NATURAL ENTERIC COCCIDIA INFECTION IN SAND RATS (PSAMMOMYS OBESUS) COLLECTED FROM NORTH COAST, EGYPT

Fatma AS Anwar¹, Gamal H Abed¹, Hossam El-Din M Omar¹,

Mohammed Tolba², Sary Kh. Abdel-Gahfar³

¹Deapartment of Zoology, Faculty of Science, Assiut University;

 ² Department of Parasitology, Faculty of Medicine, Assiut University;
 ³Department of Pathology and Clinical Pathology, Faculty of Veterinary Medicine, Assiut University

Correspondence: Fatma Elzharaa sayed anwar

Department of Parasitology, Faculty of Medicine, Assiut University

City Zip/Country: Assiut, 71515, Egypt.

Tel: 01000689092

Email: fatma.anwar@science.aun.edu.eg

Received: 31/3/2021 Accepted: 19/4/2021 Available Online: 1/6/2021

Parasitic infections in rodents are interesting area of study due to their role as reservoirs for many zoonotic parasites. The current study surveyed the intestinal parasites in 37 adult rats *Psammomys obesus* which captured from saline marsh areas of North Coast after Alexandria, Egypt by Abu Rawash hunters. Rats were investigated for intestinal protozoa with their pathological changes. Eight rats (21.6%) were infected by protozoa of total examined rats represented by *Sarcocystis* (one rat, 2.7%) and *Eimeria* (7 rats 19.9%). Examination of histological sections of small intestine revealed some developmental stages of *Eimeria sp*. Include schizont, male and female gametocytes in the lumen, mucosa and lamina propria of intestine. Degeneration, destruction, sloughing, denudation and hyperplasia of epithelium of intestinal villi were also noticed. In addition hyperplasia of Peyer's patches with lymphocytic infiltration were observed. *Sarcocysist sp.* was found in muscularis and intestinal submucosa with the resultant of destruction of the villi and crypt atrophy of the adjacent cells and discontinuity of the muscularis mucosa due to cyst formation.

Keyword: *Psammomys obesus, Eimeria sp., Sarcocystis sp.,* histopathology.

INTRODUCTION

Zoonotic diseases, especially those associated with rodents and other wildlife; pose a significant threat to human health and wellbeing (Cleaveland et al., 2001). Rodents act as a vital component in various ecosystems either acting as a prey to its predator or as a carrier and reservoir of the diseases (Okoye and Obiezue 2008). Wild rodents act as definitive and/or intermediate hosts of many endoparasites. Some naturally occurring rodent parasites are epidemiologically important and prevalent parasites of humans and domestic animals (Khatoon et al., 2004). Infection in human generally occurs directly through contact with rodent excrement, ingesting food contaminated with their fur, feet, urine or fecal dropping (Singla et al., 2008). Psammomys obesus is a diurnal gerbil's rodent whose range extends from Mauritania to Syria and Saudi Arabia, and to the Red Sea Coast of Sudan (Fichet-Calvet et al., 2000). Available literature showed no record for intestinal protozoa infecting Psammomys obesus and their pathological impacts.

Phylum apicomplexa are obligate intracellular parasites for humans and livestock (**Snow et al., 2005**). Intracellular parasites

reprogram their host for survival and reproduction (Schmid et al., 2014). Eimeria is the largest genus of the apicomplexa with more than 1,800 species (Duszynski, 2011). Eimeria life cycle has exogenous and endogenous phases. Up to 16 species of Eimeria have been described from house mice intestine (Ankrom et al., 1975). The reasons for this diversity are still elusive (Zhao and Duszynski, 2001).

Sarcocystis is a member of the Apicomplexa, class Sporozoasida, and order Eucoccidiorida and family *Sarcocystidae*. The life cycle of *Sarcocystis* is characterized by an alternation of generations, one sexual and one asexual, requiring an alternation of hosts, herbivores and omnivores as intermediate hosts and carnivores as the definitive host (**Ortega-Barria and Dominguez**, **2008; Kim et al., 2011**). *Sarcocystis* can cause pressure atrophy on the adjacent cells, abortion, eosinophilic enteritis, myositis, and subcutaneous nodules (**Kim et al, 2011; Rosenthal, 2020**).

