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Diversity of Ground Beetles (Coleoptera: Carabidae) in Conventional Potato Fields in Bulgaria: Ten Years Later

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Abstract: We studied the diversity of ground beetles in a typical potato-growing region of South-Western Bulgaria for three consecutive years (2013–2015). The aim was to discuss the present data in relation to results obtained about ten years earlier for the same agricultural area. Totally, 2058 individuals of 45 species were recorded during the period of the study. *Poecilus cupreus* (Linnaeus) and *Bembidion quadrimaculatum* (Linnaeus) were the most numerous species during each of the three years of the study. In addition, *Poecilus lepidus* (Leske) was numerous in 2014 and *Lionychus quadrillum* (Duftschmid) in 2015. More than 1300 carabid specimens were collected in June 2013, while, in June 2014 and June 2015, they were less than 400. The present results are congruent with the hypothesis that the application of systemic insecticide in 2014 and 2015 affected the community of the ground beetles by elimination of larger zoophages, thus creating conditions for recolonisation by smaller zoophages, which have high dispersal capability; however, further studies are needed to test this supposition.

Key-words: ground beetles, potato crop, agricultural practices, insecticides

Introduction

Carabid beetles play an important role in agroecosystems. They represent a highly diverse insect group, with more than 40,000 species worldwide. The family is represented by more than 600 species in Central Europe (Boháč & Černý 2012), while the known species in Bulgaria are more than 720 (Guéorguiev & Guéorguiev 1995). The vast majority of ground beetles are active predators feeding on aphids, beetles (including Colorado potato beetle larvae), molluscs, hemipterans, lepidopterans and thrips, among others. They can consume up to their body weight daily. Part of them are also omnivorous or partially herbivorous (Rondon et al. 2013).

Preserving biodiversity as spatiotemporal insurance for important ecosystem services such as biological control may be critical for coping with environmental changes (Tscharntke et al. 2005). Ground beetles are successfully used as bioindicators for agroecosystem quality and crop practices application (Lemic et al. 2017). According to Avgin & Luff (2010), ground beetles have high sensitivity to anthropogenic changes of habitats as well as to environmental changes (Maelfait & Desender 1990).

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In 2000 and 2004, Kalushkov et al. (2009) studied the diversity of ground beetles in genetically modified (Bt) and conventional (non-Bt) potato fields. It was found that Bt potato did not have a negative effect on the diversity and abundance of carabid beetles. The authors concluded that the application of foliar insecticides in conventional fields did not have a negative effect either. Our aim was to study the diversity of ground beetles in the same potato region approximately ten years later.

Materials and Methods

During the years 2013, 2014 and 2015, we studied the carabid diversity in conventional potato fields located in a typical potato-producing region near Samokov (South-Western Bulgaria, 900 m a.s.l.). The whole experimental potato area was nearly 170 ha; it was divided into three plots by a road (5–6 m wide). Experiments were carried out in three plots with area of 40 ha in 2013, 80 ha in 2014 and 50 ha in 2015. Potato cultivars were VR801 and Alexia. They were irrigated using a drip pipe.

The arthropods were collected from ten pairs of pitfall traps sampled three times per season (June, July and August). Two white plastic cups, inserted next to each other, with volume of 0.5 L, height of 120 mm and opening diameter of 90 mm were used. The pitfalls contained ethyleneglycol/water (1:1) and a drop of detergent as preserving solution. The traps were laid and left for one week each month. The trap pairs were 15 m apart in three rows, each also 15 m apart, situated in the centre of the field. They were placed among the potato plants in rows.

In 2013, the foliar insecticides Calipso 480SC (thiacloprid), Dursban 4E (chlorpyrifos) and Actara 25 WG (thiamethoxam) were applied in June, and Picador 20SL (imidacloprid) and Actara were applied in July. During the next growing years 2014/2015, systemic insecticide Monceran G 370 FS (pencycuron and imidacloprid) were applied directly to the tubers during the planting. The foliar insecticides were reduced in comparison with 2013 and in July 2014, Calipso and Agria 1050 (cypermethrin and chlorpyrifos) were used, where as in June 2015 the plants were treated with Proteus 110 OD (delthamethrin and tiacloprid) and in July with Agria (Radkova et al. 2017).

