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# Tujerodni invazivni hrošč semenar *Megabruchidius dorsalis* (Chrysomelidae) živi v Sloveniji

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**Izvleček.** Azijski hrošč semenar *Megabruchidius dorsalis*, ki se razvija v semenih azijskih vrst gledičevk (*Gleditsia*), je bil v Evropi prvič zabeležen v Italiji leta 1989, v Sloveniji pa prvič v Mariboru leta 2017. Pojavlja se še v 15 drugih evropskih državah. V novem okolju je vrsta preskočila na dva nova gostitelja iz Severne Amerike, trnato gledičevko (*Gleditsia triacanthos*) in rogovilarja (*Gymnoladus dioicus*), sicer oba sorodna azijskim vrstam gledičevke. Med našo raziskavo smo nabrali zrele stroke trnate gledičevke in rogovilarja na različnih lokacijah po osrednji in severovzhodni Sloveniji z namenom raziskati razširjenost *M. dorsalis* zunaj prvih opisanih lokacij v Mariboru. Stroke smo gojili v gojitvenih vrečah pod stalnimi razmerami v laboratoriju. Vrsto *M. dorsalis* smo potrdili v šestih krajih po Sloveniji. Na 36 lokacijah (97 %) smo našli izhodne luknje na strokih in semenih, značilne za *M. dorsalis*, žive ali mrtve hrošče pa smo našli na 26 lokacijah (70 %).

Ključne besede: Coleoptera, Bruchinae, trnata gledičevka, *Gleditsia triacanthos*, rogovilar, *Gymnocladus dioicus*, tujerodna vrsta

**Abstract. The alien invasive seed beetle *Megabruchidius dorsalis* (Chrysomelidae) established in Slovenia –** The Asian seed beetle *Megabruchidius dorsalis*, which develops inside the seeds of native *Gleditsia* species, was first recorded in Europe in Italy in 1989. In Slovenia, it was first recorded in Maribor in 2017. Its presence was confirmed in 15 other European countries. In the new range, the seed beetle switched to two new hosts from North America, the honey locust (*Gleditsia triacanthos*) and the Kentucky coffeetree (*Gymnocladus dioicus*) both related to the Asian *Gleditsia* species. Mature *G. triacanthos* and *Gy. dioicus* seed pods were collected from the ground at locations in the central and the northeastern parts of Slovenia to investigate the presence of *M. dorsalis* outside the first described locations in Maribor. Seed pods were kept in bags under constant conditions in laboratory. Exit holes of seed pods and seeds typical of *M. dorsalis* were found at 36 locations (97%), while alive or dead *M. dorsalis* beetles were recorded at 26 locations (70%).

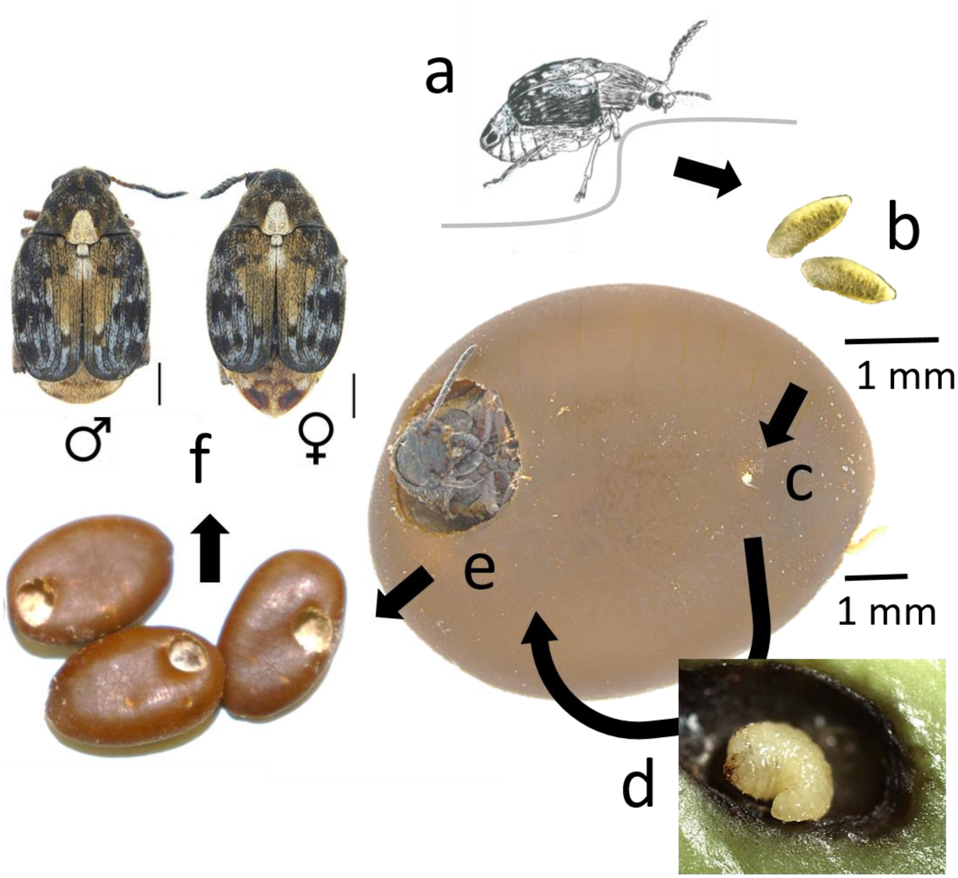
Key words: Coleoptera, Bruchinae, the honey locust, *Gleditsia triacanthos*, the Kentucky coffeetree, *Gymnocladus dioicus*, alien species

# Uvod

Razširjanje tujerodnih vrst pomeni eno izmed največjih bioloških groženj za biodiverziteto po vsem svetu (Early et al. 2016). Naraščanje številčnosti in hitro širjenje tujerodnih vrst nas spodbuja, da spremljamo in beležimo njihovo pojavljanje tudi lokalno. S tem prispevamo k boljšemu razumevanju vpliva, ki ga imajo v novem okolju. *Megabruchidius dorsalis* (Fåhræus, 1839) je hrošč, predstavnik semenarjev (Chrysomelidae, Bruchinae), ki izvira iz vzhodne Azije, kjer se ličinka razvija v semenih avtohtonih vrst azijskih gledičevk (*Gleditsia* L.) (Tuda & Morimoto 2004). V Evropi je bil hrošč prvič zabeležen v Italiji leta 1989 (Migliaccio & Zampetti 1989). Od takrat se po Evropi uspešno širi in so ga potrdili še v 16 drugih evropskih državah (v Albaniji, Avstriji, Bosni in Hercegovini, Črni gori, Franciji, na Hrvaškem, Madžarskem, v Nemčiji, na Poljskem, v Romuniji, Rusiji, na Slovaškem, v Sloveniji, Španiji, Švici in Ukrajini), pri čemer je bilo več kot 80 % teh najdb potrjenih po letu 2010 (Šipek et al. 2022). Seznam tujerodnih hroščev Slovenije iz leta 2012 sicer navaja pet vrst iz skupine Burchinae, med katerimi pa še ni vrste *M. dorsalis* (Vrezec et al. 2012). V Sloveniji je bil prvič zabeležen leta 2017 v Mariboru (Šajna 2019).

V novem območju razširjenosti je vrsta preskočila na dva nova gostitelja, in sicer na severnoameriški drevesni vrsti, trnato gledičevko (*Gleditsia triacanthos* L.) (Migliaccio & Zampetti 1989) in v redkih primerih rogovilarja (*Gymnocladus dioicus* (L.) K. Koch) (Callot & Wagner 2016), ki ju v Evropi sadijo predvsem v okrasne namene. Trnata gledičevka je sorodna azijskim vrstam gledičevke, obe vrsti z rogovilarjem pa pripadata poddružini metuljnic Caesalpinioideae (DC, 1825), ki vključuje okoli 160 rodov večinoma tropskih in subtropskih dreves (Bruneau et al. 2001).

Ugotovljeno je bilo, da ima *M. dorsalis* več generacij na leto (Kurota & Shimada 2001, 2002), različne razvojne stopnje se lahko pojavljajo tudi sočasno (Šajna 2019). Začetek razvojnega kroga se ujema s produkcijo in zorenjem plodov in semen gostitelja (Kurota & Shimada 2002). Samice avgusta odložijo jajčeca prve generacije na sveže stroke s semeni. Ličinke zvrtajo luknjo skozi strok in v seme, kjer se prehranjujejo z založnim tkivom, razvijajo se in rastejo, dokler se ne zabubijo. Pred stanjem bube imajo štiri razvojne faze (Kurota & Shimada 2001). Odrasli hrošči prve generacije prilezejo iz semen med koncem septembra in začetkom novembra in lahko v tem stadiju prezimijo ali se razvijejo ličinke druge generacije, ki potem prezimijo v eni od štirih razvojnih stopenj (Kurota & Shimada 2001). Odrasli osebki druge generacije se izležejo med aprilom in majem ter približno sočasno z odraslimi osebki prve generacije, ki so prezimili kot odrasli, odložijo jajčeca na lanske stroke, ki so čez zimo odpadli z dreves in vsebujejo zelo suha in trda semena (Kurota & Shimada 2001). Odrasli hrošči tretje generacije prilezejo iz semen junija in julija in nekateri lahko odložijo jajčeca četrte generacije na preostala semena. Samice tretje in četrte generacije nato odložijo jajčeca prve generacije nove sezone na nove, sveže stroke na drevesih (Kurota & Shimada 2001). Odrasli hrošči se lahko razmnožujejo brez prehranjevanja (Takakura 1999). Razvojni krog *M. dorsalis* (Sl. 1) traja približno 50 dni pri temperaturi 24 °C in dolžini osvetlitve 16:8 (dan:noč), kar je najhitreje med hrošči semenarji (Kurota & Shimada 2001, 2002).




**Slika 1.** Razvojni krog hrošča semenarja *Megabruchidius dorsalis.* Samica (a) odloži jajčeca (b) na strok. Ličinka, ki se izleže, potuje do semena, vanj zvrta luknjo (c), se prehranjuje z založnim tkivom semena in se razvija (d). Zadnji stadij ličinke se zabubi in preobrazi v odraslega hrošča, ki dokončno pregrize semensko lupino (e) in zapusti seme (f). Fotografije in risba: N. Šajna.

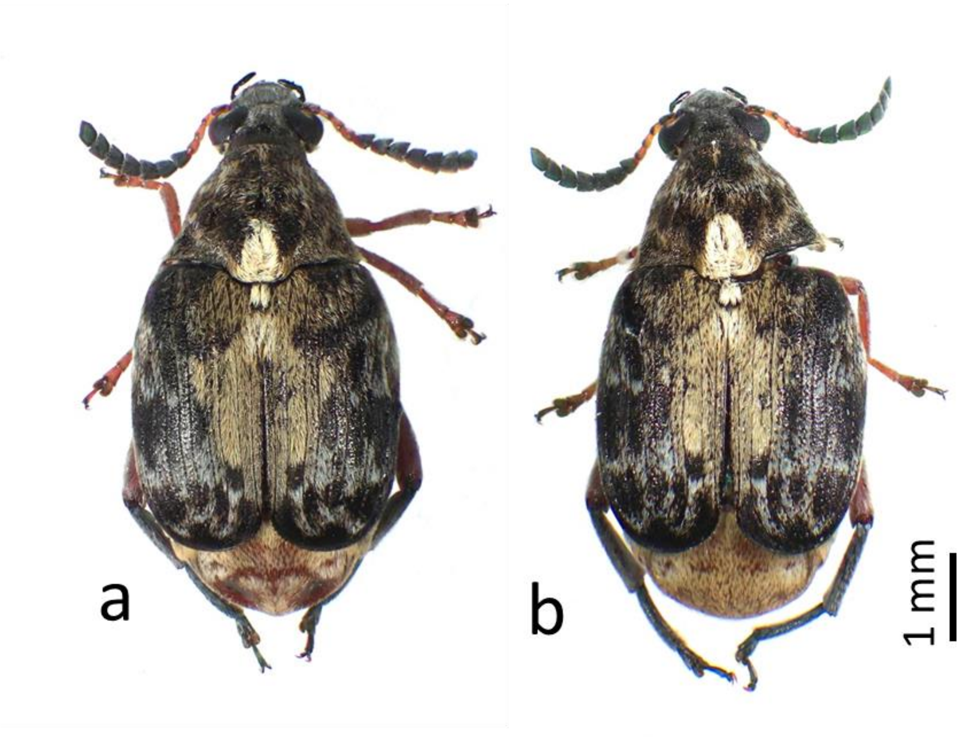
**Figure 1.** Life cycle of the seed beetle *Megabruchidius dorsalis.* Female (a) lays the eggs (b) on seed pods. Larva then travels to the seed and drills a hole into the seed to enter it (c). Larva feeds on the cotyledons and develops (d). Final larval instar pupates and transforms into an adult beetle which chews a circular exit hole (e) and leaves the seed (f). Photos and drawing: N. Šajna.

Namen študije je bilo raziskati, ali ima *M. dorsalis* v Sloveniji stabilne populacije in ali se vrsta pojavlja poleg prve opisane lokacije tudi drugod po Sloveniji, hkrati pa preveriti, katere gostitelje uporablja za svoj razvoj.

# Materiali in metode

Zrele stroke trnate gledičevke smo nabrali s tal pod drevesi na 35 lokacijah po Sloveniji (5 lokacij v Ljubljani, 26 v Mariboru, 1 v Pesnici pri Mariboru, 1 v Petanjcih, 1 v Radencih in 1 v Arboretumu Volčji Potok; Tab. 1). Suhe stroke rogovilarja smo nabrali na 2 lokacijah v Sloveniji (1 lokacija v Petanjcih in 1 v Arboretumu Volčji Potok). Stroke smo nabrali oktobra in novembra leta 2019, maja in julija 2020 ter avgusta 2021. Nabrani material smo bodisi pregledali v laboratoriju takoj in izluščili semena ter popisali hrošče, ki smo jih pri tem našli, bodisi smo material prenesli v gojitvene vreče in nato spremljali pojavljanje živih hroščev do dne, ko smo vreče odprli in pregledali stroke še za morebitne mrtve hrošče (za datum nabiranja in pregleda glej Tab. 1). Gojitvene razmere v laboratoriju so bile 22 °C, dolžina osvetlitve 8:16 (dan:noč).

