

The current distribution and status of the Hermann's tortoise, *Testudo hermanni boettgeri* (Reptilia, Testudines, Testudinidae) in Croatia

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Abstract

Hermann's tortoise (*Testudo hermanni*) is listed as "Near threatened" in the IUCN Red list of endangered species. The importance of protecting the Hermann's tortoise populations and its habitats have led to the inclusion of the species within CITES Convention (Annex II), Annex A of EU Wildlife Trade Regulation, Annex II of the Bern Convention and Annexes II and IV of the EU Habitats Directive. To assess the distribution and status of the eastern Hermann's tortoise (*Testudo hermanni boettgeri*) in Croatia, historical and recent records were gathered and analyzed. The species was recorded in all three biogeographical regions in the country, but it's native to the Mediterranean and a small part of the Alpine region. With the increase of recent surveys and the use of citizen science platforms, the known range of the species in Croatia was increased by 35.8% and is now encompassing 123 10 × 10 km EEA reference grid cells. Most records (66%) originate from lower elevations (up to 199 m), and the highest was recorded at 570 m. Sparse forests are the most preferred habitats, followed by semi-open habitats, such as grasslands and shrubs. The most serious threat to the species is natural succession due to the increased abandonment of traditional farming and grazing. Other threats include touristic infrastructure and urban development, transportation, illegal collecting, and invasive species. The Area of Occupancy calculated using 2×2 km grids resulted in an AOO of 1,372.00 km², while Extent of Occurrence (EOO) is calculated to be 18,145.07 km². The current network of National protected areas includes 14% of the species' AOO while the designated Natura 2000 areas include 29.30% of its AOO. We propose to designate an additional 10 Natura 2000 areas to help with the long-term protection of the species.

Key Words

Area of Occupancy, Extent of Occurrence, habitat preferences, island populations, protected areas, range, threats

Introduction

Hermann's tortoise, *Testudo hermanni* is a western Palearctic tortoise species, distributed in southeastern Europe (Cheylan 2001). Two subspecies are recognized, *T. hermanni hermanni* Gmelin, 1789 distributed in the west, and *T. hermanni boettgeri* Mojsisovits, 1889 in the east Europe (Wermuth 1952; Cheylan 2001; Fritz et al. 2006). *T. hermanni boettgeri* has a continuous distribution in the Balkan Peninsula (Cheylan 2001; Fritz and Havaš 2007) including Croatia, Bosnia and Herzegovina, Montenegro, Serbia, Kosovo, North Macedonia, Albania, Romania, Bulgaria, Greece (Bour 1997) and Turkish Thrace (Türkozan et al. 2019). The main range of the species is in areas with a Mediterranean climate and humid continental climate. It inhabits a variety of habitats, from meadows, dry rocky pastures, macchia, forest edges, forest, thickets, and Mediterranean shrubs to rural and agricultural landscapes (Rozylowicz and Dobre 2010; Del Vecchio et al. 2011; Stojadinović et al. 2017; Nikolić et al. 2020).

The species is listed as "Near threatened" according to the IUCN Red list of endangered species (van Dijk et al.

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2004), mostly because of ongoing habitat modification and destruction, the international pet trade, and over-collecting (Cheylan 1984; Lambert 1984; Stubbs and Swingland 1985; Guyot and Clobert 1997; Gibbons et al. 2000; Mazzotti 2004; Stanford et al. 2020). The species is included in CITES Appendix II and Annex A of EU Wildlife Trade Regulation 338/97 as early as 1975 and is strictly protected under the Bern Convention and the EU Habitats Directive (92/43 EEC, Annex II, IV) (Epstein 2014). In Croatia, it is strictly protected under the Law on Nature Protection (NN 80/13, 15/18, 14/19, 127/19) and its status in the country should be monitored and reported to the European Union according to the Article 17 of the Habitats Directive.

In Croatia, the species inhabits a narrow coastal strip along the Adriatic coast, from Istria peninsula in the north to Dubrovnik city in the south, including some of the Adriatic islands (Cheylan 2001; Ljubisavljević et al; 2014; Jelić et al. 2015). Some records from Croatia are of non-native individuals. This species has a long tradition of being collected for the pet trade and transported. Accordingly, some individuals have escaped or have been released on purpose in the continental part of Croatia as well as on some Adriatic islands (Ljubisavljević et al. 2014; Jelić et al. 2015). Occurrence data in Croatia was provided by many authors but is mostly incomplete and fragmentary: Germar 1817; Kolombatović 1882, 1886; Depoli 1898; Werner 1902, 1908; Rössler 1904; Radovanović 1951; Pavletić 1964; Bruno and Maugeri 1977; Bruno 1980; Mršić 1987; Veith 1991; Schmidtler 1999; Sehnal and Schuster 1999; Cheylan 2001; Rathbauer 2002; Fritz et al. 2006; Schweiger 2006; Tóth et al. 2006; Grbac 2009; Bertolero et al. 2011; Koren 2012; Lauš 2012; Žagar et al. 2013; Ljubisavljević et al. 2014). Although the broad distribution of the species is known (Lončar 2005; Ljubisavljević et al. 2014; Jelić et al. 2015; Zadravec and Gambiroža 2019), the detailed distribution taking into considering extensive data from multiple unpublished sources has not been published so far. Moreover, a general lack of knowledge of population size and density, demographic structure, habitat preferences, species protection, and main threats make it difficult to carry out better science-based conservation management in Croatia.

The aim of this paper is to accumulate data to aid in the reevaluation of the conservation status based on IUCN criteria, including (1) range at a small scale (10×10 km), (2) Area of occupancy and Extent of occurrence, (3) habitat preferences, (4) coverage through the existing Natura 2000 network and nationally designated protected areas (5) national threat status.