All specimens were identified up to the species level (Table 1) by the first author using Hůrka (1996). The material was preserved in the collection of the National Museum of Natural History, Sofia. The dominance values of (presented in percentage shares) of a particular carabid species in the community were calculated according to Tischler (1949) for soil invertebrates, modified by Sharova (1981) into 5 categories as follows: eudominants (with degree of dominance over 10%), dominants (5 to 10%), subdominants (2 to 5 %), recedents (1 to 2 %) and subrecedents (< 1 %).

Dynamic density per 100 trap-days was calculated according to the formula Dd = (Ni/Tr*dn)*100, where 'Dd' was the dynamic density, 'Ni' was the number of specimens of the respective species, 'Tr' was the number of the traps and 'dn' was the number of trap-days.

The carabid community structure was compared by calculating Sørensen similarity index using the formula Is = (2c/a+b)*100, where 'Is' was the Sørensen index, 'c' was the number of common species obtained in both years, 'a' was the number of species from the one-year investigation, 'b' was the number of species from the investigation in another year. Coincidence of identity of 100 % meant that there was no difference between the compared species in the community, and 1 % meant that the species complex from the two observations were completely different from one another.

The data obtained were analysed using one-way analysis of variance (ANOVA) and the Tukey's post-hoc test. The calculations of the dominance values and the dynamic density were done using Excel sheets. The calculation of Sørensen similarity index and ANOVA (with the Tukey post-hoc tests) were done using PAST software, v. 3.04 (Hammer et al. 2001).

Results

Totally, 2058 individuals of 45 species were recorded during the period of the study (2013–2015, see Table 1).

In 2013, 1368 specimens of 24 species were collected. Two species were eudominants – *Poecilus cupreus* with 54.82% and *Bembidion quadrimaculatum* with 20.25%, one was dominant – *Anisodactylus signatus*, three were subdominants – *Harpalus distinguendus*, *H. rufipes* and *Microlestes seladon*, and 17 species were subrecedents (< 1%; Table 1). *Poecilus cupreus* had a dynamic density of 178.57 per 100 days (nearly two specimens per day). Four other species had dynamic density > 10.0 per 100 trap-days – *B. quadrimaculatum, H. distinguendus, H. rufipes* and *M. seladon* (Table 1).

In 2014, there were only 337 specimens of 28 species. The analysis of community structure showed the presence of three eudominants – *P. lepidus*, *P. cupreus* and *B. quadrimaculatum*, two dominants – *Microlestes minutulus* and *L. quadrillum*,

Table 1. Diversity of ground beetles in conventional potato fields during the season 2013, 2014 and 2015

