# **Original paper**

# Investigation of flukes (*Fasciola hepatica* and *Paramphistomum* sp.) parasites of cattle in north-eastern Algeria

# Mohamed Nadir MEGUINI<sup>1</sup>, Souad RIGHI<sup>2</sup>, Mehdi BOUCHEKHCHOUKH<sup>2</sup>, Shehrazed SEDRAOUI<sup>2</sup>, Ahmed BENAKHLA<sup>2</sup>

<sup>1</sup>Department of Veterinary Sciences, Institute of Veterinary and Agronomic Sciences, Mohamed Cherif Messadia University of Souk-Ahras, B.P. 1553, Route de Annaba, 41000, Algeria <sup>2</sup>Department of Veterinary Sciences, Faculty of Sciences and Life, University Chadli Bendjedid El Tarf, BP 73, El Tarf 36000, Algeria

Corresponding Author: Mohamed Nadir Meguini; e-mail: nadirmeguini@gmail.com

**ABSTRACT.** Fasciolosis due to *Fasciola hepatica* is one of the dominant pathologies in Algeria. On the other hand, gastroduodenal paramphistomosis are little studied and little known. Our work consisted of an epidemiological survey in the Souk-Ahras region to investigate these two parasites in cattle. Regarding the epidemiological investigation, it took place in the Souk-Ahras abattoirs, where 530 cattle were inspected for the presence of both parasites. The prevalence of fasciolosis was 12.3%, while the prevalence of paramphistomosis was 7.9% in cattle. Elderly animals were more infected than those under two years of age and females were more infected than males and for both parasites. Therefore, it is important to take into consideration both parasitic diseases and appropriate control measures are strongly recommended to improve cattle productivity.

Keywords: cattle, fasciolosis, paramphistomosis, epidemiology, Souk-Ahras, Algeria

### Introduction

Fasciolosis and paramphistomosis are particularly severe helminthoses that often affect ruminants [1,2]. Considered as one of the major parasitic diseases in north-eastern Algeria [2,3], they are mainly caused by the development of *Fasciola hepatica* in the hepatic parenchyma, the bile ducts, and *Paramphistomum* sp. in the gastric tract [1–5].

Alongside *Fasciola gigantica*, the *F. hepatica* flukes are widespread trematodes in temperate countries and tropical highlands [1–6]. Its life cycle involves *Galba truncatula*, an amphibious gastropod mollusc, as an intermediate host, and the liver and bile ducts of a ruminant as definitive location and host [7]. Humans are also involved but are considered accidental hosts [8]. On the other side, paramphistomosis is digestive helminthosis due to the presence in the abomasum and the small intestine of immature forms of trematodes while the

adult pathogenic forms are living in the rumen [1-10].

*F. hepatica* and the gastroduodenal parasite *Paramphistomum* sp. are sharing the same intermediate host (*G. truncatula*) which makes the epidemiology of diseases caused by these two parasites quite close to each other [7–11]. Often neglected and underestimated by breeders because of their subclinical manifestation, fasciolosis and paramphistomosis are responsible in Algeria for severe economic losses, and several zootechnical and sanitary effects [5–11].

Anthelmintic treatments remain the most used way against these parasites. However, the massive and uncontrolled use of these products has unfortunately led to the installation of a phenomenon of chemoresistance as it has been demonstrated, in Algeria, for the Albendazole specialties [12].

The Souk-Ahras region climate and the large rural population devoted to cattle breeding play a



Figure 1. Presentation of the study area

preponderant role in the accomplishment of the life cycles of these trematodes. However, the lack of data in this region about these two parasites and their intermediate host has led us to conduct an epidemiological in the slaughterhouse investigation in order to update the knowledge on these two flukes.

