

## **SARCOSPORIDIOSIS AS AN EMERGING ZOO NOTIC DISEASE**

**By**

**Sabry, M. A; and Shalaby, H.A\***

*Department of Zoonoses, Fac. Vet. Med., Cairo Univ., Egypt.*

*\*Dept. Parasitology and Animal Diseases, National Research Center, Dokki,*

### **SUMMARY**

*The present study determines the relation between the high incidence of animal sarcosporidiosis and its reflection on human sarcocystis in Egypt. This was adopted using modified dot-ELISA technique for diagnosis of specific anti-bodies in sera of animals and man as well as examination of slaughtered animal muscles.*

*The incidence of infection in slaughtered buffaloes under 2 years old was 10.6% while it was 69.5% in animals over 7 years old using macro & microscopic techniques for examination.*

*This percentage was increased up to 19.33% (at age under 2 years old) and 88.5% (over 7 years old) by using dot-ELISA technique on sera of the same animals.*

*Serological examination of selected group of patients revealed that 37.5% of patients complained of myositis accompanied with eosinophilia as well as 8.33% of patients complained of digestive disturbances had detectable anti-sarcocystis antibodies in their sera without presence of any parasitic stages in their stool. It was worthy to mention that dot-ELISA succeeded to exclude cross reaction between anti-Sarcocystis antibodies and that of other tissue parasites related to the previous symptoms as, *Trichinella spiralis*, *Cysticercus bovis*, *C. cellulosa*, *Toxocara canis* and *Toxoplasma gondii* as well as two common diseases infect human liver include schistosomiasis and viral hepatitis.*

*This study reflected the value of Sarcocystis as an emerging zoonotic problem especially with presence of high incidence of infection in animals around human. This could increase suitability of human to infect by new species of this parasite other than that already known.*

## INTRODUCTION

Sarcosporidiosis is a common coccidian parasitic disease of animals caused by different species of *Sarcocystis*. This parasite has a mandatory two-host life cycle infect a wide range of domestic and wild animals.

It is belonging to the class sporozoa of phylum Apicomplexa. The sexual stages are in the intestine of a carnivore (final host) and asexual stages are in the vascular system and musculature of herbivore (intermediate host) (Prestwood et al., 1980). On the other hand, Rommel (1989) recorded that, not only carnivorous and omnivorous mammals but also birds of prey, owls and reptiles are definitive hosts of *Sarcocystis* species.

Certain species of *Sarcocystis* can cause reduced weight gains, low feed efficiency, anorexia, anemia, abortion, debilitation and mortality in livestock (Dubey and Fayer, 1983).

In Egypt, *Sarcocystis* is one of the most prevalent parasitic infections of buffaloes. Its incidence in slaughtered animals (by micro & macroscopic cysts) was very high figured up to 94% and 97.7% in buffaloes less than 2 years and over 5 years old, respectively (Nassar, 1982). Mohamed (1988) mentioned that esophagus was the main predilection site for sarcosporidiosis infection in female buffaloes slaughtered in Cairo abattoirs. He added that, the percentage of infection reached to 66.66% and 92.08% in animals less than 2 years and that over 5 years old respectively. The rate of infection was 76.77 % in Assiut abattoirs (Said, 1996), while it was 72.63 %, in slaughtered buffaloes at Qena Governorate using ELISA technique (Fawaz, 1998).

On the other side, zoonotic value of this parasite is still under investigation, although humans may serve as intermediate or definitive hosts for different *Sarcocystis* species. This was emphasized by Cook and Zumla (2003), who cleared that only the sexual stage of these species had been demonstrated in the intestine of humans.

Arness et al. (1999) mentioned that humans might become accidental intermediate hosts for a number of species of *Sarcocystis* (*Sarcocystis bovi hominis* and *S. sui hominis*) and the infection was acquired by these two species in humans by eating

insufficient cooked meat of infected beef or pork, respectively. They added that, *Sarcocystis* stages had been demonstrated in human tissues. A total of 52 confirmed cases of human muscular *Sarcocystis* were reported worldwide, including reports of 41 skeletal and 11 myocardial muscle infections. Of these, 10 were associated with myalgia and / or myositis and 13 (25 %) were acquired in Malaysia. Peripheral blood eosinophilia had been reported in two previous cases of *Sarcocystis* myositis.

