ISSN: 1220-3173; E-ISSN: 2457-7618

PARASITIC INVESTIGATIONS IN ENTERIC SYNDROME IN KIDS

INVESTIGAȚII PARAZITARE ÎN SINDROMUL ENTERIC LA IEZI

Olimpia IACOB[1),*)], F. BOGOS[2)], Larisa IVĂNESCU[1),*)], Gabriela Victoria MARTINESCU[1)], Raluca MÎNDRU[1)], D. ACATRINEI[1)], L. MIRON[1)]

ABSTRACT | REZUMAT

The enteric syndrome is the common response of the intestine to aggressions exerted by different etiological agents. The investigations were carried out in the Vaslui area on 155 privately owned kids with the aim of specifying the aetiology of persistent digestive disorders. The kids, aged between 3 and 90 days, were examined clinically and paraclinically. Clinically, changes in the general condition, deviation, apathy, lethargy, inappetence, refusal of food, polydipsia, isolation from the group, abdominal bloating, diarrheal emissions with watery faeces, whitish sometimes with streaks of blood, were observed. Paraclinically, faecal samples were taken from each sick subject and deposited in sterile containers. The samples (50) were examined coproscopically, by qualitative (Wisconsin, Willis) and quantitative (Mc.Master) flotation methods, to highlight specific parasitic elements (cysts, oocysts, eggs); coproantigenic, using the *Giardia* Ag VetExpert Rapid Test Kit and, in stained faecal smears by the modified Ziehl-Neelsen method, for the identification of *Cryptosporidium* oocysts. Examination and photomicrographs were done on a Leica 750 photon microscope using 100x, 200x, and Immersion magnification. Coproscopic results revealed oocysts belonging to species of the *Eimeria* genus, with a medium intensity (OPG: 500-1000), and a prevalence of 24%. Coproantigenic, the species *Giardia duodenalis* was identified in kids, with a prevalence of 14%. In smears, *Cryptosporidium* spp. was identified in kids, with a prevalence of 26 %. The associated evolution of the three parasite genera was found in 10% of the examined samples. These results confirm the multiple parasitic aetiology of enteric syndrome in kids and the negative results obtained in antibiotic therapy. Attention is also drawn to the omission of the parasitic aetiology of the digestive syndrome, which affects young animals in their first months of life, with important economic losses.

Keywords: kids, enteric syndrome, *Cryptosporidium, Giardia, Eimeria*

Sindromul enteric este răspunsul comun al intestinului față de agresiunile exercitate de diferiți agenților etiologici. Investigațiile s-au desfășurat în zona Vaslui, pe un efectiv de 155 iezi proprietate particulară, cu scopul de a preciza etiologia tulburărilor digestive trenante. Iezii, cu vârsta cuprinsă între 3 - 90 zile, au fost examinați clinic și paraclinic. Clinic, s-au observat modificarea stării generale, abatere, apatie, stare de torpoare, inapetență, refuzul hranei, polidipsie, izolarea față de grup, balonament abdominal, emisiuni diareice cu fecale apoase, albicioase uneori cu striuri de sânge. Paraclinic, de la fiecare subiect bolnav s-au prelevat probe de fecale care s-au depus în coprocultoare sterile. Probele (50) au fost examinate coproscopic, prin metode de flotație calitative și (Wisconsin, Willis) și cantitative (Mc. Master), pentru evidențierea elementelor parazitare specifice (chisturi, oochisturi, ouă); coproantigenic, prin utilizarea Kitului de testare rapidă *Giardia* Ag VetExpert și, în frotiuri de fecale colorate prin metoda Ziehl-Neelsen modificată, pentru identificarea oochisturilor de *Cryptosporidium*. S-au analizat intensivitatea (OPG) și prevalența (E%) elementelor parazitare. Examinarea și microfotografierea s-au făcut la microscopul fotonic Leica 750, utilizând magnitudinea 100x, 200x, și Imersia. Rezultatele coproscopice au relevat oochiști aparținând speciilor din genul *Eimeria*, cu o intensivitate medie (OPG: 500-1000), și o prevalență de 24%. Coproantigenic, specia *Giardia duodenalis* a fost identificată la iezi, cu o prevalență de 14 %. În frotiuri, *Cryptosporidium* spp. a fost identificat la iezi, cu o prevalență de 26 %. Evoluția asociată a celor trei genuri de paraziți a fost constatată la 10 % din probele examinate. Aceste rezultate confirmă etiologia parazitară multiplă a sindromului enteric la iezi și rezultatele negative obținute în terapia cu antibiotice. De asemenea, se atenționează asupra omisiunii etiologiei parazitare a sindromului digestiv, care afectează tineretul animal în primele lor luni de viață, cu importante pierderi economice.

