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Fytotelmy rastliny *Dipsacus fullonum* ako významný kolonizačný prvok prostredia pri disperzii vírnikov

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Abstract. Phytotelmata of the plant *Dipsacus fullonum* as an important colonizing element for the dispersion of rotifers

The wild teasel (*Dipsacus fullonum*) represents an overlooked landscape element in our conditions, but from the point of view of the biodiversity of specific groups of invertebrates it is non-negligible. Unspecified representatives of the Bdelloidea order reached the highest abundance in the phytotelms of the wild teasel. We found three species from the Monogononta group (*Lecane bulla*, *Lecane inermis*, *Colurella obtusa*). In the case of *L. bulla*, a significantly higher abundance was found in the upper part of the plant, where the phytotelms volume of water was smaller than in the lower part of the plant. The species composition was the same in all monitored localities, all identified taxa are considered as a cosmopolitan.

Key words: Rotifers, Phytotelma, *Dipsacus*, Anhydrobiosis

Úvod

Telmy majú rôznorodý pôvod; prirodzený, keď vzniknú bez vplyvu ľudských aktivít, a menej často antropogénny, keď sa o ich vznik pričíní človek. Podľa toho, aké sú veľké, ako dlho sa v nich udrží voda a v akom zemepisnom pásme vznikli, sa mení aj druhové spektrum organizmov, ktoré ich osídľujú (Májsky 2022). Fytotelmy sú telmy v, alebo na rastlinách. Rastlinné telmy vznikajú najčastejšie v pazuchách listov alebo v dutých stonkách rastlín. Väčšinou ide o rastliny zo skupiny Bromeliaceae alebo rôzne krčiazkovité rastliny. Oboňa & Svitok (2012) definujú fytotelmy širšie, radia sem množstvo rastlín, ktoré nemajú stromový vzrast, no sú schopné zadržiavať dažďovú vodu, čím vytvárajú zaujímavý, často extrémny vodný ekosystém. Môžeme sem zaradiť aj rastlinu *Dipsacus fullonum* L. – štetka planá (Obrázok 1). Táto rastlina má ideálne protistojné zrástanie listy, v ktorých sa pri daždi akumuluje voda. Voda v rastline môže zotrvať niekoľko hodín až niekoľko dní, čo závisí od veľkosti telmy, vlhkosti vzduchu, teploty a ďalších faktorov. Objem listovej nádržky môže predstavovať maximálne niekoľko decilitrov. Typickými sú pre ne periodicita, prehrievanie vody, hypoxia, nedostatok potravy a taktiež nedostatok priestoru, čo zvyšuje konkurenciu a ovplyvňuje ich spoločenstvá. Keďže fytotelmy na štetkách sú závislé na zrážkach, celý ekosystém je nestabilný. Vyparovaním vody celé prostredie na určitý čas (spravidla do nasledujúcich zrážok) mizne. Ani takéto pravidelné vznikanie a zánikanie ekosystému však nebráni živočíchom obsadiť tento mikrohabitat (Kanašová et al. 2018, 2020). Ide o extrémny vodný ekosystém, ktorý si vyžaduje extrémne spôsoby prežitia. Medzi takéto zaraďujeme anhydrobiózu. Ekológia fytoteliem a organizmov žijúcich v nich je bezpochyby dôležitá. Hoci ide o mikrokozmy (Epler & Janetzky 1998) alebo extrémne malé habitaty, aj tu sa uplatňujú ekologické faktory a procesy. Veľmi dôležité a často spomínané sú disperzia, kolonizácia, medzidruhové a vnútrodrohové vzťahy a zakladateľský efekt. Kombinácia týchto procesov

a faktorov formuje štruktúru spoločenstva a rovnováhu druhového zloženia tu žijúcich organizmov. Fytotelmy sú veľmi vhodné na vedecký výskum pre rôzne svoje atribúty. Sú početné, vo väčšine majú malé rozmery a objem, ľahko odoberieme vzorky alebo ich celý objem. Nevýhoda je, že spoločenstvá organizmov v nich sú jednoduché, a tak výsledky z nich nemôžeme aplikovať na zložitejšie spoločenstvá (Maguire 1971). Výskumu fytoteliem na Slovensku sa v minulosti venovali Oboňa & Svitok (2012). Spomínajú potenciál fytoteliem v rámci šírenia rôznych chorôb a patogénov, najmä prostredníctvom vektorov ako sú komáre rôznych taxonomických skupín. Vírniky vo fytotelmách na Slovensku spomína Fogašová et al. (2022). V telmách u nás nepôvodného druhu rastliny, *Sarracenia purpurea* (L.) zistili prítomnosť pätnástich taxónov skupín Rotatoria, Ciliophora, Flagellata, Nematoda, Tardigrada a Diptera. Smith et al. (2014) sa venuje téme vírnikov v dočasných vodných masách všeobecnejšie. Zaoberá sa vyschnutými jazerami, antropotelmami, fytotelmami a mnohými ďalšími dočasnými masami vody. Hoci dočasné vody nájdeme takmer všade, líšia sa v geografickom pôvode, veľkosti, konektivitě, hydroperióde a biologickom zložení. Všetky tieto vody však po určitom období vyschnú alebo zamrznú a obnovia sa, ak sa podmienky zlepšia. Hydroperióda je v niektorých týchto biotopoch pravidelná a cyklická, v iných je sporadická. Hoci je tomu tak, vírniky sa vyskytujú aj v týchto biotopoch. Hoci sú spoločenstvá vírnikov v dočasných vodách vystavené jedinečným selekčným tlakom, prítomné druhy zdieľajú mnohé spoločné adaptačné reakcie (Smith et al. 2014). Anhydrobióza môže byť pre druhy, ktoré sú jej schopné, prospešná (Caprioli & Ricci 2005; Covino & Ricci 2005). Priemerná plodnosť vírnikov sa nikdy neznížila v dôsledku anhydrobiózy, ale je buď rovnaká alebo vyššia ako u hydratovaného vírnika. Zdá sa, že bdeloidné populácie profitujú z anhydrobiózy. Zistilo sa, že zdatnosť bdeloidných

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vírnikov klesá, ak sú populácie udržiavané hydratované počas niekoľkých generácií, avšak neklesá, ak sú populácie cyklicky vysušované. Predpokladá sa, že anhydrobióza môže byť zásadnou udalosťou pre dlhodobé prežitie bdeloidných populácií. Vysušenie spôsobuje časový posun vo veku bdeloida, ktorý nezohľadňuje čas strávený v anhydrobióze podľa modelu „Sleeping Beauty“ (Caprioli & Ricci 2005).



Obrázok 1. Fytotelma na rastline *Dipsacus fullonum*.

Materiál a metódy

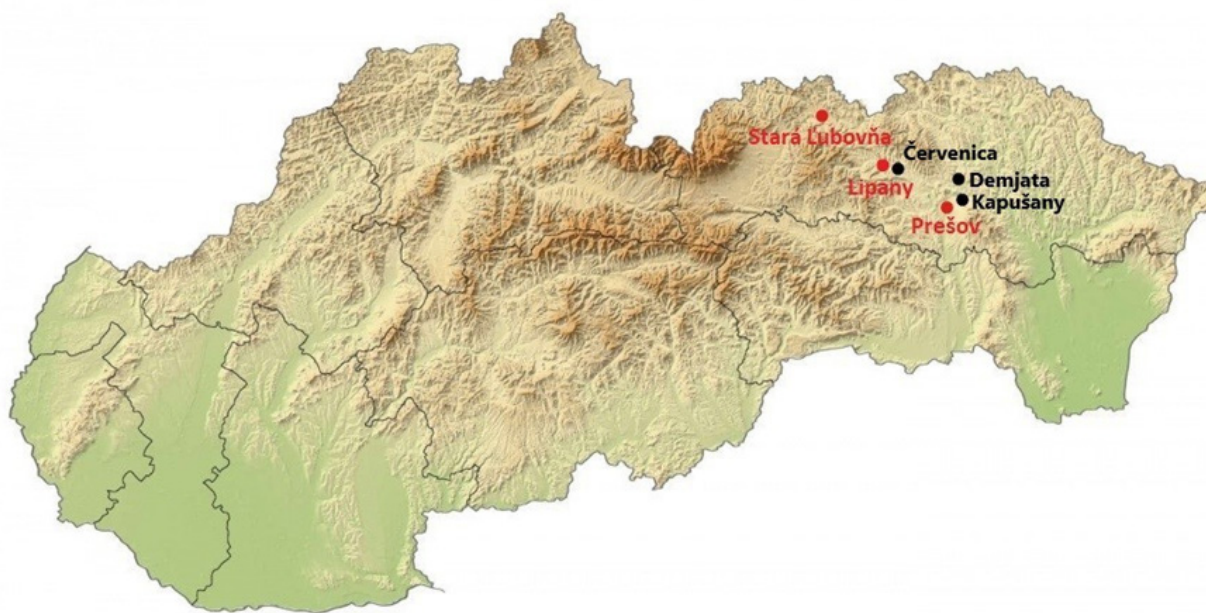
Charakteristika lokality

Odberové lokality (Obrázok 2) boli celkovo tri (Červenica, Demjata, Kapušany). Lokalita #1 Kapušany sa nachádza priamo v obci, #2 Červenica a #3 Demjata mimo obce. Rozdiel medzi najvyššie položenou lokalitou a najnižšie položenou lokalitou je ~100 metrov. Najvyššie položená lokalita bola Červenica (377 m n. m.). Najnižšia, Kapušany, dosahovala nadmorskú výšku 262 m n. m. Lokalita Demjata

dosahovala nadmorskú výšku 314 m n. m. Lokalitu Červenica, z jednej strany ohraničuje potok Hanigovianka, z druhej strany futbalové ihrisko, za ktorým je hlavná cesta do obce Hanigovce. Pri budovaní tohto ihriska vznikol medzi hracou plochou ihriska a potokom násyp zeminý vysoký ~1m. Vzorkované rastliny rástli na východnej strane tohto násypu, resp. na strane hraničiacej s potokom, nie s hracou plochou ihriska. Rastliny sa nenachádzali na celom násype, iba na časti dlhej asi 50 metrov. Lokalita Demjata sa nachádzala uprostred lesného porastu. Bola to lesná čistinka zo všetkých strán obkolesená lesným porastom. Stredom tejto čistinky prechádzali dve cesty využívané na ťažbu dreva.

Odber vzoriek a vyhodnocovania vzoriek

Odbery vzoriek vody z pazúch listov rastlín rodu *Dipsacus* sme realizovali v roku 2022, v troch termínoch (12.9., 22.9., 7.10.). Celkovo sme odobrali 60 vzoriek (v každom termíne 20 vzoriek). Vzorky vody sme odoberali v roku 2022, konkrétne v mesiacoch september a október. Vzorky boli odoberané v poludňajších až popoludňajších hodinách. Prvý odber bol ukončený až za súmraku. Vzorky sme odoberali pomocou zariadenia, ktoré pozostávalo z ručnej pumpy, zbernej nádoby o objeme 100 ml a gumenej hadičky. Hadička sa jedným koncom pripojila k zbernej nádobke uzavretej gumovým uzáverom s dvoma otvormi. Na druhý otvor gumenej zátky sa pripojila zdravotnícka striekačka s objemom 150 ml. Druhý koniec spomínanej hadičky slúžil ako nasávací otvor pre odsatie vody z pazúch rastliny. Každá rastlina bola rozdelená na hornú a dolnú polovicu. Voda z pazúch hornej polovice rastliny sa zlievala do jednej vzorky a voda z dolnej polovice rastliny predstavovala druhú vzorku. Po odsatí vody do nádoby sa táto voda preliala do odmerky s objemom 240 ml, pre stanovenie

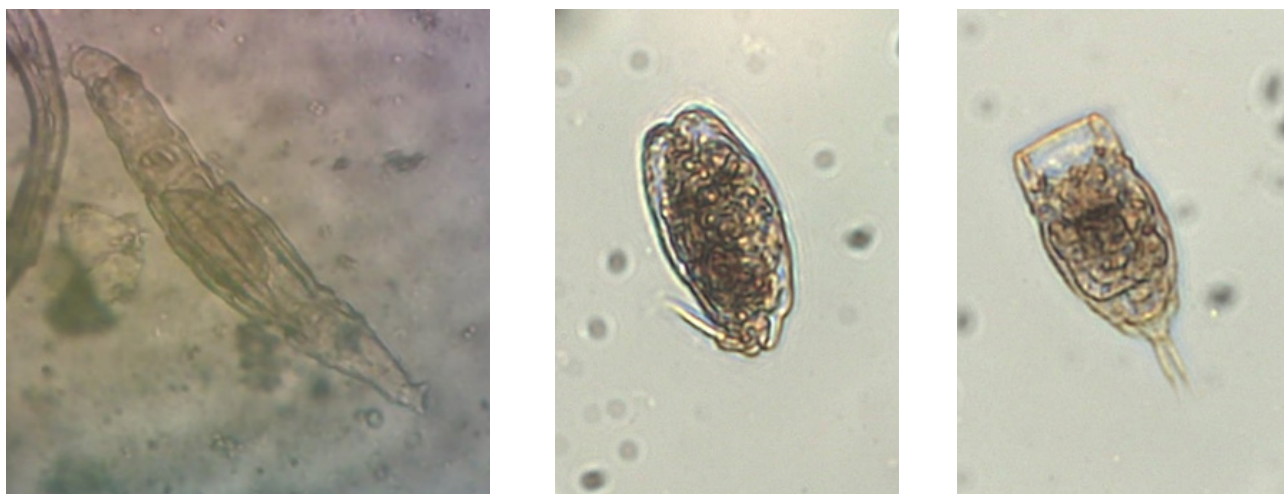


Obrázok 2. Mapa odberových lokalít (čiernou sú označené odberové lokality, červenou najbližšie väčšie sídla).

objem odobranej vzorky vody. Hodnoty sme zapísali a následne sme preliali daný objem vody do planktonickej sieťky, na spodnej strane s výpustným kohútikom. Získaná vzorky vody o objeme 5 ml bola zafixovaná 98% etanolom v pomere 2:1 (etanol:vzorka). V laboratóriu sme jednotlivé vzorky kvalitatívne a kvantitatívne taxonomicky determinovali pod mikroskopom v Kolkwitzovej komôrke prekrytej krycím sklíčkom. Pracovali sme so svetelným mikroskopom Nikon Labophot-2, vždy s tým istým zväčšením o hodnote 10/0,25. Druhovú zloženie sme určovali podľa kľúča Fauna ČSR Vířníci - Rotatoria (Bartoš 1959).

