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## Search of pattern in presence of epiphytic lichens growing in an urban environment – case study in Prešov city (Slovakia)

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### Abstract

Lichens are sensitive organisms, widely applied in air quality assessments and monitoring programs around the world. Traffic emissions or particulate matter influence the pattern in presence of lichens on roadside trees. The research of epiphytic lichens in Prešov city (Slovakia) was conducted in 17 near-road localities (three trees per locality). The traffic density, selected tree parameters, lichen density, and presence in research squares (15 x 15 cm) on 4 sides of the tree (side in the direction of traffic, in the opposite direction of traffic, the side facing towards the road and the side facing away from the road) were analysed. Three lichen species were most often determined on the trees: *Xanthoria parietina* (L.) Th. Fr., *Phaeophyscia orbicularis* (Necker) Moberg, and *Physcia adscendens* (Th. Fr.) H. Olivier. The most common lichen was the *Ph. orbicularis* accounted for up to 86% of all records. The highest presence was confirmed in localities with the highest traffic density. Based on the results it is obvious that atmospheric pollution alters lichen communities and therefore can be effectively used as biological indicator of air quality.

**Keywords:** urban area, *Phaeophyscia*, *Physcia*, *Xanthoria*, traffic density

### Introduction

It is well established that lichens are sensitive to a wide range of habitat changes, most of them man-driven (Pinho et al. 2004). Changes in lichen diversity are used as indicators of environmental conditions and have been widely applied in air quality assessments and monitoring programs around the world (Llop et al. 2012). In Europe is often used method LBI (evaluation of the lichen biodiversity index, e.g. Cioffi 2009; Giordani et al. 2002; Caoduro et al. 2014, etc.) and European guideline for mapping lichen diversity (e.g. Asta et al. 2002). The point quadrat method (e.g. Dunford et al. 2006; Martinez et al. 2006) or lichen based index (e.g. Will-Wolf et al. 2015) are used less often.

Tolerance to most of the heavy metals and the slow growth rate are the main factors which make lichens good indicators or/and monitors of metal pollution (e.g., Seaward & Letrouit-Galinou 1991; Paoli et al. 2019; Tarawneh et al. 2021). Biomonitoring with lichens can be done according Branquinho (2001) in three ways: using variations in diversity and/or abundance, using variations in physiological parameters, or using lichens as accumulators of pollutants. Lichen diversity has been used as an indicator for monitoring the effects of air pollution in urban areas since 20<sup>th</sup> century (Llop et al. 2012). Variations in lichen biodiversity may be due to changes in microclimatic conditions particularly light, water and nutrients. These alterations may be driven by local sources of disturbance, such as roads or farms, different land uses or habitat fragmentation (e.g., Jonsson & Jonsell 1999; Sillett & Goslin 1999; Moen & Jonsson 2003; Paoli et al. 2013; Asta et al. 2019).

It has been found in earlier studies that air quality in Central Europe has significantly deteriorated during second half of the 20<sup>th</sup> century (Lisowska 2011; Maňková et al. 2017). Traffic-related pollutants are considered one the biggest contributors to the urban air pollution (Stenson et al. 2021). The main pollutants in urban environments, roadsides and areas near the roads are the gases emitted from motor vehicles, such as sulphur dioxide (SO<sub>2</sub>), carbon monoxide (CO) and nitrogen dioxide (NO<sub>2</sub>) (e.g., Gilbert et al. 2007, Marmor & Randlane 2007). Traffic emissions or particulate matter influences the diversity and abundance of lichens on roadside trees. Non-tolerant lichens react to the presence of contaminants by inhibiting the growth, on the contrary pollution-tolerant species, thrive in such an environment (Marmor & Randlane 2007; Stapper 2012).

This study aims to i) determine the most frequently occurring epiphytic lichen taxa growing around the roads in the city of Prešov using modified quadrat method, ii) find out the relationship between the presence of epiphytic lichens and their orientation to the direction of traffic, iii) determine the relationship between the traffic density and the epiphytic lichens presence.

### Materials and methods

A simple study of epiphytic lichens to assess the environmental quality of the Prešov (Slovakia) city during May 2019 for the first time was conducted.

The roads were selected for research as follows:

at least 3 trees had to grow by the road, ideally on each side of the road;

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the trees near road had to be of the same species and approximately the same size;  
the minimum tree circumference had to be at least 80 cm (at a height of 1.3 m above the ground).

(note: The tendency should be to use only trees with acid bark, preferably of one species only; which was unrealistic for very specific urban conditions e.g. van Herk et al. 2002; Gombert et al. 2004).

A total of 17 near-road localities were selected for the research purposes. At each sampling locality three trees were randomly selected (if possible – on each side of the road – which was fulfilled only in 4 locations). For each tree, the genus of the tree, the perimeter of the trunk at 1.3 m height, and the radius of the crown were determined. Basic data on selected localities are given in Table 1.

Table 1. Basic information of sampled roads.

no.	GPS	traffic densities
001	48°59'27.3"N 21°14'02.9"E	LD
002	48°59'29.0"N 21°14'06.1"E	LD
003	48°59'29.5"N 21°14'11.0"E	LD
004	48°59'31.8"N 21°14'13.8"E	LD
005	48°59'29.5"N 21°14'14.3"E	MD
006	48°59'53.2"N 21°13'17.8"E	MD
007	48°59'41.4"N 21°13'40.2"E	LD
008	48°59'52.1"N 21°13'17.5"E	MD
009	48°59'58.9"N 21°13'33.2"E	HD
010	49°00'06.3"N 21°13'24.7"E	MD
011	49°00'14.7"N 21°13'16.3"E	HD
012	48°59'57.0"N 21°13'38.0"E	HD
013	48°59'41.5"N 21°13'54.8"E	MD
014	48°59'45.2"N 21°13'59.0"E	LD
015	48°59'42.5"N 21°14'06.6"E	LD
016	48°59'24.3"N 21°14'26.4"E	MD
017	48°59'18.9"N 21°14'12.2"E	HD

The traffic density was determined for individual roads as mentioned in Demková et al. (2019). Based on the results, the roads were divided into three traffic density categories: high traffic density (HD): > 600 cars per hour; medium traffic density (MD): 599 – 200 cars per hour; low traffic density (LD): < 199 cars per hour.

The coverage of each lichen was estimated by the modified quadrat method as follow. On each tree in 3 different heights (up to 40 cm from the ground, height approx. 130 cm from the ground and approx. 180 cm from the ground) were drawn squares measuring 15 x 15 cm. These squares were drawn on 4 sides of the tree (side in the direction of traffic (marked as 3), in the opposite direction of traffic (marked as 4), the side facing towards the road (marked as 1) and

the side facing away from the road (marked as 2). A total of 12 squares were drawn on each tree (Figure 1). Overall, it was evaluated 756 squares on 63 trees near 17 roads.



Figure 1. Ash tree with drawn squares at locality 001 – Kúpeľná street.

Subsequently, the percentage coverage of the total square area by lichen species was determined at each square (whole area = 100%, Figure 2).



Figure 2. Example of a marked square with lichens.

All information was recorded and photographically documented directly in situ. Subsequently, they were digitized in the laboratory by estimation based on visual inspection and statistically analysed. Only those species of

lichens whose coverage in the square was at least 1% were analysed. The possible influence of individual factors (e.g., traffic density, orientation to the road) was analysed using the nonparametric Kruskal-Wallis test in particular given that the distribution of data on the cover (representation) of individual species was not normal and Levene's test confirmed the inequality of deviations (inhomogeneity of variation). All statistical analyses and descriptive statistics were performed in the program JASP (2020), for creating box-diagrams (boxplots) program PAST (ver. 4.03, Hammer et al. 2001) was used.

## Results

For the research purposes, following trees growing near the roads in the Prešov city were used: ash (*Fraxinus* spp.), boxelder maple (*Negundo* sp.), maple (*Acer* spp.), linden (*Tilia* spp.), chestnut (*Aesculus* sp.) and cherries (*Cerasus* spp.). Ash was found as the most frequently occurred tree accounting for about 27% of all examined trees, followed by maples (24%), chestnuts (19%), lindens (14%), boxelder maple (11%), and cherries (5%) (Table 2).

Table 2. Number and percentage of measurements on trees of different species.

tree-species	frequency	percent
<i>Acer</i> spp.	180	23.8
<i>Aesculus</i> sp.	144	19.0
<i>Fraxinus</i> spp.	204	26.9
<i>Negundo</i> sp.	84	11.1
<i>Cerasus</i> spp.	36	4.80
<i>Tilia</i> spp.	108	14.3
Total	756	100

The perimeter of the trunk at a height of 130 cm from the ground reached values from 68 cm to 205 cm. The average circumference of the trunk was 136 cm. The radius of the crown of the examined trees reached values from 2 m to 6 m. The average radius was 5 m (Table 3).

Table 3. Descriptive statistics of the properties of sampled trees – perimeter of the trunk and radius of the crown.

	perimeter of the trunk (cm)	crown radius (m)
Valid	756	456
Missing	0.00	0.00
Mean	136	4.97
Median	136	5.00
Std. deviation	33.1	1.05
Minimum	68.0	2.00
Maximum	205	6.00

Most of the measurements were performed at the low traffic density locations (43%), followed by medium traffic density locations (33%) and high traffic density locations (24%) (Table 4)

Table 4. Number and percentage of measurements in localities with different traffic density (LD – low traffic density; MD – medium traffic density, HD – high traffic density).

traffic intensity	number of measurements	percent
LD	324	42.9
MD	252	33.3
HD	180	23.8
Total	756	100

On the examined trees, most often 3 species of lichens were found (species with a coverage of less than 1% were not included in the analyses): Maritime sunburst lichen – *Xanthoria parietina* (L.) Th. Fr., *Phaeophyscia orbicularis* (Necker) Moberg, and *Physcia adscendens* (Th. Fr.) H. Olivier. The most common lichen growing on the evaluated trees was *Ph. orbicularis* (86% of records), followed by *Ph. adscendens* (12%) and *X. parietina* (2%). We also observed the highest proportion (average, median and maximum) in all recorded lichen species (Table 5).

Table 5. Descriptive statistics of the presence of individual lichen species.

	<i>Ph. orbicularis</i>	<i>Ph. adscendens</i>	<i>X. parietina</i>
Valid	756	756	756
Missing	0.00	0.00	0.00
Mean	19.8	2.81	0.34
Median	10.0	0.00	0.00
Std. Deviation	22.8	7.98	2.45
Minimum	0.00	0.00	0.00
Maximum	100	70.0	40.0

The possible influence of individual factors (the traffic density and orientation to the road) was analysed only for the most frequently occurring species *Ph. orbicularis*. We found that the presence of *Ph. orbicularis* was influenced by the density of traffic and the orientation of the measured area towards the road (Table 6, Figure 5). The highest presence of *Ph. orbicularis* was determined in the localities with the highest traffic density (Figure 3) and in the northern and western areas of the trunk. The lowest presence of *Ph. orbicularis* was found in the south-facing areas of the trunk (Figure 4).

Table 6. The results of nonparametric Kruskal-Wallis test expressing the differences in the presence of the *Ph. orbicularis* depending on the traffic density and orientation to the road.

factor	statistic
traffic intensity	41.654
orientation-road	14.739

As can be seen from Figure 3, the presence of *Ph. orbicularis* in localities with a low density of traffic represented approximately 12% to 17% of the records, at a medium traffic density of about 18% to 25% of the records. The highest presence (23 – 30%) was recorded at the localities with high traffic density.

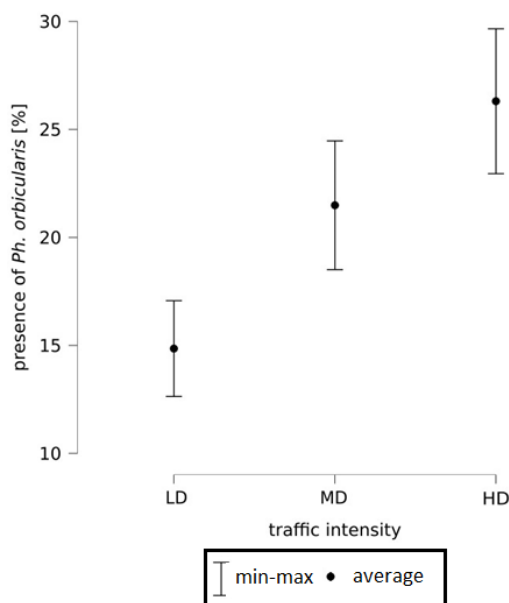


Figure 3. Presence of the species *Ph. orbicularis* in localities with different traffic density (x-axis, LD – low density, MD – medium density, HD – high density).

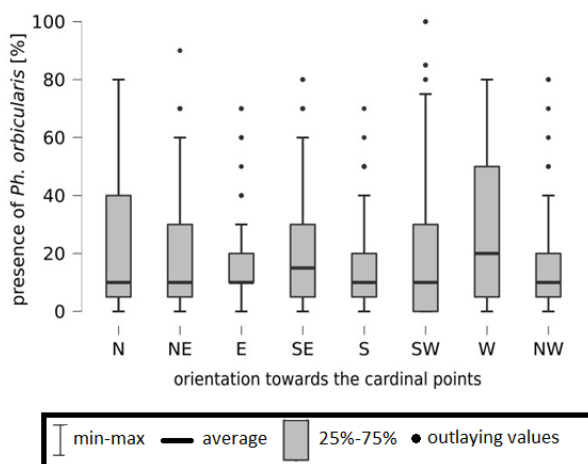


Figure 4. The presence of the *Ph. orbicularis* in squares with different orientations to the world sides (x-axis, sides of the world).

