

Non-native turtles in a peri-urban park in northern Milan (Lombardy, Italy): species diversity and population structure

CLAUDIO FOGLINI¹, ROBERTA SALVI^{2,*}

¹ Independent researcher. Via L. B. Alberti 8/A, I-20092 Cinisello Balsamo (MI), Italia

² Independent researcher. Via A. Moro 1, I-24030 Carvico (BG), Italia. *Corresponding author. E-mail: clafogli@libero.it
Both authors contributed equally to this work.

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Abstract. *Trachemys scripta* (slider turtle) and other freshwater turtle species have been traded worldwide as pets, but they are often released by owners in wild or in semi-natural habitats, especially in suburban and urbanized areas. In four artificial lakes and ponds of a Regional Park in the North outskirts of Milan (Italy, Lombardy), we trapped non native turtles using basking-traps and landing nets from April to August 2013. We captured 156 *Trachemys scripta*, 10 *Graptemys pseudogeographica* and four *Pseudemys* sp.; the capture-mark-recapture approach estimated an overall number of 224 individuals (95% C.I. 195-270). There was also a strong indirect support for a probable *in situ* reproduction, with small juveniles captured and a nest-digging female observed.

Key-words. Invasive alien species, morphometrics, non-native-turtles, reproduction, slider turtle, *Trachemys scripta*.

INTRODUCTION

The global introduction of non-native amphibian and reptile species has increased exponentially through the last 150 years (Çiçek and Ayaz, 2015). One animal group that is globally very popular in the pet trade is freshwater turtle, in particular subspecies and hybrids of the American slider turtle *Trachemys scripta*. Turtles belonging to this group are massively traded at the global scale (Ballasina, 1995; Kraus, 2009) and are usually sold when they are only few centimetres long. Due to their fast growing rate these turtles are then often released into the wild or in semi natural habitats (Crescente et al., 2014).

Trachemys scripta compete for food and basking places in wetlands where they coexist with European *Emys orbicularis* and *Mauremys leprosa* (Cadi and Joly, 2003; Perez-Santigosa et al., 2008; Ficetola et al., 2009), and massive turtle presence can influence ecosystem functioning and aquatic communities (Lindsay et al., 2013).

Free-living slider turtles' populations occur in many countries of southern Europe (Spain: Martínez-Silvestre et al., 2011; France: Cadi et al., 2004; Italy: Agosta and Parolini, 1999; Ferri and Soccini, 2003; Ficetola et al., 2003; Monti, 2010) and the ongoing climate change will probably expand the areas of suitability in the future (Ficetola et al., 2009).

In addition, following the European ban on import of *T. s. elegans* in the 1997 (European Commission, 1997), legal pet trade switched to other slider subspecies: mainly *T. s. scripta*, hybrids *T. s. scripta* x *T. s. elegans* and, but to lesser extent, *T. s. troostii* (Bringsøe, 2006). After a new European ban, extended to all *Trachemys* subspecies (European Commission, 2001 and subsequent amendments), different species of *Graptemys* and *Pseudemys* were also imported (Bringsøe, 2006).

In this study we investigated: (1) the number and size structure of turtles; (2) the presence of different species and subspecies of several non-native turtles; (3) the possibil-

ity of reproduction in the artificial lakes and ponds of the Nord Milano Park (Italy, Lombardy, Province of Milan).

MATERIALS AND METHODS

The study was carried out in the Nord Milano Park (Fig. 1), a 640 ha peri-urban Regional Park located in the North outskirts of Milan (Italy, Lombardy). The park was created on industrial brownfield during later sixties, and first reforestation dates back to 1983. During the years, woods, artificial wetlands, small lakes and ditches were added, while many others environmental improvement measures are still in progress. The presence of several non-native turtles in the park is well known, but has never been deeply investigated.

