

# Parasites of *Rana perezi* Seoane, 1885 in Ávila Province, Spain.

**Jiménez, M.S.; Zapatero, L.M. & Castaño, C.**

Departamento de Parasitología. Facultad de Farmacia. Universidad Complutense de Madrid.

Received: 11.06.01

Accepted: 11.12.01

**Abstract:** A total of 42 species of parasites including 2 species of blood bacteria, 29 protozoa, 6 trematodes, 4 nematodes and 1 leech were reported from *Rana perezi* Seoane, 1885, captured in Ávila Province (Spain). The following organisms were identified: *Aegyptianella bacterifera*, *Aegyptianella ranarum*, *Retortamonas dobelli*, *Chilomastix caulleryi*, *Hexamita intestinalis*, *Spiرونucleus elegans*, *Brugerolleia algonquinensis*, *Octomitus neglectus*, *Giardia agilis*, *Monocercomonas maculatus*, *Trichomitus batrachorum*, *Tetratrichomonas prowazeki*, *Trypanosoma rotatorium*, *Trypanosoma loricatum*, *Trypanosoma* sp.1., *Trypanosoma* sp.2., *Rhizomastix gracilis*, *Opalina ranarum*, *Opalina obtrigona*, *Cepedea dimidiata*, *Entamoeba ranarum*, amoebas of free living, *Eimeria prevoti*, *Lankesterella minima*, *Dactylosoma ranarum*, *Nyctotheroides cordiformis*, *Balantidium duodenii*, *Balantidium entozoon*, *Balantidium elongatum*, *Trichodina ranae*, *Blastocystis enterocola*, *Opisthodiscus nigrivasis*, *Haematoloechus variegatus*, *Cephalogonimus retusus*, *Gorgodera amplicava*, *Gorgoderina vitelliloba*, *Opisthioglyphe endoloba*, *Cosmocerca ornata*, *Rhabdias bufonis*, *Icosiella neglecta* and *Capillaria costacruzi*.

The ectoparasites found on many frogs were leeches, *Batracobdella algira*, which was vector for: *Icosiella neglecta*, *Lankesterella minima* and *Trypanosoma* sp. in *R. perezi* of Ávila.

**Key words:** *Rana perezi*, Protozoa, Trematoda, Nematoda, blood bacteria, Ávila.

**Resumen:** Un total de 42 especies de parásitos incluyendo 2 especies de bacterias sanguíneas, 29 especies de protozoos, 6 especies de trematodos, 4 especies de nematodos y 1 especie de sanguijuela se han encontrado en *Rana perezi* Seoane, 1885, capturada en la Provincia de Ávila (España). Los organismos identificados fueron: *Aegyptianella bacterifera*, *Aegyptianella ranarum*, *Retortamonas dobelli*, *Chilomastix caulleryi*, *Hexamita intestinalis*, *Spiرونucleus elegans*, *Brugerolleia algonquinensis*, *Octomitus neglectus*, *Giardia agilis*, *Monocercomonas maculatus*, *Trichomitus batrachorum*, *Tetratrichomonas prowazeki*, *Trypanosoma rotatorium*, *Trypanosoma* sp.1., *Trypanosoma* sp.2., *Rhizomastix gracilis*, *Opalina ranarum*, *Opalina obtrigona*, *Cepedea dimidiata*, *Entamoeba ranarum*, amoebas de vida libre, *Eimeria prevoti*, *Lankesterella minima*, *Dactylosoma ranarum*, *Nyctotheroides cordiformis*, *Balantidium duodenii*, *Balantidium entozoon*, *Balantidium elongatum*, *Trichodina ranae*, *Blastocystis enterocola*, *Opisthodiscus nigrivasis*, *Haematoloechus variegatus*, *Cephalogonimus retusus*, *Gorgodera amplicava*, *Gorgoderina vitelliloba*, *Opisthioglyphe endoloba*, *Cosmocerca ornata*, *Rhabdias bufonis*, *Icosiella neglecta* and *Capillaria costacruzi*. El ectoparásito hallado sobre algunas ranas fue la sanguijuela, *Batracobdella algira*, vector de: *Icosiella neglecta*, *Lankesterella minima* y *Trypanosoma* sp. en *R. perezi* de Ávila.