Materials and methods

Study area

Croatia is located at the western part of the Balkan Peninsula and covers an area of 56.594 km², including a number of islands. There are three biogeographical regions in the territory of Croatia: Continental, Alpine, and Mediterranean (European Environment Agency 2017). The Mediterranean region covers coastal areas and islands in the Adriatic Sea (Ostrogović Sever et al. 2021) and can be divided into three areas: Istria peninsula, Kvarner, and Dalmatia (Cvitanović 1976).

Distribution analysis

To assess the occurrence of *T. h. boettgeri* in Croatia, records until 2021 were gathered from literature, open-access databases: Biologer.hr (Živanović et al. 2018), GBIF. org, Observation.org, Balcanica.info (Balej and Jablonski 2006–2021), authors' and colleagues unpublished data. For the usage of open access databases, we followed the terms and conditions of the websites and, where necessary, asked the authors for permission. The occurrence data were preprocessed before any further analyses. In the first step, we removed literature records containing wider localities, e.g., "Dalmatia" or "Brač island" and duplicate observations.

Records were classified into two groups: native populations and introduced individuals. Areas within the main distribution range as well as islands and parts of the marginal distribution range (e.g., north-western edge of distribution), containing at least three valid literature and/or recent records were considered as belonging to the native populations. Sporadic reports of animals found in or near urban areas or gardens on the Adriatic islands or other localities outside the main distribution range of the species were considered to be introduced. In addition, every data was assigned to be from the "mainland" or an "island".

All the processed data were then used to create a distribution map of the species in Croatia on the 10×10 km EEA reference grid squares in ArcMap (v. 10.7.1, ESRI) according to the standard of the European Environment Agency (DG Environment 2017), denoting introduced and native observations. We used 10×10 km squares labeled in order to be consistent with previous publications. To compare the previous and our research, we count the number of 10×10 km EEA reference grid squares. Within the distributional data, special attention was focused on island populations, to tentatively define which island populations are native and which are introduced.

The records of native populations were further divided according to coordinate accuracy into three groups: high accuracy (exact locality), medium accuracy (within 200 m), and low accuracy (more than 200 m). Only the records with high and medium accuracy were used for the analyses of the altitude, habitat selection, coverage of the protected areas, the major threats, and estimation of the national threatened status. The elevation data were obtained by using a Digital elevation model in ArcMap (v. 10.7.1, ESRI).

Spearman's rank correlation was used to test if there is a significant correlation between the number of re-

cords of *T. h. boettgeri* and altitude using cor.test function from base R (R Core Team 2020). The visualization was made with ggscatter function from ggpubr package (Kassambara 2020).

The extent of Occurrence and Area of Occupancy according to the Red List Criteria

To estimate the spatial distribution of the species, the specie's geographical range is presented in two ways, (i) Extent of Occurrence (EOO) and (ii) Area of Occupancy (AOO) according to the IUCN Red List Categories and Criteria assessments. EOO represents the area contained within the shortest continuous imaginary boundary of the present occurrence of a taxon (IUCN 2012). As eastern Hermann's tortoise is terrestrial species, we additionally removed the marine area and estimated the 'true' EOO based just on the land territory. EOO was measured by a minimum convex polygon in which no internal angle exceeds 180 degrees and which encompasses all the current known localities (IUCN 2012). Once the localities were downloaded into the geographic information system (GIS) domain, EOO was determined by joining the distant points in all directions and then calculating the area of the resulting polygon. AOO represents the area of suitable habitat currently occupied by the taxon within its Extent of Occurrence. We estimated AOO using the 2×2 km grid cells, as recommended by the IUCN (2012). EOO and AOO where calculated only for the presumably native populations.

Habitat preferences assessment

This assessment was based on the high and medium accuracy data without segregation based on the seasonal habitat use, or sex/age-specific habitat requirements. Using spatial selection, we have combined points of records and maps of natural and seminatural non-forest and freshwater habitats of Croatia (Bardi et al. 2016) in the ArcGIS program (v. 10.7.1 ESRI), and for each record extracted one of the possible habitat types following the National Habitat Classification (NHC): (A) inland surface waters and wetland habitats, (B) inland non and poorly covered land surface, (C) grassland habitats, (D) shrub, (E) forest habitats, (I) cultivated non-forested land and habitats with weeds and ruderal vegetation, (J) constructed and industrial habitats. Accordingly, we calculated the percentage of records falling within each habitat type.

Threats on a national scale

For each assessed threat, its impact on the conservation status of the species at the national level was ranked by coding the threat as high, medium, or low according to IUCN 3.2. Criteria (IUCN 2012). The threat impact is

generally regarded as high if it directly affects the longterm viability of the species and/or its habitat causing a significant decline at the biogeographic scale. Medium impact mainly includes threats acting only regionally causing significant declines. Low impact least affects the conservation status of the species but still potentially causes population decline.

Three independent threats are known to influence the survival of tortoise populations across their range (Stanford et al. 2020):

1. Habitat loss and fragmentation.

This threat is based on three subcategories: i) Agricultural activity – which was assessed using data from the Croatian Bureau of Statistics and Environmental Protection Agency; ii) Tourist infrastructure and urban development – conclusions have been mainly reached from literature and by observations during the period 1997– 2021; iii) Transportation – conclusions have been mainly reached by authors unsystematic observation of dead animals in a car traveling at low speed (40 km/h) during the research in the period 1997–2021.