From the point of view of the presence of *Ph. orbicularis* in relation to the world sides (Figure 4), this species predominated in the west (W) and north (N), subsequently in the south-west (SW), south-east (SE) and north-east (NE). Less often *Ph. orbicularis* has occurred in the east (E), north-west (NW) and south (S), but these results are statistically insignificant.

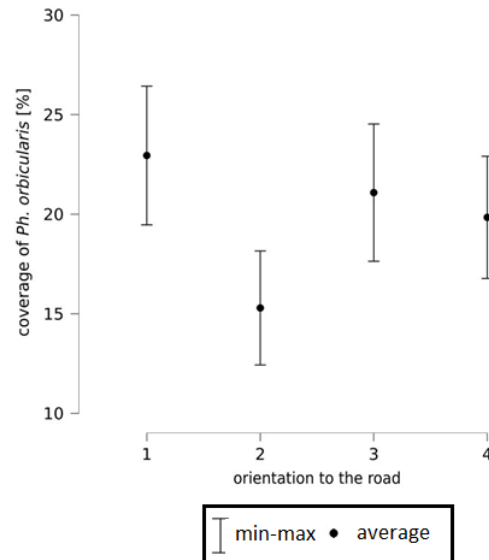


Figure 5. Coverage of *Ph. orbicularis* according to the orientation towards the road (diameters with standard error, 95% interval (side facing towards the road 1 and side facing away from the road 2, side in the direction of traffic 3, against the direction of traffic 4).

From the point of view of the cover of the species *Ph. orbicularis* and orientation to the road (Figure 5), we found, that this species grew most frequently on the side of the tree facing towards the road (19% to 27% cover), then on the side in the direction of traffic (18% to 25% coverage), followed by the opposite direction of traffic side (16% to 23% coverage). The lowest cover of this species was recorded on the side of the tree facing away from the road (12% to 18% cover).

## Discussion

Atmospheric pollution alters lichen communities, and depending on the nature and concentration of pollutants, usually results in an impoverishment in terms of richness and/or abundance (Gries 1999). In many earlier studies, lichens have been recognized and successfully utilized as biological indicators of air quality (Shukla & Upreti 2012). Demková et al. (2019) confirmed a significant positive correlation between risk elements and the traffic density. The species of lichens which were in greater numbers recorded in our study (namely *X. parietina*, *Ph. orbicularis*, and *Ph. adscendens*) were frequently represented in other European urban areas (Farkas et al. 2001; Matwiejuk & Chojnowska 2016; Djekic et al. 2017). Additionally, in the study of Piervittori & Maffei (2001) who evaluated the

presence of lichens in the urban area in Italy (the city of Aosta), the species *Ph. adscendes*, *X. parietina*, and *Ph. orbicularis* were among the most frequently recorded. The species which were recorded as the most abundant along Prešov city streets, were in earlier studies rated as the most tolerant to environmental stress or air pollution (Elsinger et al. 2007; Dymytrova 2009; Vitali et al. 2019; Owczarek-Kościelniak et al. 2020).

The most common lichen growing on the road was *Ph. orbicularis* accounted for up to 86% of all records. According to Van Herk et al. (2001) and Loppi & Pirintos (2000) the rise in the number of necrophilous lichens especially members of the Physciaceae family (including *Phaeophyscia*) has been attributed to climate change and also to dry and dusty conditions in urban centres.

Gombert et al. (2005) found, that the presence of *Ph. orbicularis* was influenced by the density of traffic and the orientation of the measured area towards the road. From the point of view of the cover of the species *Ph. orbicularis* and orientation to the road, we found that this species grew most frequently on the side of the tree facing towards the road to the road. The lowest cover was recorded on the side of the tree facing away from the road. Gombert et al. (2005) use autecological and environmental parameters for establishing the status of lichen vegetation of urban areas in France and in agreement with our results he confirmed that *Ph. orbicularis* is frequent and tolerant species. Shukla & Upreti (2012) have designated this species as frequency and abundance thermophilus and poleotolerant lichens.

The highest presence was recorded in the localities with the highest traffic density and in the northern and western areas of the trunk. It is likely that the climatically less exposed sides (west, north), which have less light and more humidity, are more suitable for the growth of lichens (Gombert et al. 2005). But, the presence, absence and the diversity of lichen species is influenced by many other characteristics such as the tree size, tree age, diameter, branch density, bark chemical properties, bark structure, its water holding-capacity, and also the position of the tree in the stand (Raniu et al. 2008; Johansson et al. 2009; van Her et al. 2002; Bäcklund et al. 2016). Some authors have stated that the pH of the bark and susceptibility to toxic substances are among the most important properties affecting the presence and composition of lichens (Jüriado et al. 2012). Transport pollution substances ( $\text{CO}_x$ ,  $\text{NO}_x$ ,  $\text{SO}_2$ ) causing acidification of the environment (Sebal et al. 2022), what can lead to the reducing bark pH. Since, according to our results, the highest presence of *Ph. orbicularis* was recorded on the high traffic density localities, with the highest abundance at the side facing the road, we assume that the recorded lichen belongs to pollution-resistant and favouring acidic environment species.

## Conclusions

In Prešov city three lichen species that most often grow on the trees near roads are *Xanthoria parietina*, *Phaeophyscia orbicularis*, and *Physcia adscendens*. The most common

lichen was *Ph. orbicularis* accounted for up to 86% of all records. We found that the presence of *Ph. orbicularis* was influenced by the density of traffic and by the orientation towards the road. The highest presence was confirmed in localities with the highest traffic densities. *Ph. orbicularis* grew most frequently on the side of the tree facing towards the road to the road, and the lowest cover of this species was recorded on the side of the tree facing away from the road.

We therefore conclude that presence and quantity of *Ph. orbicularis* growing on a tree may be a suitable indicator of traffic density, as well as traffic contamination.

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## Aktivita pôdných mikroorganizmov ovplyvnená výskytom *Fallopia japonica*

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### Abstract. Activity of soil microorganisms affected by occurrence of *Fallopia japonica*

The present work deals with the activity of microorganisms in sites that are invaded by *Fallopia japonica* in eastern of Slovakia. The aim of the study was to determine physico-chemical (soil pH, organic carbon content, total nitrogen content, soil gravimetric moisture) and microbial parameters (soil respiration, enzymes:  $\beta$ -glucosidase, FDA hydrolase, acid and alkaline phosphatase) that influence quality and health of soil ecosystem. The study was carried out within three climate localities represented by warm, mediate warm and cold conditions. Significant correlations were found between the parameters depending on location and altitude. Our results show that invasive plant species significantly alter the values of physicochemical and microbial parameters.

**Keywords:** invasive plants, physico-chemical parameters, microbial indices, enzymatic activity

### Úvod

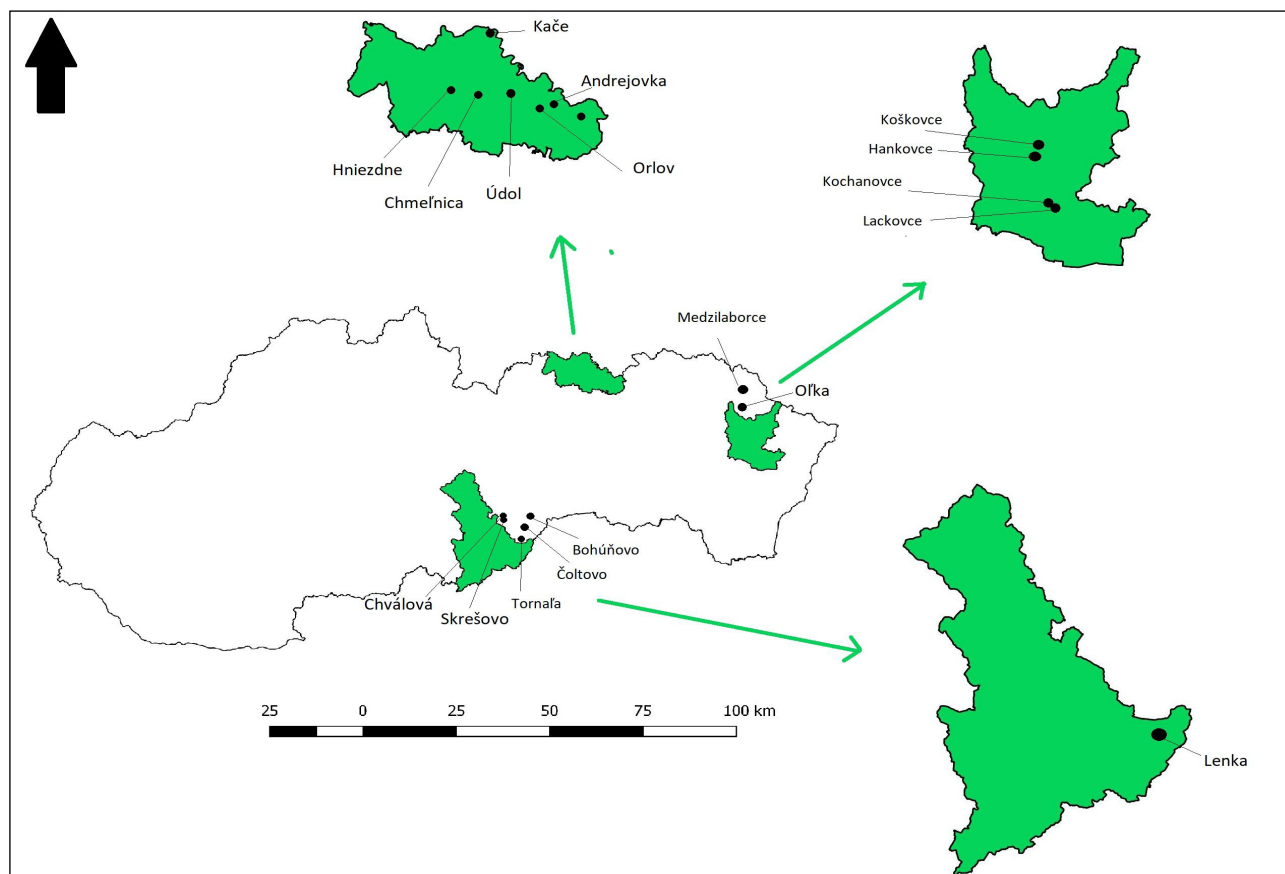
Pôda je nenahraditeľným prírodným zdrojom, ktorý umožňuje produkovať suroviny a potraviny, filtruje a zadržiava vodu, zabezpečuje kolobeh látok v prírode, udržiava diverzitu rastlín a živočíchov a formuje kvalitu životného prostredia (Fazekašová & Bobulská 2012). Každá pôda má svoje fyzikálne, biologické a chemické vlastnosti, ktoré ovplyvňujú kvalitu a zdravie pôdy (Javoreková 2008). Pod pojmom kvalita pôdy rozumieme schopnosť pôdy fungovať v rámci ekosystému, udržiavať kvalitu prostredia a podporovať zdravie rastlín, živočíchov a človeka (Yakovchenko et al. 1996). Kvalitu odvodzujeme zo zmien jej parametrov (indikátorov) (Wick et al. 2002). Často využívané sú biologické parametre pôdy (Fazekašová & Bobulská 2012), najčastejšie a najefektívnejšie sú mikrobiologické charakteristiky, ktoré vďaka svojej reaktivite, generačnej dobe a diverzite pôdnej mikroflóry umožňujú okamžitú reakciu na rôzne podnety prostredia (Kubát et al. 2002). Medzi významné mikrobiálne parametre patrí: pôdna respirácia, pôdna mikrobiálna biomasa, aktivita pôdných enzýmov a pod. (Fejér & Bobulská 2015). Pojem invázia, odvodený od lat. slova „*invado*“, znamená vpadnúť alebo násilne vstupovať (Cvachová & Gojdičová 2003). Invázie sú prítomné vo všetkých biotopoch (Záhorská 2016). Vytlačajú domáce druhy, čím menia štruktúru a funkciu pôvodného ekosystému (Nentwig 2014). Cieľom tejto práce bolo stanoviť aktivitu pôdných mikroorganizmov v porastoch invadovaných druhom *Fallopia japonica* na území troch klimatických oblastí východného Slovenska.

