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ORIGINAL PAPER

Boldness and body size of male Spanish terrapins affect their responses to chemical cues of familiar and unfamiliar males

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Abstract Recognition and avoidance of conspecifics based on chemical cues could reduce the risk of aggressive interactions between males. Success in agonistics encounters with unfamiliar males should be lower than with previously known familiar males. Then, males should avoid the chemicals from unfamiliar males with respect to those from familiar males. However, boldness and size could affect the outcome of encounters between males and, consequently, the response to chemical cues of conspecific males. We compared the time spent by male turtles *Mauremys leprosa* in water pools with chemical stimuli from unfamiliar or familiar males or with their own chemical stimuli. We also performed a behavioral test to estimate boldness of turtles in an antipredatory situation. Turtles avoided the chemicals from unfamiliar males respect to familiar ones and their own odors, but their responses depended on boldness and size of the tested turtle. Bold turtles avoided water with chemicals of unfamiliar males, but not with chemicals of familiar males, whereas shy turtles avoided chemicals of both familiar and unfamiliar males. On the other hand, large males avoided the odor from unfamiliar males, but small males did not avoid water with the odor from other males. Results suggest that male *M. leprosa* can detect chemicals released to water from conspecific males and discriminate between familiar and unfamiliar males. However, responses to these chemicals depended on boldness and body size of the responding turtle because these factors may affect intrasexual competition.

Keywords Boldness · Size · Chemical cues · Familiar recognition · Intrasexual competition

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Introduction

Animals should adopt strategies to avoid or minimize the level of aggressive interactions, thereby reducing the associated costs of fighting (Maynard Smith 1982; Huntingford and Turner 1987; Marler and Moore 1988). By assessing in advance their potential opponents and the own possibilities of winning, individuals may predict the outcome of a fight and initiate aggression or retreat (Maynard Smith 1982; Huntingford and Turner 1987). Probability of winning may depend on physical characteristics such as body size. For example, larger individual males have a greater fighting ability and are usually dominant in contests over smaller ones (e.g., López and Martín 2001). In addition, individuals may vary consistently in several personality traits (Réale et al. 2007), including boldness, which can be defined as the reaction of an individual to risky situations such as those occurring in encounters with predators or in agonistic interactions with conspecific competitors. The personality of an individual may also determine its predisposition to engage in agonistic fights and, thus, its probabilities of winning a fight. For example, personality traits are related to the rank attained in a dominance hierarchy in fish (Sundstrom et al. 2004; Colleter and Brown 2011), and boldness affects the outcome of fights in mice (Fuxjager et al. 2010).

When two individuals have already fought each other, prior experience may influence their fighting behavior when they meet again. Usually, when social relationships between two familiar males have been already established, further fights in subsequent agonistic encounters will be of lower intensity or even replaced by ritualized displays (e.g., Glinski and Krekorian 1985; Olsson 1994; Whiting 1999;

López and Martín 2001). Reduced aggressions between neighbors, the so-called "dear enemy" recognition (Fisher 1954), should prevent escalated encounters between them, minimizing the energy expended in agonistic interactions (Jaeger 1981; Glinski and Krekorian 1985; Qualls and Jaeger 1991). However, this requires that individuals were able to recognize each other. Therefore, the mechanisms that allow the recognition of familiar conspecifics (e.g., Carazo et al. 2008) might help to reduce the frequency and intensity of aggressive encounters.

Many species of reptiles present several recognition mechanisms, above all chemical systems that are widespread and are involved in many social interactions such as in sex and individual recognition, territoriality, intrasexual aggressions between males, and female mate choice (Mason and Parker 2010; Martín and López 2011). Chemical cues are used in intrasexual relationships between male lizards, where they can contribute to reduce costs of aggressive interactions (Carazo et al. 2007, 2008; Martín et al. 2007; López and Martín 2011). Responses to chemical cues of familiar and unfamiliar males should be different. For example, male Iberian rock lizards avoid scent marks from unfamiliar conspecific because the probability of success in an agonistic encounter with an unknown male is low (Aragón et al. 2001, 2003). Chemical cues are also potentially important in intraspecific communication of some turtles (Muñoz 2004; Poschadel et al. 2006; Galeotti et al. 2007; Lewis et al. 2007; Polo-Cavia et al. 2009). For example, males of some freshwater turtles can discriminate chemical cues of conspecifics in water and use this information to avoid encounters and agonistic interactions with other males (Poschadel et al. 2006; Polo-Cavia et al. 2009; Ibáñez et al. 2012). Spanish terrapins (*Mauremys leprosa*) are able to recognize several characteristics of potential opponents, such as body size, through chemicals released by these in water (Ibáñez et al. 2012).

