**Rev Inv Vet Perú 2025; 36(2): e30632**
**https://doi.org/10.15381/rivep.v36i2.30632**

## Report

# Parasitism and anatomo-histopathological lesions associated to infection by *Labiduris irineuta* in a terrestrial yellow-footed tortoise (*Chelonoidis denticulata*) (Linnaeus, 1766), San Martín, Peru: Case report

# Parasitismo y lesiones anatomohistopatológicas asociadas en la infección por *Labiduris irineuta* en una tortuga pata amarilla (*Chelonoidis denticulata*) (Linnaeus, 1766), San Martín, Perú: Reporte de caso

**Víctor H. Puicón N.[1,2]*, Alicia López F.[1,2], Camille Coomansingh Springer[3], Luis A. Gomez-Puerta[4], Alfonso Chavera C.[5]**

## Abstract

The case of a terrestrial South American yellow-footed tortoise (*Chelonoidis denticulata*) with clinical signs of decay and inappetence in a recreation center and tourist accommodation located at the entrance of the Cordillera Escalera, San Martín, Perú is reported. The animal had bilateral blepharitis and greenish ocular secretions, as well as mild respiratory sounds and marked dyspnoea on physical examination. The animal was treated in a veterinary clinic with enrofloxacin 10 mg/kg SC every 24 h and carprofen 2 mg/kg SC every 24 h, however, the animal died five days after treatment. The *post mortem* examination showed 75 parasitic specimens of roundworms in the small intestine compatible with nematodes. Severe pneumonia was found as well as small

*[1] Grupo de Investigación Parasitología Veterinaria y Zoonosis Parasitaria, Escuela de Medicina Veterinaria, Facultad de Medicina Veterinaria, Universidad Nacional de San Martín, Tarapoto, Perú*
*[2] Laboratorio de Histopatología Animal, Escuela de Medicina Veterinaria, Facultad de Medicina Veterinaria, Universidad Nacional de San Martín, Tarapoto, Perú*
*[3] Department of Pathobiology, School of Veterinary Medicine, St. George's University, Grenada, West Indies*
*[4] Laboratorio de Epidemiología y Economía Veterinaria, Facultad de Medicina Veterinaria, Universidad Nacional Mayor de San Marcos, Lima, Perú.*
*[5] Laboratorio de Histología, Embriología y Patología Veterinaria, Facultad de Medicina Veterinaria, Universidad Nacional Mayor de San Marcos, Lima, Perú.*
** Corresponding author: Víctor Humberto Puicón Niño de Guzmán; vhpuicon@unsm.edu.pe*

*Received: July 26, 2024*
*Accepted for publication: March 2, 2025*
*Published: April 30, 2025*

nodules in the intestinal mucosa. The liver was increased in size, rounded edges were evident, areas of yellowish colour focused on all lobes and macroscopic lesions of bronchopneumonia, bloody fluid in the coastal areas and hepatization of the ventral caudal lobe and hepatic steatosis were described. The histopathological lesion occurred with severe parasitic bronchopneumonia, peri bronchial lymphocytic exudate, mild interfibrillar lymphocytic infiltration of cardiac muscle and severe hepatic lipidosis.

**Keywords**: tortoise, clinic, parasitism, anatomohistopathology lesions, bronchopneumonia

## Resumen

Se reporta el caso de una tortuga terrestre de patas amarillas (*Chelonoidis denticulata*) sudamericana con signos clínicos de decaimiento e inapetencia en un centro recreacional y alojamiento turístico ubicado a la entrada de la Cordillera Escalera, San Martín, Perú. El animal presentó blefaritis bilateral y secreciones oculares verdosas, así como ruidos respiratorios leves y disnea marcada al examen físico. El animal fue tratado en una clínica veterinaria con enrofloxacino 10 mg/kg SC cada 24 h y carprofeno 2 mg/kg SC cada 24 h; sin embargo, el animal falleció cinco días postratamiento. El examen *post mortem* mostró 75 especímenes parasitarios de lombrices intestinales compatibles con nematodos en el intestino delgado. Se encontró neumonía severa, así como pequeños nódulos en la mucosa intestinal. El hígado se encontraba aumentado de tamaño, se evidenciaban bordes redondeados, áreas de coloración amarillenta focalizadas en todos los lóbulos y se describieron lesiones macroscópicas de bronconeumonía, líquido sanguinolento en zonas costeras y hepatización del lóbulo ventrocaudal y esteatosis hepática. La lesión histopatológica cursó con bronconeumonía parasitaria severa, exudado linfocítico peribronquial, leve infiltración linfocítica interfibrilar de músculo cardíaco y lipidosis hepática severa.