Výsledky

Pri prvom odbere dňa 12.9.2022 bola teplota vzduchu 19°C v Kapušanoch a Demjate, v Červenici 17,5 °C. Pri druhom odbere dňa 22.9.2022 bola v Kapušanoch a Demjate 12 °C, v Červenici 11°C a pri treťom dňa 7.10.2022 bola v Červenici 15°C, v Kapušanoch 12°C a v Demjate 13°C. V každej skúmanej vzorke sme zaznamenali prítomnosť vířníkov. Najnižšia početnosť bola vo vzorkách z posledného odboru vody 7.10.2022. V pazuchách listov sme celkovo našli 4 taxony vířníkov, bližšie neidentifikovaných zástupcov skupiny Bdelloidea a bežné, kozmopolitné druhy *Lecane bulla* (Gosse, 1851), *Lecane inermis* (Bryce, 1892) a *Colurella obtusa* (Gosse, 1886) (Obrázok 3).



Obrázok 3. Druhy, obávajúce fytotelmy; z ľava: Bdelloidea, *Lecane bulla*, *L. inermis*.

Tabuľka 1. Závislosť medzi početnosťou vířníkov a vybranými vlastnosťami prostredia.

Taxa	Objem (ml)	Výška rastliny	Teplota vzduchu	Nadmorská výška
Bdelloidea	-0,28*	-0,46**	0,05	-0,43**
<i>L. bulla</i>	-0,12	-0,27*	0,39**	-0,15
<i>L. inermis</i>	-0,06	-0,10	0,18	-0,27*
<i>C. obtusa</i>	-0,04	-0,27*	0,30*	-0,21

* $p < 0.05$, ** $p < 0.01$

Zistenú druhovú diverzitu (v absolútnych hodnotách, ako aj prepočítanú na 100 ml) sme vyhodnotili štatistickými metódami, a to: Spearmanovým korelačným koeficientom a neparametrickým Kruskal-Wallisovým testom. Spearmanov korelačný koeficient (Tabuľka 1) sme použili, aby sme zistili závislosť medzi početnosťou vířníkov a vybranými vlastnosťami prostredia. Neparametrický Kruskal-Wallisov test sme použili na určenie rozdielov početnosti vířníkov medzi jednotlivými lokalitami (Tabuľka 2), na určenie rozdielov početnosti vířníkov medzi rastlinami s rôznym počtom pazúch (Tabuľka 3), a na určenie rozdielov početnosti vířníkov v rôznych častiach rastlín (Tabuľka 4). Neparametrický Kruskal-Wallisov test sme použili na porovnanie rozdielov v početnosti dvoch základných skupín vířníkov medzi hornou a dolnou časťou rastliny (Tabuľka 5).

Početnosť vířníkov podľa Spearmanovho korelačného koeficientu negatívne korelovala s objemom vody vo fytotelme, s výškou rastliny, ako aj s nadmorskou výškou, a pozitívne s teplotou vzduchu. Môžeme teda tvrdiť, že so stúpajúcou teplotou vzduchu stúpala aj početnosť vířníkov vo fytotelmách. Významná negatívna korelácia vyšla najmä u skupiny Bdelloidea.

Tabuľka 2. Rozdiel početnosti vířníkov medzi jednotlivými lokalitami.

Bdelloidea		0.0002***
<i>L. bulla</i>	Medzi lokalitami	0.73
<i>L. inermis</i>		0.02*
<i>C. obtusa</i>		0.18

* $p < 0.05$, *** $p < 0.001$

Na lokalite #3 bola početnosť *Bdelloidea* signifikante najnižšia - v porovnaní s lokalitou #1 a #2. V početnosti *L. bulla* nebol štatisticky významný rozdiel medzi lokalitami. Na lokalite #3 bola početnosť *L. inermis* signifikantne najnižšia v porovnaní s ostatnými s lokalitami. V početnosti *C. obtusa* nebol medzi lokalitami signifikantný rozdiel.

Tabuľka 3. Rozdiel početnosti vírnikov medzi rastlinami s rôznym počtom pazúch.

<i>Bdelloidea</i>		0.12
<i>L. bulla</i>	Medzi rastlinami s rôznym počtom pazúch	0.85
<i>L. inermis</i>		0.96
<i>C. obtusa</i>		0.76

Kruskal-Wallisov test ukázal, že počet fytoetiem - pazúch na rastline nijako neovplyvňoval početnosť vírnikov.

Tabuľka 4. Rozdielov početnosti vírnikov v rôznych častiach rastlín.

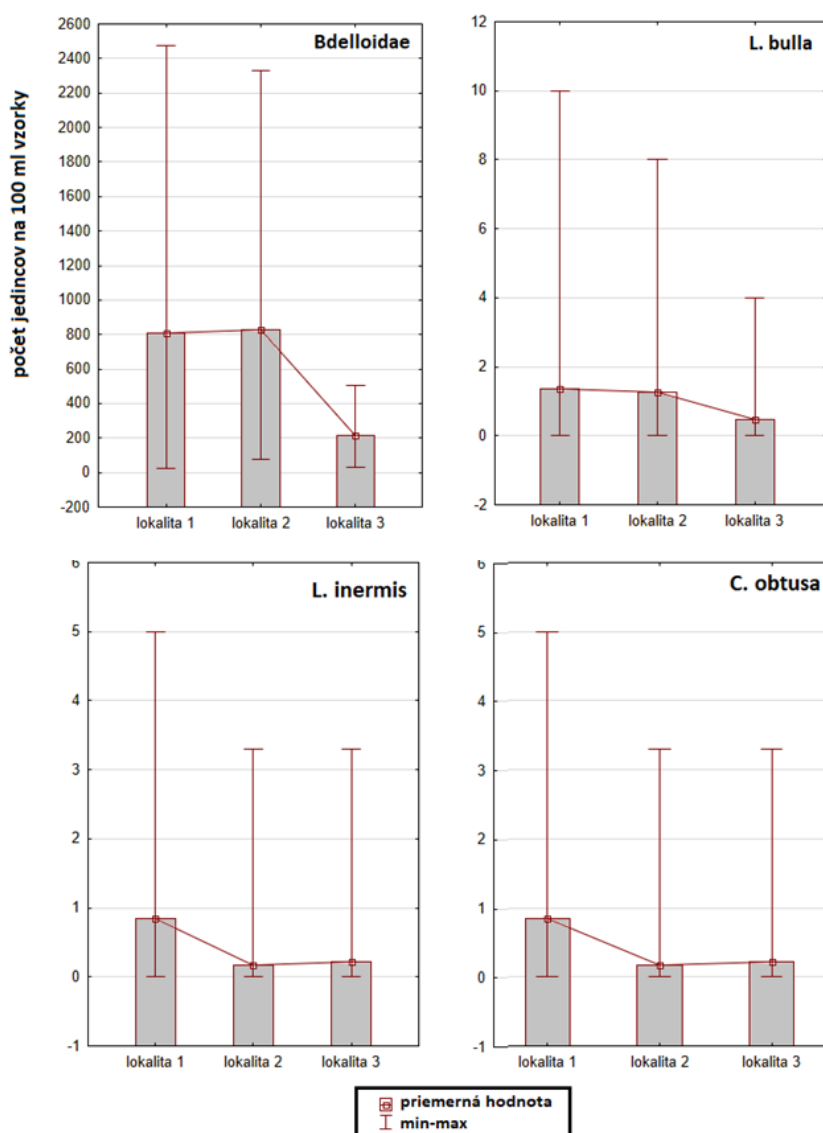
<i>Bdelloidea</i>		0.21
<i>L. bulla</i>	Medzi časťami rastlín	0.05*
<i>L. inermis</i>		0.99
<i>C. obtusa</i>		0.15

* $p < 0.05$

V prípade *L. bulla* bola signifikantne vyššia početnosť zistená v hornej časti rastliny v porovnaní s dolnou. Obrázok č. 4 prezentuje početnosť (absolútnu hodnotu) a hodnoty prepočítané na 100 ml. Najvyššiu početnosť (rádovo v stovkách) dosahovala skupina *Bdelloidea*, v absolútnych aj na 100 ml prepočítaných hodnotách. Početnosti ostatných troch druhov nedosahovali ani hodnoty 10 a vyskytovali sa len sporadicky.

Tabuľka 5 Rozdiel početnosti vírnikov v rôznych častiach rastlín.

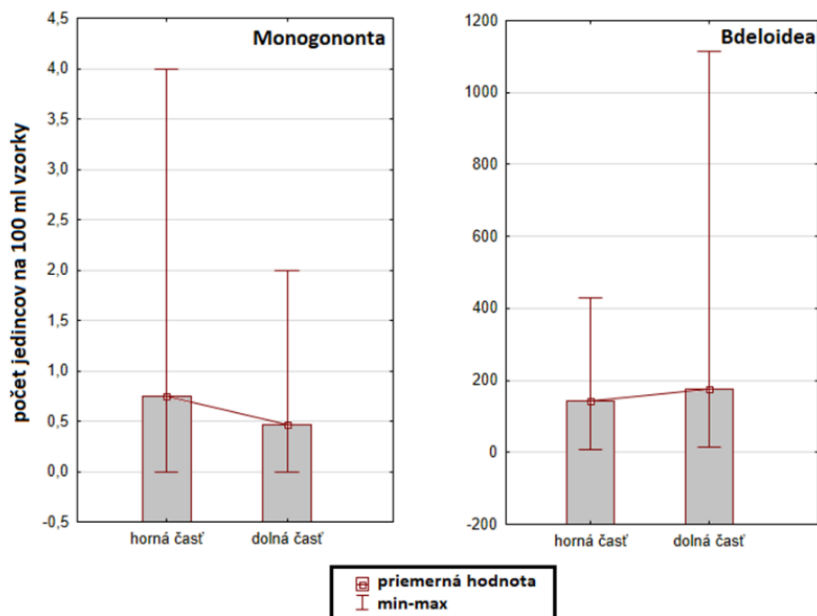
<i>Bdelloidea</i>		0.21
Monogononta	Medzi časťami rastlín	0.27



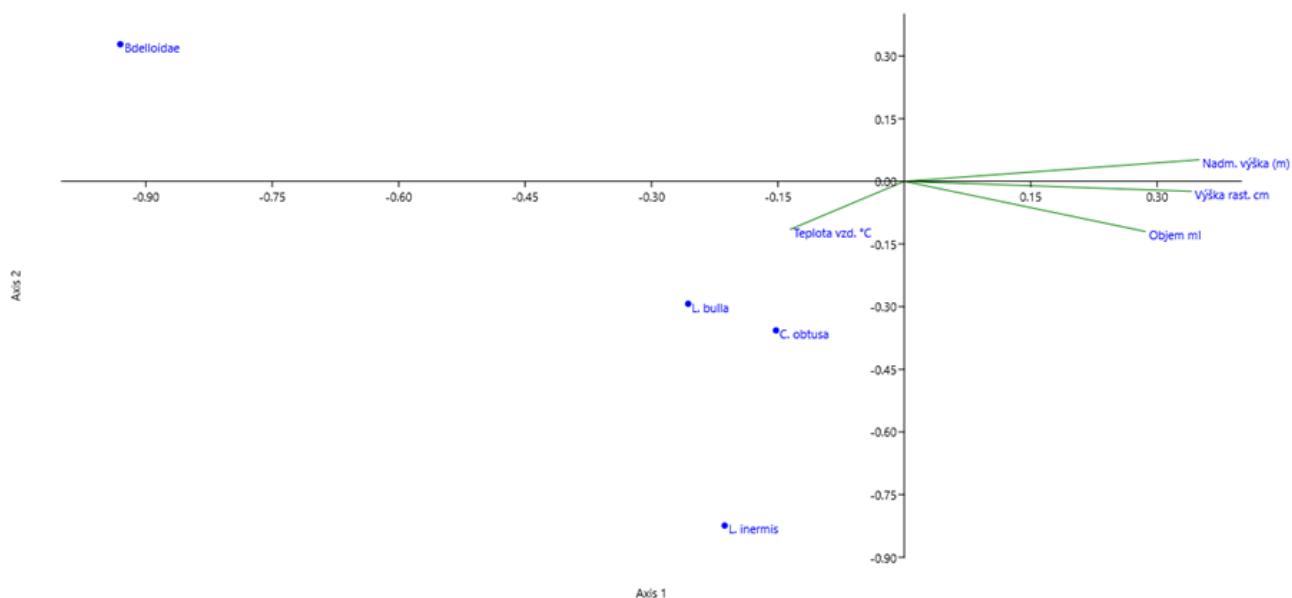
Obrázok 4. Počet jedincov na 100 ml vzorky vody a ich rozdiel medzi jednotlivými lokalitami.

