# Age Related Differences in Lipophilic Compounds Found in Femoral Gland Secretions of Male Spiny-footed Lizards, *Acanthodactylus erythrurus*

Pilar López\* and José Martín

Departamento de Ecología Evolutiva, Museo Nacional de Ciencias Naturales, CSIC, José Gutiérrez Abascal 2, 28006 Madrid, Spain. Fax: 34-91-5645078. E-mail: Pilar.Lopez@mncn.csic.es

- \* Author for correspondence and reprint requests
- Z. Naturforsch. **60 c**, 915–920 (2005); received May 13, 2005

Although chemoreception plays an important role in social organization of many lizards, only a few studies have examined the chemicals found in secretions used for intraspecific communication. We report the composition of the secretion of the femoral glands of males of the spiny-footed lizard (*Acanthodactylus erythrurus*). On the basis of mass spectra, obtained by GC/MS, we identified 45 lipophilic compounds, including several alcohols ranging from 10 to 29 carbon atoms (mainly hexacosanol and tetracosanol), steroids (mainly cholesterol and dehydrocholesterol), n-C $_9$  to n-C $_{20}$  carboxylic acids, esters of carboxylic acids, and minor components such as lactones, ketones, squalene and  $\alpha$ -tocopherol. Some of these compounds are reported for the first time in lizards. Adult and subadult males differed in the composition of secretions, with C $_9$  to C $_{15}$  carboxylic acids being more abundant in younger than in older lizards, whereas C $_{16}$  to C $_{20}$  carboxylic acids were more abundant in older lizards. Also, older lizards had significant lower proportions of cholesterol and campesterol but higher proportions of dehydrocholesterol.

Key words: Acanthodactylus erythrurus, Femoral Glands, Spiny-footed Lizards

### Introduction

Chemoreception plays an important role in social organization of many lizards (Mason, 1992; Halpern, 1992). Most species are able to detect pheromones from the skin, precloacal or femoral gland secretions (e.g., Alberts, 1993; Aragón et al., 2001a). The femoral pores are epidermal structures on the ventral surface of the thigh of many squamates connected to glands that produce copious amounts of holocrine secretion, especially in males and during the mating season (Mason, 1992; Alberts, 1993). The presence and relative concentration of pheromone components seem to vary not only between sexes but also consistently among individuals, which may convey information on the individual identity and serve a variety of functions (Alberts, 1990, 1992, 1993).

The ventral location of the femoral pores suggests that secretions are passively deposited on the substrate as lizards move through their home ranges, and, therefore, they can advertise residence in a home range, and/or can convey information about social status and competitive ability of the sender (Aragón *et al.*, 2001b, 2003; López and

Martín, 2002). Also, femoral pores secretion might transmit chemical information about a male's quality, which may be used by female lizards in their mate choice process (Martín and López, 2000; López *et al.*, 2002, 2003).

In spite of the potential importance of chemical signals in lizard intraspecific relationships, only a few studies have analyzed the chemical composition of these secretions mainly in Iguanids (Chauhan, 1986; Alberts, 1990; Weldon et al., 1990; Alberts et al., 1992; Escobar et al., 2001, 2003). Chemical data for most groups of lizards are not available, but only for skin semiochemicals of a few species (Weldon and Bangall, 1987; Mason and Gutzke, 1990). These studies have shown that femoral (or the similar precloacal) gland secretions are composed of both lipids and proteins. Lipids have a high degree of molecular diversity, which increase the potential information content of a pheromone, and are thought to be the main compounds involved in communication.

The spiny-footed lizard or common fringe-toad (A. erythrurus) is a medium-sized lacertid lizard (adult snout-vent length up to 82 mm) (Pérez-Mellado, 1998). It is the only species of its genus

present in Western Europe, where it occupies xeric, open, sandy areas with sparsely distributed vegetation (Arnold, 1987). This lizard feeds mainly on ants which it captures in the open after short, rapid attacks launched from ambushing sites (Belliure et al., 1996). There are no specific studies on detection of conspecific scents in this lizard, but, as in other lacertid lizards, the vomeronasal organ is well developed and chemoreceptor cells are abundant (Cooper, 1996); other species in the same genus are able to detect prey chemicals (Cooper, 1999), and there is a clear sexual dimorphism in femoral pores (the size of pores and the amount of secretion are higher in males; females have vestigial pores without apparent secretion). These data strongly suggests that, as in other lacertids, A. erythrurus should use chemical signals from femoral pores in some aspects of their social life. We report here the results of an analysis by gas chromatography-mass spectrometry (GC/ MS) of the lipidic fraction of femoral secretions of male A. erythrurus lizards. We specifically examined whether there were differences between adult and subadult males in the chemicals, or in their relative proportions, found in their secretions.

