

# Female preference for males that have exclusively marked or invaded territories depends on male presence and its identity in the subterranean rodent *Ctenomys talarum*

María Sol Fanjul  | María Florencia Varas | Roxana Rita Zenuto

Grupo 'Ecología Fisiológica y del Comportamiento', Instituto de Investigaciones Marinas y Costeras (IIMyC), Universidad Nacional de Mar del Plata, Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET), Mar del Plata, Argentina

## Correspondence

María Sol Fanjul, Grupo 'Ecología Fisiológica y del Comportamiento', Instituto de Investigaciones Marinas y Costeras (IIMyC), Box 1260, CP 7600, Mar del Plata, Argentina.  
Email: msfanjul@mdp.edu.ar

## Funding information

Fondo para la Investigación Científica y Tecnológica, Grant/Award Number: PICT 0998; Consejo Nacional de Investigaciones Científicas y Técnicas, Grant/Award Number: PIP 0272

Editor: L. Ebensperger

## Abstract

Territorial scent-marking provides chemical records of male competitive interactions that are available to females, who gain valuable information to assess and identify best quality partners. In this context, the solitary subterranean rodent tuco-tuco (*Ctenomys talarum*) offers excellent possibilities to evaluate the effects of male exclusive scent-marking of territories on female assessment. For evaluation, we used wild caught individuals of *C. talarum*, manipulated their scent marks within the territories in captive conditions and staged preference tests where females were able to choose between exclusive and invaded territories. The evaluation was performed in two scenarios considering the identity of the intruder scent mark: territories invaded by a strange male and territories invaded by a neighbour male. Females investigated the chemical cues deposited on the substrate of the exclusively marked territory more frequently. Next, females displayed equal interest to scent samples of both males presented in a Y-maze. Finally, when females could gain access to both individually isolated males and their scent-marked territories, they spent more time within invaded territories despite they visited them with the same frequency. Moreover, females tried to get in contact by scratching the mesh of the owner of the invaded territory more frequently. We found that females of *C. talarum* evaluate the homogeneity (exclusiveness) of scent marks within a male territory and then show preferences in relation to the identity of the intruder's scent –whether strange or neighbour.

## KEYWORDS

countermarking, female choice, neighbour males, sexual selection, subterranean rodents, territoriality

## 1 | INTRODUCTION

In mammals, the outstanding male–male competition frequently leads to the underestimation of the effect of female preference on reproductive output (Clutton-Brock & McAuliffe, 2009). Given female investment in reproduction, careful selection of the quality

of mating partners is expected (Andersson, 1994; Clutton-Brock & McAuliffe, 2009). Benefits of mate choice in mammals, especially in rodents, include female preferences on characteristics that positively impact on female's own survival, breeding success and offspring fitness (Clutton-Brock & McAuliffe, 2009). To exert such preference, females rely on different communication channels by assessing and discriminating information regarding potential mates. Chemical cues play a key role in reproductive communication (Arakawa, Blanchard,

MSF and MFV should be considered joint first author.

Arakawa, Dunlap, & Blanchard, 2008; Penn & Potts, 1998), particularly in rodents, where scents are proposed to be used instead of weaponry, ornaments and/or body size in intrasexual and intersexual selection (Penn & Potts, 1998). Scents can be produced in specialized sources (e.g., glands and skin) but they can also originate as metabolic products (e.g., urine and faeces; Eisenberg & Kleiman, 1972; Kavaliers, Choleris, Agmo, & Pfaff, 2004). Therefore, individual scents are expected to suffer changes as consequence of a complex array of factors such as a genetic profile, diet, stress levels, parasite burden and endocrine products, among others (Hurst, 2009; Penn & Potts 1998). Based on scent composition, individuals are able to discriminate conspecific individual identity, gender, reproductive condition, physiological condition and social status (Arakawa et al., 2008; Blaustein, 1981; Hurst, 2009; Tang-Halpin, 1986; Zinkevich & Vasilieva, 2001). Also, many factors are known to affect animal odours and consequently alter its attractiveness to potential mates (Hurst, 2009); some of these factors could be diet quality (Ferkin, Sorokin, Johnston, & Lee, 1997), parasitic load (Kavaliers & Colwell, 1995) or homogeneity of scent marks within a territory (Johnston, Sorokin, & Ferkin, 1997; Ferkin, 1999). There is homogeneity of scents marks when the marks within a given territory belong exclusively to the owner. In contrast, heterogeneity of scent marks corresponds to the case when territory marks were partially countermarked by an intruder (Ferkin, 1999; Johnston et al., 1997).

Scent-marking, the spatiotemporal pattern of scent deposition by an individual within a territory, is an important source of information. Scents deposited along trails, runways and prominent objects in the habitat not only allow the orientation of owners within their territories (Gosling, 1982) but also provide information to conspecifics about the competitive abilities of the owner (Fisher, Swaisgood, & Fitch-Snyder, 2003; Hurst & Rich, 1999). In territorial species, scent-marking allows to identify neighbours and consequently minimize aggressiveness during encounters ("dear enemy" hypothesis; Temeles, 1994). The capacity of an individual to saturate a territory with its own scents could be used as a signal of competitive and territory defence proficiency (Rich & Hurst, 1998). Only individuals that successfully defend a territory, and prevent intruders to overlay their own scents (Rich & Hurst, 1998), can ensure that their own scent predominates in it (Hurst, 1993). This also allows females to identify a male as the territory owner, by contrasting their body scents with the predominating scents in the area (Gosling, 1982, 1990; Hurst, Thom, Nevison, Humphries, & Beynon, 2005; Rich & Hurst, 1998), and consequently assess the quality of a potential mate (Rich & Hurst, 1999). In this way, chemical records of competitive interactions left on the substrate by scent-marking of a territorial male and by challenging countermarks of competitors are available to females which gain valuable information to discriminate between partners of varying quality.

Subterranean rodents are a particular group of species, mostly solitary and highly territorial (Lacey, Patton, & Cameron, 2000). Given the reduced encounter ratio of individuals, communication between such unsocial animals is challenging. Accordingly, both scent-marking and the use of chemical signals to evaluate conspecifics are

highly expected. Furthermore, chemical communication is enhanced by natural selection in environments where other communicative channels (e.g., visual) are precluded (Bennett, Faulkes, & Molteno, 2000; Francescoli, 2000). As scent cues persist in the habitat in the absence of the sender, it is not required that both the sender and the receptor be active at the same time (Francescoli, 2000). Up to the present, many studies support the importance of chemical communication in subterranean rodents, in the context of conspecifics and through mate recognition (e.g., *Nannospalax ehrenbergi*, Heth, Nevo, & Todrank, 1996; Zuri, Gazit, & Terkel, 1997; *Heterocephalus glaber*, Clarke & Faulkes, 1999; *Fukomys anelli*, Bappert, Burda, & Begall, 2012). Nonetheless, female assessment of potential mates using scent marks as signals of male competitive territorial ability has been unexplored.

The subterranean rodent *Ctenomys talarum* is an excellent model to study the importance of male scent-marking on female preferences. This solitary species aggressively defends its individual territory (Zenuto, Vasallo, & Busch, 2002). The mating system is polygynous (Zenuto, Lacey, & Busch, 1999a): males do not participate in parental care (Zenuto et al., 2002), sexual dimorphism exists, males are larger than females (Zenuto, Malizia, & Busch, 1999b), and female-biased sex ratios in adults but not in juveniles were found in populations with high densities, which could result—at least partly—in male–male aggression (Busch, Malizia, Scaglia, & Reig, 1989). In captive conditions, male dominance status is established during aggressive encounters, sometimes involving physical injuries (Zenuto et al., 2002). The dominant male deters others and utters its characteristic territorial vocalization (tuc-tuc); it finally scent marks (using urine, faeces and anogenital rubbing) the subordinated male burrow, who is forced to leave the territory (Zenuto et al., 2002). Male dominance is strongly related to the access to females (Zenuto et al., 2002). The use of chemical signals is conspicuous, and it has been reported in different contexts: territorial marking and defence (Zenuto, 2010; Zenuto et al., 2002), reproduction (Fanjul & Zenuto, 2008; Zenuto et al., 2002) and female mate choice (Fanjul & Zenuto, 2013, 2017; Zenuto, Estavillo, & Fanjul, 2007). Indeed, tuco-tucos advertise individual identity, gender and reproductive status by means of chemical cues contained in urine, faeces and glandular secretions (Zenuto & Fanjul, 2002; Zenuto, Fanjul, & Busch, 2004; Zenuto, Vasallo, & Busch, 2001). Next to burrow entrances, it is easy to detect substrate mounds heavily mixed with faeces and small pieces of grasses that may allow individuals to evaluate burrow owners from the surface (Zenuto, personal observation). Courtship involves individual assessment by body contact, vocalization and olfaction, particularly involving the progressive assessment of scents left in the substrate, then body inspection and, finally, anogenital area sniffing (Zenuto et al., 2002). Female mate choice includes the assessment of different male traits; females prefer "novel" (Fanjul & Zenuto, 2013; Zenuto et al., 2007), dominant males (Fanjul & Zenuto, 2017) and those bearing specific MHC alleles (Cutrera, Fanjul, & Zenuto, 2012).