Cuvinte cheie: iezi, sindrom enteric, *Cryptosporidium, Giardia, Eimeria*

1) "Ion Ionescu de la Brad" University of Life Sciences, Faculty of Veterinary Medicine, Iasi, Romania
2) S.C. Vanbet S.A., Sălcioara, Vaslui, Romania
*) *Corresponding authors: larisa.ivanescu@iuls.ro; olimpia.iacob@iuls.ro*

In Europe and globally, investigations into enteric syndromes in goats, including parasitic diseases, are crucial due to their impact on animal health and potential zoonotic risks (4). A systematic review highlighted

the complexity of zoonotic enteric parasitic diseases that affect humans and animals. These parasites span across various taxa such as helminths (cestodes, nematodes), and protozoa (*Cryptosporidium, Giardia*), which are transmitted through contaminated food, water, and soil, among other pathways. The review underscored the evolving risks and disease patterns due to human-animal contact, environmental changes, and dietary practices, emphasizing the importance of tailored One Health interventions for communities with close animal contact, such as nomadic and pastoralist populations (21). The discussed global context stresses the importance of continuous surveillance, research, and management strategies for parasitic infections in livestock (5). These efforts are vital for mitigating health risks to animals and humans, especially in regions where traditional livestock management practices prevail. Generally, goats are more resistant to the aggression of pathogens than sheep, but much more sensitive to fluctuations in climate factors (cold and humidity) (13). Public Health Ontario also addresses the broader context of enteric and food-borne diseases, highlighting the ongoing need for robust public health measures, surveillance, and research to combat these infections (Public Health Ontario) (1). Such efforts are critical for understanding and managing the health challenges posed by enteric parasites in goats and other livestock, with implications for animal and human health alike (1). The genera *Eimeria*, *Cryptosporidium*, and *Giardia* are recognized as the main causes of the diarrheal syndrome in kids, lambs, and calves up to about 3 months of age, causing significant economic losses to livestock farmers (2). *Cryptosporidium* and *Giardia* are known causes of diarrhoea in young ruminants, but their impact on the productivity of sheep and goats post-weaning is less understood, with infections often being asymptomatic and concurrent with other pathogens. Recent studies suggest a link between these protozoan parasites and reduced growth in animals, indicating a need for comprehensive research to inform management practices and address both farm profitability and public health concerns (19). The enteric syndrome is the common response of the intestine to aggressions exerted by different etiological agents (3). In a study assessing gastrointestinal parasitic infections among goats in Giza Governorate, Egypt, it was found that 89.33% of goats were infected with gastrointestinal parasites (GIP), with coccidian infection being the most prevalent at 76.89% (17). The study highlighted a significant association between coccidian infections and the age of goats, with yearlings showing the highest prevalence. Mixed infections were common, indicating a broad range of parasitic challenges affecting goats in the region. *Giardia duodenalis*, a significant protozoan parasite, affects both humans and animals, leading to symptoms like diarrhoea and weight loss, and can cause growth retardation in children even without symptoms. This study focuses on identifying the prevalence of *Giardia duodenalis* assemblages in sheep and goats in Shiraz, Iran, to inform preventive measures, acknowledging that assemblage E predominates in livestock, while assemblages A and B are more common in human infections, with B being particularly virulent (10,11). In southern Romania, a high prevalence of cryptosporidiosis was diagnosed among young sheep and goats with diarrhoea, with rates as high as 63.71% and 89.34% respectively (8). Seasonal variations affected prevalence rates in goats at pasture, and molecular studies highlighted *C. parvum* as the dominant strain, underscoring the widespread impact of this parasite in livestock under 3 months old across multiple regions, including western and northwestern Romania (14).