In this context, here, we examined whether male *M. leprosa* turtles were able to recognize and distinguish familiar from unfamiliar males via chemical cues released to water, and whether male turtles may modify their space use based on this information alone. We hypothesized that males should avoid pools containing water with chemicals from unfamiliar males respect to pools with water with chemicals from familiar males or with water with their own odor. We also investigated the effects of body size and boldness of male turtles on their response to chemical cues of familiar and unfamiliar conspecific males. Several field studies have shown that larger turtles displaced smaller individuals more often than the converse during basking activity (Bury and Wolfheim 1973; Bury et al. 1979; Lovich 1988; Lindeman 1999). Small male *M. leprosa* may prevent agonistic encounters, where they could have previous disadvantage, detecting and avoiding the chemicals released by larger and stronger males (Ibáñez et al. 2012). In this study, to avoid the effect of size differences, each male always was tested against the odor of other males of similar size (see "Methods"). We predicted that large unfamiliar males, but not small unfamiliar males, could represent a potential dangerous situation during competition for resources or agonistic interactions between males. Thus, we expected that large males avoided the chemical cues from large unfamiliar males, but not the chemical cues from large familiar males or their own odor. In contrast, small males might not need to avoid the odors from other small unfamiliar or familiar males because aggression levels between small males would be low.

Coevolution of personality traits could be driven by predation regime (Huntingford 1976; Dingemanse et al. 2007), but it also could be driven by competition between conspecifics, potentially by sexual selection (Colleter and Brown 2011). In lizards, males with larger heads are more dominant in agonistic interactions (López and Martín 2002) and spent less time inside the refuge after a predatory attack (López et al. 2005). Bold individuals with respect to social interactions may be bold also under threatening situations because of high testosterone levels (Huntingford 1976; Kagan et al. 1988; Tulley and Huntingford 1988; Godin and Crossman 1994). Personality shown during antipredator responses is often related to social dominance in agonistic encounters (López et al. 2005; Colleter and Brown 2011). Thus, we expected that turtles that showed a bold or shy behavior during antipredator test should act equally in response to chemical cues of other males. Hierarchy formation in turtles depends on the ability to obtain food and appeared stable over time (Froese and Burghardt 1974). We expected that during the familiarization process, bold male turtles became dominant or, at least, more successful in competition than shy males. Thus, bold male turtles should avoid the chemicals from unfamiliar males, but not from familiar males because they could have established a previous hierarchy with them, and these bold turtles would be very likely dominant. In contrast, shy turtles should avoid the chemicals from any kind of male because they are likely to be dominant on shy males.

Methods

Animals and study sites

The Spanish terrapin (*M. leprosa*) is a thermophile medium-sized semiaquatic turtle, which is found in southwestern Europe and northwestern Africa (Busack and Ernst 1980; Da Silva 2002). It preferably inhabits ponds and streams with riparian vegetation, being less common in large rivers and reservoirs. Males are sexually mature at 7 years old, with about 12 or 14 cm carapace length (Pérez et al. 1979).

During March 2011, we used funnel traps to capture adult male turtles in several small streams, ponds, and tributaries of the Guadiana River located within dehesa oaklands (Alconchel and Olivenza, Badajoz Province, SW Spain). We used a modified version of the traditional underwater funnel traps, which include a mesh chimney that reaches from the body of the trap to the surface allowing the turtles to come to the surface to breath (Kuchling 2003; T & L Netmaking, Mooroolbark, Victoria, Australia). We baited traps with sardines and protected them from terrestrial predators by waiting in the proximity. We revised the traps every hour to collect turtles. All collected turtles were alive and did not show any sign of being stressed inside the traps.

Turtles were transported to "El Ventorrillo" Field Station, near Navacerrada (Madrid Province) where experiments were conducted. Turtles were housed in individual outdoor plastic aquaria (60×40×30 cm) containing water and rocks that allow turtles to bask out of water. The photoperiod and temperature were the same as those of the surrounding area. Turtles were fed three times a week with minced meat, earthworms, and a commercial compound of fish "pellets." Turtles were held in captivity for more than 2 weeks before starting the experiments to allow acclimation to captivity conditions. All individuals were healthy and in good condition during the tests, and at the end of experiments, all of them had maintained or increased their body mass. Turtles were then returned to the exact locations of capture.

Male turtles were measured with a metal ruler (precision 0.1 mm). Carapace length (i.e., the greatest straight-line distance from the anterior end to the posterior end of the shell) ranged between 165 and 197 mm ($\overline{X} + 1\pm1$ standard error (SE)=176± 0.2 mm, N=20). Individuals with carapace lengths below and above the median in the sample were classified as "small" (<176 mm, N=10) and "large" (>176 mm, N=10), respectively.

Familiarization procedure

Familiarity was established by housing pairs of male turtles together in the same aquaria for 2 weeks before the experiments began. Each pair of "familiar males" was composed of two turtles of similar body size from the same capture site (Olivenza or Alconchel). During the familiarization period, turtles basked and ate normally. Individuals basked on the brick, alone, or stacked one on the top of the other, given the small emerged area available for basking. Thus, one turtle could displace the other during basking activity (Lindeman 1999; Cadi and Joly 2003; Polo-Cavia et al. 2010). Similarly, sometimes, one turtle displaced or bit the other while competing for food items (Polo-Cavia et al. 2011). These observations indicated that males interacted and competed for the resources during this period. However, turtles did not show any signs of stress (e.g., stopping or reducing their eating or basking activity), and all maintained good condition.

