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DIVERSITY OF ORTHOPTERA IN THE SOUTH-WESTERN KARST-REGION OF SLOVENIA WITH NOTES ON ACOUSTICS AND SPECIES IDENTIFICATION

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Dedicated to Professor Norbert Elsner and Professor Nada Gogala; Norbert Elsner initiated these excursions, organized them with high enthusiasm and took part up to 2008; we mourn his early death in 2011; Nada Gogala was a wonderful host for us and many students therefore giving a brilliant example for Slovenian hospitality; we mourn her unexpected death in 2013.

Abstract - An area around Brje pri Komnu in the Karst region of Slovenia has been visited between 1995 and 2009 usually for about 10 days in June or July. During these visits Orthoptera have been studied in some detail emphasizing species identification and acoustic behaviour. A species list is provided and – focussing on sibling species – some identification problems and aspects of biology are described. Supplementary lists of cicada and bird species encountered at the same time are provided as well.

KEY WORDS: Orthoptera, Karst, Bioacoustics, Barbitistes, Pachytrachis, Slovenia

Izvleček - RAZNOVRSTNOST RAVNOKRILCEV V JUGOZAHODNEM KRAŠKEM DELU SLOVENIJE Z ZAPISKI O AKUSTIKI IN DOLOČANJU VRST V obdobju med leti 1995 in 2009, navadno po 10 dni v juniju oz. juliju, smo obiskovali področje v okolici Brja pri Komnu na slovenskem Krasu. Raziskovali smo ravnokrilce (Orthoptera) s poudarkom na identifikaciji vrst in njihovem akustičnem vedenju. V članku predstavljamo seznam vrst, predvsem v primeru sestrskih vrst pa tudi posamezne probleme njihove identifikacije in vidike biologije. Dodajamo tudi vrstni seznam ptic in škržadov, na katere smo naleteli v času raziskav.

KLJUČNE BESEDE: Orthoptera, Kras, Bioakustika, Barbitistes, Pachytrachis, Slovenija

Introduction

Due to the constantly growing pressure of human civilization on nature, studying biodiversity has gained increasing interest during the last decades (e.g. Singh 2002, Luck 2007). A common scheme that crystallizes from many studies is - not unexpectedly – that a landscape rich in structures and plant diversity is able to house more animal species, like arthropods, than agriculturally intensely used areas (often with large areas of monocroppings; e.g. Perfecto et al. 1997, Hong-Jiao et al. 2007, Marini et al. 2008). The Karst region of Slovenia lies in the South-Western part of the country and includes a large region with predominantly rural appearance. Here, small vineyards, small farms for various crops and bushy areas or smaller forests are interspersed in a hilly landscape (see e.g. Gogala 2003). Such regions should be able to house certain richness in biodiversity - in plants as well as in animals (Tuyet 2001, Griffith et al. 2004, Clements et al. 2006). Since agriculture is rather extensive and since in the Karst region no superficial waters occur, the fields and meadows are not eutrophic and more or less dry grassland is widespread. This seems to be ideal for many Orthoptera species (e.g. Sergeev 1998, Braschler et al. 2009). We studied the occurrence of Orthoptera (= Saltatoria; grasshoppers, crickets, bushcrickets and allies) with a focus on the area around Brje pri Komnu, which we visited for yearly excursions between the years 1995 and 2009. The excursion with advanced students from the Georg-August-University in Göttingen, Germany, typically took place for ten days between mid-June and mid-July. Occasional visits at later times of the year occurred as well. The excursions focussed on acoustically active animals; therefore, in addition to Orthoptera, we also noted the presence of singing cicadas and of birds (see supplementary tables).

Material and Methods

The area visited is indicated in Fig. 1. We visited the site at the following times: June 17-25 1995, June 23–30 1996, June 21–29 1997, June 21–28 1998, July 4–11 1999 and August 7, 8 and 21 1999, June 16–25 2000, July 15-24 2001, July 18–28

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Fig. 1: Map of the Karst region of Slovenia with the gross area studied delimited by the red line (license: GURS).