### Materiál a metódy

Odbery pôdných vzoriek boli realizované v troch klimatických oblastiach (Obrázok 1): Stará Ľubovňa

(okolie rieky Poprad), Humenné (okolie rieky Laborec) a Rimavská Sobota (okolie rieky Slaná). Pre vyššie uvedené lokality platí, že boli výrazne invadované rastlinou *Fallopia japonica*. Na všetkých troch výskumných lokalitách bolo stanovených 20 odberných miest (10 pre invadované a 10 pre neinvadované porasty) s rozmerom 1 m x 1 m, čo predstavovalo celkovo 60 pôdných vzoriek. Každá vzorka pozostávala z troch čiastkových vzoriek odobratých z hĺbky 0,1 – 0,2 m, ktoré boli následne po odbere vzoriek homogenizované. Pred samotnými analýzami boli všetky vzorky preosiate cez sito s veľkosťou ôk 2 mm a uložené v chladničke až do samotných analýz. Naším cieľom bolo stanoviť fyzikálno-chemické a mikrobiálne parametre (pH, obsah organického uhlíka, celkový dusík, gravimetrická vlhkosť pôdy, pôdna respirácia, enzýmy:  $\beta$ -glukozidáza, FDA hydroláza, kyslá a zásaditá fosfatáza). Aktivita jednotlivých pôdných enzýmov bola stanovená za použitia konkrétnych substrátov, tlmivých roztokov a podmienok jednotlivých metodických postupov (Eivazi & Tabatabai 1988, Green et al. 2006, Grejčovský 1991). Pre každú aktivitu pôdneho enzýmu sa vykonala zodpovedajúca kontrola rovnakou analytickou metódou, ale bez pridania substrátu v okamihu začatia enzymatickej reakcie. Aktivita enzýmov bola stanovená spektrofotometricky (UV Spektrofotometer 1800, Shimadzu) vytvorením kalibračnej krivky. Jednotlivé koncentrácie enzýmov boli stanovené pri rôznych vlnových dĺžkach (464 nm –  $\beta$ -glukozidáza, 490 nm – FDA hydroláza, 510 nm – fosfatázy). Zistené údaje boli štatisticky vyhodnotené v programe STATISTICA 12 a všetky štatistické operácie boli vykonané v programe PAST 4.03. Dáta boli pred samotnou analýzou logaritmicke transformované.

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Obrázok 1. Mapa odberných miest na Slovensku.

### Výsledky a diskusia

#### Hodnotenie fyzikálno-chemických a mikrobiologických parametrov

V Tabuľke 1. uvádzame získané výsledky fyzikálno-chemických a mikrobiálnych parametrov, kde porovnávame vzorky pôdy na lokalitách invadovaných rastlinou *Fallopia japonica* (F) a kontrolné vzorky pôdy bez invázie (C). Podľa Stefanowicz et al. (2016) rastlinné invázie môžu ovplyvniť vlastnosti pôdy ako

napr. pH, vlhkosť pôdy, štruktúru a funkciu pôdných mikroorganizmov. Na základe dosiahnutých výsledkov môžeme konštatovať, že pôsobenie inváznej rastliny v pôde signifikantne mení hodnoty sledovaných pôdných parametrov. Priemerné hodnoty pôdnej reakcie, obsah organického uhlíka a aktivity FDA sú štatisticky významne vyššie v invadovaných pôdach v porovnaní s pôdami neinvadovanými ( $p < 0,05$ ).

Tabuľka 1. Sledované pôdne parametre invadovaných (F) a neinvadovaných (C) pôd.

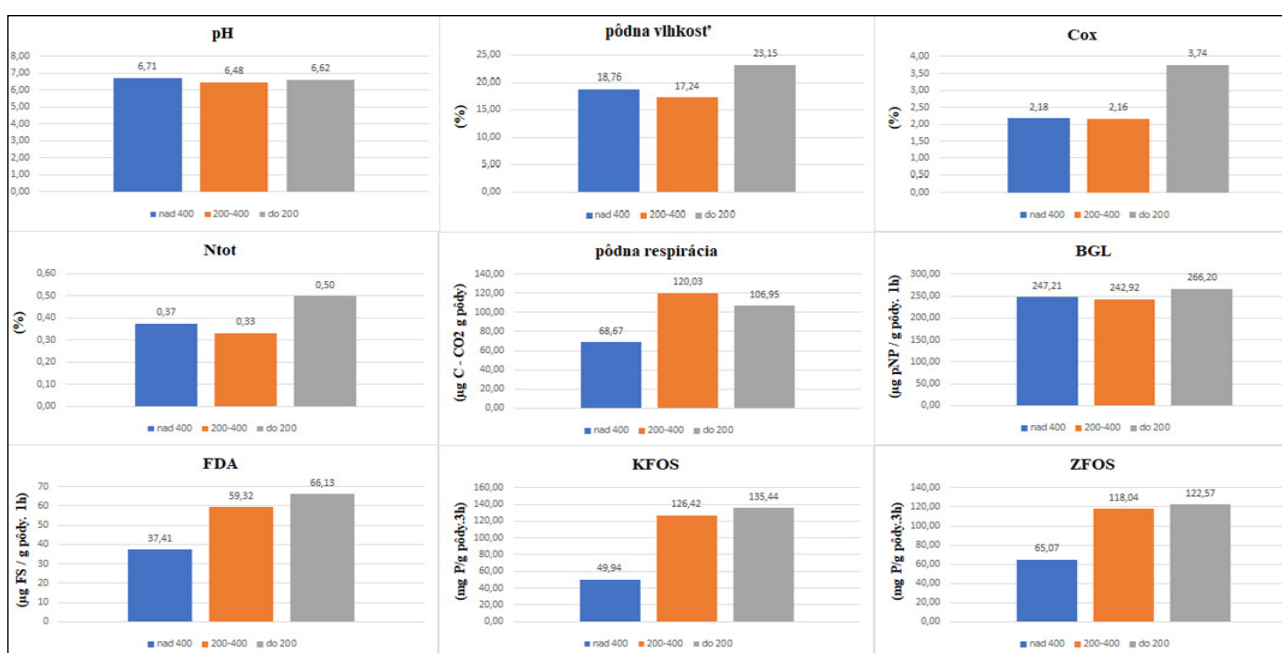
F/C		pH	Pôdna vlhkosť (%)	C <sub>ox</sub> (%)	N <sub>tot</sub> (%)	Pôdna respirácia (μg C-CO <sub>2</sub> /g pôdy)	BGL (μg pNP/g pôdy.1h)	FDA (μg FS/g pôdy.1h)	KFOS (mg P/g pôdy.3h)	ZFOS (mg P/g pôdy.3h)
F	min	5,83	6,58	1,20	0,15	48,6	161	27,4	10,9	41,1
	max	7,55	41,5	7,34	0,81	129	463	110	254	194
	PR	6,96	20,7	3,09	0,39	84,3	263	63,3	107	110
C	min	4,49	5,21	0,55	0,09	82,2	133	21,1	10,2	12,8
	max	7,3	29,1	3,11	0,57	201	343	89,9	213	196
	PR	6,32	17,3	1,84	0,34	195	251	55,5	181	157

F – vzorky pôdy invadované rastlinou *Fallopia japonica*, C – kontrolné vzorky bez invázie, PR – priemerné hodnoty, pH – pôdna reakcia, C<sub>ox</sub> – obsah organického uhlíka, N<sub>tot</sub> – obsah celkového dusíka, BGL – β-glukozidáza, FDA – fluoresceín diacetát, KFOS a ZFOS – kyslá a zásaditá fosfatáza.

Pôdna reakcia (pH) ovplyvňuje mikrobiálnu aktivitu a tým aj aktivitu enzýmov v pôde (Tobiašová 2020). Štúdia Acosta-Martínez & Tabatabai (2000) podporuje názor, že pH je dôležitým ukazovateľom kvality a zdravia pôdy. Pôdna vlhkosť je momentálny obsah vody v pôde v určitom čase, vyjadrený v percentách k hmotnosti/objemu suchej zeminy (Varga 2015). Pasternáková (2015) uvádza, že vyššie hodnoty vlhkosti pôdy sa predpokladajú na miestach napadnutých inváznymi rastlinami, kvôli plytkým koreňom a tým nižšiemu prísunu vody. Podľa Fejéra & Bobuľskej (2015) sa pri vyššej pôdnej vlhkosti obsah uhlíka zvyšuje, čo korešpondovalo s našimi dosiahnutými výsledkami. Medzi dôležité biogénne prvky patrí uhlík a dusík a podľa Pilkovej (2018) sa obsah dusíka uvádza v spojitosti s obsahom oxidovateľného uhlíka,

vyjadreného v pomere C:N. Najjednoduchším a najlepšie interpretovateľným mikrobiálnym parametrom je podľa Javorekovej (2008) mikrobiálna respirácia.

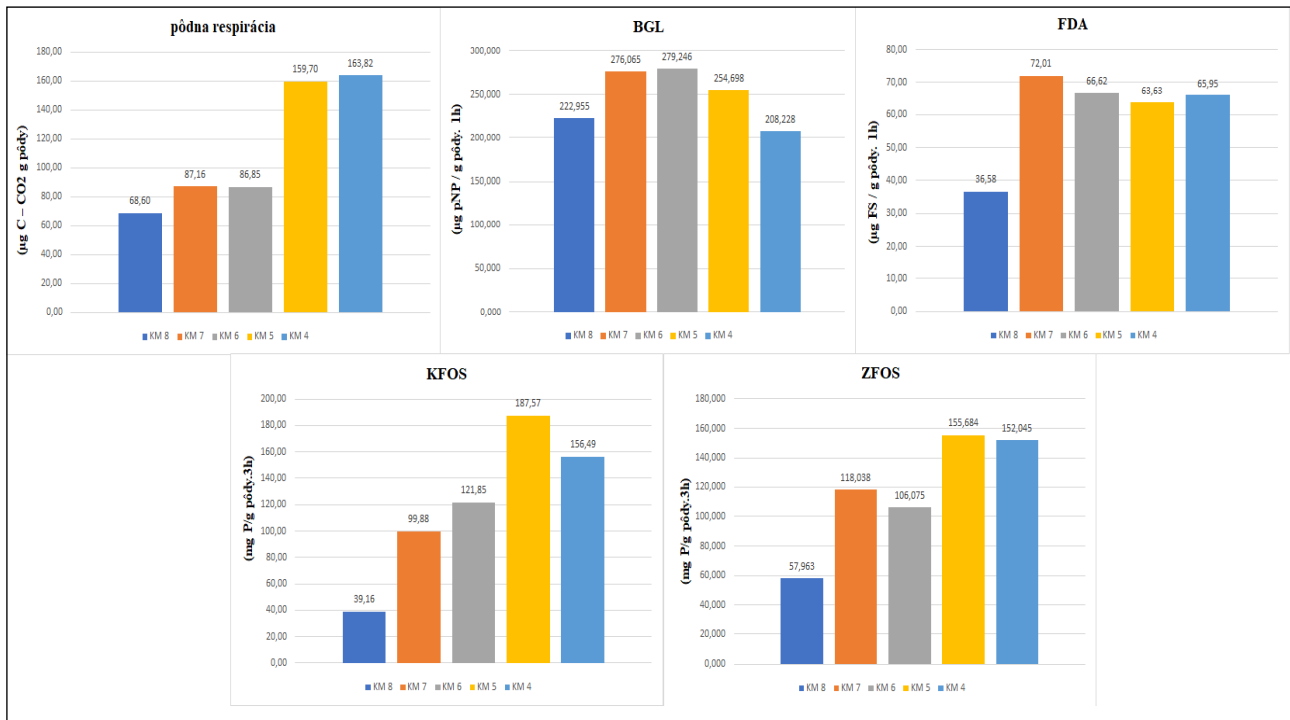
V predkladanej práci sme zisťovali vplyv nadmorskej výšky na sledované pôdne parametre zobrazené na Obrázku 2. Hodnotili sme všetky pôdne parametre bez ohľadu na lokalitu. Boli stanovené tri kategórie: do 200 m n. m., 200 – 400 m n. m. a nad 400 m n. m. Štatisticky významné rozdiely ( $p < 0,05$ ) boli zaznamenané pri všetkých pôdnych parametroch s výnimkou pôdnej reakcie (pH) a  $\beta$ -glukozidázy. Podľa Kumara et al. (2019) je nadmorská výška hlavným faktorom, ktorý ovplyvňuje biodiverzitu a fyzikálno-chemické vlastnosti pôdy. V ich štúdiu boli zaznamenané vyššie hodnoty pH vo vyšších nadmorských výškach, čo korešponduje s našimi výsledkami.



Obrázok 2. Priemerné hodnoty pôdnych parametrov podľa nadmorskej výšky.

Následne sme sledovali súvis medzi sledovanými pôdnymi parametrami a klimatickými regiónmi. Bolo stanovených päť klimatických regiónov: od klimaticky teplej po chladnú oblasť a hodnotených bez ohľadu na invadované územie a lokalitu a vymedzených podľa určitých kritérií (Obrázok 3). Klimatický región 4 a 5 (Rimavská Sobota) je charakterizovaný ako najteplejší a veľmi suchý spomedzi sledovaných regiónov. Klimatický región 6 a 7 (Humenné) je charakterizovaný ako teplý a mierne suchý región. Klimatický región 8 (Stará Ľubovňa) je charakterizovaný ako najchladnejší spomedzi sledovaných regiónov s najvyšším úhrnom zrážok.

Na Obrázku 3 sú zobrazené priemerné hodnoty mikrobiologických parametrov podľa klimatického regiónu. Štatisticky významne najvyššie hodnoty ( $p < 0,05$ ) boli preukázané pri pôdnej vlhkosti, organickom uhlíku a celkovom dusíku v klimatickom regióne 6 a 7. Rastúce klimatické zmeny výrazne menia aktivitu pôdnych enzýmov a tým aj kolobeh látok v pôde (Cao et al. 2021) a vývoj organického uhlíka (Sobocká et al. 2019). Na Obrázku 3. sú zobrazené priemerné hodnoty mikrobiologických parametrov podľa klimatického regiónu. Pri pôdnej respirácii, FDA hydrolýze, kyslej a zásaditej fosfatáze boli preukázané štatisticky významne najnižšie hodnoty parametrov ( $p < 0,05$ ) v klimatickom regióne 8, ktorý bol charakterizovaný ako pomerne chladný.