# **Materials and Methods**

### Study area

This study was carried out in the extreme northeastern Algeria, in the Souk-Ahras region (Fig. 1). This region is geographically divided into three different zones: firstly, the northern zone is

Table 1. Origin, sex and age of slaughtered cattle at the slaughterhouse in the Souk-Ahras region

Origin	Number of cattle	Sex		Age		
		Male	Female	1-3 years	3-5 years	Over 5 years
Merahna	61	57	4	57	0	4
Ouillen	225	219	6	217	1	7
Henancha	28	13	15	12	1	15
Mechrouha	41	8	33	8	2	31
Souk-Ahras	30	16	14	15	2	13
Taoura	71	49	22	45	4	22
Ouled Driss	46	20	26	18	4	24
Tiffech	26	12	14	12	2	12
Khemissa	2	0	2	0	1	1
Total	530	394	136	384	17	129

Pathologies	Infected cattle	Prevalence (%)	
Fasciolosis	65	12.26	
Paramphistomosis	42	7.9	
Associated cases	17	3.2	

Table 2. Prevalence of fasciolosis and paramphistomosis in cattle slaughtered at the Souk-Ahras slaughterhouse

characterized primarily by mountains, cold-dry climate, heavy rainfall exceeding 700 mm/year, with an extensive cattle breeding; then the median zone consists of plains with a subhumid climate, a pluviometry inferior to 700 mm/year, and a semiintensive cattle breeding. Finally, the south zone is represented by large areas with a semi-arid hot-dry climate and low rainfall levels i.e., < 400 mm/year, it is known for sheep and goat farming [13].

### Study animals

The study was conducted in the slaughterhouse in the Souk-Ahras region. All sacrificed cattle come from neighbouring localities; mainly the northern and median zones. To evaluate the importance of parasitism, three epidemiological factors were gathered: the origin, the sex, and the age of each slaughtered animal. To do so, three age categories were defined: young (1–3 years), intermediate (3–5 years) and over five years old.

Of the 530 slaughtered cattle, 225 (42.5%) were from Ouillen, 71 (13.4%) from Taoura, 61 (11.5%) from Merahna, 46 (8.7%) from Ouled Driss, 30 (5.6%) from Souk-Ahras, 26 (4.9%) of Tiffech and 2 (0.4%) were from Khemissa.

Males and young's (1-3 years) constituted the dominant population with 394 (74.3%) and 384 (72.4%), respectively. They were followed by animals over five years old 129 (24.3%) (Tab. 1).

The livers of the slaughtered animals were inspected following a rigorous protocol that started by reporting any changes of shape, colour, volume, or consistency of the inspected livers. Then, two incisions were realized: the first incision was long and shallow; it was realized at the junction of the two main lobes of the liver and near the bifurcation of the biliary trunk while the second was short and shallow and made at the base of Spiguel lobe. The *F. hepatica* worms were extracted by exerting manual pressure on the bile ducts (Fig. 2).

The rumen and reticulum of these animals were also inspected (Fig. 3). An opening was performed along the great curvature, then the ruminal content



Figure 2. Fasciola hepatica infection



Figure 3. Adult *Paramphistomum* sp. attached to the reticulum mucosa

was emptied by returning it.

It was finally rinsed with fresh water and the presence of adult *Paramphistomum* sp. attached to the mucosa was explored [2].

### Statistical analysis

Statistical analyses were carried out using the SPSS v24.0 software (IBM SPSS Statistics for Windows, Version 24.0) and the Pearson's chisquared " $\chi^2$ " test was used to compare the overall prevalence of *F. hepatica* and *Paramphistomum* sp. according to the season, the region, the sex, and age of the cattle.

Parasites Intensity	Low	Medium	Hight
Fasciola hepatica	$\leq 20$	20–50	$\geq 50$
Paramphistomum sp.	$\leq 100$	100-1000	$\geq 1000$





Figure 4. The intensity of fasciolosis and paramphistomosis in cattle

### Results

### Prevalence and intensity of infection

Of the 530 slaughtered cattle, 65 (12.3%) were infected with *F. hepatica* while 42 (7.9%) with *Paramphistomum* sp. (Tab. 2). These two trematodes were associated with 3.2% of the cases and evolve with low and medium intensities.