2003 (extremely hot and dry year), June 18–27 2006, June 16-27 2008, July 5–14 2009. Typical views and habitats of the area are shown in Fig. 2.

Orthoptera were studied by listening to their songs and recording them as well as by catching them, determining their species identity and mostly releasing them in the field, while some were preserved for collection or later identification. For studying we used magnifying glasses (10x) and stereo-microscopes (WILD M3, WILD M5, ZEISS Stemi SV 6). Taxonomic literature with keys used for identification was Harz (1969, 1975), Us (1992), Ragge and Reynolds (1998), Bellmann (1993), Corey and Thorens (2001). Orthopteran taxonomy and systematics largely follows Eades et al.

(2015). For recording we used a set of two microphones, one specialized for lower frequencies, one for ultrasound, together with a parabolic screen from flexible PVC, amplifier and headphones (Telinga ProIII, Sweden). The microphone output was in some occasions connected to an ultrasound detector (Petterson D120, Sweden). Alternatively, we used a Brüel & Kjaer ½" condenser microphone (4130) and a portable Brüel & Kjaer amplifier type 2804. Recordings were made either on a DAT-recorder (Pioneer D-C88, 96 kHz sampling rate) or on various Mini-Disc-recorders. The recorded songs were analysed on an oscilloscope (Hameg 205) or after digitisation (Cool Edit, Syntryllium) on notebooks. Photographs were taken using a Nikon-SLR with macro-lens or a Canon digital camera (Powershot 3, S5 IS). Cicadas were identified by their songs according to Gogala (1998) and a PC-presentation kindly provided by Matija Gogala. For bird identification several standard field guides were used.

Results and discussion

List of species, with some notes on their acoustic signals and behaviour

Table 1 and 2 give a list of Orthoptera including the years, when they were encountered. Altogether we identified 58 species: 5 species of crickets, 30 species of other Ensifera (mostly bush crickets) and 23 species of Caelifera. The species observed most frequently, and also the dominant species in the majority of years (see also Fig. 3), were the cricket Melanogryllus desertus (Pallas, 1771), the phaneropterid katydids Barbitistes ocskavi (Charpentier, 1850) and Poecilimon ornatus (Schmidt, 1850), the tettigoniid bush crickets Eupholidoptera schmidti (Fieber, 1861), Bicolorana kuntzeni (Ramme, 1931), Pachytrachis striolatus (Fieber, 1853), Pholidoptera littoralis (Fieber, 1853), and Tettigonia viridissima (Linnaeus, 1758), and the acridid grasshoppers Euchorthippus declivus (Brisout de Barneville, 1848), Odontopodisma schmidti (Fieber, 1853), Omocestus rufipes (Zetterstedt, 1821) and Stenobothrus rubicundus (Germar, 1817). Further characteristic species, but typically occurring in smaller numbers, are the bush crickets Decticus verrucivorus (Linnaeus, 1758), Tettigoniidae, Barbitistes versini (Brunner, 1878) and Polysarcus denticauda (Charpentier, 1825), both Phaneropteridae, the acridid grasshopper Kisella irena (Fruhstorfer, 1921) and the pamphagid grasshopper Prionotropis hystrix hystrix (Germar, 1817). Interestingly, they represent a diverse group of species not only in their ecology and general behaviour, but also seen under the light of acoustic communication. A student report from the research camp Dekani 2004 (Koce, 2005) listed 22 species in two localities close to our research area including only one species that we did not find, namely the tettigoniid Modestana modesta (Fieber, 1853).