2. Hunting and collecting of the species.

All the available CITES data sets were used (http:// trade.cites.org/en/cites_trade/) from which all the declared records of legal import/export available for T. hermanni between 1975 and 2021 were extracted. In our study, the records of legally exported individuals are incomplete because many of the individuals were traded internationally before their inclusion into CITES. Also, there are no records of tortoises in the CITES database from the period 1990-1995 for Croatia, probably due to the Croatian War of Independence (1991-1995). For illegal actions, e.g. sales and export attempts, State Inspectorate, Sector for Environmental Protection Supervision, Nature Protection, and Water Law Supervision was contacted but no data set was available. Accordingly, the media articles and stories that contained reports of the seizures of the species from 2000 to 2021 were reviewed. The resulting dataset was analyzed by the number of traded individuals regulated and not regulated under CITES. The legal exports were additionally categorized by country and purpose of trade (P-personal; T-commercial; E-educational) per year.

3. Invasive species.

This threat was derived from literature sources and limited observations by the authors.

Protected areas important for the species conservation

To assess the coverage and importance of existing protected areas for the long-term survival of the target species, two types of protected areas were used: nationally designated protected areas (NPA's) and Natura 2000 network. NPA's included protected areas designated under national legislation for Nature Protection of the Republic of Croatia and comprised six categories: National Park, Nature Park, Significant landscape, Special reserve, Horticultural monument, and Park Forest. The Natura 2000 network includes sites designated under the Habitats Directive (Sites of Community Importance and Special Areas of Conservation-SCI).

To calculate the total surface of areas under current legal protection where the species is present, we initially wanted to use the national habitat map of the Republic of Croatia to determine the surface area of suitable habitats occupied by the species within the protected areas. However, it is not accurate enough for this type of analysis, since many habitats where the species was found are incorrectly classified (e.g., grasslands are classified as forests) due to low precision of habitat polygons borders and the results were not satisfactory. Accordingly, we used AOO, which is based on actual records and therefore shows much more accurately where the species is present. Hence, for this analysis, 2×2 km squares calculated for the Area of Occupancy analysis were used and overlaid with both types of protected areas (available from BIOPORTAL, https://www.bioportal.hr/gis/). Only the protected areas that overlap with AOO were used for consequent analysis. From the overlap, the surface (km²) was calculated for AOO and NPA's, as well as for AOO

and Natura 2000. In the end, a joined analysis was performed for AOO and the joined protected areas with the exclusion of the overlapping areas between NPA's and the Natura 2000 network.

In Croatia, *T. h. boettgeri* is a conservation priority species in 15 Natura 2000 sites (Anonymous 2019). To assess if the designated Natura 2000 areas comply with the minimum required 20–60% coverage of the species range in the country the additional analysis was performed for these 15 Natura 2000 sites by overlapping with AOO (Anonymous 2014).

Results

Distribution

In total, 980 individual records of *T. h. boettgeri* during the period 1853–2021 were found. Concerning the origin of the records, 269 are literature data and 711 are new and previously unpublished records of the species in the country (Suppl. material 1). Most records, 926 (94.3%) were attributed to native population. Within this survey, the species is for the first time reported from the Alpine biogeographical region with six records.



Figure 1. The current distribution of *T. h. boettgeri* in Croatia. Solid red circles without dots represent new, unpublished data ascribed to the native populations, whereas red circles with a dot represent unpublished data ascribed to anthropogenic introductions. Green crosses represent literature data ascribed to the native populations, whereas white crosses represent literature data ascribed to anthropogenic introductions.

Out of 926 records relating to the native populations, 820 of them had high or medium coordinate accuracy, while 106 were imprecisely georeferenced records that were excluded from additional analyses. Out of the records of the native populations with high and medium accuracy, most belong to Dalmatia, 727 records (88.7%), followed by Kvarner, 57 records (7%), and Istria, 36 records (4.3%).

In total, the native populations of the species were recorded within $123 \ 10 \times 10$ km EEA reference grid squares (Fig. 3), of which it was for the first time recorded in 44 of them, which is an increase of 35.8%.

According to the records, the species is present in all three biogeographical regions in Croatia: the Continental, Mediterranean and Alpine (Fig. 1) but native populations are present only in the latter two. Within the Mediterranean biogeographical region, *T. h. boettgeri* is present both on the mainland, from the Istria peninsula in the north, along the Adriatic cost to Dubrovnik city in the south, and at 27 islands, eight of which are inhabited by presumably native populations: Krk, Cres, Pag, Vir, Zlarin, Hvar, Korčula, and Mljet (Suppl. material 2). The presence of the species on islands was ascertained with 28–51 records for the islands Krk, Cres, Pag, and Mljet, and with 5–9 records for islands Vir, Hvar, and Korčula. For island Zlarin, three valid records are confirmed in 2019 (Suppl. material 2). In the Alpine

region, species presence was confirmed through six records in the borderline area with Mediterranean region, in the vicinity of Knin. The northernmost and southernmost localities where the species is recorded in Croatia were, respectively, Buje (Istria peninsula), and Vitaljina (Konavle, Dalmatia).

The introduced individuals were recorded in Continental and Mediterranean biogeographical regions. Altogether, 14 records from eight localities are currently known from the Continental region: Zagreb with surroundings, Samobor (this study), Velika Gorica (this study), Turopolje (this study), Kalnik, Bjelovar (this study), Grbavac (this study) and Majur. In the Mediterranean biogeographical region, the introduced individuals are present on 19 islands with 41 records (Suppl. material 2). Three islands belong to the Kvarner area, while the rest are situated in the Dalmatian area.