**Palabras claves:** *Rana perezi*, Protozoos, Trematodos, Nematodos, bacterias sanguíneas, Ávila.

## 1. Introduction

Our goal was to identify the parasites of *Rana perezi* Seoane, 1885 (greenfrog indigenous to Spain and France) in Ávila Province (lat. 40° 40' N, long. 4° 38' W), Spain, in order to contribute to the knowledge of the parasite fauna of our country.

Corresponding author: L.M. Zapatero  
Departamento de Parasitología. Facultad de Farmacia (Universidad Complutense de Madrid).  
Pza. Ramón y Cajal, s/n. 28.040, Madrid. Spain.  
Tlfno.: +34 91 394 18 18. Fax: +34 91 394 18 15  
E-mail: Zapater@farm.ucm.es

Ávila province is characterized by its extensive plain to the North (inside the plateau of the Duero) and its great surface of saw to the South (inside the Cordillera Central). It is divided in 6 natural regions:

- Plain: Arévalo-Madrigal or La Moraña and Ávila.
- Saw: Barco de Ávila-Piedrahita, Gredos, Valle del bajo Alberche and Valle del Tiétar.

The frogs were captured of five villages, which belong to the two regions of the plain:

- La Moraña: *San Pedro del Arroyo* (in the river Arevalillo and pools), *Albornos* (river Arevalillo), *Santo Tomé de Zabarcos* (river Arevalillo).

- Ávila: *Niharra* (river Adaja), *Marlín* (springs).

The parasites were compared with other mentioned in the current literature by:

Alexeieff (1911) in *Rana* sp. Ballesteros (1945) in *Rana esculenta* (it is likely that it was *R. perezi* because there is not *R. esculenta* in the Iberian Peninsula) and *Rana temporaria* of Málaga (Carratraca). Barta (1991) in *Rana clamitans* and *Rana catesbeiana* of Canada. Carrera et al. (1987) in *R. perezi* e *Hyla meridionalis* of Canary Islands (Tenerife, Gomera). Combes and Gerbeaux (1970) in *R. perezi* of Gerona (Segre e Isobol). Comber and Knoepffer (1965) in *Rana iberica*, *Rana ridibunda*, *R. perezi* and *Bufo bufo* of Ávila (Sierra de Gredos). Combes and Sarrouy (1971) in *R. perezi* of Soria. Da Cunha (1950) in *Rana* sp. of Portugal. Desportes (1941) in *Rana* sp. of France. Desser et al. (1993) in *R. clamitans* and *R. catesbeiana* of Canada. Fernández (1984) in *Bufo* sp. of Valencia. Fernández-Galiano (1947) in *Rana* sp. González Castro (1942) in *R. esculenta* (likely: *R. perezi*) of Granada. Lluch et al. (1986b) in *R. perezi* of Levante (Valencia and Alicante). Lluch et al. (1990) in *R. temporaria* of the Central Pirineo (Aragón) and Oriental Pirineo (Cataluña). López-Neyra (1947) in *R. esculenta* (likely: *R. perezi*) and *R. temporaria* of Granada and Madrid. Madrigal et al. (1987) *R. perezi* of Canary Islands. Martínez-Fernández et al. (1988) in *R. ridibunda* of Salamanca and León. Navarro et al. (1988) en *R. iberica* of Western Sistema Central, *R. temporaria* of Oriental Prepirineo and Pirineo and Central Pirineo and *R. perezi* of Western Sistema Central, Central Pirineo and Levante. Oliveira-Rodrigues et al. (1973) in *R. esculenta* (likely: *R. perezi*) of Portugal (Coimbra). Rodrigues et al. (1972) in *R. esculenta* (likely: *R. perezi*) of Portugal. Travassos (1932) in *Rana* sp. of Portugal.

## 2. Materials and Methods

432 individuals (352 fully grown specimens, 39 juveniles and 41 tadpoles) of *R. perezi* of Ávila were manually collected during a period between 1993 and 1995.

