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Population structure of *Emys orbicularis* in syntopy and allotopy with *Mauremys leprosa*

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Abstract: Since the last decade several sites have been sampled with baited traps or visual surveys for the detection of *E. orbicularis*. Most presence locations consisted of syntopic populations of *E. orbicularis* and *M. leprosa*. At the majority of these sites *E. orbicularis* captures or sightings consisted exclusively of a small number of individuals and mainly adults. At the few sites where allotopic populations were found, juveniles and subadults were commonly observed and captured. Restricting the analysis to sites where more complete surveys were carried out, there are significant differences on size structure and age class proportions between syntopic and allotopic populations. Although limited by the amount of samples, we wonder if this is a direct effect of *M. leprosa* on *E. orbicularis* through species interaction or else the effect of different habitat requirements. This is an important issue for the conservation of *E. orbicularis* in Portugal. In fact, it is essential to know the role of *M. leprosa* on the abundance and life history parameters of *E. orbicularis* populations for the implementation of adequate conservation measures.
Key words: age structure, *Emys orbicularis*, *Mauremys leprosa*, size structure.

Resumen: Estructura poblacional de *Emys orbicularis* en sintopía y alotopía con *Mauremys leprosa*. – En la última década varios lugares se han muestreado con trampas cebadas o mediante avistamientos para la detección de *Emys orbicularis*, resultando en la mayoría de los casos situaciones de sintopía con *Mauremys leprosa*. En la mayor parte de estos casos, las localizaciones de *Emys orbicularis* fueron escasas y referidas a individuos adultos. En los pocos lugares en que se encontraron las poblaciones en alotopía, se encontraron y capturaron juveniles y subadultos. El estudio manifiesta diferencias significativas en la estructura y clases de edad entre las poblaciones sintópicas y alotópicas. Aunque con la limitación de la cantidad de muestras, cabe preguntarse si esto es un efecto directo de interacciones con *Mauremys leprosa* o consecuencia de requerimientos ambientales diferentes. En todo caso, para la aplicación de medidas de conservación adecuadas en Portugal es esencial conocer la influencia de *Mauremys leprosa* en las poblaciones de *Emys orbicularis*.
Palabras clave: estructura poblacional, *Emys orbicularis, Mauremys leprosa*.

INTRODUCTION

Age structure is one of the most important demographic issues for turtle conservation (GIBS & AMATO, 2000) as a consequence of life history traits such as their high longevity, delayed sexual maturity and great change of survival rates from hatchling to the adult stages. Life table analysis coupled with sensitive analysis has been for that reason a very useful tool for turtles' demographic predictions under varying scenarios (CONGDON *et al.*, 1993, 1994; BLAMIRES *et al.*, 2005). Since turtle's survivorship and fecundity are more related to size than to age (KENNETT, 1996; SPENCER & THOMPSON, 2005), population models in which life cycle is described in terms of size classes or developmental stages (CASWELL, 2001) are probably more adequate than classical life table analysis to predict population trends. A characteristic of long-lived organisms is that population's response after a perturbation can be very gradual (HEPPELL *et al.*, 1996; GIBS & AMATO, 2000). However, shifts in age structure often occur in a much shorter time frame than abundance (CASWELL, 2001) and therefore they are a better indicator of short-term changes in survival rates and fecundity (HOLMES & YORK, 2003). In this study we assume that size structure can be used as an estimate for assessing and comparing populations' condition. Under this assumption we test whether there are differences between the status of syntopic and allotopic populations of a species.