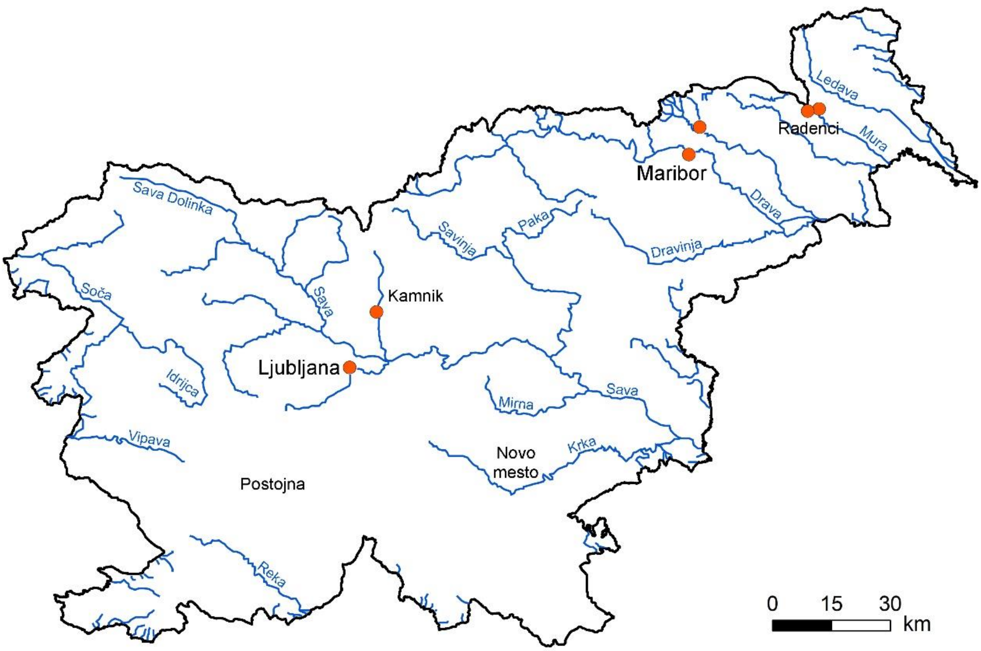
Hrošče smo določili pod stereo zoom mikroskopom SMZ-161-TLED (Motic®) glede na objavljene določevalne znake (Rheinheimer 2014, Pintilioaie et al. 2018) in zabeležili število samcev in samic (Sl. 2; Tuda & Morimoto 2004). Vsem najdenim osebkom razen dvema mrtvima, ki sta bila poškodovana, smo določili spol. Fotografije so bile posnete s kamero Axiocam 208 Color (Zeiss).



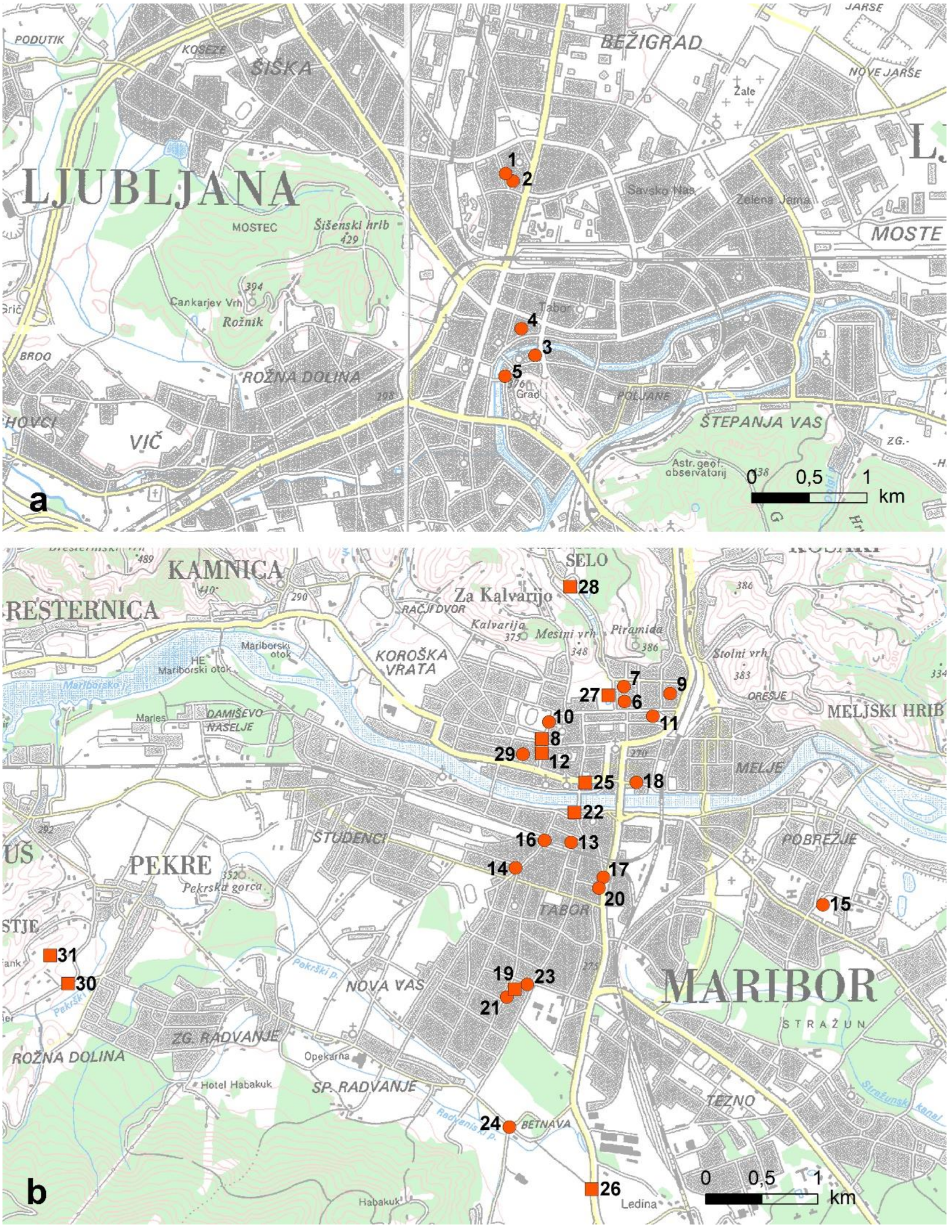

**Slika 2.** Odrasla samica (a) in odrasel samec (b) *Megabruchidius dorsalis* z dorzalne strani. Fotografiji: N. Šajna.
**Figure 2.** Adult female (a) and adult male *Megabruchidius dorsalis* (b) from dorsal side. Photo: N. Šajna.

# Rezultati

Žive hrošče *M. dorsalis* smo potrdili v šestih krajih, ki vključujejo dve največji mesti (Ljubljana, Maribor) in manjše kraje v Sloveniji (Sl. 3). Natančneje smo na vseh 35 lokacijah z drevesi trnate gledičevke in na eni od dveh lokacij z drevesi rogovilarja našli stroke in semena omenjenih dreves z znaki pojavljanja *M. dorsalis* – značilne izhodne luknje na semenih in luknje na strokih. Vrsto smo potrdili na 25 lokacijah (71 %) v semenih trnate gledičevke in na 1 lokaciji v semenu rogovilarja (Tab. 1). V Ljubljani smo hrošča potrdili na vseh obiskanih lokacijah (Sl. 4a), v Mariboru pa na 16 lokacijah (62 %) (Sl. 4b). Na vseh drugih pregledanih lokacijah v Mariboru (10 lokacij; 38 %) smo našli stroke z izhodnimi luknjami, značilnimi za *M. dorsalis* (Sl. 2). Nobenih znakov o pojavljanju vrste nismo našli le na strokih oziroma semenih rogovilarja v Arboretumu Volčji Potok. Žive hrošče smo našli na drugi lokaciji v Arboretumu v strokih trnate gledičevke. V Petanjcih smo leta 2019 našli mrtvega odraslega hrošča v semenu rogovilarja, čeprav je bilo na odpadlih strokih rogovilarja veliko izhodnih lukenj, značilnih za *M. dorsalis*. Na isti lokaciji smo leta 2021 potrdili še dva odrasla samca v stroku trnate gledičevke. V Radencih smo leta 2020 našli žive hrošče, leta 2021 pa le značilne izhodne luknje na strokih. Na vseh lokacijah, razen na semenih rogovilarja v Arboretumu, smo našli tudi nekaj semen z dvema izhodnima luknjama in na lokaciji 6 eno seme trnate gledičevke s tremi izhodnimi luknjami.




**Slika 3.** Razširjenost hrošča semenarja *Megabruchidius dorsalis* v Sloveniji.
**Figure 3.** Distribution of the seed beetle *Megabruchidius dorsalis* in Slovenia.




**Slika 4.** Lokacije s potrjenim pojavljanjem hrošča semenarja *Megabruchidius dorsalis* (krog) ali le z značilnimi izhodnimi luknjami na strokih *Gleditsia triacanthos* (kvadrat): a) v Ljubljani, b) v Mariboru. Vir podlage: Geodetska uprava Republike Slovenije.

**Figure 4.** Locations in Ljubljana with confirmed presence of the seed beetle *Megabruchidius dorsalis* (circle) or typical exit holes on seed pods of *Gleditsia triacanthos* (square): a) in Ljubljana, b) in Maribor. Map source: The Surveying and Mapping Authority of the Republic of Slovenia.

**Tabela 1.** Podatki o najdenih hroščih *Megabruchidius dorsalis* in značilnih znakih na strokih in semenih na lokacijah po Sloveniji. Lokaciji z *Gymnocladus dioicus* sta označeni z zvezdico, LJ = Ljubljana, MB = Maribor, + označuje, da je bil hrošč potrjen v letu 2018 (Šajna 2019); ? – nedoločeno; * – opažene le izhodne luknje na stroku.

**Table 1.** Data on the found adult *Megabruchidius dorsalis* beetles and characteristic signs on seeds and seed pods at locations across Slovenia. Asterisk indicates locations with *Gymnocladus dioicus*, LJ = Ljubljana, MB = Maribor, + denotes beetles recorded in the year 2018 (Šajna 2019); ? – undetermined; * – only exit holes on seed pod observed.

| Št. lokacije | Kraj, ulica, habitat | Koordinate (°N, °E) | Datum nabiranja / Datum pregleda | Št. strokov | Št. hroščev |
|---|---|---|---|---|---|
| 1 | Drevored ob parkirišču, Einspielerjeva ul., LJ | 46.065478, 14.506590 | 24.10.2019 08.06.2020 | 12 | 1♀ |
| 2 | Drevored ob cesti in pločniku, Hranilniška ul., LJ | 46.065065, 14.507103 | 24.10.2019 08.06.2020 | 15 | 1♂ |
| 3 | Nasad, osrednja tržnica, LJ | 46.051071, 14.509462 | 24.10.2019 12.06.2020 | 15 | 12♂, 33♀ |
| 4 | Drevo na parkirišču, Trubarjeva c., LJ | 46.052384, 14.507902 | 25.08.2021 07.10.2021 | 16 | 2♂, 2♀ |
| 5 | Tlakovan drevored, Cankarjevo nabrežje, LJ | 46.049081, 14.505697 | 25.08.2021 07.10.2021 | 8 | 2♂, 3♀ |
| 6 + | Drevored ob ulici z živo mejo, Prešernova ul. I, MB | 46.563764, 15.651622 | 22.10.2019 08.06.2020 | 35 | 43♂, 43♀ |
| 7 + | Drevored ob ulici z zelenico, Prešernova ul. II, MB | 46.565040, 15.651574 | 22.10.2019 25.10.2019 | 5 | 3♂, 2♀ |
| 8 + | Manjši park, Strossmayerjeva ul. / Gosposvetska c., MB | 46.560736, 15.641827 | 06.11.2019 08.06.2020 | 4 | 0* |
| 9 + | Drevored ob igrišču, Dominkuševa ul., drevored ob igrišču, MB | 46.564291, 15.656894 | 22.10.2019 12.06.2020 | 51 | 45♂, 62♀ |
| 10 + | Drevored ob cesti in ograjeni zelenici, Strossmayerjeva ul., MB | 46.562133, 15.642809 | 22.10.2019 12.06.2020 | 43 | 102♂, 105♀ |
| 11 + | Drevored ob cesti in parku, Trg Borisa Kidriča, MB | 46.562527, 15.654963 | 22.10.2019 23.10.2019 | 21 | 2♀ |
| 12 + | Drevored ob cesti in pločniku, Strossmayerjeva ul., MB | 46.559560, 15.641952 | 22.10.2019 23.10.2019 | 3 | 0* |
| 13 | Nasad skupine dreves, Magdalenski park, MB | 46.552501, 15.645272 | 06.11.2019 08.06.2020 | 5 | 1♂ |
| 14 | Parkirišče med bloki, Goriška ul., MB | 46.540497, 15.638442 | 06.11.2019 08.06.2020 | 9 | 0* |
| 15 | Drevored med cesto in zelenico, Ul. Veljka Vlahoviča, MB | 46.547285, 15.674567 | 23.10.2019 08.06.2020 | 11 | 10♂, 6♀, 1? |
| 16 | Drevesa na zelenici, Ul. Moše Pijada, MB | 46.552661, 15.642126 | 06.11.2019 08.06.2020 | 4 | 1♀ |
| 17 | Drevored ob zelenici, Titova c., MB | 46.549583, 15.648960 | 06.11.2019 08.06.2020 | 9 | 1♀ |
| 18 | Drevo pred blokom, Ul. Kneza Koclja, MB | 46.557250, 15.652965 | 06.11.2019 08.06.2020 | 11 | 1♀ |
| 19 | Drevored ob cesti, Gorkega ul., MB | 46.550776, 15.639266 | 06.11.2019 08.06.2020 | 9 | 1♂ |