The above-mentioned traits highlight the importance of territoriality for *C. talarum*, offering an excellent opportunity to evaluate

the effects of male exclusive scent-marking on female assessment of potential partners. Particularly, we aimed to evaluate whether females are able to discriminate male territorial competitive ability to maintain an exclusive territory and to show preferences accordingly. The male territorial competitive ability was evaluated by means of the scent marks available in the territory, whether exclusively marked by the owner or countermarked by another male. As such condition is considered indicative of male competitive ability, we predicted that females would prefer males that hold exclusively marked territories. To assess this, we used wild caught individuals of *C. talarum*, manipulated their scent marks within the territories in captive conditions and staged preference tests using odours from a pair of males (those with invaded or exclusive territories). Females were then able to choose between exclusive territories (those containing only scent marks from the owner within a territory) and invaded territories (those containing scent marks from the owner plus an intruder within a territory) in a device where males were present but confined. In this study, we used wild animals; hence, natural variation in several conditions such as parasite load, dominance status, genetic profile and health was expected between males. To avoid possible bias of female preference based on other qualities rather than the presence or absence of intruder countermarks, males were used twice, once as owners of invaded and once as owners of exclusively marked territories. Also, we evaluated whether the identity of the intruder's scent mark affects the response of a female towards a territory; for this, we assembled territories invaded by an unknown male and territories invaded by a neighbour male. Based on the assumption that for territorial males, unknown males pose greater threat than territory-holding neighbours (Zenuto, 2010), females are expected to use the identity of the intruders to assess male competitive abilities.

## 2 | MATERIAL AND METHODS

### 2.1 | Animal capture and housing conditions

We used live traps to capture mature *C. talarum* (tuco-tucos) individuals at Mar de Cobo, Buenos Aires province, Argentina, (37°45'S, 57°26'W). Tube-shaped traps were inserted into animal's burrow systems showing fresh surface mounds indicative of recent excavatory activity. Mature females (Malizia & Busch, 1997) were captured during their nonbreeding season (March to May) to avoid the influence of previous reproductive activity, while males were captured during the reproductive season (June to December). We transported all animals to the animal laboratory, where each tuco-tuco was individually housed in a plastic cage (42 × 34 × 26 cm) with a wire-mesh lid and wood shavings for bedding. We fed them daily with fresh food (carrots, sweet potatoes, catalogna chicory, corn, mixed grasses and sunflower seeds) ad libitum to secure water provision, as *C. talarum* does not drink free water. All individuals were maintained in the same animal room where temperature and photoperiod were automatically controlled (25 ± 1°C; nonbreeding 12L: 12D; breeding 14L: 10D). Experiments were carried out during the breeding season from

11 a.m. to 16 p.m., as *C. talarum* individuals show an asynchronous and arrhythmic activity pattern, both in laboratory and field conditions (Cutrera, Antenucci, Mora, & Vassallo, 2006; Luna, Antinuchi, & Busch, 2000). At the end of the experiments the animals were returned to their site of capture. We used disposable gloves in all instances of sample collection and during the experimental trials. All equipment used during the study was washed with tap water and odourless glassware cleaner, wiped with 95% EtOH and allowed to air dry to ensure that no trace odours from previous trials remained. We employed a total of 44 females (average time of residence in the laboratory was 3 months) and 60 males (average time of residence in the laboratory was 2 weeks). We performed a total of 44 trials. We used 44 females, 22 pairs of males that were used twice (each male once as an owner and once as an intruder), plus 16 males used only as scent donors.

### 2.2 | Experimental design

Our experimental design involved consecutive preference tests of the same females at three signalling scenarios. (a) We tested female preference for exclusive or invaded, but otherwise equal, territories (owners temporarily removed). After an hour of territory exploration and recognition, (b) we tested female preference for the scent samples of both territory owners. (c) Finally, preference tests involved both males, each one confined within its corresponding territory (exclusive or invaded) sensu Rich and Hurst (1998). We used two different types of scent invasion: in a first experiment, artificial invasion of the territory was made with scents of a third unknown male, and in a second experiment, we used the scent of the male owner of the exclusive territory, as may occur more frequently among neighbours.

### 2.3 | Establishment of male territories

Prior to preference tests, each individual—one female and two males—was placed individually in a Perspex box with freshly scented bedding (soiled shavings) from its own home cage, and left to habituate for 1 hr before the test began (Zenuto & Fanjul, 2002). Bedding in each home cage was changed each week in a manner that soiled shavings carried recently deposited (fresh) scents and others deposited at most a week ago. Then each male was temporarily removed and two different types of territories were established: exclusive and invaded ones. The exclusive territory remained with no manipulation, exclusively containing the owners' odour. We artificially scent marked the invaded territory with odours from another male. We staged this artificial scent-countermarking by placing a sample (200 cm<sup>3</sup> approximately) of fresh soiled shavings with urine, faeces and bodily secretions obtained from the current home cage of another male (which inhabited its cage for at least 7 days). In this manner, we avoided any modification that a natural countermarking may contain, such as signals derived of dominance, stress or anxiety of an individual exposed to scents from a competitor (Rich & Hurst, 1998). Male pairs did not differ in more than 10% of body mass and we assured that males within pairs were unfamiliar with each other

(distance between their respective sites of captures had to exceed 50 m; Zenuto, 2010). To control for possible individual differences in attractiveness, males within pairs were also used in their reversed roles of owners of invaded and exclusive territories (Rich & Hurst, 1998).

To evaluate whether the ecological identity (thereafter identity) of the countermarking male affects female response, we staged two different types of invaded territories. In the first experiment ( $n = 26$ ), our countermark scent was a sample from a third unknown male, and in the second experiment ( $n = 18$ ), we used a scent sample from the neighbouring male (i.e., the owner of the exclusive territory).

## 2.4 | Female preference test

To induce female receptivity, 24 hr before the beginning of the trials, females were injected intraperitoneally with 10  $\mu\text{g}/100$  g liveweight of water-soluble estradiol benzoate (Estradiol 10, Rio de Janeiro Laboratories) as indicated for induced ovulator species (Bakker & Baum, 2000).