In terms of altitudinal range, most records (66%) originate from lower elevations (between 0 m and 99 m), and 20% of records originated from 100–199 m. Above 200 m there were only 115 (14%) records. The locations with the highest altitude where the species was recorded are Vrataruša, Mt. Velebit: 570 m in Kvarner area and Polača, Mt. Dinara: 517 m in the Dalmatia area. Strong, significant correlation was found between the number of records of *T. h. boettgeri* and altitude (Spearman correlation test, p < 0.05, R = -0.67) (Fig. 2).



Figure 2. Relationship between the number of records of the species and the altitude (m).

The extent of Occurrence and Area of Occupancy according to the Red List Criteria

To get more precise information, the marine area was excluded and the corrected EOO was calculated to 18,145.07 km² (Fig. 3). Still, this is a significant overestimate as it includes unsuitable areas such as highest mountains (e.g., Učka, Velebit, Dinara, and Biokovo).

The Area of Occupancy calculated using 2×2 km grids resulted in an AOO of 1,372.00 km².

Habitat preferences assessment

Our analysis showed that the species has the highest affiliation with forests (33%), followed by cultivated non-forested land and habitats with weeds and ruderal vegetation (24.5%), and grassland habitats (20.9%). The remaining habitats included shrubs (14.7%), constructed and industrial habitats (4.7%), inland surface waters and wetland habitats (1.6%), and inland non and poorly covered land surface (0.3%).

Threats on a national scale

All three main threats: (1) habitat loss and fragmentation, (2) hunting and collecting of the species, and (3) invasive species were identified in Croatia.

Within the threat of habitat loss and fragmentation, tourist infrastructure and urban development, as well as transportation were assessed to have a medium impact while agricultural activity does not affect or minimally affects the species. Tourist infrastructure and urban development have a medium impact mostly due to the increasing development in the coastal areas, predominantly because of high touristic pressure and the increase in private accommodation and accompanying urban development. Concerning transportation, the whole species range is highly fragmented due to large numbers of local and regional roads, including highways, which impacts the population of the species. During the period 1997–2021 we observed more than 50 dead on road individuals highlighting this threat.

Hunting and collecting was assessed to have a medium impact and is reflected in the number of exported (legal or trafficked) individuals. Over the period 1997–2021, a total number of 3.291 individuals were exported from Croatia. Of those, 1.644 records were legally exported under the CITES regulation to eight countries, with the highest numbers being exported to Slovenia, Austria, and the Czech Republic for commercial and personal purposes of translocation (Suppl. material 3). The remaining 1.647 individuals were illegally trafficked from or through Croatia in the period 2000–2021. In respect to the timeframe, during the period 1997–2006, there were 33 reported legal exports and 22 illegally trafficked individuals, while in the period 2007–2016 the exports increased to amount of 3.083 individuals, of which 1.610



Figure 3. Geographic ranges of the species in Croatia using Extent of Occurrence (EOO), Area of Occupancy (AOO), and 10×10 km EEA reference grid squares.

were legally exported and 1.473 illegally trafficked. After 2017, there was a decrease in the number of exports from Croatia, counting 153 individuals respectively, of which only one was legally exported (Suppl. material 3).

The threat of invasive species affects the species with medium impact. Two main invasive species were determined as the most significant threats, wild boar, *Sus scrofa* Linnaeus, 1758, and the small Indian mongoose, *Herpestes javanicus* (É. Geoffroy Saint-Hilaire, 1818). In the area of southern Dalmatia, at least five turtle nests were observed to be destroyed by the mentioned species. Also, at least ten observed adult tortoises across the range were recorded to bear marks that could be attributed to wild boars.

Protected areas

Within Croatia, the eastern Hermann's tortoise occurs in 24 NPA's and 37 Natura 2000 areas (of which it is a target species in 15 of them). The total surface area of the species' AOO is 1,372.00 km², of which 191.48 km² (14%) is included in those 24 NPA's, while the coverage of the Natura 2000 network is 517.71 km² (37.73%). The 15 areas designated for the species cover only 401.92 km² (29.30%) of the species AOO. In addition, for two out of the 15 Natura 2000 sites designated for the species (both sites are on island Pag: HR4000018 and HR4000019), there is neither literature nor recent records of the species' presence. NPA's and Natura 2000 overlap on 22.9% of the territory. The coverage of both NPA's and the whole Natura 2000 network (excluding the overlaps between them) is 546.73 km² (40%) (Fig. 4).

Discussion

Distribution

The general distribution of *T. h. boettgeri* in Croatia is known for some time (Ljubisavljević et al. 2014; Jelić et al. 2015; Zadravec and Gambiroža 2019). Previous to our study, the species has been known from 79 10 × 10 km EEA reference grid squares (Jelić et al. 2015), while now it has increased to 123 10 × 10 km EEA reference grid squares. This increase of 35.8% is due to more intense herpetological research conducted in recent years and the development of citizen science.

The distribution of *T. h. boettgeri* in the eastern Mediterranean (including Croatia) is not homogeneous and is influenced both by the Mediterranean climate and zones of high mountains (Haxhiu 1998; Fritz et al. 2006). Our results confirmed that the natural range of the species in Croatia mostly stretches along the Adriatic coast, within the Mediterranean biogeographic region (Ljubisavljević et al. 2014). Regarding the distribution limits, the eastern boundary mostly follows the boundary between the Mediterranean and Alpine biogeographical regions and is determined by orographic parameters, e.g., Ćićarija and Učka mountain range in the north and the Dinaric Alps



Figure 4. The original Natura 2000 sites and NPA's and Natura 2000 network priority for species.

in Kvarner and Dalmatia (Fig. 1). The inland extension of the species in Croatia, including the records from the Alpine region, are also conditioned by the Mediterranean climate. The same is true for inland regions in Bosnia and Hercegovina, Serbia, Kosovo, Montenegro, North Macedonia, Romania and Bulgaria (Ljubisavljević et al. 2014).