| Št. lokacije | Kraj, ulica, habitat | Koordinate (°N, °E) | Datum nabiranja / Datum pregleda | Št. strokov | Št. hroščev |
|---|---|---|---|---|---|
| 20 | Skupina dreves pred bloki, Titova c., MB | 46.548789, 15.648475 | 06.11.2019 14.11.2019 | 5 | 1♂ |
| 21 | Zelenica med bloki, Goriška ul., MB | 46.540136, 15.637672 | 06.11.2019 14.11.2019 | 5 | 1♂ |
| 22 | Nasad dreves na zelenici in pešpoti, Taborska ul., MB | 46.554849, 15.645719 | 06.11.2019 13.11.2019 | 15 | 0* |
| 23 | Zelenica pred blokom, Ljubljanska ul. / c. Proletarskih brigad, MB | 46.541026, 15.639939 | 06.11.2019 14.11.2019 | 4 | 3♂, 2♀ |
| 24 | Drevored ob cesti in potoku, Streliška c. / Razvanjska c., MB | 46.529293, 15.637645 | 06.11.2019 08.06.2020 | 10 | 13♂, 11♀ |
| 25 | Nov nasad mladih dreves ob parkirišču, Glavni trg, MB | 46.557277, 15.647003 | 27.05.2020 27.05.2020 | 1 | 0* |
| 26 | Drevesa ob cesti, spontano pojavljanje ob ograji, Tržaška c. / Papeževa c., MB | 46.524454, 15.647432 | 27.05.2020 27.05.2020 | 4 | 0* |
| 27 | Zelenica ob Akvariju, Mestni park, MB | 46.564339, 15.649823 | 06.11.2019 13.11.2019 | 2 | 0* |
| 28 | Drevo na zelenici, Mestni park, za Tremi ribniki, MB | 46.573085, 15.645468 | 26.05.2020 26.05.2020 | 2 | 0* |
| 29 | Drevored ob cesti, Smetanova ul., MB | 46.559538, 15.639713 | 06.11.2019 13.11.2019 | 6 | 2♂, 1♀ |
| 30 | Drevo ob cesti in spontano pojavljanje v mejici, Hrastje, MB | 46.541552, 15.586798 | 09.11.2019 09.11.2019 | 8 | 0* |
| 31 | Sajena drevesa ob hiši, Hrastje, MB | 46.543653, 15.584493 | 09.11.2019 09.11.2019 | 2 | 0* |
| 32 | Drevored ob cesti, Pesnica pri MB | 46.609782, 15.679781 | 24.10.2019 08.06.2020 | 20 | 34♂, 39♀ |
| 33* | Parkovni nasad, Vrt spominov in tovarištva, Petanjci* | 46.648896, 16.076874 | 24.10.2019 08.06.2020 | 11 | 1? |
| | | | 28.07.2020 12.10.2020 | 5 | 0* |
| 34 | Parkovni nasad, Vrt spominov in tovarištva, Petanjci | 46.648896, 16.076874 | 28.07.2020 - ni strokov | 0 | 0 |
| | | | 24.08.2021 07.10.2021 | 7 | 2♂ |
| 35 | Parkovni nasad skupine dreves, Mestni park, Radenci | 46.643342, 16.042843 | 28.07.2020 12.10.2020 | 10 | 14♂, 25♀ |
| | | | 24.08.2021 07.10.2021 | 10 | 0* |
| 36 | Park, Arboretum Volčji Potok | 46.186142, 14.609954 | 24.10.2019 08.06.2020 | 10 | 45♂, 33♀ |
| 37* | Park, Arboretum Volčji Potok* | 46.187791, 14.612125 | 24.10.2019 08.06.2020 | 11 | 0 |

# Razprava

Tujerodna vrsta azijskega hrošča semenarja *M. dorsalis* je v Evropi preskočila na nov, ozkosorodni vir hrane, in sicer na gojeno severnoameriško vrsto okrasnega drevesa trnata gledičevka. Posledično je *M. dorsalis* v Evropi naturalizirana vrsta, se uspešno spontano širi in pridobiva značilnosti tujerodne invazivne vrste (Horvat & Šajna 2021a, Šipek et al. 2022). Iz vseh lokacij pojavljanja v Evropi poročajo, da so njegove populacije stabilne. Naši rezultati prav tako kažejo, da ima invazivna vrsta *M. dorsalis* stabilne populacije tudi v Sloveniji. Na vseh lokacijah so bile vidne sledi prisotnosti vrste, kot so izhodne luknje odraslih hroščev na strokih in semenih trnate gledičevke. Na 68 % pregledanih lokacijah so bili potrjeni tudi živi ali mrtvi odrasli hrošči. Na dveh lokacijah v Mariboru, kjer smo našli le značilne izhodne luknje na strokih trnate gledičevke (lokaciji št. 8 in 12), so bili živi hrošči najdeni leta 2018 (Šajna 2019). Nadalje se kaže, da se v Evropi *M. dorsalis* občasno razvije tudi v semenih rogovilarja (Callot et al. 2016, György & Tuda 2020). To je bilo opaženo tudi v naši raziskavi, kar bi lahko invazivni potencial vrste še okrepilo. Bistvena razlika med tema drevesnima vrstama, ki bi lahko bili pomembni za razvojni krog *M. dorsalis*, je, da se stroki trnate gledičevke ne razprejo, stroki rogovilarja pa se ob določeni stopnji zrelosti razprejo tako, da semena padejo na tla. Samice v naravi jajčeca odlagajo na stroke (Kurota & Shimada 2001). Za zdaj še ni znano, ali samice v naravi zaznajo, da v stroku ni semen, in ali odlagajo jajčeca tudi na semena, ki padejo na tla. V laboratoriju so namreč jajčeca odložile na stene steklene posode, v kateri so jih gojili, kljub temu, da so imele v posodi na voljo semena trnate gledičevke (Fritzsche et al. 2016). Ličinke so nato same poiskale semena (Fritzsche et al. 2016). Druga pomembna razlika, ki bi lahko vplivala na razvoj hrošča, je debelina semenske lupine. Semena rogovilarja imajo debelejšo semensko lupino kot semena trnate gledičevke, kar bi lahko vplivalo na uspešnost ličink pri vrtanju v seme. Tretja razlika je velikost semena. Po do sedaj objavljenih podatkih so na Madžarskem našli seme rogovilarja s tremi izhodnimi luknjami, značilnimi za *M. dorsalis* (György & Tuda 2020), v Mariboru pa seme trnate gledičevke z dvema razvitima odraslima hroščema (Šajna 2019). V tej raziskavi smo našli tudi seme trnate gledičevke s tremi izhodnimi luknjami, kar pomeni, da bi se teoretično v semenu rogovilarja, ki je večje od semena trnate gledičevke in ima posledično več založnega tkiva oziroma hrane za ličinko hrošča, lahko razvili tudi več kot trije hrošči.

Percepcija o tujerodni vrsti *M. dorsalis* se v Evropi spreminja. V starejši literaturi se omenja kot aklimatizirana tujerodna vrsta, v novejši literaturi kot naturalizirana, vse pogosteje pa se jo omenja tudi kot invazivno tujerodno vrsto. Za zdaj se *M. dorsalis* ne obravnava kot škodljivo tujerodno vrsto, saj ni neposredne škode za samo drevo in s tem pojavljanje *M. dorsalis* ne povzroča finančne škode za vzdrževanje zasajenih dreves in zatiranje hroščev v urbanem okolju ni potrebno. Tudi naravovarstveno vrsta ni pretirano zanimiva, saj se hrani s semeni gojenih dreves, ki niso ogrožena. Žal še vedno marsikje po Evropi napačno gledajo na vrsto *M. dorsalis* kot na popestritev lokalne favne (npr. Reinhardt et al. 2020).

Glede na to, da v zadnjem času v Evropi vse pogosteje opisujejo tudi gostiteljsko rastlino *G. triacanthos* kot invazivno tujerodno vrsto (pregled literature v Horvat & Šajna 2021b), vse kaže, da bo vrsto *M. dorsalis* treba obravnavati kot invazivno tujerodno vrsto. Še posebej zato, ker so odnosi med hroščem *M. dorsalis* in novo gostiteljsko rastlino trnato gledičevko kompleksni in presegajo herbivorijo (Horvat & Šajna 2021b). V določenih okoljskih razmerah se lahko namreč med tema vrstama vzpostavi mutualizem, saj ličinka z vrtanjem v seme povzroči poškodbo, ki lahko omogoči nabrekanje in kalitev semena gostiteljske rastline (Horvat & Šajna 2021b). Trnata gledičevka ima namreč zelo trda semena s fizično dormanco, kar pomeni, da lahko kalijo šele, ko nastanejo fizične poškodbe na semenski lupini (Baskin & Baskin 1998, Ferreras & Galetto 2010).

Za razvoj invazivnosti naturalizirane tujerodne vrste so pomembni procesi prenašanja (disperzije) in širjenja velikosti populacije (Richardson et al. 2000). Disperzija hroščev semenarjev ni dobro raziskana, še posebej ne v urbanih okoljih (Or & Ward 2003). Čeprav je hrošč *M. dorsalis* dober letalec, ne vemo, kako daleč lahko potuje po zraku. Predpostavljamo, da se v mestnih središčih lahko razširja vsaj na tri načine: (1) spontano z disperzijo odraslih osebkov, (2) antropohorno – s pomočjo človeka, ki hote in nehote prenaša stroke na nove lokacije, in (3) s trgovanjem okuženih semen. Zato bi bilo zanimivo raziskati, kako sorodne so si populacije *M. dorsalis* iz različnih krajev v Sloveniji.

V preučevanju bioloških invazij je pogosto omenjano »pravilo desetine« (Bright 1998), ki posplošuje, da se naturalizira le vsaka deseta tujerodna vrsta in da le desetina teh postane invazivna. Toda prav zaradi težko napovedljivih interakcij, ki se lahko vzpostavijo, ko se neka tujerodna vrsta naturalizira, smo mnenja, da velja pozorno spremljati tudi pojavljanje tujerodnih vrst, ki še ne izkazujejo invazivnosti.

# Summary

The East Asian seed beetle (Coleoptera, Chrysomelidae, Bruchinae) *Megabruchidius dorsalis* (Fåhræus, 1839) has been recorded in 17 European countries (Šipek et al. 2022). In the native area, *M. dorsalis* develops inside the seeds of native *Gleditsia* species (Tuda & Morimoto 2004), but in the new range it switched to two North American trees the honey locust (*Gleditsia triacanthos* L.) (Migliaccio & Zampetti 1989) and the Kentucky coffeetree (*Gymnocladus dioicus* (L.) K. Koch) (Callot et al. 2016). *M. dorsalis* has a multivoltine life cycle (Kurota & Shimada 2001, 2002) and different developmental stages can occur simultaneously (Šajna 2019). Females lay eggs on seed pods (Kurota & Shimada 2001). Larvae drill a hole into the seed and feed on the cotyledons (Kurota & Shimada 2001). Adult beetles emerge approximately after 50 days, which is fastest among seed beetles (Kurota & Shimada 2001, 2002). The aim of the study was to investigate the presence of *M. dorsalis* at different locations in Slovenia.

Mature *G. triacanthos* seed pods were collected from the ground at 35 locations and mature *Gy. dioicus* seed pods at 2 locations. Seed pods were kept in bags in laboratory at 22°C and under a 8 h:16 h light:dark regimen. Emerged beetles were determined under a stereomicroscope SMZ-161-TLED (Motic®) (Rheinheimer 2014, Pintilioaie et al. 2018) and the number of males and females (Tuda & Morimoto 2004) was recorded per location.

Exit holes on seed pods and seeds typical of *M. dorsalis* were found at all (35, 100%) *G. triacanthos* locations and one (50%) *Gy. dioicus* location. Adult *M. dorsalis* beetles were found at 26 (71%) *G. triacanthos* locations and one (50%) *Gy. dioicus* location. In Ljubljana, the species was confirmed at five (100%) locations and in Maribor at 16 (62%) locations. The beetle was also confirmed in Radenci, Petanjci, Pesnica pri Mariboru and in the Arboretum Volčji Potok. Several seeds of both trees with two exit holes and one seed of *G. triacanthos* with three exit holes were also found.

Alien seed beetle *M. dorsalis* successfully switched food source in Europe, specifically to the North American ornamental tree *G. triacanthos*. Consequently, it became naturalized in Europe and spontaneously expanded its range (Horvat & Šajna 2021a, Šipek et al. 2022). Our results suggest *M. dorsalis* has stable populations in Slovenia. Additionally, *M. dorsalis* can also develop inside the seeds of *Gy. dioicus* (Callot et al. 2016, György & Tuda 2020), as also shown in this study, which could further enhance the invasive potential of the species. Furthermore, indications show the relationship between *M. dorsalis* and *G. triacanthos* are complex and transcends herbivory (Horvat & Šajna 2021b). Larva entrance hole can be sufficiently damaging to the seed coat to enable seed imbibition and germination (Horvat & Šajna 2021b). When alien species become naturalized in new area, interactions with other alien or native species are difficult to predict, hence monitoring of all alien species regardless of their invasiveness status is advisable.