Gottstein, (1995), stated that *Sarcocystis* was relatively harmless intestinal parasite in humans. The man, from tropical areas, could also become an intermediate host for certain *Sarcocystis* species, which potentially represented a source of opportunistic infection and disease in areas with increasing HIV prevalence.

Diagnosis of animal *Sarcocystis* based mainly on microscopic and macroscopic examination of muscle. While its detection in human depended on serological techniques. Assessment of an easily applied serological technique as dot ELISA, especially if it is able to exclude the cross-reaction with other muscle parasites, may be aid in specific diagnosis of this disease between animal and human.

In this respect, Pappas et al (1983) described a more rapid, economical, direct, visual read, and improved ELISA technique for the diagnosis of visceral leishmaniasis as microenzyme-linked immunosorbent assay (dot-ELISA). Zimmerman et al (1985) and De Morilla et al (1989) used this technique for diagnosis of other parasites. They cleared that nanogram quantities of parasite antigen, were dotted onto very small piece of nitrocellulose membranes, considered enough to obtain a marked accurate diagnosis for the parasite directly.

The present study was initiated to throw more light on value of *Sarcocystis* as zoonotic parasite transmitted from buffaloes (main herbivores host in Egypt) to human. This from the aspect of its distribution in slaughtered buffaloes muscles, presence of its specific anti-bodies in the same animal sera and its role as a cause of myositis and digestive disturbances in selected group of patients using dot ELISA technique.

This study also focussed on minimizing the cross reactivity with the most common cyst-forming parasites in human and animal as *Toxoplasma*, *Cysticercus bovis & cellulosae*, *Toxocara canis* and *Trichinella spiralis*. This was done by using specific reference hyper-immune sera experimentally prepared in rabbits versus each parasite.

## **MATERIALS AND METHODS**

### **Examination of slaughtered buffaloes:**

Through monthly visits to El-basateen abattoir during the year 2003, a total of 350 random samples from esophagus, heart, masseter muscle and blood of slaughtered buffaloes at two age groups as under two years and that over 7 years old were collected. Also ten serum samples were collected from slaughtered buffalo's calves free from muscle infection and used as negative control.

The collected muscle samples were subjected to macroscopic and microscopic examination after compression of small pieces using trichoscope glass plates for detection of muscle cysts according to Mohamed (1988).

### **Human Samples:**

Eighty eight human serum and fecal samples (of both sexes with age ranged from 22-53 years old) were selected from patients complained of symptoms and signs related to sarcocystosis infection from the outpatient clinics and patients referred to *Parasitology* labs of the Research Institute of Ophthalmology (Giza), Kasr El-Aini Hospital and from some private clinics in Cairo. These patients were complained with myositis & eosinophilia (32 cases) as well as digestive disturbances (24 cases). Moreover, 12 patients harboring *Schistosoma mansoni* eggs in their faeces, 10 serum samples of Viral hepatitis "C" PCR positive patients and 10 serum samples from healthy individual were used as control for accuracy of the test.

### **Antigens preparation:**

#### **1-Sarcocystis and Toxoplasma antigen:**

According to the technique described by Gasbarre et al. (1984) and Hong Moon, (1987)., The bradyzoites of *Sarcocystis* were extracted from macroscopic cysts of natural infected bovine esophagus (identified as *Sarcocystis bovi-hominis*) by crushing in 0.01M phosphate buffered saline (PBS) pH 7.4. Also *Toxoplasma*

(RH) strains tachyzoites were collect from the peritoneal fluid of experimentally infected mice. After washing by centrifugation, the bradyzoites and tachyzoites were ruptured separately in few amount of PBS by repeated freezing thawing (3times).

The contents were sonicated after this using "Cole parmer ultrasonic Homogenizer " under 150 watt interrupted pulse output at 50% power cycle in ice bath. The suspension was centrifuged at 10 000 rpm at 4C for one hour. This supernatant was collected and dialyzed overnight in refrigerator against PBS, pH 7.2 using a dialysis membrane (6000 to 8000 molecular weight cut off). The dialyzed antigen was stored at -70 ° C until use.