In the present work study the identification of protozoan species naturally infecting sand rats (*Psammomys obesus*) collected from North Coast after Alexandria, Egypt and their pathological impacts on the small intestine.

MATERIALS & METHOD

A-Animals:

In the current study, a total of 37 adult sand rat (*Psammomys obsesus*) weighting 80-171 g were captured from saline marsh areas (Navigations) of North Coast after Alexandria, Egypt by the help of Abu Rawash hunters. Rats were housed in cages and were kept in a room temperature with normal 12 hour light/12 hour dark cycle. They were allowed to acclimatize for one day before sacrificed, and they received their special food plant and tap water throughout the study.

B- Parasitological investigations:

For parasitological studies, faeces of rats were collected intestine. Direct examination and sedimentation from large methods were used (Garcia, 2006). Each sample was processed and examined immediately after collection, by routine direct fecal smear microscopy and concentration techniques using normal saline and Lugol's iodine preparations to record the prevalence of intestinal parasites. Direct wet mount using thin emulsion of small amount of faeces, few drops of saline sometimes add Lugol's iodine had been done. Also, sedimentation techniques (formol ether) methods were used (Garcia, 2006).

C-Specimens processing for pathological examination:

After scarifying the rats, small pieces of each part of small intestine were fixed in 10% neutral buffered formalin for 24 hours. After fixation, all tissue specimens were routinely processed for conventional histopathological examination by light microscopy

(Olympus CX31 with Digital Camera, Japan) according to (Bancroft and Stevens, 1982).

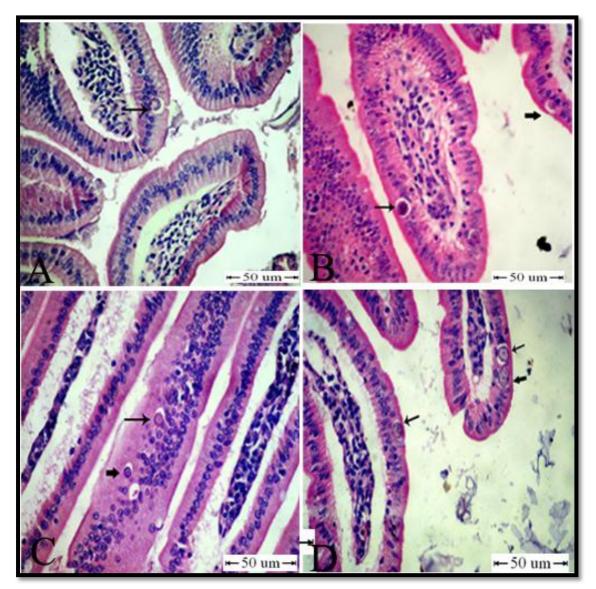
RESULTS

Examination showed no oocyst stage of the collected fecal samples of sand rats. In the present study, two distinctive gametocytes were morphologically identified. Macromeronts were found mainly within the lamina propria, however, micromeronts were found in lamina propria and mucosal epithelial cells. In heavily infected rats, both macro and micromeronts were found in intestinal villi and schizont stage observed in epithelium of intestinal villi (**Plate 1**).

Microscopic examination of small intestine revealed presence of some morphologically developmental stages of Eimeria including male and female gametocyte, and schizont stage in the lumen, mucosa and lamina propria of intestine. The pathological lesions of small intestine revealed degeneration of the intestinal mucosa, Also, there were pressure atrophy of the adjacent intestinal cells, sloughing, destruction and denudation of the intestinal epithelium due to presence of female gamete and schizont stages, with enteritis and presence of hyperplasia of peyer's patches. These infections were usually associated with epithelial hyperplasia (Plate 2).

The lamina propria of affected villi was often contained a mild to moderate increase in numbers of lymphocytes. Necrotic epithelial cells were also seen in some areas (**Plate 2**).

Moreover microscopic examination of small intestine revealed presence of *Sarcocystis sp.* throughout the entire intestinal mucosa and their submucosal and muscularis layers. The histopathological lesions included inflammatory cell reaction, enteritis, destruction of the intestinal epithelium of the villi and crypt. Severe necrotic area in villi and discontinuity of the muscularis mucosa associated with hyperplasia of epithelium of intestinal villi were also observed (**Plate 3**).