The counting sessions of this study were focused into four wetlands: three artificial lakes (Cinisello, Suzzani and Bresso Lakes) and one ditch-like site (Breda Ditch) (see Fig. 1 legend). To avoid problems with double-counts and with low detectability (i.e., water turbidity, lack of basking sites, aquatic macrophytes cover) turtles were captured with basking traps from April to August 2013: each trap was visited every two or three days between 11:00 AM and 03:00 PM, when the basking activity is greater (Cadi and Joly, 2000). During each visit additional turtles were captured with landing nets.

Parameters such as the straight line plastron length (SPL), the minimum straight carapace length (SCLmin) and the straight carapace width (SCW) were measured with callipers (Bjorndal and Bolten, 1989) by the same operator. Each individual was weighted and sexed using secondary sexual characteristics such as foreclaws and tail length (Readel, 2008). Specimens with SPL less than 150 mm (Gibbons, 1990) and with doubtful elements were considered unsexed. Each animal was also unambiguously marked with a unique notches combination on the marginal carapace scutes.

The release of invasive turtles after marking was an exceptional procedure, necessary to collect the basic knowledge for a pilot capture-recapture study; this was essential to develop and refine an effective control or eradication plan in the study area.

Given the fact that capture and recapture invariably occurred for each individual within the same site, a close populations scenario was assumed. This assumption is supported by the fact that trapping sites are not connected and are separated by wooded areas, lawns, footpaths and bikeways. For this reason, in order to estimate the overall number of turtles, the capture-mark-recapture (CMR) model for close populations (JHE Closed Population Model Estimation) provided by the software Noremark (White, 1996) was used.

