E75°01′04.4"	3338	540	Salix spp, Myricaria elegans, Ephedra intermedia (35%)	40-50
K762+740-K763+300	N36°51′30.0" E75°02′04.0"	3361	260	Salix spp, Ephedra intermedia (30%)	10-15
K766+200-600	N36°51′27.4" E75°03′49.5"	3415	600	Salix spp, Ephedra intermedia (15%)	20-25
K770+800-K771+300	N36°52′01.3" E75°06′41.5"	3490	700	Salix spp, Ephedra intermedia (15%)	30-40
K773+860-K774+050	N36°52′18.6" E75°08′22.0"	3554	400	Salix spp, Ephedra intermedia, Myricaria elegans (10%)	5-10
K776-K777Barkhon Valley	N36°52′33.8" E75°09′47.8"	3604	700	Salix spp, Ephedra intermedia (45%)	70-80

Table II.- Eleven locations of potential habitat/safe passages of Himalaya Ibex along KKH

found to have decreased quickly after recent KKH construction as compared with previous KKH construction. The main reason for the population decrease was found to be poaching by humans (Schaller, 2007). The construction of the KKH dramatically facilitates access to the national park for visitors. The need to protect wildlife in the KNP has recently attracted international attention (Schaller, 2007). Following the park management strategies employed in Canada and the USA (Eagles and McCool, 2002; Eagles *et al.*, 2001) visitors to the KNP should be limited to protect the wildlife inhabiting the KNP. Our continued studies of wildlife found along the KKH in the KNP will provide the research necessary to support this aim.

The method of modifying the highway crosssection designs and optimizing construction regimes has been used in Ji-yan expressway in Jilin Province, China successfully (Lu and Chen, 2010), by use of which along KKH in KNP, at least 4,200m² habitat/vegetation has been reserved in the pateriform ecological ditch, we recommend that this method should be spread in similar environmental condition in Northern area in Pakistan.

Acknowledgements

The authors thank Professor Abudukadir Ablimit of the Institute of Ecology and Geography of Chinese Academy of Sciences for his vital support and guidance. This research is funded by the Western China Communications Construction and Technology Project (Grant No. 2008 318 221 56) and International S&T Cooperation Program of China (Grant No. 2012DFA20980).

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(Received 16 June 2012, revised 27 August 2012)

Pakistan J. Zool., vol. 44(5), pp. 1457-1461, 2012.

Illegal Mass Killing of Indian Pangolin (*Manis crassicaudata*) in Potohar Region, Pakistan

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> Abstract.- Massive and ruthless killing of Indian pangolin (*Manis crassicaudata*) was recorded in Potohar region of Pakistan. From January 2011 to May 2012; 118 individuals were killed brutally including from districts

Chakwal (n=60), Attock (n=25), Jhelum (n=19) and Rawalpindi (n=14). Nomads and local hunters have been found directly involved in the illegal trade of the animal with a selling price of Rs.10,000-15,000/- per animal (US\$ 108 to 163) depending upon its size. The captured live pangolin is boiled in water tank to remove its scales, the rest of the scale-less dead body being thrown away. It is suspected that its scales have a high demand in the illegal local as well international markets; to be used in manufacturing bullet-proof jackets and in traditional Chinese medicines.

Key words: Pangolin, Potohar, arid environment, illegal hunting, trade.

Pangolins are inimitable mammals having rigid keratinized protective scales around their body. There are eight extant species of pangolins, restrained to Asia and Africa (Lekagul and McNeelv, 1977). The sole species that occurs in is the Indian pangolin Pakistan (Manis crassicaudata). Its populations are reported from few localities of the country, including the Potohar Plateau. This species is adapted to desert regions, locally distributed in Pakistan and prefers more barren hilly districts (Roberts, 1997). This mammal species is under massive hunting pressure due to its demand in the market (Broad et al., 1988). Its scales are believed to have traditionally medicinal importance, magical powers (Israel et al., 1987) and ornamental uses (Prater, 1980). Its flesh and fats are also used in medicines (Indian Wildlife Club Ezine, 2004), and skin to manufacture clothes and shoes (Broad et al., 1988). The animal species and its products are also traded internationally to various countries (Broad et al., 1988; Nowak, 1999).