MATERIALS AND METHODS

The research was carried out in the Vaslui area on a herd of privately owned goats. The 155 kids, aged between 3 and 90 days, were examined clinically and paraclinically. The clinical examination was done through a general inspection of the herd and an individual examination of sick kids. For paraclinical examination, 50 faecal samples were taken from each sick subject and deposited in sterile containers. The samples were examined using the coproscopic method, through qualitative (Wisconsin, Willis) and quantitative (Mc.Master) flotation methods, to highlight specific parasitic elements (cysts, oocysts, eggs). The coproantigenic method was also used, using the *Giardia* Ag VetExpert rapid test kit. The VetExpert test is based on the immunochromatographic method (IC). Immunochromatography is a widely-used immunological method; it is a so-called "wet method", which means that a precise result remains as long as the nitrocellulose membrane is wet, and this time period is specified in the leaflet attached to each test. Faecal smears were stained by the modified Ziehl-Neelsen method to identify *Cryptosporidium* oocysts. Diff Quick staining was used to detect trophozoites and cysts of *Giardia* spp. Examination and photomicrographs were performed on a Leica 750 photon microscope using 100x, 200x, and immersion magnification.

RESULTS AND DISCUSSION

Clinical results

Changes in the general condition, deviation, apathy, lethargy, inappetence, refusal of food, polydipsia, isolation from the group, abdominal bloating, diarrheal emissions with watery faeces, whitish, sometimes streaks of blood, were observed in the sick kids (Fig. 1).

Fig. 1. The clinical signs of enteric syndrome in kids

Coproscopic results revealed oocysts belonging to species of the *Eimeria* genus (Fig. 2(a, b, c, d, e, f, g, h.), with a medium intensity (OPG: 500-1000), and a prevalence of 24%.

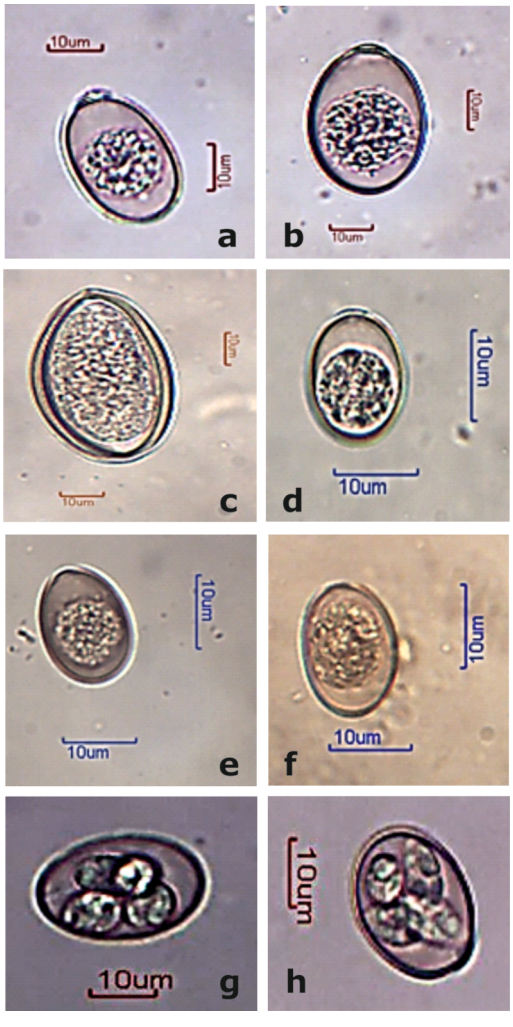

Fig. 2. *Eimeria* spp, immature (a, b, c, d, e, f) and mature (g, h.) oocysts (Willis, 400x)

Coproantigenic results

Coproantigenic, the species *Giardia duodenalis* was identified in kids, with a prevalence of 14% (Fig. 3).

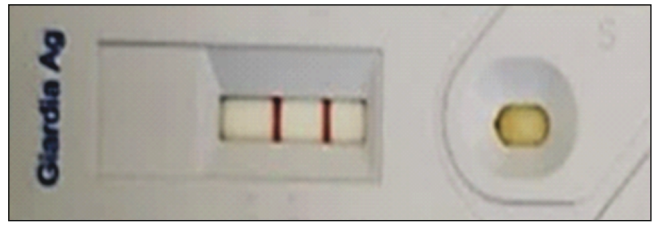

Fig. 3. Coproantigenic result, using the *Giardia* Ag VetExpert Rapid Test Kit

In coloured faecal smears

In smears, *Cryptosporidium* spp. was identified in kids, with a prevalence of 26 % (Fig. 4. a, b).