Aj keď nebol preukázaný signifikantný rozdiel, na obrázku č. 5 môžeme vidieť, že čo sa týka skupiny Monogononta, početnosť bola mierne vyššia v hornej časti rastlín, zatiaľ čo početnosť Bdelloidea bola vyššia v dolnej časti rastlín. Tieto výsledky nemusia byť

smerodajné, nakoľko početnosti Monogononta boli zaznamenané v jednotkách. RDA analýza (Obrázok 6) ukázala vplyv teploty prostredia na početnosť zástupcov skupiny Monogononta.



Obrázok 5. Počet jedincov v spodnej a hornej časti rastliny zoskupených do dvoch základných skupín na 100 ml vzorky vody.



Obrázok 6. RDA analýza (závislosť medzi environmentálnymi parametrami a početnosťou zástupcov skupiny Monogononta).

Diskusia

Telmy sú charakteristické špecifickými, až extrémnymi podmienkami prostredia. Životnosť vodného útvaru, jeho kapacita a povrchová plocha vplýva na druhové zloženie

zooplanktonického spoločenstva (Ermokhin & Evdokimov 2009). Na druhej strane prostredie fytozoidov, a iných dočasných vôd môže byť pre zooplanktón výhodné, nakoľko

zooplanktón nie je vystavený tlaku zooplanktivorných rýb. No aj v týchto vodných útvaroch je určitý predачný tlak prítomný. Tento tlak vytvárajú rôzne vývinové štádiá rôznych druhov hmyzu (Brendonck et al. 2002). Predačný tlak môže ovplyvňovať početnosť vírnikov vo vzorkách, ako aj druhové zloženie zooplanktónu. Niektorí autori považujú dočasné vodné útvary za nebezpečné, pretože môžu byť rezervuármi arbovírusov (Williams 2005). Doposiaľ bolo publikovaných niekoľko prác, ktoré sa venujú rastlinám rodu *Dipsacus* a ich fytotelmám, ale nevenujú sa vírnikom, ale iným skupinám živočíchov, napr. skupine Diptera. Iné práce sa venujú fytotelmám, ale nie na rastlinách rodu *Dipsacus*, ale rastlinám skupiny Bromeliaceae alebo rastlinám rodu *Sarracenia*. Rastline rodu *Sarracenia* sa venoval výskum autorov Fogašová et al. (2022), pričom výsledky nimi publikované uvádzajú prítomnosť vírnikov vo fytotelmách tejto rastliny. Táto v našich podmienkach exotická rastlina vystavená vonkajšiemu prostrediu na Slovensku obsahovala vo svojich telmách druhy *Lecane bulla*, *L. inermis* a *Colurella obtusa*. Spomínaný výskum je príbuzný s našim tým, že bol robený na podobnej lokalite. Lokalita umiestnenia sledovaných Sarracinií bola veľmi blízko našich odberových lokalít, a od toho sa môže odvíjať aj druhové zloženie spoločenstva vírnikov vo fytotelmách. Rovnaké druhy sa vyskytovali aj vo fytotelmách rastlín *Dipsacus*. Fytotelmy v listoch tiež vytvára *Dipsacus laciniatus* (L.). Práve táto rastlina a jej fytotelmy boli skúmané v USA. Zisťovala sa prítomnosť larií a vajíčok rôznych druhov komárov. Z výsledkov vyplýva, že vo veľkom počte skúmaných rastlín a ich fytotelmi sa vajíčka alebo larvy komárov nenašli vôbec (Baumgartner 1986). Akum et al. (2001) sledovali vírniky v telmách vytvorených v opadanom ovocí. Celkovo tu našli 76 druhov vírnikov, najpočetnejšie zastúpenie mali Lecanidae a Collurellidae. Pri vyhodnocovaní našich vzoriek sme našli aj taxóny, ktoré sú schopné anhydrobiózy. Výskum Ricci (1998) potvrdzuje, že bdeloidné vírniky sú schopné anhydrobiózy. Predpokladá sa, že anhydrobiózy boli schopné všetky druhy vírnikov, niektoré druhy však túto schopnosť druhotne stratili. Medzi bdeloidnými a monogonontnými druhmi je rozdiel. Monogonontné druhy sú schopné anhydrobiózy (dormancie) len ako pokojové vajíčka, zatiaľ čo bdeloidné druhy môžu do tohto stavu upadnúť prakticky kedykoľvek počas svojho životného cyklu a úspešne sa z neho zotaviť (Ricci 2001).

Tieto výsledky ukazujú pozitívnu koreláciu medzi početnosťou vírnikov a teplotou vzduchu. RDA analýza potvrdila, signifikantný vplyv teploty na početnosť skupiny Monogononta, no nie na skupinu Bdelloidea. Môžeme to porovnať so štúdiou, ktorá sa tiež zaoberala zooplanktónom, nie však vírnikmi. Daný výskum zistil, že veslonôžky sa vyskytujú častejšie v chladnejšej vode vo fytotelmách bromélií, na rozdiel od teplejšej, slnku vystavenej vode (Lopez & Rios 2001). Na broméliách sa uskutočnil aj ďalší výskum mikrobioty obývajúcej fytotelmy tejto rastliny. Zameral sa na viacero faktorov, medzi nimi aj na teplotu vody. Výskum Antonetti et al. (2021) potvrdzuje pozitívnu koreláciu teploty vody vo fytotelmách s počtom živočíchov

v nich žijúcich. Výskum dendrotelmách, ktorý publikoval Devetter (2009) potvrdzuje pozitívnu koreláciu teploty vody a početnosti bdeloidných vírnikov. Ďalší výskum ukazuje u vírnikov lepšiu toleranciu na zvýšenie teploty vody, ako na jej zníženie. Pri prenesení do chladnejšej vody (18°C) z vody o teplote 23°C po niekoľkých hodinách žilo len asi 50 % vírnikov, pri prenesení do teplejšej vody (28°C) významný pokles početnosti nebol pozorovaný (Battaglene et al. 2000). Teplota prostredia a od nej sa odvíjajúca teplota vody môže mať signifikantný vplyv na početnosť vírnikov vo fytotelmách. Teplota je v tomto prípade faktor, ktorý vplyva na mikrohabitaty omnoho viac ako na veľké vodné masy. Výsledok nestabilnej, nízkej termickej kapacity malého objemu vody. Kruskal-Wallisovým testom sme zistili, že počet fytotelmi na rastline nijako neovplyvňoval početnosť Monogononta ani Bdelloidea. Toto zistenie bolo v rozpore s našou hypotézou, podľa ktorej viac fytotelmi znamená väčší objem vody, väčší životný priestor, stabilnejšie ekologické podmienky a viac vírnikov. V prípade porovnania počtu vírnikov v hornej a dolnej časti vyšli buď štatisticky nevýznamné rozdiely, resp. pri *L. bulla* bola väčšia početnosť v hornej časti rastliny. Toto znovu vyvracia predpoklady, že v dolnej časti rastliny by mala byť početnosť vírnikov vyššia. Tieto predpoklady sa zakladajú na skutočnosti, že v dolnej časti rastliny sú zvyčajne väčšie listy, tým aj väčšie fytotelmy, v ktorých je väčší objem vody. Mali by teda vytvárať viac životného priestoru pre vírniky. Bledzki & Ellison (1998) naznačujú, že spolužitie *S. purpurea* a *Habrotrocha rosa* (Donner, 1949) (zástupcu skupiny Bdelloidea) môže byť obojstranne výhodné. *S. purpurea* poskytuje životný priestor, *H. rosa* poskytuje rastline fosfor a dusík. Poznáme dve skupiny rastlín, ktoré využívajú pre svoju výživu pasce vo fytotelmách. Jedna skupina produkuje toxické, kyslé šťavy a tráviace enzýmy, druhá skupina, kam patrí rod *Sarracenia*, tieto látky neprodukuje, ich telmy obsahujú len dažďovú vodu. Vo fytotelmách rastlín *Sarracenia* sa vyskytuje množstvo vírnikov. Výskum Adlassing et al. (2011) potvrdzuje, že živočchy žijúce v takýchto telmách majú s hostiteľskou rastlinou mutualistický vzťah, redukujú veľkosť koristi, produkujú anorganické látky pre rastlinu a asimilujú atmosférický dusík. To platí aj pre vírniky. Daufresne et al. (2008) uvádzajú, že vírniky majú k rastline *S. purpurea* parazitický vzťah. Fytotelmy sa nevyskytujú len u *S. purpurea*, ale aj u *Sarracenia flava*. U *S. flava* sa našlo vo fytotelmách množstvo vírnikov v anhydrobióze. Po rehydratácii sa vrátili k aktívnemu životu (Baldwin & Menhinick 2000). V Severnej Amerike sa uskutočnil výskum autorov Buckley et al. (2003), v ktorom sledovali celé potravné siete vo fytotelmách u *S. purpurea*. Výskum prebiehal v dvoch priestorových rovinách – porovnávali sa siete medzi fytotelmami v rámci jednej lokality a medzi viacerými lokalitami ako celkami. Bohatosť druhov v oboch rovinách sa zvyšovala so zemepisnou šírkou, a to v dôsledku rastúcej druhovej bohatosti nižšej trofickej úrovne. Vplyv na to mohlo mať aj to, že smerom na sever sa znižovala početnosť vrcholového predátora (larií komára *Wyeomyia smithii*) v týchto mikrohabitatoch (Buckley et al., 2003). Určité druhy

vírníkov sa pravidelne vyskytujú u konkrétneho druhu rastliny vytvárajúcej fytootelmy. Môžeme preto usudzovať, že daný druh vírnik je pre rastlinu špecifický. Toto tvrdenie podporuje výskum zo Severnej Ameriky. Vzorkované boli rastliny *S. purpurea* na východnom pobreží od New Jersey po mesto Georgia. *Habrotrocha* cf. *rosa* sa vyskytovala v 70 % z 225 vzoriek (Duffield et al. 1997).

Záver

V pazuchách listov sme celkovo našli 4 taxony vírnikov, bližšie neidentifikovaných zástupcov skupiny Bdelloidea a bežné, kozmopolitné druhy *Lecane bulla* (Gosse, 1851), *Lecane inermis* (Bryce, 1892) a *Colurella obtusa* (Gosse, 1886). Na základe stanovených hypotéz sme predpokladali, že pozitívnu koreláciu s prítomnosťou a početnosťou vírnikov budú mať viaceré faktory, ako objem fytootelmy, ich poloha v rámci rastliny, výška rastliny a podobne. Z týchto faktorov však mala pozitívnu koreláciu len teplota vzduchu. Početnosť vírnikov negatívne korelovala s objemom fytootelmy. Druhové zloženie medzi lokalitami sa nelíšilo, početnosť, naopak výrazne. Toto by mohlo súvisieť s mechanizmami disperzie jednotlivých druhov. Druhy schopné anhydrobiózy sa dokážu šíriť vetrom aj na veľké vzdialenosti (Jenkins & Underwood 1998). Predpokladali sme, že objemom väčšie telmy, v dolnej časti rastliny budú mať vyššiu relatívnu početnosť vírnikov, túto hypotézu sme, keďže u skupiny vírnikov Monogononta, konkrétne u druhu *L. bulla* vyvrátili. Zaznamenali sme pravý opak, početnosť bola vyššia v hornej časti rastliny. Výskyt a početnosť vírnikov vo fytootelmách *D. fullonum* ovplyvňujú rôzne faktory prostredia. Zásadným a limitujúcim faktorom pre živočíšnych obyvateľov fytootelmy je však prítomnosť vody, resp. vysychanie.

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The moth flies (Diptera: Psychodidae) of different ecosystems of the karst Barrois plateau, France

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Abstract

The psychodid fauna of the karst Barrois plateau in France is documented for the first time. Eight moth flies species were recorded in five different ecosystems. *Trichomyia urbica* Haliday in Curtis, 1839, and *Mormia revisenda* (Eaton 1893) are rare European species. On the other hand, *Psychodocha cinerea* (Banks 1894) and *Psychodocha gemina* (Eaton 1904) are very common and occur in all the ecosystems studied. Karst landscapes represent a very interesting and at present highly endangered ecosystem that deserves our attention.

Keywords: Biodiversity, faunistics, Karst, Trichomyiinae, Psychodinae, Mormiina, Trichopsychodina, Psychodini, Europe, Palearctic Region

Introduction

According to Hamilton-Smith (2001), it is important to think of karst areas not just as caves or a landscape, but rather as “a karst system, incorporating component landforms as well as life, energy, water, gases, soils and bedrock”, simply the karst ecosystem (see also in Eberhad 1994).