For the two trematodes, the infection intensity was estimated by determining the number of observed parasites (Tab. 3; Fig. 4).

# Receptivity factors

Seasonal follow-up of the two parasitic diseases (Fig. 5) indicates that fasciolosis infection rates vary between 12% in winter and 14.4% in summer while



Figure 5. Prevalence of fasciolosis and paramphistomosis according to the seasons



Figure 6. Prevalence of fasciolosis and paramphistomosis according to cattle origin



Figure 7. Prevalence of fasciolosis and paramphistomosis according to cattle sex

for paramphistomosis, the highest rates were observed in spring (22.7%) and summer with 12.6%. Nevertheless, no paramphistomosis infection cases were reported in winter.

Monitoring the overall prevalences depending on the animal's origins (Fig. 6) showed that most of fasciolosis (21%) and paramphistomosis (18%) cases were observed in the Mechroha region. Nonetheless, low prevalences were observed in Ouillene (1%) and Khemissa (1%) regions for fasciolosis and Ouled Driss (2%) for paramphistomosis. These results allow us to define a statistically significant ( $\chi^2$  test,  $\rho$ =0.001) influence of the region on infection rates.

Females were more parasitized than males with

respectively 31.6% for fasciolosis and 23.5% for paramphistomosis (Fig. 7).

For both trematodes, cattle aged between three and five years were most frequently infected with *F*. *hepatica* (35.3%) and *Paramphistomum* sp. (29.4%) (Fig. 8). However, the lowest prevalence rates (5.7% and 2.1% respectively) were recorded for cattle of 1–3 years category. Likewise, the sex and age of the animals interfere significatively ( $\chi^2$  test,  $\rho$ =0.001) with the infesction rates.

Monitoring the monthly kinetics of bovine fasciolosis showed an annual activity, with a high prevalence in September (26.3%) and a low rate in November (Fig. 9). On the other hand, bovine



Figure 8. Prevalence of fasciolosis and paramphistomosis according to cattle age categories



Figure 9. Monthly prevalence of fasciolosis in cattle

paramphistomosis monthly prevalences evolve between April and August with stable infection rates (5%). However, after this period, the prevalences decline, and no cases were encountered until March (3%) (Fig. 10).

The influence of the period on prevalences was statistically significant for paramphistomosis ( $\chi^2$  test,  $\rho$ =0.222) while for fasciolosis there was no relation between months and prevalences ( $\chi^2$  test,  $\rho$ =0.002).

### Discussion

Fasciolosis prevalence in the Souk-Ahras region (12.3%) is considered as low when compared to

prevalences reported in several Algerian regions such as El Tarf where the prevalence was 52.4% in 2012 [2] and 26.7% in 2018 [11]; and Jijel with 27.2% in 2004. However, in a recent study conducted in several slaughterhouses of Bejaïa region, the prevalences were very low (2.8%) [3].

Not far from Algeria, the infection was reported in cattle in other Maghreb countries such as Morocco (10.4%), and Tunisia (12.6%) [14,15]. Nevertheless, various surveys on fasciolosis were conducted in Africa with prevalences varying from 12.3% to 30.4% in Egypt [16,17], 7.1% to 15.3% in Nigeria [18,19], and 37.1% in Zimbabwe [20].

Regarding paramphistomosis, the prevalences were also considered as low (7.9%) when compared



Figure 10. Monthly prevalence of paramphistomosis in cattle

to those observed in El Tarf (19.3%) and Jijel (14.5% and 14.6%) [2–5]. Nevertheless, the results presented in this study were close or lower to those found in other countries such as France (20%) [21], Zimbabwe (27%) [22], Egypt (47.1%) [23], and Ethiopia (51.8%) [24].