**Palabras clave:** tortuga, clínica, parasitismo, anatomía, histopatología, lesiones, bronconeumonía

## Introduction

*Chelonoidis denticulata* is a medium to large-sized terrestrial tortoise whose length is 82 cm and 60 kg in weight, although they are usually found in nature with a size of 40 cm and 15 kg in weight (Vogt, 2008). Currently, this species is in a vulnerable IUCN status (SERFOR, 2025) and is present in several of the Amazon basins in the Loreto region (Ferronato y Morales, 2012), Ucayali, Madre de Dios (Ferronato y Morales, 2012; Catenazzi *et al*., 2013), Huánuco (Lehr, 2006), and Pasco (Ferronato *et al*., 2011). Currently, *C. denticulata* has also been reported in several temporary custody areas in the San Martín region.

Human activity severely impacts various populations of land tortoises; factors such as mascotism, as well as beliefs and myths, have contributed to the endangerment of this species within the San Martín region (Zariquiey *et al*., 2016). In the wild, reptiles live in large areas, which exempt them from the stress process, but predisposing them to be easy prey (Zacarías *et al*., 2016). Furthermore, in the San Martín region, wildlife animals such as yellow-footed tortoise are maintained in rescue and custody centers, as well as in ecotourism recreation centers duly certified by the competent authorities according to the wildlife government regulations (OSINFOR, 2020). However, factors such as high population density, interaction with humans and other unusual animal species,

inadequate diet, inadequate temperatures, etc. are generators of stress, this being a state in which it can generate adverse effects in prolonged or sustained periods, potentially causing diseases due to infectious and parasitic agents (Oliveira *et al*., 2001). or even generate irreversible damage or the animal's death (Arrojo, 2002).

Concerning gastrointestinal parasitism in yellow-footed tortoises in Peru, the presence of parasitic species of the genera *Klossinemella, Atractis*, *Ophidascaris* and *Labiduris* has been reported in the Loreto region (Julca *et al*., 2014), *Klossinemella conciliates*, *Paraorientatractis semiannulata*, and *Angusticaecum brevispiculum* in both Pucallpa and Yurimaguas (Salizar y Sánchez, 2007), *Sauricola* sp. in Lima (Chávez *et al*., 2015). The Chelidae family covers the most significant number of helminths (Chapin, 1925), being in nematodes, the reported species *L. gulosa* (Rudolphi, 1819, Schneider 1866), *L. irineuta* (Costa, 1961) and *L. zschokkei* (Linstow, 1899) in tortoises *C. denticulata*, which have been reported in the states of Pará and Rio de Janeiro, Brazil (Costa, 1961; Mascarenhas y Müller, 2021) and the city of Iquitos, Peru (Julca *et al*., 2014). The objective of this case study was to report a case of parasitism by *Labiduris irineuta* in a yellow-footed tortoise (*C. denticulata*) in the San Martín region and to describe lesions associated to its infection.

## Case Description

### History and Clinical Examination

A male tortoise (*Chelonoidis denticulata*), approximately eight years old, was kept at a tourist recreation center at the entrance to the Cordillera Escalera, district of La Banda de Shilcayo, San Martin, Peru. The animal was found isolating itself from the other specimens in its breeding pool, appeared depressed, and displayed respiratory-related clinical signs. Information collected from the center stated that no similar cases previously occurred, nor have there been reports of mortality. New animals have not been introduced to the creep for the last two years.

The clinical examination was carried out, and the animal presented bilateral blepharitis, greenish ocular secretions, mild respiratory sounds, and marked dyspnea with the opening of the oral cavity during breathing. According to the clinical assessment, the animal was transferred to a veterinary clinic located in Tarapoto City, where it was treated with enrofloxacin 10 mg/kg SC every 24 h and carprofen 2 mg/kg SC every 24 h. However, there was no evidence of recovery, and the animal died five days after the start of therapy. The recreation center notified the national competent authority and the necropsy was conducted.

### Macroscopic Examination

Approximately 2 h after the death of the animal, the *postmortem* examination was carried out in the Animal Histopathology Laboratory of the Professional School of Veterinary Medicine, National University of San Martín, Tarapoto, where the macroscopic evaluation of the lesions was performed and samples collected for laboratory analysis.