In the Iberian Peninsula two freshwater turtle coexist: the European pond turtle, *Emys orbicularis* (L., 1758), and the stripe-necked turtle, *Mauremys leprosa* (Schweigger, 1812). The geographical range of *E. orbicularis* in the Iberian Peninsula is very fragmented and greatly overlaps with that of *M. leprosa*. Exceptions are Galicia and Navarra communities of northern Spain where *E. orbicularis* extends its range beyond that of *M. leprosa* (AYRES & CORDERO, 2001; KELLER & ANDREU, 2002). On the other hand, *E. orbicularis* is absent from vast regions of southern Spain where *M. leprosa* occurs, such as eastern Andaluzia and Murcia (DA SILVA, 2002). While *E. orbicularis* has a predominant European distribution, *M. leprosa* has an Iberian and North African distribution, which probably explains these 'exclusion' areas. At a finer spatial scale *E. orbicularis* is much less abundant than *M. leprosa* at sites where syntopic populations occur (ARAÚJO *et al.*, 1997; SEGURADO & ARAUJO, 2004). More recently, many aquatic habitats in Iberia were colonized by exotic freshwater turtles, namely the red-eared slider, *Trachemys scripta elegans* (PLEGUEZUELOS, 2002), which probably will have an important impact on the populations of native species (CADI & JOLY, 2003, 2004). Nevertheless, in Portugal no exotic turtles were found at most sampled sites until now, including those that in recent years have been sampled more exhaustively.

Although it has never been tested, there is some empirical evidence for the age structure of syntopic populations of *E. orbicularis* in Portugal being more skewed towards a predomination of adults than that of allotopic populations. If the presence of *M. leprosa* affects the age structure of *E. orbicularis* this could either mean that that some kind of interaction between species was taking place or simply that syntopic populations occur at suboptimal habitats for *E. orbicularis*. Patterns of species co-occurrence in Portugal have been analysed elsewhere (SEGURADO & ARAÚJO, 2004; SEGURADO & FIGUEIREDO, 2007). Overall, these studies suggest that (1) there is a wide overlap between species both on their geographical distributions and coarse scale habitat preferences and (2) differences in the use of space are mainly expressed at the microhabitat scale.

Testing if and how the presence of *M. leprosa* directly affects *E. orbicularis* populations' parameters (e.g. recruitment) is an important issue for the conservation of *E. orbicularis* in Portugal. If such effect were confirmed as an important factor on the distribution, abundance or other life history parameters of *E. orbicularis*, then ecological information on *M. leprosa* would also be required for an adequate conservation planning for this species.

In this study we first analyse the species' co-occurrence pattern in Portugal using up to date information on species presence. We then test for differences between syntopic and allotopic populations of *E. orbicularis* using several population descriptors, mainly those derived from size structure.

MATERIALS AND METHODS

Since the last decade several sites have been sampled for the detection of *E. orbicularis* and *M. leprosa* in Portugal (ARAÚJO *et al.*, 1997; SEGURADO, 2000; SEGURADO & ARAÚJO, 2004). Sampling was based on two techniques: 1) visual surveys consisting of approximately 200m transects along the shorelines of streams, ponds or reservoirs; 2) trapping sessions using baited hoop nets at a restricted number of sites. A total of 326 sites were sampled from 1996 until 2005. No exotic freshwater turtles were captured or observed at these sites.

Comparisons between populations' age class distribution were restricted to six syntopic populations and eight allotopic populations (Table 1), at sites where at least a three-day trapping session was undertaken somewhere between April and July and more than four individuals were caught. A sample size of four individuals is considered as the minimum number for body size characterization (BROWN *et al.*, 1999). In order to check whether if it is minimally acceptable to use samples of four individuals to estimate age proportions, the following resample procedure was performed: (1) one thousand subsamples with sizes from 2 to 15 individuals were taken from the largest sample available for a single population (N = 98), that included a proportion of juvenile turtles of 0.18; (2) median values of juvenile proportion were calculated for each sample size. Samples sizes of 2 and 3 individuals yielded median values equal to zero, the sample size of four individuals yielded a median value of 0.25 and sample sizes from five to 15 yielded median values that oscillated between 0.13 and 0.21. Therefore we assumed that a sample of four individuals was the number that could provide proportion estimates with an acceptable probability of being roughly correct. Even so, 42.1% of the four individuals' subsamples yielded no juveniles and thus there is always the danger of juvenile proportions being underestimated using such small samples.