Through these prevalences, great variability in infection rates can be observed. This variability could be due to several factors, such as the breeding environment of the animals, their age, the biotopes of the snails, the climate, and even the lack of pharmaceutical products used against these trematodoses.

The disparity of the results recorded in our region and those of El Tarf and Jijel can mainly be explained by the fact that the last two regions are temperate wetlands with agro-climatic and ecological characteristics favoring the development of *G. truncatula*.

During our investigation, both trematodoses evolved with low intensities. Our results in terms of paramphistomosis corroborate those previously reported where 56.2% (Jijel) and 53.5% (Mila) of examined ruminants were infected with less than 100 *Paramphistomum* sp. [25]. Similar findings were reported in France and Ireland [21–26] while a parasitic load of 270.46  $\pm$  471.947 was noticed in Ethiopia [27].

These differences in the intensity of infection can be related to several factors such as the abundance of favourable biotopes for *G. truncatula*, the poor health status of the animals, and the application of treatments.

Through the seasonal evolution of fasciolosis, we revealed two infection peaks: the first one

occurred in summer (14.4%) while the second in winter (12%). For paramphistomosis, we observed only one peak in spring (22.7%) and the absence of affected animals in winter. In this context, the risk periods for fasciolosis have been already determined in Algeria and precisely the Jijel region [28]. The authors studied *G. truncatula* main characteristics and monitored their population dynamics. The study showed the presence of two annual generations of molluscs (from October to December and from April to May) therefore two cycles of *F. hepatica* in molluscs.

Besides, high summer prevalences of fasciolosis were already highlighted in studies conducted in Ethiopia and Nigeria. According to these research works, these results may be related to a long prepatent period [29,30].

On the other hand, this investigation allowed us to determine two areas where the cattle are more affected by fasciolosis and paramphistomosis, namely the Northern area and the middle one. This zone-effect phenomenon could be explained by the richness of these two areas in water resources (two large dams, two small dams, and four large hill reservoirs) and their sub-humid climate, thus, favorable biotopes for the development of snails.

In our study, we have highlighted a statistically significant relationship between sex and the infection rate of cattle. Moreover, previous studies have already shown that females were statistically more infected than males [21–32]; this could be related to the immune status of the females during pregnancy and lactation where they could be more prone to infection [33,34].

Moreover, young males of the Souk-Ahras

region were kept in the stable, especially in the region of Ouillene which is known for the fattening and the breeding of bull calves. In this region, females are put to grass more quickly (without any prophylactic plan). Consequently, the males are slaughtered in good health, while the females are usually sent to the slaughterhouse for several health conditions.

On the other side, our findings regarding the effect of age on the receptivity of cattle are in line with previous reports that consider aged cattle to be the main reservoirs of helminthic diseases [35,36]. Indeed, it has been already highlighted that the seizure of parasitized livers increases with advancing age. Previous studies have linked that to the predisposition of aged cattle to health problems and mainly parasitic infections by the weakening of their immune system. However, other reports stated that young cattle (< 2 years old) are more prone to infection [37] and that animals will become immunized and less exposed to infections when aged [38,39].

Regarding paramphistomosis, our results are in line with previous reports which indicate that old cattle are more affected [5–27], mainly due to the weakening of the immune system of the animal [40].

Based on the results of this study, it is concluded that cattle are mostly infected with trematodes parasites. It can be concluded that, despite the limited use of inspection data from slaughterhouses, the information collected can be very useful for a better understanding of the epidemiological data of the parasites studied, in view of a better management of the parasitoses caused.