- *Pholidoptera littoralis* is widely distributed in the study area and typically hidden in the vegetation on or close to the ground. Singing males have been reported at maximum heights between 0.6 and 1.2 m (Keuper et al. 1986). Males can be heard all day and parts of the night and show a clear incitement to songs of other males. They usually do not sing synchronously but repeatedly in sequence with three to five males involved. These sequences (e.g. which male starts) appear to be stable at certain times, Andreas Stumpner, Nataša Stritih, Oliver Mai, Sven Bradler: Diversity of Orthoptera in the south-western Karst-region of Slovenia

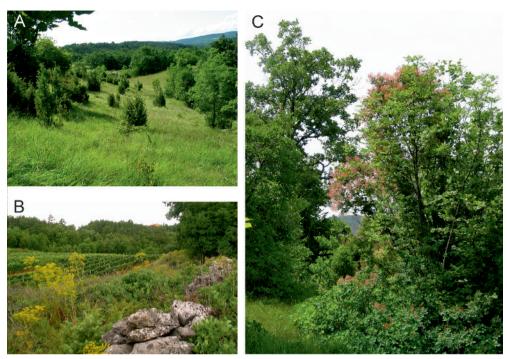


Fig. 2: Photos of typical habitats in the Karst region housing numerous Orthoptera. A) Meadows with bushes on slopes; these are typically used for making hay only. B) Vineyards with the typical stony fences around them. Also some relatively loose forests are found, in which e.g. caves with *Troglophilus neglectus* may occur or bushes with *Pholidoptera griseoaptera* or grassy areas with *Gomphocerus rufus*. C) Bushes (*Cotinus, Fraxinus*) in which the *Barbitistes* species are very typical, but also *Tettigonia viridissima* or *Eupholidoptera schmidti*.

but hierarchies, when tested with the same individuals over several days, are not stable. This behaviour appears to be common in Pholidopterini (Jones 1966) and involves some degree of aggressive rivalry and territoriality. It is also found in the more alpine relative *Pholidoptera aptera* (Fabricius, 1793), an abundant species in the Kucelj area North of the region studied here.

- *Bicolorana kuntzeni* (synonymous: *Metrioptera kuntzeni*; Fig. 3I) is mostly singing during the days. Its song spectrum lies mainly in the ultrasound. If one uses an ultrasound detector, the scenery during the day is dominated by *B. kuntzeni*'s nearly uninterrupted song (reminding of a helicopter).

- *Poecilimon ornatus* (Fig. 3 A-C) is a widespread species with reduced wings – more frequently found in the denser and slightly more humid vegetation. Males sing very short one-syllabic songs with receptive females responding (see Heller and von Helversen 1986, Helversen et al. 2001). Acoustic communication may occur over 20 m or so.



Fig. 3: Photos of various orthopteran species: A) – C) Females of *Poecilimon* ornatus (Po); D) Female of *Polysarcus denticauda* (Pd); E) Male and F) female, carrying a male of *Prionotropis hystrix* (Ph); G) Male of *Odontopodisma schmidti*; H) Female and male of *Kisella irena*; I) Female of *Bicolorana kuntzeni*; K) Male and L) – N) females of *Stenobothrus rubicundus* (Sr).

- The two *Barbitistes* species B. *ocskayi* and *B. yersini* (see Fig. 4 and below), brachypterous as well, are living in bushes and show extreme variations in density over the years (mainly occurring in June as adults). In some years they are rarely found, in other years one may find several individuals, often sitting less than 20 cm apart in each bush of the many host plants they may occur on. In both species males mainly sing during the days and early night and receptive females may reply to males many meters away.

- Stenobothrus rubicundus (synonymous: S. rubicundulus; Fig. 3K-N) is a species with mainly montane distribution, but in Slovenian Istria it also occurs down to about 250 m above NN (Koce, 2005). Its behaviour is quite spectacular, with males courting in the sun and producing song flights in which the forewings are clapped together producing loud sound (see e.g. Elsner and Wasser 1995). It occurs in high densities in meadows. Its colouration (especially among females) may vary widely from green/brown, to combinations of brown to mostly purple (Fig. 3).

- The species with the most complex song – only paralleled by songs of some cicadas or grasshoppers – is the large *Polysarcus denticauda*. This rare insect is adult early in the year. Males sing their multi-component song often also during walking (see Heller 1988, Kalmring et al. 1997). Females (Fig. 3D) can easily be confused with female *Poecilimon ornatus* (see below and Fig. 3A-C).