Individuals were measured and permanently marked by marginal notching, according to a coding system for future recognition. Age classes were based on straight-line carapace length (CL) according to KELLER (1997) classification for the populations of Doñana National Park, Andalusia. Three age classes were considered: juveniles (CL < 115mm), subadults (CL = 115-120 for males and CL = 115-130 for females) and adults. Comparisons were made between syntopic and allotopic populations for the following

TABLE 1. Sampling sites and main habitats.

TABLA 1. Lugares de muestreo y hábitats principales.

Sampling site	Type	Habitat
Ludo	Syntopic	Stream / canals
Budens	Syntopic	Stream
Excomungado	Syntopic	Permanent pond
Cobres	Syntopic	Stream
Apariça	Syntopic	Lake (Dam)
Tornada	Syntopic	Marsh / canals
Longueira	Allotopic	Pond system
Malhão	Allotopic	Pond system
Abobeleira	Allotopic	Stream
Freitas	Allotopic	Permanent pond
Vidoeiro	Allotopic	Pond / stream
Calvão	Allotopic	Stream
Terva	Allotopic	Pond / stream
Prado	Allotopic	Pond system

population structure parameters: (1) total proportion of juveniles, (2) proportion of young-of-the-year juveniles, (3) proportion of adults, (4) sex ratio (5) mean carapace length, (6) kurtosis and (7) skewness of body size distribution. Skewness is a measure of lack of symmetry of a distribution while kurtosis is a measure of whether the data are peaked or flat relative to a normal distribution (SOKAL & ROHLF, 1995). Whether the total proportion of juveniles and proportion of young-of-the year differed or not between syntopic and allotropic populations was also tested. Whenever necessary, arcsine and logarithmic transformations (SOKAL & ROHLF, 1995) were previously applied to, respectively, proportion and non-proportion data (Table 1).

RESULTS

Locations with *E. orbicularis* (18 sites) represented approximately 15% of the sites containing freshwater turtles, among which about 58% consisted of syntopic populations, while 42% were allotopic populations of *E. orbicularis*.

The vast majority of sites where freshwater turtles occurred consisted of allotopic populations of *M. leprosa*. At the majority of sites where species co-occurred, captures or sightings of *E. orbicularis* consisted exclusively of a small number of individuals. The highest absolute frequencies of *E. orbicularis* were found in the allotopic populations. While the syntopic populations were occasionally found throughout the geographical range of *M. leprosa*, the few allotopic populations of *E. orbicularis* were essentially found at two distinct regions: at the far north of Portugal and in the coastal pond systems in Southwest coast (Fig. 1).

Size distributions suggest that syntopic populations tend to show skewed age structures towards adults and less size variability (Fig. 2). This tendency is confirmed by both the mean values of carapace length and the measures of skewness and kurtosis of size distribution for each kind of population

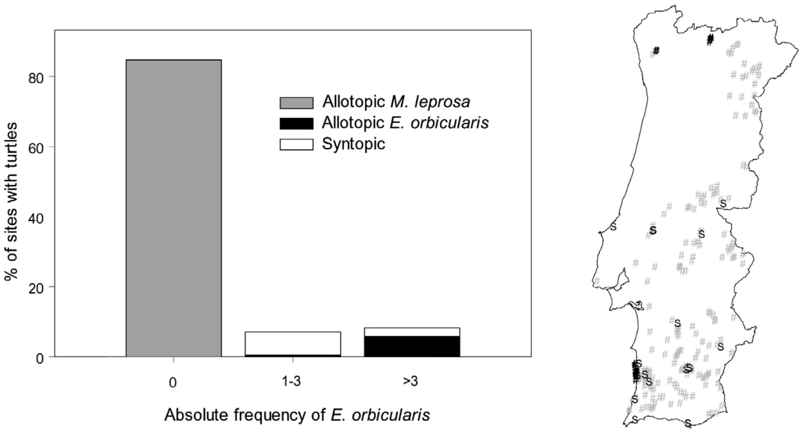

FIGURE 1. Relationship between the absolute frequency *of E. orbicularis* and the % of sites where turtles of either species were observed, and the correspondent geographic locations of each kind of population (allotopic *E. orbicularis*, allotopic *M. leprosa* and syntopic populations).