### **2-Cysticercus antigen:**

Live Cysts of *C.bovis* and *C.cellulosae* were collected from infected heart muscles of cattle and pigs respectively from Cairo abattoir. Crude whole cyst antigen was prepared by homogenization, sonication of the cysts in PBS, pH 7.4 and centrifugation at 10.000.rpm for one hour at 4 ° C as described by Cheng and Ko (1991).

### **3- Toxocara canis antigen:**

According to Kagan et al. (1958), the anterior parts of freshly extracted *Toxocara canis* worms collected from scarified natural infected dogs, were cut out, washed repeatedly in 0.01M PBS pH 7.4 then homogenized (ULTRA- TURRAX Janke and Kunkel KG) with PBS at 6000 r.p.m. for 20 minutes in ice bath.

The supernatant was separated by centrifugation (10.000 rpm) for one hour at 4°C. and stored as crude antigen at - 70 °C until use.

### **4- Trichinella spiralis larval antigen:**

*Trichinella spiralis* infective stage muscle larvae were collected from *T.spiralis* infected albino mice, 35 days post-infection. The larvae were separated by pepsin digestion. They were washed using PBS, pH 7.4 and used for production of crude antigen after homogenization and sonication according to Soliman and El-Bahy (1998).

The protein content in the previous prepared antigens were measured by the method of Lowry et al.(1951) and stored at -70 °C until used.

**Preparation of hyper-immune sera:**

Rabbit hyper-immune sera were raised against the previous prepared antigens according to Langley and Hillyer (1989) via initial subcutaneously injection in an equal volume of Freund's complete adjuvant and three consecutive intramuscularly injections in an equal volumes of Freund's incomplete adjuvant during 60 days.

**Stool examination:**

The collected stool samples were examined using Fluke finder technique according to Welch et al (1987) for diagnosis of large size parasitic eggs. The other parasitic eggs, oocysts and cysts were diagnosed using concentration flotation technique according to Solusby (1982).

**Dot-ELISA technique:**

The technique described by Attallah *et al.* (1999) after modification was adopted. According to preliminary block titration, the optimum dilutions of the antigens reference sera and all reagents were determined. According to this, five microliters of each tested antigen containing 1-2  $\mu\text{g}$  protein was dropped on 6 mm diameter nitrocellulose membrane discs (sigma) in flat bottom ELISA plate. The discs were dried, then blocked using 3% bovine serum albumin for 30 min. The discs were washed in phosphate buffered saline (pH 7.4) containing 0.05% Tween 20 (PBS-T20) and dried again. Subsequently, 20  $\mu\text{l}$  of sera (tested and control) diluted at 1:50 and 1:100 in PBS-T20 was applied on top of the dotted samples in doublets. The plates were incubated at room temperature for one hour. After washing, 50  $\mu\text{l}$  of horseradish peroxidase conjugated protein A (sigma) at 1:1000 dilution was added to each well and incubated for 30 min. at room temperature. After washing, the discs were immersed each in 50 $\mu\text{l}$  of freshly prepared substrate solution (4-chloro-1-naphthol 340  $\mu\text{g}/\text{ml}$  substrate buffer with 0.03% hydrogen peroxide solution). Color developed within 15 min. and the reaction was stopped with distilled water. The discs were dried and kept in the dark. The purple color that was produced in positive test results varied in its intensity from weak to strong. A colorless or diffuse spot was produced in the case of a negative test result as in fig.(1).

## RESULTS AND DISCUSSION

*Sarcocystis* is an Apicomplexa parasite need two hosts, the final is carnivore shed the infective oocysts in its feces and a herbivore one act as intermediate, contain chronic cysts in its muscle (Arness, 1999).

Macroscopic and microscopic examination of compressed oesophageal, heart & masseter muscles of two buffaloes group slaughtered in Cairo abattoir was done, table (1). This examination revealed incidence of *Sarcocystis* oesophagus (less than two years old), this incidence increased to 69.5% in buffaloes over 7 years old.

Although, the incidence of *Sarcocystis* infection especially in old aged buffaloes is high, but it is lower than that recorded by Nassar, (1982) (97.7%) and Mohamed (1988) (92.08%). This may be due to improving in veterinary control program against stray dogs and the variation in number of examined animals.

Concerning examination of heart and masseter muscles of the same animals, it evidenced that, only 2.66% (under 2 years old) and 6.0% (over 7 years old) of them were harbored *Cysticercus bovis*.