# Zahvala

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# Under the bridge: marking sites prove the presence of Eurasian otter *Lutra lutra* in the Poljanska Sora River valley, Slovenia

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**Abstract.** Knowledge on distribution of the Eurasian otter (*Lutra lutra*) in Slovenia in the past was very limited, due to the lack of studies of this species. Hunting and persecution reduced the species distribution area and densities. After its legal protection in 1973, the species seems to have increased in population size. A few studies had shown the Eurasian otter repopulating the Alpine region. To confirm the presence of the species in the Slovenian Pre-Alps, we searched for otter scat – spraint, on six tributaries to the Poljanska Sora River of Polhograjsko hribovje, the Pre-Alpine mountains around Polhov Gradec. Otter marking sites were exclusively in the riverbed (100 %), with a great majority found under the bridges (83 %). Using camera traps, we were able to collect video evidence of the otter reproduction and confirm that the species is reproducing in the studied region. Since this area could act as a corridor for the otter's recovery in the Adriatic River basin, it is crucial to continue to monitor its presence and the potential expansion to the west of the country.

Key words: *Lutra lutra,* recolonisation, alpine streams, marking behaviour, ecology

**Izvleček. Pod mostom: markiranje evrazijske vidre *Lutra lutra* potrjuje njeno stalno prisotnost v Poljanski dolini, Slovenija –** Znanje o pretekli razširjenosti evrazijske vidre (*Lutra lutra*) v Sloveniji je zaradi pomanjkanja študij pomanjkljivo. Lov in preganjanje živali sta pripomogla k upadu številčnosti osebkov in območja razširjenosti. Z zavarovanjem vrste leta 1973 se je populacija sčasoma ponovno prostorsko in številčno razširila. Nekaj študij tudi nakazuje na ponovno poselitev Alpskega območja. Za potrditev vidre v slovenskem predalpskem prostoru smo iskali iztrebke vider – vidrekov na šestih pritokih Poljanske Sore s Polhograjskega hribovja. Mesta iztrebljanja vidre so bila izključno v strugi potoka (100 %), ki so bili večinoma pod mostovi (83 %). Prek uporabe avtomatskih fotopasti smo zabeležili video dokaze o reprodukciji in s tem potrdili, da razmnoževanje že poteka v predalpskem prostoru. Ker je območje študije koridor do Jadranskega povodja, je to lahko pomembno območje za povezovanje in okrevanje populacije v Jadranskem povodju. Zato je ključno za nadaljnje spremljanje širjenja proti zahodu države.

Ključne besede: *Lutra lutra*, rekolonizacija, alpski potoki, markiranje, ekologija

# Introduction

In the mid-20th century, the Eurasian otter (*Lutra lutra*) was considered a widespread species in Slovenia, which was hunted and prosecuted. This decimated its numbers, pushing the species close to extinction in many parts of the country (Kryštufek 1991, Balestrieri et al. 2015). Lack of studies limited the possibility to evaluate their true extent at the time. The only available data about the historical otter distribution were hunting bags, showing a decimation of population in Slovenia since 1940 (Kryštufek et al. 1986). After the legal protection of the species in 1973, hunting bags were not available anymore and no monitoring was established ever since, even after 2004 (Hönigsfeld Adamič 2003) when Slovenia adopted the EU legislation and listed the Eurasian otter as a Natura 2000 species in 12 reference areas (Hönigsfeld Adamič 2003). Thus, the knowledge of the species distribution in Slovenia is currently restricted to data acquired from locally limited small scale studies (Hönigsfeld Adamič 2003, Hönigsfeld Adamič et al. 2011).

The Poljanska Sora River Valley is one of many Alpine river valleys for which reliable data on the presence of otters are lacking (Kryštufek 2001). In 2019, however, some information about observations of otters in the Poljanska Sora River were published in a local fishermen news websites (Križnar 2019). These observations indicate the Eurasian otter might have returned to the Slovenian Alpine region, completing the findings from Austria where the return of the species was the fastest amongst the Alpine countries (Kranz & Poledník 2020, Loy & Duplaix 2020). The closest area to the Poljanska Sora River Valley, where the presence of the Eurasian otter has been confirmed, is the Ljubljansko barje with tributaries from southern slopes of Polhograjsko hribovje, the Pre-Alpine mountains around Polhov Gradec (Kryštufek 2001, Hönigsfeld Adamič et al. 2009, Hönigsfeld Adamič et al. 2011).

The most straightforward and cost-effective method for detecting the presence of otters is to search for their scat, also called spraints. These are usually found in the proximity of their dens, called holts, or at feeding sites (Kruuk 2006). A high number of spraints indicates regular presence of otters in an area (Guter et al. 2008). The relationship between otter densities and spraint abundance can vary between different habitats and seasons (Lampa et al. 2015, Sittenthaler et al. 2020). As otters are territorial, spraints are used for marking their territory, as well as for other communication among the individuals living on the same shared territory, like establishing and/or maintaining social hierarchy, key food sources, or finding a mate (Trowbridge 1983, Kruuk 2006, Remonti et al. 2011). Some otter territories are used by several females at the same time with different and exclusive core areas, usually around holts, where they spend most of their time. Territories of males, which are usually larger, overlap with several female's territories (Kruuk & Moorhouse 1991). Due to a solitary lifestyle, two or more otters seen together are usually a female with her young (Kruuk 2006).

Sprains are easily identified by their shape and odour, which resembles fish oil. They are mostly deposited on surfaces or objects, which are standing out from their surroundings, such as large rocks striking form the water, confluences, grass tussocks in the riverbed, or on the riverbanks (Lampa et al. 2015). For example, otters living in slow-flowing rivers in karst poljes usually deposit spraints on the grass tussocks, meadows or moles on the riverbank (Krofel & Potočnik 2016). Human structures such as bridges with concrete shelves and other surfaces underneath can also provide an important site for marking, as they protect the spraints from weather conditions (Hönigsfeld Adamič 2003). An otter marking site is considered as an area where spraints are deposited within 1 m or are grouped (Kruuk et al. 1986). These sites can be used repeatedly by same individuals or even by several generations (Hönigsfeld Adamič 2003). Besides spraints, otters can also chemically communicate via urinating and cheek rubs (Hönigsfeld Adamič et al. 2011).

An initial discovery of signs indicating the presence of otter in the nearby area of the Poljanska Sora River Valley in spring 2020 (the Mavelščica stream; Kraševec own unpublished data) motivated us to explore the presence of the species in the Poljanska Sora River Valley and determine whether reproduction occurs. We first systematically searched for otter spraints in the fast-flowing alpine streams to confirm the otter presence in the study area. Then we analysed otter marking behaviour through listing accurate locations of the spraints in the fast-flowing alpine streams and comparing them to the ones typical for lowland slow-flowing water habitats (e.g. grass tufts on stream banks).

# Materials and methods

## Study area

We conducted our study in in the Poljanska Sora River Valley (Central Slovenia) on small alpine streams, all of them being southern tributaries to the Poljanska Sora River between Gorenja vas and Škofja Loka. The streams all originate in the northern slopes of Polhograjsko hribovje, which is largely protected as a Polhograjski Dolomiti Landscape Park. The mountains range consisting of dolomite and limestone is situated in the central part of Slovenia between the Poljanska Sora River in the north, the Gradaščica River to the south, and to the Brebovščica stream to the west (Fig. 1). The Poljanska Sora River is the only larger body of water in the area, being a small to medium sized river (with average discharge of approximately 10 $m^3/s$) with strong torrential character (ARSO 2021). It is one of the Sava River tributaries, part of the Danube River basin.

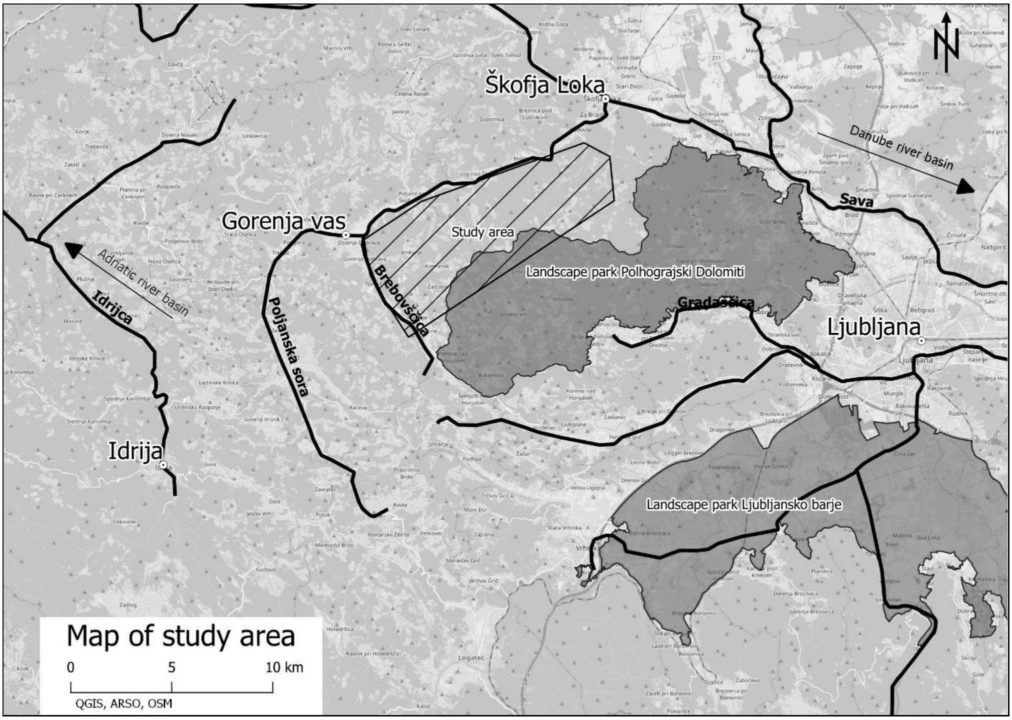


**Figure 1.** A map showing the overview of the study area and nearby protected areas of nature. Only major rivers important for the understanding of the study are marked down, and the Brebovščica stream limiting the study area. Flows towards the Adriatic and Danube River basins are also shown.
**Slika 1.** Zemljevid s prikazom območja raziskave ter bližnjih zavarovanih območij narave. Prikazane so samo glavne reke, ki so pomembne za razumevanje raziskave, ter potok Brebovščica, ki omejuje študijsko območje. Označeni sta tudi smeri toka proti Jadranskemu oziroma Donavskemu povodju.

The studied streams are fast and cascading in the upper parts with deep valleys and very steep, impassable banks in some parts, which distinguish them greatly from the lowland slow-flowing waters. Lower parts of the streams are also relatively fast flowing with easily accessible riverbanks. The widths of the streams range from 1 to 5 metres, with average discharge 0.15 $m^3/s$ and with strong torrential character. Streambanks are mostly covered with mixed forest in the upper parts, whereas the lower parts of the valley also include cultivated fields, artificial landscapes with human infrastructure, houses, and small villages. Some of the streams are channelized in parts. No stagnant waters exist in the vicinity.

## Study design

Field work was carried out on July 20 and 21, 2020. We searched for spraints under bridges, the most probable location for finding otter marking sites (Kruuk 2006). Each bridge represented a sampling site. Additionally, we surveyed 50 m upstream and 50 m downstream from the sampling site, both in the streambed and on the streambanks, in the width of 6 m from the stream (Fig. 2). The standard otter survey method recommends 600 m transects, but we chose 100 m transects, which represent more than 60% probability of detection if otter is present (Mason & Macdonald 1987).

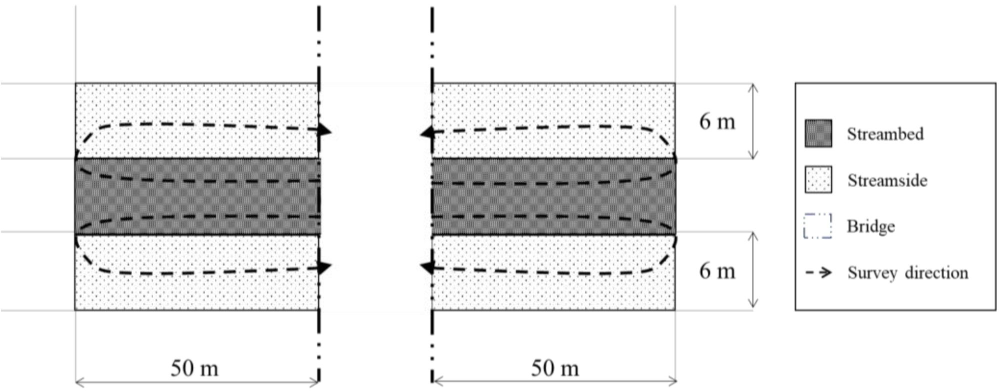


**Figure 2**. Otter spraint survey protocol scheme. We searched the streambed (dark grey), and 6 metres of streambank (light grey) 50 metres upstream and downstream from the bridge (dots and lines).
**Slika 2.** Shematski prikaz protokola iskanja vidrinih iztrebkov. Preiskali smo strugo (temno siva) in 6 metrov široki pas brežine (svetlo siva), 50 metrov gorvodno in dolvodno od mostu (črtkana črta s pikami).

The field work was carried out on 29 sampling sites on 6 different streams (Brebovščica, Todražica, Hotoveljščica, Sovpat, Bodoveljščica and Hrastnica). Most of the sampling sites were located downstream on parts of the stream that were closer to the outflow to the Poljanska Sora River. Our highest bridge was at an altitude of 611 metres and the lowest at an altitude of 366 meters. The number of sampling sites per waterbody was between 2 and 9 (Tab. 2). We chose sampling sites regardless of the type (wooden, concrete, etc.) and size of the bridge, i.e., culverts, up to 6 metres in length, and minor bridges, ranging from 6 up to 60 metres in length. The bridges were mostly of local importance. We had selected our sampling sites prior to the field survey. If it turned out that the water underneath our chosen sampling site was not accessible, we surveyed the adjacent sampling site instead. All sampling sites on the same water body were surveyed in the same day. At each sampling site (e.g., bridge), we categorized artificial changes of the streambed based on anthropogenic changes made to the banks (Natural, banks on both sides of the stream are not modified by humans; Moderately modified, banks are modified by less than 50% of our surveyed area or only one bank is almost completely modified by humans, while the other is strictly not; Artificial, banks are almost completely modified by humans), and the streamside land use (Meadow, area adjacent to the stream is covered mostly by meadows and/or fields; Forest, area adjacent to the stream is covered mostly by forest and/or shrubs; Artificial, area adjacent to the stream is mostly modified by humans) (Tab. 1).