- Odontopodisma schmidti (Fig. 3G) and Kisella irena (Fig. 3H) are living in bushes, which is quite unusual for grasshoppers. Neither of them produces songs. *Kisella* is often seen in pairs, with mate guarding males sitting on females (Fig. 3H). Also the large and spectacular grasshopper *Prionotropis hystrix hystrix* is often encountered as such pairs in the field (Fig. 3E, F), with the considerably larger females carrying a male on the back. It is found on few places only like on exposed meadows close to Škofi, but never in large numbers. A conspicuous structure well seen on the outer cuticle of the second abdominal segment is the "Krauss' organ", which appears to have a function in stridulation (Massa 2012).

Some comments on species identification

The majority of species are easy to identify, while for some species identification is more difficult due to sibling species.

- Phaneropteridae: Barbistini: Barbitistes

Barbitistes ocskavi and B. versini are two members of the Barbitistini of great morphological similarity. The easiest criterion for species identity is the song, described in detail by Meyer and Stumpner (2000). Even though their distribution indicates slightly different preferences of the two species, we could not identify any clear criterion, what parameters differed. Both species have been found on a number of bushes, which they obviously also use as forage plants (see also Fig. 2); these are mainly Cotinus coggygria and Fraxinus ornus, but also thirteen more plant species (Asparagus acutifolius, Cornus mas, Crataegus spec., Juniperus communis, Ligustrum vulgare, Prunus mahaleb, Quercus pubescens, Paliurus spina-christi, Pistacia terebinthus, Rubus spec., Ulmus spec., Viburnum lantana, Vitis vinifera). On a single bush both species regularly occur in immediate vicinity. Nevertheless, in all the years we found no indication that hybridisation might occur – neither from morphology, nor from songs. The overall shape of the body differs in the two species, with B. versini having a more rounded back and B. ocskayi having an even to saddle like back (especially obvious in some males; Fig. 4A, D, E). The most prominent character to discriminate among the males of the two species is a protrusion of the subgenital plate, which is very prominent in B. ocskavi, but much less clearly expressed in B. versini

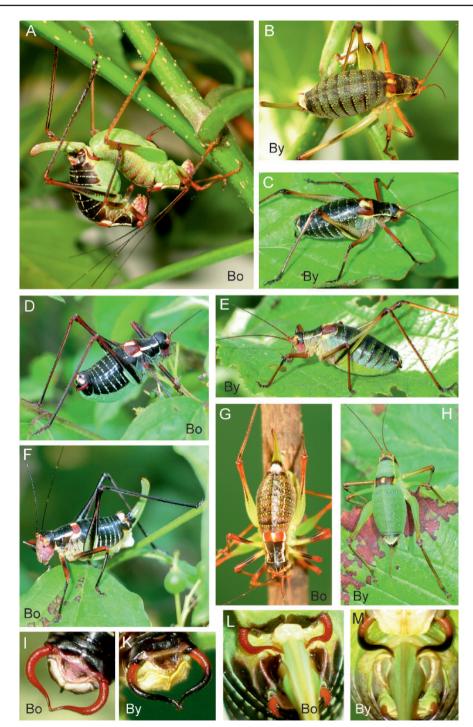


Fig. 4: Photos of *Barbitistes ocskayi* (Bo) and *B. yersini* (By) giving examples for typical differences in colouration, shape of male terminal segment and cerci. A) Precopulatory behaviour of *B. ocskayi*. The male cerci are attached to the female ovipositor and the male subgenital plate is introduced into the female genital opening. B) – E) Examples for different colouration. F) A dark *B. ocskayi* female with a spermatophore attached to its ventral base of the ovipositor. G) A light-coloured female *B. ocskayi* sitting on a male in the precopulatory phase. I), K) Tip of male abdomen with cerci (note the difference in colouration and the much more elevated protrusion on the subgenital plate in *B. ocskayi*). L), M) Attachment of the male cerci to the female base of the ovipositor in both species.