We considered as "unfamiliar males" those that had not been maintained together in the same aquaria during the 2 weeks prior to the experiments and that had been captured in two different study sites (Alconchel or Olivenza), which are separated by 25 km, within the same turtle population. In similar terrapin species, home range size of males varied between 0.7 and 5.6 ha (Schubauer et al. 1990; Rowe and Dalgarn 2010). Thus, with this procedure, we were confident that males from the different capture sites had not had any interaction in the field before being captured. We cannot rule out the possibility of differences between capture locations in chemical composition of the pheromones released to water by male turtles, which might confound population with the time spent together in the familiarization treatment. However, genetic- or phenotypic-dependent differences in chemical secretions of turtles between study sites are highly unlikely. This is because both capture sites belong to the same turtle population, which is distributed uniformly through the study area occupying several freshwater systems interconnected by a main river and with almost identical ecological conditions. Therefore, we considered that any differences between study sites should be very small and should not affect to our experiment.

Boldness

The boldness of male turtles was estimated by simulating a predatory attack, turning over them onto their own shell with the plastron upside. Thus, we reproduced a high-risk situation where the turtle had to turn itself back to its normal position (i.e., righting response) before escaping of possible following attacks by the predator (Martín et al. 2005). We considered two stages in the righting response of turtles, the appearance time, and the mechanical righting time (see below). The first time represents the behavioral decision of turtles on when starting (Martín et al. 2005), while the second one depends mainly on physical traits and physiological state of turtles (Steyermark and Spotila 2001; Elnitsky and Claussen 2006). During the appearance time, turtles entirely withdrawn into their shell had no visual information of the predator surroundings. Turtles must accurately assess the costs of remaining overturned vs those of returning to natural position and properly determine the optimal time to initiate righting, basing on this trade-off (Martín et al. 2005). Similarly, turtles and other animals adjust the time spent inside refuges or withdrawn within the shell, so that the optimal time to emerge occurs when the costs of staying equal the costs of leaving (Sih 1997; Martín et al. 2005; Cooper and Frederick 2007; Polo-Cavia et al. 2008). Specifically, overturned turtles may suffer increased predation exposure, overheating, or difficulties to breath, but righting response requires a particular energy effort related with variables such as health and nutritional state or temperature, which may also vary over time. In this way, when starting to right (i.e., the duration of the appearance time), this may be determined by the probability of righting success by turtles, which depends on individual intrinsic variables and external factors that affect their physiological response.

All the tests were performed at midday, and the air temperature was always around 25 °C (digital thermometer, accuracy of ±0.1 °C). Before the beginning of the tests, turtles were in their individual own home aquaria, where they could bask and attain an optimal body temperature. To standardize the trials, the same person (AI) performed all the tests and always followed the same procedure. The experimenter gently took a turtle from its home aquaria and handled it briefly while moving it to an open grassy area located 5 m from the aquaria. Thus, the time of handling was similar for all the tests. Then, the turtle was placed on the ground onto its carapace with the plastron upside. Turtles typically spent some time entirely withdrawn in the shell, after which they put the head partially out and scanned the surroundings. Then, turtles put the head, neck, and legs entirely out from the shell and started to right. From a hidden position and always at the same distance (5 m) from the tested turtle, the experimenter used binoculars to observe it and, by using a stopwatch to the nearest second, recorded the time spent hidden into the shell until the turtle emerged from the carapace, and the eyes could be seen from above (appearance time) and the total time taken by the turtle to turn back (righting time). Each turtle was tested and turned down at only one time. Because the appearance time represents the time spent into the refuge (shell) before starting to escape (Martín et al. 2005; Polo-Cavia et al. 2008), we used this time to calculate a "boldness index." Shy males should spend more time hidden inside the refuge (i.e., shell) and delay the start of an active escape from the predator in comparison to bold males, which should have shorter appearance times. Thus, we used the median of the appearance time to classify males as "shy" (i.e., values above the median) or "bold" (i.e., values below the median).

Choice of water pools

We analyzed the use by male *M. leprosa* of pools with water containing (1) their own chemical stimuli, (2) chemical stimuli of familiar males, or (3) chemical stimuli of unfamiliar males. The experiments were conducted outdoor using two artificial pools (two plastic containers of 60×40×15 cm) connected by stone ramps that allowed turtles to move easily from one pool to another and to bask out of water (Ibáñez et al. 2012). In each trial, one of the pools contained clean water, and the other was filled with water containing chemical stimuli. Clean water was obtained from a nearby mountain spring stream that did not contain turtles. Water with different chemical stimuli came from the individual containers where turtles were kept separately. To obtain the water with chemical stimuli, we filled individual containers with clean water to about half of the container (18 l) and left the turtle there for 3 days. During this time, we did not provide food to avoid possible contamination of water with food chemicals. Thus, clean water was impregnated only with the scent of each turtle (Ibáñez et al. 2012).