N	Species	2013			2014			2015		
		n	%	Dd	n	%	Dd	n	%	Dd
Brachinini (carnivorous)										
1	*Brachinus crepitans* (Linnaeus, 1758)	10	0.73	2.38						
Carabini (carnivorous)										
2	*Carabus cancellatus* Illiger, 1798							1	0.28	0.24
3	*C. granulatus* Linnaeus, 1758				2	0.59	0.48			
4	*C. intricatus* Linnaeus, 1760							1	0.28	0.24
5	*C. violaceus* Linnaeus, 1758							2	0.57	0.48
Clivinini (carnivorous)										
6	*Clivina fossor* (Linnaeus, 1758)	4	0.28	0.95						
Bembidiini (carnivorous)										
7	*Bembidion bualei* Jacquelin du Val, 1852							11	3.12	2.62
8	*B. lampros* (Herbst, 1784)				3	0.89	0.71	4	1.13	0.95
9	*B. properans* (Stephens, 1828)	1	0.07	0.24	1	0.30	0.24	11	3.12	2.62
10	*B. quadrimaculatum* (Linnaeus, 1760)	277	20.25	65.95	72	21.36	23.34	104	29.46	24.76
11	*B. subcostatum* (Motschulsky, 1850)	6	0.44	1.43	5	1.48	1.19			
Trechini (carnivorous)										
12	*Trechoblemus micros* (Herbst, 1784)							1	0.28	0.24
13	*Trechus quadristriatus* (Schrank, 1781)	4	0.29	0.95	2	0.59	0.48	13	3.68	3.09
Harpalini (omnivorous)										
14	*Anisodactylus signatus* (Panzer, 1796)	85	6.21	2.02	1	0.30	0.24	12	3.40	2.86
15	*Harpalus affinis* (Schrank, 1781)	3	0.22	0.71	1	0.30	0.24			
16	*H. cupreus* Dejean, 1829	2	0.15	0.48						
17	*H. distinguendus* (Duftschmid, 1812)	54	3.95	12.86				3	0.85	0.71
18	*H. rufipes* (DeGeer, 1774)	68	4.97	16.19	8	2.37	1.90	5	1.42	1.19
19	*H. smaragdinus* (Duftschmid, 1812)	1	0.07	0.24	2	0.59	0.48			
20	*H. sulphuripes* Germar, 1823				3	0.89	0.71			
21	*Ophonus laticollis* Mannerheim, 1825	1	0.07	0.24						
22	*O. rufibarbis* (Fabricius, 1792)							1	0.28	0.24
Platynini (carnivorous)										
23	*Anchomenus dorsalis* (Pontoppidan, 1763)				2	0.59	0.48	2	0.57	0.48
Pterostichini (carnivorous)										
24	*Poecilus cupreus* (Linnaeus, 1758)	750	54.82	178.57	62	18.40	14.76	57	16.15	13.57
25	*P. lepidus* (Leske, 1785)				82	24.33	19.52	7	1.98	1.67
26	*P. versicolor* (Sturm, 1824)				10	2.97	2.38	10	2.83	2.38
27	*Pterostichus cursor* (Dejean, 1828)				1	0.30	0.24			
28	*Pt. melanarius* (Illiger, 1798)	12	0.88	2.86	4	1.19	0.95	2	0.57	0.48
29	*Pt. strenuus* (Panzer, 1796)				1	0.30	0.24			
Zabrini (phytophagous)										
30	*Amara aenea* (DeGeer, 1774)	6	0.44	1.43						
31	*A. apricaria* (Paykull, 1790)							6	1.70	1.43
32	*A. aulica* (Panzer, 1796)	1	0.07	0.24				3	0.85	0.71
33	*A. chaudoiri* Schaum, 1858				2	0.59	0.48	1	0.28	0.24
34	*A. fulva* (O.F. Müller, 1776)	1	0.07	0.24	3	0.89	0.71	6	1.70	1.43
35	*A. majuscula* (Chaudoir, 1850)	15	1.10	3.57						
36	*A. plebeja* (Gyllenhal, 1810)	1	0.07	0.24	1	0.30	0.24			
37	*A. serdicana* Apfelbeck, 1904	7	0.51	1.67				1	0.28	0.24
38	*A. similata* (Gyllenhal, 1810)				2	0.59	0.48			
39	*Zabrus tenebrioides* (Goeze, 1777)				2	0.59	0.48			

Table 1. Continuation

N	Species	2013			2014			2015		
		n	%	Dd	n	%	Dd	n	%	Dd
Lebiini (carnivorous)										
40	*Lionychus quadrillum* (Duftschmid, 1812)	3	0.22	0.71	26	7.72	6.19	57	16.15	13.57
41	*Microlestes minutulus* (Goeze, 1777)				28	8.31	6.67	6	1.70	1.43
42	*M. seladon* Holdhaus, 1912	55	4.02	13.10						
Sphodrini (carnivorous)										
43	*Calathus fuscipes* (Goeze, 1777)				4	1.19	0.95	7	1.98	1.67
44	*C. melanocephalus* (Linnaeus, 1758)	1	0.07	0.24	6	1.78	1.43	19	5.38	4.52
45	*Laemostenus punctatus* (Dejean, 1828)				1	0.30	0.24			
Species: Specimens:		24 species 1368 specimens			28 species 337 specimens			27 species 353 specimens		
TOTAL		45 species, 2058 specimens								

two species were subdominant – *H. rufipes* and *P. versicolor*, and 19 were subrecedents. Three species had dynamic density > 10.0 per 100 trap-days –*B. quadrimaculatum, P. cupreus* and *P. lepidus*; Table 1). For *P. cupreus*, this parameter was much lower in comparison with 2013.

In 2015, totally 353 individuals of 27 carabid species were recorded. *Bembidion quadrimaculatum, P. cupreus* and *L. quadrillum* were eudominants, one species was dominant – *Calathus melanocephalus*, five were subdominants – *Bembidion bualei, B. properans, Trechus quadristriatus* and *P. versicolor*, seven were recedents and 11 subrecedents (Table 1). The species with dynamic density > 10.0 per 100 trap-days were the eudominants *B. quadrimaculatum, P. cupreus* and *L. quadrillum*.

In the period of the study, the carnivorous ground beetles dominated (mainly members of tribes Pterostichini and Bembidiini), followed by representatives of the omnivorous tribe Harpalini and the phytophagous tribe Zabrini.