(Fig. 4I, K). This protrusion obviously has a function in precopulatory sperm removal, when the subgenital plate is introduced into the female genital opening (von Helversen and von Helversen 1991, see also Fig. 4A, L). The distal part of the male cerci, which are used for grabbing the female ovipositor at its base (see Fig. 4A, L, M), is less evenly curved on the outside in B. ocskavi than in B. versini. In addition to the body shape and colouration (see below), the posterior margin of the hindwings differs in both sexes, being rather straight in *B. ocskavi* and more curved in *B. versini* (Fig. 4 B-H). Both species vary extensively in colouration from nearly completely light green (only females) to mostly black or brown with dark green or red portions (Fig. 4 A-H). This general variation seems to be correlated with density, as in years with high density (distances of below about 1 m between individuals are frequent) darker individuals are more frequent, while in years with low density (often only single individuals are seen on one bush) lighter colours prevail (e.g. Applebaum and Heifertz 1999). This is corroborated by rearing tests with eggs collected from wild captures: these animals reared in the laboratory always were extremely dark. Nevertheless, there are some consistent differences in colouration between the two species. B. ocskayi always have a black ring or dorsal and ventral spot on the pro- and mesofemur. Proximal and distal of this spot, the femur is red in dark animals, proximally red or greenish in light animals (Fig. 4D, F, G). In mostly green individuals the dark spots may be reduced to minimal size on the ventral femur. In contrast, B. yersini does not have a separate dark spot, but the femur gets increasingly darker towards the knee. The proximal tibia is dark as well (Fig. 4B, C, E). In light green animals at least the area around the tympana is dark (Fig. 4H). Only in very dark *B. versini* the femur may be dark up to the middle area where B. ocskayi has a dark spot. All in all, the colours of B. ocskayi are brighter and more brilliant, while those of *B. yersini* are more moderate. *B. ocskayi* may have artificially looking "plastic"-green colour, most strongly expressed on the ventral abdomen (see Fig. 4A, D), which is never seen in *B. versini* whose abdomen usually is some shade of (greenish) yellow. The epiproct of *B. ocskayi* is distinctly lighter coloured as a whole than the surrounding cuticle, often nearly pure white, while the epiproct of B. yersini is often as dark as the neighbouring cuticle and lighter only at the edges; in light green animals it is more yellow to greenish (Fig. 4B, C, F-K). This is more obvious in females than in males. Light markings on wings have similar colouration as the epiproct, so that wings of *B. ocskavi* are more red with white, those of *B. versini* are more brownish

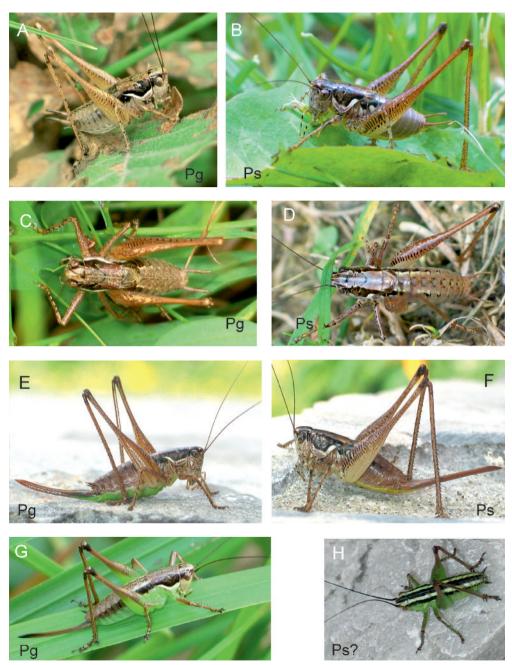


Fig. 5: *Pachytrachis gracilis* (Pg) male (A, C) and female (E, G) and *P. striolatus* (Ps) male (B, D) and female (F). H) A typically coloured smaller immature *Pachytrachis*, likely *P. striolatus*.

red with yellow (e.g. Fig. 4C, D). The antennae are annulated with alternating dark and lighter rings. In *B. ocskayi* the lighter rings are whitish (to reddish in very dark animals) and much smaller than the long darker rings. In *B. yersini* the lighter rings are always reddish and even smaller than in *B. ocskayi* and therefore less obvious.

