Ultrastructural morphology of the Myxobolus dermatobius *Ishii 1915 (Mixosporea: Myxobolideae) microspores infecting eyes of Nile tilapia (*Oreochromis niloticus*) in Egypt*

Nosseur M. El-Sayed*

Department of Parasitology, Faculty of Veterinary Medicine, Zagazig University, Egypt.

**Corresponding author at: Department of Parasitology, Faculty of Veterinary Medicine, Zagazig University, Egypt. E-mail: nosseur@gmail.com.*

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Myxosporidae, Ultrastructure, *Myxozoa*, Nile tilapia (*Oreochromis niloticus*), Transmission electron microscopy.

Summary
A total of 1,000 cultured Nile tilapia (*Oreochromis niloticus*) were collected from different governmental and private fish farms and examined for detection of myxosporean parasites infection. The infected fishes showed slight unilateral exophthalmia with whitish cyst in the eye. Numerous white cysts like plasmodia of *Myxobolus dermatobius* were recovered from the eye of the examined fishes with low prevalence rate (1%). Small intact cyst was isolated, fixed in 3% glutaraldehyde in 0.1 M sodium cacodylate (pH 7.4) and prepared for transmission electron microscopy examination. Ultrathin sections myxospores of *M. dermatobius* revealed pair of capsulogenic cells at the apical pole of the developing myxospore. Single sporoplasm containing a single nucleus and sporoplasmosomes fills nearly all the space beneath the polar capsules. The later were pyriform in shape, each one had homogenous dense core and 4 turns of polar filaments. Ultrastructural characteristics of the present myxospore were described and discussed in detail.

Introduction

Fishes are considered as one of the most important sources of animal protein all over the world. Because of numerous lakes, seas and a long river, Egypt has a very diversified fauna of fresh and marine water fishes. Vision either in wild or farmed fishes is quite different from mammalians as it has to be adapted to acquatic environment (Lee 2002). The awareness of parasites that affect fish health, growth, and survival is increasing together with a developing interest for the fish farming and production. Knowing fish parasites becomes important for a rapid and correct diagnosis. Early diagnosis is a prerequisite for implementing preventive measures, which are the best way to reduce infection spread (Abdel Ghaffar *et al.* 2012, Morsy *et al.* 2012).

Myxosporidea have a great importance in ichtyopathology. They are frequently described in freshwater, brackish and marine fishes. Myxosporean parasites are the most important fish pathogens and more than 2,300 species have been reported from marine and freshwater fishes in several global areas (Manrique *et al.* 2015, Manrique *et al.* 2016). Myxosporea infecting fishes are a group of parasites responsible for myxosporidiosis, a serious disease of fishes (Adriano *et al.* 2009, Morsy 2010). *Myxobolus* Butschli, 1882, is one of the largest genus of myxosporean groups with approximately 856 species. Species of *Myxobolus* infect fishes from all over the world (Eiras *et al.* 2014). They can infect a diverse set of specific tissues including specifically the tegument, eyes, gills, glands, gonads, kidneys, muscle, digestive tract, and nervous system (Lorna and Dykova 1992). The *Myxobolus dermatobius* (Ishii, 1915) (*Syn. M. dermatobia*) in Nile tilapia (*Oreochromis niloticus*) causes petechial to focal haemorrhages in orbit, exophthalmia and unilateral eye opacity especially in advanced cases (Abdel-Aal 2002) while *M. dermatobia,* isolated from *Tilapia zillii* at Giza province, causes unilateral eye opacity (Mohamed *et al.* 2004).

The ultrastructural morphology of myxosporean species has been widely studied (Lorn and Dykova 1992). However, in Egypt, few species only have been ultrastructurally described. In this study the ultrastructural morphology of *M. dermatobius* detected in Nile tilapia is described.

Materials and methods

A thousand of live Nile tilapia were collected from different governmental and private fish farms in Sharkia Governorate, Egypt. The collected fishes were transported to the laboratory and dissected. The different organs were examined macroscopically and microscopically for detection of any visible myxosporean cysts.

Several small intact cysts with minimum surrounding tissue isolated from ten positive fish samples were fixed in 3% glutaraldehyde in 0.1 M sodium cacodylate (pH 7.4) for at least 24 hours, washed in the same buffer and post-fixed with 2% OsO_4 in the same buffer. The specimens were dehydrated in series of graded ethanol, transferred to propylene oxide and, finally, embedded in araldite. Ultrathin sections were stained with uranyle acetate and lead citrate and observed in a Philips (208) TE Moperated at 80-100 kV (Vital *et al.* 2003).

Results

The prevalence of infection with *M. dermatobius* was 1% (10 out of 1,000). It was found in the form of whitish cyst in the eye of Nile tilapia. Slight exophthalmia was observed in the infected eye. The myxospores were recovered from their original plasmodia found in the eye of Nile tilapia (Figure 1).