The calculation of the Sørensen index showed high similarity (61.8%) between community structure in 2014 and 2015 (Table 2). The similarity between 2013 and 2014 and between 2013 and 2015 growing seasons was over 50%, too (53.8% and 50.9%, respectively).

There was a significant difference between carabids abundance in the three years of the current survey ($F_{2,87} = 42.66$, p>0.001), mainly due to the difference in abundance, which was much higher in 2013 in comparison with the two other years (Tukey`s pair-wise comparisons: p<0.001 for both). On the other hand, no difference in abundances between 2014 and 2015 were found. Actually, the numbers of these two samples were almost the same (Tukey`s pair-wise comparisons: p=0.85).

Table 2. Similarity (Sørensen index) of carabid fauna in 2013–2015.

//////	2013	2014	2015
2013	//////	53.8 %	50.9 %
2014	53.8 %	//////	61.8 %
2015	50.9 %	61.8 %	//////

Discussion

The aim of our investigation was to describe the ground beetles diversity in conventional potato fields located in a typical potato region. The ground beetles collected in the pitfall traps were mainly carnivorous. The abundance of the carabid beetles was the highest in June and decreased in July and August. In June 2013, more than 1300 carabid specimens were caught, while in June 2014 and June 2015 – less than 400. These differences were likely due to the different insecticides used in 2013 compared to those used in 2014 and 2015.

In 2014, the traps were exposed one month after and, in 2015, two months after potatoes were planted. During this one-/two-month period, systemic insecticide probably had intoxicated the soil around potato tubers and had affected the carabid fauna. Nevertheless, the total number of species was higher (but not statistically significant) in 2014 and 2015 compared to that in 2013 (Table 1).

During a study in the same potato region in 2000 and 2004, the pitfall traps were exposed from the beginning of June till the end of August (for ten weeks in 2000 and for eight weeks in 2004) and the average number of collected specimens per week was 131 in 2000 and 168 in 2004 (Kalushkov et al. 2009). These values were similar to those we recorded in 2014 and 2015, whereas in 2013 we col-

Table 3. Carabid species collected in potato fields during the first study (2000 and 2004) and the present study (2013–2015).

Species present in both studies	Species caught only in 2013, 2014, 2015	Species caught only in 2000 and 2004
Bembidion lampros	*Brachinus crepitans*	*Leistus ferrugineus*
B. properans	*Carabus cancellatus*	*Bembidion femoratum*
B. quadrimaculatum	*C. granulatus*	*Blemus discus*
Poecilus cupreus	*C. intricatus*	*Amara equestris*
P. versicolor	*C. violaceus*	*A. eurynota*
Pterostichus melanarius	*Clivina fossor*	*A. ingenua*
Amara aenea	*Bembidion bualei*	*Harpalus autumnalis*
A. apricaria	*B. subcostatum*	*H. rubripes*
A. aulica	*Trechoblemus micros*	*Dolichus halensis*
A. majuscula	*Trechus quadristriatus*	*Microlestes maurus*
A. plebeja	*Harpalus cupreus*	
A. similata	*H. sulphuripes*	
Anisodactylus signatus	*Ophonus laticollis*	
Harpalus affinis	*O. rufibarbis*	
H. distinguendus	*Anchomenus dorsalis*	
H. rufipes	*Poecilus lepidus*	
H. smaragdinus	*Pterostichus cursor*	
Calathus fuscipes	*P. strenuus*	
C. melanocephalus	*Amara fulva*	
Microlestes minutulus	*A. chaudoiri*	
	A. serdicana	
	Zabrus tenebrioides	
	Lionychus quadrillum	
	Microlestes seladon	
	Laemostenus punctatus	

lected 1368 carabid specimens over a period of three weeks (average 456 per week). Ten species caught in 2000 and 2004 were not recorded during the present study (Table 3). The water between potato rows remained for more than 24 hours and probably had a negative effect on ground beetles. Aquino et al. (2013) evaluated the effects of an irrigation system on the community structure of arthropods in a coastal sage-scrub chaparral habitat and found that morphospecies richness, relative abundance and taxonomic representation at the ordinal level were higher at the control (non-irrigated) site than at the irrigated site.

The detailed comparison between the carabid diversity shows that the total numbers of the caught species in the period of 2013–2015 (45 species) is higher than in 2000 and 2004 (30 species). Most of the newly appeared species are subrecedents.