In the *postmortem* evaluation, no lesions were evident in the stomach at the level of the gastric mucosa. At the small intestine level, the digested food contained many adult parasitic forms: whitish in color, round, and measured 5 mm in length (Figure 1A). The serosal hemoglobin imbibition was observed as a *postmortem* change lesion, with whitish and protruding nodulations of 0.5 to 2.5 mm in length at the mucosa (Figure 1B) and serous layers (Figure 1C).

The liver was increased in size, rounded edges were evident, areas of yellowish color focused on all lobes (Figure 1D), and at the cut surface showed the presence of fat droplets and on the scalpel edges. Additionally, hypostasis was apparent at the level of the right lobe as a *postmortem* change lesion.

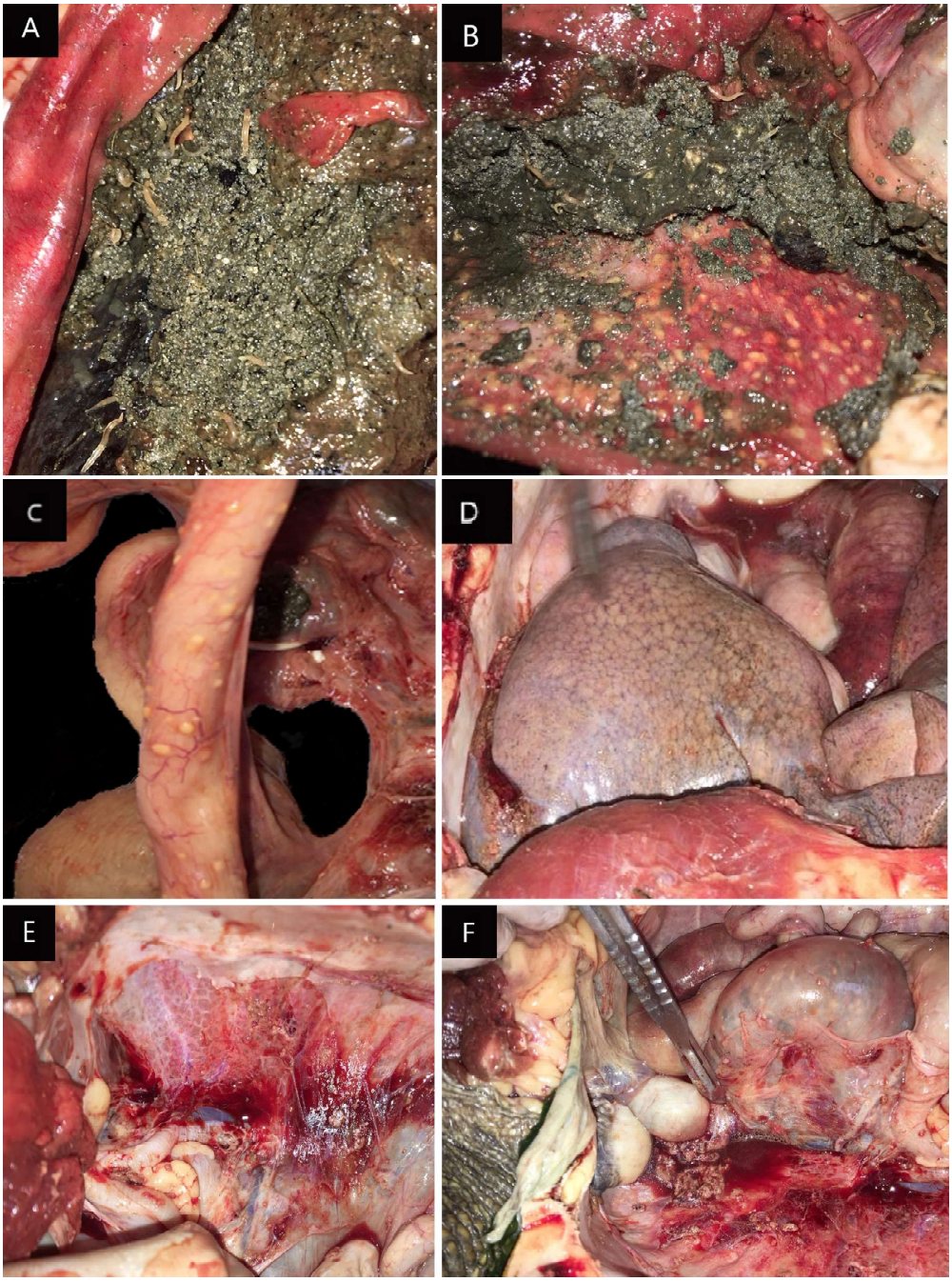


Figure 1. Presence of parasites and anatomopathological lesions in the intestine and abdominal cavity of a tortoise (*Chelonoidis denticulata*). A. *Labiduris irineuta* at the level of the intestinal contents (small intestine). B. Nodulations at the level of the intestinal mucosa. C. Multiple nodulations at the level of the serosa of the small intestine. D. Hepatic lobe with yellowish