## **Material and Methods**

### Lizards capture and measurements

We captured by noosing 16 male A. erythrurus during June 2004 in different places in a siliceous and sandy large area in the southwest of the Madrid province (Central Spain) at an average elevation of 600 m. The area was covered by an open "dehesa" woodland characterized by Quercus ilex forest with an understory of dispersed low shrub cover (mainly Lavandula stoechas and Thymus mastichina), and extensive areas of bare sand substrates between isolated bushes (Martín and López, 2002). Lizards were weighed and their snoutvent length (SVL) was measured. Two clear age categories were found in the population; young, but reproductively mature subadults [body mass: mean  $\pm$  SE = (4.7  $\pm$  0.4) g, range = 3.7-5.5 g; SVL:  $(64.0 \pm 1.9)$  mm, range = 61-69 mm; n = 8] and older adults [body mass: mean  $\pm$  SE = (11.1  $\pm$ 0.4) g, range = 10.5-12.5 g; SVL:  $(82.0 \pm 1.2)$  mm, range = 80-85 mm; n = 8] (see Castilla *et al.*, 1992; Pérez-Mellado, 1998).

Lizards had an average ( $\pm$  SE) of (23.6  $\pm$  0.4) (range = 22–26) femoral pores on each leg, which did not significantly vary between age classes

(Mann-Whitney U-test, Z=1.44, p=0.15). We extracted the femoral pores secretion by gently pressing with forceps around the femoral pores, and collected the secretion directly in glass vials with Teflon-lined stoppers. Vials were stored at -20 °C until analyses. We also used the same procedure on each sampling occasion but without collecting secretion to obtain blank control vials that were treated in the same manner to compare with the lizard samples. Lizards were released to their initial sighting location prior to the capture. Before the analyses we added  $250 \,\mu$ l of n-hexane (Sigma, capillary GC grade) to each vial.

# GC/MS

Samples were analyzed using a Finnigan-ThermoQuest Trace 2000 gas chromatograph (GC) fitted with a poly(5% diphenyl/95% dimethylsiloxane) column (Supelco, Equity-5, 30 m length × 0.25 mm ID, 0.25-µm film thickness) and a Finnigan-ThermoQuest Trace mass spectrometer (MS) as detector. Sample injections (2  $\mu$ l of each sample dissolved in n-hexane) were performed in splitless mode, using helium as the carrier gas, with injector and detector temperatures at 270 °C and 250 °C, respectively. The oven temperature program was as follows: 50 °C isothermal for 10 min, then increased to 280 °C at a rate of 5 °C/min, and then isothermal (280 °C) for 30 min. Mass spectral fragments below m/z = 39 were not recorded. Impurities identified in the solvent and/or the control vial samples were not reported. Initial identification of secretion components was done by comparison of mass spectra in the NIST/EPA/NIH 1998 computerized mass spectral library. When possible, identifications were confirmed by comparison of spectra and retention times with those of authentic standards. Authentic samples were purchased from Sigma-Aldrich Chemical Co.

# Data analyses

The relative amount of each component was determined as the percent of the total ion current (TIC). Then, we selected the peaks that represented > 0.05% relative peak area and that were present in all the individuals to reduce the number of variables to be used in multivariate statistical analysis (Dietemann *et al.*, 2003). The relative areas of the 29 selected peaks were restandardized to 100% and transformed following Aitchison's formula:  $Z_{ij} = \ln(Y_{ij}/g(Y_j))$ , where  $Z_{ij}$  is the stand-

ardized peak area i for individual j,  $Y_{ij}$  is the peak area i for individual j, and  $g(Y_j)$  is the geometric mean of all peaks for individual j (Aitchison, 1986). The homogeneity of variance of these variables was tested with Levene's test, and Bonferroni's correction was applied. The transformed areas were used as variables in a principal component analysis. The five principal components (PC) extracted (which explained 95% of variance) were used as independent variables in a multivariate analysis of variance (MANOVA) to determine whether subadult and adult lizards groups differed in the abundances of compounds.