Much of the study of karst biota has focused on macrocaverns, simply because these are the only caverns directly accessible to biologists (Hamilton-Smith 2001). However, Howarth (1983) demonstrated the immense importance of smaller cavities as habitats for invertebrates. Most biological work has focused on subterranean fauna, particularly bats and troglobitic invertebrates. There are, however, a number of other extremely important and often neglected karst-dependent species, such as microbiota, plants, vertebrates and invertebrates, which are dependent on the karst vegetation or are otherwise restricted to karst areas (Hamilton-Smith 2001). Many of the threats to karst biota are large-scale events that threaten the very integrity or even survival of the karst itself (Watson et al. 1997). These include: i.) total destruction of the karst as a result of mining, quarrying, submersion of water reservoirs; ii.) major land or hydrological disturbance, e.g., monoculture forestry, quarrying, land clearing, construction, waste disposal or other landfills; iii.) pollution from sewage and domestic drainage, farm or industrial waste, hydrocarbons from fuel spills or microbial contamination; and iv.) human entry to caves or other uses, e.g., military, religious observance and monuments, sanatoria, research, tourism, etc. (e.g. Watson et al. 1997; Hamilton-Smith 2001). From the point of view of the biodiversity of the family

Psychodidae, there are only a few notes devoted to this group of organisms in karst ecosystems in Europe (e.g., Ježek 1982; Ježek et al. 2014), especially from caves, e.g., Ježek (1983, 1990), Košel & Horváth (1996), Ježek & Omelková (2012), Omelková & Ježek (2012), etc.

The main goal of this work is to describe the biodiversity and seasonal changes of the family Psychodidae in different ecosystems of the karst Barrois plateau in France.

Material and methods

Sampling was carried out in 2022 at three different sites: Mussey station (48°48'23.4"N 5°04'40.2"E, 204 m a.s.l.), Ville-sur-Saulx station (48°42'45.6"N 5°03'14.7"E, 207 m a.s.l.), and Robert-Espagne station (48°44'52.0"N 5°01'49.0"E, 160 m a.s.l.), all in the forest, on a karst plateau in north-eastern France, west of Bar-le-Duc, prefecture of the Meuse department (Figure 1).

The climate is oceanic with continental tendencies, an average annual temperature of 11°C and regular monthly rainfall (annual average of 845 mm). This region suffers from the effects of climate change, with inter-seasonal heatwaves damaging the forest ecosystem, and unusual tornadoes causing flooding at the resurgences in the valley of the Saulx River, which crosses the karst Barrois plateau. A miniature CDC light trap (a miniature light trap with an attractant to lure flies into the collection chamber) was used for sampling, powered by new Simply LR20/D alkaline batteries. A detailed overview of the investigated ecosystems is in Table 1.

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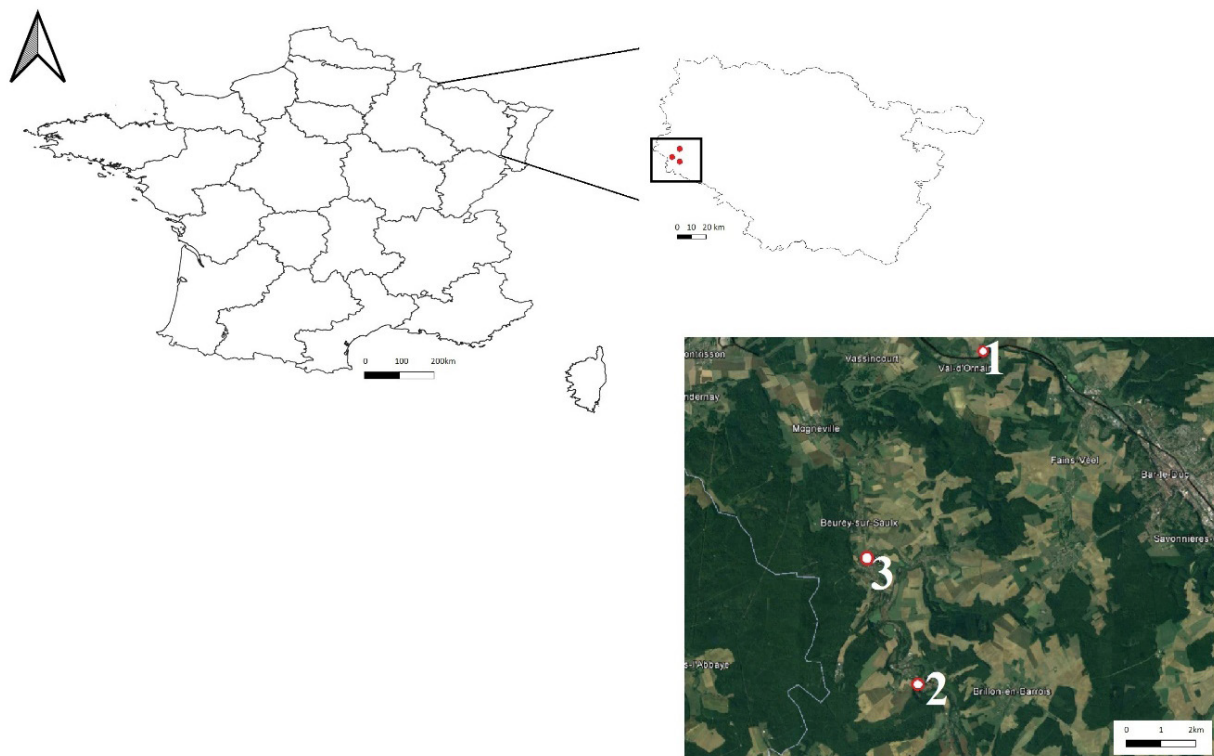


Figure 1. Map showing the sampling sites on the karst Barrois plateau, France. Prepared by Lenka Demková. 1: Mussey station, 2: Ville-sur-Saulx station, 3: Robert-Espagne station.

Table 1. Detailed overview of the investigated ecosystems.

Station	Ecosystem	Short description and collecting methods
Mussey station	undergrowth	CDC at 1.20 m from the ground at the bottom of a unique sinkhole (a funnel-shaped collapse 10 to 15 m deep, found in karst regions, the bottom is blind, with a tangle of vegetation and trees)
	sinkholes	varying depth and appearance Perini (Figure 2), Blaireau, Huss, and Petit bois communal
Ville-sur-Saulx station	undergrowth	same location of the CDC
	trench	Figure 4; path 400 m long, 15 m wide, and 5 m deep, dug by quarrymen to reach the limestone layer, along which the various entrances to the underground quarry can be seen; CDC always placed in the same location
	underground quarries	entrances to Agouti 1 (Figure 3), 2, and 3 on the right side, and two others on the left side; CDC placed at each entrance
Robert-Espagne station	chasms	La Laie (Figure 5), Paul, Ohzi, and Renaissance

Each ecosystem was sampled during four periods: May, July, September, and November. The captured moth flies (approximately 340 specimens in total) were preserved in 70% ethanol in the field by the second author and subsequently identified by the first author. The studied material is deposited in the alcohol collection of insects (Diptera) at the University of Prešov.



Figure 2. The Perini sinkhole in old-growth forest, with large oak trees and a low density of growing trees in the understory.



Figure 3. Entrance to the Agouti 1 underground quarry. The entrance is blocked by the soil from the erosion of the hill since the quarry was closed.



Figure 4. The trench is still covered with vegetation despite the winter. On the left, in the background in the middle of the path, is a CDC trap. On the right and halfway up, another CDC at the entrance to a quarry, nommée ‘the first on the left’.



Figure 5. The ‘La laie’ chasm in Robert-Espagne, a classically shaped chasm.

Identification keys used: Vaillant (1971–1983), Szabó (1983), Withers (1989) and original papers with species descriptions (Ježek 1977, 1983, 1985, 1990; Ježek & Goutner 1993; Omelková & Ježek 2012). The nomenclature and classifications are modified using Ježek & Omelková (2012), Oboňa & Ježek (2014), Kroča & Ježek (2015, 2019, 2022) and Ježek et al. (2021b).

Results and Discussion

During 2022, five different types of ecosystems were sampled in three different locations: chasm, undergrowth, underground quarry, sinkhole and trench. A total of eight species of family Psychodidae were recorded. Faunal data from the same ecosystems are summarised in Table 2.

Table 2. Alphabetic overview of species of the family Psychodidae recorded in the studied ecosystems.

	Chasm	Undergrowth	Underground quarry	Sinkhole	Trench
<i>Logima zetterstedti</i> Ježek, 1983	*		*		
<i>Mormia revisenda</i> (Eaton, 1893)	*	*	*		
<i>Philosepedon humerale</i> (Meigen, 1818)	*				
<i>Psychodocha cinerea</i> (Banks, 1894)	*	*	*	*	*
<i>Psychodocha gemina</i> (Eaton, 1904)	*	*	*	*	*
<i>Psychodula minuta</i> (Banks, 1894)	*	*	*		*
<i>Tinearia alternata</i> (Say, 1824)		*			
<i>Trichomyia urbica</i> Haliday in Curtis, 1839			*		

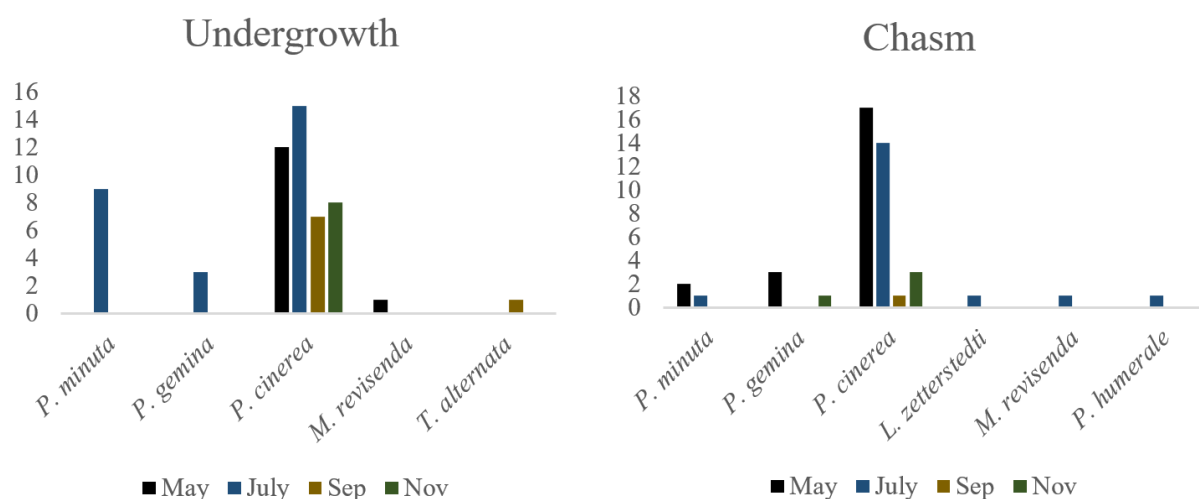


Figure 6. An overview of the occurrence of the moth flies in chasms and in undergrowth. The x-axis shows the presence of species in the months of May, July, September and November; the y-axis shows the abundance of species.

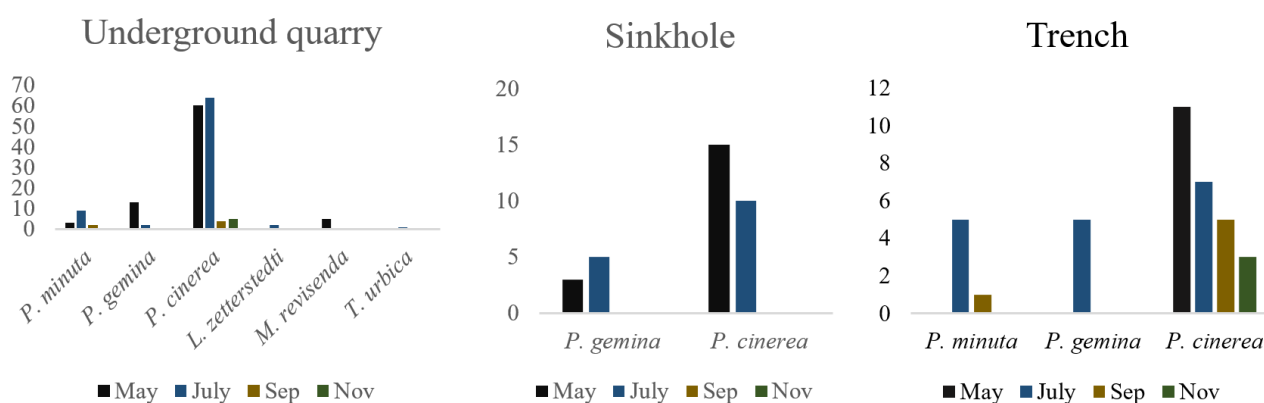


Figure 7. An overview of the occurrence of moth flies in underground quarries, sinkholes and trenches. The x-axis shows the presence of species in the months of May, July, September and November; the y-axis shows species abundance.

In the chasms, 44 moth flies belonging to five species were collected. *P. cinerea* dominated with 35 individuals. This species was present in chasms throughout the year. In the undergrowth, 56 moth flies belonging to five species were collected. Again, *P. cinerea* dominated with 42 individuals. This species was present in undergrowth throughout the year. An overview of the occurrence of other species is given in Figure 6.