Ten species caught in 2000 and 2004 were not recorded during the present study (Table 3). These were *Leistus ferrugineus* (Linnaeus), *Bembidion femoratum* Sturm, *Blemus discus* (Fabricius), *Amara equestris*, *A. eurynota*, *A. ingenua*, *Harpalus autumnalis*, *H. rubripes*, *Dolichus halensis*, *Microlestes maurus*). Among the abundant species, only *Harpalus rufipes* showed a considerable difference. It was eudominant in 2000 and 2004, with 24 % and 15 %, respectively, while in 2013–2015, its representation varied between 1 % and 5 %.

Poecilus cupreus and *B. quadrimaculatum* were eudominant species during all three years of the study. In addition, *P. lepidus* was eudominant in 2014 and *L. quadrillum* was eudominant in 2015. In 2000 and 2004, the eudominant species were *P. cupreus* and *H. rufipes*, respectively (Kalushkov et al. 2009). *Poecilus lepidus* and *L. quadrillum* were not recorded in 2000 and 2004. On the other hand, *P. lepidus* was more abundant in 2014 but no specimens were collected in 2013 and only seven specimens were caught in 2015. *Harpalus distinguendus* was subdominant in 2000 (9 %) but missed in pitfall traps in 2014 and only three specimens were caught in 2015. Most probably, these differences are due to different agro-ecological conditions during both periods. In 2000 and 2004, potatoes were abundantly irrigated every two weeks and only foliar sprays were used. In 2013-2015, drip pipe system was used for irrigation and insecticides were used systemically in 2014 and 2015.

The carabid community structures monitored now and ten years earlier correspond to the descriptions by Baranová et al. (2013), where the carabid community from arable land was composed of a small number of dominant species represented by a high number of specimens. On the other hand, the community contained numerous accidental, subrecedent species, represented by a small number of specimens. The presence of accidentally occurring species represented by few individuals probably depends mostly on the environmental conditions in a particular year (Baranová et al. 2013).

According to Solga (2008), the most abundant carabid species in potato fields of Central and Northern European are *P. melanarius*, *H. rufipes* and other taxa of the genera *Bembidion* Latreille, 1802 and *Harpalus* Latreille, 1802, which usually occur in high abundances. In Hungary, *H. rufipes* and *B. crepitans* dominated potato fields (Dudas et al. 2016). In the Czech Republic, Poland, Germany, Latvia, Slovakia and Croatia, *P. cupreus* was among the dominant species (Boháč & Černý 2012, Kozewska et al. 2014, Imler 2003, Bukejs 2005, Baranová et al. 2013, Lemic et al. 2017). The same species is also among the most abundant ground beetle species in maize, beet and oilseed rape fields, too (Imler 2003, Sehnal et al. 2004, Meissle et al. 2012, Lemic et al. 2017). In 2001, we studied ground beetle diversity in potato fields in the region of Ihtiman (W Bulgaria, 600 m a.s.l, 30 km from Samokov, see Kalushkov et al. 2009). In this region, only 11 specimens (1.11 %) of *P. cupreus* were caught and dominant species were the omnivorous *H. rufipes, H. distinguendus* and *Zabrus spinipes* (Fabricius). Therefore, we believe *P. cupreus* could be used as a bioindicator for monitoring the effects (both positive and negative) of the changes related with the agricultural practices and climatic conditions in the Samokov Region.

Application of foliar pesticides has no adverse impact on the abundance of ground beetles (Kozewska et al. 2014). Ground-dwelling carabids are protected from the foliar insecticides by soil particles and damages are not serious. Therefore, they are not suitable biotic indicators of insecticide impact in potato fields (Masato et al. 2010). However, according to latter authors, larger zoophages are more sensitive to pesticide application and after their elimination, competitive interactions between predators are weaker and the rate of recolonisation by smaller zoophages, which have high dispersal capability, increases. In 2000 and 2004, we have studied the ground beetles fauna not only in conventional potato fields but also in an experimental Bt field. No negative effects on ground beetles by Bt potatoes or by the foliar insecticidal treatments applied in conventional fields have been detected.

According to the present results, we could speculate that the systematic insecticide use probably has caused adverse impacts on ground-dwelling carabid beetles but we need more studies to prove this.

In terms of biodiversity, we could conclude that the carabid community had not changed significantly in the period of ten years. Our study will contribute to future programs related to fauna conservation in Bulgaria and in Europe. *Poecilus cupreus* could be used as a bioindicator for monitoring the effects of the changes related with the agricultural practices and climatic conditions in the studied potato region.

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