**Table 1.** Category of streambank modification and type of surroundings 6 metres from the edge on each sampling site, with given proportions (n=116). Darker colour of the field matches the higher percentage.
**Tabela 1.** Kategorije spremenjenosti rečne brežine in tip okolice 6 metrov od roba struge na vsakem vzorčnem mestu, s podanimi deleži (n=116). Temnejša obarvanost polj se ujema z višjim deležem.

| **Type of streambank/ Tip okolice** / **Streambed modification/ Spremenjenost struge** | Forest/ Gozd (n = 50) | Meadow/ Travnik (n = 31) | Artificial/ Umetno (n = 35) | Total/ Skupno |
|---|---|---|---|---|
| Natural/Naravno (n = 80) | 42.2 | 17.2 | 9.5 | 68.9 |
| Moderately modified/Zmerno spremenjeno (n = 14) | 0.9 | 2.6 | 8.6 | 12.1 |
| Artificial/Umetno (n = 12) | 0.0 | 6.9 | 12.1 | 19.0 |
| Total/Skupaj | 43.1 | 26.7 | 30.2 | |

We identified the spraints following the descriptions from Krofel & Potočnik (2016). Then we determined if the location of found spraints was a marking site or not, based on whether a single spraint or a group of them was found. Finally, we set up 3 camera traps (MAGINON 4WKD) as a supplemental method to monitor the otter presence (for locations see the description of Fig. 4).

# Results

We confirmed the presence of otters where otter spraints were detected, hence we confirmed the presence of otter under 17 out of 29 sampling sites (Fig. 4), located on 4 out of 6 surveyed streams (Tab. 2). On one of these sampling sites (site number 1) where spraints indicated otter presence, we also video recorded two otters at the same site together (Fig. 5).

**Table 2.** An overview of the distribution of sampling sites along selected streams, their full lengths, and the number of found marking sites and collected samples.
**Tabela 2.** Podrobnejši prikaz razporeditve števila vzorčnih mest, markirnih mest, št. vzorcev in dolžin obravnavanih vodotokov.

| Stream/Potok (km) | No. of sampling locations/ Št. vzorčnih mest | No. of marking sites/ Št. markirnih mest | No. of samples/ Št. vzorcev |
|---|---|---|---|
| Brebovščica (9.4) | 9 | 2 | 66 |
| Todražica (3.8) | 2 | 2 | 15 |
| Hrastnica (9.3) | 2 | 0 | 8 |
| Sovpat (2.8) | 4 | 0 | 0 |
| Hotoveljščica (3.1) | 4 | 0 | 0 |
| Bodoveljščica (7.1) | 8 | 2 | 25 |
| **Sum/Skupno** | 29 | 6 | 114 |

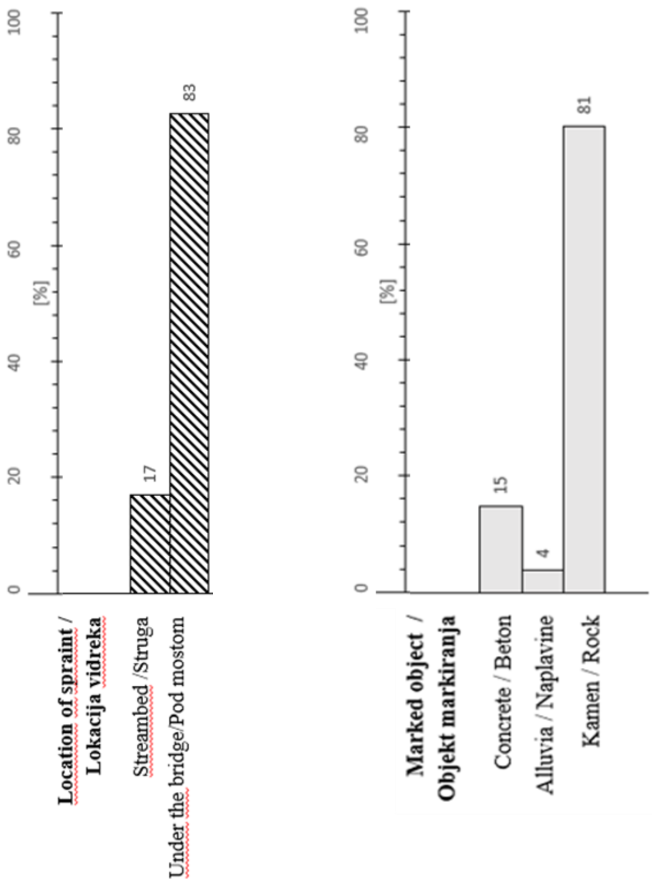


**Figure 3.** Location of otter spraint (left), and marked objects (right), according to the frequency of occurrence in percentage (%) of occurrence of all spraints (n = 114).
**Slika 3.** Lokacije vidrinih iztrebkov (levo) in objekti markiranja (desno), izraženi v deležu (%) vseh iztrebkov (n = 114).

In total, we found 114 otter spraints, with 7 ± 5 spraints per sampling site (Tab. 2). The majority of them were located under bridges (83%) or in the streambed nearby (17%). We did not find any spraints outside the streambed. At this point it must be stressed that almost half of the riverbanks were not surveyed because of their inaccessibility for the research team. On the other half, which was surveyed, we did not manage to find any spraints. The ones found in the streambed were mainly deposited on rocks (Fig. 3), but concrete shelves under bridges or other concrete structures and river debris were also used for defecating. Sprints were found individually (70%), or in groups on marking sites (30%), consisting of sprints which were undoubtedly of different age.

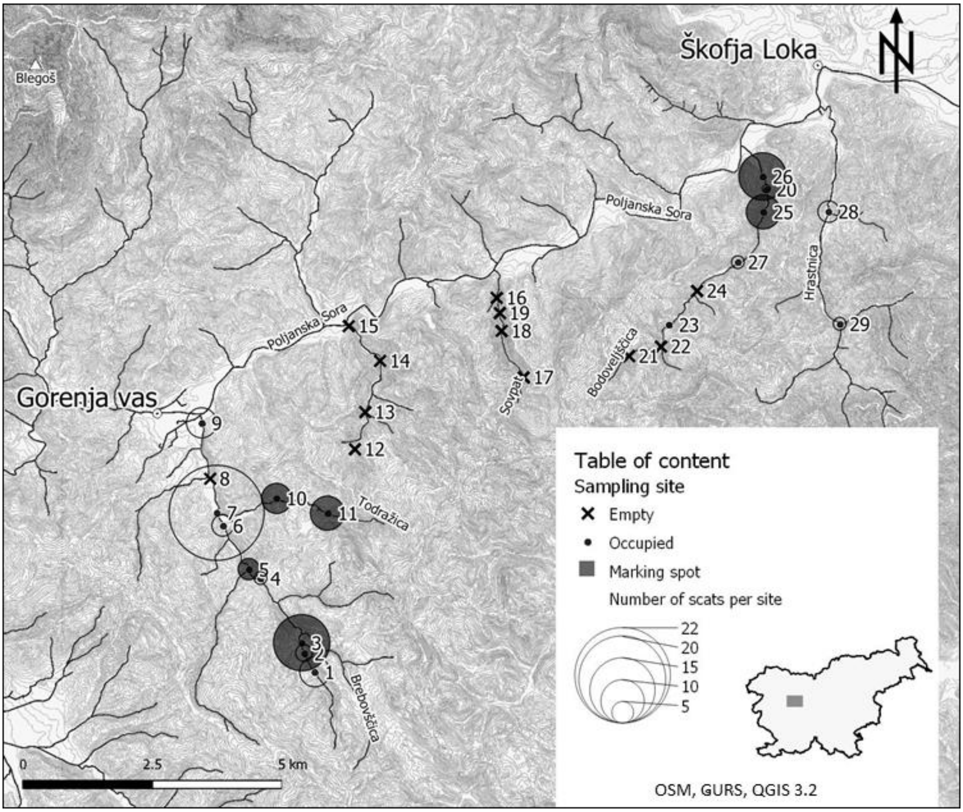


**Figure 4.** A map showing localities of field sampling sites in the Poljanska Sora River Valley, between Gorenja vas and Škofja Loka. The size of the circles corresponds with the number of otter (*Lutra lutra*) spraint found on the location. The crosses mark sampling sites without any spraint found. Dark colouring indicates the presence of a marking spot. Sampling was carried out under roadway bridges as well as 50 metres upstream and downstream. We set up camera traps on sampling sites 1, 3 and 10.

**Slika 4.** Zemljevid s prikazom lokacij mest terenskega vzorčenja v Poljanski dolini med Gorenjo vasjo in Škofjo Loko. Velikost točk ponazarja število vidrinih iztrebkov (*Lutra lutra*) na vzorčnem mestu. Križci ponazarjajo prazna vzorčna mesta, brez najdenih iztrebkov. Temna obarvanost označuje markirna mesta. Vzorčenje je potekalo pod cestnimi mostovi in 50 metrov tako dolvodno kot gorvodno. Avtomatske fotopasti smo namestili na vzorčna mesta 1, 3 in 10.

**Figure 5.** Screenshot from camera trap video, showing two otters present at the same time, probably a mother with her young. Video was recorded 20.7.2020 on the Brebovščica stream.
**Slika 5.** Zajem zaslona videoposnetka avtomatske foto pasti, na katerem sta vidna dva osebka, najverjetneje samica in njen mladič. Posneto 20. 7. 2021 na potoku Brebovščica.

The vast majority (68.9%) of streambank sampling sites were classified as »Natural«, followed by »Artificial« (19%) and »Moderately modified« (12.1%). The main type of habitat in the surroundings was the »Forest« category (43.1%), followed closely by the categories »Artificial« (30.2%) and »Meadow« (26.7%) (Tab. 1).

# Discussion

Despite the short duration of our study, we were able to show that the Eurasian otter is widely present in the Poljanska Sora River Valley. This is also confirmed by the fisherman's occasional observations from the previous year (Križnar 2019). It cannot be assessed whether the otters had been present in the area long before, due to the lack of studies (Kryštufek 1991, 2001). Based on our study, we can confirm their presence via target studies of marking sites on different tributaries, originating in Polhograjsko hribovje, of the Poljanska Sora River.

Due to the fact that our study area lacked information on otter presence (Kryštufek 1991, Kryštufek 2001), the number of streams and sampling sites (Fig. 4) confirming the presence of the species in this alpine valley is encouraging. Our study area is comparable to the habitat in Austria, where otter density studies related to the spraint density, which were done and confirmed with genetics (Sittenthaler et al. 2020).

According to the Austrian studies, we estimate 0.16 – 0.28 otters per km of waterway could inhabit the Poljanska Sora River Valley. This means that regarding the spraint distribution in our study area (35.5 km of waterways), the area could potentially be inhabited by 6 to 10 otters. Video record of two otters confirmed that reproduction already occurs (Kruuk 2006).

Our results of assessment of the habitat around sampling sites indicate a rather well-preserved habitat for the otter (Tab. 1). For otter to be able to colonise an area, it is important that the streambanks and the adjacent areas are natural to at least some extent. Dense and woody streambank vegetation and canopy (shrubs and trees), bare roots and stones that form holes in the riverbanks (potential holts), as well as inaccessibility to humans, are some of the most important factors determining the suitability of habitat for the Eurasian otter (Mason & Macdonald 1987).

Due to the wide distribution of otters in the Poljanska Sora River basin, it could be predicted that the species distribution could soon extend from the Danube River basin and connect with the Adriatic river basin where the species was absent or only occasionally present (Gregorc & Nekrep 2010, Hönigsfeld Adamič et al. 2010, Balestrieri et al. 2015). The connection and extension of the range would significantly improve the species conservation status in Slovenia.

Contrary to some publications stating that otters use mainly grass tufts located on riverbanks and adjacent areas as marking sites (Krofel & Potočnik 2016), our results show that otters living in fast-flowing alpine streams with steep riverbanks exclusively use the streambed area, more accurately, the sections under bridges, as their preferred sprainting sites (Fig. 3). First, the alpine environment is topographically very different from a typical karst or other lowland area where most of the studies were done (Krofel & Potočnik 2016). Otters might not be able to reach the area surrounding the stream because of the steep slope of the streambanks, or because otters mostly use the streambed, sprainting in the streambed could be more successful for effective communication between individuals. Moreover, otters prefer to spraint where their chemical signal can persist for as long as possible (Hutchings and White 2000). Bridges provide cover from precipitation and heat (Hönigsfeld Adamič 2003) and spraints are therefore probably emitting chemical signal for longer periods than those deposited in places with no cover. The use of rocks and concrete structures under the bridges, as the most common, but not exclusive, surface for droppings, can also be confirmed in the variety of other studies (Macdonald 1980, Kruuk 2006, Hönigsfeld Adamič et al. 2011). It must be highlighted, however, that in many cases the streams were followed by nearby roads (not directly quantified, but also categorised as »Artificial« surrounding, Tab. 1), making a part of the area adjacent to the stream completely unsuitable for a typical otter marking site, typical in the lowland slow-flowing water habitats. On the other hand, no spraints were detected even in the meadows representing more than a quarter of the streambanks and often adjacent to a naturally shaped streambed (Tab. 1).