- Phaneropteridae: Barbistini: Poecilimon ornatus/ Polysarcus denticauda

Males of the two large species have a number of obvious differences: The most prominent and eponymous for *P. denticauda* is the elongated subgenital plate standing up between the cerci. Females of the two species can more easily be confused. *P. denticauda* females are larger and have a long pronotum, which is longer than the fore tibiae. As a consequence, also the short forewings are nearly completely covered by the pronotum, while the distal part of the front wings is well visible in *P. ornatus* females. *P. denticauda* females (and males) in the study area have been always green (but are reported to vary widely with a tendency to be dark in populations of high density, e.g. Chobanov and Heller, 2010), while those of *P. ornatus* can be green to colourful green/yellow/brown to very dark (Fig. 3A-C).

- Tettigoniinae: Platycleidini: Pachytrachis

Two species of *Pachytrachis* are regularly found in the area, *P. gracilis* and *P. striolatus* (Fig. 5). There are some reliable morphological characters (see also Ingrisch and Pavićević 2010), mostly the shape of the male cerci (straight, slightly thickened at the tip in *P. gracilis*, but slightly curved, flattened in the middle in *P. striolatus*) and the shape of the female ovipositor (upwards curved in *P. gracilis*, but straight, with an oblique end – ventral tip longer – in *P. striolatus*) (see Fig. 5). Already Brunner von Wattenwyl (1861) mentioned some difference in colouration, with *P. gracilis* having a more yellowish margin on the pronotum and more uniformly coloured hind femora than *P. striolatus*. We, however, found that the easiest accessible and quite reliable character in adults is the colour of the ventral abdomen: green in *P. gracilis* and yellow in *P. striolatus*. Whether this is true also for instars, we do not know. At least larger instars of *P. gracilis* are green over most of the ventral body (see Fig. 5), but we could not reveal their species identity.

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Table 1: List of Ensifera in the Western Karst region of Slovenia. The years in which the respective species was encountered, is indicated as grey box with |v| = only larval stages found; on the right a more generalized abundance is indicated (r = rare, only single individuals encountered; += few individuals, but seen on several locations; ++ = typically more than one individual seen on one spot; +++ = many individuals seen on one spot, interindividual distance often less than 50 cm. + to +++ = in some years only few individuals, in others many; ??? means: this species may be more abundant later in the year. R, V, E1, E2 following the species names are criteria for listing endangered species (red lists of endangered species of Slovenia; URRS, št. 82/2002) with R = rare, V = vulnerable, E1 = critically endangered, E2 = strongly endangered.

Crickets:	95	96	97	98	99	00	01	03	06	09	abundance
Gryllotalpa gryllotalpa											r
Gryllus campestris											++
Melanogryllus desertus											+++
Modicogryllus (Tartarogryllus) burdigalensis											+
Oecanthus pellucens											+
other Ensifera:	95	96	97	98	99	00	01	03	06	09	
Acrometopa servillea/ macropoda (V)	1v		1v			1v			1v		+ to ++
Barbitistes ocskayi											r to +++
Barbitistes yersini											r to +++
Bicolorana kuntzeni											+++
Cyrtaspis scutata								lv			???
Decticus albifrons										1v	r
Decticus verrucivorus											+ to +++
Ephippiger discoidalis											r
Eupholidoptera schmidti (= Eupholidoptera chabrieri)											++ to +++
Leptophyes laticauda			1v			1v					r to ++
Meconema thalassinum											r
Pachytrachis gracilis											+
Pachytrachis striolatus	1v					1v			1v		++
Phaneroptera nana							1v				r
Pholidoptera fallax											r
Pholidoptera griseoaptera											+
Pholidoptera littoralis											(+ to) +++
Platycleis grisea (/albopunctata)											r
Platycleis (undetermined)				lv	lv	lv					++
Poecilimon elegans (V)										lv	+
Poecilimon ornatus (V)											r to +++

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Polysarcus denticauda								r to ++
Rhacocleis germanica								+
Ruspolia nitidula					lv		lv	+
Saga pedo (E1)		lv	lv					r
Sepiana sepium				lv		lv		+ to ++
Tettigonia viridissima								+++
Troglophilus neglectus *								r to +++
Tylopsis liliifolia (V)			lv	lv			lv	+
Yersiniella raymondi								r

* In case of *Troglophilus neglectus* (cave cricket) data relate mostly to detection of the species in an artificial tunnel close to Brje pri Komnu (about 200 m towards Gorjansko, left to the main road).