Each focal male was tested in three different trials: "clean water vs own," "clean water vs familiar male," and "clean water vs unfamiliar male." To remove possible effects due to differences in size between the focal male and the male donor of the chemical stimuli, each male was tested with chemicals of other males of similar size. Differences in carapace length between the focal male and the male donor of the chemical stimuli were very small and ranged between +10 and −13 mm in the familiar treatment and between +29 and −13 mm in the unfamiliar treatment. These differences did not significantly differ between treatments (GLMM; $F_{1,19}$=1.64, P=0.21) (see below). All experimental treatments were presented to each individual in random order and balanced. The positions of the pools were also randomized. The experiments were spaced in time at least 2 days, and no individual participated in one experiment on two successive days to avoid the possible stressful effects on the next test. Tests were only made on sunny days when the turtles were active, excluding very cloudy and rainy days. We monitored the temperature of pools at the beginning of the experiments (with a digital thermometer, accuracy of ±0.1 °C), the illumination intensity, and the depth of the water column to ensure that these conditions were similar in both pools during all trials (DeRosa and Taylor 1980). After each test, all pools and ramps were cleaned with clean water and allowed to dry outdoor to avoid contamination with other smells.

Before starting each trial, turtles were allowed to bask for at least 2 h in their home containers. At the beginning of each test, a turtle was gently placed in the middle of the stone ramp linking the two experimental pools, to avoid any initial bias in choosing a pool or another. Turtles were usually very active, shifting between pools more frequently at the beginning of the experiment and less frequently as time passed by. We used the instant registration procedure, monitoring each turtle from a hidden position for 2 h and noting the location of the individual every 5 min (24 total records for each turtle in each test). In each record, if the turtle was inside one of the two pools (clean or chemical stimuli), it was designated as having chosen temporarily that particular pool, whereas if the position of the turtle was not clear (e.g., on the ramp), it was designated as having made no choice (Ibáñez et al. 2012).

We evaluated the "preferences" of each turtle for each chemical stimulus as the proportion of time (excluding time in the "no-choice" places) that each turtle spent in each pool (clean vs chemical stimulus). Preliminary experiments showed no significant differences in the time spent by turtles in two pools with clean water (Ibáñez et al. 2012).

Statistical analyses

To analyze differences in use of water pools with different chemical cues across treatments by the same individual subject turtle, we ran a three-way repeated measure analysis of variance (ANOVA) with the experimental treatment ("own" vs "familiar" vs "unfamiliar") as a within-subject factor and the size and boldness of the responding turtles as two between-subject factors. We transformed data (square root) to normalize them before analysis (verified by Shapiro–Wilk tests), and we also tested the homogeneity of variances (Levene's test). Post hoc analyses were performed using Fisher's least significance difference tests when ANOVAs showed significance.

Results

Boldness

Appearance times of turtles ranged between 2 and 305 s ($\overline{X}+1$ SE = 68 + 21s). There were no significant differences in appearance time between males from the two study sites (one-way ANOVA; $F_{1,18}$=0.22, P=0.64). Body size (carapace length) was not significantly related with the appearance time (r=0.20, N=20, $F_{1,18}$=0.78, P=0.39). For further analyses, males (N=10) with appearance times below the median (i.e., <23 s) were classified as bold, whereas males (N=10) with appearance times above the median (i.e., >23 s) were classified as shy. Righting time ranged between 12 and 736 s ($\overline{X}+1$SE = 189 + 51s) and was positively and significantly related with the appearance time (r=0.83, N=20, $F_{1,18}$=40.54, P<0.0001). Thus, appearance and righting times could describe the same shy–bold continuum within this turtle species.

Responses to chemical stimuli

There were no significant overall differences among experimental treatments in the time spent by male turtles in the pool with chemical stimuli (Table 1; Fig. 1). There were no significant differences between shy and bold individuals or between small and large individuals in the overall time spent in water with scent of conspecifics (Table 1). However, boldness and body size affected significantly the responses to the different treatments as shown by the significant interaction terms between treatment and boldness and between treatment and size (Table 1). The interaction between size and boldness was not significant (Table 1).

Thus, with respect to boldness of turtles, shy turtles tended, although not significantly, to spend more time in the pool with their own chemical stimuli than in the familiar (Fisher's test, P=0.08) or unfamiliar treatments (P=0.06), which did not significantly differ (P=0.75) (Fig. 1a). In contrast, bold turtles spent significantly more time in the pools with scent of familiar males than in pools with scent of unfamiliar males (P=0.02) or with their own odor (P=0.047), which did not significantly differ (P=0.70) (Fig. 1a). In addition, shy turtles spent significantly less time than bold ones in pools with chemicals of familiar males (P=0.03), but they did not significantly differ in the other treatments (P>0.20 in both cases) (Fig. 1a).