FIGURA 1. Relación entre la frecuencia absoluta de *E. orbicularis* y el porcentaje de enclaves en los que se observaron galápagos de cualquier especie con indicación de su situación geográfica (poblaciones alotópicas de *E. orbicularis*, alotópicas de *M. leprosa* y sintópicas).

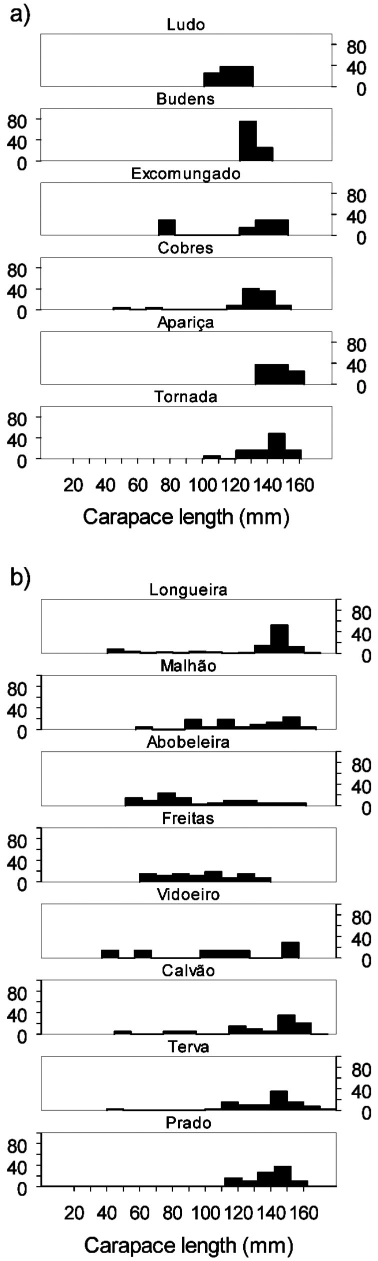

FIGURE 2. Size distributions of *E. orbicularis* at each sampling site that entered the analysis (the y-axis are the percentages of absolute frequencies), in syntopic (a) and allotopic (b) populations.

FIGURA 2. Distribuciones de tamaño de *E. orbicularis* en cada enclave incluido en el análisis (el eje y indica porcentajes de frecuencias absolutas), en poblaciones sintópicas (a) y alotópicas (b).

(Table 2). However, no significant differences were found for these values between syntopic and allotopic populations (ANOVA; mean carapace length, $F1,12 = 1.380$, $p = 0.263$; skewness, $F1,12 = 0.279$, $p = 0.607$; kurtosis, $F1,12 = 0.004$, $p = 0.949$).

Juveniles of *E. orbicularis* were captured at four sites with syntopic populations (Table 3) and at all sites with allotopic populations of *E. orbicularis*. Young-of-the-year individuals were found at a single syntopic site and at six allotopic sites. At three allotopic sites juveniles represented the most abundant age class, while among syntopic sites adults predominate at all but one site where subadults prevailed (Ludo).

According to the mean percentage of *E. orbicularis* captures (Fig. 3), the allotopic populations tend to possess a lower percentage of adults and a higher percentage of juveniles (51% and 37%, respectively) when compared to syntopic populations (80% and 10%, respectively). No marked differences were observed for subadults. However, for both the total proportion of populations with juveniles and the proportion of populations with young-of-the-year, the differences between populations' types were not significant (Fisher's exact test, two-tailed, $p > 0.1$).

A significant difference between syntopic and allotopic was confirmed for the proportion of young-of-the-year individuals (ANOVA, $F1,12 = 5.602$, $p = 0.036$), while for the total proportion of juveniles a near significant difference was found (ANOVA, $F1,12 = 4.103$, $p = 0.066$). For the proportion of adults no significant difference between the two kinds of population was found (ANOVA, $F1,12 = 2.406$, $p = 0.147$).