Obrázok 3. Priemerné hodnoty mikrobiologických parametrov podľa klimatického regiónu.

## Záver

Cieľom tejto práce bolo stanoviť aktivitu mikroorganizmov v porastoch invadovaných *Fallopia japonica* na území Slovenska. Odbery pôdných vzoriek boli realizované v troch klimatických oblastiach a na všetkých troch lokalitách bolo stanovených 20 odberných miest (10 pre invadované a 10 pre neinvadované alebo kontrolné vzorky pôdy). Naším cieľom bolo stanoviť fyzikálno-chemické a mikrobiálne parametre, ktoré ovplyvňujú kvalitu pôdy (pôdna reakcia, gravimetrická vlhkosť pôdy, obsah organického uhlíka, celkový dusík, pôdna respirácia a enzýmy:  $\beta$ -glukozidáza, FDA hydroláza, kyslá a zásaditá fosfatáza). Z našich výsledkov vyplýva, že pôsobenie inváznej rastliny v pôde mení hodnoty pôdných sledovaných parametrov. Taktiež pôsobenie klimatických podmienok a rôzne nadmorské výšky majú vplyv na mikrobiálnu aktivitu a ostatné pôdne parametre.

## Podakovanie

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## *Sarracenia* – an exotic hostel for European aquatic invertebrates?

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### Abstract

Pitcher plants provide temporal aquatic habitats, known as phytotelmata, which can be colonized by aquatic invertebrates. In this study, we tested, if phytotelma provided by an exotic pitcher plant *Sarracenia purpurea* L. could be colonized by native aquatic organisms. Thus, we exposed three plants to natural conditions for three months from June to August in E Slovakia and sampled the contained water in monthly intervals. As many as 15 taxa of the groups Rotatoria, Ciliophora, Flagellata, Nematoda, Tardigrada, and Diptera colonized the phytotelmata. The Rotifera species belonged to *Lecane bulla* (Gosse 1851), *L. inermis* (Bryce 1892), and *Colurella obtusa* (Gosse 1886). Dipterans were represented by families Ceratopogonidae, Chironomidae, Psychodidae, and Sciomyzidae. The results confirmed that phytotelmata provided by non-native plant species can potentially be colonized by native aquatic organisms, even though some microorganisms could have been remains of a previous colonization.

**Key words:** phytotelmata, pitcher plants, exotic plant species, cultivation, new environment, colonization, invertebrates, Insecta

### Introduction

Small periodic waters are undeniably interesting but often overlooked ecosystems. Kitching (2000) and Williams (2006) developed their detailed classification. An interesting type of small periodic aquatic habitats are phytotelmata i.e., small temporary habitats provided by water-filled cavities of terrestrial plants. In the climate conditions of Europe, compared to the tropics, the variability of these ecosystems is much smaller. In Europe, phytotelmata occur on plants of the genus *Dipsacus* and natural occurrence of tropical plants forming phytotelmata e.g., from Bromeliaceae or Nepenthales is fully excluded. On the other hand, representatives of these groups are often present as ornamental houseplants throughout Europe (e.g., Kitching 2000).

The natural area of distribution of *Sarracenia purpurea* L. is limited to the eastern part of the USA and Canada (Juniper et al. 1989) where it grows on low-nutrient soils in boggy locations, but allochthonous populations have been reported from Britain and Ireland (Walker 2014), Sweden (Almborn 1983), Switzerland (Marabini 1994; Parisod et al. 2005), and Germany (Marabini 1994; Fürsch 2001; Gebühr et al. 2006). The pitchers of this carnivorous plant are formed in tubular leaves that work as passive pitfall traps, even though at least one species produces attractant substances (Miles et al. 1975). A combination of downward-pointing hairs, waxy sections of the inner walls, the low surface tension of the chamber fluid, and the presence of insect-paralyzing compounds allows to retain prey species (Folkerts 1999). The type of prey captured is largely related to leaf morphology and varies among *Sarracenia* species, with spiders being the prevailing prey for *S. purpurea* (Williams 2006). The decomposition of prey residuals

occurs mainly by microbial extracellular enzyme activity (Bradshaw & Creelman 1984), although some *Nepenthes* and most *Sarracenia* species also produce digestive enzymes (Juniper et al. 1989). Despite the carnivorous nature of the plant, a number of invertebrate species have become residents in *Sarracenia* pitchers, even though the mechanism of the survival of these organisms is not fully understood. Most of the pitcher colonizers profit from the food resources i.e., the accumulation of the bodies of prey. Typical inhabitants of this habitat are dipterans, such as mosquitos (*Wyeomyia smithii* (Coquillett, 1901)), chironomids (*Metriocnemus knabi* Coquillett, 1904), sciarids and phorids (e.g., Aldrich 1916; Jones 1918; Buffington 1970; Cameron et al. 1977; Rymal & Folkerts 1982; Bradshaw 1983; Milne et al. 2008; Hamilton & Duffield 2002). From microscopic organisms there were recorded mites, microcrustaceans, rotifers, nematodes, and protozoans (e.g., Aldrich 1916; Hegner 1926; Goss et al. 1964; Addicott 1974; Petersen et al. 1997; Hamilton et al. 2000). Top predators, such as Odonata or Megaloptera were recorded from *Sarracenia* pitchers by Mather (1981), Rymal & Folkerts (1982), and Corbet (1983). Bradshaw (1980) and Cameron et al. (1977) examined the temperature and oxygen regimes within the pitcher liquid of *S. purpurea*, while Fish & Hall (1978) measured pH in such environments. The unique environment of pitcher plants was studied by e.g., Wray & Brimley (1943), Judd (1959), Paterson (1971), Fish & Hall (1978), Kingsolver (1979), Rymal & Folkerts (1982).

The main goal of this work was to find out whether this non-native ecosystem can be colonized by native insect species in European climatic conditions.

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## Material and methods

The research was carried out in the vicinity of the village Tulčík, eastern Slovakia (49.0882949 N, 21.3136972 E, 286 m a.s.l.) in 2022. There are no permanent aquatic ecosystems near the site, only seasonal waters, so-called anthropotelmata (rainwater tanks, etc.). Three plants of the genus *Sarracenia* (Figure 1) were used for the experiment.



Figure 1. One from the tree experimental plants *Sarracenia purpurea* L.

At the beginning of the experiment (7.5.2022), the plants were completely drained of old water that they contained and filled with rainwater free of organisms after filtering through a plankton net. The plants prepared in this way were then placed outside in the nature and left to self-develop. Subsequently, the plants were checked at monthly intervals during the three-month experiment (7.6., 7.7. and 7.8.). The last date, however, was without water, since owing to the extremely hot summer the phytotelmata naturally dried up. At each inspection, the number of leaves that formed phytotelma was counted, water was removed from each plant and its total volume was measured. Water samples were subsequently fixed with 96% ethanol and transferred to the laboratory for further processing. The amount of pumped water from phytotelmata was always filled up with new filtered water as described above.

In laboratory the water samples were thoroughly examined under a stereomicroscope and the representatives of insects were picked. Subsequently, water samples were examined once again under high magnification for aquatic microorganisms.

Insects were identified according to Rozkošný (1980) and Nilsson (1997), rotifers using keys of Bartoš (1959) and Segers (1995), and for microinvertebrates guide of Sládeček & Sládečková (1997) was used.

## Results

The volume of water changed over time. While in June there was about 350 ml of water contained in the pitchers, one month later less than half of this water was present in the pitcher leaves. In August there was no water present. The basic overview of the development (number and volume) of phytotelmata (Figure 2) in the three examined plants in different sampling dates is given in Table 1.

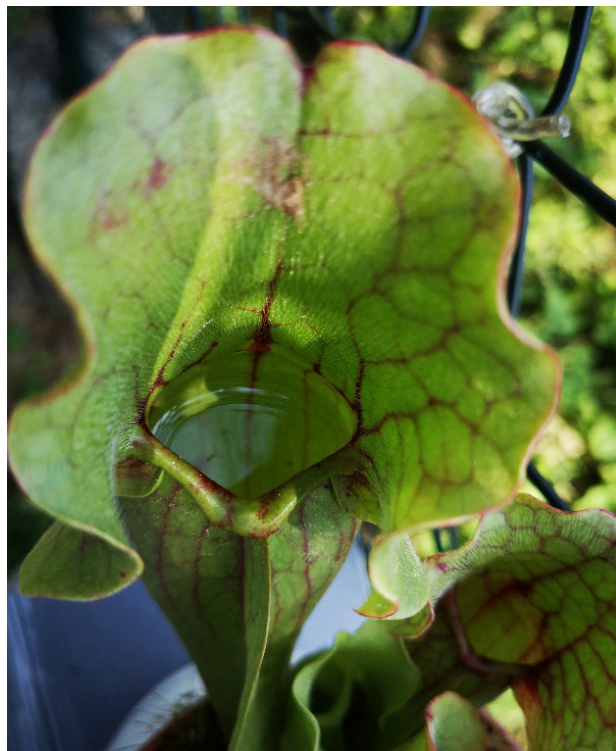


Figure 2. Phytotelmata created in *Sarracenia purpurea* L.

Table 1. Overview of the number of pitcher leaves and contained water volume in different sampling dates.

Sampling date/ experimental plant No.	7.6.2022		7.7.2022	
	No. leaves*	Volume (ml)	No. leaves*	Volume (ml)
1	11	105	13	40
2	9	108	7	50
3	11	143	11	70
Total	31	356	31	160

\* Leaves with water

A total of 15 taxa were recorded in the pitcher phytotelmata during the three-month period (Table 2). While seven taxa were collected at the first sampling date, 14 taxa were found one month later. Six taxa overlapped between the two samplings. We primarily focused on the composition and dynamics of the Rotifera and Diptera fauna. From the rotifer fauna, we recorded only bdelloids (mainly juvenile individuals) with a frequency of ~1.3 ind./leaf in June. One month later, in July, we found a significantly larger amount of deposited organic material and, at the same time, an increase in both the number of individuals and the species diversity of rotifers (Bdelloidea 108.5; *Lecane bulla* (Gosse 1851) 147.7 ind/leaf; *L. inermis* (Bryce 1892) 124.5 ind/leaf; *Colurella obtusa* (Gosse 1886) 2.3 ind/leaf. From dipterans, a representative of the family Psychodidae (*Psychoda* sp.) and Chironomidae (*Paratanytarsus* sp.) were recorded in June, while Ceratopogonidae (*Dasyhelea* sp.), Sciomyzidae and Chironomidae (*Paratanytarsus* sp. and *Chironomus* sp.) in July. Representatives of Tardigrada, Nematoda, Ciliophora (e.g., *Colpidium* sp.) and Flagellata (e.g., *Katodinium* sp.) were also recorded.

## Discussion

Dipteran larvae are the most frequent inhabitants of pitchers. Fish & Hall (1987) confirmed three dipteran families, Chironomidae, Sarcophagidae and Culicidae. In the present study, larvae of four families were recorded. Chironomidae were represented by genera *Chironomus* and *Paratanytarsus*. *Chironomus* is one of the most species-rich genera of the family with many opportunistic species that may be early colonizers after a significant environmental change (Brooks 1997). It was also a common inhabitant in fountains of some European cities (Hamerlík & Brodersen 2010; Oboňa et al. 2017), and frequently one of the first colonizers after their fill-up. Moreover, it was the most abundant and frequent chironomid in water-filled garden barrels in eastern Slovakia (unpublished data), so this could be the colonization source of the plants in our case, since natural water bodies did not occur close to the experimental set-up.

Table 2. List of collected taxa in different sampling dates.

Taxon	7.6.2022	7.7.2022
<b>Diptera</b>		
<b>Ceratopogonidae</b>		
<i>Dasyhelea</i> sp.	-	3 larvae
<b>Chironomidae</b>		
<i>Chironomus</i> sp.	-	1 larva
<i>Paratanytarsus</i> sp.	1 larva	4 larvae
<b>Psychodidae</b>		
<i>Psychoda</i> sp.	2 larvae	-
<b>Sciomyzidae</b>		
Sciomyzidae indet.	-	2 larvae
<b>Rotatoria</b>		
<b>Bdelloidea</b>	1.3 ind/leaf	108.5 ind/leaf
unidentified bdelloids	(*)	(*)
<i>Habrotrocha</i> sp.	(*)	(*)
<b>Lecanidae</b>		
<i>Lecane bulla</i> (Gosse, 1851)	-	147.7 ind/leaf
<i>Lecane inermis</i> (Bryce, 1892)	-	124.5 ind/leaf
<b>Lepadellidae</b>		
<i>Colurella obtusa</i> (Gosse, 1886)	-	2.3 ind/leaf
<b>Ciliophora</b>		
<i>Colpidium</i> sp.	-	*
<b>Flagellata</b>		
<i>Katodinium</i> sp.	-	*
<b>Nematoda</b>	*	*
<b>Tardigrada</b>	*	*

(\*) since most individuals (>90%) were juveniles or morphologically unidentifiable specimens, the numbers could not be relevantly quantified to this level

Occurrence of *Paratanytarsus* in the pitchers is interesting, since the genus has not been known as an exceptionally good colonizer. *Paratanytarsus grimmii* (Schneider 1885) was, however, recorded in some fountains (Hamerlík & Brodersen 2010). This parthenogenetic species, known as a pest through its ability to breed in water distribution system, was hypothesised to originate from the tap water network, however, in nature it may occupy hyporheic habitats. Unfortunately, in our case the species and its source of colonization remain unknown.