color and visualization of hepatic lobules. E. Presence of bloody fluid in the coastal areas and adhesions between the lungs, parietal and visceral pleura and digestive tract. F. Adhesions and hepatization of the ventral caudal lobe

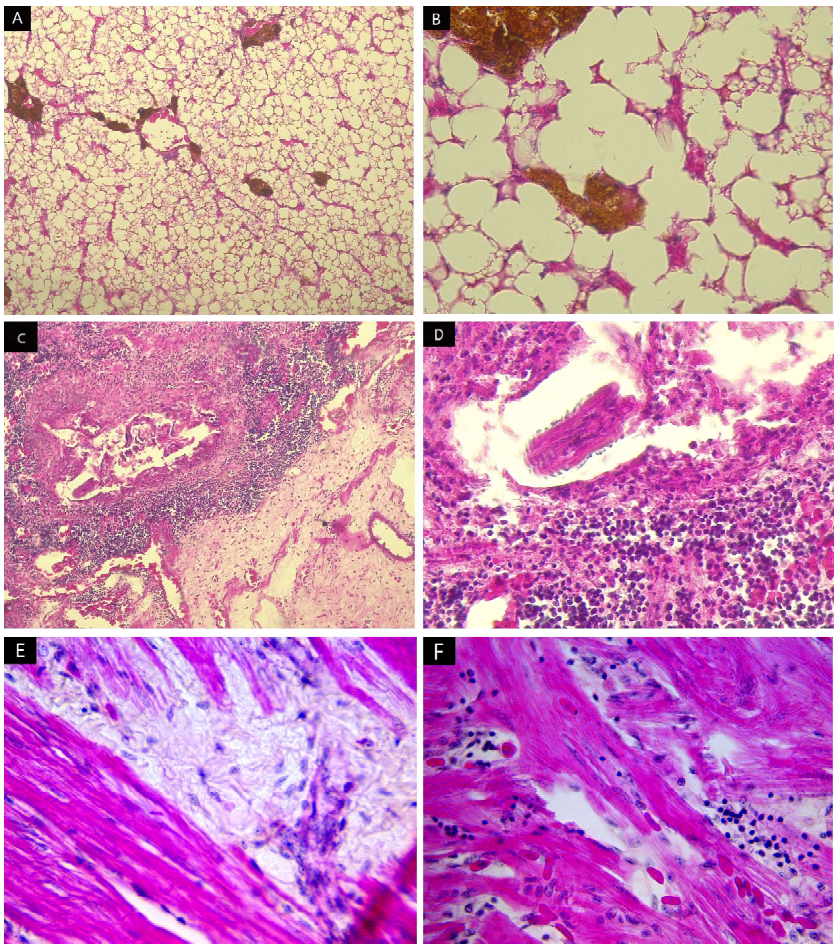


Figure 2. Histopathological lesions in *Chelonoidis denticulata*. A. Liver. Severe hepatic steatosis (10X). B. Liver. Melanomacrophage nodules in the liver parenchyma (40X). C. Lung. Peribronchial lymphocytic exudate. (10X).D. Lung. A thickened cuticle scalloped fragment within a necrotizing bronchiolar luminal epitelium intermingled within severe exudate surrounding (40X)E. Heart. Fibroblasts between muscle fibers. (40X) F. Heart. Mild interfibrillar lymphocytic infiltrates the cardiac muscle (40X). H&E

In the dissection of the laryngeal area, the observation of copious mucoid secretion at the level of the upper respiratory tract was evident. At the level of both lungs, an intense reddish color was evident throughout the lung surface, with bloody fluid and blood clots with fibrinous precipitation that generated a strong adhesion between the lung surface and the walls of the gastrointestinal tract (Figures 1E, 1F).