Table 1 Results of a repeated measures three-way ANOVA comparing the time spent by male turtles in pools with water with scent from conspecifics among the different treatments (within factor) and the effects of boldness and body size (between effects) of focal males on their responses to the different treatments

	F	*df*	*P*
Intercept	641.71	1,16	**<0.0001**
Treatment	2.17	2,32	0.13
Boldness	0.20	1,16	0.66
Size	0.0015	1,16	0.97
Treatment × boldness	3.39	2,32	**0.046**
Treatment × size	3.47	2,32	**0.043**
Boldness × size	0.28	1,16	0.60
Treatment × boldness × size	0.13	2,32	0.87

Significant effects (P<0.05) are indicated in bold

On the other hand, the body size of males also affected their responses to the different treatments (Table 1; Fig. 1b). Small males did not significantly differ in the time spent in the different pools with chemical stimuli among treatments (Fisher's tests, all P>0.27) (Fig. 1b). In contrast, large males spent significantly less time in the pools with scent of unfamiliar males than in pools with scent of familiar males (P=0.027) or their own chemicals (P=0.003), which did not significantly differ (P=0.41) (Fig. 1b). However, small and large males did not significantly differ in the overall time spent in the pool with chemical stimuli in the different treatments (own, P=0.10; familiar, P=0.93; unfamiliar, P=0.07).

Discussion

Our results confirm that male *M. leprosa* can detect the chemicals released to water from conspecific males. Responses to chemicals from familiar and unfamiliar males depended on boldness and body size of the responding turtle. Male *M. leprosa* classified as bold and those classified as large (both traits are independent) spent less time in water pools containing chemical cues from unfamiliar males than in the pools containing chemical cues from familiar males. These results suggest that male *M. leprosa* turtles are able to discriminate between scents of familiar and unfamiliar males. Similarly, in *Gopherus agassizii* turtles, chin gland secretions of turtles allow discrimination between familiar and unfamiliar males (Alberts et al. 1994). The ability of territorial males to discriminate between

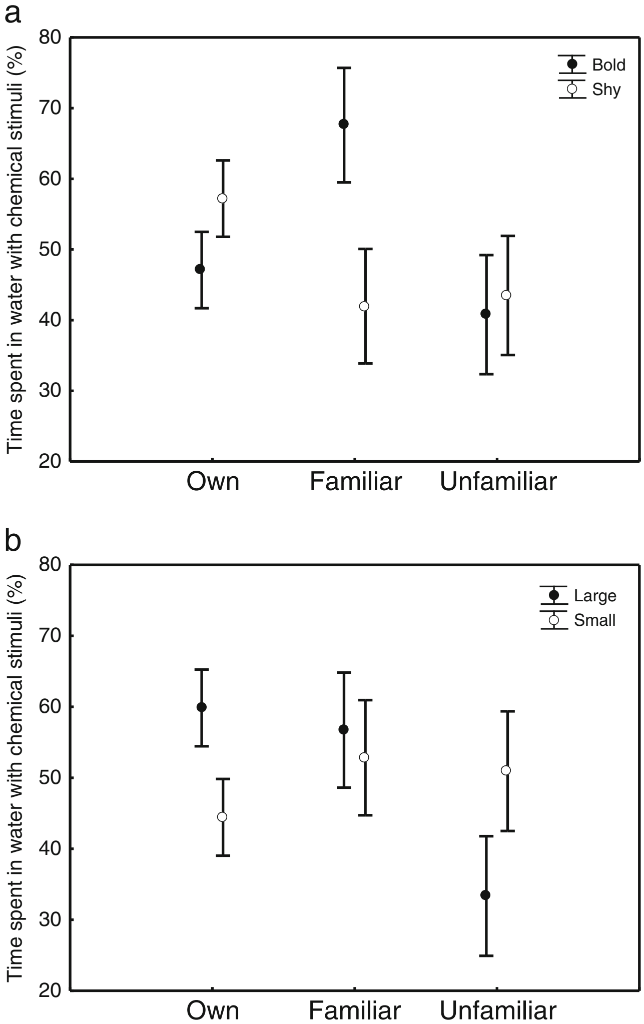

Fig. 1 Percent time ($\overline{X} \pm$ SE) spent by male turtles in pools with water with conspecific chemicals in the different treatments depending on **a** the boldness (shy vs bold) or **b** the body size (small vs large) categories of the responding male

neighbors (familiar) and non-neighbors (unfamiliar) might help to stabilize social systems by reducing the frequency and intensity of aggressive encounters between males (Glinski and Krekorian 1985). Thus, the results obtained in this study suggest that recognition of familiar and unfamiliar males by male turtles *M. leprosa* could prevent escalated agonistic interactions between males.