Although no significant differences were found between the sex ratios of syntopic and allotropic populations of *E. orbicularis* (ANOVA, $F1,12 = 2.391$, $p = 0.148$), the

TABLE 2. Sex ratio, mean and range of carapace length (CL, in mm), mean kurtosis and mean skewness of *E. orbicularis* in syntopic and allotopic populations (SD between brackets).

TABLA 2. Sex ratio, media y rango de la longitud del espaldar (en mm), curtosis y asimetría de las poblaciones de *Emys orbicularis* sintópicas y alotópicas (SD entre paréntesis).

	Sex ratio	Mean CL	Range CL	Kurtosis	Skewness
Syntopic	2.68 (2.61)	135.50 (13.98)	45 - 160.2	1.56 (3.27)	1.29 (0.88)
Allotopic	1.09 (0.46)	120.45 (40.27)	37 - 175	1.11 (2.94)	1.01 (0.72)

TABLE 3. Absolute frequency of sex-age-classes of *Emys orbicularis* at each site (the number of young-of-the-year individuals are between brackets).

TABLA 3. Frecuencia absoluta de las clases de edad y sexo de *Emys orbicularis* en cada enclave de muestreo (el número de juveniles del año se muestra entre paréntesis).

Site	Adult females	Subadult females	Adult males	Subadult males	Juvenile
Allotopic populations					
Prado	8	1	8	1	1
Alto da Abobeleira	7		4		31 (5)
Poço das Freitas		3	3	2	19
Rib. do Vidoeiro		1	2		4 (2)
Rio Calvão	4	4	9		3 (1)
Sapiões - Terva	13	8	15		4 (1)
Longueira	42	1	36		19 (9)
Malhão	3	4	7		8 (1)
Syntopic populations					
Ludo		4	1		3
Budens		1	3		
Cobres	18	2	26		4 (2)
Apariça96	2		6		
Tornada	2		15		1
Excomungado	2		3		2

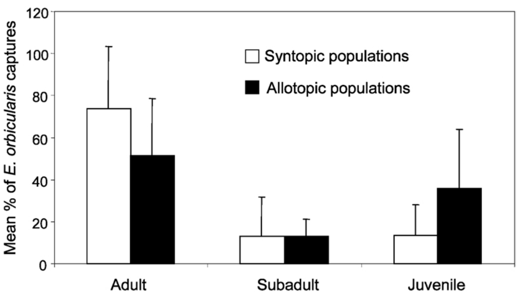

FIGURE 3. Proportion of adults, subadults and juveniles of *E. orbicularis* at allotopic and syntopic populations.

FIGURA 3. Proporción de adultos, subadultos y juveniles de *E. orbicularis* en las poblaciones sintópicas y alotópicas.

mean sex ratio of allotropic populations is consistently much lower (mean = 1.09, SD = 0.46) than that of syntopic populations (mean = 2.68, SD = 2.61; Table 2).

DISCUSSION

A few differences between syntopic and allotopic populations of *E. orbicularis* were found for some population structure parameters, which might suggest that allotopic populations are healthier than syntopic populations. Whether such differences are a consequence of species interaction that somehow would have a negative impact on the recruitment rate of *E. orbicularis* populations, or else the result of distinct environmental optima between species, remains unanswered after the present study.

The most significant result was a markedly lower proportion of juvenile individuals, specially the young-of-the-year, found in syntopic populations. The low frequency of juveniles is not uncommon in other populations, either in syntopy with *M. leprosa* (e.g. KELLER *et al*., 1998; BERTOLERO, 1999) or in allotropy (e.g. MAZZOTTI, 1995), and might be attributed to sampling bias or low recruitment rates. However, as the same capture techniques were adopted on both kinds of population, the problem of sampling bias should be obviated, although it is arguable whether differences on sampled habitat between syntopic and allotropic sites could affect juvenile detectability.