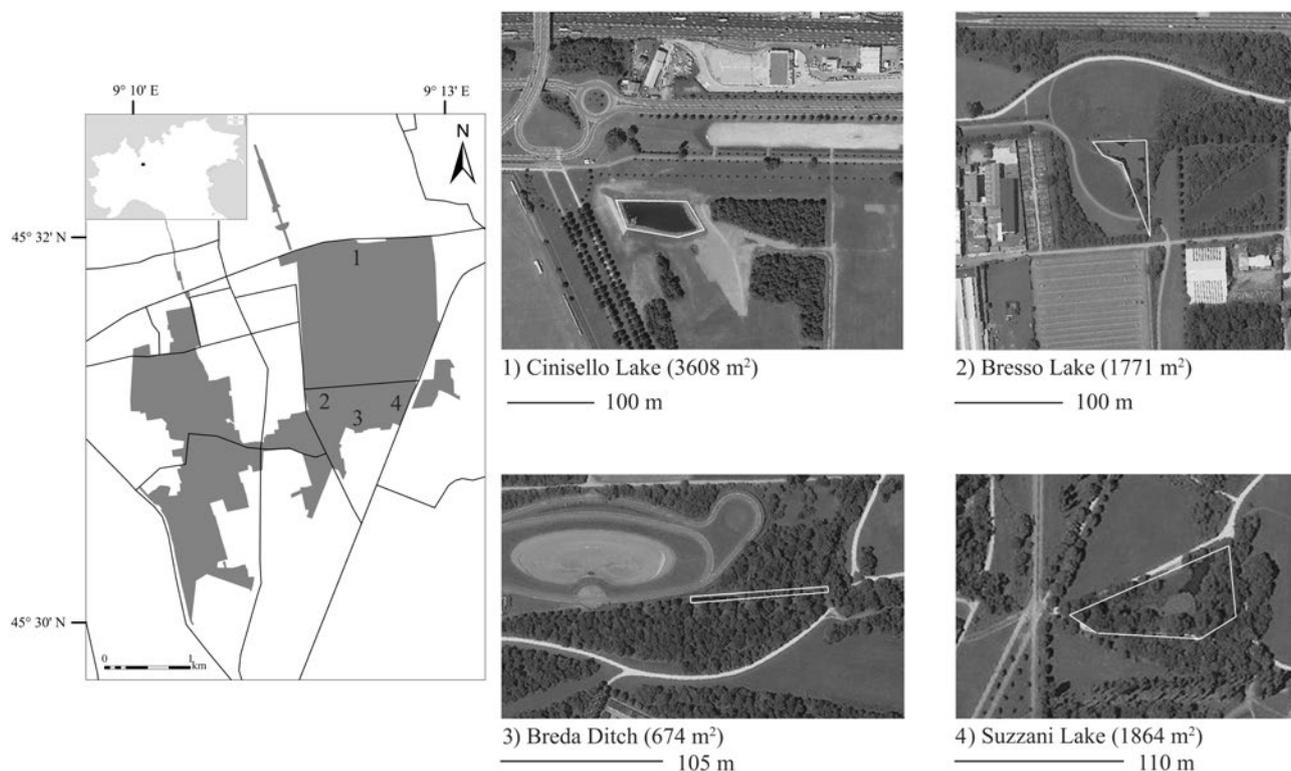


Fig. 1. Study area (black box, not scaled). The position and a detailed view of each wetland is given by numbers: 1) Cinisello Lake; 2) Bresso Lake; 3) Breda Ditch; 4) Suzzani Lake. Wetland boundaries are marked in white (modified from www.d-maps.com and GeoPortale Regione Lombardia).

Table 1. Number and sex of non-native turtles captured in each pond, grouped by taxa. M=male; F=female; n.d.=unsexed.

Wetland	<i>T. s. elegans</i>			<i>T. s. scripta</i>			<i>T.s. troostii</i>			<i>T. s. hybrids</i>			<i>Pseudemys</i> sp.			<i>G. pseudogeographica</i>		
	M	F	n.d.	M	F	n.d.												
Suzzani Lake	0	2	4	0	1	11	0	0	3	3	6	27	0	1	2	0	0	4
Bresso Lake	2	5	3	0	1	14	0	1	1	1	7	34	0	0	0	0	1	5
Breda Ditch	0	0	0	0	0	4	0	0	2	0	1	11	0	1	0	0	0	0
Cinisello Lake	0	0	0	0	0	4	1	0	0	0	0	8	0	0	0	0	0	0

Table 2. Morphometrics (mean \pm SD and range) of captured specimens, grouped by taxa. SPL = Straight line Plastron Length; SCL_{min} = minimum straight carapace length; weight. N may vary, as some data is missing.

	<i>T. s. elegans</i>	<i>T. s. scripta</i>	<i>T. s. troostii</i>	<i>T. s. hybrids</i>	<i>Pseudemys</i> sp.	<i>G. pseudogeographica</i>
SPL (mm) \pm SD	143.19 \pm 43.64	100.57 \pm 32.31	112.86 \pm 44.95	108.14 \pm 38.57	189.00 \pm 46.91	105.80 \pm 40.58
n	16	35	7	97	4	10
min. – max. (mm)	43 – 210	42 – 183	50 – 168	26 – 188	123 – 223	53 – 167
SCL _{min} (mm) \pm SD	156.31 \pm 45.99	111.97 \pm 35.18	120.71 \pm 50.45	116.67 \pm 40.49	206.25 \pm 47.67	119.60 \pm 44.81
n	16	35	7	97	4	10
min. – max. (mm)	47 – 221	46 – 200	53 – 180	31 – 201	140 – 242	61 – 187
weight (g) \pm SD	751.67 \pm 506.78	347.06 \pm 287.41	382.14 \pm 274.89	407.18 \pm 302.53	1437.50 \pm 807.65	360.00 \pm 281.66
n	15	34	7	94	4	10
min. – max. (g)	100 – 2000	50 – 1600	50 – 850	50 – 1500	400 – 2150	50 – 800

RESULTS

Species richness and abundance

We captured and individually marked, weighted and sexed 156 slider turtles: 16 red-eared slider *Trachemys scripta elegans*, 35 yellow-bellied slider *T. scripta scripta*, 7 Cumberland slider *T. scripta troostii* and 98 *T. scripta* hybrids. Specimens were classified as hybrids when they showed a mix of elements from parental species (Seidel et al., 1999; for details see supplementary Fig. S1).