Even though we initially planned to visit more streambanks, they could not be accessed due to dense vegetation or steep slopes of the riverbanks. Consequently, it is possible we might have missed some sprainting sites outside the streambed. We, however, do not expect that otters would choose such areas for marking, as they prefer to spraint in conspicuous areas where their signal will be available, accessible and obvious to other otters (Gorman & Trowbridge 1989).

Further genetic analyses would give a better insight in the detailed demographic parameters of the otter population in the area (Sittenthaler et al. 2020), while further (at least local) scale studies such as ours could help reveal the expansion of otter towards the Adriatic river basin. We suggest that our study offers a good start of otter studies in the area, and that additional surveys of the Eurasian otter should be done in summer, or in winter to early spring. The tendency of otters marking in that time is up to ten times more often (Kruuk 1992) and the river's still unfoliaged vegetation allow us easier access for surveying a larger area of streambanks. To evaluate the status of the otter population on the national scale, extensive national monitoring scheme should be established.

# Povzetek


Na območju desnih pritokov Poljanske Sore, ki izvirajo v severnem delu Polhograjskega hribovja (Sl. 1), smo med 20. in 21. julijem 2021 opravili raziskavo ekologije evrazijske vidre (*Lutra lutra*). Namen raziskave je bil potrditi njeno prisotnost, ugotoviti njeno razširjenost ter način markiranja in potencialno zaznati razmnoževanje na območju.

Za kune (Mustelidae), med katere sodijo tudi vidre, je značilen kemičen način markiranja, kjer prek iztrebljanja na zelo očitnih mestih, kot so kamni, štori in ožine, označujejo svoj teritorij. Prav to lastnost velja izkoristiti med številnimi raziskavami vider ob domnevi, da iztrebki v določenem območju pomeni tudi prisotnost vidre (Kruuk 2006). Za zaznavanje vidrinih iztrebkov, t. i. vidrekov, smo uporabili prilagojeno standardizirano metodo za ugotavljanje prisotnosti vidre. Namesto priporočenih 600-metrskih smo opravili zgolj 100 metrov dolge in 12 metrov široke transekte ob mostovih (Sl. 2) (Mason and Macdonald, 1987). Kljub temu to še vedno ustreza več kot 60-odstotni verjetnosti za zaznavo vidrekov, če so le ti na območju opaženi (Mason & Macdonald 1987). Vsak transekt je bil razdeljen na več delov, in sicer most, breg in struga (Sl. 2). Slednja sta bila kategorizirana glede na oceno naravnega stanja posameznega dela vodotoka (Tab. 1). Naše študijsko območje je zajemalo 6 vodotokov, in sicer Brebovščico, Todražico, Hotoveljščico, Sovpat, Bodoveljščico in Hrastnico.

Vidro smo potrdili na 4 od 6 vodotokov oz. na 17 od 29 lokacij (Tab. 2). Vsega skupaj smo zaznali 114 vidrekov, od katerih so bili prav vsi (100 %) v strugi, večinoma (83 %) pod mostovi (Sl. 3). Prav tako smo z uporabo avtomatskih foto pasti v objektiv na lokaciji št. 1 ujeli dve vidri, skoraj zagotovo samico z njenim mladičem (Sl. 5). Evrazijska vidra je namreč samotarska vrsta in opažanje dveh ali več vider hkrati ponavadi pomeni, da gre za samico z njenim mladičem oz. mladiči (Kruuk 2006).

Z našo raziskavo smo potrdili prisotnost ter razmnoževanje evrazijske vidre in zabeležili njeno razširjenost na južnih pritokih Poljanske Sore (Sl. 4). Ugotovili smo veliko preferenco do markiranja v strugi (Sl. 3). Ugotovili smo, da se slednje namreč precej razlikuje od načina markiranja, ki je značilen za nižinske vodne habitate, kjer se vidra približno v dveh tretjinah primerov iztreblja na območju ob vodotokih (breg ali zaledje), kot je prikazano v Krofel & Potočnik (2016). Domnevamo, da gre pri tem za razliko v topografiji območja, saj so v nižinskem območju bregovi bolj dostopni kot v predalpskem. To dejstvo dokazuje, da ob popisu tudi za nas skoraj polovica bregov ni bila dostopna, prav tako najverjetneje tudi za vidre. Takšna gostota iztrebkov in markirnih mest v podobnem habitatu v Avstriji pomeni, da bi na kilometer vodotoka lahko živelo od 0,16 do 0,28 vider (Sittenthaler et al. 2020), zato bi v južnih pritokih Poljanske Sore, ki skupaj tvorijo 35,5 km vodotokov, lahko živelo od 6 do 10 vider.

Genetske analize bi omogočile boljši vpogled v podrobne demografske parametre populacije vidre na tem območju, medtem ko bi nadaljnje (vsaj lokalne) ekološke študije, kot je naša, pomagale razkriti širjenje vidre v jadransko porečje. Za podrobnejšo obravnavo stanja populacije vidre v Sloveniji pa je nujna vzpostavitev nacionalnega monitoringa vrste.

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# Asiatic fern *Cyrtomium fortunei* J. Sm. (Dryopteridaceae) – a new naturalized fern in the flora of Slovenia

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**Abstract.** A naturalized population of Asiatic fern *Cyrtomium fortunei* J. Sm. was found in the surroundings of Nova Gorica. It is the first record of the species in the country, although the locality is an expected prolongation of the secondary distribution range with the species' concentration in southern foothills of the Carnic Prealps (N Italy). The locality is characterized by mild temperate submediterranean climate and slightly degraded forest vegetation of the alliance *Carpinion orientalis*, where the fern occurs in the herb layer. Local occurrence of the fern represents the easternmost point of its secondary distribution range in Northeastern Italy, although this could possibly be connected with the immediate vicinity of the graveyards at the top of the slope.

Key words: biological invasions, invasive alien plant species, naturalization, ferns

**Izvleček. Azijska praprot *Cyrtomium fortunei* J. Sm. (Dryopteridaceae) – nova naturalizirana vrsta praproti v Sloveniji –** V okolici Nove Gorice je bila najdena naturalizirana populacija azijske praproti *Cyrtomium fortunei* J. Sm. kar je prvi podatek o uspevanju te vrste v Sloveniji. To nahajališče je hkrati pričakovani podaljšek zgostitve sekundarnega pojavljanja te vrste na južnem vznožju Karnijskih predalp (S Italija). Za nahajališče te vrste je značilno blago zmerno toplo submediteransko podnebje; gozdna združba, v katere podrasti je rastišče, pripada degradiranemu gozdu iz zveze *Carpinion orientalis*. Pojavljanje te praproti na tem mestu dosega skrajni vzhodni del njenega sekundarnega areala v severovzhodni Italiji, morda pa je povezano tudi z neposredno bližino pokopališča na vrhu pobočja.

Ključne besede: biološke invazije, invazivne tujerodne rastlinske vrste, naturalizacija, praprotnice

# Introduction

Naturalized alien floras are focus of research and nature conservation concern for the last decades. Thousands of alochtonous flowering plant species have been established and hundreds of them became invasive in Europe, but only about ten alien fern species were successful during colonization of the European continent (Prelli & Boudrie 2021). In Slovenia, *Azolla filiculoides* can be found as a pest in some garden pools, *Cystopteris bulbifera* is escaping from the Alpine botanical garden Juliana, both of American origin, and an ephemeral occurrence of some *Nephrolepis* sp. was detected on a derelict house in Ljubljana about 10 years ago. None of the mentioned three alien ferns occur with completely naturalized populations.

Genus *Cyrtomium* has about 35 species, mostly confined to E and SE Asia. Its centre of diversity is in SW China (Zhang & Barrington 2013). Some of the species are popular ornamental plants and can also escape from cultivation in suitable ecological conditions. In Europe, two *Cyrtomium* species are reported as being locally naturalized, i.e. *C. falcatum* (L. f.) C. Presl and *C. fortunei* (Prelli & Boudrie 2021). Representatives of the genus are easily distinguished from European native ferns, their fronds are imparipinnate with large, more or less asymmetric pinnae and even larger terminal segment slightly lobed basally. Abaxial surface of pinnae is covered with many dispersed sori with almost circular peltate and persistent indusia. Pinnae have anastomozing venation and acuminate apex. The only native fern that can superficially resemble any of the *Cyrtomium* species is *Polystichum lonchitis* (L.) Roth.

Two *Cyrtomium* species which are reported as locally naturalized in Europe are easily distinguished from each other, *C. falcatum* has leathery fronds with 4-10 (-12) pairs of pinnae, distinctly shiny, margins undulate or coarsely dentate, and it can be found in ruderal sites like derelict stone walls or ruins. On the other hand, *C. fortunei* has papery fronds with (8-)10-25 pairs of pinnae, not shiny, margins minutely crenulate-denticulate, and it has also been recorded in natural vegetation on limestone and sandstone (Yatskievych 1993, Zhang & Barrington 2013). Another difference is in the karyology of the two species, *C. falcatum* is reported to be diploid, tetraploid and apomictic triploid, whereas *C. fortunei* is triploid with apogamic apomyxis (Iwatsuki 1995, Ootsuki et al. 2012). Peroni & Peroni (2000) checked the micromorphology of leaf epidermis for distinguishing the two species and they proposed measurements of stomata as reliable trait for identification. As the material of both species belonged to triploids, we have no information about stomata measurements in 2x and 4x populations of *C. falcatum*. Apomictic propagation can partly explain success of *C. fortunei* in other continents (Krippel & Thommes 2021).

*C. fortunei* has erect rhizome, densely covered with dark brown scales present also in basal part of stipe. Stipe is 12-26 cm long and 2-3 mm wide at base with a longitudinal furrow (U-shaped in transection). Frond lamina oblong-lanceolate, 20-40 cm long and 6-12 cm wide, 1-imparipinnate, rhachis and pinnae abaxially glabrous or with sparse linear scales resembling simple trichomes. Lateral pinnae 7-16 (-29) pairs, alternate, with short petioles, lanceolate to falcate; middle pinnae 5-8 × 1.2-2 cm, base oblique, acroscopic part with weakly developed blunt auricle, basiscopic part cuneate, margins entire or sometimes serrulate, apex acuminate; terminal pinna triangularly-ovate-lanceolate, sometimes lower portion with 1 or 2 lobes, 3-6 × 1.5-3 cm; papery, venation anastomosing to form 4 or 5 rows of areoles on each side of pinna midrib, each areole with 1 or 2 included free veinlets. Sori irregularly scattered on abaxial surface of pinnae; indusia peltate, circular, margins entire (description mostly from Zhang & Barrington 2013).

In its native range (China, NE India (Manipur), Japan, S Korea, Nepal, Thailand, N Vietnam), the species is reported for limestone crevices in open areas or forests in altitudinal range between 100 and 2,400 m a.s.l. Its occurrence in Iran is also reported as spontaneous (Gholipour & Greuter 2010), which would change the native distribution range of the species (and genus). Even if that would be the fact, known European naturalized populations are more than 2,000 km away, with their occurrence distinctly linked to escapes from cultivation.

Neophytic distribution range of *C. fortunei* comprises some parts of N America (mostly SE of US) and parts of Europe (Gholipour & Greuter 2010). In Europe, the genus has been represented mostly by escapes of *C. falcatum*, but in the last decades there have been several records of established populations of *C. fortunei*. There are only a couple of scattered records from the first half of the 20$^{th}$ century (Pignatti et al. 1983, Krippel & Thommes 2021), but in the last four decades the process of naturalization took place in several parts of Europe. It was discovered naturalized in NE Italy (Friuli-Venezia Giulia, Monte De Ragogna; Pignatti et al. 1983, Pignatti 2017) and in the following years several populations were recorded concentrated mostly in the southern foothills of the Carnic Prealps, where the species has become naturalized (Bona et al. 2005). Similar pattern of naturalized occurrence follows in the western direction towards Lombardy (Martini et al. 2012) and Novara (Aeschimann et al. 2004). In addition to that, it has been recorded in many scattered localities in N Italy, where its occurrence is mostly explainable by cultivation in the vicinity. The oldest record of the species for N Italy dates back to 1912 (Bona et al. 2005). In Switzerland, populations are well established in the sub-mediterranean part of Tessin/Ticino (Eggenberg et al. 2018), a direct prolongation of the secondary distribution range in Lombardy, and there are still some further local records in central and western Switzerland (Infoflora 2021). It has not been reported by previous floristic monographs (before 2010). Its habitat is mentioned as wet walls and rocks in the *Cystopteridion* vegetation (Infoflora 2021). In France (Tison & Foucault 2014), it is reported as rare species in the Alpes Maritimes around Nice, where it has been recorded in lowlands (100-200 m a.s.l.), naturalized on shaded tuff walls in humid local conditions, but its local spreading can be interpreted as invasive in some of the narrow valleys (Krippel & Thommes 2021). In the Benelux area, *C. fortunei* is reported as »rare escape from cultivation but increasingly found since 2001« (Verloove 2010), and in addition to ruderal places it has already been recorded in natural locality with terrestrial population in woodland in Moen (Verloove 2010). In Nederland, it has been reported in ruderal places (Denters & Verloove 2007-2008) and similarly in Luxemburg (Krippel & Thommes 2021) and England (Page 2005).

Cultivation of *C. fortunei* as ornamental plant has a long history. As early as 1904, it was mentioned as hardy plant suitable for outdoor planting (Kunert 1902). It has not been so popular as the closely related *C. falcata* (Studnička 2009), but it is mentioned as suitable for herb layer under trees or shrubs with need of winter protection and possibility of propagation by spores, sensitive to winter wet weather (Jäger et al. 2008). Also in Slovenia, *C. fortunei* was mentioned as a house pot plant in some of the translated handbooks (Spangenberg 1976, Vermeulen 2005), but it is not possible to interpret this fact as being connected to naturalized occurrence of the species. In comparison with some other European countries, Slovenian horticultural trade is not very developed and diversity of available plants is comparably low. Among ferns and fern allies, there are normally less than 10 species available for outdoor gardening in bigger garden centres or from internet providers. And the two mentioned *Cyrtomium* species are often the only really exotic ones.