#### Results

A total of 45 major lipophilic compounds was identified in femoral gland secretions of A. erythrurus (Table I). Alcohols (72.2%), steroids (19.6%) and carboxylic acids (6.4%) and their esters (0.4%) were the main components, but we found also lactones (1.0%), squalene (0.3%), two ketones (0.04%), and  $\alpha$ -tocopherol (0.04%).

Multivariate analyses on the five PC for the 29 shared major compounds showed that there were significant differences in composition between age classes (MANOVA, Wilks'  $\lambda = 0.023$ ,  $F_{5,10} = 83.36$ , p < 0.0001), which were due to differences in the PC-2 (protected ANOVA,  $F_{1,14} = 54.84$ , p < 0.0001). Thus, according to the correlations of compounds with the PC-2, subadults have higher portions of nonanoic, dodecanoic and tetradecanoic acids, squalene, cholesterol and campesterol, and lower portions of hexadecanoic, octadecadienoic, octadecenoic, octadecanoic acids, and of cholesta-5,7-dien-3-ol and eicosanol than adults.

Free alcohols ranging from 10 to 29 carbon atoms were the main components in the femoral secretions of A. erythrurus. Hexacosanol and tetracosanol were the most abundant ones. Hexacosanol was present in all individuals and showed little variation (range = 50-59%, coefficient of variation = 5.2%). Neither there were significant differences between age classes in the proportion of these two main chemicals.

Cholesterol and cholesta-5,7-dien-3-ol (= dehydrocholesterol) were the main steroids found in all secretions, but there was a high interindividual variability (cholesterol: range = 4-23%, coefficient of variation = 66.4%; dehydrocholesterol: range = 4-15%, coefficient of variation = 31.4%).

Older lizards had significant lower portions of cholesterol, but higher portions of dehydrocholesterol. We also found other steroids in significant quantities, such as campesterol, mainly in subadults, ergosta-dien-ol, and an unidentified steroid.

Carboxylic acids found in femoral secretions of A. erythrurus ranged between n- $C_9$  and n- $C_{20}$ , with dodecanoic, hexadecanoic, and octadecadienoic acid being the more abundant. However, there were significant differences between age classes (see above) with  $C_9$  to  $C_{15}$  carboxylic acids being more abundant in younger than in older lizards, whereas  $C_{16}$  to  $C_{20}$  carboxylic acids were more abundant in older lizards.

#### Discussion

It was surprising to find such a high portion of alcohols in secretions of this lacertid lizard. These long-chain alcohols may form waxy esters, which can give consistence to femoral secretions (hexacosanol is also known as 'cery alcohol', and tetracosanol as 'lignoceric alcohol'). Alcohols have only been reported previously as minor components from femoral gland secretions of L. monticola, another lacertid lizard (López and Martín, 2005), and in extracts from the skin of *Iguana* iguana (Roberts and Lillywhite, 1980) and of several snakes (see review in Mason, 1992). In contrast, other lizards, mainly Iguanids, have alkanes, carboxylic acids and steroids as the main compounds of glandular secretions, but not alcohols (Alberts, 1990; Weldon et al., 1990; Alberts et al., 1992; Escobar et al., 2001, 2003). Alcohols are also abundant in paracloacal gland secretions of several species of crocodiles (Shafagati et al., 1989; Dunn et al., 1993; García-Rubio et al., 2002). Thus, different compounds may be characteristics of some groups of reptiles, but be absent in others. The presence of different compounds may have a phylogenetic effect, but it might also be linked to different environmental conditions, or to the way how these chemicals are used in communication (e.g., as scent marks on different substrates, or as chemicals sampled directly from the individuals). This raises the need of more studies analyzing the chemical compounds in secretions of diverse groups of lizards.

Cholesterol was found in abundance in most lizard species, both in the skin (Weldon and Bangall, 1987; Mason and Gutzke, 1990) and in femoral and precloacal gland secretions (Alberts *et al.*,

Table I. Lipophilic compounds found in femoral secretions of spiny-footed subadult and adult male lizards, *Acanthodactylus erythrurus*. The relative amount of each component was determined as the percent of the total ion current (TIC) and reported as the average  $(\pm 1 \text{ SD})$ .