The frogs were euthanized, necropsied and examined for the presence of blood, cloacal, lung, liver, muscle, intestine and urinary bladder parasites using a light microscope or lens.

Films of blood and liver and the contents of the cloacal, intestine and urinary bladder were prepared and stained while fresh contents were examined by light microscopy to observe the morphology and motility of the protozoa. The blood, liver and urinary bladder films were air dried, fixed in methanol, and stained with Giemsa's stain. The cloacal and intestine films were stained with Giemsa-Suarez Peregrín's

stain and Bodian's protargol, which stains the rows of kinetics of opalinids and ciliates. Each film was scanned at low power for several minutes for large parasites and then examined for the oil immersion 100x objective for small or intracellular parasites.

Diamond's SN B9 culture (for trypanosomes) and Boeck and Drbohlav culture (for fecal protozoa) were also used.

Trematodes were separated of the walls of lung, cloacal, intestine and urinary bladder, fixed in formol 10% (during 1 day), stained with hydrochloric alcoholic carmine, mounted with Balsam of Canada and looked through a lens.

Nematodes were collected of muscle, cloacal and lung, fixed in alcohol 70°, mounted and cleared with Amman Lactophenol and examined for a lens.

## 3. Results

A total of 42 species of parasites including 2 species of blood bacteria, 29 protozoa, 6 trematodes, 4 nematodes and 1 leech were observed in *R. perezi* of Ávila.

All the frogs were parasited (100% prevalence): with a minimum of 1 and a maximum of 21 different species. Protozoa were present in 100 %, nematodes in 69 %, blood bacteria in 29.6 % and trematodes in 27.1 % of the frogs. 761 trematodes and 1032 nematodes (844 female and only 188 males) were collected, showing a mean intensity of 4.1 trematodes and 3 nematodes per infested frog

The following organisms were identified:

-Blood bacteria: *Aegyptianella bacterifera* (Labbé, 1894) Barta, 1989 and *Aegyptianella ranarum* Desser, 1987 (family Anaplasmataceae Philip, 1957).

-Protozoa: *Retortamonas dobelli* Bishop, 1931 and *Chilomastix caulleryi* Alexeieff, 1909 (family Retortamonadidae Wenrich, 1932); *Hexamita intestinalis* Dujardin, 1841; *Spironucleus elegans* Lavier, 1936; *Brugerolleia algonquinensis* Desser, Hong, Siddall and Barta, 1993; *Octomitus neglectus* (Lavier, 1936) Grassé, 1952 and *Giardia agilis* Kunstler, 1882 (family Hexamitidae Kent, 1880); *Monocercomonas maculatus* Carrera-Moro, 1987 (family Monocercomonadidae Kirby, 1944) Honigberg, 1963); *Trichomitus batrachorum* (Perty, 1852) Honigberg, 1963 and *Tetratrichomonas prowazeki* (Alexeieff, 1909) Alexeieff, 1911 (family Trichomonadidae (Chalmers and Pekkola, 1918) Honigberg, 1963); *Trypanosoma rotatorium* Mayer, 1843; *Trypanosoma loricatum* Mayer, 1843; *Trypanosoma* sp.1. and *Trypanosoma* sp.2. (family Trypanosomatidae Doflein, 1911); *Rhizomastix gracilis* (*Incertae sedis*); *Opalina ranarum* (Ehrenberg, 1832) Purkinje and Valentin, 1835; *Opalina obtrigona* Stein,

Table 1. Blood parasites.

	Prevalence (%)	Intensity (1-3)	New Host ( <i>R. perezi</i> )	New Area (IP/S)
<b>BACTERIA</b>				
<i>Aegyptianella bacterifera</i>	28.0	1.4	R	IP
<i>Aegyptianella ranarum</i>	9.7	1.2	R	IP
<b>PROTOZOA</b>				
<i>Brugerolleia algonquinensis</i>	1.2	2.0	R	IP
<i>Trypanosoma rotatorium</i>	28.0	1.4	R	IP
<i>Trypanosoma loricatum</i>	23.6	1.4	R	IP
<i>Trypanosoma sp.1.</i>	22.0	1.9	R	IP
<i>Trypanosoma sp.2.</i>	3.7	1.4	R	IP
<i>Lankesterella minima</i>	41.4	1.6	R	IP
<i>Dactylosoma ranarum</i>	5.3	1.3	R	IP
<b>NEMATODES</b>				
<i>Icosiella neglecta (microfilariae)</i>	3.2	2.7		

Prevalence (%): percentage of parasitism.