Killing of pangolin species were reported from Sumatra, Indonesia (Sopyan, 2009) and in Peninsular Malaysia (Chin and Pantel, 2009). Data obtained from the press and enforcement authorities have shown that about 30,000 pangolins were apprehended in Southeast Asia, between the years 2000 and 2007 (Chin and Pantel, 2009) and at least about 700 skins of Indian Pangolin were exported to the USA in 1983 and 5023 skins during the 1980-1982 period (TRAFFIC, 2000). In Pakistan, traditional local ethno-medical practitioners (hakims) believe that pangolin is a valuable source

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of traditional medicines (Roberts, 1997).

Indian pangolin is a protected species and placed in Category three of the "Third Schedule" of Punjab Wildlife Acts and Rules (1975). It is also listed in Appendix-II of Convention on International Trade in Endangered Species (CITES, 2000). But in spite of its protected status it is being illegally captured, brutally killed and traded for its precious scales throughout the Potohar Plateau, which is a semi-arid zone in the country and it lies in the north part of the Punjab province. The Plateau comprises of four districts *viz.*, Jhelum, Chakwal, Rawalpindi and Attock. The total area of the Plateau including all districts is approximately 25,000 km².

Methodology

The authors collected data regarding illegal capture, killing and trade of this animal species in the study region through field visits and interviews with the local people. Interviewees belonged to different backgrounds including shop keepers, farmers, nomads, business men and students.

Results and discussion

The information generated revealed that two major groups were involved in hunting and killing of the animal species; nomads and trained local hunters. There was low number of respondents among nomads probably due to the fear that they would be reported to the authorities pertaining to the wild animals. Only seven out of twenty interviewees (7/20) admitted to hunt Indian pangolin. The others did not accept hunting but it was suspected that all respondents were actually involved in hunting as their kids were even aware of the trade and provided information about it. However, some of the local hunters in the study area were also found involved in subsistence hunting of Indian pangolin.

Results showed a total of n=118 individuals of Indian pangolin (*Manis crassicaudata*) which were recovered in the form of dead bodies, skeletons, scale jackets, live captured or reportedly killed and or traded in the Potohar Plateau during the 17 months (from January 2011 to May 2012) of the current study period (Fig.1). A total of 118 individuals of Indian pangolin have been confirmed to be captured, killed. The maximum numbers of kills have occurred in district Chakwal (n=60) at ten different sites (Table I). The most miserable incident was the recovery of n=45 scale-less decaying dead bodies of this animal species (in March 2012) which were found dumped (in jute sacks) inside an un-used railway tunnel near Chakwal city (Fig. 2 A and B). It appeared from the gross examination of the dead bodies that all those were thrown away after removing their body scales. The dead bodies also included eight (n=8) juveniles.



Fig. 1. Numbers of individuals (n=118) of Indian pangolin (*Manis crassicaudata*) which were recovered in the form of dead bodies, skeletons, scale jackets, live captured or reportedly killed during the current study period.

Another alarming incident was the transport of about 24 kg scales of Indian pangolin from Chakwal to Islamabad during February 2012 (the local grain mill owner reported where the sellers weighed the scales and handed them over to the buyers). Both the events seem to be correlated and the massive quantity of scales that were transported out of city were most probably removed from the same 45 dead bodies which were recovered from the railway tunnel in March 2012. There are published reports that Indian pangolin is mainly hunted for its scales (Nowak, 1991), which on average extracted by an adult pangolin weigh about 1 kg (Indian Wildlife Club Ezine, 2004). The wildlife office of district Chakwal was informed about the situation and few people have been arrested and being trialed in the court.

The brutal killing of Indian pangolin in Potohar region was inadvertently stumble upon in November 2011, when skeletons of six individuals along with a scale-less dead body of Indian pangolin were recovered around the huts of nomads in the

District	Place /Site	No. of dead bodies recovered(n)	No. of skeletons	No. of Scale jackets/Scales	Reported live captures(n)	Reported killings (n)
Chakwal	Mureed	01 (male)				
(n-60)	Chumbi Surla	01 (male)	-	01 kg scales	-	_
(11-00)	Thoha Mehram	02	-	-	-	_
	Tala gang	02				
	Jubairpur	05	-	-	-	-
	Sardhi, Kallar Kahar	02	-	-	-	-
	Ratta Sharif,	02	-	-	-	-
	Kallar Kahar					
	Thai,	-	-	-	-	-
	Kallar Kahar					
	Talagang city	02	-	-	-	-
	Chakwal city	-	-	24 kg scales	-	-
	Railway tunnel, Chakwal	45 (scale-less dead bodies)	-	-	-	-
Rawalpindi (n=14)	Kanyat Ladhu (Gujar Khan)	01		-	-	-
	Chakri village (motorway)	01	05	01 jacket of scales	01(baby	01
			skeletons		pangolin) + 01(female)	
	Banni gala (Islamabad)	-	-	-	03	-
Jhelum	PD khan	-	-	-	02	10
(n=19)	Diljaba Domeli		-	01 jacket of scales	-	-
	Farash Pamal (Sohawa)	01	-	-	-	-
	PD Khan	02	-	01 jacket	02	-
Attock	Dhoke Hafizabad (Jand)	-	-	-	02	-
(n=25)	Chakki (Pindi Gheb)	-	-	-	01 Juvenile	-
	Mera sharif (Pindi Gheb)	02	-	-	-	-
	Haddowali (Jand)	-	-	-	10	-
	Minimal (Jand)	-	-	-	10	-
Total (n=118)		67	05	03 Jackets	32	11