When it comes to the food source of the recorded chironomids, it is likely that similar to the larvae of *Metriocnemus knabi* found previously in pitcher plants, they feed on the food remains and detritus on the bottom of the pitcher.

We are aware that in the case of microorganisms, it is challenging to decide whether the plants were

newly colonized, or they are remains of the previous environment. It is likely that draining the old water from the pitchers did not remove the microscopic organisms completely, which may have easily survived in the newly added water. Many of the taxa identified in our study survive unfavourable conditions in various stages and that once suitable conditions are restored, the 'dormant' stages are reactivated. If this is the case, the colonisation of the plants occurred much earlier and not in the new environment, during the experiment. The bdelloid rotifer, *Habrotrocha rosa* Donner 1949 is considered as a common and dominant inhabitant of *Sarracenia purpurea* leaves by many authors (Bateman 1987; Błędzki & Ellison 1998, 2003; Kneitel & Miller 2002; Petersen et al. 1997). *H. rosa* was found also in the pitchers of the allochthonous *S. purpurea* populations in Germany (Gebühr et al. 2006). Petersen et al. (1997) found two other bdelloid species (*Macrotrachela quadricornifera* Milne, 1886, and *Adineta steineri* Bartos, 1951) and two individuals of the genus *Lecane* in *S. purpurea* leaves. Błędzki & Ellison (2003) added three more species to the list of *Sarracenia* dwellers, *Cephalodella anebodica* (Berzins 1976), *Lecane lunaris* (Ehrenberg 1832) and *Notholca acuminata* (Ehrenberg 1832).

Based on the above results, we can conclude that *Sarracenia purpurea* phytotelmata represent an interesting ecosystem, with a potential for a suitable free ecological niche even in a new environment, and which is colonized by similar groups of organisms as in the original ecosystems. However, presence of some microscopic organisms could be the result of a previous colonisation that occurred in the original environment.

## Conclusion

Common inhabitants of the water-filled pitchers of *S. purpurea* were rotifers, dominated by bdelloid rotifers, as well as *Lecane bulla* and *L. inermis*, and larvae of dipterans, especially Chironomidae. Our results indicate that the biodiversity of the pitcher-occupying communities increase with retention time of the water and consequently the accumulation of sediment. Longer and more complex survey will be needed to describe the succession of the aquatic communities in pitcher plants.

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## Color aberration by the Eurasian woodcock (*Scolopax rusticola* L.)

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### Abstract

In Hungarian and international ornithology literature certain colour variations of Eurasian woodcock (*Scolopax rusticola*) and their pattern are described, but the literature on the topic is rather incomplete and often contradictory. This article deals only with colour changes caused by genetic disorders i.e., with mutations that affect pigmentation. These include melanism, i.e., brown mutation, dilution, gray discoloration, leucism and Ino mutation. The descriptions of the colour variants help to identify the mutations by briefly explaining physiological processes in the background, pointing out uncertainties and the misinterpretation of the different mutations. In addition to a literature review of the mutations we illustrate the most significant colour varieties with photos, and we present our own research results on colour aberration. The large-scale analysis of the variability of colors and patterns was supported by the Woodcock Monitoring Project managed by the Hungarian Hunters Association since 2010. Based on countrywide Woodcock wing collection, as part of the biometric module of monitoring, we examined 12 078 wing samples from 2010 to 2018. Only six specimens (0.05%) were found in the entire data set showing deficient pigmentation of some degree or pastel colour.

**Keywords:** *Scolopax rusticola*, Woodcock curiosities, color aberration, colour variability, pigmentation disorders

### Introduction

The plumage colour variability in birds is not rare, in polymorphic species forms with deviating colours are generally regarded as varieties (van Grouw 2013). In the case of the Woodcock (*Scolopax rusticola* Linnaeus 1758 - Scolopacidae), however unique coloured specimens occur only very rarely. The aim of the study is to represent a uniform system of colour deviations occurring by Woodcock and to resolve the related contradictions in the professional literature. Representing the basic characteristics of the most common mutations promotes the use of correct terminology and makes easier to identify them. It is to be noted that there are also known colour deviations which are difficult to distinguish from one another (van Grouw 2006, 2013). Since pigmentation disorders, as well as any disorder of varying quality and quantity in melanin production, can lead to similar coloration, a visual examination is not consistently sufficient to precisely describe the lesions (van Grouw 2006, 2013). The classification of curiosities in taxidermic collections is also made more difficult by the fact that the feather loses its fine, detailed pattern over time, as pigments are also destroyed by light, however, there is often the only possibility of examining unique specimens in such collections or photos of their materials.

### Pigmentation process

To correctly identify these colour mutations, is important to know which pigments define the species-specific normal colour of feathers and the physiognomy of pigmentation process disturbances. The multistage chemical process of the melanin formation as well as the pigmentation process itself became known in the second half of the 20<sup>th</sup> century (Mason 1953; Lubnow 1963). The typical colour and pattern of Woodcock are determined

by two types of melanin - eumelanin and pheomelanin. Melanocytes are formed by melanoblasts that develop in the embryonic spinal cord at an early embryonic stage and then spread during transport processes to the skin and feather follicles where they may produce melanin. Both forms of colour-determining melanins are not necessarily present in the plumage of all species (Lubnow 1963). In the species of the Corvidae family only eumelanin is found in the plumage, while both melanins are present in the feathers of the Woodcock. In this case feathers can be characterized by certain patterns and/or colour differences caused by different amount, oxidation state and specific distribution of eumelanin and pheomelanin. During the development of the species-specific pattern and colouration, eumelanin is mainly concentrated in the centre while pheomelanin on the edges of the feathers. The central compound in the chemical process of melanogenesis is the amino acid tyrosine, pigment formation occurs through its oxidation and is regulated by the tyrosinase enzyme (van Grouw 2006). Melanin polymer molecules are oxidized during the process. However, the degree of oxidation can vary, and thus the intensity of the colour produced. Black is the most oxidized form, while brown indicates a weaker oxidation state. Depending on the concentration and distribution of the different melanin types in the feather, eumelanin is responsible for the black, gray and dark brown shades. Pheomelanin produces a red-brown hue at high concentrations and a yellowish-brown colour at lower concentrations. The common presence of the two melanins leads to a combination of the colours determined by them, resulting in a normal coloration and pattern typical for the species (van Grouw 2006). It should be noted hereby that any abnormality during

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the melanoblast spread, or melanin synthesis may affect the final pigmentation of the plumage. Of course, the deviations do not always have a genetic background, unusual colour changes can also develop due to external factors. In the ornithological literature, the nomenclature of lesions indicating colour varieties are mainly based on the mutations of rock doves (*Columba livia* Gmelin 1789) (van Grouw 2013).

### Mutations leading to colour varieties

Woodcock's most extensive knowledge of colour varieties was published in the journal of the Club National des Bécassiers, La Mordorée, in France (see e.g., Boidot 2002a–2015; Chantron 2019; Jarry 2018 etc.). Although the uniquely coloured Woodcocks are presented, they are not comprehensively systematized and the genetic and physiological reasons for the lesions are not presented. In our study we performed the systematization of colour deviations due to genetic mutations in this species. According to the literature, the following colour deviations occur in Woodcock: melanism, melanin thinning (pastel, Isabella), brown mutation Ino and leucism. In addition, some special colours (agate, opal) have also been described.

### Melanism

Melanism is the most common polymorphism in birds (Mundy 2006), but in Woodcock, the lesions associated with an increase in the amount of melanin are rather infrequent pigment disorders. No birds are known in which the concentrations of both melanins were increased at the same time (van Grouw 2013). If the amount of pheomelanin remains at a normal level with increased eumelanin concentrations, the colour induced by pheomelanin is hardly or even not recognisable, which means that the bird will turn completely black as the brown / gray patterns are barely noticeable. This mutation appears in two forms:

(1) *Eumelanism* – a lesion associated with an increase in the amount of eumelanin that results in a black-tinted appearance of the bird's plumage. Eumelanism is the most common melanistic lesion (Van Grouw 2006).

(2) *Pheomelanism* – an increase in the amount of pheomelanin, which shifts the colour of the bird's plumage more towards red-brown than the normal wild-dominant colour.

In French literature, Boidot (2009a) distinguishes the following two main categories in case of Woodcock melanism:

(1) *Jet-black mutation* – affects the entire plumage, the legs are dark, steel gray, the nails black. In this case, the brown colour is completely absent, so the wild dominant pattern does not appear due to the lack of transverse markings on the feathers. Jet-black plumage has not been observed in the Woodcock until now (Boidot 2009b, 2012).



Figure 1. A pheomelanistic Woodcock described as a transitional form of the gray colour variety. (Boidot 2009a) (Photo: J-P. Boidot).

(2) *Transient black mutation* – the black mutation is to a greater or lesser extent present in the plumage, the brown colour is still visible in some places, so that the classic wild-dominant pattern is locally recognizable (especially on the lower body and on the wings with the exception of the primaries and secondaries). But these transitive forms appear also very rarely in Woodcock (Boidot 2012). According to Boidot's (2009b) hypothesis, the melanistic mutation can occur in two ways, depending on the initial melanisation sequence in the feather: type I: brown/black and type II. black/brown.

*In the first type* the brown markings in the patterning are replaced by black melanin during the development of the feather. Oxidized black eumelanin is the only pigment synthesized in the early stages of feather growth. Later, however, some brown, red pigments are occasionally incorporated, resulting in a light brown or very fine beige pattern on the tip or the edge of the feathers. *In the second type*, the process begins with a brown/black sequence. The patterns of the feathers are getting blacker as the oxidation process continues, and the markings in the patterning are thickening during the melanisation. Even the lightest areas will be filled with dark pigment and the feather will be deeply black tinted as it grows. The brown patterns change to gray while the gray patterns turn to black. In the first type, it is remarkable that the melanisation affects feathers from the upper body, with very few feathers from the lower body. In the second case Boidot (2009b), believes that the entire plumage

is concerned by the melanisation. During the first type the melanisation process is slowed down, disrupted, or blocked by genes responsible for pigment polarity. In connection with the phenomenon declares Boidot (2012), that Woodcock specimens even with a patterned melanistic character are extremely rare.

Boidot (2009b) suggests the following classification based on the low number of known melanistic birds:

Black / melanistic plumages

Brown / black pattern-dependent melanisation

Black plumage pattern

Black / brown melanisation independent of the pattern

1. Transient black, upper body intensely, lower body not significantly melanized
2. Limited / partial black, intensive melanisation in the upper or lower body



Figure 2. Special black coloured Woodcock from France. (Boidot 2009b) (Photo: M. Baril).

### Brown mutation

The brown colour change is caused by a qualitative decrease in eumelanin. In case of this mutation the amount of eumelanin remains unchanged by decreasing oxidation degree. The hereditary, incomplete oxidation of eumelanin causes black feathers to turn dark brown, while the colour induced by unchanged quality and quantity of the pheomelanin dominates (Kopf 1986). In the Woodcock, caused by the presence of both melanins, this mutation is less pronounced than in species whose feathers only contain eumelanin. This phenomenon gives the birds different colour intensity in patches, caused by melanin amounts present in various oxidation states. Overijssel (1996) refers to this change as „biscuitcoloured” which deviation also occurs in Woodcock (Url. 1; Url. 2; Url. 3; Url. 4; Boidot 2012; Vorontsov 2014). There is a mentionable important fact that eumelanin in different oxidation degrees is responsible for the different shades

of brown, so eumelanin is not only by deviations present in feathers in different qualities and concentrations. The decrease in quality of eumelanin in feathers causes sensitivity to sunlight, which effects the pigments a rapid and strong fading. In case of such a very pale, almost white “brown mutant” bird, it is worth looking at the lower layer under the contour feathers as well as the plumage on the inner, covered parts. The brown mutation is widespread among bird species and is also one of the most common lesions in the Woodcock, but extremely light, almost white “brown mutants” are very rare. Inheritance of brown mutation is linked to sex chromosomes. In birds, males have homogametic (ZZ) females heterogametic (WZ) sex chromosomes, which explains why most brown mutant birds female are, since in their case the brown mutation appears even by one altered gene sequence (Van Grouw 2006, 2013).

### Changes associated with melanin dilution

The melanin dilution is defined by Kopf (1986) as a quantitative decrease, which means, that the quality, i.e. the oxidation state of the pigment does not change during the mutation, but the concentration can decrease significantly. As a result, a “thinned”, lighter shade can be observed compared to the original colour. Two types of this lesion can be distinguished:

(1) *Isabella plumage* – defined by French experts as a separate colour category, but aware of the biological causes that lead to the development of the lesion, Van Grouw (2006) classifies it among melanin dilution mutations. In contrast to pastel mutations, the Isabella mutation results in a quantitative decrease of eumelanin, only one of the two melanins, the black/dark brown feathers turn gray, while the concentration of pheomelanin causing the red/brown colour does not change. According to Boidot (2012), two mutations occur in this case, the number of brown pigments decreases, but the black colour is missing because of the incomplete oxidation of eumelanin, which results in a lighter so-called Isabella plumage.