Intensity (1-3): between 1 (few specimens) and 3 (a lot of specimens).

New Host: R (*Rana perezi*).

New Area: IP (Iberian Peninsula) and S (Spain).

(Railliet, 1916) Travassos, 1925); *Rhabdias bufonis* (Schränk, 1788) Stiles and Hassall, 1905 (family: Rhabdiásidae Railliet, 1915); *Icosiella neglecta* (Diesing, 1851) Seurat, 1917 (family: Onchocercidae Leiper, 1911); *Capillaria costacruzi* Travassos, 1932 (family: Trichuridae Railliet, 1915).

-Leech: *Batracobdella algira* (Moquin-Tandon, 1846) Autrum, 1936 (family: Glossiphoniidae).

The prevalence, intensity, new host and new area records of the parasites are summarized in Table 1, 2, 3, 4, 5, 6 and 7.

#### 4. Discussion

Of all species, 11 are new host records: *Aegyptianella bacterifera*, *Aegyptianella ranarum*, *Hexamita intestinalis*, *Brugerolleia algonquinensis*,

Table 2. Cloaca parasites.

	Prevalence (%)	Intensity (1-3/nº)	New Host ( <i>R. perezi</i> )	New Area (IP/S)
<b>PROTOZOA</b>				
<i>Retortamonas dobelli</i>	21.1	1.5		IP
<i>Chilomastix caulleryi</i>	0.5	2.5		IP
<i>Hexamita intestinalis</i>	34.5	1.2	R	IP
<i>Spironucleus elegans</i>	81.9	1.8		IP
<i>Octomitus neglectus</i>	38.7	1.8		IP
<i>Monocercomonas maculatus</i>	46.3	1.6		IP
<i>Trichomitus batrachorum</i>	55.8	1.5		IP
<i>Tetrastrichomonas prowazeki</i>	5.1	1.0		IP
<i>Rhizomastix gracilis</i>	0.9	1.5		IP
<i>Opalina ranarum</i>	30.6	1.4		IP
<i>Opalina obtigona</i>	38.7	2.1		IP
<i>Cepedea dimidiata</i>	39.8	1.7		IP
<i>Entamoeba ranarum</i>	20.4	1.2	R	IP
<i>Amoebas of free living</i>	5.3	1.5		IP
<i>Eimeria prevoti</i>	5.6	2.0	R	IP
<i>Nyctotheroides cordiformis</i>	60.0	1.2		IP
<i>Balantidium entozoon</i>	80.3	1.3		IP
<i>Balantidium elongatum</i>	7.9	1.1	R	IP
<i>Blastocystis enterocola</i>	51.6	2.3		IP
<b>TREMATODES</b>				
<i>Opisthodiscus nigrivasis</i>	11.6	1.4		
<b>NEMATODES</b>				
<i>Cosmocerca ornata</i>	62.0	3.0		
<i>Capillaria costacruzi</i>	5.6	5.9		S

Prevalence (%): percentage of parasitism.

Intensity (1-3/nº): Protozoa (1-3): between 1 (few specimens) and 3 (a lot of specimens). Trematodes and Nematodes (nº): number of specimens (mean intensity of parasitism per infested frog).

New Host: R (*Rana perezi*).

New Area: IP (Iberian Peninsula) and S (Spain).

Table 3. Small intestine parasites.

	Prevalence (%)	Intensity (1-3/nº)	New Host ( <i>R. perezi</i> )	New Area (IP/S)
<b>PROTOZOA</b>				
<i>Giardia agilis</i>	2.3	2.8		IP
<i>Balantidium duodenii</i>	70.8	1.3		IP
<b>TREMATODES</b>				
<i>Cephalogonimus retusus</i>	14.1	7.1		
<i>Opisthioglyphe endoloba</i>	1.4	2.7		

Prevalence (%): percentage of parasitism.