The records from Croatia mostly originate from an altitude lower than 500 m, the same as in Italy (Romano et al. 2013) and Slovenia (Krofel et al. 2009). In contrast, the species reaches higher altitudes, up to 1500 m, in southern European countries, due to the greater influence of the sub-Mediterranean climate on the southern mountain chains (Cheylan 2001; Mazzotti 2006; Vetter 2006; Romano et al. 2013; Couturier et al. 2014; Ljubisavljević et al. 2014; Celse et al. 2018; Duro et al. 2021). All the existing records on higher elevations are in the central and southern mountain slopes of Kvarner and Dalmatia, which are also influenced by the Mediterranean climate.

The distribution of the species in the northern part of the Adriatic coast, especially in the Istria peninsula, is still not well understood (Ljubisavljević et al. 2014; Žagar et al. 2019). Some authors refer that those populations were introduced from southern parts of the Adriatic coast by Catholic monks and gentry who breed them for consummation (Sajović 1913; Tome 1996; Krofel et al. 2009; Ljubisavljević et al. 2014) or were merely released or escaped pets (Žagar et al. 2019). The records from northern parts of Croatian Istria mostly refer to single specimen observations and can be attributed to the runaway individuals, as no historical records from this region exist. The oldest finding for the area north of Limska Draga originated from 2004 (this survey). However, records for the southern part of Croatian Istria, including the area from Limska Draga to the south, exist for the whole century (Krumbach 1918). Certainly, further genetic research will reveal the origin of Istrian populations, as also suggested for Slovenian Istria (Žagar et al. 2019). Therefore, as the Istrian population is noted as native (Jelić et al. 2015), the authors accepted it as native until proven otherwise.

Records from Dalmatia in the period from 2000 until today have greatly increased, as opposed to the Kvarner area, where most of the recent records are related to islands. Only a few recent data, mostly sporadic, are available for the mainland part of Kvarner (this study). Most likely this is the result of fewer research activities in the mainland Kvarner area, and the general trend of Mediterranean species being present only on the narrow coastline (Jelić et al. 2015).

The individuals recorded in the Continental region and individuals from 19 Adriatic islands in the Mediterranean region are considered to be introduced. In the Continental region individual specimens were mostly found in, or around larger cities, especially the capital of Zagreb, the largest city in Croatia (Bašić 2003). These individuals are most probably pets intentionally or accidentally released into nature, as almost all individuals were found in gardens or parks (Jelić et al. 2015). In Italy, reproductive populations were reported from a few urban parks (Bologna et al. 2003; Rugiero 2004; Rugiero and Luiselli 2006) but in the continental part of Croatia their reproduction has never been observed. The probable low survival of introduced individuals in the continental part of Croatia can be linked to climatic conditions which are not optimal for successful reproduction and winter hibernation. Importantly, the optimal temperature range for successful hibernation in *T. hermanni* is between 4–7 °C (McCormack 2016) and for successful breeding is between 23–34 °C (Eendebak 2001), while the mean annual temperature of the coldest months in the continental part of Croatia is between -5 °C and 5 °C and the mean annual temperatures during the nesting activities peaks are around 17 °C (Zaninović et al. 2008).

The historical presence of the species on the Adriatic islands was reported by many authors (Suppl. material 2), while the review of species distribution on Adriatic islands was never conducted in detail. Jelić et al. (2015) mentioned seven islands with native populations (Krk, Cres, Pag, Korčula, Mljet, Lastovo, and Zlarin), while other authors cite nine islands, of which some were considered introduced (Lončar 2005; Ljubisavljević et al. 2014). Within our study we found that T. h. boettgeri is present on 27 Adriatic islands. The native populations are present on eight islands according to the criterium of at least three valid literature and/or recent records (Suppl. material 2). In comparison with Jelić et al. (2015), we determined two more islands with native populations: Vir and Hvar due to the increased number of individuals observed in natural habitats. At the same time, we excluded Lastovo from the list because almost all individuals were found in gardens or olive groves where they were probably kept as pets.

On the other 19 islands, the populations are considered introduced. The high number of islands with introduced individuals is associated with the tendency of local inhabitants to collect and transport individuals of the species. Their survival on the islands is most probably related to typical Mediterranean warm climate which is predominant on the islands (Meliadou and Troumbis 1997; Kryštufek and Kletečki 2007).

On Rab island the species was always recorded individually and most often in settlements (Tvrtković et al. 2012). During this survey systematical research of the herpetofauna of island Rab was conducted for more than 20 days, but no individuals were recorded (Jelić et al. 2016; Sučić et al. 2018; Štih et al. 2018). If the individuals on the island still exist, they are most probably introduced ones.

The records from island Plavnik date back to the 1980s (Bruno 1980, 1988), and the same record was cited by many authors (Cheylan 2001; Tóth et al. 2006, 2017; Tóth 2018). The herpetofauna of island Plavnik was repeatedly surveyed (Tóth et al. 2006, 2017; Tóth 2018) but the species was never recorded, and the authors mentioned that the species on the island is most probably introduced.

For the islands, Ugljan, Pašman, Čiovo, Šolta, Šćedro and Brač only single records exist, and they were accordingly categorized as introduced. Although intense herpetological research on a national level was carried out on the above mentioned islands, except Šćedro, no individuals were recorded (Žagar et al. 2013; Jelić et al. 2016).