(2) *Pastel mutation* – caused by a quantitative decrease in both eumelanin and pheomelanin. Black feathers turn gray, while red-brown feathers yellowish-brown. The extent of pigment concentration decrease in Woodcock is varying widely, which affects plumages faded to different degrees. According to Boidot (2008a, b, c), the changing melanin concentration is also responsible for pattern formation in different shades. The pastel mutation actually leads to a normally patterned, but to different degrees faded bird. The plumage of birds with heavy melanin dilution are often almost white as the low pigment concentration in the feathers fades further by sunlight.



Figure 3. Woodcock with Isabella plumage from Italy. (Url.5). (Photo: P. Pennacchini).

According to the French literature, melanin dilution occurs simultaneously with further mutations causing colour lesions and induce together a colour change. An example of this is the *black pastel* (Boidot 2013a; Pennacchini 2013; Maltot 2017) colour variety. In these birds, a significant part of the body plumage is white due to a strong melanin dilution. Patterns of normal coloured plumage are often completely absent, but melanin responsible for the black colour is present in high concentrations, in a highly oxidized state in some regions of the plumage. The peculiarity of this colour variety is confirmed by the low number of known black pastel birds in the Palearctic (Bende & László 2019a, 2020a,b, 2022).



Figure 4. Black pastel Woodcock from Hungary. (Photo: Zs. Marton).

The first known mutated brown pastel Woodcock was exhibited in the collection of the Rothschild Zoological Museum accompanied by a pure white specimen (Anonym 2018). In these birds, due to the very low oxidation level of eumelanin the black colour does not appear at all, stronger or weaker brown shades dominate depending on the concentration. The pattern of these birds corresponds to that of classic wild-dominant birds but is paler due to the lower pigment concentration.

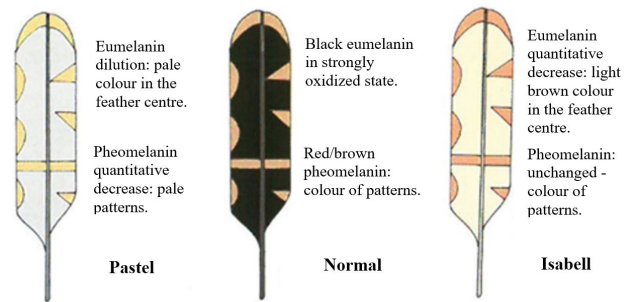


Figure 5. Classic wild-dominant, pastel and Isabella mutation of Woodcock. (after Boidot 2008c).

### Gray mutation

The gray colour variety results from the strong pheomelanin decrease, leading to a suppression of brown and red shades and the more intense appearance of gray and black tones (van Grouw 2006). Among the special pastel birds, the French experts also described the medium gray and gray pastel types (Boidot 2012). According to Boidot's (2012) assumption and to Grouw's (2006) assertions, there is a connection between the origin of the black and gray colour varieties, because both lesions are caused by the inhibition of pheomelanin, whereby gray shades are emphasized. Only two such specimens are known from the literature (Chantron 2019; Url. 5.). It is important to point up that schizochromism in which one of the melanins is completely missing is not identical to gray mutation.



Figure 6. Gray Woodcock. (Chantron 2019) (Photo: P. Chantron).

### Albinism and leucism

Some information about the colour and pattern variability of the Woodcock is known from the second half of the 1800s. These usually report on birds with a lack of pigmentation with very pale patterns on the feathers. Other specimens are known with basic white plumage with very few dark patterns or patchy unpigmented, so-called “varicoloured” Woodcocks. However, these designations are ambiguous in many cases contradictory.

According to the definition of Fox & Vevers (1960) albinism means the deficiency of both pigments in plumage, skin and in the iris caused by the inherited lack of the enzyme tyrosinase. In this case, the pigments are not only missing in the plumage (white colour), but also in legs, claws and eyes. Real albino specimens are very rare among wild birds because they have no depth perception with eyes without pigments. There are not any known reports of true albino Woodcock. In the published, white-feathered specimens, the eyes, the skin and the naked horny structures have always been pigmented. Accordingly, with the correct terminology, these partially unpigmented specimens are not albinos, but leucistic mutants which means white specimens with some pigmentation.

In the case of leucism, the enzyme tyrosinase is present so that melanin is produced in the body of these birds. The pigment deficiency occurs in the feathers because the pigment-producing melanoblasts cannot reach certain parts of the body (in extreme cases the entire surface of the skin) due to a congenital and hereditary defect. Pigment cells are therefore absent from the skin or part of it, where they would normally supply pigment material to the growing feather. The size of the body surface covered with white feathers shows a varied picture, from a few white feathers (partly leucistic) to full plumage; in the latter the skin appendages are also colourless. Leucistic birds always have dark eyes and pigmented bills, legs and claws (van Grouw 2006). White groups of feathers caused by leucism usually have bilateral symmetry, so some white primaries appear on either side (van Grouw 2013). At first, describing leucism, it was assumed to be caused by malnutrition (Rollin 1964), but later it came to prove of a mutation of genetic origin behind the phenomenon.



Figure 7. Rare leucistic Woodcock from France. (Boidot 2008a) (Photo: M. Tauxe).

### Ino mutation

The Ino mutation arises from a strong qualitative reduction in eumelanin and pheomelanin. In contrast to melanin dilution, in this case both melanins are present in normal amounts, but in an incompletely oxidized state, so that brown eumelanin and pale pheomelanin lead to this colour. The Ino mutation is based on a single gene linked to the sex chromosome in each species (Van Grouw 2013). The mutation can lead to a variety of phenotypes depending on melanin oxidation grade. The oxidation degree of melanin varies, so that black eumelanin can be dark to very light brown, whereas reddish-brown pheomelanin is always very pale or even barely visible. The plumage has a faint pattern, particularly at higher levels of eumelanin, which is typically noticeable in species where the feathers usually contain white parts, like the feathers of Woodcock. According to Boidot (2014), Ino is characterized by a special melanisation in which eumelanin, often in a strongly oxidized state, accumulates in the plumage of certain body regions, especially in the wings, at the tip of the feathers. The mutant Ino bird has reddish eyes, although the iris pigments are not absent, but melanin is in a slightly oxidized state. Due to the available pigment materials, these birds have much better sight than an Ino. It is to claim that the red-eyed white birds registered under natural conditions are certainly INOs and not albinos. This recessively inherited mutation occurs only in females (Van Grouw 2013).

English specialist literature (Van Grouw 2013) distinguishes between two categories:

(1) *Ino-Light* – significant quality decrease in of both melanins due to insufficient oxidation grade. The original black colour turns very light brownish cream, while the original red yellowish-brown colour is barely visible. The eyes, beak, and legs are pink.

(2) *Ino-Dark* – strong quality reduction of both melanins due to insufficient oxidation state. The original black colour becomes light brown while the red yellowish-brown colour changes to brown cream. The beak and legs are also creamy brown coloured (Van Grouw 2013).

French literature (Boidot 2014) also distinguishes two main categories:

(1) *Standard Ino plumage* – the pattern of the upper parts of the body is characterized by a complete lack of brown colour. The concentration of pigment granules occurs in more distant parts of the body. The body is typically white with a fuzzy pattern. The beak and legs are yellow, the tips of the wing feathers show a disproportionate colour change without wild dominant patterns.

(2) *Transitional Ino plumage*

*Type 1* – blurred pigmentation is characteristic in the upper body. The wings are evenly dark gray coloured due

to a pigment lack. The legs and beak are yellow.

*Type 2* – only in the upper part of the bird's body and only to a negligible extent, but the discoloration with a blurred contour in the white colour is present. Discoloration is mainly seen on the end or side of the wings, leg, and beak of the bird.

### Further mutations in the Plumage of Woodcock

#### *Opal*

The opal mutation prohibits the appearance of the brown colour, since it is coupled with the inconsistently positioned quantitative decrease in eumelanin. It differs from pastel in that the pigment concentration in the centre decreases more intensely than at the edges of the feather (BOIDOT 2008c). Boidot considers the simultaneous appearance of the opal and pastel mutations is possible, which leads to an even more intense decrease in the pigment concentration. As a result of the opal mutation, the melanins responsible for the brown colour decrease quantitatively, which leads to a bluish effect. The pattern usually appears in light brown and with a blurred contour, which is most noticeable on the wing plumage. This recessive mutation is described by Boidot (2006) for Woodcock using unusual wing patterns from two different countries (Russia, France).

#### *Agate plumage*

The mutation, which leads to a quantitative reduction in melanin giving the brown colour, does not change the quality of the melanin. A characteristic of the agate mutation is the simultaneous presence of eumelanin and pheomelanin, like black and "diluted" brown pigments (Boidot 2002a, 2008a). Boidot (2003) reported that the agate and opal mutations can even occur simultaneously.

### Some literature discrepancies regarding colour variations

#### *Melanism*

Finding that the markings are reduced through progressive melanisation or during pattern independent incorporation of dyes is fraught with uncertainty if it is relied only on phenotypic traits. In this way, the system of melanistic categories (partially black, transient black, limited black, black in pattern) constructed by Boidot (2009b) is also uncertain.

With regard to the hypotheses on the melanisation process described by Boidot (2009b), it should be pointed out that in type I it is not known whether the colour modification is caused by a change in the oxidation state and / or an increase of melanin concentration. According to Van Grouw (2006), the two melanins (eumelanin and pheomelanin) cannot be substituted, so that the appearance of the colour determined by them is ruled by their oxidation level and relative concentration. In type II, the exact genetic background of the concentration and/or oxidation state of the weakly oxidized eumelanin and pheomelanin resulting the brown colour is not known, which means that the reduction processes behind the species-specific wild-dominant patterns are also

not clear identified. It can even be difficult to determine, especially in specimens with partially pattern-deficient melanistic plumage, whether the lesion is indeed caused by a mutation. In case of many bird species, it has been proven that non-mutational lesions may also underlie this phenomenon e.g.: illness, malnutrition. Unfortunately, the problem of partial melanism in Woodcock is still unclear, although Boidot (2009b, 2011) considers this as a common phenomenon especially in the coverts of flight feathers. Between 450 and 500 samples of this wing type have already been collected by French experts. However, there are only a few literature references to larger lesions affecting other areas of the body's plumage.

#### *Lesions caused by melanin dilution*

French ornithologists investigating colour variations of Woodcock use pastel shades to differentiate further each of the classic colour categories such as black, agate, brown (classic brown and reddish brown) and Isabella pastel (Boidot 2008a). In French literature, Isabella-lesion is classified as one of the main categories (Boidot 2003b, 2007; Jarry 2018) and Isabella pastel often described as its colour variant based on French and Italian Woodcock specimens (Boidot 2008e, 2009a). In the case of Isabella, this description of the colour variant is questionable, as Isabella is formed by quantitative eumelanin reduction, while pastel by the combined quantitative reduction of eumelanin and pheomelanin, therefore, the amount of either or both melanins varies, so the mutation can be either Isabella or pastel.



Figure 8. Woodcock specimen described as Isabella pastel. (Boidot 2009a) (Photo: M. J. Bernard).

Boidot (2008b) points out the uncertainty of the distinction between Isabella and pastel colour variants when publishing a photo by Fernan Blandin (England) and one by Alain Chalopin (France) of pastel Woodcock specimens. While describing the birds, Boidot (2008c) reports that the categories Isabella and pastel are often confused. In our opinion, the combined interpretation of these two phenomena is by definition contradicting, and the phenotypic features seen in photographs of preparations of these birds are not sufficient to clarify this problem.

French experts describe the so-called ‘agate’ mutation as a separate colour category. In this case, the mutation reduces the concentration of brown melanins, resulting in a lighter plumage. In our opinion, the agate lesion cannot be distinguished from the range of melanin dilution lesions based only on phenotypic appearances. It is also questionable whether this mutation can appear in a pastel version, however, in the case of a Woodcock shot in Ireland, Boidot (2008a) assumes a double melanin dilution and reports it as agate pastel. The agate-opal lesion was described by Boidot (2006) based on a single special sample of Woodcock.



Figure 9. Agate-opal coloured Woodcock with light gray, bluish plumage. (Boidot 2002a) (Photo: M. M. Boulanger).

Quantitative as well as qualitative disorders in melanin production often result in very similar pale colour changes, so the visual description of these mutations is rather uncertain, which Boidot (2002a) also draws attention to. The assessment of this complex mutation is made even

more difficult by the fact that it is the only known specimen in which the simultaneous occurrence of “partial agate plumage” and “opal mutation” has been described.

The literature is also often contradictory in the description of the black-pastel colour variant, since several specimens discussed among Ino mutants are clearly corresponding to the black-pastel category due to their appearance (Boidot 2003a, 2014). The classification of the unusually coloured Woodcock, recommended by Alain Le Coniac to Boidot, shot in the hunting season 2003/04 in Russia is also ambiguous, as Guy Hellequin describes the plumage as pastel mutation, while Boidot considers the opal mutation to be likely. Therefore, the opinion of the experts on the classification of individual specimens is also not clear even for these two lesions.