Intensity (1-3/nº): Protozoa (1-3): between 1 (few specimens) and 3 (a lot of specimens). Trematodes (nº): number of specimens (mean intensity of parasitism per infested frog).

New Host: R (*Rana perezi*).

New Area: IP (Iberian Peninsula) and S (Spain).

*Trypanosoma rotatorium*, *Trypanosoma loricatum*, *Entamoeba ranarum*, *Eimeria prevoti*, *Lankesterella minima*, *Dactylosoma ranarum* and *Balantidium elongatum*; all species of bacteria and protozoa are new area records in the Iberian Peninsula and *Capillaria costacruzi* is a new area record in Spain (it was cited in Portugal).

In *Rana perezi* of Ávila we have found blood parasites and *Rhizomastix* sp., lung trematodes and nematodes, and muscle nematodes which have not been cited in *Rana perezi* of the Canary Islands (Carrera et al., 1987), what can be due to the absence of their intermediate hosts in the islands.

We have also observed an increasing prevalence of parasites with increasing sizes (ages) of these frogs suggesting that longer exposure to water makes these species more likely to acquire parasites.

The identified species were similar to those already reported in the current literature except for those species which are discussed below:

The dimensions of *B. algonquinensis* of Canadian amphibians (*Rana clamitans* and *Rana catesbeiana*) were: 8.4 x 3.2 mm and the recurrent flagellum was about 1.5 times the length of the body (Desser et al., 1993). *B. algonquinensis* of our frogs measured 9 x 8 mm and the recurrent flagellum 9 mm. Therefore our specimens are broader and with a recurrent flagellum shorter than Canadian ones. This can be due to the different geographical area or the different host.

The only species cited previously in the genus *Rhizomastix* Alexeieff, 1911 is *Rhizomastix gracilis* Alexeieff, 1911 from the intestine of axolotls and the larval *Tipula abdominalis* (Alexeieff, 1911; Mackinnon, 1912 and Geiman, 1932). The specimens reported by Ludwig (1946) varied at random from 3.7 mm to 10 mm in length and from 2 mm to 4 mm in

width, with an average of 7.5 x 2.5 mm and the flagellum averaged at least twice the body size; rounded individuals averaged 4.0 mm in diameter. However, all of our specimens are rounded or a bit elongated but bigger than the previously mentioned rounded ones (9 x 7 ± 2 mm) and the flagellum is not twice the length of the body (13 ± 5 mm). This can be due to the different host or geographical area or perhaps it is a new specie or even a new parasite but it would have to be confirmed by electronic microscopy.

In the blood, we have identified 4 species of trypanosomes: *Trypanosoma rotatorium*, *Trypanosoma loricatum* and two undetermined species which have been designated as: *Trypanosoma* sp.1. *Trypanosoma* sp.2. This species presents differences in morphology with other cited species and they will be described as new species in a next article. A mixed infection (of 2 to 4 species) was frequently observed.

We observed epimastigote stages in the leech, *Batracobdella algira*. Other species found in this leech were sporozoites of *Lankesterella minima* and the filarial larvae of *Icosiella neglecta*.

In fecal samples we have seen two kinds of amoebae: *Entamoeba ranarum* and other smaller ones which we have designated as amoebas of free living.

The dimensions of *Dactylosoma ranarum* described by Barta (1991) of Canadian amphibians were: primary merozoites: 4.3 ± 0.4 x 1.3 ± 0.3 mm and gamonts: 7.0 ± 0.7 x 3.4 ± 4 mm, whereas our merozoites were larger (5 ± 1 x 2 ± 1 mm) and the gamonts were twice the length of Canadian ones (14 ± 1 x 2 ± 1 mm) but to be able to consider it a new specie it would have to be confirmed by electronic microscopy.

We found *Blastocystis* in the frogs and we observed that they had a different morphology (they were much bigger) than *Blastocystis hominis* (Stenzel and Boreham, 1996) and different optimal culture requirements in fecal

Table 4. Urinary bladder parasites.