The most flies were collected the underground quarries – 170 moth fly individuals in total, belonging to six species. *P. cinerea* dominated with 133 individuals. In sinkholes, only 33 moth flies belonging to two species were collected. None of the captured species occurred in sinkholes throughout the year. In the trenches 37 moth fly samples were caught belonging to three species. *P. cinerea* dominated with 26 samples. This species was present here throughout the entire year. An overview of the occurrence of other species is in Figure 7.

There are five species in the flat undergrowth of the plateau; the psychodid fauna of the chasms and underground quarries is slightly richer compared to the undergrowth which characterises the natural environment of the forest which is particularly evident from Figure 7 (Sinkhole and Trench). In general, it can be noted (see also Table 2) that the fauna is remarkably similar (although there are no harvests during the winter period). The chasms, although more exposed to bad weather, provide shelter for the same number of species as the undergrowth. If we consider common undergrowth dipterans from other families found in this biotope, such as *Limonia nubeculosa* Meigen, 1804 (Diptera: Limoniidae), we observe that its outbreaks in September 2021 and 2022 were as abundant at the entrance of underground quarries as above chasms, confirming the shelter role of the latter.

Recorded species

From eight recorded species, one belongs in subfamily Trichomyiinae, and seven in subfamily Psychodinae (one from Mormiina, one from Trichopsychodina, five from Psychodini).

Subfamily Trichomyiinae

Trichomyia urbica Haliday in Curtis, 1839

A rare European and Transcaucasian species (Ježek et al. 2021a). The larvae are xylophagous and occur in habitats with decaying cellulose (Ježek & Omelková 2012). Only one individual of this species was found, in July in an underground quarry (site Ville sur Saulx).

Subfamily Psychodinae

Subtribe Mormiina

Tribe Mormiini

Mormia revisenda (Eaton, 1893)

A rare European species (Ježek et al. 2018, 2020; Morelli & Biscaccianti 2021). This species was recorded in a chasm (1 specimen), undergrowth (1 ind.) and an underground quarry (4 ind.) in May and in July.

Subtribe Trichopsychodina

Tribe Paramormiini

Philosepedon humerale (Meigen, 1818)

It is known from the Western Palearctic and the Afrotropical region (Afzan & Belqat 2016; Ježek et al. 2020). The larvae were found in dead snails of various species (Beaver 1972; Ježek 1985). This species was only recorded in a chasm (1 individual) in July.

Tribe Psychodini

Logima zetterstedti Ježek, 1983

A common European, Transcaucasian and West Siberian species (e.g. Ježek et al. 2021a,b; Gibernau & Albre 2022). The larvae are saprobionts, inhabiting the soil of paddocks, drains and water pipes, dead fungi (genus *Craterellus* Pers.), excrements of vertebrates, banks of polluted water reservoirs, manure and decaying plant material, etc. (e.g. Jung 1956; Wagner 1977; Ježek 1983). This species was recorded only in a chasm (1 specimen) and the underground quarry (2 ind.), both in July.

Psychodocha cinerea (Banks, 1894)

A very common cosmopolitan species (e.g. Ježek et al. 2017, 2021a,b, 2023). The larvae have been recorded in rotten vegetables, in an algae cover of a trough with water in shady places, in sewage works, in paddocks in sludge, in manure, in sewers, on toilets, water pipes, etc. (Crisp & Lloyd 1954; Jung 1956; Duckhouse 1966; Wagner 1977; Ježek 1990). This species was recorded in large numbers in all the ecosystems studied. Most specimens were caught in May and June. The abundance of the species always decreased in September and November, which can be related, for example, to the availability of resources and environmental conditions, but also interspecies competition.

Psychodocha gemina (Eaton, 1904)

A common European and Transcaucasian species (Ježek et al. 2021a). The larvae live in the damp mud of paddocks, in manure, sewers, on toilets, sewage works, water pipes, among decaying leaves on the banks of ponds and near springs (Jung 1956; Ježek 1990). This species was found in small numbers in all the ecosystems studied. In none of the monitored ecosystems was this species present throughout the season.

Psychodula minuta (Banks, 1894)

A generally common Holarctic species (Ježek et al. 2017). Larvae are saprobionts that develop in decaying vegetation, caves, damp places, mostly in dung or animal faeces, especially of vertebrates. Adults are often found on the banks of ponds, forest streams, swamps, gutters, outflows from water reservoirs and in waste pits (Jung 1956; Quate 1960; Wagner 1977; Ježek 1990). This species was recorded in smaller numbers in all the ecosystems studied except sinkholes. In none of the monitored ecosystems did this species occur throughout the season.

Tinearia alternata (Say, 1824)

A cosmopolitan and euryvalent species (Kročá & Ježek, 2019; Ježek et al. 2021b, 2023). Larvae can be found in sewage treatment plants, on grave beds, in organically polluted water, on faeces, on fresh potato litter and in alder swamps. Adults occur on the banks of streams and ponds, on the walls of buildings and stables or in nests and burrows of small mammals (e.g. Ježek 1977; Semelbauer et al. 2020; Oboňa et al. 2022; Roháček et al. 2022). This species (only 2 individuals) was only found underground (in July and September) at the site Ville-sur-Saulx.

As the karst environment is very sensitive to disturbance, once a strong disturbance occurs in a karst ecosystem, it will undergo reverse succession, and both its recovery and restoration may be difficult. Therefore, biodiversity is an important factor in the maintenance of existing karst landscapes and provides the basis for the recovery of degraded karst ecosystems. Monitoring could provide scientific data to help maintain biodiversity in karst areas (Li et al. 2013).

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The variability of chaetotaxy of *Lipoptena fortisetosa* Maa, 1965 (Diptera: Hippoboscidae)

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Abstract

Keds (Diptera: Hippoboscidae) are a very specific and undoubtedly interesting family of ectoparasitic dipterans. A morphometric analysis of the setae of *Lipoptena fortisetosa* (Maa, 1965) revealed considerable variability, with many setae often missing in each specimen. Therefore, it is advisable to select a smaller number of landmarks that are consistently present in all individuals. The analysis of the location of such head and thorax landmarks in *L. fortisetosa* indicated a significantly higher variability in females. The “ideal” individual is always characterized by 6 setae on the head and 22 on the thorax (4 humeral setae, 4 laterocentral setae, 8 postalar setae and 6 scutellar setae).

Key words: louse flies, ectoparasites, morphometry, setae, thorax, head

Introduction

The dipteran family Hippoboscidae (louse flies – ectoparasites of birds or keds – ectoparasites of mammals) is an interesting object of research (e.g., Dibo et al. 2022; Andreani et al. 2022; Yatsuk et al. 2023; Tiawsirisup et al. 2023, etc.). The most important defining features of this family include host affinity and morphological characteristics, such as the presence, absence or reduction of the wing, veins and microtrichia on the wing, body shape and colour, and the presence of setae and other structures on the body, etc. (Povolný & Rosický 1955; Maa 1965; Chalupský 1980; Chalupský & Povolný 1983; Hutson 1984; Dosnažov 1987; Oboňa et al. 2022, etc.). The genus *Lipoptena* Nitzsch, 1818, is represented by two species in Slovakia (Sychra 2009; Oboňa et al. 2019a). The first, *L. fortisetosa* Maa, 1965, is a relatively common but non-native species in Slovakia (Figure 1, right), distributed in the eastern Palaearctic region. In the past, it was frequently misidentified with *L. cervi* (Linnaeus, 1758), and appears to have a western distribution limit in Central

Europe. This species is an ectoparasite of Cervidae and also attacks humans and birds (e.g., Ducháč & Bádr 1998; Oboňa et al. 2019a, 2022). The second species is *L. cervi*, a relatively common species native to Central Europe, distributed in the Palaearctic region and introduced into the Nearctic region. Similarly as *L. fortisetosa*, it is an ectoparasite of Cervidae and can also attacks humans (Křištofik 1998; Oboňa et al. 2019a).

To a large extent, the main differences between the two species are in the chaetotaxy of the head and thorax (mesonotum) (Maa 1965; Chalupský 1980; Dosnažov 1987; Ducháč & Bádr 1998; Salvetti et al. 2020; Oboňa et al. 2022). The chaetotaxy, especially on the thorax, is relatively variable; therefore, we decided to analyse this variability in more detail.

The main aims of this work are i) to analyse the chaetotaxy of *L. fortisetosa* and ii) to evaluate the differences in chaetotaxy between the sexes of this species in Slovakia.



Figure 1. Photographs of specimens of *Lipoptena cervi* (Linnaeus, 1758) (left) and *Lipoptena fortisetosa* Maa, 1965 (right).

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Material and methods

The majority of the material was collected during field excursions in different parts of Slovakia (mainly from humans). The samples are deposited in the Laboratory and Museum of Evolutionary Ecology, Department of Ecology, University of Prešov (LMEE PO), see Oboňa et al. (2019a). Chaetotaxy analysis was performed on *L. fortisetosa* specimens deposited at LMEE PO. All specimens were photographed from the back under the same conditions using a Motic trinocular microscope with a camera (10x magnification). The photographs were then sorted by location, collection date and sex. Prior to data collection, tps files were created from the photographs using the program tpsUtil (ver. 1.75). All setae on the thorax (20 landmarks) were first selected. Consequently, the data were standardized in MorphoJ (ver. 1.06d) and analysed in the same program using the PCA (Principal Component Analysis) method, with graphical output. Landmarks were highly variable and several were often absent in many individuals. Their number was reassessed and the procedure was repeated only on the landmarks present in all individuals (10 landmarks on the head and 15 landmarks on the thorax, Fig. 2), so that they could be evaluated in the tpsDIG1 program (ver 1.40). In the PAST software (ver. 3.18; Hammer et al. 2001), the data were standardized to Procrust coordinates, thus eliminating the influence of position, size and rotation. In the same software, the data were analysed using the Principal Component Analysis (PCA) method, generating a graphical output of the analysis (scatterplot) with 95% ellipses (the probability that landmarks placed on other specimens of the sampled population will be within the ellipse is 95%).

PCA searches for hypothetical variables (components) that explain most of the variance in the measures (Davis 1986; Harper 1999). These variables are linear combinations of the original variables and this method is mainly used to reduce the data set to two components and to produce a graphical output (Peres-Neto et al. 2003). In order to visualize the relative deformations, the deformation energy matrices were calculated from the eigenvectors and then represented as deformations of the mesh, showing how the object would look if its relative deformation values (relative wraps-score) were at opposite ends of one of the relative deformation axes and at zero on the other axes. The percentage of the total variance expressed by each component was also calculated. The graphical output of the eigenvalues for each component was complemented by a “broken stick” – a curve showing the predicted eigenvalues in a random model, which helps to assess the number of significant components (above the intersection point). The XY plot, supplemented by a 95% ellipse, was used to illustrate the location of landmarks and their variability within the set groups.

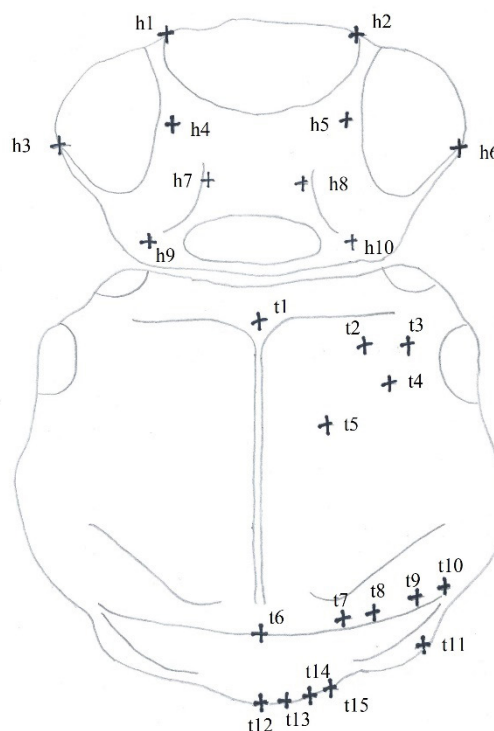


Figure 2. Scheme of the most suitable landmarks on the head (h1 – h10) and thorax (t1 – t15) of *L. fortisetosa*.

Results and Discussion

A total of 48 specimens (26 females and 22 males) of *L. fortisetosa* were analysed. The number of landmarks varied among specimens, as many were missing many of the setae. This may have biased the analysis, and no significant relationship was found between morphometrics and collection time. Variable landmarks were therefore excluded from the analysis. The generated chart, completed with 95% ellipses (Figure 3 head, Figure 4 body), confirmed the appropriate selection of landmarks. The placement of landmarks was almost identical in both sexes (Figures 5, 6), although the variability was higher in females, as indicated by the red ellipse consistently being larger than black one).

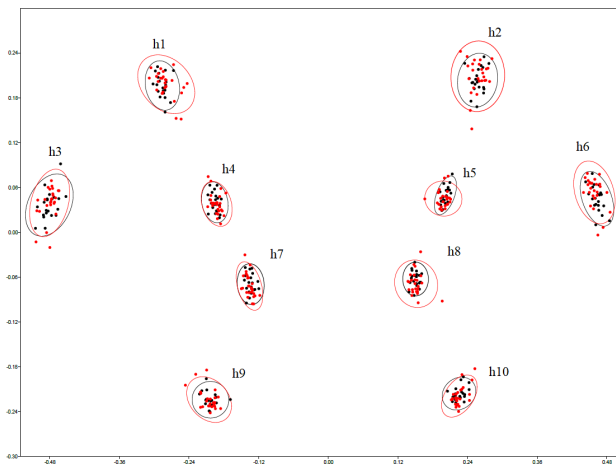


Figure 3. XY plot supplemented by 95% ellipses showing the location of landmarks and their variability within defined head groups of *L. fortisetosa* (red dots – female, black – male).