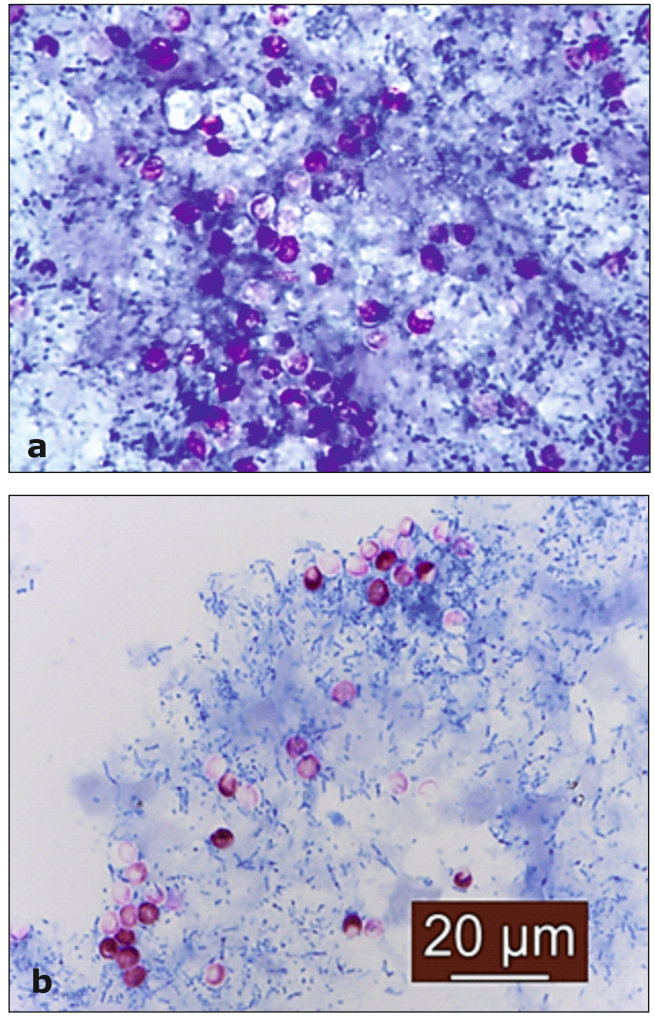

Fig. 4. a, b. *Cryptosoporidum* spp: oocysts identified in diarrheal faeces, in kids with enteric syndrome (Ziehl-Neelsen, Imm x1000)

The co-evolution of parasite species in the same sick subjects: the associated evolution of the three genera of parasites: *Eimeria, Cryptosporidium*, and *Giardia* was found in 10% of the samples examined (5/50). In animal rearing units, the enteric syndrome occurs in young animals during the first months of life with a-