	Prevalence (%)	Intensity (1-3/nº)	New Host ( <i>R. perezi</i> )	New Area (IP/S)
<b>PROTOZOA</b>				
<i>Trichodina ranae</i>	58.6	2.3		IP
<b>TREMATODES</b>				
<i>Gorgodera amplicava</i>	1.9	7.1		
<i>Gorgoderina vitelliloba</i>	7.9	4.8		

Prevalence (%): percentage of parasitism.

Intensity (1-3/nº): Protozoa (1-3): between 1 (few specimens) and 3 (a lot of specimens). Trematodes (nº): number of specimens (mean intensity of parasitism per infested frog).

New Host: R (*Rana perezi*).

New Area: IP (Iberian Peninsula) and S (Spain).

Table 5. Lung parasites.

	Prevalence (%)	Intensity (nº)	New Host ( <i>R. perezi</i> )	New Area (IP/S)
<b>TREMATODES</b>				
<i>Haematoloechus variegatus</i>	3.2	1.7		
<b>NEMATODES</b>				
<i>Rhabdias bufonis</i>	7.9	4.5		

Prevalence (%): percentage of parasitism.

Intensity (nº): number of specimens (mean intensity of parasitism per infested frog).

New Host: R (*Rana perezi*).

New Area: IP (Iberian Peninsula) and S (Spain).

protozoa culture (Boeck and Drbohlav culture) where they grew best at 24° C rather than at 37° C. We, therefore, think that there are two different species. We have also seen that our specimens are very similar to the organisms described by Alexeieff in 1911, so we have decided to take up the name of *Blastocystis enterocola* Alexeieff, 1911 for *Blastocystis* of *R. perezi* of Ávila.

In Sierra de Gredos (Ávila) two *Haematoloechus* species have been cited, *H. variegatus* (Rudolphi, 1819) Looss, 1899 in *Rana perezi* (Combes and Knoepfler, 1965) and *H. carbonelli* Lluch, Navarro and Pérez-Soler, 1991 in *Rana iberica* and *Bufo bufo gredosicola* (Lluch et al., 1991). The measurements of our specimens were more similar to *H. variegatus* than *H. carbonelli*, so we consider that they belong to *H. variegatus*.

In 1942, González Castro observed slight morphometrical differences between *Gorgodera circava* Guberlet, 1920 (in American frogs) and specimens in *R. esculenta* (likely: *R. perezi*) of Granada (Spain) and he created a new variety “*Gorgodera circava var. granatensis* González-Castro, 1942”. However, the literature “mistakenly” collected this variety as a new species “*Gorgodera granatensis* González-Castro, 1942”. The measurements of our specimens were smaller than Spanish ones and more similar to American ones. So we consider that they belong to *Gorgodera*

Table 6. Liver parasites.

	Prevalence (%)	Intensity (1-3)	New Host ( <i>R. perezi</i> )	New Area (IP/S)
<b>PROTOZOA</b>				
<i>Lankesterella minima</i>	41.4	1.6	R	IP

Prevalence (%): percentage of parasitism.

Intensity (1-3): between 1 (few specimens) and 3 (a lot of specimens).

New Host: R (*Rana perezi*).

New Area: IP (Iberian Peninsula) and S (Spain).

Table 7. Muscle parasites.

	Prevalence (%)	Intensity (1-3)	New Host ( <i>R. perezi</i> )	New Area (IP/S)
<b>NEMATODES</b>				
<i>Icosiella neglecta</i>	3.2	2.7		

Prevalence (%): percentage of parasitism.

Intensity (nº): number of specimens (mean intensity of parasitism per infested frog).

New Host: R (*Rana perezi*).

New Area: IP (Iberian Peninsula) and S (Spain).

*circava*, nowadays, synonymous species of *Gorgodera amplicava* Looss, 1899.

We also agree with Combes and Knoepfler (1965 and 1972-1973), Combes and Sarrouy (1971) and Lluch et al. (1986a), that the genus *Gorgodera* has a high variability in the characters considered to be of systematical value and that an exhaustive review of the genus is necessary.