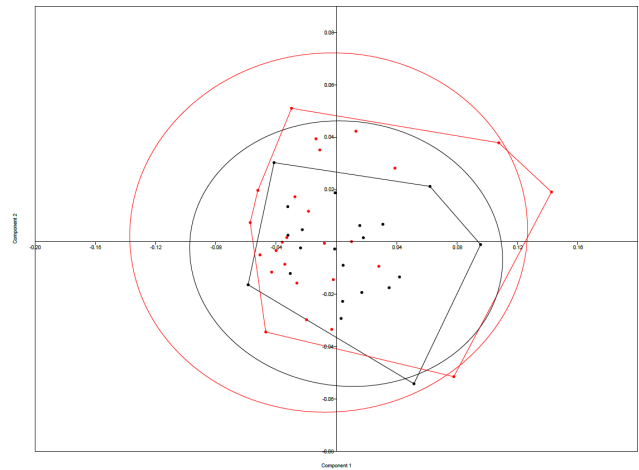


Figure 5. Scatter plot with 95% ellipse on the head of *L. fortisetosa* (red dots – female, black dots – male).

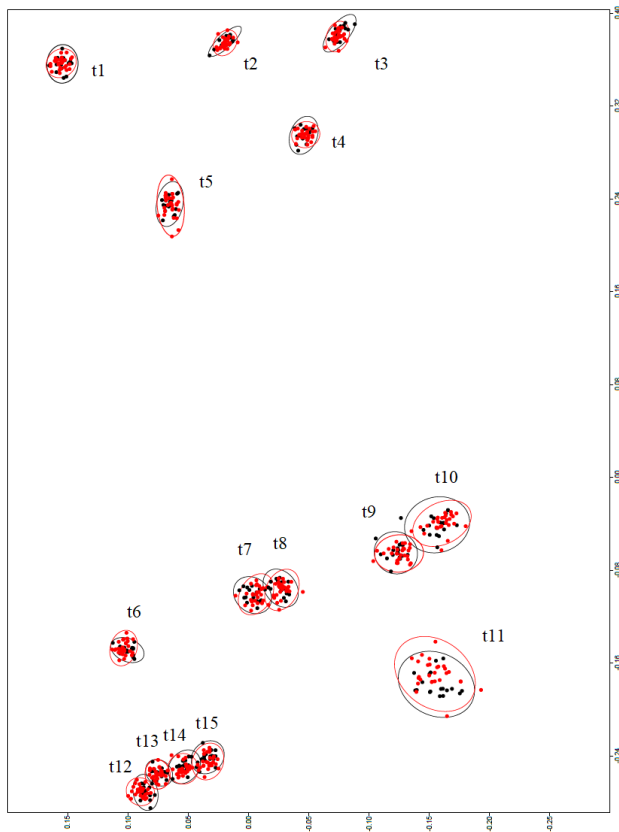


Figure 4. XY plot supplemented by 95% ellipses showing the location of landmarks and their variability within defined thorax groups of *L. fortisetosa* (red dots – female, black – male).

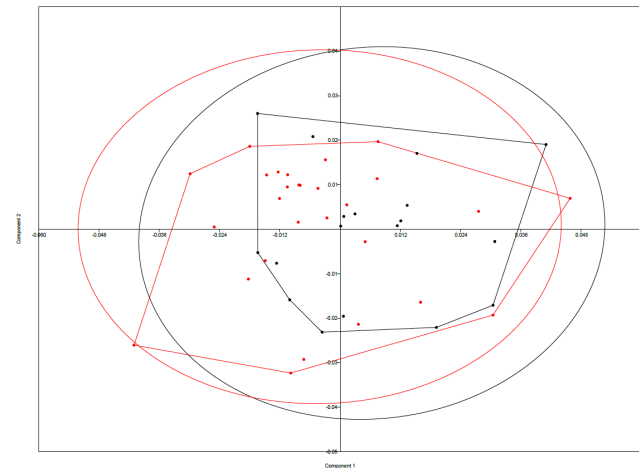


Figure 6. Scatter plot with 95% ellipses on the thorax for *L. fortisetosa* (red dots – female, black dots – male).

The percentage of total variance accounted for by each component was also evaluated (Figures 7 and 8). The graphical output was supplemented by a curve showing the predicted values of the significant components (above the intersection point).

The graphical output showed that despite the higher variability of females (Figure 5 head, Figure 6 body), males and females did not differ significantly in the morphometric features observed. A greater difference was found for the head landmarks, where 4 points (18%) of the analysed female specimens were outside 95% of the male ellipse.

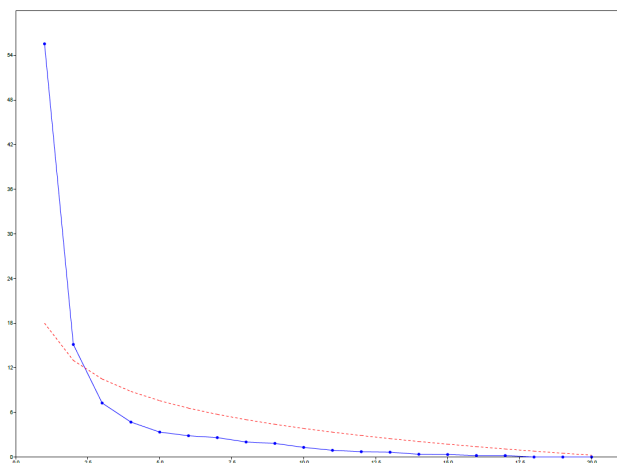


Figure 7. Percentage of the total variance with a curve showing the predicted values of significant components (above the crossing point) on the head of *L. fortisetosa*.

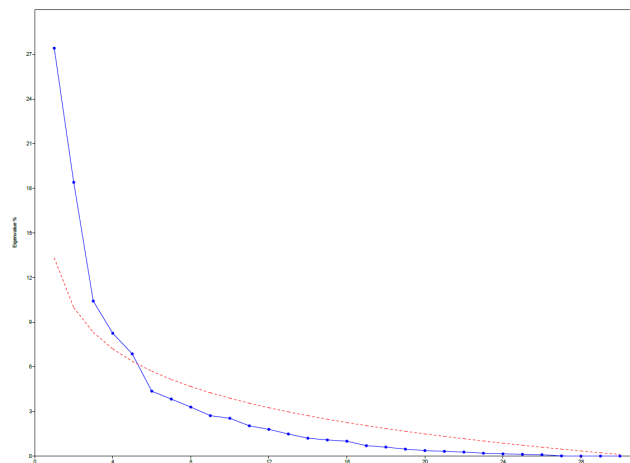


Figure 8. Percentage of the total variance with a curve showing the predicted values of significant components (above the crossing point) on the thorax of *L. fortisetosa*.

The chaetotaxis of the species *L. fortisetosa* is quite variable, as shown in the above results. However, when suitable landmarks (present in all individuals) are selected and a sufficient amount of material is processed, the results clearly confirm that males and females are similar in chaetotaxy, but greater variability was noted in females of *L. fortisetosa*. For the species *L. fortisetosa*, the landmarks in Figure 9 appear to be the most suitable for further possible analysis.

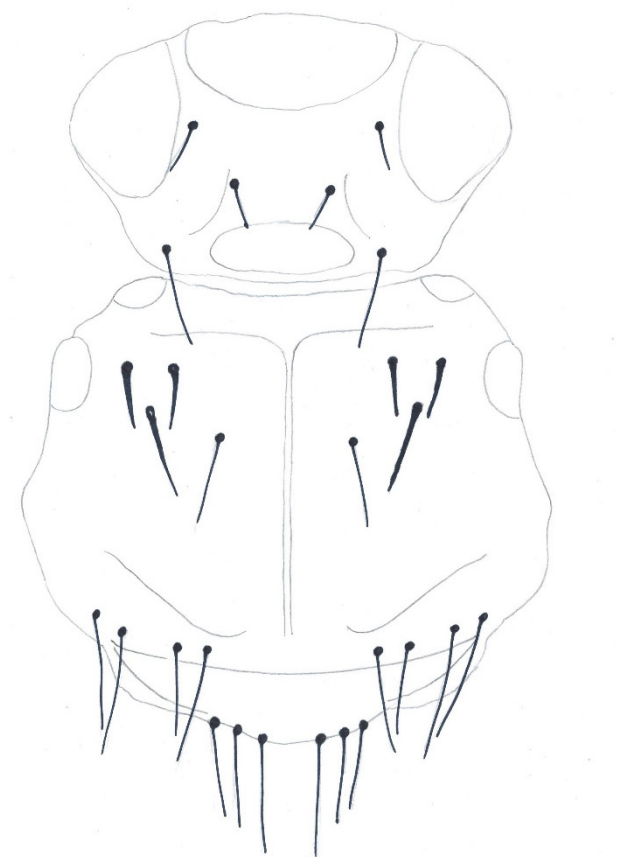


Figure 9. Scheme of the “ideal” specimen of *L. fortisetosa*.

The chaetotaxy of the head and thorax of the species *L. fortisetosa* is quite variable (even in terms of length and thickness of individual setae); the “ideal” individual has 6 setae on the head and 22 on the thorax. At the front of the mesonotum 2 setae (on each side – humeral setae), below them, approximately in the middle of the mesonotum – 2 laterocentral setae on each side. On the dorsum of the mesonotum 8 posterior setae, and on the scutellum with 6 scutellar setae (Figure 9).

Similar results were published by Maa (1965), Chalupský (1980), Dosnažov (1987) and Ducháč & Bádř (1998). Therefore, it is necessary to select appropriate landmarks that are present in all specimens examined. The ideal landmarks for *L. fortisetosa* are the points in Fig. 2 or the setae in Fig. 9. These points were present in both sexes, although greater variability was observed in the females. It is interesting to note that certain shape variables were also recorded in the photographed specimens (e.g., in the scutellum).

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Bird mortality near roads in the Sabinov district, Slovakia

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Abstract

This study presents findings from a pilot investigation regarding the presence of bird carcasses in close proximity to roadways in the district of Sabinov, located in eastern Slovakia. Over the period of 2016 to 2021, a total of 29 bird carcasses were identified through mapping. The highest frequency of carcasses was observed during April and May (exceeding 60% of the total). Based on zoological classification, the carcasses were categorized into 6 orders, 13 families, and 18 species. Carcasses of the following species were repeatedly found: *Emberiza citrinella* Linnaeus, 1758 (5 birds), *Turdus merula* Linnaeus, 1758 (4 birds), and *Turdus pilaris* Linnaeus, 1758 (4 birds). Based on our pilot study results, it appears that bird-car collisions are more likely to occur in areas with vertical barriers on both sides of the road and where landscape structures such as non-irrigated arable land and a discontinuous urban fabric are dominant.

Key words: Aves, Passeriformes, carcasses, landscape structure, vertical barrier

Introduction

Car traffic significantly eases people's lives. However, it also has negative effects on the environment (Demková et al. 2019). One negative effect is that it poses a significant threat to wildlife like birds in the surrounding areas (Kambourova-Ivanova et al. 2012; Ďula 2013; Loss et al. 2014; Garcês et al. 2020). Birds are the most common animals killed on roads (Kambourova-Ivanova et al. 2012). According to Loss et al. (2014), many factors affect the incidence of birds colliding with vehicles (see Figure 1).

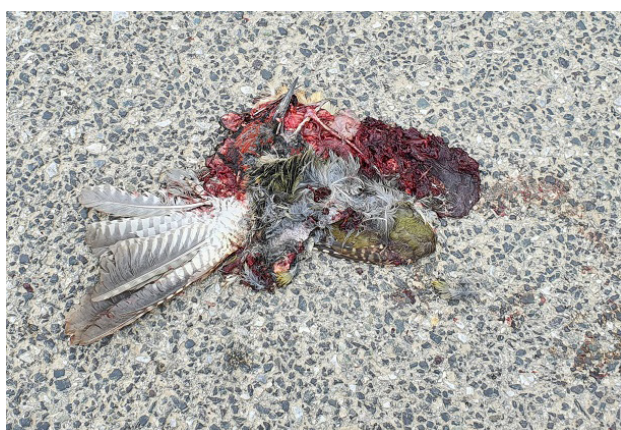


Figure 1. The corpse of a male European Green Woodpecker (*Picus viridis* Linnaeus, 1758) found at site 22.

The main factors are the speed and volume of traffic (Case 1978) and the presence of young birds (e.g. Erritzoe et al. 2003; Gunson et al. 2010; Boves & Belthoff 2012). The highest number of victims on motorways are passerine birds, with the highest number of individuals killed in

March and June (Kambourova-Ivanova et al. 2012).

The main objective of this work was to obtain the first information on bird carcasses near roads in eastern Slovakia in order to describe their biodiversity and to try to determine the landscape structure that influences the presence of these carcasses.

Material and methods

The cadastral surveys were carried out randomly during extensive cycling mapping activities in eastern Slovakia, Sabinov district, between 2016 and 2021. Only roads where cycling was possible were documented. Each carcass was documented (GPS position, date, time, landscape and systematic classification of the bird were recorded).