46 Fatma AS Anwar, Gamal H Abed, Hossam El-Din M Omar, Mohammed E marghany, Sary Kh. Abdel-Gahfar

Plate (1): Photomicrographs of small intestine from naturally infected rats with *Eimeria sp.* showing developmental stages of *Eimeria sp.* (A) male gametocyte (thin arrow), (B) Schizont stage (thin arrow) and (C and D) female gametocyte (arrows) associated with hyperplasia of epithelium of intestinal epithelium and inflammatory cell infiltrates.

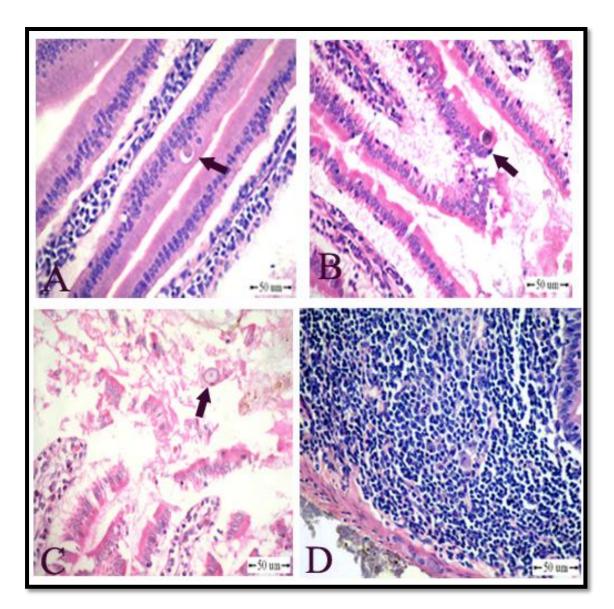


Plate (2): Photomicrographs of small intestine from naturally infected rats with *Eimeria sp.* showing: (A) Male gametocyte (thick arrow) with hyperplasia of epithelium of intestinal villi, presence of lymphocytes infiltration, (B) Schizont (thick arrow), with degeneration of epithelium of intestinal villi and lymphocytes infiltration, (C) Presence of female gametocyte (thick arrow) with

sloughing, destruction and denudation of the villus epithelium. (**D**) Hyperplasia of Payer's patches.

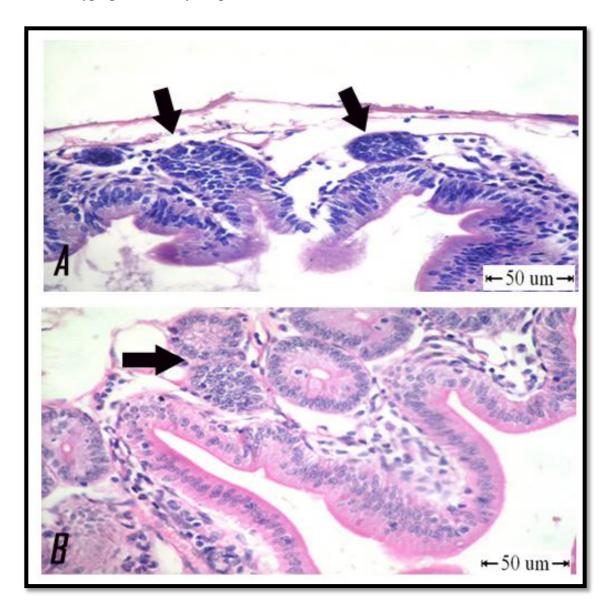


Plate (3): Photomicrographs of small intestine from naturally infected rats with *Sarcocysist sp.* sporulated cyst observed in submucosal and muscularis layer (thick arrows). (A) Necrosis of the villi and crypt associated with hyperplasia of the intestinal epithelium. (B) Inflammatory cell reaction at the lamina propria, associated with hyperplasia of epithelium of intestinal epithelium.