RTa [min]	Compound	Subadults	Adults
19.5	Decanol	$0.06 \pm 0.04$	$0.06 \pm 0.08$
25.3	Nonanoic acid	$0.16 \pm 0.07$	$0.03 \pm 0.02$
25.9	Undecanol	$0.05 \pm 0.08$	$0.01 \pm 0.01$
26.0	Tridecanone	_	$0.01 \pm 0.01$
31.2	Dodecanol	$0.04 \pm 0.04$	$0.01 \pm 0.01$
32.9	Dodecanoic acid	$2.52 \pm 0.92$	$0.35 \pm 0.08$
33.8	Dodecenoic acid	$0.25 \pm 0.17$	$0.11 \pm 0.06$
37.4	Tetradecanoic acid	$0.07 \pm 0.03$	$0.03 \pm 0.01$
38.8	Tetradecanoic acid, 1-methylethyl ester	_	$0.01 \pm 0.01$
39.5	Pentadecanoic acid	$0.02 \pm 0.02$	$0.01 \pm 0.01$
40.1	4-Hydroxy-dodecanoic acid, γ-lactone	$0.55 \pm 0.37$	$0.51 \pm 0.34$
40.3	Nonadecanone	$0.06 \pm 0.02$	$0.02 \pm 0.01$
41.0	Hexadecenoic acid	$0.03 \pm 0.06$	$0.03 \pm 0.01$
41.4	Hexadecanoic acid	$0.84 \pm 0.14$	$1.61 \pm 0.56$
43.4	Heptadecanoic acid	_	$0.02 \pm 0.01$
43.9	Octadecanol	$0.08 \pm 0.06$	$0.05 \pm 0.04$
44.2	4-Hydroxy-hexadecanoic acid, γ-lactone	$0.54 \pm 0.24$	$0.27 \pm 0.09$
44.7	9,12-Octadecadienoic acid	$0.29 \pm 0.13$	$1.73 \pm 0.79$
44.8	Octadecenoic acid	$0.28 \pm 0.12$	$0.72 \pm 0.20$
44.9	Oleic acid	$0.25 \pm 0.20$	$0.51 \pm 0.17$
45.2	Octadecanoic acid	$0.64 \pm 0.22$	$1.28 \pm 0.35$
47.4	Eicosanol	$0.07 \pm 0.07$	$0.28 \pm 0.06$
47.6	Eicosatetraenoic acid, ethyl ester	$0.02 \pm 0.03$	$0.22 \pm 0.18$
47.9	4-Hydroxy-octadecanoic acid, methyl ester	$0.05 \pm 0.03$	$0.05 \pm 0.05$
48.6	Eicosanoic acid	$0.46 \pm 0.13$	$0.62 \pm 0.15$
50.7	Docosanol	$0.30 \pm 0.23$	$0.85 \pm 0.32$
51.3	4-Hydroxy-octadecanoic acid, γ-lactone	$0.06 \pm 0.06$	$0.02 \pm 0.01$
51.8	Docosanoic acid	$0.06 \pm 0.04$	$0.10 \pm 0.03$
53.8	Tetracosanol	$10.31 \pm 2.94$	$13.36 \pm 2.72$
55.0	Unidentified steroid	_	$0.03 \pm 0.03$
55.7	Squalene	$0.42 \pm 0.48$	$0.09 \pm 0.05$
56.3	Cĥolesta-3,5-diene	_	$0.10 \pm 0.09$
56.7	Hexacosanol	$54.34 \pm 3.27$	$53.23 \pm 2.65$
57.2	Cholesta-5,7,9(11)-trien-3-ol	$0.18 \pm 0.13$	$0.10 \pm 0.03$
57.8	Cholesta-5,7-dien-3-ol, acetate	$0.01 \pm 0.03$	$0.04 \pm 0.03$
60.2	Nonacosanol	$5.24 \pm 0.75$	$5.98 \pm 0.87$
60.8	Cholesterol	$12.05 \pm 7.51$	$6.00 \pm 1.82$
61.0	$\alpha$ -Tocopherol	$0.07 \pm 0.09$	$0.01 \pm 0.01$
61.7	Cholesta-5,7-dien-3-ol	$8.52 \pm 3.33$	$11.13 \pm 2.58$
63.5	Campesterol	$0.37 \pm 0.20$	$0.06 \pm 0.03$
64.3	Ergosta-5,8-dien-3-ol	$0.28 \pm 0.20$	$0.21 \pm 0.11$
65.2	Hexadecyl hexadecenoate	$0.25 \pm 0.50$	_
65.9	γ-Sitosterol	$0.04 \pm 0.09$	_
66.4	Octadecyl octadecenoate	$0.10 \pm 0.21$	$0.01 \pm 0.14$
67.2	4,4-Dimethyl-cholesta-5,7-dien-3-ol	_	$0.01 \pm 0.02$

<sup>&</sup>lt;sup>a</sup> RT: Retention time.