 Table I. Illegal capture and killing record of Indian pangolin (Manis crassicaudata) in Potohar Plateau of Pakistan from January 2011 to May 2012.

surroundings of village Chakri near Motorway (M-2). All animals seemed to have been killed for their scales while the dead bodies were thrown in the open which were decaying (Table I; Fig.2 C and D). Similarly, 19 individuals of this species from Jhelum and 25 individuals from Attock have been reported to be killed, and or live captured during the study period for trade purpose of its scales.

In April 2012, important information was received from China on a seizure that occurred in China where the suspect had apparently sourced the pangolin scales from Pakistan. A Chinese passenger "Zhou" was found to carry 12 bags of Pangolin scales, weighting 25.4 kg. Zhou confessed to getting the pangolin scales when he worked in Pakistan and planning to sell them in China (http://www. customs.gov.cn/publish/portal0/tab39267/info36527 6.htm). The case has been transferred to the antismuggling department of Shenzhen Bay Customs for further investigation. These recovered scales apparently seem to be the same that were collected from the Chakwal district of the study region in February 2012, where they were transported to Islamabad. This fact shows that trade in the Indian pangolin scales have become trans-national from Pakistan.

The question is why the animal species is being captured from wild and massively killed in the



Fig. 2. A. Shows a railway tunnel around Chakwal city where 45 scale-less dead bodies of Indian pangolin (*Manis crassicaudata*) were recovered which were dumped in sacs. B. Dead bodies of the animals uncovered from the sacs. C. Skeletons of six dead specimens recovered from around the huts of nomads at Chakri site (near motorway M-2) of district Rawalpindi. D. A dead body recovered from around the nomad's huts at same Chakri site, note that all body scales had been removed.

region? There are few possibilities; one of the strongest reasons is its scales. It has also been revealed from local reliable sources that scales of Indian pangolin are transported to two main cities in the country viz., Islamabad and Lahore and it is suspected that these are being used in manufacturing bullet-proof jackets. This seems probable in the scenario of terrorism in the country for the past 10 years where the demand of such jackets must have increased many folds. Besides, its scales are also being used by local practitioners for making traditional medicines, since these are believed to be aphrodisiac. The scales of Indian pangolin are used either as whole, or in powdered form to prepare traditional medicines (CITES, 2000). In Bangladesh, the species is regularly collected in hill forest areas

for its scales and possibly has disappeared in many parts mainly due to hunting (Khan, 1985; CITES, 2000). There is limited evidence of trade, either legal or illegal, in Manis crassicaudata, but very little is known about its status across its range, or how well it adapts to human threats (CITES, 2000). There is evidence that the species is being severely impacted by hunting in India and at the same time its status in Bangladesh, Pakistan, and Sri Lanka appears to be decreasing. Given the level of trade that appears to be occurring in other Asian pangolin species (especially Manis javanica and Manis pentadactyla), it is reasonable to assume that as these two species become rarer and more difficult to obtain, that more trade could shift to Manis crassicaudata (CITES, 2000).

In Pakistan, this species is protected under the Islamabad Wildlife (Protection, Preservation. Conservation, and Management) Ordinance, 1979 and the North-West Frontier Province Wildlife (Protection, Preservation, Conservation, and Management) Act, 1975 (Molur, 2008). Despite the regulations, in all the four districts of the Plateau, nomads living in the open surrounding areas of the cities and villages, and also trained hunter groups have been found to be involved in capturing and killing of Indian pangolins. They dig out the animal from its burrow, capture it and then put it in the boiling water tank/bath to remove its scales which are sold at high price. Sometimes, the live animal is sold at a rate of Rs.10,000 to 15,000 per animal (US\$ 108 to 163 per animal). The local people who cooperate with these nomads are also paid per animal.