DISCUSSION

Rodents have an important role as hosts of many parasitic agents. Study on their parasites in every geographical area has medical and veterinary importances to prevent transmission of diseases to human and domestic animals (**Kia et al., 2010**). This is the first report identifying infection of sand rat (*Psammomys obesus*) with *Eimeria sp.* and *Sarcoystis sp.* because we cannot find any other reports in the available literature related to coccidian intestinal infection.

Identification of cysts of *Sarcocystis sp.* and different developmental stages of *Eimeria sp.* had been observed with microscopic examination in small intestine of sand rats. The protozoan parasite *Eimeria* sp. is common in the small intestines of wild rats and is considered mildly pathogenic in small numbers. This coccidian has been reported in rodents from various parts of the world including Japan (Kasai, 1978), Colombia (Bonfante et al., 1961), Brazil (Chagas et al., 2017). Similar to our observation (Kasai, 1978) found that *Rattus norvegicus*, and *Rattus rattus*, were infected with *Sarcocystis muris*, *Eimeria nieschulzi*, *Eimeria separata*, *Eimeria miyairii*. The incidence rate of *Sarcocystis*

muris was found to increase with age. Coccidia occurred more frequently in young rats than in old ones.

Moreover, **Chinchilla et al.** (2013) reported that dusky rice rats, *Melanomys caliginosus* (Rodentia, Cricetidae, Sigmodontinae), in a Biological Reserve in Costa Rica to be infected with 2 Eimeria species 8 (89%) which and in the western United States, Baja California, and northern Mexico (Stout & Duszynski, 1983). A high prevalence of the coccidian was found in the studied rats at São Paulo Zoo (Chagas et al., 2017); similar Also, another study conducted in Brazilian animal houses keeping animals intended for research projects: it was found that 60% harboured coccidians (Gilioli et al., 2000).

In the present study, these are the first eimerians described from this rodent genus. However, the parasite was not identified to species level. Identifying coccidian species can be challenging due to their small size and the necessity to first sporulate them in a sodium dichromate solution. From the previous studies the locality of different species of rats correlated to the infection of rats with Eimeria specially the coast country with a high prevalence of the coccidian was recorded.

In the present study we found *Eimeria sp.* in the small intestine in 7 out of 37 examined rats (18.9%). Small intestine of the infected rats showed mucosal degeneration, atrophy of villi and intestinal glands, pressure atrophy of the adjacent cells, hyperplasia of epithelium and Peyer's patches, sloughing, destruction and denudation of the villi due to presence of female

Similarly, Blagburn and Todd gamete and schizont stages. (1984) found histopathological changes in the small intestine of infected Eimeria Mus musculus by vermiformis includes neutrophilic and mononuclear cell infiltrations; villus atrophy and crypt hyperplasia with necrotic debris filled the cryptal and intestinal lumina. Also, Patra et al. (2011) found severe damage of the lamina propria of the intestinal mucosa with numerous coccidian developmental stages in the epithelium of small intestine in naturally infected rats by Eimeria nieschulzi. Lindsay and Todd (1993) and Decker et al. (2001) found Eimeria sigmodontis and Eimeria tuskegeensis in infecting Kangaroo rats (Dipodomys ordii and Dipodomys merriami) and mentioned that their pathogenicity are moderate.

In the present study we found that fat sand rat infected by *Sarcocystis* (one rat 2.7%) in the entire intestinal mucosa and their muscularis layer. *Sarcocystis cymruensis* was originally described in Norway rats in England (**Ashford 1978**). Subsequently, this parasite was reported from a Norway and black rat in Egypt (**Jäkel et al. 1996**). In the present study histopathological observation of rats infected by *Sarcocystis* include pressure atrophy of the adjacent cells of intestine, enteritis, destruction of the villi and crypt. These observations were in agreement with **El Kersh et al. (2016)** who found histopathological changes in the intestinal tissues of rats infected by *Sarcosystis*. In our study observed *Sarcosystis sp.* in mucosa, submucosa and muscularis with slight infiltration of inflammatory cells. Similarity, **Jäkel et**

al. (1997) identified Sarcocystis spp. in the skeletal muscles of wild rodents (*Rattus spp. and Bandicota indica*) mainly captured in the central plains of Thailand. Moreover, Smith and Frenkel (1978) found sarcocysts in skeletal muscles lab mice housed in the same room as cats that had shed sporulated sporocysts of *Sarcocystis muris*.