Table 2: List of Caelifera in the Western Karst region of Slovenia. For abbreviations see table 1.

Caelifera:	95	96	97	98	99	00	01	03	06	09	abundance
Aiolopus strepens											r
Anacridium aegypticum (R)											r
Arcyptera fusca											r to +
Calliptamus italicus				lv					lv		+ to +++
Chrysochraon brachyptera (many holopterous!)											++
Chorthippus biguttulus											+
Chorthippus brunneus											+
Chorthippus dorsatus											r
Chorthippus parallelus											r
Euchorthippus declivus											+++
Gomphocerus rufus											+
Kisella irena (= Miramella irena)											+
Myrmeleotettix maculatus											r
Odontopodisma fallax											r
Odontopodisma schmidti											++
Oedipoda caerulescens	lv								lv		+
Omocestus haemorrhoidalis											
Omocestus rufipes											+ to +++
Pezotettix giornae											r
Prionotropis hystrix hystrix (E2)											+
Stenobothrus lineatus											+ to ++
Stenobothrus nigromaculatus ssp. istrianus											r
Stenobothrus rubicundus (syn. S. rubicundulus)											++ to +++

Supplementary material: Table S1 gives a list of singing cicadas noted in the area

Species	in how many years detected (out of 10):
Cicada orni	5
Cicadetta tibialis (new: Cicadivetta tibialis)	1
Lyristes plebejus	5
Tettigetta brullei (new: Tettigettula pygmea)	5
Tettigetta dimissa (new: Dimissalna dimissa)	5
Tettigetta argentata (new: Tettigettalna argentata)	4
Tibicina haematodes	5

Table S2 gives a list of birds noted in the area

Species Non passerines	in how many	Song birds	(no. of years):		
	years (out of 9):	Sitta europaea	3		
Buteo buteo	8	Certhia brachydactyla	1		
Pernis apivorus	3	Aegithalus caudatus	2		
Accipiter nisus	3	Turdus merula	6		
Streptopelia decaocto	7	Turdus viscivorus	1		
Streptopelia turtur	6	Erithacus rubecula	7		
Columbus livia domestica	2	Muscicapa striata	8		
Caprimulgus europaeus	9	Phoenicurus ochruros	4		
Otus scops	6	Luscinia megarhynchos	5		
Strix aluco	1	Lanius collurio	5		
Upupa epops	6	Silvia atricapilla	6		
Cuculus canorum	5	Hippolais polyglotta	2		
Apus apus	7	Acrocephalus palustris	1		
Apus albus	1	Oriolus oriolus	9		
Dendrocopos major	5	Sturnus vulgaris	4		
Dendrocopos minor	1	Corvus cornix	5		
Picoides viridis	3	Corvus corax	7		
Jynx torquila	1	Garrulus glandarius	7		
		Pica pica	3		
Song birds	(no. of years):	Carduelis carduelis	9		
Lullula arborea	6	Carduelis chloris	6		
Motacilla alba	9	Serinus serinus	8		
Motacilla cinerea	1	Fringilla coelebs	8		
Delichon urbica	8	Coccothraustes coccothraustes	8		
Hirundo rustica	8	Emberiza cia	7		
Phylloscopus collybita	7	Emberiza cirlus	9		
Phylloscopus trochilus	1	Emberiza (Miliaria) calandra	3		
Parus major	8	Passer domesticus	6		
Parus caeruleus	8	Passer italiae (?)	5		
Parus palustris	1	Passer montanus	2		
Parus cristatus	1	L	,		
Parus ater	3				