# Material and methods

Plant material was sampled during routine field floristic mapping aimed at *Buddleja davidii* populations. All the previously reported localities of that invasive alien shrub scattered in lowland parts of Slovenia were systematically visited, vegetation relevés done and in the vicinity additional populations of *B. davidii* searched for in suitable sites. Systematic sampling took place in summer 2021 and will go on until 2023. Collected herbarium vouchers are deposited in herbaria LJU (University of Ljubljana, Biotechnical Faculty) and LJS (Jovan Hadži Institute of Biology, Scientific Research Centre of the Slovenian Academy of Sciences and Arts). Identification of plant material was done using many literature sources (Yatskievych 1993, Peroni & Peroni 2000, Verloove 2010, Zhang & Barrington 2013, Tison & Foucault 2014, Prelli & Boudrie 2021).

# Results and discussion

Finding of *C. fortunei* in the SW part of Slovenia has not been unexpected, as the fern has been naturalized in several localities of Northern Italy, where it was first recorded more than a century ago. Slovenian locality is almost on the Italian border, with the Soča/Isonzo River crossing the state border just about 1 km SW from the spot.

Slovenian population was discovered as locally naturalized in herb layer of forest on the left bank of the Soča River (0047/2 Slovenia: Nova Gorica, Solkan, Goriščlittle, forest on NW slopes above the Soča River, ~70 m a.s.l., 45,976912° N 13,649904° E, leg. N. Jogan, 2. 7. 2021). Local population has about 100 plants with ripe fronds (Fig. 1) and covers an area of about 100 $m^2$. Nearest known populations on the Italian side of the border are in the vicinities of Cividale del Friuli around 15 km to the NW and Trieste about 35 km to the SE (Bona et al. 2005). Forest above the Soča River bank is slightly degraded due to a walking path and nearby settlement on the top of the slope. As a consequence, there are several invasive alien species in the forest as e.g. *Buddleja davidii*, but the overall impression of the locality is that ground layer of the forest vegetation is quite natural. At the top of the slope, there is a WW1 military graveyard now maintained as a park, as well as a bigger civil cemetery. Graves in Slovenia are often decorated with planted ornamental plants, flower bouquets, mourning wreaths and other flower arrangements, which could possibly be the source of the discussed fern population merely 100 m away.

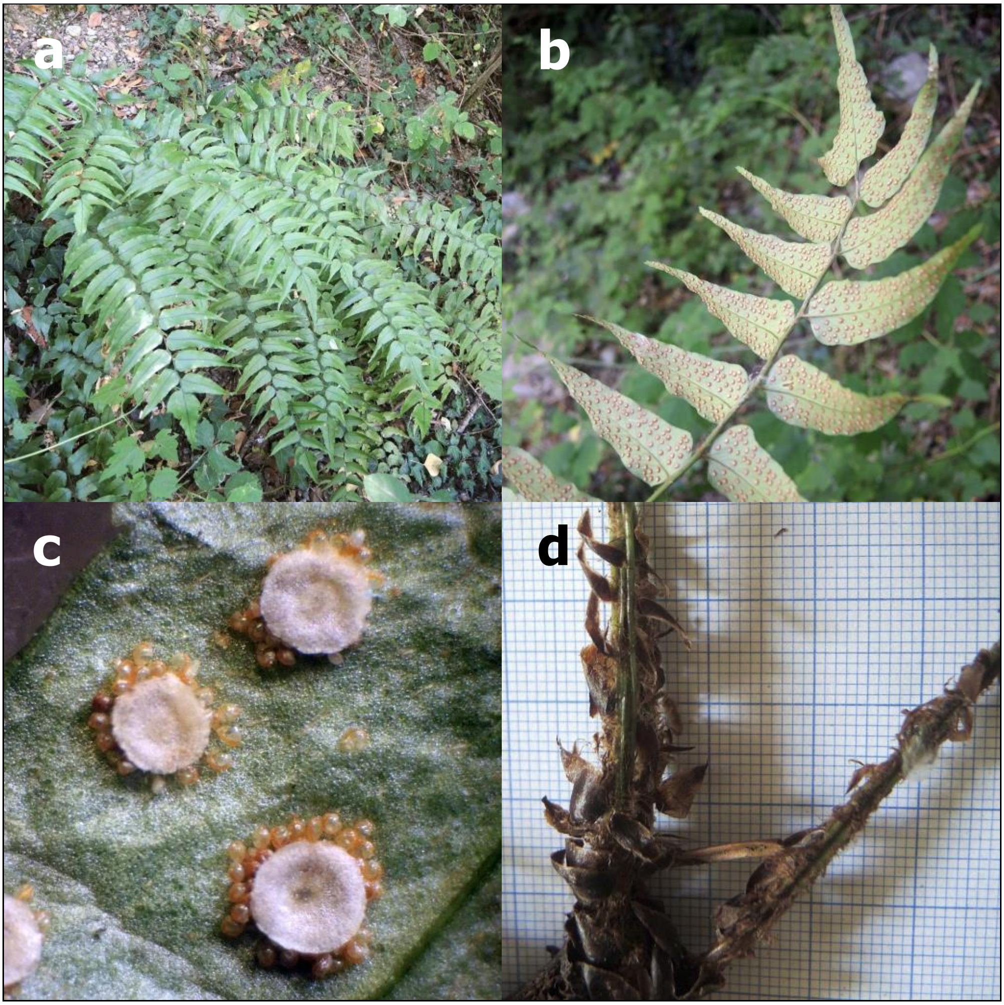


**Figure 1.** a) Asiatic fern *C. fortunei* in the locality near Nova Gorica; b) abaxial surface of frond with typical sori pattern; c) sori with peculiar persistent circular peltate indusia; d) dark scales at the basal part of stipe (Photo: N. Jogan).
**Slika 1**. a) Azijska praprot *C. fortunei* na nahajališču blizu Nove Gorice; b) spodnja stran lista s tipičnim vzorcem trosišč; c) trosišča s prepoznavnimi dežnikasto oblikovanimi obstojnimi zastiralci; d) temne luske pri dnu listnega peclja (Foto: N. Jogan).

Climatic conditions in that particular part of Slovenia are very mild (https://meteo.arso.gov.si/met/sl/climate/maps/), with average annual temperature around 13°C, total annual precipitation close to 2,000 mm with most intense autumnal rains and almost without snow cover (less than 5 snowy days per winter).

Vegetation in the discussed locality is slightly degraded forest that could be classified into the alliance *Carpinion orientalis* (*Quercetalia pubescenti-petreae, Quercetea pubescentis*). There is evident influence of riverine forest nearby. Forest community is shaded and moist with ruderal species indicating human impact (as explained above). Substrate is conglomerate, soil is rich in sand (due to the Soča River floods), elevation 88 m above sea level, exposition W, inclination: 30°, average height of the tree layer: 20 m, average height of the shrub layer: 5 m, coordinates (WGS84): 45,976944 N, 13,650000 E, plot area: 100 m², date: 2.7.2021. Tree layer (80% coverage): *Quercus pubescens* 3, *Ostrya carpinifolia* 2, *Populus nigra* 2, *Hedera helix* +, *Robinia pseudoacacia* +; shrub layer (50%): *Sambucus nigra* 3, *Rubus fruticosus* agg. 3, *Euonymus europaeus* 1, *Laurus nobilis* 1, *Ruscus aculeatus* 1, *Acer campestre* +, *Asparagus acutifolius* +, *Clematis vitalba* +, *Cotinus coggygria* +, *Crataegus monogyna* +, *Fraxinus* ornus +; herb layer (60%): *Hedera helix* 3, *Cyrtomium fortunei* 2, *Lamium maculatum* 1, *Lamium orvala* 1, *Acer campestre* +, *Asplenium trichomanes* +, *Brachypodium sylvaticum* +, *Calamagrostis varia* +, *Campanula rapunculoides* +, *Clematis vitalba* +, *Fraxinus ornus* +, *Geranium robertianum* +, *Helleborus odorus* +, *Hepatica nobilis* +, *Laurus nobilis* +, *Lonicera caprifolium* +, *Mycelis muralis* +, *Rubus fruticosus* agg. +, *Tamus communis* +, moss layer (15%).

Spread of alien plants in natural vegetation could present a threat to local habitat type structure and biodiversity. Bearing in mind more than four decades of sub-spontaneous spread of *C. fortunei* to suitable natural habitats in the wider area of southern Alpine foothills, on one hand we could say that it is not possible to prevent spread of tiny spores to the neighbouring habitats as long as the fern is cultivated as an ornamental and maybe used also as decoration in flower bouquets (and hence appearing in graveyards, garden garbage hips, etc.). On the other hand, after all these decades the populations of the discussed fern are still small and scattered, so at the moment it is not possible to recognize it as a distinct threat to the local biodiversity. But for the future, it can be expected that naturalization process would not go on and in some of the next phases invasiveness of the species could emerge.

As the newly discovered population is clearly naturalized and there are more of them in the neighbouring areas of Italy, it seems highly probable that *C. fortunei* has already been established locally also in some other suitable sites in the Sub-Mediterranean part of Slovenia. It would be even possible that it has been overlooked due to general neglect for ferns as they are often treated as superficially similar. Care should be taken during floristic exploration of shaded stony forests in the river valleys in Goriška Brda and in the Vipava Valley, where comparable micro-ecological conditions could be available.

At the end, as a newly naturalized fern in Slovenian flora, the species would deserve a vernacular name. Availability of names used by Slovenian gardeners is scarce, either they provide it as a »fern« (praprot), simply transliterated name into »cirtomium«, some use only the name of a cultivar (»clivicola«), or pragmatically sell the fern under English vernacular names (holy fern) or simply translate one of the vernacular names in other languages (e.g. »lažni aspidij« as a translation of false aspidium). Obviously a new name has to be coined. If we would like to derive the name from Latin, the author of the name Cyrtomium (Presl 1836) used the Greek term kyrtos in the meaning of arch, curved as an arch, to refer to the anastomosing leaf nervature. In fact, in the discussed species leaf venation is so obscure that anastomoses are hardly visible. But a distinct trait of the genus is also the shape of pinnulae that are asymmetrically curved in the form of a schyte blade, again resembling an arch shape. Locally, a small traditionally used knife with curved blade is called »fovč«, so using that term as a linguistic root we propose the name »fovčevka« as a vernacular name for the genus *Cyrtomium*. Naming the species is easier, as it is dedicated to the 18th Century botanist Robert Fortune (Prelli & Boudrie 2021), so the Slovenian name would then be »Fortunova fovčevka«.

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FIELD NOTE

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# First sighting of 'white' and 'black' olms (*Proteus anguinus*) in the same spring

## Prvo opažanje belega in črnega močerila (*Proteus anguinus*) v istem izviru

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The olm (*Proteus anguinus*) is the largest inhabitant of caves and subterranean waters along the Dinaric Karst (Sket 1997). Across its range, distinct evolutionary lineages are found (Gorički & Trontelj 2006, Trontelj et al. 2009), with most of them exhibiting highly troglomorphic features, including the absence of pigmentation and eyes, an elongated snout and limbs (referred to as 'white olms'). However, a single lineage (*P. anguinus parkelj*) in south-eastern Slovenia has presumably retained ancestral features, including a dark colouration, visible eyes, a shorter snout and limbs, named the 'black olm' (Arntzen & Sket 1997, Bizjak Mali & Sket 2019, Ivanović et al. 2019). Most of the lineages are allopatric, with non-overlapping distributional ranges. In the region of Bela krajina, however, the white olm lineage of southeastern Slovenia comes into contact with the black olm (Gorički et al. 2017). While the southeastern Slovenian lineage is distributed across a large part of Dolenjska and has been reported from over a hundred locations, the black lineage has a very limited distribution and is only known from 14 locations (Fig. 1; Hudoklin & Aljančič 2017, SubBioDB 2021). Direct contact between these two distinct lineages had not been observed before, but a study based on environmental DNA (eDNA) found that a sample from a spring close to the river Dobličica (Šprajcarjev zdenec) contained the DNA of both lineages (Gorički et al. 2017). The distance between the closest locations, where the black and white olms have been visually verified, is around 1.5 km in linear distance (Gorički et al. 2017, Hudoklin & Aljančič 2017, Năpăruș-Aljančič et al. 2017). However, linear distances are not necessarily representative or even meaningful for hydrological connections. A study on hydrological connections in the Dobličica catchment showed that the water from Otovski breg reappears in springs near the Dobličica (Habič et al. 1990). Therefore, while both eDNA data and hydrological connections suggest that black and white olms could have come into contact, this has not been confirmed by field observations as yet. Genetic and morphological data did not reveal any hybrids between black and white olms, although there are too few samples available from the potential contact zone to entirely dismiss sporadic hybridization (Gorički et al. 2017).

We report herewith on the first field observations of black and white olms at the same location, the Planinec spring. During the survey of springs potentially inhabited by black olms (Gorički et al. 2017), on 3.6.2021, we observed a young medium-sized black olm (~15 cm) in Planinec spring at 1:00 in the night. Together with the previous sighting (Gorički et al. 2017), this is the second visual confirmation of the black olm occurring in this spring. On the second survey of this spring on 12.10.2021, a white olm was observed at 22:40. In the late-night hours, at 00:40, presumably the same white olm was observed again. After waiting and checking the spring again at 1:15, the olm was not observed anymore. During the third survey on 25.3.2022, a white olm was observed and caught for closer inspection (Fig. 2). After measuring its length and body mass it was released back into the spring. On the fourth survey on 11.5.2022, three black olms were observed. During all surveys, olms reacted to our white lights and immediately retreated to the subterranean part of the spring. The only exception was the white olm that was caught and inspected on 25.3.2022. The spring was overgrown with plants at the time, and the olm swam into these plants on the shore of the spring.