However, responses to conspecific chemical cues were not similar for all males. Boldness affected responses to chemical cues of familiar and unfamiliar male turtles. Shy turtles spent more time in the pool with their own chemical, avoiding pools with chemicals from either familiar or unfamiliar males. In contrast, bold males preferred the chemicals of familiar males to the chemicals of unfamiliar males. In any case, shy males spent less time than bold ones in the pools with chemicals from familiar males. Boldness is associated with dominance in fishes and other animals (Sundstrom et al. 2004; López et al. 2005; Colleter and Brown 2011). In the white-footed mice (*Peromyscus leucopus*), shy or bold personality types modulate winning ability in a context manner, suggesting that this species uses a conditional strategy to appropriately gate displays of social aggressions (Fuxjager et al. 2010). The results found in our experiment showed that male turtle's boldness in antipredatory contexts could also be related with boldness in male–male interactions. Thus, shy–bold behavior could influence the selection of pools with chemicals from other males. Hierarchy formation during familiarity process could explain avoidance of the familiar chemicals by the shy males that could had been in a previous disadvantageous situation in agonistic encounters and competition for the resources. Otherwise, bold males preferred to stay in pools with odors of familiar males, probably because they could be victorious in agonistic interactions. In some spiders, previous experience during the male interactions could influence the outcome of the future fights and could learn to be winners or losers (Whitehouse 1997). In turtles, competition for resources such as food (Polo-Cavia et al. 2011) or basking sites (Cadi and Joly 2003; Polo-Cavia et al. 2010) could explain hierarchy formation during the familiarization process in our study. Also, familiar males could have established previously in the field dominance relationships between them because they were captured in the same or very nearby locations. Thus, both in the field and during the familiarization procedure, turtles could have experienced several fights and/or aggressions. In this context, bold turtles could be more successful during agonistic interactions or competition for food or basking sites than the shy males. Both shy and bold males had the same avoidance responses of the odors of unfamiliar males, suggesting that chemicals from unknown individual males could represent a potential dangerous situation for all males. An alternative explanation could be that, because the unfamiliar male came from a different capture site than the tested male, there might be interpopulational differences in chemical composition of sex pheromones. Thus, bold males could prefer to occupy the pools with water with chemicals of males of the same population (i.e., familiar) than the pools with water of chemicals of males of other populations (i.e., unfamiliar) because a new different smell could represent a risky situation independently of the familiarization procedure followed in this experiment.

Size and relative size in male *M. leprosa* seem to be the most important factors or, at least, more important than other traits such as body condition and health state, in modulating responses to chemicals of conspecific males (Ibáñez et al. 2012). Previous studies showed that smaller males avoid using water pools with chemicals of large males, which, in contrast, prefer to use water pools with chemicals of smaller males (Ibáñez et al. 2012). Similar results were observed in the turtle *Emys orbicularis* (Poschadel et al. 2006). The results of the current experiment suggest that only large males seem to recognize and avoid the water pools with chemicals from unfamiliar males. The apparent lack of discrimination between familiar and unfamiliar chemical cues by small males could be explained because, in our study, the small males always were tested against the odor of other males of similar small size, and large males were always tested with odors of males of similar large size. In agreement with these results, several field studies have shown that larger turtles displaced smaller individuals more often than the converse during basking activity (Bury and Wolfheim 1973; Bury et al. 1979; Lovich 1988; Lindeman 1999). Thus, the pheromones released in water from large unfamiliar males, but not necessarily from small unfamiliar males, could inform other males about a threatening situation. This difference would lead to size-dependent responses to conspecific cues (Aragón et al. 2000). These results suggest that familiarity with neighbors could allow inferior competitors to reduce the frequency and intensity of interactions (Aragón et al. 2001, 2007; López and Martín 2011).

Our results provide evidence that boldness and size could influence intrasexual competition between males. Recently, it has been shown that variation in personality may affect female mate preferences (David and Cezilly 2011). In the same way, male–male interactions in the Spanish terrapin could depend on the individual personality along the shy–bold axis and consequently affect their responses to conspecific chemicals. Nevertheless, this is the first study to demonstrate both chemical discrimination between familiar and unfamiliar males in a freshwater turtle as well as the importance of some traits such as size and boldness in the responses to conspecific chemical cues. Future studies should focus on the influence of boldness on sexual selection and on the role of chemical cues in modulating these processes.

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Ethical standards Captures and experiments complied with all current laws of Spain and the Environmental Agency (“Consejería de Industria, Energía y Medio Ambiente”) of the “Junta de Extremadura” Local Government (permit number CN0008/11/ACA).

References

Alberts AC, Rostal DC, Lance VA (1994) Studies on the chemistry and social significance of chin gland secretions in the desert tortoise, *Gopherus agassizii*. Herpetol Monogr 8:116–124

Aragón P, López P, Martín J (2000) Size-dependent chemosensory responses to familiar and unfamiliar conspecific faecal pellets by the Iberian rock-lizard, *Lacerta monticola*. Ethology 106:1115–1128

Aragón P, López P, Martín J (2001) Chemosensory discrimination of familiar and unfamiliar conspecifics by lizards: implications of field spatial relationships between males. Behav Ecol Sociobiol 50:128–133

Aragón P, López P, Martín J (2003) Differential avoidance responses to chemical cues from familiar and unfamiliar conspecifics by male Iberian rock lizards (*Lacerta monticola*). J Herpetol 37:583–585