**Microscopic Examination**

Sampling tissues with macroscopic lesions were sectioned and fixed in 10% buffered formalin and then processed using the histopathological standard procedure consisting of staining with hematoxylin and eosin (H&E) and examined using an optical microscope (Leica DM 750®, USA).

At the microscopic examination level, it was evident that the liver parenchyma had lost its trabecular cytoarchitecture (Figure 2A). The hepatocytes are severely swollen, and the cytoplasm contains unique vacuoles with precise edges and an empty appearance with round to elliptical nuclei of homogeneous size in marginal position. Multiple large cells containing cytoplasm loaded with light brown granular pigment suggestive of melano macrophages, nuclei that are difficult to discern, and congested sinusoid capillaries are observed (Figure 2B). In lungs, moderate peribronchiolar and subpleural multifocal lymphocytic inflammatory cells infiltrating septa and severe congestion were evident. A fragment with a fringed cuticle, thickened external membrane, and muscles without internal organs was observed in a necrotic bronchus. However, it could not be corroborated if it was an arrested parasitic larva (Figure 2C, 2D). At the heart level, mild myocyte hypertrophy, a few foci of mild lymphocytic inflammatory cells, and sporadic fibrous connective tissue infiltration areas were evident (Figure 2E, 2F).

The histopathological diagnoses concluded severe hepatic steatosis, moderate acute interstitial lymphocytic (non-suppurative) pneumonia, and mild chronic multifocal non-suppurative myocarditis.

**Parasitological Examination**

Feces were collected to identify parasitic forms by direct smear and flotation technique. A total of 175 adult parasitic specimens were collected and sent to the Veterinary Epidemiology and Economics Laboratory of the Faculty of Veterinary Medicine of the Universidad Nacional Mayor de San Marcos (Lima, Peru). For taxonomic identification, nine specimens (five females and four males) were clarified by Lactofenol de Amaan to allow the observation of morphological and morphometric characteristics using identification keys, according to Vicente *et al*. (1993) and Anderson *et al*. (2009). Identification was based on the anterior end, cuticle, and sub ventral lips at the level of the anterior end (Figure 3). Measurements were taken of the specimens, length, width, and esophageal bulb, as well as the spicules of males.

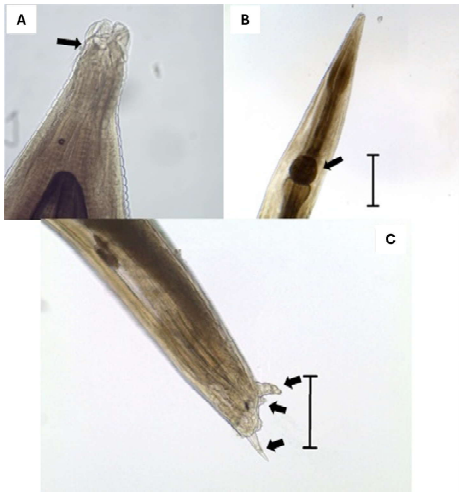


Figure 3. A. *Labiduris irineuta.* Anterior end showing the subventral lips (arrow) 40X. B. Presence of esophageal portions. Scale bars = 500 ìm. C. Posterior end showing the terminal appendage and ventrolateral papillae in the male (arrows). Scale bars = 250 µm

Coprology revealed larval forms in feces (2-3 larvae/ 40X). In adult nematodes, the parasites had sub ventral lips at the anterior end, an esophagus with a thickened and sclerotized anterior part (esophageal bulb), and a thinner posterior portion. Likewise, in the terminal portion of the male, a terminal appendix and ventrolateral papillae were evident. The average measurements were as follows: Length: 4773 µm (minimum: 4283 µm, maximum 5468 µm), width: 528 µm (minimum: 496 µm, maximum 546 µm), length of the esophageal bulb: 219 (minimum: 203 µm, maximum 227 µm), width of the esophageal bulb: 209 (minimum: 196 µm, maximum 221 µm) and spicule: 407 (minimum: 398 µm, maximum 421 µm). Therefore, due to the above characteristics (Table 1), the species correspond to *Labiduris irineuta*.