In conclusion, the present study gives the first overview on the endoparastic protozoa infection of trapped rodents from saline marsh areas (Navigations) of North Coast after Alexandria. Two genera of parasites infecting this species of rats were observed. Further studies should be conducted in other parts of Egypt to discuss the most significant factors predicting the route of infections, the prevalence and intensity of infection of the parasites and their relation to the habitat of the host.

REFERENCES

Ankrom, S. L., Chobotar, B., & Ernst, J. V. (1975). Life cycle of Eimeria ferrisi Levine & Ivens, (1965) in the mouse, Mus musculus. *The Journal of Protozoology*, 22(3), 317-323.

Ashford, R. W. (1978). Sarcocystis cymruensis n. sp., a parasite of rats Rattus norvegicus and cats Felis catus. Annals of Tropical Medicine & Parasitology, 72(1), 37-43.

Bancroft, J. D., & Stevens, A. (1982). Theory practice of histological examination. New York.

Blagburn, B. L., and Todd JR, K. S. (1984). Pathological Changes and Immunity Associated with Experimental Eimeria Vermiformis Infections in Mus Musculus 1. *The Journal of protozoology*, *31*(4), 556-561.

Bonfante, R., Faust, E.C., Giraldo, L.E., (1961). Parasitologic surveys in cali, department del valle, Colombia. IX. Endoparasites of rodents and cockroaches in ward silor, cali, Colombia. J. Parasitol. 47 (5), 843–846.

Chagas, C.R.F., Gonzalez, I.H.L., Favoretto, S.M., Ramos, P.L., (2017). Parasitological surveillance in a rat (Rattus norvegicus) colony in S~ao Paulo Zoo animal house. Ann. Parasitol. 63 (4), 291–297.

Chartier, C., & Paraud, C. (2012). Coccidiosis due to Eimeria in sheep and goats, a review. *Small Ruminant Research*, *103*(1), 84-92.

Chinchilla, M., Valerio, I., Sánchez, R., González, A., Martínez, L., & Duszynski, D. W. (2013). Two new Eimeria spp.(Apicomplexa: Eimeriidae) from the dusky rice rat, Melanomys caliginosus, Tome 1860, in Costa Rica. *The Journal of parasitology*, *99*(1), 82-84.

Cleaveland, S., Laurenson, M.K., Taylor, L.H., (2001). Diseases of humans and their domestic mammals: pathogen characteristics, host range and the risk of emergence. R. Soc. 356, 991–999. https://doi.org/10.1098/rstb.2001.0889.

Daly, M., & Daly, S. (1973). On the feeding ecology of Psammomys obesus (Rodentia, Gerbillidae) in the Wadi Saoura, Algeria. Mammalia, 37(4), 545-561.

Decker, K. H., Duszynski, D. W., & Patrick, M. J. (2001). Biotic and abiotic effects on endoparasites infecting Dipodomys and Perognathus species. *Journal of Parasitology*, 87(2), 300-307.

 Duszynski, D.W., (2011). Eimeria. In: ELS. John Wiley & Sons, Ltd,

 Chichester,
 UK,
 pp.1192–

 1196.https://doi.org/10.1002/9780470015902.a0001962.pub2.

El Kersh, W. M.; Afifi, A. F., El Aswad, B. E. W., & Hussein, N. M. A. (2016). A histopathological and ultrastructural study on experimental murine sarcocystosis. Menoufia Medical Journal, 29(4), 862.

Fichet-Calvet, E., Jomâa, I., Ben Ismail, R., & Ashford, R. W. (2000). Patterns of infection of haemoparasites in the fat sand rat, Psammomys obesus, in Tunisia, and effect on the host. *Annals of Tropical Medicine & Parasitology*, *94*(1), 55-68.

Garcia, L. S. (2006). *Diagnostic medical parasitology*. American Society for Microbiology Press.

Gilioli R., Andrade L.A.G., Passos L.A.C., Silva F.A., Rodrigues D.M., Guaraldo A.M.A. 2000. Parasite survey in mouse and rat colonies of Brazilian laboratory animal houses kept under different sanitary barrier conditions. Arquivo Brasileiro de Medicina Veterinaria e Zootecnica 52: 33-37.