Our experimental apparatus consisted of Perspex boxes (45 cm  $\times$  30 cm  $\times$  30 cm) and Y tubes (tubes of 10 cm diameter and 25 cm length each arm) with sliding doors that strategically allowed or prevented the entrance of the female to the testing target at each trial phase (see Figure 1 for details). During the first phase, females explored exclusive and invaded territories for an hour. We recorded female interest in each territory as: the total time the female spent exploring each territory (*time*), frequency in which females visited each territory (*visits*) and the frequency of sniffing the substrate (*sniff shavings*). After that, and when the female returned to its box, we initiated phase 2 (closing door #1 and opening door #2, Figure 1) where the scents of each territory owner were presented in a Y-maze. Scent samples consisted of Petri dishes filled with male odour-saturated soiled shavings and covered by a wire mesh with a central hole (this allowed females to come in contact with the scent sample and thus perceive volatile and nonvolatile compounds, please see Zenuto et al., 2004 for further details) placed as an end cap of each Y-maze arm (Fanjul & Zenuto, 2013). Female interest in each scent sample was registered during 7 min test period as: the total time the female spent at each arm (*time*), the frequency of visits to each arm (*visits*) and the frequency in which it scratched the mesh of scent samples (*scratch mesh*). Finally, during the third phase, females were allowed to explore the territories with the owner present in it by opening door #1 and closing door # 2 (Figure 1). Males remained confined within each territory in a Perspex tube, separated by a wire mesh, allowing females to use chemical, vocal and visual communication channels to evaluate them but avoiding further scent-marking and female coercion (Fanjul & Zenuto, 2013). We registered the interest of the female in each territory and its owners during 15 min test period as: the total time the female spent exploring each territory (*time*), frequency in which females visited each territory (*visits*), frequency scratching the mesh of each male confinement (*scratch mesh*), sniffing each male (sniff male) and exposing her rump towards each male (*expose rump*). All trials were recorded using a digital HD

Handycam (Sony HDR-XR100) and evaluated later. The experimental sequence designed resemble a natural condition for a highly territorial and aggressive species like *C. talarum* where a female is likely to obtain information from odours left in the substrate near the burrows previously to enter in contact with individuals. Thus, this protocol allows us to better understand how information gathered from the environment would influence female preferences and decisions.

At each level of assessment (phase), we compared the interest of a female in the exclusive vs. the invaded territory/owner scent using Paired-sample *t* test or its nonparametric analogue if assumptions were not met (Zar, 2010). Results were reported as means  $\pm$  SD. In all cases, the critical significance level was set at  $p \leq 0.05$ . Along with the exact *p*-values, we reported the observed effect sizes according to recommendation for behavioural sciences (Nakagawa, 2004; Stoehr, 1999). Effect sizes are "a scale-free value that measure, in terms appropriate to it, the discrepancy between H0 and the H1" (Cohen, 1992). We calculated effect size, standardized Cohen's  $d_z$ , using the free program GPower 3.1.1 (by F. Faul, Universität Kiel, Germany; <http://www.psych.uni-duesseldorf.de/abteilungen/aap/gpower3/>) (McDonald, 2014).

## 2.5 | Ethical note

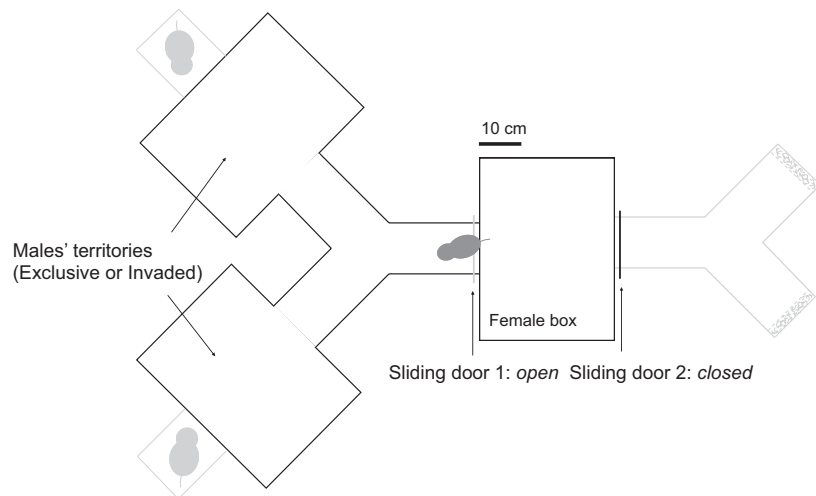
We adhered to the 2012 Revised International Guiding Principles for Biomedical Research Involving Animals developed by the Council for International Organizations of Medical Sciences (CIOMS) and the International Council for Laboratory Animal Science (ICLAS).

## 3 | RESULTS

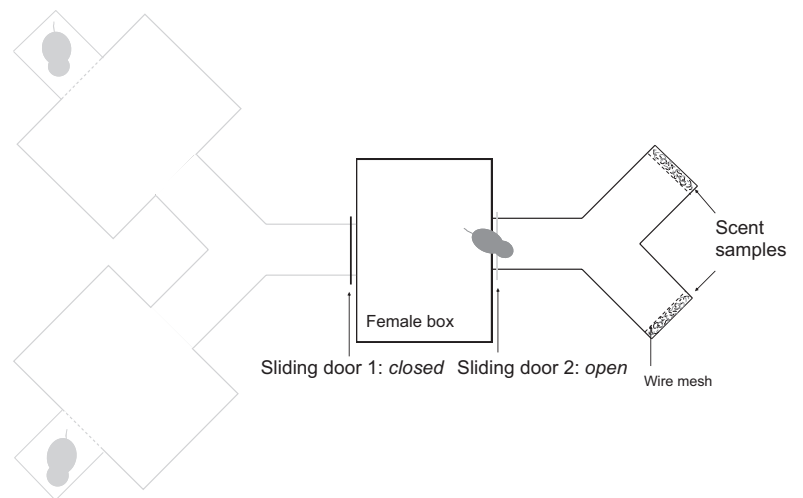
### 3.1 | Invasion by a strange male

In the first phase, females stayed in both territories a similar amount of time (Paired *t* test  $t_{25} = 1.276$ ,  $p = 0.743$ , Cohen's  $d_z = 0.422$ ; Figure 2a) and visited both territories at a similar frequency ( $t_{25} = -0.237$ ,  $p = 0.814$ , Cohen's  $d_z = 0.046$ ; Figure 2b). However, females investigated chemical cues deposited on the floor of the exclusively marked territory more frequently than in that invaded by a strange male (Wilcoxon matched-pairs test:  $Z_{25} = 2.490$ ,  $p = 0.013$ , Cohen's  $d_z = 0.434$ ; Figure 2b). During the second phase, females displayed equal interest to scent samples of both males: the owners of exclusive and invaded territories (time  $Z_{25} = -0.381$ ,  $p = 0.713$ , Cohen's  $d_z = 0.610$ ; visits,  $Z_{25} = -0.618$ ,  $p = 0.551$ , Cohen's  $d_z = 0.116$ ; Scratch mesh,  $Z_{25} = -0.390$ ,  $p = 0.706$ , Cohen's  $d_z = 0.294$ ; Figure 2c,d). In the later phase (Figure 2e,f), when females explored the territories in the presence of the owner, they spent more time in the invaded territories (Figure 2e, Paired *t* test  $t_{25} = 2.460$ ,  $p = 0.021$ , Cohen's  $d_z = 0.481$ ) despite they visited both with the same frequency (Wilcoxon matched-pairs test:  $Z_{25} = -1.648$ ,  $p = 0.103$ , Cohen's  $d_z = 0.386$ ). Females sniffed the males ( $t_{25} = 1.950$ ,  $p = 0.062$ , Cohen's  $d_z = 0.382$ ) and exposed their rump ( $Z_{25} = -1.802$ ,  $p = 0.094$ , Cohen's  $d_z = 0.4984$ ) slightly more frequently to the owner of the invaded territories, but no statistical

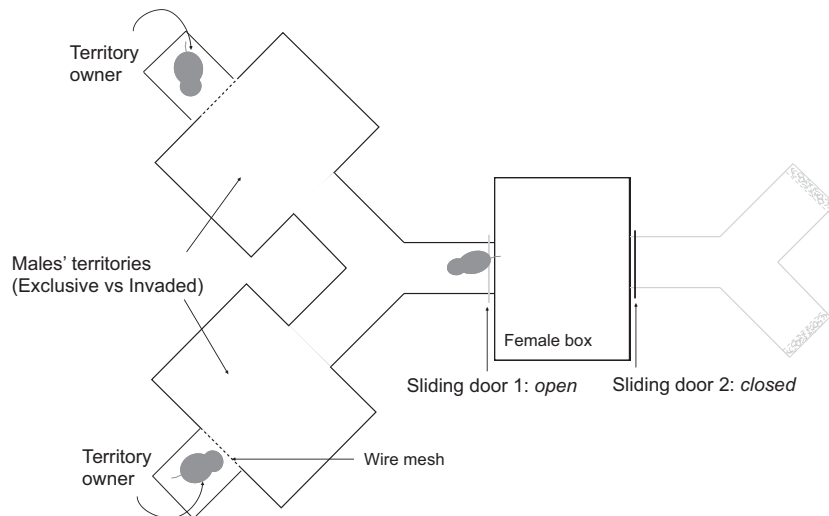
### Phase 1: Exploration and preference of territories (1 hour)