Table 1. Morphometric measurements of *Labiduris* species applied in the identification of *Labiduris irineuta* (Vicente *et al*., 1993)

| | *L. gulosa* | *L. irineuta* | *L. zschokkei* |
|---|---|---|---|
| | | Male | |
| Length (mm) | 6.0-7.00 | 5.3 | 7.25 |
| Width (µm) | 480 | 320 | 560 |
| Esophageal bulb (Length, µm) | - | 184-210 | 300-400 |
| Esophageal bulb (Width, µm) | - | 184-210 | 300-400 |
| Spicule (µm) | 425 | 323-352 | 450 |

## Discussion

In the present study, a case of moderate parasitism by *Labiduris irineuta* (Costa, 1961) could be determined in a motel tortoise (*Chelonoidis denticulata*) in a custody center in the San Martín region. A previous study carried out on the same species in the city of Iquitos, Loreto, determined the presence of the three species of *Labiduris*, which indicated that in tissues injured by Atractidae parasites, granulomas were found with the presence of nematodes distinguishable by their cuticle and digestive system; however, it could not be determined which species it belonged to (*Labiduris* or *Atractis*) (Julca *et al*., 2014). In the present study, lymphocytic foci with linear structures of thickened cuticle could be observed but without details of the digestive or another system due to tissue necrosis at the lung level, although no eosinophilic cells were observed in these foci. In this sense, Julca *et al*. (2014) mention it as one of the lesions found caused by parasites in the gastrointestinal tract. It should be noted that in the present study, macroscopically, serous nodulations at the intestinal level were determined similarly.

Still, in addition, lymphocytic foci at the lung level could suggest an ectopic lesion of the parasite. However the lack of an eosinophilic exudate, which is characteristic of parasitic processes in tortoises (Romero y López, 2023) and the lack of tissue details required for the identification of a parasite (cuticle, cuticular structures, digestive system composed of columnar cells, platymyarian muscles) as demonstrated in other similar studies (Rideout *et al*., 1987), it could not be shown if this parasitic species causes such lesion outside the gastrointestinal tract.

In the necropsy, it was determined that the cause of death was due to an acute pneumonic process that was corroborated in the histopathology, whose initial clinical suspicion was of a bacterial and or viral agent, so the treatment of choice was the application of a broad-spectrum antibiotic such as enrofloxacin (5 mg/kg). It should be noted that the animal was already suffering from a prior weakening state since the parasite load could have generated hypoproteinemia with the consequent weakness and exhaustion of the immune response.

The origin of the agent that could have developed in the lungs is unknown since bacteriological analyses and antibiograms could not be performed due to the lack of veterinary diagnostic laboratories available during the pandemic period where the case occurred. However, it is worth highlighting those infectious agents are usually transmitted through food, so added to a chronic parasitism condition and an immune system whose innate immunity, although robust, the adaptive (cellular) immunity is primitive and fluctuates

according to seasonal and sexual variations (Ghorai *et al*., 2018).

These factors can cause mortality in these animals, in which the impact of various diseases is not yet adequately understood, but both morbidity and mortality have already been reported in nematodes of the genera *Proatactis*, *Labiduris* and *Atractis* considering them as serious pathogens for tortoises *C. denticulata* and *C. carbonaria* in countries such as Brazil, USA, Grenada, and Peru (Rideout *et al*., 1987; Springer *et al*., 2020; Mascarenhas and Müller, 2021). It is concluded that, although the findings were not evident from parasitism by *Labiduris irineuta*, it can be deduced that moderate to severe parasitism generates death in animals presenting lesions associated with immunosuppression, weakness and secondary infections.