It is necessary to mention that, *C.bovis* recorded in aged groups were calcified. Also, a percentage of 4.0% and 6.0 % of these animals judged as suffered from partial liver fibrosis, as in table (1). On the other hand, there was no cases of mixed infection were recorded in these selected animal groups.

Serological examination of *Sarcocystis* infection in the same animal groups by detection of specific anti-parasite anti-bodies in their sera was done using dot ELISA technique (table 2). The results cleared higher rate of infection than that recorded by the first method, it figured up to 21.3 % & 91.0% at 1:50 serum dilution then decreased to 19.33 % and 88.5% with increasing the serum dilution to 1:100 in both age groups respectively. This was accompanied by absence of cross reaction with *T.spiralis* antigen (at 1:100 serum dilution) and it was decreased from 6.0% & 11.0% to 5.33% & 8.5% versus *Toxoplasma* antigen with increasing serum dilution in both age groups respectively. Sera of 5 & 21

animals appear to have specific anti-*C.bovis* antibodies (3.33% & 10.5%) in their sera for both tested age groups, respectively. These anti-*C.bovis* antibodies did not diminish with increasing the serum dilution.

Presence of other anti-bodies in sera of these examined animals than that of *Sarcocystis* may be reflect a true undiagnosed infection by other parasites specially where they still present even with increase the tested serum dilution from 1:50 to 1:100. This was agreed with Lund and Fayer, (1977).

It was interested to mention that there is no positive ELISA reading could be detected in sera of 10 healthy calves tested as control.

Concerning the specificity of the used ELISA technique in exclusion of the cross reaction between the related tissue parasites, findings in table (3) revealed that the used dot-ELISA technique succeed in sensitive detection (100%) of anti-*Sarcocystis* anti-bodies in sera of tissue positive animals at 1:100 serum dilution. This level of serum dilution was able to completely diminish the cross-reaction with *Toxoplasma* antigens in the tested sera, but 3 animals (1.9 %) showing positive results versus *C.bovis* antigen.

The same phenomenon was true for *C.bovis* specific anti-bodies versus its homologous antigen at 1:50 serum dilution (100% specificity). The percentage of these positive animals were decreased to 75% by increasing the serum dilution to 1:100. this decrease may be due to weak anti-*C.bovis* anti-bodies in animals harboring calcified cysts. Also 3 animals (18.75%) from that infected by *C.bovis* gave positive results (at 1:50 serum dilution) versus *Sarcocystis* antigen. This may be reflect true undiagnosed *Sarcocystis* infection specially where it did not diminished with increasing the serum dilution to 1:100.

These findings are agreed with Hong Moon (1987) where he exclude cross reaction between *Sarcocystis* and *Toxoplasm* infections in pigs by increasing serum dilution.

Concerning the value of the used dot-ELISA technique in detection of the specific anti-*Sarcocystis* anti-bodies in sera of selected group of patients complained by specific symptoms at



1:50 & 1:100 serum dilution (tables 4&5). The seroprevalence of *Sarcystis* infection was 37.5% in-patients complained with myositis and eosinophilia and 8.33% in-patients complained with digestive disturbances.

From the obtained results, it is clearly evidence that dot ELISA technique succeeded in excluding cross-reaction in the suspected examined patient versus *Toxocara canis* and *Trichinell spiralis* antigens at the tested low and high serum dilution. This was agreed with that previously described by Piekarski et al. (1978).

On the other side, no cross-reaction was recorded between *Sarcocystis* antigens and the anti-bodies present in sera of *Schistosoma* and hepatitis infected patients. These data proved that previously mentioned by Ibarra et al. (1998) about the sensitivity & specificity of Dot-ELISA in diagnosis of parasitic infection.

These results were disagreed with Khalil et al., (1990) concerning presence of cross reaction (using indirect ELISA technique) between *Fasciola gigantica* antigen in patient sera of other disease such as schistosmiasis, amoebic liver abscess, trichinosis, hydatidosis, cysticercosis, viral hepatits, leuckaemia and rheumatiod arthritis and sera of normal controls also. These data clear the efficacy of the used dot-ELISA technique in diagnosis of the specific *Sarcocystis* antibodies in sera of suspected patients under the tested condition.