### References

- [1] Bussiéras J., Chermette R. 1995. Abrégé de parasitologie vétérinaire: Helminthologie vétérinaire. Service de parasitologie, Ecole Nationale Vétérinaire d'Alfort (in French).
- [2] Boucheikhchoukh M., Righi S., Sedraoui S., Mekroud A., Benakhla A. 2012. Principales helminthoses des bovins: enquête épidémiolo-gique au niveau de deux abattoirs de la région d'El Tarf (Algérie). [Main cattle helminthosis: epidemiological investigation at two slaughters in the area of El Tarf (Algeria)]. *Tropicultura* 30: 167-172 (in French with summary in English)..
- [3] Ayad A., Benhanifia M., Balla E., Moussouni L., Ait-Yahia F., Benakhla A. 2019. A retrospective survey of fasciolosis and hydatidosis in domestic ruminants based on abattoirs' data in Bejaia province, Algeria.

Veterinaria 68: 47-51.

- [4] Elliott T., Kelley J., Rawlin G., Spithill T. 2015. High prevalence of fasciolosis and evaluation of drug efficacy against *Fasciola hepatica* in dairy cattle in the Maffra and Bairnsdale districts of Gippsland, Victoria, Australia. *Veterinary Parasitology* 209: 117-124. doi:10.1016/j.vetpar.2015.02.014
- [5] Titi A., Mekroud A., Chibat M., Boucheikhchoukh M., Zein-Eddine R., Djuikwo-Teukeng F.F., Vignoles P., Rondelaud D., Dreyfuss G. 2014. Ruminal paramphistomosis in cattle from northeastern Algeria: prevalence, parasite burdens and species identification. *Parasite* 21: 50. doi:10.1051/parasite/2014041
- [6] Novobilský A., Novák J., Björkman C., Höglund J. 2015. Impact of meteorological and environmental factors on the spatial distribution of *Fasciola hepatica* in beef cattle herds in Sweden. *BMC Veterinary Research* 11: 128. doi:10.1186/s12917-015-0447-0
- [7] Jones R.A., Williams H.W., Dalesman S., Brophy P.M. 2015. Confirmation of *Galba truncatula* as an intermediate host snail for *Calicophoron daubneyi* in Great Britain, with evidence of alternative snail species hosting *Fasciola hepatica*. *Parasites and Vectors* 8: 656. doi:10.1186/s13071-015-1271-x
- [8] Cwiklinski K., O'Neill S., Donnelly S., Dalton J. 2016. A prospective view of animal and human Fasciolosis. *Parasite Immunology* 38: 558-568. doi:10.1111/pim.12343
- [9] Huson K.M., Oliver N.A., Robinson M.W. 2017. Paramphistomosis of ruminants: an emerging parasitic disease in Europe. *Trends in Parasitology* 33: 836-844. doi:10.1016/j.pt.2017.07.002
- [10] Khedri J., Radfar M.H., Borji H., Mirzaei M. 2015. Prevalence and intensity of *Paramphistomum* spp. in cattle from South-Eastern Iran. *Iranian Journal of Parasitology* 10: 268-272.
- [11] Ouchene-Khelifi N., Ouchene N., Dahmani H., Dahmani A., Sadi M., Douifi M. 2018. Fasciolosis due to *Fasciola hepatica* in ruminants in abattoirs and its economic impact in two regions in Algeria. *Tropical Biomedicine* 35: 181-187.
- Bentounsi B. 2001. Parasitologie vétérinaire: helminthoses des mammifères domestiques.
  [Veterinary parasitology: helminthosis of domestic mammals]. Départment Sciences Vétérinaires, Université Mentouri, Algeria (in French). http://livre21.com/livref/F5/F005116.pdf
- [13] Yosmane R., Mebirouk-Boudechiche L., Chaker-Houd K., Abelmadjid S. 2019.Typologie des élevages bovins laitiers de la région de Souk-Ahras (Algérie). *Canadian Journal of Animal Science* 99: 620-630 (in French). https://doi.org/10.1139/cjas-2017-0179
- [14] Moukrim A., Rondelaud D. 1991. [Initial epidemiological data on a focus of animal fascioliasis in the Oued Massa Valley (Morocco)]. *Revue de Médecine Vétérinaire* 11: 839-843 (in French with summary in English).