Aragon P, Lopez P, Martin J (2007) Familiarity modulates social tolerance between male lizards, *Lacerta monticola*, with size asymmetry. Ethol Ecol Evol 19:69–76

Bury RB, Wolfheim JH (1973) Aggression in free-living pond turtles (*Clemmys marmorata*). Bioscience 23:659–662

Bury RB, Wolfheim JH, Luckenbach RA (1979) Agonistic behavior in free-living painted turtles (*Chrysemys picta bellii*). Biol Behav 4:227–239

Busack SD, Ernst CH (1980) Variation in Mediterranean populations of *Mauremys* Gray 1869 (Reptilia, Testudines, Emydidae). Ann Carnegie Mus 49:251–264

Cadi A, Joly P (2003) Competition for basking places between the endangered European pond turtle (*Emys orbicularis galloitalica*) and the introduced red-eared slider (*Trachemys scripta elegans*). Can J Zool 81:1392–1398

Carazo P, Font E, Desfilis E (2007) Chemosensory assessment of rival competitive ability and scent-mark function in a lizard, *Podarcis hispanica*. Anim Behav 74:895–902

Carazo P, Font E, Desfilis E (2008) Beyond ‘nasty neighbours’ and ‘dear enemies’? Individual recognition by scent marks in a lizard (*Podarcis hispanica*). Anim Behav 76:1953–1963

Colleter M, Brown C (2011) Personality traits predict hierarchy rank in male rainbow fish social groups. Anim Behav 81:1231–1237

Cooper WE Jr, Frederick WG (2007) Optimal time to emerge from refuge. Biol J Linn Soc 91:375–382

Da Silva E (2002) *Mauremys leprosa* (Schweiger, 1812). Galápago leproso. In: Pleguezuelos JM, Márquez R, Lizana M (eds) Atlas y libro rojo de los anfibios y reptiles de España. Asociación Herpetológica Española-Ministerio de Medio Ambiente, Madrid, pp 143–146

David M, Cezilly F (2011) Personality may confound common measures of mate-choice. PLoS One 6:e24778

DeRosa CT, Taylor DH (1980) Homeward orientation mechanisms in three species of turtles (*Trionyx spinifer*, *Chrysemys picta*, and *Terrapene carolina*). Behav Ecol Sociobiol 7:15–23

Dingemanse NJ, Wright J, Kazem AJN, Thomas DK, Hickling R, Dawnay N (2007) Behavioural syndromes differ predictably between 12 populations of three-spined stickleback. J Anim Ecol 76:1128–1138

Elnitsky MA, Claussen DL (2006) The effects of temperature and inter-individual variation on the locomotor performance of juvenile turtles. J Comp Physiol B 176:497–504

Fisher J (1954) Evolution and bird sociality. In: Huxley JS, Hardy AC, Ford EB (eds) Evolution as a process. Allen and Unwin, London, pp 71–83

Froese AD, Burghardt GM (1974) Food competition in captive juvenile snapping turtles, *Chelydra serpentina*. Anim Behav 22:735–740

Fuxjager MJ, Montgomery JL, Becker EA, Marler CA (2010) Deciding to win: interactive effects of residency, resources and ‘boldness’ on contest outcome in white-footed mice. Anim Behav 80:921–927

Galeotti P, Sacchi R, Pellitteri-Rosa D, Fasola M (2007) Olfactory discrimination of species, sex, and sexual maturity by the Hermann’s tortoise *Testudo hermanni*. Copeia 2007:980–985

Glinski TH, Krekorian CO (1985) Individual recognition in free-living adult male desert iguanas, *Dipsosaurus dorsalis*. J Herpetol 19:541–544

Godin JGJ, Crossman SL (1994) Hunger-dependent predator inspection and foraging behaviors in the threespine stickleback (*Gasterosteus aculeatus*) under predation risk. Behav Ecol Sociobiol 34:359–366