The results in table (5) cleared that one patient (3.12%) harboring positive anti-bodies versus *Toxoplasma* antigen and another one showing positive ELISA reading versus *C.cellulose* antigen. According to (Lund & Fayer, 1977) this may be reflect a condition of true inapparant infection by these parasite where this anti-bodies titer still showing positive ELISA reading even after increasing the serum dilution as described previously by Hong Moon (1987).

It is necessary to mention that there is no other parasitic stages could be detected in stool of the examined patients, also there is no positive ELISA reading recorded versus sera of healthy control people. (table,5)

Concerning the used technique and according to De Morilla et. al, (1989), ELISA is a sensitive serological test able to analyze many samples simultaneously. However, it needs some sophisticated equipment. The new modified dot-ELISA was more economic, more suitable for accurate diagnosis using very few amounts of reagent specially in case of small amount of valuable purified antigen, and very convenient for field study where the results can be read visually.

For conclusion the detected level of anti-*Sarcocystis* antibodies in sera of the examined patients specially that complained with myositis associated with eosinophilia, reflect the role of this parasite as an emerging zoonotic problem specially with presence of high incidence of infection in the surrounding buffaloes. The high incidence of infection in abattoir may lead to widespread of the disease in carnivores around this patient. This can support the idea of Arness et al.(1999) about the suitability of human to act as intermediate host for more species of *Sarcocystis* than that already known.

### REFERENCES

- Arness, M.K.; Brown, J.D.; Dubey, J.P.; Neafie, R.C. and Granstrom, D.E. (1999): An outbreak of acute eosinophilic myositis attributed to human *Sarcocystis* parasitism. Am.J.Trop.Med.Hyg., 61 (4):548-553.
- Attallah, A.M.; Ismail, H. ; El Masry, S.A.; Rizk, H.; Handousa, A.; El Bendary, M.; Tabll, A. and Ezzat, F. (1999): Rapid detection of *Schistosoma mansoni* circulating antigen excreted in urine. J. Clin. Microbiol., 37 : 354-357.
- Cheng, R. W. and Ko, R. C. (1991). Cross-reaction between crude antigens of larval *Taenia solium* (*Cysticercus cellulosae*) and other helminthes of pigs. Vet. Parasitol.,39 : 161-170.
- Cook, G. C. and Zumla, A. I. (2003). Manson's Tropical Diseases.(2<sup>nd</sup> edition) Elsevier Science Ltd, Robert Stevenson House,1-Leith Walk, Edinburgh EH1 3AF.
- De Morilla, A. C.; Gomez, A. Bautista, C. R. and Morilla, A.(1989). Evaluacion de un antígeno somatico y uno metabolico de *Fasciola hepatica* en diferentes pruebas inmunologicas par el diagnostico de la fascioliasis en bovinose. Tec. Pecu. Mex.,44: 41-45.
- Dubey, J.P. and Fayer, R.( 1983): Sarcocystosis. Br.Vet.J., 139:371- 377.
- Fawaz, A.A.( 1998): Incidence of *Toxoplasma* and Sarcosporidia in Slaughtered animals in Qena Governorate. Ph.D Thesis. Faculty of Vet. Med. Assiut University
- Gasbarre, L.C.; Suter, P. and Fayer, R. (1984): Humeral and cellular immune responses in cattle and sheep inoculated with *Sarcocystis*. Am.J.Vet.Res., 45: 1592-1596.