- [15] Hamed N., Ayadi A., Hammami H. 2014. Epidemiological studies on fasciolosis in northern Tunisia. *Revue de Médecine Vétérinaire* 165: 49-56.
- [16] Atef E.S., Salah W., Haridy F., Mohamed S., Rifaat M., Tosson A. 2002. Fascioliasis among live and slaugthered animals in nine centers of dakahlia governorate. *Journal of the Egyptian Society of Parasitology* 32: 47-58.
- [17] Elshraway N.T., Mahmoud W.G. 2017. Prevalence of fascioliasis (liver flukes) infection in cattle and buffaloes slaughtered at the municipal abattoir of El-Kharga, Egypt. *Veterinary World* 10: 914. doi:10.14202/vetworld.2017.914-917
- [18] Liba J.W., Atsanda N.N., Francis M.I. 2017. Economic loss from liver condemnation due to fasciolosis in slaughtered ruminants in Maiduguri abattoir, Borno State, Nigeria. *Journal of Advanced Veterinary and Animal Research* 4: 65-70. doi:10.5455/javar.2017.d192
- [19] Afolabi O.J., Olususi F.C. 2016. The prevalence of fascioliasis among slaughtered cattle in Akure, Nigeria. *Molecular Pathogens* 7: 1-5. doi: 10.5376/mp.2016.07.0001
- [20] Pfukenyi D.M., Mukaratirwa S.A. 2004. Retrospective study of the prevalence and seasonal variation of *Fasciola gigantica* in cattle slaughtered in the major abattoirs of Zimbabwe between 1990 and 1999. *Onderstepoort Journal of Veterinary Research* 71: 181-187. doi:10.4102/ojvr.v71i3.258
- [21] Szmidt-Adjidé V., Abrous M., Adjidé C., Dreyfuss G., Lecompte A., Cabaret J., Rondelaud D. 2000. Prevalence of *Paramphistomum daubneyi* infection in cattle in central France. *Veterinary Parasitology* 87: 133-138. doi:10.1016/S0304-4017(99)00168-5
- [22] Dube S., Tizauone M. 2014. Paramphistomes in Matebeleland South Province Zimbabwe and their effect on aspects of blood plasma composition in infected cattle. *IOSR Journal of Agriculture and Veterinary Science* 7: 133-138. doi:10.9790/2380-0721133138
- [23] Abu-Elwafa S., Al-Araby M. 2008. Parasitic helminths among animals slaughtered at Dakahlia Province abattoirs. *Mansoura Veterinary Medical Journal* 10: 93-104.
- [24] Ayalew G, Tilahun A., Aylate A., Teshale A., Getachew A.A. 2016. Study on prevalence of *Paramphistomum* in cattle slaughtered in Gondar Elfora Abattoir, Ethiopia. *Journal of Veterinary Medicine and Animal Health* 8: 107-111. doi:10.5897/JVMAH2016.0458
- [25] Titi A., Mekroud A., Sedraoui S., Vignoles P., Rondelaud D. 2010. Prevalence and intensity of *Paramphistomum daubneyi* infections in cattle from north-eastern Algeria. *Journal of Helminthology* 84: 177-181. doi:10.1017/s0022149x09990502
- [26] Toolan D.P., Mitchell G., Searle K., Sheehan M.,

Skuce P.J., Zadoks R.N. 2015. Bovine and ovine rumen fluke in Ireland Prevalence, risk factors and species identity based on passive veterinary surveillance and abattoir findings. *Veterinary Parasitology* 212: 168-174.