cute or chronic evolution (7). Without knowing the aetiology, antibiotics are usually administered, and the restoration of health is delayed due to numerous biotic and abiotic factors being involved. It is possible that parasitic protozoa were favoured by empiric antibiotic treatments. A correct understanding of the role of protozoan infections on animal health has implications for goat management to reduce the negative impact on farm profitability, animal welfare, and public health risk (16). The involvement of parasitic agents in triggering the enteric syndrome in goats is highlighted by numerous authors (6, 18). The enteric syndrome can be induced by each genus of protozoa, evolving as a standalone parasitosis or in association as a parasitic complex. The prevalence of species from the genus *Eimeria* varies depending on the age of the goats. Thus, in adult goats, O`Callaghan (1989) mentions a prevalence of 97%. The identified species were *E. hirci* (82 %), *E. arloingi* (81%), *E. ninakohlyakimovae* (51%), *E. alijevi* (49%), *E. caprina* (32%), *E. christenseni* (29%), *E. caprovina* (12%), *E. jolchijevi* (9%), and *E. apsheronica* (6%). In the research conducted by Cavalcante et al. (2012) (6), it is shown that the overall prevalence of species from the genus *Eimeria* was 91.2%, specifying that in adult goats the prevalence was 72%, and in kids, 28%. Healthy goats can quite well resist *Eimeria* infections without showing clinical signs, but stressful situations can break the host-parasite balance, leading to clinical eimeriosis or economic losses (6). Young animals are more sensitive due to the reduced capacity for intestinal mucosa regeneration, and with its proliferation, haemorrhagic enteritis often occurs (12). In our research, the overall prevalence of *Eimeria* species in kids was 24%, similar to the values reported by Calvacante et al. (2012). Some studies emphasize that the prevalence of diarrhoea was 21.1% higher for *Cryptosporidium* positive samples (prevalence of 30.3%) compared to *Cryptosporidium* negative samples (prevalence of 9.2%). *C. parvum* was present in 9.9% of positive samples, and *C. ubiquitum* accounted for 16.4% (16). Contact with animals has been identified as a risk factor in many studies, and therefore, zoonotic transmission plays a major role in the epidemiology of cryptosporidiosis (20). In sheep and goats, *C. parvum, C. ubiquitum*, and *C. xiaoi* are the predominant species, but a number of other species, including C*. bovis, C. hominis, C. andersoni, C. suis, C. muris, C. fayeri, C. baileyi, C. ryanae, C. scrofarum, C. canis, C. occultus, Cryptosporidium* sheep genotype I and rat genotype II have been occasionally reported (20). *Cryptosporidium hominis* and *C. parvum* are responsible for about 95% of human infections, followed by *C. meleagridis, C. felis, C. canis, and C. ubiquitum* (20). *Cryptosporidium spp.* cause severe damage to the intestinal mucosal epithelium, by weakening microvilli and increasing the infiltration of mononuclear cells into the *lamina propria*, leading to the occurrence of enteritis (9) The application of whole-genome sequencing (WGS) and amplicon NGS in future studies is important for more accurate tracking of transmission and understanding the mechanisms behind host specificity (20).The average prevalence of *G. duodenalis* infection was 5.0% (34/678) by microscopy after staining with Lugol's iodine, with 5.6% (30/539) for sheep versus 2.9% (4/139) for goats (22). The immune response shows some variability in the host's response either at the individual level or due to internal (genetic) or external factors (physiological status, nutrition), suggesting the possibility to exploit and manipulate this response. There is good evidence from field studies to suggest that a better nutritional plan and a healthy water supply could contribute to improving the resistance of goats (15, 22).

CONCLUSIONS

The obtained results confirm the multiple parasitic aetiology of enteric syndrome in kids and explain, in general, the negative results obtained in antibiotic therapy. In this context, the omission of the parasitic aetiology of the digestive syndrome affecting kids in the first months of life is highlighted, along with the significant economic losses and the risk of contamination of workers with species from the *Cryptosporidium* and *Giardia* genera.

REFERENCES

1. *Al-Habsi K., Yang R., Ryan U., Miller D.W., Jacobson C.*, (2017), Morphological and molecular characterization of three Eimeria species from captured rangeland goats in Western Australia. Vet Parasitol Reg Stud Reports, 9:75-83
2. *Al-Habsi K., Yang R., Williams A., Miller D., Ryan U., Jacobson C.*, (2017), Zoonotic Cryptosporidium and Giardia shedding by captured rangeland goats. Vet Parasitol Reg Stud Reports, 7:32-35
3. *Asghari A., Mahdavi F., Shamsi L., Motazedian M.H., Asgari Q., Shahabi S., Ghalehbin B.M., Sadrebazzaz A.*, (2022), Prevalence and molecular characterization of *Giardia duodenalis* in small ruminants of Shiraz, southwestern Iran: A zoonotic concern, Comp. Immunol. Microbiol. Infect. Dis., 86:101819
4. *Barnes A.N., Davaasuren A., Baasandagva U., Gray G.C.*, (2017), A systematic review of zoonotic enteric parasitic diseases among nomadic and pastoral people. PLoS ONE, 12(11):e0188809
5. *Broglia A., Kapel C.*, (2011), Changing dietary habits in a changing world: emerging drivers for the transmission of foodborne parasitic zoonoses. Vet