- Gottstein, B.(1995):** Cyst-forming coccidia: *Toxoplasma*, *Neospora*, *Sarcocystis*. Schweiz Med. Wochenscr, May, 6; 125.
- Hong-Moon, M. H. (1987).** Serological cross-reaction between *Sarcocystis* and *Toxoplasma* in pigs. The Korean J.of Parasitology. , 25 : 188-194.
- Ibarra, F. ; Montenegro, N. ; Vera, Y. ; Boulard, C. ; Quroz, H., Flores, J. and Ochoa, P. (1998).** Comparison of three ELISA tests for seroepidemiology of bovine fascioliosis. Vet. Parasitol., 77 : 229-236.
- Kagan, I.G.;Jeska, E.L. and Gentzkow, C.J.(1958):** Serum agar double diffusion studies with *Ascaris* antigens: Assay of whole worm and tissue antigen complexes. Journal of Immunology, 80 : 400-406.
- Khalil, H.M., Abdel-Aal T.M., Makhalel, M.K., Abdallah, H.M., Fahmy, I.A. and El-Zayyat, E.A. (1990):** Specificity of crude and purified *Fasciola* antigens in immunodiagnosis of human fascioliasis. J. Egypt. Soc. Parasit,20(1): 87-94.
- Langley, R. J. and Hillyer, G. V. (1989).** Detection of circulating parasite antigen in murine fascioliasis by two-site enzyme-linked immunosorbent assays. Am. J. Trop. Med. Hyg., 41 : 472-478.
- Lowry, O.H.; Rosenbrough, N.J.; Farr, A.L. and Randall, R. (1951):**Protein measurement with the phenol reagent. J.Biol. Chemis., 193 : 265-275.
- Lunde, M.N. and Fayer, R; (1977):** Serological test for antibody to *Sarcocystis* in cattle. J.Parasitol. 63 (2): 222-225
- Mohamed, W.S.(1988):** Muscular parasites of buffaloes. M.V.Sc., Thesis. Faculty of Vet. Med. Cairo University.
- Nassar, A.M.(1982):** Studies on *Sarcocystis* infecting buffaloes in Egypt. Ph.D. Thesis. Faculty of Vet. Med. Cairo University.
- Pappas, M. G.; Hajkowski, R. and Hockmeyer. W. T. (1983).** Dot enzyme-linked immunosorbent assay (dot-ELISA): a microtechnique for the rapid diagnosis of visceral leishmaniasis. J. Immunol. Methods, 46: 205-214.
- Piekarski, G., Heydorn, A.O., Aryeetey, M.E. and Kimmig, P. (1978):** Clinical parasitological and serological investigations in sarcosporidiosis (*Sarcocystis suihominis*)of man. Immun Infekt. 6: (4): 153-159.
- Prestwood, A.K.; Cahoon, R.W. and McDaniel, H.T. (1980):** *Sarcocystis* infections in Georgia swine. Am.J.Vet.Res., 41:1,876-1,881.
- Rommel, M.(1989):** Recent advances in the knowledge of the biology of the cyst-forming coccidia. Angew Parasitol., Aug;30(3):173-183.
- Said, M.S. (1996):** Muscular parasites in slaughtered animals in Assiut Governorates. Ph.D Thesis. Faculty of Vet. Med. Assiut University.
- Soliman,G.A. and El-Bahy, N.M. (1998):**The therapeutic efficacy of Doramectin, Ivermectin and levamisole against different stages of *Trichinella spiralis* infection in rats. Vet.Med.J.,Giza (4) A :527-541.Prestwood et al., 1980
- Soulsby, E. J. (1982).** Helminthes, Arthropods and Protozoa of Domesticated Animals (text book). 6<sup>th</sup> edition, Bailliere Tindall and Cassell, London.
- Welch, S.; Malone, J. and Geaghan, H. (1987).** Herd evaluation of *Fasciola hepatica* infection in Louisiana cattle by an ELISA. Am. J. Vet. Res., 48 : 345-347.
- Zimmerman, G. L.; Kerkvliet, N. I. and Cerro, J. E. (1985).** Blood parameters of lambs experimentally infected with *Fasciola hepatica*. Int. Goat Sheep Res., (5) : 1513-1515.

Table (1): Incidence of tissue parasites detected in examined buffaloes

Examined Organs	No. Exam.	Sarcocystis muscle cysts		Cysticercus bovis		LIVER FIBROSIS	
		No. Infect.	%	No. Infect.	%	No. Infect.	%
under two years	150	16	10.6%	4	2.66%	6	4.0%
Over 7 years	200	139	69.5%	12 *	6.0%	12	6.0%
Total	350**	155	44.28%	16	4.57%	18	5.14%

\* These cysts are calcified. \*\* no mixed infection.

Table (2): Serological diagnosis of tissue parasites in the examined animals using dot ELISA technique.