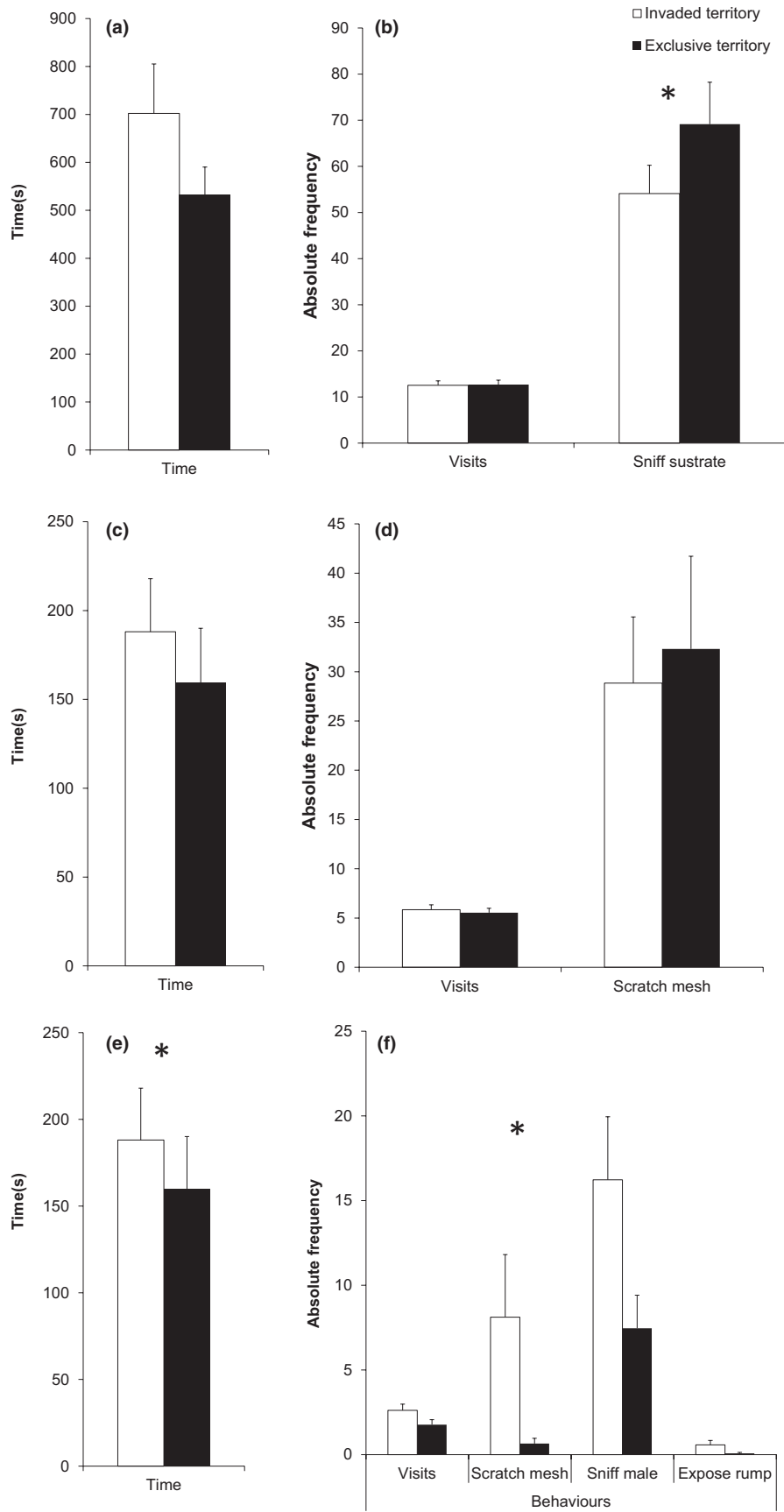
### Phase 2: Preference of male owner's scent (7 min)



### Phase 3: Preference of territories + owners present



**FIGURE 1** Schematic representation of the experimental apparatus used in female preference trials for exclusive and invaded conditions involving three consecutive levels of assessment. Phase 1: Scent-marked territories with male owners temporarily removed. Phase 2: Male odours, consisting of shavings soiled with urine, faeces and presumably other bodily secretions collected from males' housing cages. Phase 3: Territories with male owners confined through a wire mesh. Grey drawn sections represent areas banned to females at a given phase



**FIGURE 2** Interest devoted by tuco-tuco females towards territories scent marked exclusively (black bars) or invaded (white bars) with a strange male scent (Treatment 1). Time (a) and frequency (b) of behaviours recorded during exploration of each territory. Time (c) and frequency (d) of behaviours displayed during evaluation of owner scent samples. Time (e) and frequency (f) of behaviours displayed during exploration of each territory with owners presence. Data are shown as mean ( $\pm$ SE), \* $p < 0.05$ , Paired  $t$  tests or its nonparametric equivalent (Wilcoxon matched-pair tests) if assumptions were not met ( $n = 26$ )

differences were detected. Also, females tried to get in contact with the owner of the invaded territory by scratching the mesh more frequently ( $Z_{25} = -2.581$ ,  $p = 0.007$ , Cohen's  $d_z = 0.650$ ).

### 3.2 | Invasion by neighbouring male

During the first phase, females visited, smelled the substrate and stayed a similar amount of time in each territory (Time: Paired  $t$  test,  $t_{17} = 0.912$ ,  $p = 0.375$ , Cohen's  $d_z = 0.214$ ; visits: Wilcoxon matched-pair test:  $Z_{17} = -0.350$ ,  $p = 0.735$ , Cohen's  $d_z = 0.09$ ; Sniff shavings:  $t_{17} = -1.702$ ,  $p = 0.107$ , Cohen's  $d_z = 0.401$ ; Figure 3a,b). Later, in the second phase, females explored with same interest the scent samples of both males (time,  $t_{16} = -0.346$ ,  $p = 0.734$ , Cohen's  $d_z = 0.083$ ; visits,  $Z_{16} = -1.116$ ,  $p = 0.273 = 0.259$ ; Scratch mesh,  $t_{16} = -0.631$ ,  $p = 0.537$ , Cohen's  $d_z = 0.152$ ; Figure 3c,d). In the last phase, when females explored both territories in the presence of the owners of the territory, females did not show a preference for a territories, neither in time ( $t_{17} = -0.087$ ,  $p = 0.932$ , Cohen's  $d_z = 0.020$ ; Figure 3e) nor in frequencies (visits:  $Z_{17} = 1.262$ ,  $p = 0.217$ , Cohen's  $d_z = 0.275$ ; sniff male:  $t_{17} = -0.123$ ,  $p = 0.904$ , Cohen's  $d_z = 0.028$ ; expose rump:  $Z_{17} = 0.00$ , Cohen's  $d_z = 0.065$ ,  $p = 1.000$ , scratch mesh:  $Z_{17} = 0.028$ ,  $p = 0.978$ , Cohen's  $d_z = 0.059$ ; Figure 3f).

### 3.3 | Intruder's identity

Finally, we also compared females' responses between treatments, that is, responses to neighbour-invaded territories vs. to stranger-invaded territories. Females displayed the same interest to both types of invaded territories in all three phases evaluated. When exploring territories (Phase 1) females displayed similar proportion of visits ( $t$  test,  $t_{42} = -0.410$ ,  $p = 0.684$ , Cohen's  $d_z = 0.126$ ), time devoted to exploration (Mann-Whitney  $U_{(18, 26)} = 224$ ,  $p = 0.821$ , Cohen's  $d_z = 0.501$ ) and/or sniffing shavings ( $t_{42} = -0.078$ ,  $p = 0.938$ , Cohen's  $d_z = 0.023$ ) in both types of the invaded territories. During the evaluation of male scents (Phase 2), females also displayed the same proportion of visits ( $t_{41} = -0.131$ ,  $p = 0.896$ , Cohen's  $d_z = 0.041$ ), time exploring ( $U_{(17, 26)} = 209$ ,  $p = 0.775$ , Cohen's  $d_z = 0.213$ ) and scratching the mesh ( $U_{(17, 26)} = 183$ ,  $500$ ,  $p = 0.411$ , Cohen's  $d_z = 0.197$ ) in both types of invaded territories. During the last phase, for the evaluation of territories with the presence of the owner, there were no differences either (time:  $U_{(18, 26)} = 183,500$ ,  $p = 0.231$ , Cohen's  $d_z = 0.383$  sniff male:  $U_{(17, 24)} = 185$ ,  $500$ ,  $p = 0.626$ , Cohen's  $d_z = 0.63$ ), but the proportion of certain behaviours showed a slightly nonsignificant tendency: visits ( $t_{42} = 1.957$ ,  $p = 0.057$ , Cohen's  $d_z = 0.593$ ), exposure of rump ( $U_{(7, 11)} = 24$ ,  $500$ ,  $p = 0.189$ , Cohen's  $d_z = 0.644$ ) and scratching of mesh ( $U_{(14, 15)} = 82$ ,  $p = 0.289$ , Cohen's  $d_z = 0.436$ ).