1992; Escobar *et al.*, 2001; López and Martín, 2005). It has been suggested that the abundance and ubiquity of this component may help to constitute an unreactive apolar matrix that might deliver the compounds that are the true semiochemicals (Escobar *et al.*, 2003). Additionally, interesting is the presence in femoral secretions of dehydrocholesterol, a precursor for vitamin  $D_3$ , which is

often found in the skin, where it will transform into vitamin  $D_3$  after exposition to sun UVB irradiation (Carman *et al.*, 2000).

Similar long-chain carboxylic acids have also been found in other lizards as common and major constituents of skin (Mason and Gutzke, 1990; Weldon *et al.*, 1990) and femoral and precloacal glands (Alberts *et al.*, 1992; Escobar *et al.*, 2001;

López and Martín, 2005). In these studies the range in the number of carbon atoms in carboxylic acids varied between species (e.g. n-C<sub>14</sub> to n-C<sub>24</sub> in. I. iguana, or n- $C_3$  to n- $C_{26}$  in Liolaemus spp.). Presumably this would reflect the environment where each lizard species lives. Thus, it might be expected that under higher environmental temperatures, lizards had a larger portion of compounds of lower volatility and higher chemical stability (Escobar et al., 2001, 2003). Because A. erythrurus inhabit arid areas where temperatures are high, we should expect a high number of carbon atoms in carboxylic acids. Our study show that C<sub>9</sub> to C<sub>15</sub> carboxylic acids were more abundant in younger than in older lizards, whereas  $C_{16}$  to  $C_{20}$  carboxylic acids were more abundant in older lizards. If scent marking is used by this lizard, then the persistence of scent marks would be more important for reproductive-territorial (= older) males than for younger-floater males. Thus, older males should increase the portion of low volatile chemicals in their femoral secretions. In contrast, lipids in femoral secretions of I. iguana did not differ between adult and juvenile males (Weldon et al., 1990), but all carboxylic acids found in this species have low volatility (see above).

Other minor components were also found in femoral secretions of *A. erythrurus* lizards. Squalene was also found in femoral gland secretions of the lacertid, *L. monticola* (López and Martín, 2005), but not in gland secretions of Iguanid lizards (Weldon *et al.*, 1990; Escobar *et al.*, 2001). Squalene, however, is found in the skin of male and female leopard geckos (Mason and Gutzke, 1990) and of male garter snakes (Mason *et al.*, 1989), and also in paracloacal glands of crocodiles (García-Rubio *et al.*, 2002). In snakes, squalene

Aitchison J. (1986), The Statistical Analysis of Compositional Data. Chapman and Hall, London.

Alberts A. C. (1990), Chemical properties of femoral gland secretions in the desert iguana *Dipsosaurus dorsalis*. J. Chem. Ecol. **16**, 13–25.

Alberts A. C. (1992), Pheromonal self-recognition in desert iguanas. Copeia **1992**, 229–232.

Alberts A. C. (1993), Chemical and behavioral studies of femoral gland secretions in iguanid lizards. Brain Behav. Evol. **41**, 255–260.

Alberts A. C., Sharp T. R., Werner D. I., and Weldon P. J. (1992), Seasonal variation of lipids in femoral gland secretions of male green iguanas (*Iguana iguana*). J. Chem. Ecol. **18**, 703–712.