Recommendations

The matter of illegal capture and ruthless killing of Indian pangolins in the Potohar Plateau demands immediate conservation measures, failing which the animal species may very soon be vanished from the study region. The results of the current study also suggest to some extent, reviewing the IUCN status of this animal species to change it from the category "Near Threatened (because it is likely to become endangered in the near future)" to the category "Endangered (having high risk of extinction in the wild)" (IUCN, 2012) and inclusion of this species from Appendix-II to Appendix-I of the CITES, although it requires further data from other parts of the country as well other countries.

Acknowledgement

The authors are grateful to the Higher Education Commission (HEC), Islamabad, Pakistan for providing necessary funds for the current research work.

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(Received 3 August 2012, revised 28 August 2012)

Pakistan J. Zool., vol. 44(6), pp. 1462-1464, 2012

First Record of Tibetan Lark (*Melanocorypha maxima*) in Pakistan

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> Abstract.- A 2-week long avian survey in Shimshal valley in Khunjerab National Park was conducted during July 2009 to establish the baseline about the existence, distribution and current status of different bird species. All the potential habitats were visited and 48 avian species belonging to 9 orders and 24 families were recorded. While observing the avian species in the area, a flock of eight Tibetan larks (Melanocorypha maxima) was surprisingly observed which makes the first record of Tibetan lark not only in Shimshal valley but also in the country, thus extending the distribution range of the species further westwards along the Karakorum mountain ranges and adding to the avian fauna of the country.

Key words: Khunjerab National Park, Pakistan Wildlife Foundation, Shimshal, Tibetan Lark, WWF Pakistan

The study area is a part of the Khunjerab National Park (KNP) which includes three main valleys; Khunjerab, Ghujerab and Shimshal. Khunjerab valley starts from Sost village and ends at the Khunjerab pass on Pak-China border. The Ghujerab valley which forms the main tributary of Khunjerab River provides a link between the valleys of Khunjerab and Shimshal. Shimshal valley, located in the Karakorum Mountain Ranges, starts at Shimshal village (N 36° 26' 16.5 E 75° 19' 05.8) and ends at Shimshal-Pamir Lakes about 48 km from Pak-China border (N 36° 26' 26.8 E 75° 40' 53.4). The valley is around 50 km long and covers different habitat types with different elevations ranging from 3,078 m above sea level at Shimshal

village to 4,731 m at Shimshal-Pamir Lake. The Lake is around 275 km from Gilgit city and the road conditions up to the lake include; 170 km *i.e.* up to the Passu is Karakorum Highway (KKH) metalled road, from Passu to Shimshal village around 55 km is jeep-able while from Shimshal village to the Shimshal Lake around 50 km is the pony track. It takes around 8 hours from Gilgit to reach Shimshal village while from Shimshal village to Shimshal-Pamir Lake, it is very tough, dangerous and tiresome track of around three days although the distance is just about 50 km.

Larks belong to order Passeriformes and family Alaudidae. There are overall 92 species of larks occurring worldwide (Wikipedia, 2010). Roberts (1991), Mirza (2007) and Grimmett et al. (2008) have described 17 lark species in Pakistan; all categorized as least concern by IUCN (2010). Tibetan lark (Melanocorypha maxima) has been reported from Bhutan, China and India (Wikipedia, 2010); whereas Grimmett et al. (2006) have reported it as a resident bird in Laddakh and Sikkim in India. Khan (2006) and Qureshi et al. (2011) reported 40 bird species from Shimshal including one lark species, horned lark (Eremophila alpestris). Ali and Ripley (1972) have reported 39 lark species from Indian sub-continent including India, Pakistan, Nepal, Sikkim, Bhutan and Ceylon and have described two sub-species of Tibetan lark, Melanocorypha maxima maxima (Sikkim long billed calandra lark) and Melanocorypha maxima holdereri (Ladakh long billed calandra lark). Melanocorypha maxima holdereri has been reported from Ladakh at an altitude of 4300 m whereas Melanocorypha maxima maxima has been reported from northern Sikkim and northern Bhutan in the Tibetan plateau above 3600 m. According to the available literature, Tibetan lark (Melanocorypha maxima) has never been reported from Pakistan.

Materials and methods

All the potential habitats were visited during dawn and dusk and most of the birds were identified without using binoculars. Tibetan lark was observed, first with naked eye by third author and later by first and second authors and then its morphology was studied by all using binoculars.