## 4 | DISCUSSION

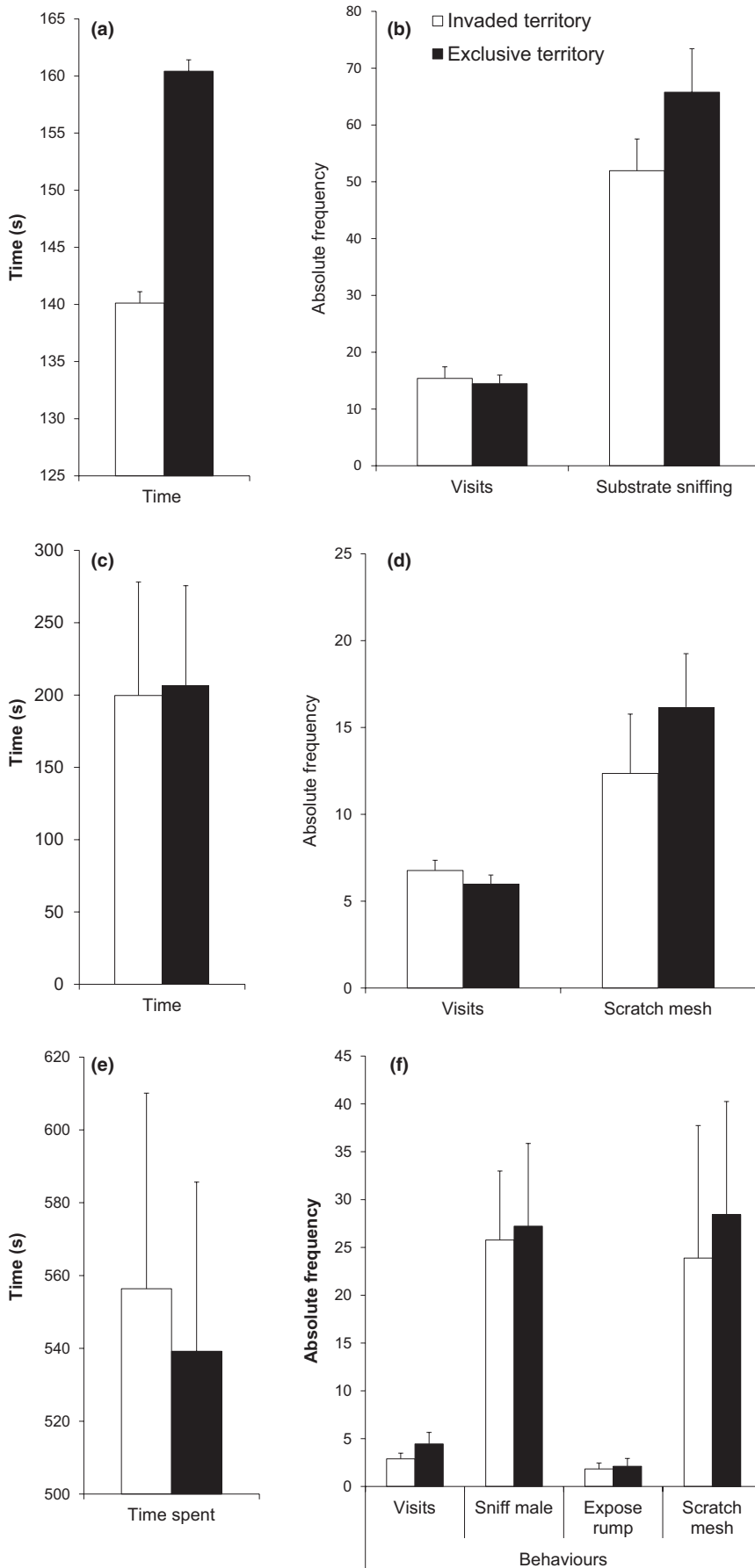
Females of *C. talarum* use information about the homogeneity of scent marks within a male territory, preferring the exclusively marked ones. But when the male owner is present in its scent-invaded

territory, visual and auditory signals may be used by the female to reevaluate the initial information gathered, changing its response accordingly. Moreover, females recognize the identity of the intruder's scent—stranger or neighbour—and respond differentially, both to the scent-marked territories and to males in relation to this condition. To accomplish the discrimination of male competitive relationships, females use multiple cues and then exert their preferences. Despite most studies consider only one characteristic, the evaluation of the quality of a potential partner is of such importance that the use of multiple cues is required (Künzler & Bakker, 2001). Individuals rely on signals and cues that additively reinforce each other, increasing the amount and reliability of the recorded information (Candolin & Reynolds, 2001).

### 4.1 | If the intruder is an strange male

During the first phase, when females explored male territories, they devoted more time investigating exclusively marked ones. Such investigatory bias in females of many species are reported to correspond to their actual mating preferences (e.g., Egid & Brown 1989) including tuco-tucos (Fanjul & Zenuto, 2013, 2017). Preference for the exclusively marked territory is in agreement with our initial prediction and supported by other studies (e.g., Hurst, 1993; Thomas, 2002; Thonhauser, Raveh, Hetttyey, Beissmann, & Penn, 2013). A male that owns a territory and is capable of preventing a competitor to trespass and deposit a different scent would account for its superior competitive abilities (Gosling & Roberts, 2001; Hurst & Rich, 1999). In some cases, this competitive ability to hold a high-quality territory give back some direct benefits to the progeny -like food resources or proximity to mates. In others, it would result in genetic benefits if sons inherit their father's competitive abilities to defend the territory (Wong & Candolin, 2005). For polygynous rodents with no paternal care, like *C. talarum*, females are expected to increase their fitness by choosing males that provide these genetic benefits. In tuco-tucos, territorial establishment and defence are crucial to males in order to successfully monopolize females. The performance in such critical task is affected by male fighting ability and strength (Becerra, Echeverría, Casinos, & Vassallo, 2012; Zenuto et al., 2002) and territorial scent-marking (Zenuto, 2010). Experimental evidence obtained in laboratory enclosures resembling the natural social environment conditions, showed that females of *C. talarum* mate exclusively with the dominant male (Zenuto et al., 2002). This occurred after a male established its territory and dominance condition by fighting and threatening the other males in the enclosure, effectively imposing and precluding any access of other males to females (Zenuto et al., 2002). Indeed, even when females were free to choose between different males, they preferred such territorial and dominant males (Fanjul & Zenuto, 2017). Thus, the obtained evidence indicates that holding a territory would be an important proxy of male quality that females will evaluate.