doi:10.1016/j.vetpar.2015.07.040

- [27] Sintayehu M., Mekonnen A. 2012. Prevalence and intensity of *Paramphistomum* in ruminants slaughtered at Debre Zeit industrial abattoir, Ethiopia. *Global Veterinaria* 8: 315-319.
- [28] Mekroud A., Benakhla A., Benlatreche C., Rondelaud D., Dreyfuss G. 2002. First studies on the habitats of *Galba truncatula* (Mollusca Gastropoda: Lymnaeidae), the snail host of *Fasciola hepatica*, and the dynamics of snail populations in Northeastern Algeria. *Revue de Médecine Vétérinaire* 153: 181-188.
- [29] Yemisrach A., Mekonnen A. 2012. An abattoir study on the prevalence of fasciolosis in cattle, sheep and goats in Debre Zeit town, Ethiopia. *Global Veterinaria* 8: 308-314.
- [30] Ejeh E., Paul B., Lawan F., Lawal J., Ejeh S., Hambali I. 2015. Seasonal prevalence of bovine fasciolosis and its direct economic losses (del) due to liver condemnation at Makurdi abattoirs north central Nigeria. Sokoto Journal of Veterinary Sciences 13: 42-48.
- [31] Ulayi B., Umaru-Sule B., Adamu S. 2007. Prevalence of *Dicrocoelium hospes* and *Fasciola gigantica* infections in cattle at slaughter in Zaria, Nigeria. *Journal of Animal and Veterinary Advances* 6: 1112-1115.
- [32] Yatswako S., Alhaji N.B. 2017. Survey of bovine fasciolosis burdens in trade cattle slaughtered at abattoirs in North-central Nigeria: the associated predisposing factors and economic implication. *Parasite Epidemiology and Control* 2: 30-39. doi:10.1016/j.parepi.2017.02.001
- [33] Soulsby E., Mönnig H. 1982. Helminths, arthropods and protozoa of domesticated animals. Bailliere Tindall and Cassel Ltd., London.
- [34] Van Veen T.S. 1997. Sense or nonsense? Traditional methods of animal parasitic disease control. *Veterinary Parasitology* 71: 177-194.
- [35] Titi A., Mekroud A., Chibat Mel H., Boucheikhchoukh M., Zein-Eddine R., Djuikwo-Teukeng F.F., Vignoles P., Rondelaud D., Dreyfuss G. 2014. Ruminal paramphistomosis in cattle from northeastern Algeria: prevalence, parasite burdens and species identification. *Parasite* 21: 50. doi:10.1051/parasite/2014041
- [36] Mazeri S., Rydevik G., Handel I., Barend M., Sargison N. 2017. Estimation of the impact of *Fasciola hepatica* infection on time taken for UK beef cattle to reach slaughter weight. *Scientific Reports* 7: 1-15. doi:10.1038/s41598-017-07396-1

- [37] Mekroud A., Benakhla A., Vignoles P., Rondelaud D., Dreyfuss G. 2004. Preliminary studies on the prevalences of natural fasciolosis in cattle, sheep, and the host snail (*Galba truncatula*) in north-eastern Algeria. *Parasitology Research* 92: 502-505. doi:10.1007/s00436-004-1072-1
- [38] Sánchez-Andrade R., Paz-Silva A., Suárez J., Panadero R., Pedreira J., López C., Díez-Baños P., Morrondo P. 2002. Influence of age and breed on natural bovine fasciolosis in an endemic area (Galicia, NW Spain). *Veterinary Research Communications* 26: 361-370. doi:10.1023/a:1016290727793
- [39] Díaz P., Lomba C., Pedreira J., Arias M., Sánchez-

Andrade R., Suárez J., Díez-Baños P., Morrondo P., Paz-Silva A. 2006. Analysis of the IgG antibody response against Paramphistomidae trematoda in naturally infected cattle: application to serological surveys. *Veterinary Parasitology* 140: 281-288. doi:10.1016/j.vetpar.2006.04.007

[40] Euzeby J. 1971. Les fascioloses hepato-biliaires des ruminants domestiques. *Les Cahiers de Medecine Veterinaire* 40: 249-258 (in French).

Received 13 December 2020 Accepted 01 July 2021