## References


1. ***Anderson RC, Chabaud AG, Willmott S. 2009.*** Keys to the nematode parasites of vertebrates: Archival volume. Cabi: 463 p.
2. ***Arrojo L. 2002.*** Parásitos de animales silvestres en cautiverio en Lima, Perú. Rev Peru Biol 9: 118-120.
3. ***Catenazzi A, Lehr E, May RV. 2013.*** The amphibians and reptiles of Manu National Park and its buffer zone, Amazon basin and eastern slopes of the Andes, Peru. Biota Neotrop 13: 269-283.
4. ***Chapin EA. 1925.*** Nematode parasites of the Brazilian land tortoise, *Testudo delltieulala*. Proc U.S. Nat Museum 65: 1-6.
5. ***Chávez C, Laura, Serrano-Martínez E, Tantaleán M, Quispe M, Casas G. 2015.*** Parásitos gastrointestinales en reptiles en cautiverio en Lima Metropolitana. Rev Inv Vet Perú 26: 127-134. doi: 10.15381/rivep.v26i1.10909
6. ***Costa SCG. 1961.*** Nota prévia sobre um novo nematódeo do gênero *Labiduris* Schneider, 1866 (Atractoidea). Atas Soc Biol Rio J 4: 17-18.
7. ***Ferronato BO, Molina FB, Molina F, Espinosa R, Morales V. 2011.*** New locality records for chelonians (Testudines: Chelidae, Podocnemididae, Testudinidae) from Departamento de Pasco, Peru. Herpetol Notes 4: 219-224.
8. ***Ferronato BO, Morales VM. 2012.*** Biology and conservation of the freshwater turtles and tortoises of Peru. Reptiles Amphib 19: 103-116.
9. ***Ghorai SM, Priyam M. Reptilia: 2018.*** Reptilia: cellular immunity in reptiles: perspective on elements of evolution. In: Advances in comparative immunology. p 773-791.
10. ***Julca R, Casas E, Chavera A, Sánchez L, Sánchez N, Batalla L. 2014.*** Anatomopathological description of lesions of gastrointestinal helminths in motelo tortoises (*Chelonoidis denticulata*). Rev Inv Vet Perú 25: 37-50.
11. ***Lehr E. 2006.*** Book Review-Amphibians of an Amazonian blackwater pond in Peru: Schluter, A. Copeia 4: 834-836.
12. ***Mascarenhas CS, Müller G. 2021.*** Checklist of helminths associated with continental testudines from South America. Neotrop Helminthol 15: 97-126.
13. ***Oliveira T, Moreira N, Eizirik E, Gonçalves R, Crawshaw P, Morato RG. Order Carnivora, Family Felidae (cats). In: Fowler ME, Cubas Z (eds). 2001.*** Biology, medicine, and surgery of South American wild animals. USA: Wiley-Blackwell. p 291-300.
14. ***[OSINFOR] Organismo de Supervisión de los Recursos Forestales y de Fauna Silvestre. 2020.*** Fauna Silvestre en el Perú – Procesos de supervisión, fiscalización y normativa. Lima (Perú): OSINFOR. [Internet]. Disponible en: https://cdn.www.gob.pe/uploads/document/file/1013398/Publicable-Fauna-Silvestre-.pdf
15. ***Rideout BA, Montali RJ, Phillips LG, Gardiner CH. 1987.*** Mortality of captive tortoises due to viviparous nematodes of the genus *Proatractis* (Family Atractidae). J Wildlife Dis 23: 103-108. doi: 10.7589/0090-3558-23.1.103

16. ***Romero-Mera A, López-Flores A. 2023.*** Valores hematológicos de la tortuga de pata roja en cautiverio, mediante hemogramas en la región San Martín. Rev Vet Zootec Amazonica 3: e505.
17. ***Salizar P, Sánchez L. 2007.*** Nuevos registros de nematodos en dos especies de tortugas (Reptilia: Testudines) en el Perú. Neotrop Helminthol 1: 43-45.
18. ***[SERFOR] Servicio Nacional Forestal y de Fauna Silvestre. 2025.*** Guía para la identificación de tortugas terrestres y de agua dulce del Perú. Lima, Perú. SERFOR. 69 p.
19. ***Springer CC, Kinsella M, Vasuki V, Sharma RN. 2020.*** Gastrointestinal parasitic nematodes in pet red-footed tortoises (*Chelonoidis carbonaria*) from Grenada, West Indies. Heliyon 6: e04119. doi: 10.1016/j.heliyon.2020.-e04119
20. ***Vicente J, Oliveira H, Corrêa D, Magalhâes R. 1993.*** Nematoides do Brasil. Parte III: Nematóides de Repteis. Rev Bras Zool 10: 19-168. doi: 10.1590/S0101-81751993000100003
21. ***Vogt RC. 2008.*** Amazon turtles. Lima, Perú: Grafica Biblos. 104 p.
22. ***Zacarias GG, Diaz Gomez JM, de la Fuente MS. 2016.*** Biología, ecología, paleontología y filogenia de nuestra tortuga chaqueña (*Chelonoidis chilensis*): pequeño pariente de las tortugas terrestres de las islas Galápagos. Temas de Biología y Geología del NOA 6: 8-21.
23. ***Zariquiey C, Murillo Y, Cavero N, Chávez G. 2016.*** Guía de reconocimiento. Herramienta para el control del tráfico ilegal de tortugas terrestres y de agua dulce del Perú. Lima, Perú: Wildlife Conservation Society. 50 p .