The location of the carcass was also used to determine the secondary landscape structure and its possible influence on the characteristics assessed. For research purposes, the flight distance of the birds was set at 150 m from the carcasses. Within this radius, the percentage of the following classes of land cover structure elements were determined according to the CORINE Land Cover (CLC) (2018): (2) discontinuous urban fabric, (3) industrial or commercial units, (12) non-irrigated arable land, (18) pastures, (20) complex cropping patterns, (21) land predominantly used for agriculture with significant areas of natural vegetation. The CLC (2018) is coordinated by the European Environment Agency (EEA) under the EU Copernicus programme and is implemented by national teams under EEA management and quality control. It consists of an inventory of land cover in 44 nomenclature classes, with a minimum mapping unit of 25 hectares and a minimum mapping width of 100 metres. Based on

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this vector data (see also Oboňa et al. 2022), we clipped a 150 m radius at each locality and, using the geographic information system QGIS, we counted the proportion of each class in the focus area. The 150 m radius was chosen in consultation with ornithologists.

The presence of vertical barriers was also assessed: (Ver 2) vertical barrier on both sides, (Ver 1) vertical barrier on one side, (Ver 0) vertical barrier not present. (Vertical barriers are obstacles close to the road with a height of at least 2 m and an area of at least 1 m²).

Simple descriptive statistics were calculated and graphs were generated using LibreOffice software. PCA analysis was performed using PAST software (version 4.13; Hammer et al. 2001). The map was created in QGIS

(version 3.22.4-Białowieża) using the standard OSM layer (OpenStreetMap; <https://www.openstreetmap.org>).

Two distant sites with specific species were not compatible with the others and were therefore used in the analyses of the effects of landscape structure and roadside vertical barriers.

Results and Discussion

A total of 29 carcass samples were documented between 2016 and 2021 (Figure 2; see also Supplement 1). According to the date, the bird carcasses were found from March to September. Of the total number, 34% of all the carcasses were found in April, 31% in May, 13% in June, 10% in July and 4% in March, August and September. The difference in seasonal timing also appears to be an important factor, as confirmed by other studies (e.g. Erritzoe et al. 2003).

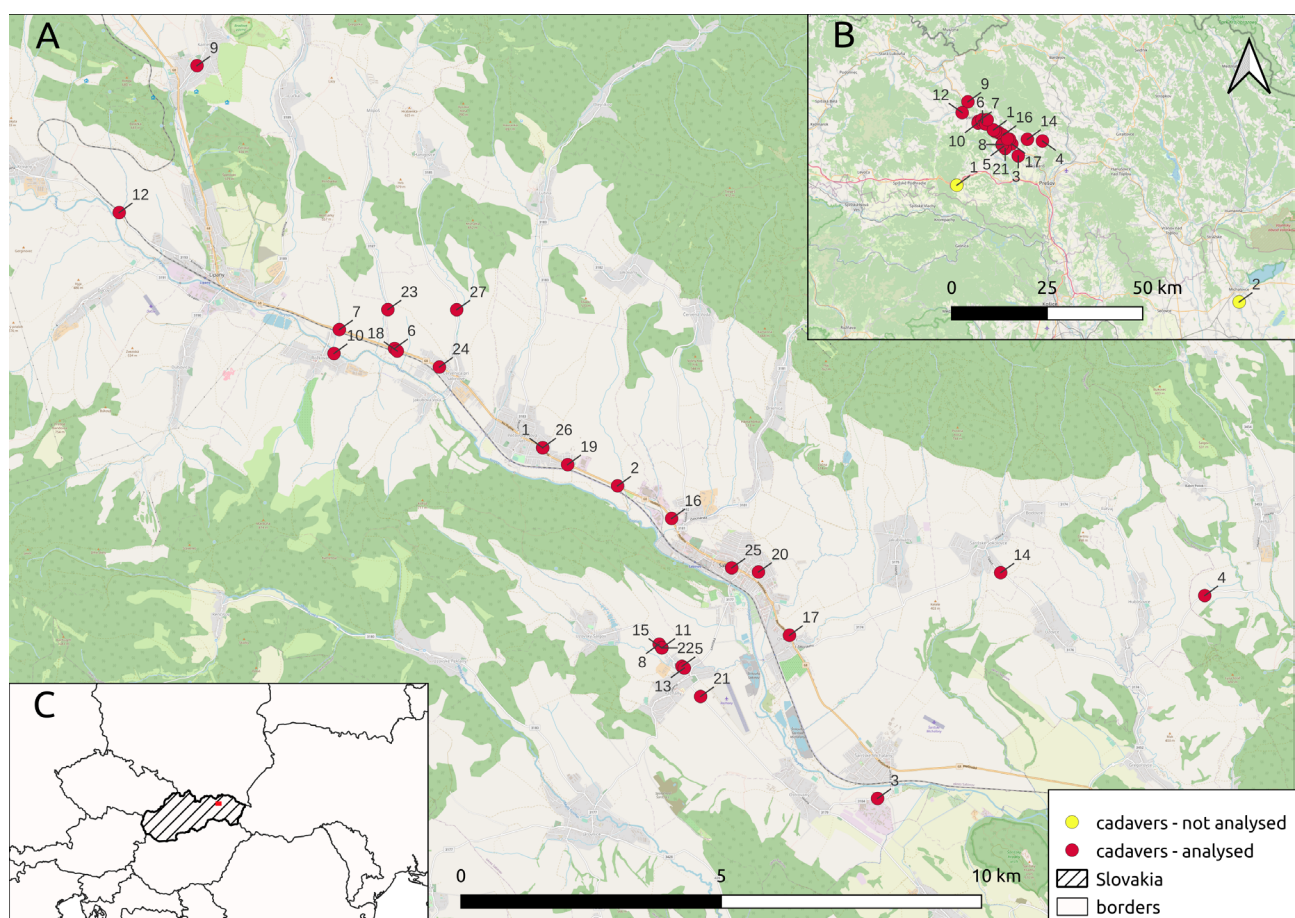


Figure 2. Map of the sites. A – sites included in the analyses. B – sites with both analysed and unanalysed sites. C – overview map.

According to the taxonomic classification, the bird carcasses belong to 6 orders. Most of the carcasses belonged to the family Passeriformes (79%), followed by Piciformes (7%), Accipitriformes, Anseriformes, Galliformes and Strigiformes (4% each). A total of 13 families and 18 species were recorded. The repeated occurrence of carcasses was recorded for the species *Emberiza citrinella* Linnaeus, 1758 (5 individuals), *Turdus merula* Linnaeus, 1758 and *Turdus pilaris* Linnaeus, 1758 (4 each).

When assessing the presence of vertical barriers near the road at the site of the carcass, we found that 65% of Passeriformes carcasses were found at sites with vertical barriers on both sides of the road (Ver 2). Much fewer carcasses were found at sites with vertical barriers on only one side of the road (Ver 1) or with no vertical barriers (Ver 0). In general, carcasses were also found at sites with vertical barriers on both sides (Figure 3).

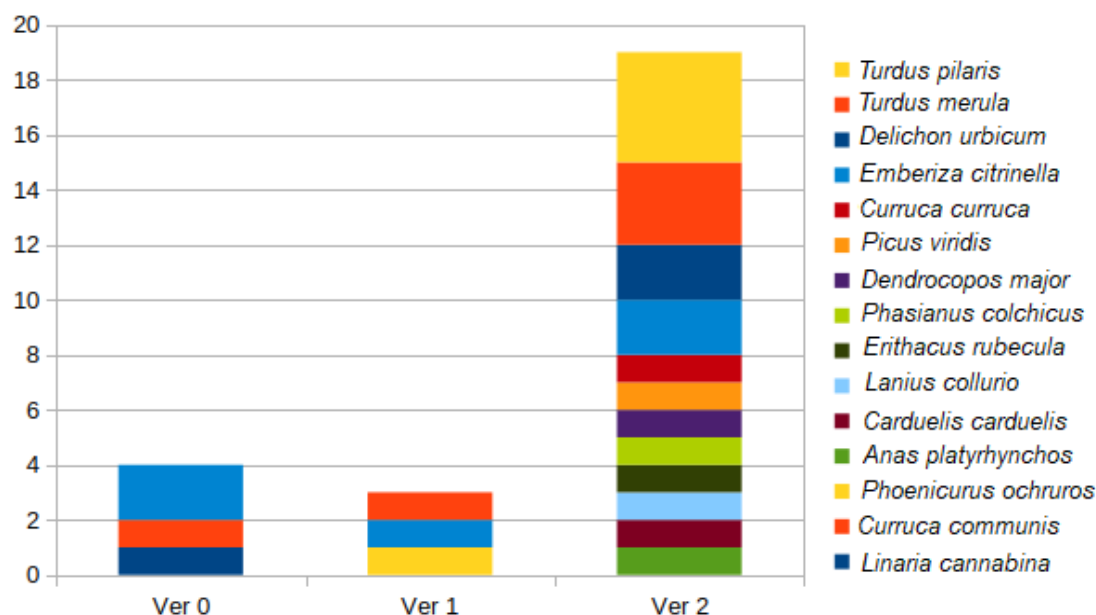


Figure 3. Carcass abundance of each species at sites with different presence of vertical barriers along the sides of the road. Ver 0 – no vertical barrier. Ver 1 – vertical barrier on one side. Ver 2 – vertical barriers on both sides.

When considering secondary landscape structures and their possible influence on the presence of Passeriformes carcasses, it seems that non-irrigated arable land (12) and discontinuous urban fabric (2) are the landscape structures with the strongest influence (Figure 4) and with the most frequent occurrence of these carcasses. We did not observe any significant pattern in the influence of landscape features at the species or family level. However, our results are probably largely influenced by the small number of samples and the presence/absence of landscape structures

in this small study area. According to Ďula (2013), most collisions between cars and birds are related to adjacent vegetation and habitat type. Clevenger et al. (2003) stated that birds die more often in the open country than on roads crossing the forest or ecotone, which is consistent with our findings. Various vertical barriers around roads, e.g. bushes and trees, pose a risk especially for young birds, but they also allow birds to nest and are a source of food or a resting place for them (Havlín 1987; Seiler 2001; Erritzoe et al. 2003; Ďula 2013).

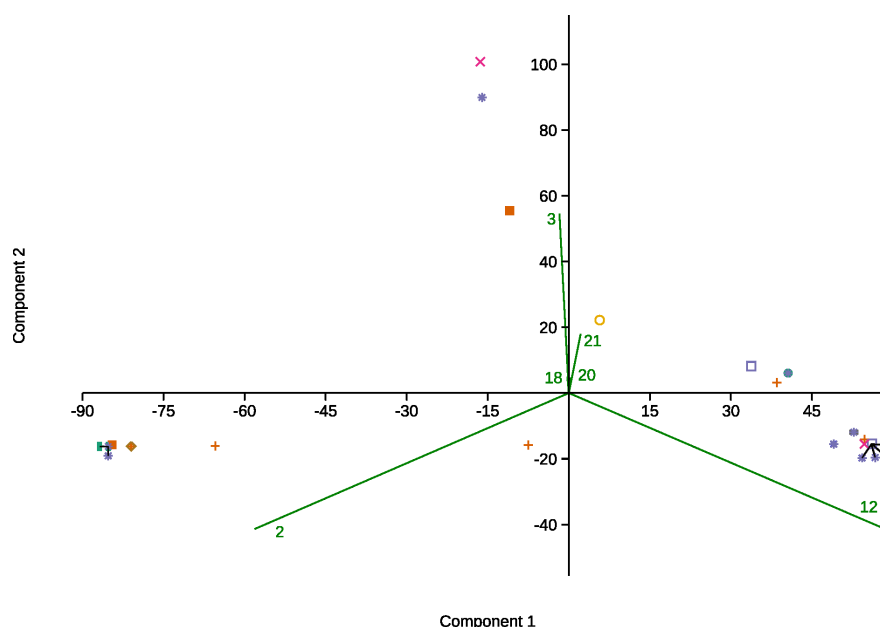


Figure 4. Presence of carcasses at sites with different landscape structures. PCA analysis (variance-covariance matrix, disregarding groups, PC1 explains 66.71% of the variance; symbols represent carcasses of particular species; numerical factor codes: 2 – discontinuous urban fabric, 3 – industrial or commercial units, 12 – non-irrigated arable land, 18 – pastures, 20 – complex cropping patterns, 21 – land mainly used for agriculture with significant areas of natural vegetation). Some symbols have been moved out of the clusters to make the image clearer and are connected to the original location by a black line.

At the species level, we observed differences in the presentation of carcasses. Three species, *E. citrinella*, *T. merula* and *T. pilaris*, collided repeatedly with cars in the study area. The collisions of *E. citrinella* are probably not related to the presence of a vertical barrier (Ver 1 – 1 carcasses, Ver 2 – 2, Ver 0 – 2; Figure 3). The collisions of *T. merula* are probably related to the presence of a vertical barrier on both sides (Ver 2 – 85%; Figure 2). It seems, that non-irrigated arable land (12) is the landscape structure with the most frequent occurrence of these carcasses and this structure seems to have the strongest impact on the traffic mortality of this species among the ones analyzed (Figure 4). The collisions of *T. pilaris* are probably also related to the presence of a vertical barrier on both sides (Ver 2 – 100%). However, the carcasses of this species occurred in different country structures. As with previous results, our findings are likely to be influenced by the small sample size and the presence/absence of landscape structures in this small study area. The validity of our findings is therefore limited and cannot be generalized without confirmation over a larger sample and area.