Jäkel T, Burgstaller H, Frank W. (1996). Sarcocystis singaporensis: studies on host specificity, pathogenicity, and potential use as a biocontrol agent of wild rats. J Parasitol 82:280–287.

Jäkel, T., Khoprasert, Y., Sorger, I., Kliemt, D., Seehabutr, V., Suasa-Ard, K., & Hongnark, S. (1997). Sarcosporidiasis in rodents from Thailand. *Journal of wildlife diseases*, *33*(4), 860-867. KASAI, Y. (1978). Studies on helminth and protozoan parasites of rats in Sapporo. *Japanese Journal of Veterinary Research*, *26*(1-2), 31-31.

Kasai, Y., (1978). Studies on helminth and protozoan parasites of rats in Sapporo. Jpn. J. Vet. Res. 26 (1-2).

Khatoon N, Bilqees FM, Shahwar D, Rizwana AG (2004). Histopathologic alterations associated with Syphacia sp. (Nematode) in the intestine of Nesokia indica. Turkish Journal of Zoology 28, 345–351.

Kia, E. B., Shahryary-Rad, E., Mohebali, M., Mahmoudi, M., Mobedi, I., Zahabiun, F., & Vatandoost, H. (2010). Endoparasites of rodents and their zoonotic importance in Germi, Dashte–Mogan, Ardabil Province, Iran. *Iranian Journal of Parasitology*, *5*(4), 15.

Kim, T. H., Han, J. H., Chang, S. N., Kim, D. S., Abdelkader, T. S., Seok, S. H., & Park, J. H. (2011). Detection of sarcocystic infection in a wild rodent (Apodemus agrarius chejuensis) captured on Jeju island. *Laboratory animal research*, 27(4), 357.

Kim, T. H., Han, J. H., Chang, S. N., Kim, D. S., Abdelkader, T. S., Seok, S. H., & Park, J. H. (2011). Detection of sarcocystic infection in a wild rodent (Apodemus agrarius chejuensis) captured Jeju on island. *Laboratory* animal research, 27(4), 357.Ivens, 1965 in the mouse, Mus musculus. The Journal of Protozoology, 22(3), 317-323. Levine, N. D. (1986). The taxonomy of Sarcocystis (protozoa, apicomplexa) species. The Journal of parasitology, 372-382.

Lewis, D. C., & Ball, S. J. (1983). Species of Eimeria of small wild rodents from the British Isles, with descriptions of two new species. *Systematic parasitology*, *5*(4), 259-270.

Lindsay, D. S., & Todd, K. S. (1993). Coccidia of mammals. *Parasitic* protozoa, 4, 89-131.

Mendelssohn, H., & Yom-Tov, Y. (1999). A report of birds and mammals which have increased their distribution and abundance in Israel due to human activity. *Israel Journal of Zoology*, *45*(1), 35-47.

Okoye, I. C., & Obiezue, R. N. N. (2008). A survey of the gut parasites of rodents in Nsukka ecological zone. *Animal Research International*, 5(2).

Ortega-Barria, E., & Dominguez, E. (2008). Sarcocystis Species. In *Principles and Practice of Pediatric Infectious Disease* (pp. 1266-1267).

Patra, G., Ali, M. A., Chanu, K. V., Lalsiamthara, J., Kataria, J. L., Malsawmkima, D., & Devi, L. I. (2011). Molecular diagnosis of natural infection with Eimeria nieschulzi in laboratory rats. *Research Journal of Parasitology*, *6*, 43-52.

Perry, B. D., & Randolph, T. F. (1999). Improving the assessment of the economic impact of parasitic diseases and of their control in production animals. *Veterinary parasitology*, *84*(3-4), 145-168.

Rosenthal, B. M. (2020). Sarcocystosis. In *Hunter's Tropical Medicine* and *Emerging Infectious Diseases* (pp. 821-824). Content Repository Only!. Schmid, M., Heitlinger, E., Spork, S., Mollenkopf, H. J., Lucius, R. and Gupta, N. (2014). Eimeria falciformis infection of the mouse caecum identifies opposing roles of IFN γ -regulated host pathways for the parasite development. Mucosal immunology, 7(4), 969-982.