We also captured, measured and marked 10 false map turtle *Graptemys pseudogeographica* (supplementary Fig. S2) and cooter *Pseudemys* sp. (supplementary Fig. S3). Details about *sex-ratio* and species repartition into each sampling site are shown in Table 1, while details about morphometrics are shown in Table 2.

Using the acquired data, the following overall population was estimated with Noremark software (95 % confidence interval in parentheses): 13 (13) turtles in the Cinisello Lake, 84 (73-102) in the Suzzani Lake, 106 (90-129) in the Bresso Lake and 21 (19-26) in the Breda Ditch (supplementary Table S4).

Trachemys scripta population structure

Due to the lack of data for the others turtles genera, only the population structure of *Trachemys scripta* sub-species was analysed.

Among sexed specimens, in three sampled wetlands we found a shift towards a female-biased sex-ratio. In the Suzzani and Bresso lakes (Fig. 2a and 2b, respectively), we found mainly medium-small turtles; full-grown adults are present in small number, and we found also some juveniles. Small turtles were prevalent also in the Breda Ditch, in which we found also only two juveniles and one adult female (Fig. 2c). In the Cinisello Lake, the small number of individuals does not allow a population description (Fig. 2d).

The presence of small specimens with SPL between 26 and 51 millimetres was noteworthy. Four were *T. scripta* hybrids, three of which trapped in the Bresso Lake and one in the Breda Ditch (SPL 26 to 51 mm, μ 36.25 \pm 10.56 SD). One *T. s. elegans* (SPL 43 mm) was captured in the Suzzani Lake, and one *T. s. scripta* (SPL 42 mm) was captured in the Bresso Lake (supplementary Fig. S5a and S5b). Three more *Trachemys* juveniles have SPL