When only scents of males holding exclusive or invaded territories were available, females did not display any preference. Tuco-tucos rely on chemical signals to convey information about



**FIGURE 3** Interest devoted by tuco-tuco females towards territories scent marked exclusively (black bars) or invaded (with bars) by a neighbour male (treatment 2). Time (a) and frequency (b) of behaviours recorded during exploration of each territory. Time (c) and frequency (d) of behaviours displayed during evaluation of owner scent samples; and time (e) and frequency (f) of behaviours displayed during exploration of each territory with owners' presence. Data are shown as mean ( $\pm$ SE), \* $p < 0.05$ , Paired  $t$  tests or its nonparametric equivalent (Wilcoxon matched-pair tests) if assumptions were not met ( $n = 18$ )



individuality, gender and reproductive status (Zenuto & Fanjul, 2002; Zenuto et al., 2001, 2004). However, this result does not imply that chemical signals were not enough to offer territorial information to females, as in the previous phase, they showed more interest for exclusively marked territories. Possibly, the scents of the owners of the territories does not stimulate the females to display a preference, or females only recognize an exclusive or invaded territory but were not able to attribute such condition to the males (by means of its odours) that hold those territories. Chemical signals can convey territorial dominance information in two ways: the composition of the scent itself (e.g., Jones and Nowell 1973; Novotny, Harvey, & Jemiolo, 1990) and/or the spatial and temporal pattern of scent deposition in the environment (e.g., Johnston et al., 1997; Rich & Hurst, 1998). We explored the first one in the present study (second phase of experiments) and in a previous study (Fanjul & Zenuto, 2017), and in both cases, we failed to find any preference when only scents were available for females. Thus, tuco-tucos may use the pattern of scent-marking to convey territorial dominance, as it can be observed during territorial preference in the first and third phase of experiments, that is, when cues are deposited on the substrate. Chemical assessment of territorial dominance and competitive ability appear to be more linked to the frequency of scent-marking, the spatiotemporal pattern of deposition and the presence of a competitor's scent mark (Wong & Candolin, 2005). Moreover, the "multiple sensory environments" hypothesis addresses the situation where the signal and reception are influenced by environmental conditions (Candolin, 2003; Hebets & Papaj, 2005) predicting that the receiver response will vary through different environmental contexts. This seems to be the case of *C. talarum* territorial scent-marking: the information about the territoriality status is not only conveyed as a pattern but also through an appropriate ecological context.

At last, and in contrast to the territory preference phase, when females choose between territories with the owners present in the box, they prefer males with scent-invaded territories. As aggression is tightly related to dominance and territoriality (Qvarnström & Forsgren, 1998), we would firstly hypothesize that female tuco-tucos could be avoiding highly aggressive territorial males (exclusive territory owners) to lower the risk of injuries during courtship and mating. This was also reported for other species, where females would prefer subordinated, less aggressive, males to obtain safe mating (Ophir & Galef, 2003). However, this idea cannot be supported by the fact that females of *C. talarum* prefer dominant males (Fanjul & Zenuto, 2017). In the wild, competitive interactions would occur on a regular basis and individuals of higher competitive ability would be able to deter and countermark intrusions of low quality males. This information can be used by competitors to decide whether to challenge the owner or to flee (Gosling, 1982). But also, it can be used by females to evaluate the suitability of a male, as the Competitive Countermark Hypothesis (CCH) highlights (Rich & Hurst, 1999). According to that, females would prefer a countermarked territory as they would prefer to mate males of proven competitive abilities (Rich & Hurst, 1999). Then, we propose that the presence of the male in an invaded territory would act as a countermark and then

interpreted as a handicap (Zahavi, 2008), a noncheatable proof of its competitive ability. In the present study, females would prefer males with proven abilities to maintain their own territories and repel intrusions. Such preference emphasizes once again the central role of dominance (Fanjul & Zenuto, 2017) and territory holding on male reproductive success.

#### 4.2 | If the intruder were the neighbour male

In contrast, in the second experiment, when the invasion corresponded to a neighbour's scent, females of tuco-tucos did not show any preference for the territories or males. According to the experimental design, we originally hypothesized that the neighbouring male would have a double gain: he exclusively owned a territory and could also mark in the neighbour's territory. This led us to expect that females would display a stronger preference for those males that exclusively own a territory and challenge the neighbour. The Dear Enemy Phenomenon (DEP) explains the buffered display of agonistic interactions between individuals that recognized each other as neighbours by the deposition of territorial scent marks (Temeles, 1994). In other words, neighbours would be less aggressive to each other as they pose a lower threat than wandering intruders that could be searching for a territory to settle. Evidence is abundant for such relationships (e.g., Monclús, Saavedra, & de Miguel, 2014); in tuco-tucos the DEP seems to play an important role modulating male competitive relationships (Zenuto, 2010). In the wild, the burrows of tuco-tucos are multipurpose territories as they allow to reach food resources and to find mating partners (Busch et al., 1989; Zenuto et al., 1999a). Thus, the outcome of male competition to gain access to a particular/central territory directly affects mating opportunities. In this sense, the recognition of familiar scents (Zenuto & Fanjul, 2002), that is, strange from neighbour, is critical for male-male competition (Zenuto, 2010) but also for female mate choice (Fanjul & Zenuto, 2013). It is widely known that females rely on odour cues to assess potential partners (Hurst, 2009; Penn & Potts 1998; Potts, Manning, & Wakeland, 1991). Indeed females assess male competition by means of countermarking (Wong & Candolin, 2005), as it constitutes a suitable proxy of quality for mates. The pattern of scent marks among neighbours, in a context of DEP, is also a source of information to be investigated by females. Given that neighbours do not represent a great risk for an owner, the ability to prevent access to their territory would not be a signal of quality for females. Our results then underline the importance of male competitive status by scent-marking and female recognition of the identity of challenging marks during male assessment. Thus, the recognition and evaluation of the identity on scent-countermarking, belonging to a neighbour or stranger male, seems to play an important role for females of *C. talarum*.

As the costs of competitive interactions are significant for males of *C. talarum* in regard of energy expenditure and the associated risk of losing the hierarchy and/or the territory (Zenuto, 2010; Zenuto et al., 2002), females would ensure gathering multiple honest signals to recognize high-quality potential mates and maximize progeny

success. Thus, female tuco-tucos do not only use the information about the presence of the owner and the exclusiveness of scent marks within a territory, but also recognize the identity – neighbour or stranger- of the intruder's scent. In conclusion, male–male competition can have profound effects on mate evaluation; females investigate male competitive cues to ensure mating with the best partner. For females of *C. talarum*, this means males with better genetic constitution, which is related to health (Cutrera et al., 2012), dominance (Fanjul & Zenuto, 2017) and territorial abilities (this study). A better understanding of how male competition affects females mate preference is likely to provide new insights about the complex multi-signalling process of assessing potential partners.

## ACKNOWLEDGEMENTS

We want to express our gratitude to the members of the Physiological and Behavioural Ecology Laboratory for their help in field and laboratory work. This work was supported by the CONICET [PIP 0272 granted to Roxana R. Zenuto] and FONCYT [PICT 0998 granted to C.D. Antenucci].