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Results and discussions Habitats

Shimshal valley is characterized by different habitat types including agricultural lands, dry and barren mountains, snow covered peaks, green slopes covered by small bushes, herbs and shrubs and alpine meadows. Plain areas near Shujerab Pass in the bottom of the valley have different small sized water ponds separated from the main stream. Shujerab pass is located at the top of the valley and from Shujerab pass onwards upto the Shimshal-Pamir Lake the area represents an alpine meadow which remains covered with snow around nine months in a year. July to September are the months when the white snow gets replaced by green grasses and biological activities are at the peak in the area. The peat area around the lake offers good breeding grounds for some passerine birds like horned lark. Tibetan lark was observed in this type of habitat.

Field observations

Overall 48 avian species belonging to 9 orders and 24 families were recorded during the survey with Tibetan lark recorded for the first time from the study area. A scattered flock of eight birds was observed near Gulchin Wash Top at 36° 28' 09.1 N 75° 38' 33.2 E and at an elevation of 4.623 m above sea level. Gulchin Wash Top is located near Shujerab Pass (N 36° 28' 19.5 E 75° 37' 51.6; elevation 4353 m) and about three km from Shimshal Lake (N 36° 26' 26.8 E 75° 40' 53.4; elevation 4731 m). Three field biologists observed the birds in a light snow fall near Gulchin Wash Top during their backward journey from Shimshal Lake on July 26, 2010 at 10:30 a.m. The birds were first observed by the third author and later confirmed by the first and second authors after consulting the Field Guide to the Birds of Indian Sub-continent (Grimmett et al., 2006). The birds were observed from a distance of around 30 to 40 m. Although the birds could not be trapped and photographed due to fall yet some of continuous snow their morphological characters were recorded. The size of the bird was roughly about the size of a common starling (Sturnus vulgaris). Dorsal body color was brownish with overall off-white ventral side including breast, belly and under-tail coverts. Beak was sharp, pointing and somewhat darker in color and two blackish lines were emerging towards breast from either shoulder but not joining together. Legs were also slightly dark in color. When the photographer tried to get closer to take some good quality photographs, the birds flew and landed around 60 m away. The photographer again tried to get closer but the birds flew away again and landed around 55 m away from us. This happened three times then the birds disappeared.



Fig. 1. Map of the study area: Shimshal Valley in Khunjerab National Park © WWF-Pakistan



Fig. 2. Distribution of Tibetan lark worldwide and in Pakistan (Grimmett, 2006).

Ecological linkages of Tibetan Lark

Along Shujerab Pass at an elevation 4,353 m, during a very short summer season (July to September), soft leaves and flowers of different species of grasses attract a number of insects including grasshoppers, crickets, beetles and their larvae which in turn are attractive to different birds like wagtails, redstarts and larks. Secondly, there are seven small and large sized enclosures (around 20 x 20 m^2 to $50 \text{ x} 50 \text{ m}^2$) built with stones for temporary stay during annual migration of locals both towards north in the start of summer and towards south during the start of winter. These enclosures are used by locals as a staging area for keeping thousands of yaks, sheep and goats for only a few days during migration. Following year after year migrations, the dung of yaks and droppings of sheep and goats have made a thick layer of organic matter over the soil around these enclosures which supports a number of insect species that might also be attractive for Tibetan lark in this area. Food is the basic habitat component and availability of a number of insect species in large numbers around these locations supports the existence of bird species.

Conclusion

Ali and Ripley (1972) have described the altitudinal range of *Melanocorypha maxima holdereri* between 4300 and 4600 m in Tibetan plateau and that of *Melanocorypha maxima maxima* above 3600 m in the Tibetan plateau. The species was recorded form the study area at an elevation of 4,623 m above sea level. Since detailed morphological characters of the species found in the study area could not be recorded, but based on the altitudinal range of the species in the adjoining areas in Tibetan plateau, it is presumed that the recorded species might be *Melanocorypha maxima holdereri*.

The occurrence of Tibetan lark in northern Pakistan not only adds to the avian fauna of the country but also extends its distribution range further westwards along Karakorum mountain ranges in Khunjerab National Park.

Acknowledgments

The study was sponsored by WWF-Pakistan under its Trans-Boundary Conservation Program with active participation of Forest and Wildlife Department of Gilgit-Baltistan. A number of local influential people, hunters and guides also facilitated and supported the survey team. The authors are grateful to WWF-Pakistan, officials of Forest & Wildlife Department of Gilgit-Baltistan and all those people who assisted and facilitated the survey team and sincerely acknowledge their assistance.

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(Received 11 June 2010, revised 11 August 2011)