In 1990, Moravec and Barus considered two subspecies within the species *Cosmocerca ornata* (Dujardin, 1845) Railliet and Henry, 1916: *Cosmocerca ornata ornata* Dujardin, 1845 in the temperate zone of Europe and *Cosmocerca ornata japonica* Yamaguti, 1938 in the subtropical regions of Africa and Asia. Our specimens had an intermediate value between the two subspecies due perhaps to the geographical situation of Spain (between Europe and Africa).

*R. perezi* of Ávila, like *R. temporaria* of Pirineo (Lluch et al., 1990) and *R. iberica* of Sistema Central (Navarro et al., 1988) and in contrast to *R. perezi* of Levante (Lluch et al., 1986b), does not have African influence in the trematodes (*Sonsinotrema*, *Ratzia*, *Szidatia* genus that are to be presumed with this origin).

## 5. References

- Alexeieff, A.G. 1911. Notes sur les flagellés. *Arch Zool Exper Gén*, 5, 491-530.
- Barta, J.R. 1991. The Dactylosomatidae. *Adv. Parasit.* 30, 1-37
- Ballesteros-Márquez, A. 1945. Revisión de la familia Cosmocercidae, Travassos, 1925. *Rev Ibér Parasitol*, Tomo extraordinario, 150-180.
- Carrera, M.P.; Zapatero, L.M. and Castaño, C. 1987. Protozoos parásitos de anfibios de anuros de las Islas Canarias. *Rev Ibér Parasitol*, 47, 113-119.
- Combes, C. and Gerbeaux, M.T. 1970. Recherches éco-parasi-tologiques sur l'helminthofaune de *Rana ridibunda perezi* (Amphibien Anoure) dans l'Est des Pyrénées. *Vie Milie*, 21, 121-158.