## ORCID

María Sol Fanjul  <http://orcid.org/0000-0003-1890-8183>

## REFERENCES

- Andersson, M. (1994). *Sexual selection*. Princeton, NJ: Princeton University Press.
- Arakawa, H., Blanchard, D. C., Arakawa, K., Dunlap, C., & Blanchard, R. J. (2008). Scent marking behavior as an odorant communication in mice. *Neuroscience and Biobehavioral Reviews*, 32, 1236–1248. <https://doi.org/10.1016/j.neubiorev.2008.05.012>
- Bakker, J., & Baum, M. J. (2000). Neuroendocrine regulation of GnRH release in induced ovulators. *Frontiers in Neuroendocrinology*, 21(3), 220–262. <https://doi.org/10.1006/frne.2000.0198>
- Bappert, M. T., Burda, H., & Begall, S. (2012). To mate or not to mate? Mate preference and fidelity in monogamous Ansell's mole-rats, *Fukomys ansellii*, Bathyergidae. *Folia Zoologica*, 61(Suppl), 71–83. <https://doi.org/10.25225/fozo.v61.i1.a11.2012>
- Becerra, F., Echeverría, A. I., Casinos, A., & Vassallo, A. I. (2012). Sexual selection in a polygynous rodent (*Ctenomys talarum*) – an analysis of fighting capacity. *Journal of Zoology*, 115, 405–410. <https://doi.org/10.1016/j.zool.2012.05.002>
- Bennett, N. C., Faulkes, C. G., & Molteno, A. J. (2000). Reproduction in subterranean rodents. In E. A. Lacey, J. L. Patton, & G. N. Cameron (Eds.), *Life underground: The biology of subterranean rodents* (pp. 145–177). Chicago, IL: University of Chicago Press.
- Blaustein, A. R. (1981). Sexual selection and mammalian olfaction. *American Naturalist*, 117, 1006–1010. <https://doi.org/10.1086/283786>
- Busch, C., Malizia, A. I., Scaglia, O. A., & Reig, O. A. (1989). Spatial distribution and attributes of a population of *Ctenomys talarum* (Rodentia: Octodontidae). *Journal of Mammalogy*, 70(1), 204–208. <https://doi.org/10.2307/1381691>
- Candolin, U. (2003). The use of multiple cues in mate choice. *Biological Reviews*, 78, 575–595. <https://doi.org/10.1017/S1464793103006158>
- Candolin, U., & Reynolds, J. D. (2001). Sexual signaling in the European bitterling: Females learn the truth by direct inspection of the resource. *Behavioral Ecology*, 12, 407–411. <https://doi.org/10.1093/beheco/12.4.407>
- Clarke, F. M., & Faulkes, C. G. (1999). Kin discrimination and female mate choice in the naked mole rat *Heterocephalus glaber*. *Proceedings of the Royal Society B: Biological Sciences*, 266, 1995–2002. <https://doi.org/10.1098/rspb.1999.0877>
- Clutton-Brock, T., & McAuliffe, K. (2009). Female mate choice in mammals. *The Quarterly Review of Biology*, 84, 3–27. <https://doi.org/10.1086/596461>
- Cohen, J. (1992). A power primer. *Psychological Bulletin*, 112, 155–159. <https://doi.org/10.1037/0033-2909.112.1.155>
- Cutrera, A. P., Antenucci, C. D., Mora, M. S., & Vassallo, A. I. (2006). Home-range and daily movements of the South American subterranean rodent *Ctenomys talarum*. *Journal of Mammalogy*, 87, 1183–1192. <https://doi.org/10.1644/05-MAMM-A-386R1.1>
- Cutrera, A. P., Fanjul, M. S., & Zenuto, R. R. (2012). Females prefer good genes: MHC-associated mate choice in wild and captive tuco-tucos. *Animal Behaviour*, 83, 847–856. <https://doi.org/10.1016/j.anbehav.2012.01.006>
- Egid, K., & Brown, J. L. (1989). The MHC and female mating preferences in mice. *Animal Behaviour*, 38, 548–550. [https://doi.org/10.1016/S0003-3472\(89\)80051-X](https://doi.org/10.1016/S0003-3472(89)80051-X)
- Eisenberg, J. F., & Kleiman, D. G. (1972). Olfactory communication in mammals. *Annual Review of Ecology and Systematics*, 3, 1–32. <https://doi.org/10.1146/annurev.es.03.110172.000245>
- Fanjul, M. S., & Zenuto, R. R. (2008). Copulatory pattern of the subterranean rodent *Ctenomys talarum*. *Mammalia*, 72(2), 102–108.
- Fanjul, M. S., & Zenuto, R. R. (2013). When allowed, females prefer novel males in the polygynous subterranean rodent *Ctenomys talarum* (tuco-tucos). *Behavioural Processes*, 92, 71–78. <https://doi.org/10.1016/j.beproc.2012.10.010>
- Fanjul, M. S., & Zenuto, R. R. (2017). Female choice, male dominance and condition-related traits in the polygynous subterranean rodent *Ctenomys talarum*. *Behavioural Processes*, 142, 46–55. <https://doi.org/10.1016/j.beproc.2017.05.019>
- Ferkin, M. H. (1999). Scent over-marking and adjacent-marking as competitive tactics used during chemical communication in voles. In R. E. Johnston, D. Müller-Schwarze, & P. W. Sorensen (Eds.), *Advances in chemical signals in vertebrates* (pp. 239–246). Boston, MA: Springer. <https://doi.org/10.1007/978-1-4615-4733-4>
- Ferkin, M. H., Sorokin, E. S., Johnston, R. E., & Lee, C. J. (1997). Attractiveness of scents varies with protein content of the diet in meadow voles. *Animal Behavior*, 53, 133–141. <https://doi.org/10.1006/anbe.1996.0284>
- Fisher, H. S., Swaisgood, R. R., & Fitch-Snyder, H. (2003). Odor familiarity and female preferences for males in a threatened primate, the pygmy loris *Nycticebus pygmaeus*: Applications for genetic management of small populations. *Naturwissenschaften*, 90, 509–512. <https://doi.org/10.1007/s00114-003-0465-9>
- Francescoli, G. (2000). Sensory capabilities and communication in subterranean rodents. In E. A. Lacey, J. L. Patton, & G. N. Cameron (Eds.), *Life underground: The biology of subterranean rodents* (pp. 111–144). Chicago, IL: The University of Chicago Press.
- Gosling, L. M. (1982). A reassessment of the function of scent marking in territories. *Zeitschrift für Tierpsychologie*, 60, 89–118. <https://doi.org/10.1111/j.1439-0310.1982.tb00492.x>
- Gosling, L. M. (1990). Scent marking by resource holders: Alternative mechanisms of advertising the cost of competition. In D. W. Macdonald, D. Müller-Schwarze, & S. E. Natynczuk (Eds.), *Chemical signals in vertebrates 5* (pp. 315–328). Oxford, UK: Oxford University Press.
- Gosling, L. M., & Roberts, S. C. (2001). Scent-marking by male mammals: Cheat-proof signals to competitors and mates. *Advances in the Study of Behavior*, 30, 169–217. [https://doi.org/10.1016/S0065-3454\(01\)80007-3](https://doi.org/10.1016/S0065-3454(01)80007-3)