The records from the rest of the islands: Lošinj (Tóth et al. 2009), Silba (this study), Olib (this study), Veli Iž (this study), Dugi Otok (Škvarč 2000; Schmidt et al. 2020), Žirje (Lauš 2010), Lastovo (Vervust et al. 2009; Zadravec and Gambiroža 2019), Žut (Schmidt et al. 2020) and Sestrunj (Schmidt et al. 2020), as well as Lokrum (this study) mostly originate from gardens or olive groves. With fewer than three records, and the lack of historical records, the individuals from all these islands are considered introduced.

Despite the widespread distribution of the species across islands in Croatia, the genetic structure of the island and mainland population has not been studied yet. As genetic studies are recognized to be an important method for detecting the origin of island colonization by the species (Vázquez-Domínguez et al. 2012), to completely resolve the origin of the island populations in Croatia, future studies based on mitochondrial and nuclear markers are needed as was the case on some other Mediterranean islands (Giacalone et al. 2009; Perez et al. 2014; Biello et al. 2021).

The extent of Occurrence and Area of Occupancy according to the Red List Criteria

The IUCN Red List status of *T. h. boettgeri* was assessed in 1996 and the species was considered as Near Threatened (NT) globally, as well as on the national level (Jelić et al. 2015). Our results indicated that the species has a smaller AOO in comparison with previous studies (Jelić et al. 2015), but still has a higher EOO, suggesting that the species is relatively widespread, but occurs in patchy locations within the range. To compensate for a relatively high EOO value, we calculated the "true" EOO. Nonetheless, the result clearly indicates that without excluding the marine areas from the analyses, the EOO would contain a very wide range of extensive unsuitable areas and thus not estimate that the species is under threat.

Habitat preferences assessment

The habitat preferences of the eastern populations of Hermann's tortoise have been studied in more detail only in Montenegro (Meek 1985), Serbia (Stojadinović et al. 2017; Golubović et a., 2019; Nikolić et al. 2020) and Romania (Rozylowicz and Dobre 2010; Rozylowicz and Popescu 2013). As for the Croatian populations, no previous studies on habitat preferences have been carried out, outside short-term general observations (Meek 1989). Our preliminary analysis indicated that the dominant habitat types used in Croatia are characterized by semiopen areas, such as sparse forests with herbaceous vegetation, cultivated non-forested land, shrubs, grasslands, and pastures. A similar pattern has already been observed in previous studies in Serbia (Stojadinović et al. 2017; Golubović et al. 2019; Nikolić et al. 2020). Sparse forest and shrubs provide protection from predators (Pătroescu and Rozylowicz 2007; Vilardell-Bartino et al. 2015), shade during the day (Bourn and Coe 1978) and places for egg laying (Vilardell-Bartino et al. 2015). On the other hand, more open areas, cultivated non-forested land, habitats with weeds and ruderal vegetation, and grasslands are most probably preferred for efficient thermoregulation (Zug et al. 2001; Anadón et al. 2006; Falcón and Hansen 2018) or feeding requirements (Anadón et al. 2006).

Our results are based mainly on the adult tortoise, so juveniles likely prefer different habitats with more cover to avoid predators (Gaymer 1968; Walton et al. 2019). Also, we are aware that the species could have different preferences toward a specific macrohabitat type depending on the season or sex/age-specific, as indicated in other studies, e.g., (Vilardell-Bartino et al. 2015; Stojadinović et al. 2017; Nikolić et al. 2020). Regardless, our results have provided useful insights into the habitat use of *T. h. boettgeri* in Croatia which is the first important step for the conservation of the species.

Threats on a national scale

The order Testudines is among the most threatened groups of vertebrates (Rhodin et al. 2018; Stanford et al. 2020). Still, the decline of the populations of reptiles, including the population of *T. hermanni*, can be difficult to detect and long-term studies of native populations and their association with environmental and anthropogenic factors are essential for understanding the population trends and fluctuations as well as for the development of appropriate protection measures (Todd et al. 2010). Without detailed long-term studies of the threats, the only available method is to deduce them from the available literature from other countries and personal observations. While this may be biased, it still represents a good starting point for all future threat assessments.

Habitat loss and fragmentation are mediated by numerous factors, such as agricultural expansion and intensification, urbanization, tourist infrastructure development and recreational activities, the pet trade and climate change. All these threats are causing a landscape change worldwide (Forman et al. 2003; Bürgi et al. 2004; Jaeger et al. 2007) affecting numerous wildlife species (Grift 1999; Underhill and Angold 2000; Marzluff 2001), as well as the eastern populations of *T. hermanni* (Willemsen and Hailey 1989; Hailey and Willemsen 2003; Pătroescu and Rozylowicz 2007; Türkozan et al. 2008; Rozylowicz and Dobre 2010; Ljubisavljević et al. 2011, 2014; Nikolić et al. 2018). Hence, landscape use intensification is globally one of the most significant land-use changes (Ellis et al. 2021) that directly contribute to habitat loss and modification, especially in western European countries (Couturier et al. 2014). However, in eastern European countries, an opposite trend can be observed, with the increased abandonment of traditional farming and grazing which contribute to habitat overgrowth and the decline of suitable habitats and populations (Sirami et al. 2008). The traditional agricultural land use and grazing in Croatia are in decline due to massive emigration from rural to urban areas (Obad 2021), with the consequences of the dramatic reduction of grassland areas and the spreading of shrubs and dense forests. Within the period 1980-2006 the surface of agricultural areas in Croatia was reduced by 8%, while the greatest decrease was observed in meadows and/or pastures, with 37% (Kušan 2010). The trend continued also in the period 2015-2019 (Cvjetićanin et al. 2020). This is rather concerning as 46% of the land territory of Croatia before 2010 was already covered by forests and shrubs, with the trend of further increase (Kušan 2010). The habitat overgrowing is evident across the whole range of the species in Croatia but is most noticeable in the Istria peninsula and the Adriatic islands, where parts of the land are still being cultivated, while most are under significant successional process (Lauš et al. 2019). Consequently, if these negative trends continue in the future, it will cause a serious decline of the species in Croatia.