#### *Leucism*

Woodcock with deficiency of colour of different degrees are so-called leucistic birds (Anonym 2018), but the lack of pigment in the feather can be caused not only by mutation, but also by other factors. Worthy of mentioning is the phenomenon of gradual graying, in which as a bird ages, the amount of pigment cells gradually decreases, finally the entire plumage becomes whitish. In the case of Woodcock, this phenomenon has not yet been described, but its possibility cannot be ruled out. In the case of leucism, or in extreme cases of gradual graying, the abnormal feathers are completely white because of melanin deficiency. This issue is further complicated by the fact that external, non-hereditary factors such as disease or malnutrition can also cause discoloration because of disturbed melanin synthesis, in which case pigmentation normalizes as soon as external causes disappear.

Another difficulty with describing leucism is that the extremely pale, almost white plumage may be the result of other mutations, of which the brown mutation is the most frequent (Buckley 1982; Van Grouw 2013). It is important to note that leucistic feathers are completely pigmentless, as there are also specimens whose feathers are almost completely white, but on closer inspection the normal wild-dominant pattern in light brown or silver colour is partially or fully visible.

#### *Ino mutation*

Regarding the Ino mutation two publications with contradictory findings are known from France (Boidot 2003a, 2014). Boidot (2003a) refers to a strongly white Woodcock described as a pastel, however, he states that this specimen can be classified as Ino because of its plumage. Boidot (2014) also points to a specimen previously described as Isabella mutant, whereby Ino mutation would be the correct classification according to current knowledge. In the case of the Woodcock, we do not yet have any authentic literature on the Ino phenomenon, so we consider the French classification problematic in this regard. In published descriptions, the authors usually rely on photographs of preparations, which further impedes the

description of the lesion, since its defining characteristics cannot be objectively assessed using collection materials. Lesions examined only visually often cannot be determined with absolute certainty, not even in case of a living bird or of a fresh cadaver.

Depending on the mutation scale the degree of melanin oxidation can vary significantly, so a variety of colour changes can result in a range of theoretical phenotypes, leading to a range of specimens that can be confused with many other modifications genetic origin as well (e. g. melanin dilutions, brown mutation).

In species without phaeomelanin, Ino can be distinguished from the brown mutation by foot and beak, as they are always pink due to the lack of melanins (Van Grouw 2013), however, in the case of Woodcock, both melanins are found. The correct description of this mutation is further complicated by the fact that the plumage continues to fade over time after moulting. According to Van Grouw (2013) the mutation is recessive, its inheritance is linked to the sex chromosome, so that the change occurs phenotypically only in females, i.e., without sex dimorphism in the case of living birds and without knowledge of the sex in case of preparations, the claim about the mutation cannot be trustworthy.

### Summary

Using literature data on curiosities and the results of the Hungarian Woodcock Bag Monitoring we tried to give a comprehensive picture of the colour variability in the case of Woodcock, referring to the possible reasons for the rare occurrence, as well as to the possibilities and difficulties of classification. To arrange uniquely coloured specimens in a system, described color variant categories were collected as classification options based on the nomenclature of the mutations in the English and French specialist literature. In addition, for each category, we referred to possible inconsistencies in the literature on the coloration of Woodcock.

Four categories of colour changes caused by confirmed mutations occur in Woodcock, such as melanism, brown mutation, melanin dilution (Pastel and Isabella) and leucism. The phenomenon of Ino and albinism has not been confirmed yet in this species, but their occurrence cannot be excluded. In connection to the classification, it can be stated that the correct naming and description of the colour variants occurring in Woodcock is burdened with considerable uncertainty based only on visual inspection. This is especially true for the distinguishability of melanin dilution mutations (pastel and Isabella) as well as for the distinctness of colour variants resulting from multiple mutations.

The establishment of certain categories or the description of groups within larger categories – e.g. groups of melanism described by Boidot (2009b) or the Ino categorization also reported by Boidot (2014) – is considered justified only if the given mutation has been credibly described for the species or the abnormal feathered specimen required for

exact categorization is known. It would be also important to clarify which mutations may appear together, and how they modify the colour of the specimen. It should also be elucidated which relationship exists between the individual categories (e.g., a leucistic specimen being black-pastel at the same time) since the possibility of belonging to more than one category cannot necessarily be ruled out, especially if several mutations act simultaneously. It is important to emphasize that only laboratory tests of genetic background can give credible results if accurate description of a colour variant is required.

During the investigation term 2010–2018, we observed mutations resulting in a rare colour variation (leucism and melanin dilution) in only 0.05% of the Woodcock wing samples of the 12 078 specimens examined (Bende & László 2019a, b).

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## The usability of beer traps for detecting *Xenos vesparum* parasitizing *Polistes dominula*

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### Abstract

The Strepsiptera species *Xenos vesparum* Rossi, 1793, is an endoparasite of wasps (genus *Polistes*) characterized by a specific lifecycle and significant sexual dimorphism. A total of 12 host specimens (*Polistes dominula* (Christ, 1791)) were caught using a beer trap – site Diviacka Nová Ves, period 25 July 2022 – 5 August 2022. The prevalence of *X. vesparum* were 83%. Overall, 33 parasites were present in 10 wasps (min. 1, max. 10, average 3.3). The highest number of parasites (13 specimens) was observed below terga T3 and T4. It appears that the parasite *X. vesparum* most commonly infects *P. dominula* on the dorsal side of the body under terga T3 and T4. In addition, beer traps seem to be a suitable mean of detecting and calculating the prevalence of this parasite.

**Keywords:** Strepsiptera, prevalence, European paper wasp, endoparasite, trapping, Slovakia

### Introduction

Strepsiptera are among the smallest groups (male body size 1–7 mm, female body size 2–30 mm) of holometabolous insects, with approximately 600 species described so far (e.g., Pohl & Beutel 2013). The species *Xenos vesparum* Rossi, 1793 (Insecta: Strepsiptera: Xenidae), is a noteworthy organism representing an endoparasite of the European paper wasp – *Polistes dominula* (Christ, 1791) (Insecta: Hymenoptera). The species displays a peculiar lifecycle and demonstrates extensive sexual dimorphism (e.g., Beani et al. 2005; Cappa et al. 2014; Richter et al. 2017). Both sexes develop in their host. Males pupate as adults, subsequently leave the host and actively seek females. In contrast, females are obligatory and permanent endoparasites (e.g., Beani & Massolo 2007; Beani et al. 2018). Beer traps represent a passive method commonly used to capture a wide range of insects (e.g., Manko et al. 2018; Ruchin et al. 2020a; Touroult & Witté 2020). The method has been proven to be suitable for trapping rare insects as well as for monitoring selected invasive species and pests (e.g., Dvořák et al. 2019, 2020, 2022; Dvořáková et al. 2020; Hribar 2020; Albacete et al. 2020; Lioy et al. 2020; Mariychuk et al. 2020; Ruchin et al. 2020ab; Maceda-Veiga et al. 2021; Manko et al. 2021). To date, this method is not known to have been used for the detection of *X. vesparum* in the population of *P. dominula*.

The present study aims to analyse the suitability of the beer trap method for capturing and evaluating the prevalence of *X. vesparum* endoparasites parasitizing *P. dominula*.

### Material and methods

The study was carried out between 25 July and 5 August 2022 in the village of Diviacka Nová Ves (48.749672, 18.492035). A single beer trap (for more details, see Manko et al. (2018)) was hung on *Parthenocissus* (up to a height of approx. 2 meters above the ground). After exposure, the caught specimens were removed from the trap, washed under clean water, and fixed with 96% ethanol. In the laboratory, they were determined after Richter

et al. (2017). The *P. dominula* specimens were separated and subsequently examined under a Motic SMZ-168 microscope. In each of the sampled wasps, the abdomen (this parasite does not infect other parts of the body) was thoroughly examined and the position/presence of the endoparasites was recorded.

### Results and Discussion

A total of 12 specimens of *P. dominula* were caught using the beer trap, 10 of which, were positive for *X. vesparum*. In addition, 25 specimens of the German Wasp (*Vespula germanica* (Fabricius, 1793)) were caught in the beer trap, but these were completely free of parasites.



Figure 1. Positive sample of *P. dominula* infected by *X. vesparum*.

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The prevalence of *X. vesparum* was 83%. A total of 33 parasites were present in 10 wasps (min. 1, max. 10, average 3.3). The highest number of parasites (13 specimens) was

observed below terga T3 and T4. For more details, see Table 1 and Figure 2.

Table 1. Overview of the presence of *X. vesparum* under the terga of *P. dominula*.

Specimen n.	Tergite 1	Tergite 2	Tergite 3	Tergite 4	Tergite 5	Tergite 6	total
1		1	3	2	2		8
2				2			2
3			2	1			3
4				1			1
5							0
6			6	2	2		10
7					1		1
8				1			1
9				2			2
10							0
11			2	1	1		4
12				1			1
Total	0	1	13	13	6	0	

Below tergite T3, 7 parasites occurred dorsally and 6 laterally, while under tergite T4, 11 parasites were present dorsally and only 2 laterally. Below tergite T5, 6 parasites were observed dorsally, and under tergite T2 a single parasite was found dorsally. The frequency of occurrence of the studied parasite in relation to individual tergites is depicted in Figure 2. It appears that the parasite *X. vesparum* infects *P. dominula* most commonly on the dorsal side under terga T3 and T4.

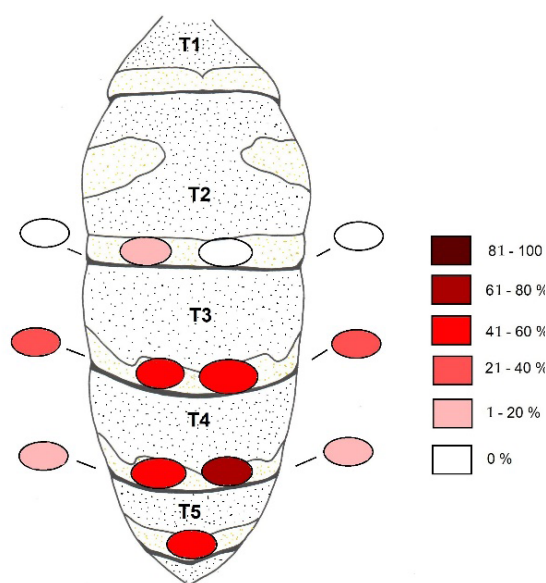


Figure 2. Scheme of infestation of *P. dominula* by *X. vesparum*. The colour scale indicates the position and frequency of occurrence in individual tergites (from 0 % to 100 %).

*Note.* Tergite 5 is relatively narrow, therefore only one point is used in the scheme to illustrate the occurrence of the parasite. In two cases there was one parasite, in two other cases two parasites closely packed together.

Findings similar to ours were reported by Reilly & McCarthy (1993), who investigated the attachment site selection in *Eylais* (Acari: Hydrachnellae) water mites, the larvae of which infect Corixidae (Hemiptera: Heteroptera). The species *Eylais* tended to attach to the abdominal terga of the hosts. While *E. infundibulifera* Koenike, 1897, infected the two anterior terga, *E. discreta* Koenike, 1897, infected terga three and four. Ramsey et al. (2019) investigated the location of *Varroa destructor* Anderson & Trueman, 2000, in honeybees. This work revealed that the parasite is not consuming haemolymph, as was previously accepted, but damages the host bees by consuming the fat body, a tissue roughly analogous to the mammalian liver.

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## Správa o opätovnom náleze *Zingel streber* (Siebold, 1863) v rieke Váh

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### Abstract. Note on the rediscovery of *Zingel streber* (Siebold, 1863) in the Váh River

The authors report a note on the first find of a Danube streber (*Zingel streber*) from the Váh River near the town of Vrútky (northern Slovakia). Specimen with a body length of 147 mm was captured on the edge of a rapid section of the river at a depth of approximately 40 cm. This report is only the second documented find directly from the Váh River in the region of northern Slovakia and at the same time from its very isolated section due to the presence of several migration barriers in this area. In the end, the distribution of this species in the western part of the Danube river basin in Slovakia is discussed, where the occurrence of this species is considered to be very rare.

**Key words:** Percidae, Danube streber, Váh River, Northern Slovakia

### Úvod

Viacere toky podhorskej zóny severozápadného Slovenska patria z ichtyologického hľadiska v porovnaní so zvyškom našej krajiny medzi menej preskúmané. Tento fakt v súčasnosti platí najmä pre povodie stredného a horného úseku Váhu (medzi Trenčínom až Ružomberkom), kde boli podrobnejšie ichtyologické prieskumy realizované zväčša len na významnejších prítokoch (Vlára, Rajčianka, Kysuca, Varínka, Turiec, Orava, prípadne na samotnom hornom úseku Váhu), aj to však prevažne len v období pred niekoľkými desaťročiami. Práve z tohto regiónu však pochádzajú viaceré mimoriadne zaujímavé nálezy chránených a ohrozených zástupcov našej ichtyofauny, akými sú hrúz Kesslerov (*Romanogobio kesslerii*), hrúz fúzatý (*Romanogobio uranoscopus*) alebo kolok vretenovitý (*Zingel streber*), ktorých výskytové záznamy sa v prevažnej miere datujú práve do obdobia minulého storočia (Balon 1956; Mišík 1959; Kirka 1967; Holčík et al. 1965; Bastl et al. 1975; Černý 1980).