- Hebets, E. A., & Papaj, D. R. (2005). Complex signal function: Developing a framework of testable hypotheses. *Behavioral Ecology and Sociobiology*, 57(3), 197–214. <https://doi.org/10.1007/s00265-004-0865-7>
- Heth, G., Nevo, E., & Todrank, J. (1996). Seasonal changes in urinary odors and in responses to them by subterranean mole rats. *Physiology & Behavior*, 60, 963–968. [https://doi.org/10.1016/0031-9384\(96\)00077-7](https://doi.org/10.1016/0031-9384(96)00077-7)
- Hurst, J. L. (1993). The priming effects of urine substrate marks on interaction between male house mice *Mus musculus domesticus*. *Animal Behaviour*, 40, 233–243.
- Hurst, J. L. (2009). Female recognition and assessment of males through scent. *Behavioral Brain Research*, 200, 295–303. <https://doi.org/10.1016/j.bbr.2008.12.020>
- Hurst, J. L., & Rich, T. J. (1999). Scent marks as competitive signals of mate quality. In R. E. Johnston, D. Müller-Schwarze, & P. Sorensen (Eds.), *Advances in chemical signals in vertebrates* (pp. 209–225). New York, NY: Plenum Press. <https://doi.org/10.1007/978-1-4615-4733-4>
- Hurst, J. L., Thom, M. D., Nevison, C. M., Humphries, R. E., & Beynon, R. J. (2005). The “scents” of ownership. In R. T. Mason, M. P. LeMaster, & D. Muller-Schwarze (Eds.), *Chemical signals in vertebrates 10* (pp. 199–208). New York, NY: Springer. <https://doi.org/10.1007/b106868>
- Johnston, R. E., Sorokin, E. S., & Ferkin, M. H. (1997). Female voles discriminate males’ over-marks and prefer top-scent males. *Animal Behaviour*, 54, 679–690.
- Jones, R. B., & Nowell, N. W. (1973). Aversive and aggression-promoting properties of urine from dominant and subordinate male mice. *Animal Learning and Behavior*, 1, 207–210.
- Kavaliers, M., Choleris, E., Agmo, A., & Pfaff, D. W. (2004). Olfactory-mediated parasite recognition and avoidance: Linking genes to behavior. *Hormones and Behavior*, 46, 272–283. <https://doi.org/10.1016/j.yhbeh.2004.03.005>
- Kavaliers, M., & Colwell, D. D. (1995). Discrimination by female mice between the odours of parasitized and non-parasitized males. *Proceedings of the Royal Society B: Biological Sciences*, 261, 31–35. <https://doi.org/10.1098/rspb.1995.0113>
- Künzler, R., & Bakker, T. C. M. (2001). Female preferences for single and combined traits in computer animated stickleback males. *Behavioral Ecology*, 12, 681–685. <https://doi.org/10.1093/beheco/12.6.681>
- Lacey, E. A., Patton, J. L., & Cameron, G. N. (2000). *Life underground: The biology of subterranean rodents*. Chicago, IL: University of Chicago Press.
- Luna, F., Antinuchi, C. D., & Busch, C. (2000). Rhythms of locomotor activity and burrow use under seminatural conditions in *Ctenomys talarum* (Rodentia, Octodontidae). *Revista Chilena de Historia Natural*, 73, 39–47.
- Malizia, A. I., & Busch, C. (1997). Breeding biology of the fossorial rodent *Ctenomys talarum* (Rodentia: Octodontidae). *Journal of Zoology*, 242, 463–471.
- McDonald, J. H. (2014). *Handbook of biological statistics* (3rd ed.). Baltimore, MD: Sparky House Publishing.
- Monclús, R., Saavedra, I., & de Miguel, J. (2014). Context-dependent responses to neighbours and strangers in wild European rabbits (*Oryctolagus cuniculus*). *Behavioural Processes*, 106, 17–21. <https://doi.org/10.1016/j.beproc.2014.04.004>
- Nakagawa, S. (2004). A farewell to Bonferroni: The problems of low statistical power and publication bias. *Behavioral Ecology*, 15, 1044–1045. <https://doi.org/10.1093/beheco/15.10.1044>
- Novotny, M., Harvey, S., & Jemiolo, B. (1990). Chemistry of male dominance in the house mouse, *Mus domesticus*. *Experientia*, 46(1), 109–113. <https://doi.org/10.1007/BF01955433>
- Ophir, A., & Galef, B. (2003). Female Japanese quail that “eavesdrop” on fighting males prefer losers to winners. *Animal Behaviour*, 66, 399–407. <https://doi.org/10.1006/anbe.2003.2230>
- Penn, D., & Potts, W. K. (1998). Chemical signals and parasite-mediated sexual selection. *Trends in Ecology and Evolution*, 13(10), 391–396. [https://doi.org/10.1016/S0169-5347\(98\)01473-6](https://doi.org/10.1016/S0169-5347(98)01473-6)
- Potts, W. K., Manning, C. J., & Wakeland, E. K. (1991). Mating patterns in seminatural populations of mice influenced by MHC genotype. *Nature*, 352(6336), 619–621. <https://doi.org/10.1038/352619a0>
- Qvarnström, A., & Forsgren, E. (1998). Should females prefer dominant males? *Trends in Ecology & Evolution*, 13(12), 498–501. [https://doi.org/10.1016/S0169-5347\(98\)01513-4](https://doi.org/10.1016/S0169-5347(98)01513-4)
- Rich, T. J., & Hurst, J. L. (1998). Scents marks as reliable signals of the competitive ability of mates. *Animal Behaviour*, 56, 727–735. <https://doi.org/10.1006/anbe.1998.0803>
- Rich, T. J., & Hurst, J. L. (1999). The competing countermarks hypothesis: Reliable assessment of competitive ability by potential mates. *Animal Behaviour*, 58, 1027–1037. <https://doi.org/10.1006/anbe.1999.1217>
- Stoehr, A. (1999). Are significance thresholds appropriate for the study of animal behaviour? *Animal Behaviour*, 57, F22–F25. <https://doi.org/10.1006/anbe.1998.1016>
- Tang-Halpin, Z. (1986). Individual odors among mammals: Origins and functions. *Advances in the Study of Behavior*, 16, 39–70. [https://doi.org/10.1016/S0065-3454\(08\)60187-4](https://doi.org/10.1016/S0065-3454(08)60187-4)
- Temeles, E. J. (1994). The role of neighbors in territorial systems: When are they “dear enemies”? *Animal Behaviour*, 47(2), 339–350. <https://doi.org/10.1006/anbe.1994.1047>
- Thomas, S. A. (2002). Scent marking and mate choice in the prairie vole, *Microtus ochrogaster*. *Animal Behaviour*, 63, 1121–1127. <https://doi.org/10.1006/anbe.2002.3014>
- Thonhauser, K. E., Raveh, S., Hettyey, A., Beissmann, H., & Penn, D. J. (2013). Scent marking increases male reproductive success in wild house mice. *Animal Behaviour*, 86(5), 1013–1021. <https://doi.org/10.1016/j.anbehav.2013.09.004>
- Wong, B. B. M., & Candolin, U. (2005). How is female mate choice affected by male competition? *Biological Reviews*, 80, 559–571. <https://doi.org/10.1017/S1464793105006809>
- Zahavi, A. (2008). The handicap principle and signaling in collaborative systems. In P. d’Ettorre & D. P. Hughes (Eds.), *Sociobiology of communication: An interdisciplinary perspective* (pp. 1–9). Oxford, UK: Oxford University Press.
- Zar, J. H. (2010). *Biostatistical analysis*. Upper Saddle River, NJ: Pearson Prentice Hall.
- Zenuto, R. R. (2010). Dear enemy relationships in the subterranean rodent *Ctenomys talarum*: The role of memory of familiar odors. *Animal Behavior*, 79, 1247–1255. <https://doi.org/10.1016/j.anbehav.2010.02.024>
- Zenuto, R. R., Estavillo, C., & Fanjul, M. S. (2007). Familiarity and mating behaviour in the subterranean rodent *Ctenomys talarum* (tuco-tuco). *Canadian Journal of Zoology*, 85, 944–955. <https://doi.org/10.1139/Z07-078>
- Zenuto, R. R., & Fanjul, M. S. (2002). Olfactory discrimination of individual scents in the subterranean rodent *Ctenomys talarum* (tuco-tuco). *Ethology*, 108(7), 629–641. <https://doi.org/10.1046/j.1439-0310.2002.00808.x>
- Zenuto, R. R., Fanjul, M. S., & Busch, C. (2004). Use of chemical communication by the subterranean rodent *Ctenomys talarum* (tuco-tuco) during breeding season. *Journal of Chemical Ecology*, 30(11), 2111–2126. <https://doi.org/10.1023/B:JOEC.0000048777.42945.e4>
- Zenuto, R. R., Lacey, E. A., & Busch, C. (1999a). DNA fingerprinting reveals polygyny in the subterranean rodent *Ctenomys talarum*. *Molecular Ecology*, 8, 1529–1532. <https://doi.org/10.1046/j.1365-294x.1999.00715.x>
- Zenuto, R. R., Malizia, A. I., & Busch, C. (1999b). Sexual size dimorphism, testes size and mating system in two populations of *Ctenomys talarum* (Rodentia: Octodontidae). *Journal of Natural History*, 33, 305–314. <https://doi.org/10.1080/002229399300434>

- Zenuto, R. R., Vasallo, A., & Busch, C. (2001). A method for studying social and reproductive behavior of subterranean rodents in captivity. *Acta Theriologica*, 46, 161–170. <https://doi.org/10.4098/0001-7051>
- Zenuto, R. R., Vasallo, A., & Busch, C. (2002). Comportamiento social y reproductivo del roedor subterráneo solitario *Ctenomys talarum* (Rodentia: Ctenomyidae) en condiciones de semicautiverio. *Revista Chilena de Historia Natural*, 75, 165–177.
- Zinkevich, E. P., & Vasilieva, V. S. (2001). Molecular approaches in chemical communication of mammals. In A. Marchlewska-Koj, J. J. Lepri, & D. Müller-Schwarze (Eds.), *Chemical signals in vertebrates 9* (pp. 53–60). New York, NY: Kluwer Academic/Plenum Publishers. <https://doi.org/10.1007/978-1-4615-0671-3>
- Zuri, I., Gazit, I., & Terkel, J. (1997). Effect of scent-marking in delaying territorial invasion in the blind mole-rat *Spalax ehrenbergi*. *Behaviour*, 134, 867–880. <https://doi.org/10.1163/156853997X00197>

**How to cite this article:** Fanjul MS, Varas MF, Zenuto RR. Female preference for males that have exclusively marked or invaded territories depends on male presence and its identity in the subterranean rodent *Ctenomys talarum*. *Ethology*. 2018;00:1–12. <https://doi.org/10.1111/eth.12761>