Transportation corridors with associated infrastructure, such as roads, railroads, and utility and service transport affect the tortoise through direct mortality from vehicle collisions (Ashley and Robinson 1996; Bennett 2017), as well as creating a barrier to movement and dispersal between habitats, which can potentially affect population diversity (Latch et al. 2011). Also, road surfaces act as heat traps, which many species of reptiles use as basking sites (Rosen and Lowe 1994) and are one of the main causes of amphibians' and reptiles' decline (Carr and Fahrig 2001; Hels and Buchwald 2001; Smith and Dodd 2003; Carvalho and Mira 2011). In Croatia, many urban corridors were created or renewed during the period 1980-1990, but this is still an ongoing process in many urban areas where the human population is increasing, especially during the tourist season (Kušan 2010). Such high-density residential areas with a developed network of roads can increase the chance of an animal being run over by a car. Even with the unsystematic recording of road mortality, this seems to be high, especially in the Istria peninsula and on the Dalmatian coast during the late spring and early summer months, which may be associated with the higher traffic flow that occurs during summer holidays (Erritzoe et al. 2003). On the other hand, traffic speed is reported as an important factor determining the mortality of many vertebrate groups on the road (Bradford et al. 2005; Grilo et al. 2010), which may also play important role in Croatia, but this should be investigated in the future.

Tourism is one of the main industries in Croatia, especially on the coast, with the tendency to intensify in the future (Jelinčić and Žuvela 2012). Accordingly, construction of tourist infrastructure is causing significant habitat changes including habitat loss and fragmentation through the removal of natural vegetation. This threat is also correlated with the next larger threat, hunting, and the collecting of species which is largely happening during the touristic season.

The members of the family Testudinidae belong to the most traded species worldwide (Luiselli et al. 2016), which is worrisome since the life-history characteristics of tortoises do not provide buffering against exclusion of individuals from population (Nikolić et al. 2018). Such overexploitation of the species significantly influences the decline in the populations (Türkozan et al. 2008; Bertolero et al. 2011; Ljubisavljević et al. 2011; Luiselli et al. 2016; Graciá et al. 2020; CITES 2021). In the territory of former Yugoslavia (whose member was Croatia until 1991) significant exports of tortoises lasted over half a century (Honegger 1974; Lambert 1984; Vetter 2006; Ljubisavljević et al. 2011; Nikolić et al. 2018). While T. h. boettgeri is a long-lived species with a long generation time and low fecundity, it is evident that this species is particularly vulnerable to over-collecting, as is the case with other tortoises and large lizards (Warwick 2014; Nikolić et al. 2018). Aside from organized trafficking, many visitors to the country, as well as residents from different regions within same country, collected tortoises during holidays and took them back to their home as presents or garden animals. This resulted in many records of the species outside its natural distribution range (Fig. 1). The current annual trafficking of the species from Croatia is largely unknown due to the lack of available databases. In this context, the data for illegal traffic presented in this paper is based on available media reports, usually confiscated during border crossings, and in many cases it is not certain whether the individuals were collected in Croatia or were just transported through Croatia. Accordingly, there is a need for establishing up to date databases that contain such data and can be used in the future for assessing the trends in the trafficking of this and other native species. Still, the observed decline shows that the national policies were partially successful in regulating the exports of the species (Suppl. material 3). Focusing on all the facts, it is challenging to assess the impact of the overexploitation in Croatia as medium or high. Indeed, the legal exports from Croatia decreased in recent years, but the illegal traffic of tortoise is still on-going and hence, its impact can be significant. In general terms, it is evident that overexploitation is more prevalent globally (Luiselli et al. 2016) than in Croatia.

Wild boar and the small Indian mongoose are the species that represent an important threat for *T. hermanni*. The wild boar is native species in Croatia but has been introduced to many Adriatic islands (Šprem et al. 2011). The mongoose has been introduced intentionally on islands Mljet, Korčula, Hvar, Čiovo, Pelješac peninsula (Tvrtković and Krystufek 1990; Barun et al. 2011) and along the Adriatic coast in Croatia at the beginning of the twentieth century (Kryštufek and Tvrtković 1992; Ćirović et al. 2011; Boršić et al. 2018) to control the populations of venomous snakes, primarily the horned viper (Vipera ammodytes) (Tvrtković and Krystufek 1990). The species did not survive at all the localities where it was introduced and currently inhabits the mentioned islands as well as the mainland from the southern Konavle area up to the river Neretva in the north, with several more recent observations north of the river, indicating the further spreading of the species. Both species are known predators of eggs, hatchlings (Corti and Zuffi 2003; Vilardell et al. 2008), and adults in other Mediterranean countries (Ballasina 1995; Budó et al. 2003; Corti and Zuffi 2003; Bertolero et al. 2007). The individuals with bite marks from wild boar were recorded also in Croatia, on the island of Mljet (Jelić et al. 2012). Additional observations of the excavation of T. hermanni nests by both mongooses and wild boars were observed by the authors in several localities in southern Dalmatia (Majkovi and Dubrovnik area). The mongoose is a target species for future insular eradication attempts in Croatia (Barun et al. 2011), as their populations tend to increase throughout the country. Both mentioned species have in many areas outside their natural distribution range in Croatia established high-density populations (Cirović et al. 2011; Sprem et al. 2011) which can to some degree jeopardize the populations of T. hermanni and other native reptiles and amphibians protected on national and international levels (EU Habitats Directive, Annexes II and IV), especially in the Adriatic islands. In the future, targeted surveys should be carried out to determine the real impact of both species on the T. hermanni populations in Croatia.