Parasitol, 182(1):2-13
6. *Cavalcante R.C.A., Teixeiraa M., Monteiro J.P., Lopes C.W.G.*,(2012),Eimeria species in dairy goats in Brazil. Vet Parasitol, 183:356-358
7. *Dărăbuş G., Imre K., Imre M., Sorescu D.I., Mederle O., Mederle N.*, (2015) Cryptosporidiosis and giardiosis in western Romania: Animal source-reservoir of infection for the human population. Sci Parasitol, 16:58-62
8. *Dărăbuş G., Lupu M.A., Mederle N., Dărăbuş R.G., Imre K., Mederle O., Imre M., Paduraru A.M., Morariu S., Olariu T.R.*, (2023), Epidemiology of *Cryptosporidium* Infection in Romania: A Review. Microorganisms, 11(7):1793
9. *El-Deeb W., Iacob O., Fayez M., Elsohaby I., Alhaider A., Mkrtchyan H.V., Ibrahim A., Alhumam N.*, (2022), Assessment of the immune response of clinically infected calves to *Cryptosporidium parvum* infection. Agriculture, 12(8):1151
10. *Elhadad H., Abdo S., Tolba M., Salem A.I.M.A., Mohamed E.A., El-Abd H.A., Taweel E.*, (2021), Detection of Giardia intestinalis assemblages A and B among children from three villages in the West Delta region, Egypt using assemblage specific primers. J Parasit Dis, 45:655-663
11. *Faridi A., Tavakoli Kareshk A., Sadooghian S., Firouzeh N.*, (2020), Frequency of different genotypes of Giardia duodenalis in slaughtered sheep and goat in east of Iran. J Parasit Dis, 44:618-624
12. *Hohšteter M.*, (2021), Enteric coccidiosis in a kid. Hrvatski Veterinarski Vjesnik/ Croatian Veterinary Report, 29(2):55-57
13. *Iacob O., El-Deeb W., Solcan G., Pavel G.*, (2016), Parasite burdens in Carpathian goats in Romania and associated haematological and biochemical parameters. In FAO Book: Sustenable goat breeding and goat farming Central and Eastern Europe countries. (Ed) Sándor Kukovics, Hungary. (Ed.) Food Agriculture Organization of the United Nations, Rome, Italy, 227-235
14. *Imre K., Dărăbuş G., Ilie M.S., Tamasy L.P.M.*, (2008), Evaluation of some diagnosis methods in cryptosporidiosis. Sci Works, Vet Med, 53:269-276
15. *Imre K., Sala C., Morar A., Ilie M.S., Imre M., Dărăbuș G.*, (2016), *Cryptosporidium* species, genotipes and subtipes contaminating European surface waters: minireview of the last decade. Rev Rom Med Vet, 26(1):46-52
16. *Jacobson C., Khalid A.H., Ryan U., Williams A., Anderson F., Yang R., Abraham S., Miller D.*, (2018), Cryptosporidium infection is associated with reduced growth and diarrhoea in goats beyond weaning. Vet Parasitol, 260:30-37
17. *Kiani-Salmi N., Fattahi-Bafghi A., Astani A., Sazmand A., Zahedi Z., Firoozi B., Ebrahimi A., Dehghani-Tafti U., Ryan F., Akrami-Mohajeri*, (2019), Molecular typing of Giardia duodenalis in cattle, sheep and goats in an arid area of central Iran. Infect Genet Evol, 75:104021
18. *O'Callaghan M.G.*, (1989), Coccidia of domestic and feral goats in South Australia. Vet Parasito., 30(4):267-272
19. *Robertson L.J., Björkman C., Axén C., Fayer R.*, (2013), Cryptosporidiosis in farmed animals. In: Cacciò S.M., Widmer G., (Eds.), Cryptosporidium: Parasite and Disease. Springer, Vienna, Austria, 149-235
20. *Ryan U., Zahedi A., Feng Y., Xiao L.*, (2021), An update on zoonotic *Cryptosporidium* species and genotypes in humans. Animals, 19(11):3307
21. *Stroud C., Kaplan B., Logan J.E., Gray G.C.*, (2016), One Health training, research, and outreach in North America. Infection Ecology & Epidemiology, 1(6)1:33680
22. *Zhang W., Zhang X., Wang R., Liu A., Shen Y., Ling H., Cao J., Yang F., Zhang X., Zhang L.*, (2012), Genetic characterizations of *Giardia duodenalis* in sheep and goats in Heilongjiang Province, China and possibility of zoonotic transmission. PLoS Negl Trop Dis, 6(9):e1826.