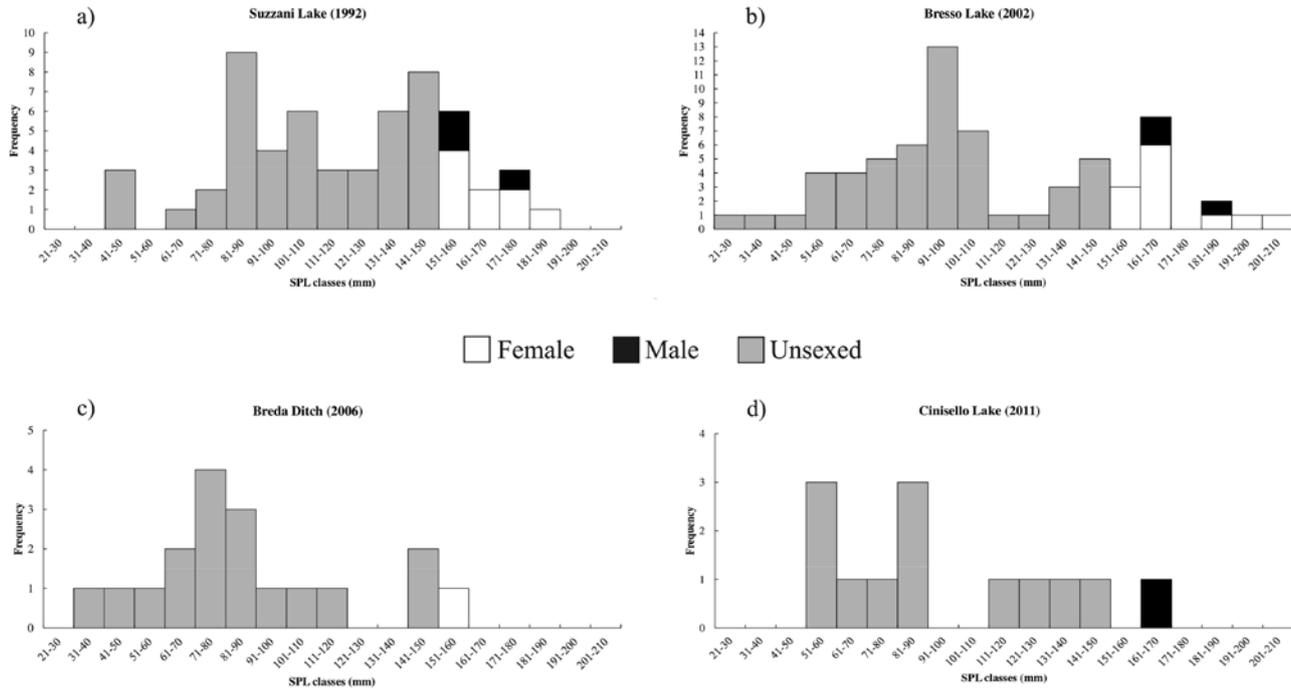


Fig. 2. Number of slider turtles, classified after their Straight Plastron Length, hosted in each wetland (completion year shown in brackets).

inside the same range, but their SCL exceed 60 mm.

DISCUSSION

Trachemys scripta has been included in the “Top 100 World’s Worst Invaders” as a result of their formerly massive import by the pet trade (Lowe et al., 2000). It is now listed among the Invasive Alien Species (IAS) of European Union concern (European Commission, 2016). *Pseudemys* and *Graptemys* have partially replaced sliders as pets-turtle, but due to their higher price they are not so common as feral individuals (Bringsøe, 2006).

Species composition and population status

Captured slider turtles were mainly *T. scripta* hybrids and *T. s. scripta*, followed by red-eared slider and just few *T. s. troostii*. Few individuals of other species (i.e., false map turtle and cooter) were also captured.

Among slider turtles, the female-biased sex ratio we found is common in non-native population, because slider turtles were reared with artificially high temperature: this accelerate their development (Prevot-Julliard et al., 2007), but increase female development in species with temperature-dependent sex determination (Godfrey et al., 2003). In suitable conditions, the unbalanced sex

ratio could increase the number of recruits of non-native population (Ficetola et al., 2009).

According to the number and the size of small turtles captured, Bresso Lake was the only wetland with apparently active recruiting: this could be due either to new release of relatively young individuals (uncommon with pet turtle) or to active reproduction (see below).

Reproduction

Several Mediterranean and southern-continental European areas currently have suitable climate conditions for slider turtles’ successful reproduction (Tzankov et al., 2015).

Concerning breeding turtles in the park, before this study there was only an anecdotic report of one hatchling with eggshell remains, ran over on the side of a bikeway (Mariani G., pers. com.). Unfortunately, because of the advanced decomposition, it was possible to identify the specimen only as belonging to the *Trachemys* genus.

The smallest slider turtles recorded (see results) can be considered clues of probably recent breeds, because their carapace lengths were similar to those reported for newly born juveniles in Ernst et al. (1994). Moreover, the juvenile *T. s. elegans* captured was almost certainly feral, because the trade of this specie in the EU was banned since 1997 (European Commission, 1997).

Although we did not record nests during this study, in July 2016 an adult female was observed during nest-digging (Gelso M.R., pers.com.; supplementary Fig. S6), and during the same month, in the Bresso Lake another juvenile of *T. scripta* was reported (Ghislandi A. and Ghislandi M., pers. com.; supplementary Fig. S7).

Conclusions

As the park is deeply engulfed in the urban matrix, no European pond turtle *Emys orbicularis* is present, but on the other end there is a small population of smooth newt (*Lissotriton vulgaris*), classified as near threatened (NT) in Italy (Rondinini et al., 2013).

For all these reasons, we find unusual that the park has not yet adopted an invasive turtle control plan, even more after the ratification of the European Regulation 1143/2014 and the European Implementing Regulation 1141/2016 (European Commission, 2014; 2016).

According to the evidences we found (i.e. reproductive population, number of potential breeding turtles), we hope that this work could represent a first step towards the adoption of a non-native turtles monitoring and control program in the area, even with eradication measures. Due to the high level of anthropization, and the easy accessibility of wetlands in the study area, we also urged the adoption of steady actions aimed at discouraging further releases by turtle owners.

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SUPPLEMENTARY MATERIAL

Supplementary material associated with this article can be found at <<http://www.unipv.it/webshi/appendix>> Manuscript number 20700.

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