Aragón P., López P., and Martín J. (2001a), Discrimination of femoral gland secretions from familiar and unfamiliar conspecifics by male Iberian rock-lizards, *Lacerta monticola*. J. Herpetol. 35, 346–350.

was thought to contribute to sex identification, as females have not this chemical in their skin (Mason et al., 1989). We also found three lactones (= furanones) which have not been previously described in lizards, but a lactone of the hydroxy octadecanoic acid was described, although later not confirmed, in a crocodile (Navajas Polo, 1982). Also, furanones have been identified as components of the sex pheromone of some insects (e.g., Farine et al., 1993; James et al., 2003). Thus, it remains possible that lactones have also some functions in communication in this lizard. We also found three ketones, which have never been found in other lizards. However, several long-chain saturated and unsaturated methyl-ketones were identified in the skin of female garter snakes having activity of sex pheromones (Mason et al., 1989, 1990). Finally, it was interesting to find  $\alpha$ -tocopherol, one of the most abundant forms of vitamin E (Brigelius-Flohe and Traber, 1999).  $\alpha$ -Tocopherol was also found in secretions from the gular and paracloacal glands of the American alligator (Weldon et al., 1987, 1988). Further studies are needed to clarify the possible role of the chemicals found in femoral secretions of this lizard in its social organization and sexual selection processes.

# Acknowledgements

We thank E. Dunkelblum and A. Guerrero for helpful comments, J. Belliure and P. L. Moreira for field assistance collecting lizards, and "El Ventorrillo" MNCN Field Station for use of their facilities. Financial support was provided by the MCYT project BOS 2002-00598 and the MEC project CGL2005-00391/BOS.

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

Aragón P., López P., and 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.

Arnold E. N. (1987), Resource partitioning among lacertid lizards in Southern Europe. J. Zool. (suppl.) 1, 739–782.

Belliure J., Carrascal L. M., and Díaz J. A. (1996), Covariation of thermal biology and foraging mode in two Mediterranean lacertid lizards. Ecology **77**, 1163–1173

Brigelius-Flohe R. and Traber M. G. (1999), Vitamin E: function and metabolism. FASEB J. 13, 1145–1155.

- Carman E. N., Ferguson G. W., Gehrmann W. H., Chen T. C., and Holick M. F. (2000), Photobiosynthetic opportunity and ability for UVB generated vitamin D synthesis in free-living house geckos (*Hemidactylus turcicus*) and Texas spiny lizards (*Sceloporus olivaceous*). Copeia **2000**, 245–250.
- Castilla A. M., Barbadillo L. J., and Bauwens D. (1992), Annual variation in reproductive traits in the lizard Acanthodactylus erythrurus. Can. J. Zool. **70**, 395–402.
- Chauhan N. B. (1986), A preliminary report on the lipid components of pre-anal gland secretion of lizards *Hemidactylus flaviviridis* and *Uromastix hardwickii*. J. Anim. Morphol. Physiol. **33**, 73–76.
- Cooper Jr. W. E. (1996), Preliminary reconstructions of nasal chemosensory evolution in Squamata. Amph.-Rept. 17, 395–415.
- Cooper Jr. W. E. (1999), Supplementation of phylogenetically correct data by two species comparison: support for correlated evolution of foraging mode and prey chemical discrimination in lizards extended by first intrageneric evidence. Oikos **86**, 97–104.
- Dietemann V., Peeters C., Liebig J., Thivet V., and Hölldobler B. (2003), Cuticular hydrocarbons mediate discrimination of reproductives and nonreproductives in the ant *Myrmecia gulosa*. Proc. Natl. Acad. Sci. USA **100**, 10341–10346.
- Dunn Jr. B. S., Weldon P. J., Howard R. W., and McDaniel C. A. (1993), Lipids from the paracloacal glands of the Chinese alligator (*Alligator sinensis*). Lipids **28**, 75–78.
- Escobar C. A., Labra A., and Niemeyer H. M. (2001), Chemical composition of precloacal secretions of *Lio-laemus* lizards. J. Chem. Ecol. 27, 1677–1690.
- Escobar C. M., Escobar C. A., Labra A., and Niemeyer H. M. (2003), Chemical composition of precloacal secretions of two *Liolaemus fabiani* populations: are they different? J. Chem. Ecol. **29**, 629–638.
- Farine J. P., le Quere J.-L., Duffy J., Semon E., and Brossut R. (1993), 4-Hydroxy-5-methyl-3(2*H*)-furanone and 4-hydroxy-2,5-dimethyl-3(2*H*)-furanone, two components of the male sex pheromone of *Eurycolis florianda* (Walker) (Insecta, Blattidae, Polyzosteriinae). Biosci. Biotechn. Biochem. **57**, 2026–2030.
- García-Rubio S., Attygalle A.B., Weldon P. J., and Meinwald J. (2002), Reptilian chemistry: volatile compounds from paracloacal glands of the American crocodile (*Crocodylus acutus*). J. Chem. Ecol. **28**, 769–781
- Halpern M. (1992), Nasal chemical senses in reptiles: structure and function. In: Biology of the Reptilia, Vol. 18 (Gans C. and Crews D., eds.). University of Chicago Press, Chicago, pp. 423–523.
- James D. G., Petroski R. J., Coss A. A., Zilkowski B. W., and Bartelt R. J. (2003), Bioactivity, synthesis, and chirality of the sex pheromone of currant stem girdler, *Janus integer*. J. Chem. Ecol. **29**, 2189–2199.
- López P. and Martín J. (2002), Chemical rival recognition decreases aggression levels in male Iberian wall lizards, *Podarcis hispanica*. Behav. Ecol. Sociobiol. 51, 461–465.