Tested Serum dilution	No. exam.	TESTED ANTIGENS							
		Sarcocystis		Toxoplasma		C. BOVIS		T. SPIRALIS	
		No. +Ve	%	No. +Ve	%	No. +Ve	%	No. +Ve	%
At 1:50	150	32	21.3%	9	6.0%	5	3.33%	2	1.3%
	200	182	91.0%	22	11.0%	21	10.5%	4	2.0%
At 1:100	150	29	19.33%	8	5.33%	5	3.33%	0.0	0
	200	177	88.5%	17	8.5%	21	10.5%	0.0	0

No positive ELISA readings recorded using 10 control calve serum samples

Table (3): Serological evaluation of cross-reactions in sera of animals containing muscle cysts using dot ELISA technique.

Examined animals	No. Exam	Tested serum dilution	Tested antigen					
			Sarcocystis Ag		C. bovis Ag.		Toxoplasma Ag.	
			No. +Ve	%	No. +Ve	%	No. +Ve	%
Animal had Sarcocystis muscle cysts	155	1 : 50	155	100%	12	7.7%	12	9.6%
		1 : 100	155	100%	3	1.9%	0.0	0.0
Animal had C.bovis in muscles	16	1 : 50	3	18.75%	16	100%	4	25.0%
		1 : 100	3	18.75%	12	75.0%	0.0	0.0

Ag. = antigen

Table (4): Incidence of anti *Sarcocystis* antibodies and their specificity in sera of examined patients at 1:50 serum dilution

Examined Patients complains	No. Exam	Antigens used for testing the present anti-bodies							
		<i>Sarcocystis</i> Ag.		<i>Toxoplasma</i> Ag.		<i>C. cellulosa</i> Ag.		<i>T. canis</i> Ag.	
		No. +Ve	%	No. +Ve	%	No. +Ve	%	No. +Ve	%
Myositis & Eosinophilia *	32	20	62.5	8	25.0	4	12.5	4	12.5
Digestive* disturbances	24	8	33.3 %	2	8.33 %	0.0	0.0	0.0	0.0
Schistosoma infected	12	2	16.6 %	2	16.6 %	2	16.6 %	0.0	0.0
Hepatitis* patients	10	2	20.0 %	2	20.0 %	0.0	0.0	0.0	0.0
Healthy control	10	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0

\*No parasite diagnostic stages recorded at fecal examination of these patients.  
No positive reaction recorded versus *Trichinella spiralis* larvae antigens

Table (5): Incidence of anti *Sarcocystis* antibodies and their specificity in sera of examined patients at 1:100 serum dilution

Examined Patients complains	No. Exam	Antigens used for testing the present anti-bodies							
		<i>Sarcocystis</i> Ag.		<i>Toxoplasma</i> Ag.		<i>C. cellulosa</i> Ag.		<i>T. canis</i> Ag.	
		No. +Ve	%	No. +Ve	%	No. +Ve	%	No. +Ve	%
Myositis and eosinophilia	32	12	37.5%	1.0	3.12	1.0	3.12	0.0	0.0
Digestive disturbances	24	2	8.33%	0.0	0.0	0.0	0.0	0.0	0.0
Schistosoma infected	12	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Hepatitis patients	10	0.0	0.0	1.0	10%	0.0	0.0	0.0	0.0
Healthy control	10	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0