Although in Portugal there is a wide overlap between species in their macro-environmental and habitat preferences (SEGURADO & ARAÚJO, 2004), recently found allotopic populations of *E. orbicularis* in the extreme north of Portugal and in the southwest coast, suggest that spatial and environmental partitioning may occur between species. This would favour the hypothesis for the eventual lower recruitment at syntopic populations being the result of environmental - related constraints. The occurrence of allotopic populations of *E. orbicularis* is probably due to distinct factors acting at distinct scales. While the northern allotopic populations of *E. orbicularis* are probably located at a bioclimatic region that is beyond *M. leprosa* ecological tolerance, the southwest coast populations occupy several temporary pond systems that probably lack habitat conditions required by *M. leprosa*. A similar situation to that found in the southwest coast of Portugal was described by KELLER (1997) in a pond system within the Doñana Natural Park, in the western coast of Andalusia. Here, both species widely coexist on permanent water bodies but *E. orbicularis* was also frequently found on small temporary lagoons where *M. leprosa* was absent. However, KELLER (1997) found an overall positive correlation between the densities of the two species.

If *E. orbicularis* pattern of occurrence is governed by a source-sink dynamics (PULLIAM, 1988), the realized niche may exceed the fundamental niche (for a review see PULLIAM, 2000), i.e. the species may frequently be found in unsuitable sites where populations are maintained by immigration and therefore showing highly skewed age structures. Indeed, factors related to habitat and landscape can seriously influence turtles' demography *per se* (MARCHAND & LITVAITIS, 2004). It is possible that most sampled syntopic populations correspond to sink habitats for *E. orbicularis*, as a result of source habitats being for some reason under-represented in the syntopic population's sample.

The more frequent male-biased sex ratio found at syntopic populations is also in agreement with evidences of lower recruitment rates. Furthermore, male-biased sex ratio could indicate that these populations tended to be maintained by immigrating individuals. In fact, because males have a longer annual activity period, they have probably more opportunities for dispersion. On the other hand, it has been reported that males of *E. orbicularis* moved between different wetland systems more often than females (MEESKE & MÜHLENBERG, 2004). However, it should be noted that it is very difficult to discern which factors contribute to biased sex ratios, since many factors

–other than differential migration between sexes– may be involved, including primary sex ratio, differential age at maturity, differential mortality and sampling bias (GIBBONS, 1990).

In spite of the above mentioned evidences for the effect of habitat on the differences found between the two kinds of populations, there are also some pieces of evidence for a possible effect of ecological interaction between the two species. First, in the southwest coast of Portugal there is a strong spatial segregation between species. While *M. leprosa* is restricted to rivers, streams and permanent ponds, *E. orbicularis* occupies predominantly temporary pond systems that usually dry out during the summer, leaving almost no available habitats during that season (P. Segurado, unpublished data). Second, species tend to segregate in streams at the microhabitat scale, with *E. orbicularis* tending also to be more abundant at more temporary microhabitats (SEGURADO & FIGUEIREDO, 2007). Third, there is evidence for allotopic northern populations of *E. orbicularis* being abundant at a wider variety of habitats, including streams and permanent ponds (P. Segurado, unpublished data), which can be due to the absence of *M. leprosa* in this region. Moreover, populations of *E. orbicularis* occurring in allopatry at other parts of its European range often show preference for large permanent wetlands (FICETOLA *et al.*, 2004). This might suggest that the realized niche of *E. orbicularis* is reduced whenever its range overlaps with that of *M. leprosa*.

Results presented here should be regarded as preliminary since analyses were based on very few samples, which might be the reason for the weak statistical significances found. Moreover samples from allotopic populations are spatially autocorrelated as these populations were restricted to very few regions. Analyses using a more complete set of Iberian populations, preferentially using a stratified scheme according to habitat type, are therefore needed to provide more consistency to these results, allowing more conclusive considerations to be drawn. On the other hand, this study was mainly based on the assumption that size structure provides a means to compare populations' health. However, studies focused on other ecological issues are also required for a proper assessment of the role of *M. leprosa* on *E. orbicularis* populations' life history. More specifically, information on differences between species in resource use, survival, fecundity, behavioural interactions and thermoregulation efficiency are needed. It is also important to assess the persistence of populations with highly skewed age structure such as those found in Portugal.

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