Huntingford FA (1976) Relationship between anti-predator behavior and aggression among conspecifics in three-spined stickleback, *Gasterosteus aculeatus*. Anim Behav 24:245–260
Huntingford FA, Turner AK (1987) Animal conflict. Chapman and Hall, New York
Ibáñez A, López P, Martín J (2012) Discrimination of conspecifics' chemicals may allow Spanish terrapins to find better partners and to avoid competitors. Anim Behav 83:1107–1113
Jaeger RG (1981) Dear enemy recognition and the costs of aggression between salamanders. Am Nat 117:962–974
Kagan J, Reznick JS, Snidman N (1988) Biological bases of childhood shyness. Science 240:167–171
Kuchling G (2003) A new underwater trap for catching turtles. Herpetol Rev 34:126–128
Lewis CH, Molloy SF, Chambers RM, Davenport J (2007) Response of common musk turtles (*Sternotherus odoratus*) to intraspecific chemical cues. J Herpetol 41:349–353
Lindeman PV (1999) Aggressive interactions during basking among four species of Emydid turtles. J Herpetol 33:214–219
López P, Martín J (2001) Fighting rules and rival recognition reduce costs of aggression in male lizards, *Podarcis hispanica*. Behav Ecol Sociobiol 49:111–116
López P, Martín J (2002) Locomotor capacity and dominance in male lizards *Lacerta monticola*: a trade-off between survival and reproductive success? Biol J Linn Soc 77:201–209
López P, Martín J (2011) Male Iberian rock lizards may reduce the costs of fighting by scent-matching of the resource holders. Behav Ecol Sociobiol 65:1891–1898
López P, Hawlena D, Polo V, Amo L, Martín J (2005) Sources of individual shy-bold variations in antipredator behaviour of male Iberian rock lizards. Anim Behav 69:1–9
Lovich J (1988) Aggressive basking behavior in eastern painted turtles (*Chrysemys picta picta*). Herpetologica 44:197–202
Marler CA, Moore MC (1988) Evolutionary costs of aggression revealed by testosterone manipulations in free-living male lizards. Behav Ecol Sociobiol 23:21–26
Martín J, López P (2011) Pheromones and reproduction in Reptiles. In: Norris DO, Lopez KH (eds) Hormones and reproduction of vertebrates, vol 3, Reptiles. Academic, San Diego, CA, pp 141–167
Martín J, Marcos I, López P (2005) When to come out from your own shell: risk-sensitive hiding decisions in terrapins. Behav Ecol Sociobiol 57:405–411
Martín J, Moreira PL, López P (2007) Status-signalling chemical badges in male Iberian rock lizards. Funct Ecol 21:568–576
Mason RT, Parker MR (2010) Social behavior and pheromonal communication in reptiles. J Comp Physiol A 196:729–749
Maynard Smith J (1982) Evolution and the theory of games. Cambridge University Press, Cambridge, UK
Muñoz A (2004) Chemo-orientation using conspecific chemical cues in the stripe-necked terrapin (*Mauremys leprosa*). J Chem Ecol 30:519–530
Olsson M (1994) Rival recognition affects male contest behavior in sand lizards (*Lacerta agilis*). Behav Ecol Sociobiol 35:249–252
Pérez M, Collado E, Ramo C (1979) Crecimiento de *Mauremys caspica leprosa* (Schweigger, 1812) (Reptilia, Testudines) en la Reserva Biologica de Doñana. Doñana Acta Vertebrata 6:161–178
Polo-Cavia N, López P, Martín J (2008) Interspecific differences in responses to predation risk may confer competitive advantages to invasive freshwater turtle species. Ethology 114:115–123
Polo-Cavia N, López P, Martín J (2009) Interspecific differences in chemosensory responses of freshwater turtles: consequences for competition between native and invasive species. Biol Invas 11:431–440
Polo-Cavia N, López P, Martín J (2010) Competitive interactions during basking between native and invasive freshwater turtle species. Biol Invas 12:2141–2152
Polo-Cavia N, López P, Martín J (2011) Aggressive interactions during feeding between native and invasive freshwater turtles. Biol Invas 13:1387–1396
Poschadel JR, Meyer-Lucht Y, Plath M (2006) Response to chemical cues from conspecifics reflects male mating preference for large females and avoidance of large competitors in the European pond turtle, *Emys orbicularis*. Behaviour 143:569–587
Qualls CP, Jaeger RG (1991) Dear enemy recognition in *Anolis carolinenis*. J Herpetol 25:361–363
Réale D, Reader SM, Sol D, McDougall PT, Dingemanse N (2007) Integrating animal temperament within ecology and evolution. Biol Rev 82:291–318
Rowe JW, Dalgarn SF (2010) Home range size and daily movements of Midland painted turtles (*Chrysemys picta marginata*) in relation to body size, sex, and weather patterns. Herpetol Conserv Biol 5:461–473
Schubauer JP, Gibbons JW, Spotila JR (1990) Home range and movement patterns of slider turtles inhabiting Par Pond. In: Gibbons JW (ed) Life history and ecology of the slider turtle. Smithsonian Institution Press, Washington, DC, pp 223–232
Sih A (1997) To hide or not to hide? Refuge use in a fluctuating environment. Trends Ecol Evol 12:375–376
Steyermark AC, Spotila JR (2001) Body temperature and maternal identity affect snapping turtle (*Chelydra serpentina*) righting response. Copeia 2001:1050–1057
Sundstrom LF, Petersson E, Hojesjo J, Johnsson JI, Jarvi T (2004) Hatchery selection promotes boldness in newly hatched brown trout (*Salmo trutta*): implications for dominance. Behav Ecol 15:192–198
Tulley JJ, Huntingford FA (1988) Additional information on the relationship between intra-specific aggression and anti-predator behavior in the three-spined stickleback, *Gasterosteus aculeatus*. Ethology 78:219–222
Whitehouse MEA (1997) Experience influences male-male contests in the spider *Argyrodes antipodiana* (Theridiidae: Araneae). Anim Behav 53:913–923
Whiting MJ (1999) When to be neighbourly: differential agonistic responses in the lizard *Platysaurus broadleyi*. Behav Ecol Sociobiol 46:210–214