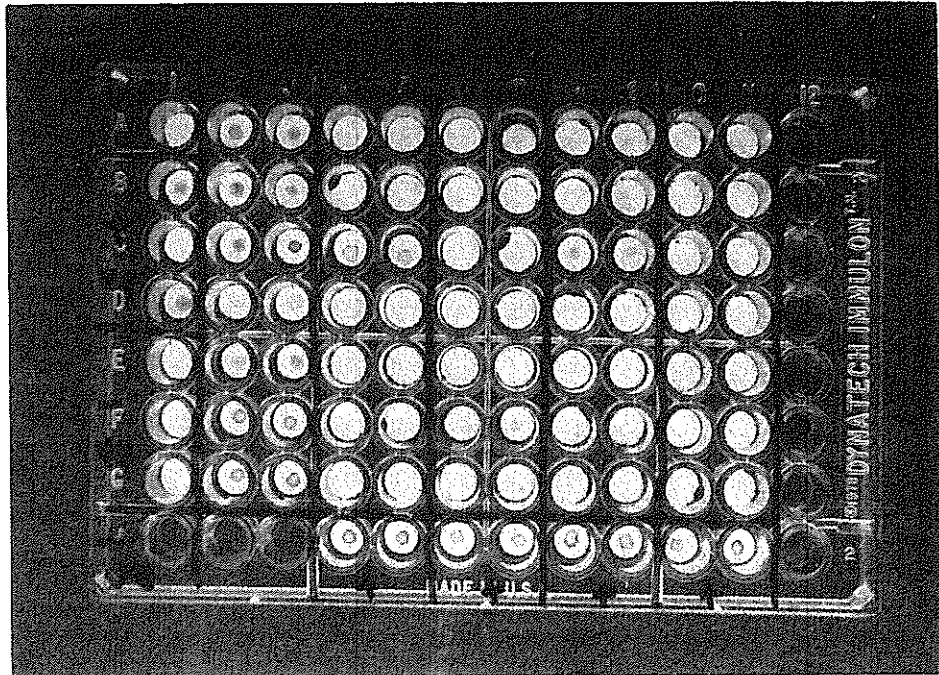


Fig. (1) Dot ELISA plate showing markedly defined blue purple circle on area of Ag application which considered as positive, while the diffused pale blue or colorless considered as negative serum sample.

### الملخص العربي

#### ساركوسبورديوزس كمرض مشترك

مها أحمد صبرى و حاتم عبدالموجود شلبي\*  
قسم الامراض المشتركة - كلية الطب البيطرى جامعة القاهرة  
قسم الطفيليات وأمراض الحيوان - المركز القومى للبحوث

مع ارتفاع نسبه الإصابة بطفيل السركوسيستس فى الجاموس المذبوح مازالت  
قدرة هذا الطفيل على النمو فى عضلات الإنسان محل دراسة. لذلك هدف هذه البحث إلى

تحديد معدل الإصابة بهذا الطفيل في الجاموس المذبوح ومن ثم انعكاس هذه النسبة على تواجد الإصابة بنفس الطفيل في أمصال و براز مجموعة من المرضى تعانى من الام في العضلات . و لهذا تم فحص عضلات و أمصال فئتين عمريتين من الحيوانات المذبوحة في مجزر القاهرة ( أقل من سنتين وأخرى أكثر من 7 سنوات) باستخدام الفحص الميكروسكوبي وكذلك تقييم كفاءة طريقة الميكرو-أليزا في التشخيص الدقيق للأجسام المناعية الخاصة بهذا الطفيل في أمصال نفس الحيوانات. واستيضاح نفس الظاهرة في أمصال مجموعه منتقاة من المرضى.

وقد ثبت وجود نسبة إصابة بلغت 10,6% ، 69,5% في عضلات المجموعتين العمريتين السابقتين على التعاقب. بينما زادت النسبة إلى 19,33% ، 88,5% باستخدام طريقة الميكرو اليزا.

و أما بالنسبة للمرضى فقد تم اختبار أمصال 32 حاله تعانى من آلام في العضلات مصحوبة بارتفاع في أعداد كرات الدم البيضاء (الأيزينوفيل) و مجموعه أخرى ( 24 حاله) تعانى من اضطرابات معديه معوية وكانت نسبه وجود الأجسام المناعية الخاصة بطفيل الساركوسست هي 37,5% ، 8,33% في المجموعتين السابقتين على التوالي. وفى نفس الوقت لم يتم إثبات وجود هذه الأجسام المناعية في أمصال الضوابط العير المصابة من الأدميين أو الحيوانات وكذلك في أمصال أشخاص مصابة بطفيل البلهارسيا أو الالتهاب الكبدي "سى".

وجدير بالذكر ان الأختبار المناعى المستخدم (طريقة الميكرو-أليزا ) كانت ذات كفاءة عاليه في التشخيص الدقيق للأجسام المناعية الخاصة بهذا الطفيل فى كل الامصال المفحوصه حيث لم يكن هناك تداخل مع مولدات الضد لبعض الطفيليات الأخرى التى تنمو فى العضلات مثل حويصلات الساركوسست والتكسوكارا والتكسوبلازما .

من هذا يتضح احتماليه اصابه الإنسان بأنواع جديدة من طفيل الساركوسيست يلعب فيها الإنسان دور العائل الوسيط علاوة على تلك التى تصيب قناته الهضمية.

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