Area important for species conservation

Although the network of protected areas is an important initiative for species and habitat conservation in Europe, some studies reveal that coverage of species and habitats by existing networks is insufficient to assure the long-term maintenance of biodiversity (Miu et al. 2020). The fact that some protected areas are or will become unsuitable for target species in the future, represents an additional concern in the preservation of species' suitable habitats (Araújo et al. 2011). In Croatia, 15 Natura 2000 sites were designated for the species, but as we stated before, for two of them (HR4000018 and HR4000019), the origin of the data on the basis of which they were designated is not clear (Jelić 2016). Although the dominant habitats present on these two Natura 2000 sites are suitable for the species (Eastern sub-Mediterranean dry grasslands and Eastern Mediterranean screes), further research is needed to confirm the species' presence.

In Croatia, the AOO area of *T. h. boettgeri* is more covered by the Natura 2000 network than NPA's, which agrees with similar studies on other European amphibians and reptiles (Sánchez-Fernández and Abellán 2015). The percentage of AOO covered by the Natura 2000 site designated for the species is 29.30%, which is more than the minimum of 20% considered in the European Union

as a guideline for nature conservation, but still at the lower limit of a sufficient degree of conservation of the habitat which is important for the species at national level (proposed range 20-60%) (Anonymous 2014). Thus, we identified 10 existing Natura 2000 sites to which T. h. boettgeri should be added as a target species. Site HR5000028 – Dinara is the only area where the species was found within the Alpine biogeographical region and should therefore be included as an area important for the species at the national level. The additional nine existing Natura 2000 sites which we identified are: HR2001021 - Lun on island Pag, HR4000005 - Privlaka Ninski zaljev-Ljubački zaljev, HR2000132 - Područje oko špilje Škarin Samograd and HR2001010 - Paleoombla-Ombla, HR2001325 - Ninski stanovi-livade, HR2001322 – Vela Traba, HR2000629 – Limski zaljev-kopno, HR2001371 - Područje oko Dobre vode, HR2000641 -Zrmanja, which in total amount to 73.51 km². With the inclusion of these areas, the coverage of the species-area by the Natura 2000 network would increase to at least 34.6%. The percentage could increase even more with additional surveys of the unexplored parts of existing Natura 2000 sites.

The existing NPA's and Natura 2000 network combined cover about 40% of the species AOO while the remaining 60% of the species area remains almost completely unprotected. *T. h. boettgeri* is a side-fidelity species with low dispersal abilities (Vilardell-Bartino et al. 2015; Türkozan et al. 2018). Consequently, if the existing protected areas do not host a sufficient percentage of the total population, it may cause a decline in the population size. Thus, conducting targeted research could lead to better protection of the species at the national level.

Conclusions

This study has provided valuable information about the current distribution, habitat preferences, and conservation status of T. h. boettgeri in Croatia. No decrease in the distribution of the species has been recorded, but rather an increase in the known range within the country. However, there are still unresolved questions, particularly regarding the origin of certain island populations, which can only be answered through the use of DNA methods. In Croatia, T. h. boettgeri faces significant threats and disturbances, such as habitat overgrowth, which could pose a risk to its survival in the future. Consequently, although based on the assessment of both the Area of Occupancy (AOO) and the "true" Extent of Occurrence (EOO), T. h. boettgeri may currently be classified as a Near Threatened (NT) species, an up-listing to the Vulnerable category (VU) could be expected in the near future. To ensure the long-term protection of this species, it is crucial to expand its coverage within the Natura 2000 network. The study suggests that 10 areas should be considered for inclusion during the reassessment of existing Natura 2000 sites. Additionally, targeted ecological surveys are needed across the species'

range in the country to better understand its ecological needs and provide a more scientifically-based approach to its protection in the future.

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Supplementary material 1

The new and previously unpublished records of *T. h. boettgeri* in Croatia

Authors: Katarina Koller Šarić, Boris Lauš, Ivona Burić, Ana Štih Koren, Toni Koren

Data type: Excel file (.xlsx)

- Explanation note: The new and previously unpublished records of *T. h. boettgeri* in Croatia according to open access databases data, the authors' unpublished field records and shared unpublished field data from colleagues. For the records of introduced individuals there is no altitudinal data available because they were not use in the analysis. Reference type (TR – Technical report; PO – personal observation; OP - Bio – online platform Biologer.hr; OP - O – online platform Observado.org; OP - G – online platform GBIF; OP - B – online platform Balcanica.info).
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- Link: https://doi.org/10.3897/herpetozoa.36.e103510.suppl1

Supplementary material 2

List of Adriatic islands with known records of *T. h. boettgeri*

Authors: Katarina Koller Šarić, Boris Lauš, Ivona Burić, Ana Štih Koren, Toni Koren

Data type: Word file (.docx)

- Explanation note: List of Adriatic islands with known records of *T. h. boettgeri*. For each island number of records and literature sources are provided. Number of records includes only the primary records from original citation, while the consecutive citations of the same records are mentioned in a literature source. The islands are arranged from the north to south.
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Supplementary material 3

The number of exported individuals regulated under CITES

Authors: Katarina Koller Šarić, Boris Lauš, Ivona Burić, Ana Štih Koren, Toni Koren

Data type: Word file (.docx)

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