Singla, N., Singla, L. D., & Kaur, R. (2008). Rodents as museum of helminthic parasites of public health importance in Punjab, India. *International Journal of Infectious Diseases*, *12*, e381-e382.

Smith, D. D. and Frenkel, J. K. (1978): Cockroaches as vectors of Sarcocystis muris and of other coccidia in the laboratory. The Journal of parasitology, 315-319.

Snow, R.W., Guerra, C.A., Noor, A.M., Myint, H.Y. and Hay, S.I. (2005). The global distribution of clinical episodes of Plasmodium falciparum malaria. Nature 434, 214–217.

Stout, C. A., & Duszynski, D. W. (1983). Coccidia from kangaroo rats (Dipodomys spp.) in the western United States, Baja California, and northern Mexico with descriptions of Eimeria merriami sp. n. and Isospora sp. *The Journal of parasitology*, 209-214.

Webster, J.P., Macdonald, D.W., (1995). Parasites of wild brown rats (Rattus norvegicus) on UK farms. Parasitology 111, 247–255.

Zhao, X., & Duszynski, D. (2001). Molecular phylogenies suggest the oocyst residuum can be used to distinguish two independent lineages of Eimeria spp. in rodents. Parasitology Research, 87(8), 638-643.

الاصابة الطبيعية للجرذان الرميلية بالاوليات المعوية التى تم اصطيادها من الاصابة الطبيعية للجرذان الساحل الشمالي، مصر

فاطمة الزهراء سيد أنور '،جمال حسن عابد'،حسام الدين محمد عمر"،محمد عيسي مرغني ⁺،ساري خليل عبد الغفار[°]

> ^امدرس مساعد تخصص طفيليات بقسم علم الحيوان - كلية العلوم – جامعة أسيوط ^٧أستاذ الطفيليات بقسم علم الحيوان - كلية العلوم – جامعة أسيوط ^٦أستاذ الفسيولوجي بقسم علم الحيوان – كلية العلوم – جامعة أسيوط ^٤أستاذ الطفيليات – كلية الطب- جامعة أسيوط

°أستاذ الباثولوجي والباثولوجية الاكلينيكة - كلية الطب البيطري- جامعة أسيوط

تعد العدوى الطفيلية في القوارض مجالًا مثيرًا للدراسة نظرًا لدور ها كعوائل مخزنية للعديد من الطفيليات الحيوانية. في الدراسة الحالية تم فحص سبعة وثلاثون جرذ رملي دهني obesus تحماسيد الإسكندرية اصطيادها من مناطق المستنقعات المالحة بالساحل الشمالي بعد الإسكندرية بمصر بواسطة صيادي أبو رواش. تم فحص براز وأمعاء الجرذان بحثًا عن البروتوزوا المعوية المرضية حيث وجد أن ثمانية جرذان فقط بنسبة 21.6٪ مصابة بنوعين من الأوليات: جرذ واحد بنسبة 2.5% مصاب بالساركوسيستس و سبعة جرزان بنسبة 19.9 % مصابة بالإيميريا . الفحص المجهري للمقاطع النسيجية للأمعاء الدقيقة أظهر وجود الشيزونت والمشيج المذكور والمؤنث لطفيل الإيميريا في تجويف الامعاء والغشاء المخاطي المبطن للامعاء الدقيقة أظهر الساركوسيستس وجدت في الطبقة العضاء الدقيقة و بينما حويصلات تشمل رشح للخلايا الليمفاوية مع تضخم الغدد الليمفاوية، وجود خلايا ميت في الطبقة المخاطية، تهتك في الطبقة العضاية نتيجة وجود حويصلات الساركوسيستس وانساخ الخلايا المبطنة للطبقة المخاطية للامعاء. وخلصت الدراسة إلي إصابة الجرذان بنوعين من الأوليات في الأمعاء الدقيقية حيث كان لهما تأثير باثولوجي علي الأمعاء الدقيقة و توصي الدراسة بفحص عدد أكبر من هذه الجرذان لما تمثلة من خطر علي صحة الحيوان والأنسان كعوائل مخزنية لهذه الطفيليات