Despite the limitations imposed by the size of the area and the sample size, our data and results suggest that the vertical barriers on the sides of the road and landscape structures such as unirrigated arable land and discontinuous urban fabric could be significant factors in increasing the rate of bird-vehicle collisions.

Acknowledgements

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Supplement 1. A detailed overview of all recorded data.

Site	2	3	LCL (%)				Date	GPS				Species	Ordo	Vertical barriers		
			12	18	20	21								Ver 2	Ver 1	Ver 0
1	97		3				3.3.2017	49.116303214679	21.068378672179			<i>Emberiza citrinella</i> Linnaeus, 1758	Passeriformes	0	0	1
2			1		99		29.4.2016	49.139464369636	21.010485872802			<i>Curruca communis</i> Latham, 1787	Passeriformes	0	0	1
3	2		30		68		5.4.2020	49.163260149993	20.937422499236			<i>Erithacus rubecula</i> (Linnaeus, 1758)	Passeriformes	1	0	0
4	5		95				27.4.2020	49.101494961874	21.105407848127			<i>Turdus merula</i> Linnaeus, 1758	Passeriformes	1	0	0
5			100				13.5.2018	49.08090052813	21.090201124726			<i>Turdus pilaris</i> Linnaeus, 1758	Passeriformes	1	0	0
6	86		14				13.5.2018	49.06254650404	21.136699900207			<i>Emberiza citrinella</i> Linnaeus, 1758	Passeriformes	0	0	1
8			98		2		7.4.2019	49.097444758014	21.222680792389			<i>Emberiza citrinella</i> Linnaeus, 1758	Passeriformes	1	0	0
9			74		26		1.4.2019	49.085276021248	21.085308775482			<i>Emberiza citrinella</i> Linnaeus, 1758	Passeriformes	1	0	0
10			95		5		26.4.2019	49.088446437124	21.080018048064			<i>Phasianus colchicus</i> Linnaeus, 1758	Galliformes	1	0	0
10			95		5		26.4.2019	49.088446437124	21.080018048064			<i>Turdus merula</i> Linnaeus, 1758	Passeriformes	1	0	0
11	97		3				25.4.2019	49.101378584479	21.169058069764			<i>Passer montanus</i> (Linnaeus, 1758)	Passeriformes	0	1	0
12	99						19.5.2019	49.119954675787	21.055262670098			<i>Curruca curruca</i> (Linnaeus, 1758)	Passeriformes	1	0	0
13	45		55				19.5.2019	49.139906565896	21.009772405207			<i>Emberiza citrinella</i> Linnaeus, 1758	Passeriformes	0	1	0
14			67		33		19.5.2019	49.143163254902	20.995267018854			<i>Linaria cannabina</i> (Linnaeus, 1758)	Passeriformes	0	0	1
15		100					19.5.2019	49.188481997683	20.95786629635			<i>Delichon urbicum</i> (Linnaeus, 1758)	Passeriformes	1	0	0
16	100						19.5.2019	49.146588162442	21.008012876091			<i>Turdus pilaris</i> Linnaeus, 1758	Passeriformes	1	0	0
17	100						24.5.2019	49.110700859248	21.082580968915			<i>Dendrocopos major</i> (Linnaeus, 1758)	Piciformes	1	0	0
18		82		18			26.5.2019	49.13673156266	21.021563690676			<i>Turdus pilaris</i> Linnaeus, 1758	Passeriformes	1	0	0
19			100				1.6.2019	49.084968602583	21.085917636928			<i>Phoenicurus ochruros</i> (Gmelin, 1789)	Passeriformes	0	1	0
20	1		99				1.6.2019	49.139052000523	20.993864223537			<i>Delichon urbicum</i> (Linnaeus, 1758)	Passeriformes	1	0	0
21	100						5.6.2019	49.102203945993	21.098376497802			<i>Turdus pilaris</i> Linnaeus, 1758	Passeriformes	1	0	0
22	100						28.6.2020	49.090614868813	21.113559557118			<i>Picus viridis</i> Linnaeus, 1758	Piciformes	1	0	0
23			100				10.7.2020	48.994520810458	20.918518290101			<i>Accipiter nisus</i> (Linnaeus, 1758)	Accipitriformes	1	0	0
24		84	5		11		2.8.2020	48.721699885128	21.921578630984			<i>Otus scops</i> (Linnaeus, 1758)	Strigiformes	1	0	0
25		13	78		9		7.4.2021	49.122847565645	21.048701986849			<i>Anas platyrhynchos</i> Linnaeus, 1758	Anseriformes	1	0	0
25		13	78		9		7.4.2021	49.122847565645	21.048701986849			<i>Turdus merula</i> Linnaeus, 1758	Passeriformes	1	0	0
26			100				15.9.2021	49.146567108406	21.026107058107			<i>Turdus merula</i> Linnaeus, 1758	Passeriformes	0	1	0
10			95		5		12.7.2021	49.088446437124	21.080018048064			<i>Lanius collurio</i> Linnaeus, 1758	Passeriformes	1	0	0
27			100				13.7.2021	49.089091367657	21.07932208496			<i>Carduelis carduelis</i> (Linnaeus, 1758)	Passeriformes	1	0	0

First record of *Neochondriodes subfasciatus* (Westwood, 1848) (Megaloptera: Corydalidae) in India

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Abstract: The occurrence of the fishfly *Neochondriodes subfasciatus* (Westwood, 1848) in India is confirmed. It is the second species of the genus *Neochondriodes* Van der Weele, 1909 known from India.

Keywords: Megaloptera, India, Bangladesh, Myanmar, distribution, new record

Introduction

Neochondriodes Van der Weele, 1909 is the largest genus of the subfamily Chauliiodinae, which includes more than 40 species and is widely distributed through tropical and temperate Asia. Eight species of *Neochondriodes* are known from India and adjacent regions of South Asia (Liu et al. 2010, 2019). These *Neochondriodes* species are narrowly distributed in the northern part of the Indian subcontinent. Six species are to be known from India: *N. fletcheri* Kimmins, 1954, *N. flinti* Liu & Hayashi, 2019, *N. indicus* (van der Weele, 1907), *N. obscurus* van der Weele, 1909, *N. simplex* (Walker, 1853), and *N. truncatus* Kimmins, 1954 (Liu et al. 2010, 2019). In this contribution, the seventh species of *Neochondriodes* is recorded from India.

Material examined

Neochondriodes subfasciatus (Westwood, 1848)
India, Meghalaya, 1 km E of Tura, 25°30'N, 90°14'E, 500–2600 m a.s.l., 13.–18.v.2002, 1 ♂ (Figure 1), M. Trýzna & P. Benda leg., L. Dvořák det. et coll. X.Y. Liu revid. via photo. The investigated material originates from the estate, and unfortunately, detailed information about the collection method has not been preserved. The examined material originates from the estate, and unfortunately, detailed information about the collection method has not been preserved.



Figure 1. Male specimen of *Neochondriodes subfasciatus* (Westwood, 1848) from Tura, India. Photo: Libor Dvořák.

Results and Discussion

In the paper on the genus *Neochondriodes* of India and adjacent regions of South Asia (Liu et al. 2010), only two records of *N. subfasciatus* were published: three specimens from Bangladesh, Sylhet and three specimens from Caty [untraceable locality]. The collecting site Caty is ambiguous, even though it is probable that the location is somewhere in northern India or its adjacent regions (Liu et al. 2010). Later on, two specimens from Myanmar, Zi Yar Dam village, were published by Liu & Dvořák (2017). The present record of *N. subfasciatus* from Meghalaya represents the first record of this species from India (Figure 2).



Figure 2. The present known distribution of *Neochondriodes subfasciatus* (Westwood, 1848). Map source: maps.google.com/.

Conclusions

Altogether, seven species of the genus *Neochondriodes* are known from India now. *Neochondriodes nepalensis* Liu, Hayashi & Yang, 2010 is known from two localities in Nepal only (Liu et al. 2010), but its occurrence in India cannot be excluded.

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First record of *Sclerodermus* Latreille, 1809 (Hymenoptera: Bethylidae) and report of the first case of human stings in Slovakia

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Abstract

The first stings by a *Sclerodermus* species in humans is reported from Slovakia, where this genus has been overlooked for a long time. It is probable that individuals of this species arrived into the house in untreated piece of wood, where they parasitised wood-boring insects.

Key words: ant-like insects, painful and extremely itchy stings, Middle Europe

Introduction

The genus *Sclerodermus* belongs to parasitoid wasps of family Bethylidae (Hymenoptera). In Europe, 23 species occur sporadically (Polaszek 2022). No species of this genus has been formally recorded in Slovakia until now (see Macek et al. 2007; Polaszek 2022).

These small- to medium-sized, frequently dark-coloured wasps are commonly known as “flat wasps” (Azevedo et al. 2018; Colombo et al. 2020; Brewer-Carías et al. 2020). The females of many bethylids are ant-like in appearance, frequently apterous, but some species are polymorphic, and females could be both apterous and macropterous (e.g., Colombo & Azevedo 2020; Brewer-Carías et al. 2020). Flat wasps parasitise larvae of Lepidoptera and Coleoptera. For example, bethyline species tend to attack microlepidopteran families, such as the Gelechiidae, Noctuidae and Tortricidae, whereas there are many reports that epyrine and pristocerine species mainly attack coleopteran families, such as the Anobiidae, Bostrychidae, Buprestidae, Cerambycidae and Tenebrionidae (Gordh & Móczár 1990). Few species have been reported to sting humans, causing slight to severe pain and allergic reactions to those who suffered the wasp attack (Oda et al. 1981; Viglizzo et al. 2002; Veraldi et al. 2010; Papini 2014; Almeida et al. 2017; Skvarla 2018; Simon et al. 2020; Brewer-Carías et al. 2020). In Europe, most cases are reported from Italy and Spain. All cases were associated with (direct or indirect) handling of wood (Veraldi et al. 2010; Papini 2014).

The knowledge about the clinical picture of *Sclerodermus* human stings and the presence of this genus in Slovakia has not been published yet.

Material examined

Slovakia, Košice, 16.1.2022: 4♀ (Laboratory and Museum of Evolutionary Ecology, University of Prešov) (Figure 1). Notes: Small wasps, larger than 3.0 mm. Body slender and flattened. Fully described in Azevedo et al. (2018). *Sclerodermus* (apterous females) was recorded in the

house (a family house) close to the terrarium containing leopard gecko *Eublepharis macularius* (Blyth 1854) (Eublepharidae). In the summer, a piece of wood from the forest around Košice city (48°42'11"N, 21°17'25"E) was placed in the terrarium (Figure 2).

First record of human stings in Slovakia – case description

Individuals of *Sclerodermus* were registered and collected for the first time after stinging a person living in the household. The first, but not clarified, sting appeared 13 January 2022 on the left forearm of the keeper, apparently due to accidental squeeze of the insect during manipulation of the terrarium equipment. Because of the small size of the causative agent, it was overlooked. It was unexpectedly painful and extremely itchy. After few days, moving insects were observed in terrarium. They were crawling on the sand substrate and on the geckoes, also. The gecko was probably stung (a small swelling in the head area and scratching was observed). During the following night, more bites occurred on the keeper, who slept near the terrarium. Five intense pruritic lesions suggestive of insect bites were localised on the left arm. Insects that were smaller than gaps in terrarium aeration system had escaped to adjacent furniture and invaded the room. The “corpus delicti” was found next day when the keeper noticed small wasps on her nightwear, with other two bites to the breasts and one in abdomen located. The stings produced itchy papular lesions (Figure 3) similar to those reported for *S. domesticus* (e.g., Serini et al. 2010; Veraldi et al. 2010; Viglizzo et al. 2002). These were healed with topical corticosteroid creams such as Triamcinolonacetonid and Betamethason. For the first three days, they were combined with oral antihistaminic treatment (Desloratadin tbl.), due to vehement pruritus and pain. The topical therapy lasted for 10 days.

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Figure 1. *Sclerodermus* female specimen from the stung person.



Figure 2. A terrarium and a piece of wood that is most likely the source of the pest.



Figure 3. Papular lesions caused by *Sclerodermus* stings.

Conclusions

The genus *Sclerodermus* has been overlooked in Slovakia for a long time. The fact that the first discovery is published only now is probably due to the lack of experts who would deal with this group in Slovakia. Although we publish the first documented case of human stings, rare stings may have occurred in the past. However, stung people and certainly medical doctors cannot identify the origin of the stings. Even if individuals are observed or caught, misidentification probably occurs due to the small size and similarity with ants. A person can most often come into contact with wasps in everyday life, e.g. untreated old or antique furniture (e.g. in Italy, Spain, etc. there have been several cases of dermatitis associated with these wasps (Veraldi et al. 2010; Papini 2014)), but also wooden panelling, floors, ceilings, beams in old houses. A high-risk group, in addition to the general public, is e.g. restorers working with wood, carpenters, antique dealers, etc.

Stings were unexpectedly painful and extremely itchy. The source of contamination was identified in a worm-eaten piece of wood from the forest placed in a terrarium. Although the first documented case of human stings

is published here, bites may have occurred in the past, albeit rarely. Physicians, dermatologists, medical staff, and public health entomologists, as well as specific categories of workers, should be made aware of the risk of exposure to *Sclerodermus* stings.

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