- López P. and Martín J. (2005), Chemical compounds from femoral gland secretions of male Iberian Rock lizards, *Lacerta monticola cyreni*. Z. Naturforsch. **60 c**, 632–636.
- López P., Muñoz A., and Martín J. (2002), Symmetry, male dominance and female mate preferences in the Iberian rock lizard, *Lacerta monticola*. Behav. Ecol. Sociobiol. **52**, 342–347.
- López P., Aragón P., and Martín J. (2003), Responses of female lizards, *Lacerta monticola*, to males' chemical cues reflect their mating preference for older males. Behav. Ecol. Sociobiol. **55**, 73–79.
- Martín J. and López P. (2000), Chemoreception, symmetry and mate choice in lizards. Proc. R. Soc. London Ser. B **267**, 1265–1269.
- Martín J. and López P. (2002), The effect of Mediterranean dehesa management on lizard distribution and conservation. Biol. Conserv. **108**, 213–219.
- Mason R. T. (1992), Reptilian pheromones. In: Biology of the Reptilia, Vol. 18 (Gans C. and Crews D., eds.). University of Chicago Press, Chicago, pp. 114–228,
- Mason R. T. and Gutzke W. H. N. (1990), Sex recognition in the leopard gecko, *Eublepharis macularius* (Sauria: Gekkonidae). Possible mediation by skin-derived semiochemicals. J. Chem. Ecol. **16**, 27–36.
- Mason R. T., Fales H. M., Jones T. H., Pannell L. K., Chinn J. W., and Crews D. (1989), Sex pheromones in snakes. Science **241**, 290–293.
- Mason R. T., Jones T. H., Fales H. M., Pannell L. K., and Crews D. (1990), Characterization, synthesis, and behavioral response to sex pheromone in garter snakes. J. Chem. Ecol. **16**, 2353–2369.
- Navajas Polo C. (1982), Procedimiento para la Obtención de una Lactona y el Producto Obtenido. Cuban patent #21412.
- Pérez-Mellado V. (1998), Acanthodactylus erythrurus (Schinz, 1834). In: Reptiles, Fauna Ibérica, Vol. 10 (Salvador A., ed.). Museo Nacional de Ciencias Naturales, Madrid, pp. 167–175.
- Roberts J. B. and Lillywhite H. B. (1980), Lipid barrier to water exchange in reptile epidermis. Science **207**, 1077–1079.
- Shafagati A., Weldon P. J., and Wheeler J. W. (1989), Lipids in the paracloacal gland secretions of dwarf (*Paleosuchus palpebrosus*) and smooth-fronted (*P. trigonatus*) caimans. Biochem. Syst. Ecol. **17**, 431–435.
- Weldon P. J. and Bangall D. (1987), A survey of polar and nonpolar skin lipids from lizards by thin-layer chromatography. Comp. Biochem. Physiol. **87B**, 345–349.
- Weldon P. J., Shafagati A., and Wheeler J. W. (1987), Lipids in the gular gland secretion of the American alligator (*Alligator mississippiensis*). Z. Naturforsch. 42 c, 1345–1346.
- Weldon P. J., Shafagati A., and Wheeler J. W. (1988), Lipids from the paracloacal glands of the American alligator (*Alligator mississippiensis*). Lipids **23**, 727–729.
- Weldon P. J., Dunn B. S., McDaniel C. A., and Werner D. I. (1990), Lipids in the femoral gland secretions of the green iguana (*Iguana iguana*). Comp. Biochem. Physiol. B **95**, 541–543.