Based on their eDNA study, Gorički et al. (2017) found that the spring Šprajcarjev zdenec (600 m upstream) contained DNA of both black and white olms, although neither were visually confirmed. However, it was suspected that additional sampling may likely result in further springs nearby with co-occurring olms, potentially by passive dispersal of eDNA. Since Planinec is downstream of Šprajcarjev zdenec, passive dispersal of eDNA to Šprajcarjev zdenec is not likely. Therefore, it can be assumed that black and white olms come into contact in at least these two springs, and probably also in the springs in between.

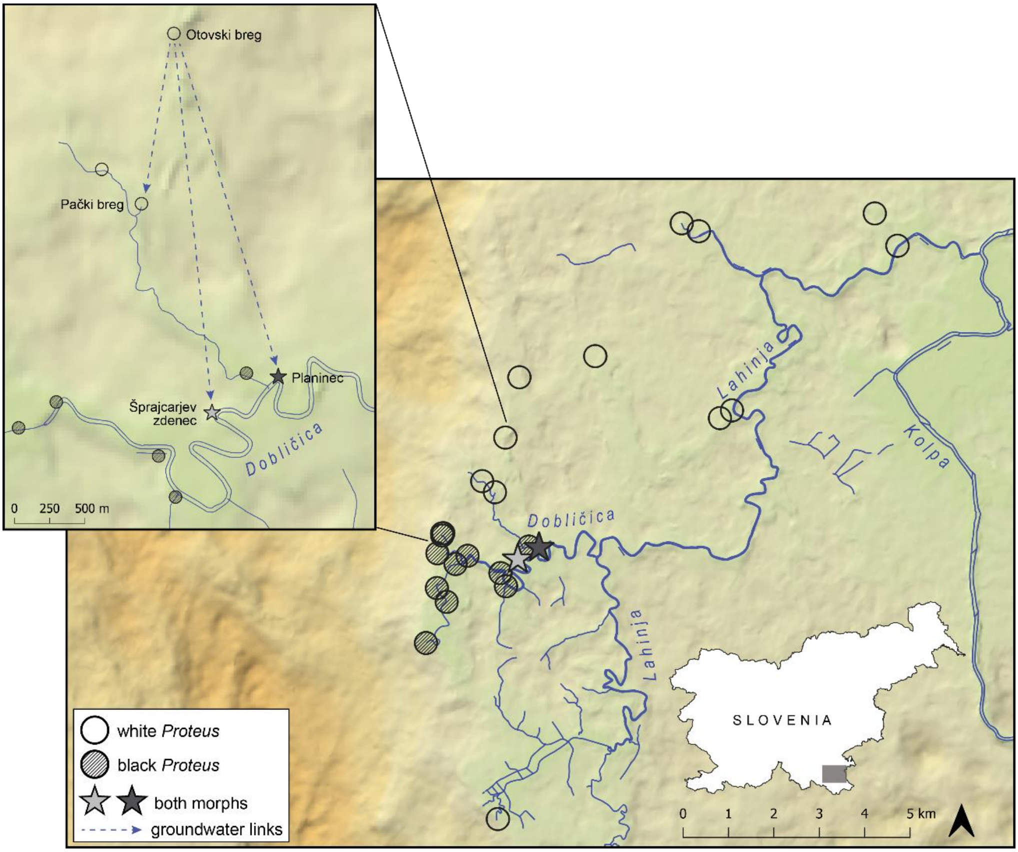


**Figure 1.** Distribution of the white olm (*P. anguinus*) and the black olm (*P. anguinus parkelj*) in Bela krajina, Slovenia. Data retrieved from SubBio Database (SubBioDB 2021) and Gorički et al. (2017). Unreliable and false data according to Hudoklin and Aljančič (2017) excluded. Map produced with QGIS version 3.10.12, using Copernicus data and information funded by the European Union - EU-DEM (European Union 2016) and rivers (Geodetska uprava Republike Slovenije 2010) as base layers. In the top left corner, a magnification of the proposed contact zone between black and white olms is shown, including known groundwater connections between Otovski breg and springs near the river Dobličica (Habič et al. 1990).

**Slika 1.** Razširjenost belega (*P. anguinus*) in črnega močerila (*P. anguinus parkelj*) v Beli krajini, Slovenija. Podatke smo pridobili iz Baze o podzemnem živalstvu (SubBioDB 2021) in prispevka Gorički in sod. (2017). Podatke, ki so bili glede na Hudoklin in Aljančič (2017) nezanesljivi ali napačni, smo izključili. Zemljevid je izdelan s programom QGIS verzija 3.10.12. Kot osnovna sloja smo uporabili podatke Copernicus in informacije, ki jih financira Evropska unija - EU-DEM (European Union 2016) in sloj rek (Geodetska uprava Republike Slovenije 2010). V zgornjem levem kotu je povečano območje kontaktne cone belega in črnega močerila, vključno z znanimi podzemnimi vodnimi povezavami med Otovskim bregom in izviri ob reki Dobličici (Habič in sod. 1990).

Our observations are an important confirmation of the existence of a contact zone between black and white olms in the northern part of the Dobličica catchment. Based on previous knowledge of hydrogeological connections and eDNA, combined with our new observations, the contact zone probably extends at least between the two mentioned springs near the Dobličica and Otovski breg, covering approximately 0.7 km$^2$ (Fig. 1). Although extensive research has been carried out in the area following the discovery of the black olm in 1986 (Sket & Arntzen 1994), there is still not much data on the occurrence of olms in springs located within the presumed contact zone. Thus, future research may show the extent of the contact zone and overlap between the two lineages, and whether they are potentially able to hybridize.

**Figure 2.** The spring Planinec, where black and white olms were observed. The arrow marks the hole to which the olms retreated after we enlightened the spring. In the top right corner, the photograph of a white olm observed on 25 March 2022 in the spring is shown. Photo: Ester Premate.

**Slika 2.** Izvir Planinec, kjer smo opazili belega in črnega močerila. Ko smo izvir osvetlili, sta se v obeh primerih umaknila v podzemlje skozi luknjo, označeno s puščico. V zgornjem desnem kotu je fotografija belega močerila, opaženega 25. marca 2022. Foto: Ester Premate.

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# The first record of the Cumberland slider *Trachemys scripta troostii* (Thunberg & Schoepff, 1792) (Testudines: Emydidae) in Slovenia

## Prva najdba okrasne gizdavke *Trachemys scripta troostii* (Thunberg & Schoepff, 1792) (Testudines: Emydidae) v Sloveniji

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The pond slider (*Trachemys scripta*) is a freshwater turtle species native to the South-Eastern part of USA, but has also been introduced to many other countries around the world through pet trade (Escoriza et al. 2021b). Due to its colourful appearance, it has been the most numerous turtle species in pet shops for decades with millions of hatchlings reared every year on turtle farms. Their owners are eventually releasing many turtles to the nature, after overgrowing their confinements or simply becoming too difficult to keep (Cadi & Joly 2004). Pond sliders established populations in wetlands around the world (Espindola et al. 2019) and have a negative effect on indigenous turtles (Cadi & Joly 2004, Escoriza et al. 2021a). Today, pond sliders are banned from sale in many countries worldwide. In Slovenia, the sale of this turtle species was banned in 2017, after it was proven that it reproduced (Vamberger et al. 2012) and established vital populations in the country (Standfuss et al. 2016).

In their natural range, the pond slider has three accepted and one putative subspecies that differ in colouration of the neck and plastron and the shape and texture of the carapace (Praschag et al. 2017, Vamberger et al. 2020). The nominal subspecies, the yellow-eared slider (*T. s. scripta*) is distinguishable by a vertical yellow S-shaped stripe behind its eye, a yellow coloured plastron with only a few small black markings and a less elongated and more domed carapace (Praschag et al. 2017, Ernst & Lovich 2009). The commonest subspecies in pet trade was the red-eared slider (*T. s. elegans*), having a red horizontal stripe on its neck and a yellow plastron with large black spots on each scale (Praschag et al. 2017, Ernst & Lovich 2009). Cumberland slider (*T. s. troostii*) generally resembles *T. s. elegans*, but is distinguishable by its narrower horizontal neck stripe that starts with pale green or yellow colour and fades into dark red towards the end of the neck. Their plastron is yellow with medium-sized black spots on each scale (Carr 1937, Ernst & Jett 1969, Parham et al. 2020, Vamberger et al. 2020). The fourth subspecies, the Western slider, has only recently been proposed based on a DNA study of the *T. scripta* species (Vamberger et al. 2020). It was considered a distinct morph of the *T. s. elegans* and some authors considered it the result of hybridization with adjacent taxa. The Western slider has, in general, more elongated head than *T. s. elegans*, the red neck stripe is smaller and often split into two patches and the black spots on the plastron scales are the largest of all *T. scripta* subspecies. The status of these alleged subspecies remains to be resolved in further studies. Considering the species natural ranges, the Cumberland slider has the smallest natural range. It inhabits only the upper reaches of the Cumberland and Tennessee Rivers, its range overlapping with *T. s. scripta* and *T. s. elegans* only in the southernmost part of its range (Ernst & Lovich 2009, Vamberger et al. 2020).

Despite the recent ban on selling pond sliders in Slovenia, this species has already spread through most of the country and established several large reproducing populations (Standfuss et al. 2016). One of such population inhabits the waterways and wetlands in the Vipava Valley in Western Slovenia. One of the goals of the VIPava project (a nature conservation project, financed by EU Cohesion funds, link: https://www.projektvipava.si/), led by The Fisheries Research Institute of Slovenia, is to improve the habitat conditions for the highly protected European pond turtle (*Emys orbicularis*), the species included in the Appendix 2 of the Habitats Directive (OJ EC 1992). This native freshwater turtle has small fragmented populations in the Vipava Valley, concentrated in and around small ponds, channels and oxbow lakes close to the Vipava River (Vamberger et al. 2015). Removing pond sliders from waterbodies close to the Vipava River is one of the actions to improve the habitat for the European pond turtle. Invasive turtles currently represent one of the major ecological threats to the indigenous European pond turtle by competing for sources of food and most importantly basking places (Cadi & Joly 2004). Since the pond sliders grow bigger and are much more active and aggressive, they can easily compete for the best basking places in the water bodies, at least under controlled conditions in the laboratory (Cadi & Joly 2004).

During our fieldwork carried out in 2017, we focused on catching and removing pond sliders from water bodies, known to be inhabited by the European pond turtles, or larger ponds, where significant numbers of reproducing pond sliders had been detected (Standfuss et al. 2016). The turtles were caught with fishing pods for catching crayfish and with basking traps. In 20. 9. 2019, we sampled the Vogršček water reservoir in the central Vipava Valley. Among a total of 16 turtles caught (15 *T. s. elegans*, 1 *T. s. elegans* × *T. s. scripta*), one individual of the captured turtles generally resembled the *T. s. troostii* subspecies (Fig. 1). The individual was an adult female with carapace length of 186 mm and a weight of 1001 grams. The neck stripe was thin and yellow to pale green in colour that faded into dark green, without any shades of red or orange. The carapace was light green with a pattern of lighter and darker green stripes and curves, and was not domed as in *T. s. scripta.* The surface of the carapace was rough. The plastron was yellow with medium sized black markings on each scale. The colouration of the front legs was typical of the *T. s. troostii* subspecies with broad yellow parallel stripes (Fig. 1). The age of the individual was estimated at 18–20 years by counting the growth rings on its plastron. It is uncertain whether it had been released from captivity or hatched in the wild, but judging by the size of the turtle, its general good condition and lack of any damage or scars it was most likely the former case.

Despite no DNA sample was collected, the typical morphological characters enabled reliable subspecies identification. It is unclear how the *T. s. troostii* subspecies came to Slovenia, since it is the least known and attractive of the four pond slider subspecies, and there is no record of it ever being sold in pet shops in the country. One possible explanation is that it was imported on a personal request or even came to Slovenia from the neighbouring Italy.

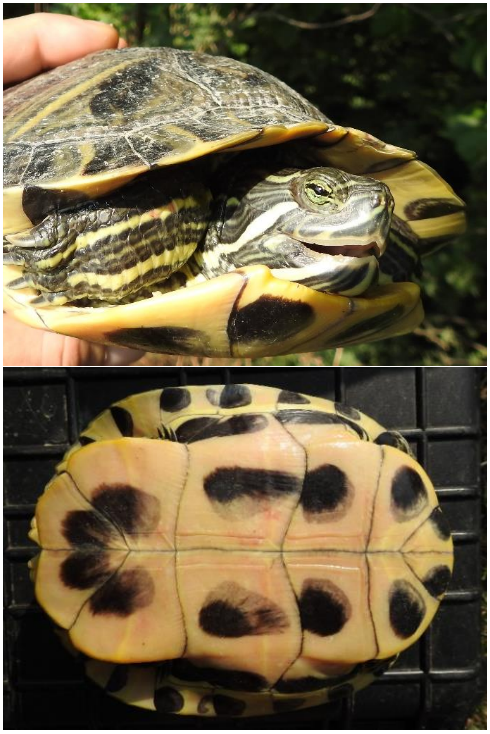

**Figure 1.** Photos of head (top) and plastron (below) of a *T. s. troostii* individual, caught in Vogršček in September 2019. Notice the thin yellow to pale-green neck stripe and medium sized black spots on plastron (Photo: ZZRS).
**Slika 1.** Fotografiji glave (zgoraj) in plastrona (spodaj) osebka *T. s. troostii*, ujetega septembra 2019 v Vogrščku. Opazne so tanka rumena do bledo zelena proga na vratu ter srednje velike črne lise na luskah plastrona (Foto: ZZRS).

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