- Combes, C. and Knoepffler, L.P. 1965. Sur quelques plathelminthes d'amphibiens et de reptiles de la Sierra de Gredos (Espagne). *Vie et Milieu*, 16, 487-495.
- Combes, C. and Knoepffler, L.P. 1972-1973. Helmintes parasites de *Rana ridibunda ridibunda* Pallas 1771 sur les rives iraniennes de la Mer Caspienne. *Vie Milieu*, 23, 329-334.
- Combes, C. and Sarrouy, H. 1971. Helmintes de *Rana ridibunda perezi* (Amphiba) dans la région de Soria. *Rev. Ibér. Parasitol.*, 31, 115-119.
- Da Cunha, X. 1950. *Trichodina ranae* n. sp., un urcéolaire parasite de la vessie urinaire de la Grenouille. Ed. Coimbra Editora, L. da. Coimbra.
- Desportes, C. 1941. Nouvelles recherches sur la morphologie et sur l'évolution d'*Icosiella neglecta* (Diesing, 1851), Filaire commune de la grenouille verte. *Ann Parasitol Hum Comp*, 18, 46-66.
- Desser, S.S.; Hong, G. H.; Siddall, M.E. and Barta, J.R. 1993. An ultrastructural study of *Brugerolleia algonquinensis* gen. nov., sp. nov. (Diplomonadina; Diplomonadida), a flagellate parasite in the blood of frogs from Ontario, Canada. *Eur J Protistol*, 29, 72-80.
- Fernández, P. 1984. Contribución al conocimiento de la helmintofauna del género *Bufo*. Tesis de Licenciatura. Ciencias Biológicas. Universidad de Valencia.
- Fernández-Galiano, D. 1947. Observaciones citológicas sobre las opalinas. *Bol R Soc Española Hist Nat* (Biol.), 1, 353-422.
- Geiman, Q.M. 1932. The intestinal protozoa of the larvae of the crane-fly, *Tipula abdominalis*. *J Parasitol*, 19, 173.
- Gonzalez Castro, J. 1942. Descripción de una variedad de *Gorgodera circava* (Guberlet, 1920) en la vejiga urinaria de la *Rana esculenta* en Granada. *Rev Ibér Parasitol*, 2, 310.
- Lluch, J.; Navarro, P. and Izquierdo, S. 1990. Sobre la helmintofauna de *Rana temporaria* Linnaeus, 1758 en el Pirineo español. *Rev Esp Herpetol*, 67-79.
- Lluch, J.; Navarro, P. and Pérez-Soler, P. 1991. *Haematoloechus carbonelli* sp.n. (Haematoloechidae: Plagiorchiata) un nouveau Trématode parasite d'Amphibiens de la Péninsule ibérique. *Rev Sui Zool*, 98, 255-260.
- Lluch, J.; Roca, V. and Navarro, P. 1986a. Contribución al conocimiento de la helmintofauna de los herpetos ibéricos. III Digenea Paramphistomidae, Hemiuridae, Gorgoderidae, Plagiorchiidae, Haematoloechidae and Cephalogonidae de *Rana perezi* Seoane, 1885 (Amphibia: Ranidae). *Rev Ibér Parasitol*, 46, 387-392.
- Lluch, J.; Roca, V. and Navarro, P. 1986b. Helmintoфаuna de anfibios del Levante Ibérico. Trematodos de *Rana perezi* Seoane, 1885. *Bol R Soc Esp Hist Nat*, (Biol.), 81, 43-49.
- López-Neira, C.R. 1947. Helmintos de los vertebrados ibéricos. Patronato Santiago Ramón y Cajal, C.S.I.C., Granada.
- Ludwig, F. W. 1946. Studies on the protozoan fauna of the larva of the crane-fly, *Tipula abdominalis*. I Flagellates, amoebae, and gregarines. *Trans Am Microscop Soc*, 65, 189-214.
- Mackinnon, D.L. 1912. Protists parasites in the larvae of the crane-fly, *Tipula* sp. Prelim. Note. *Parasitol*, 4, 175-189.
- Madrigal, M.J.; Pedrero, L. and González, P. 1987. Aislamiento de amebas de vida libre en ejemplares de *Rana perezi* de distintos puntos de las Islas Canarias. *Rev Ibér Parasitol*, Vol. Extraordinario, 13-15.
- Martínez-Fernández, A.R.; Simón-Vicente, F. and Cordero del Campillo, M. 1988. On the morphology of *Opisthodiscus nigroviridis* (V. Mehely, 1929) Odening, 1959 (Trematoda: Paramphistomidae) of *Rana ridibunda*. *Rev Ibér Parasitol*, 48, 9-16.
- Moravec, F. and Barus, V. 1990. Some nematode parasites of frogs from Papua New Guinea and Australia. *Acta Soc Zool Bohemoslov*, 54, 268-286.
- Navarro, P.; Izquierdo, S.; Pérez-Soler, P.; Hornero, M. J. and Lluch, J. 1988. Contribución al conocimiento de la helmintofauna de los herpetos ibéricos. VIII. Nematoda Ascaridida Skrjabin et Schultz, 1940 de *Rana* spp. *Rev. Ibér. Parasitol.*, 48 (2), 167-173.
- Navarro, P.; Lluch, J.; Pérez-Soler, P. and Izquierdo, S. 1988. Estimaciones respecto a la helmintofauna de *Rana perezi* Seoane, 1885. *Rev Esp Herpetol*, 3, 69-74.
- Oliveira-Rodriguez, H.; Sodré-Rodrigues, S. and Cristofaro, R. 1973. ContribuVao para o estudo dos Trematodeos parasitos de *Rana esculenta* L. em Portugal metropolitano. *Atas Soc Biol R de J*, 16, 47-52.
- Rodrigues, H.O.; Rodrigues, S.S. and Cristofaro, R. 1972. ContribuVao para o estudo dos nematódeos parasitos de *Rana esculenta* L. en Portugal metropolitano. *Atas Soc Biol R de J*, 16, 21-26.
- Stenzel, D.J. and Boreham, P.F.L. 1996. *Blastocystis hominis* revisited. *Clin Microbiol Rev*, Oct, 563-584.
- Travassos, L. 1932. Sobre dois parasitos de batraquios de Portugal. *Bol Biol R de J*, 21, 61-64.