Práve 1 exemplár kolka vretenovitého bol zaznamenaný dňa 20. mája 2022 pracovníkmi MsO SRZ Martin v starom koryte Váhu v katastri obce Turčianske Kľačany (49°06'44.7"N 18°56'35.2"E), pri odlove generačného materiálu mreny severnej (*Barbus barbus*) pre účely jej následnej umelej reprodukcie. Vzhľadom na vzácnosť tohto druhu v hornej časti povodia Váhu ako aj jeho citlivosť voči antropogénnym vplyvom, považujeme za dôležité informovať o tomto náleze detailnejšie v kontexte doterajších poznatkov o jeho rozšírení.

### Materiál a metódy

Samotný Váh v oblasti Turčianskej kotliny môžeme charakterizovať ako typickú podhorskú rieku (hyporitrál), spadajúcu v rámci ichtyologickej klasifikácie do mrenového pásma. Staré koryto Váhu si v tejto oblasti zachováva pomerne prirodzený charakter so striedaním plytších torentilných a hlbších fluviatilných úsekov. Priamo na mieste lokality nálezu dominuje výrazne perejný úsek

s hĺbkou do 40 cm, ktorý pozvoľna prechádza do pomalšie prúdového úseku a následne ostro padá do tichej hĺbočiny pri ľavom brehu (Obrázok 1).



Obrázok 1. Lokalita nálezu kolka vretenovitého na starom koryte rieky Váh v blízkosti obce Turčianske Kľačany (Foto: Richard Štencel).

Pri love bol použitý batériový elektrický agregát (typ SAMUS 1000, 12 V, 50 A, 2-99 Hz). Ďalší odlov zameraný na získanie početnejšieho materiálu predmetného druhu bol uskutočnený 25. mája 2022 s použitím dvoch elektrických agregátov rovnakého typu. Tento pokus nebol úspešný, avšak v jeho priebehu bolo sledované celkové druhové spektrum ichtyofauny sledovaného úseku a taktiež boli pomocou multifunkčného prístroja HACH SL1000 zaznamenané nasledovné fyzikálno-chemické ukazovatele vodného prostredia: teplota vody = 13.7 °C; obsah rozpusteného kyslíka (O<sub>2</sub>) = 10.32 mg.l<sup>-1</sup>; pH = 6.83; obsah železa (Fe) = 0.01 mg.l<sup>-1</sup>; amoniak (NH<sub>3</sub>) = 0.01 mg.l<sup>-1</sup>; uhličitany (CaCO<sub>3</sub>) = 99 mg.l<sup>-1</sup>. Nakolko kolok vretenovitý predstavuje v zmysle platnej legislatívy (Vyhláška č. 170/2021

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Z. z., ktorou sa vykonáva zákon č. 543/2002 Z. z. o ochrane prírody a krajiny v znení neskorších predpisov) chránený druh so stanovenou spoločenskou hodnotou, ktorý je navyše aj druhom európskeho významu, bol tento jedinec po odmeraní dĺžky tela (SL) a následnej fotodokumentácii (Obrázok 2) vypustený na miesto pôvodného výskytu.



Obrázok 2. Ulovený exemplár kolka vretenovitého (Foto: Rudolf Martiniak).

### Výsledky a diskusia

Ulovený exemplár mal dĺžku tela 147 mm čo na základe poznatkov o veku a raste tohto druhu v blízkom Turci (Mužík 1997) zodpovedá IV. vekovej kategórii. V priebehu opakovaného odlovu bol na danej lokalite zaznamenaný aj výskyt ďalších 12 druhov rýb. Spolu s výsledkami predošlých ichtyologických prieskumov tak bolo v dotknutej oblasti Váhu zdokumentovaných celkovo 22 druhov (Tabuľka 1).

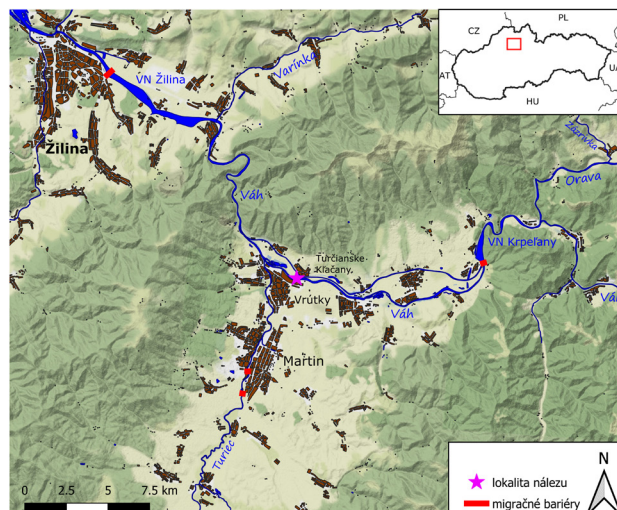
Kolok vretenovitý sa na území Slovenska vyskytuje len v dunajskom systéme (povodie Dunaja a Tisy). Kým v minulosti bol tento druh rozšírený vo väčšej časti podhorskej zóny tokov (Hensel 1979), dnes je jeho areál na Slovensku výrazne obmedzený. Pod túto skutočnosť sa podpísali najmä fragmentácia tokov, znečisťovanie vôd (na ktoré je tento druh obzvlášť citlivý) ako aj ťažba materiálov zo štrkových lavíc, ktoré predstavujú habitat pre jeho reprodukciu ako aj samotnú existenciu. V povodí Tisy vo východnej časti Slovenska je areál kolka situovaný prevažne do podhorských úsekov tunajších riek (najmä Topľa a Ondava) (Košťath et al. 2006), ale vyskytuje sa aj v spodných úsekoch Rimavy a Slanej v západnej časti povodia (Hajdú 2014). Podľa autorov však aj v tejto oblasti došlo v minulom storočí vďaka viacerým vodohospodárskym úpravám tokov a nadmernému znečisťovaniu k výraznému úbytku lokalít s jeho prirodzeným výskytom. V západnej časti dunajského povodia bol kolok vretenovitý v minulosti zistený zväčša len v dolných, resp. stredných úsekoch prítokov Dunaja, akými sú rieky Ipeľ, Hron, Váh alebo Morava, prípadne aj v samotných prietochných ramenách Dunaja pred vybudovaním VD Gabčíkovo (Kux & Weisz 1962, 1964). V súčasnosti absentujú doklady o výskyte tohto druhu z oblasti dolného Váhu a Moravy, hoci podľa Horvátha et al. (2012) je najmä v rieke Morava veľký predpoklad jeho výskytu vzhľadom na viaceré záznamy z vyššie položených úsekov rieky na území Českej republiky. V čiastkovom povodí Váhu bol kolok v minulosti zaznamenaný v jeho dolnej časti pri Piešťanoch (Mišík 1958) a následne až v prítokoch podhorskej zóny – v Turci (Bastl et al. 1975), Orave (Balon 1956), resp. Čiernej Orave pred vybudovaním VN Orava (Holčík et al. 1965). Hensel (1979) uvádza, že koncom 19. storočia bol zistený aj v rieke

Tabuľka 1. Zoznam zaznamenaných druhov rýb na predmetnom úseku rieky Váh.

Druh	Kux & Weisz (1964)	Mužík (2005)	Križek & Štencel (2022)	naše údaje
<i>Alburnus alburnus</i>	+	+	+	+
<i>Alburnoides bipunctatus</i>		+	+	+
<i>Anguilla anguilla</i>		+	+	
<i>Barbus barbus</i>		+	+	+
<i>Barbatula barbatula</i>	+	+	+	+
<i>Blicca bjoerkna</i>			+	
<i>Cobitis elongatoides</i>				+
<i>Cottus gobio</i>	+		+	+
<i>Cottus poecilopus</i>	+			
<i>Esox lucius</i>		+		
<i>Gobio obtusirostris</i>	+	+	+	+
<i>Chondrostoma nasus</i>	+	+	+	+
<i>Leuciscus leuciscus</i>	+	+	+	
<i>Lota lota</i>	+	+		
<i>Perca fluviatilis</i>	+	+		
<i>Phoxinus phoxinus</i>	+		+	+
<i>Proterorhinus semilunaris</i>			+	+
<i>Rutilus rutilus</i>	+	+	+	
<i>Salmo trutta m. fario</i>		+	+	
<i>Squalius cephalus</i>	+	+	+	+
<i>Thymallus thymallus</i>	+	+	+	
<i>Vimba vimba</i>		+	+	+
<i>Zingel streber</i>				+

Kysuca (podľa autora bol jedinec deponovaný v Maďarskom národnom múzeu v Budapešti). Populácia kolka v rieke Turiec je rovnako otázná. Na lokalitách kde sa v minulosti pravidelne vyskytoval (Bastl et al. 1975; Mužík 1996; Bastl & Holčík 1997; Stráňai & Andreji 2002), dnes absentuje (vlastné nepublikované pozorovania autorov). Zontág (2000) koncom minulého storočia zaznamenal jeho hojný výskyt vo viacerých prítokoch Turca priamo v intraviláne mesta Martin (vzdialenosť lokality nálezu od ústia Turca do Váhu je približne 2.2 km), odkiaľ bol v počte niekoľko desiatok kusov introdukovaný aj do pôvodného areálu na sútoku Váhu a Oravy, resp. aj do oblasti pod sútokom Váhu a Lubochnianky (okres Ružomberok). Úspešnosť tejto introdukcie však zostala otázná. Zatiaľ posledným publikovaným údajom o výskyte kolka priamo vo Váhu zostáva záznam Mužíka (2012) z jesene roku 2010, ktorý ho ulovil pod sútokom s riekou Oravou. Rovnako vzácny bol tento druh i v iných tokoch podhorskej zóny západnej časti dunajského systému na Slovensku. V regióne stredného Pohronia sa podľa Dudicha (1956) vyskytoval na úseku medzi Zvolenom a Banskou Bystricou. Aktuálne práce (Škovranová et al. 2012; Varga et al. 2019) už prítomnosť kolka na strednom úseku Hrona nepotvrdili. Z dolného úseku pod Želiezovcami jeho výskyt uvádza Škovranová et al. (2012). Podobne izolovaná populácia existovala aj v hornej časti rieky Nitra v okolí Prievidze (Kux & Weisz 1964; Sedlár 1969). Podľa Stráňai (1995) však koncom 70. rokov 20. storočia z tejto oblasti postupne vymizol. Na základe týchto poznatkov preto môžeme tieto lokálne populácie pokladať za pravdepodobne vyhynuté. Azda najväčší areál v rámci tokov dunajského povodia vykazuje kolok vretenovitý aktuálne v rieke Ipeľ, kde je pravidelne zaznamenávaný od dolného úseku nad ústím do Dunaja až takmer po Lučenec (Weiperth et al. 2020). Aj v tomto prípade však autori konštatujú, že tento druh je pre povodie Ipeľa veľmi vzácny. Náš nález v danej situácii predstavuje mimoriadnu zaujímavosť, najmä z pohľadu izolovanosti lokality medzi viacerými neprekonateľnými migračnými bariérami (Obrázok 3). Na rieke Váh je to vodné dielo Krpeľany spolu s nižšie položeným VD Žilina, na Turci malá vodná elektrárň (MVE) Martin, resp. ďalšia vyššie položená nepriechodná hať s mlynským náhonom v Martine.

Tento fakt tak môže svedčiť o určitej adaptabilite tohto druhu na narušený hydrologický režim v rieke Váh, ktorý spôsobuje výrazné kolísanie vodnej hladiny zapríčinené práve manipuláciou viacerých vodných diel. Na druhej strane, práve fragmentácia toku a narušenie hydrologického režimu mohli byť príčinou výrazného úbytku početnosti druhu v kontraste s obdobím pred výstavbou vodných diel. Pre posúdenie veľkosti tunajšej populácie a jej vekového zloženia bude potrebné realizovať podrobnejší ichtyologický prieskum rieky Váh v danej oblasti. Významným dôvodom pre detailnejší monitoring je aj fakt, že kolok vretenovitý predstavuje prioritný druh európskeho významu, pre ktorého ochranu je možné vyhlásiť samostatné chránené územie (územie európskeho



Obrázok 3. Mapa záujmového územia s vyznačenou lokalitou zaznamenaného jedinca (Autor: Peter Križek).

významu). Na základe informácií Štátnej ochrany prírody je aktuálne na Slovensku vyhlásených 24 takýchto území, medzi ktoré okrem iných patria aj rieky Orava, Turiec alebo horný úsek Váhu v okolí Ružomberka. Na základe súčasného nálezu môžeme taktiež predpokladať, že izolované populácie kolka vretenovitého v oblasti regiónu stredného Považia v obmedzenej miere stále pretrvávajú, avšak čelia viacerým typom ohrozenia. Okrem už vyššie spomenutého narušeného vodného režimu vplyvom manipulácie vodných diel a viacerých nepriechodných migračných bariér, predstavuje riziko aj možnosť náhleho mimoriadneho zhoršenia kvality vody a čoraz výraznejší dopad klimatickej zmeny na vodné toky, prejavujúci sa najmä v letných mesiacoch rapidným znížením prietokov. Náš súčasný záznam o výskyte kolka v dotknutom úseku Váhu taktiež vytvára predpoklad výskytu ďalších ohrozených reofilných druhov rýb v tejto oblasti (*Romanogobio uranoscopus*, resp. *R. kesslerii*), ktoré tu neboli zdokumentované už niekoľko desaťročí. V tomto smere by mohla pomôcť aj výraznejšia aplikácia progresívnych molekulárnych metód (environmentálna DNA), ktorá umožňuje detekciu druhov aj bez ich priameho fyzického ulovenia (Ficetola et al. 2008).

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