Each myxospore contains a pair of capsulogenic cells, two peripherally arranged valvogenic cells and one sporoplasm cell. Capsulogenic cells are found at the apical pole of the developing spore and, together with the sporoplasm, forms a central core that is ensheathed by valvogenic cells. These cells give rise to the two shell valves surrounding each spore and the sutural ridge joining the valves. The differentiation of the capsulogenic cells starts with appearance of a club-shaped structure 'capsular primordium' (Figures 2 and 3).

Sporoplasm fills nearly all the space beneath the polar capsules. It contains a nucleus, small vesicles

Figure 1. *Specimen of Nile tilapia (*Oreochromis niloticus*), showing white plasmodia and exophthalmia (arrows) in the eye due to* Myxobolus dermatobius *infection.*

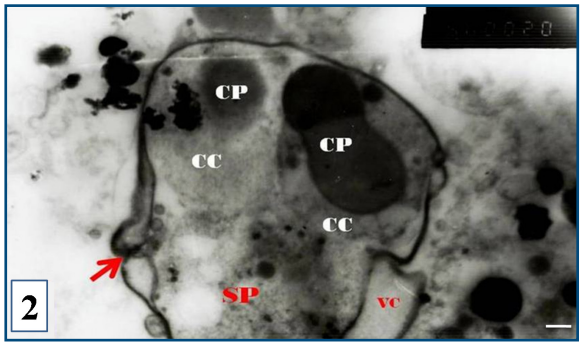

Figure 2. *Transmission electron microscopy of* M. dermatobius *premature myxospore parasite from the eye of* Oreochromis niloticus *showing two capsulogenic cells (CC) containing capsular primordium (CP), sporoplasm (SP), valvogenic cell (VC), and suture valve (arrow).* Scale bar = 300 nm.

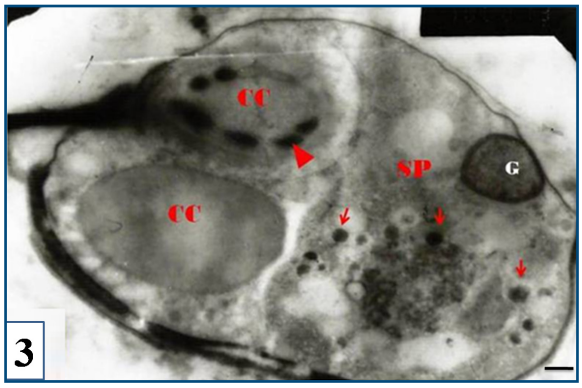

Figure 3. *Transmission electron microscopy of* M. dermatobius *nearly mature myxospore parasite from the eye of* Oreochromis niloticus *showing two capsulogenic cells (CC), primordia of polar filaments (arrow head), sporoplasm (SP) with sporoplasmosomes (arrow) and glycogen body (G).* Scale bar = 300 nm.

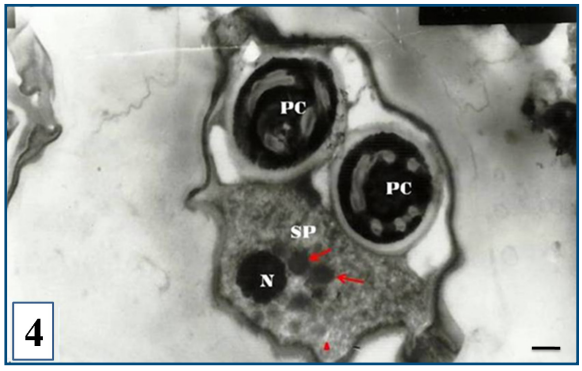

Figure 4. *Transmission electron microscopy of the transverse section of* M. dermatobius *mature, parasite from the eye of* Oreochromis niloticus, *showing two polar capsules (PC), sporoplasm (SP) containing sporoplasmosomes (arrows), one nucleus (N), polar filaments and small vesicles (arrow head).* Scale bar = 300 nm.

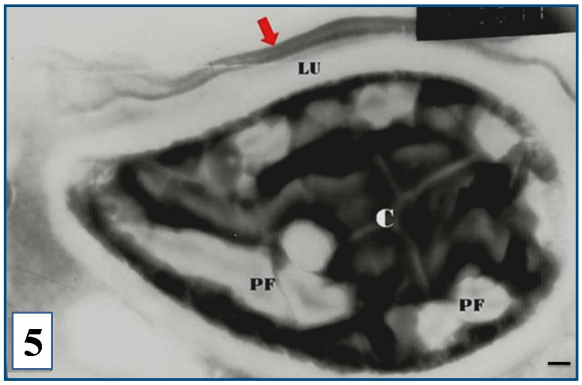

Figure 5. *Longitudinal section through* Myxobolus dermatobius *well developed polar capsule showing an electron-dense outer layer (arrow); a central translucent layer (LU) and inner dense core (C) with polar filament coils (PF).* Scale bar =500 nm.

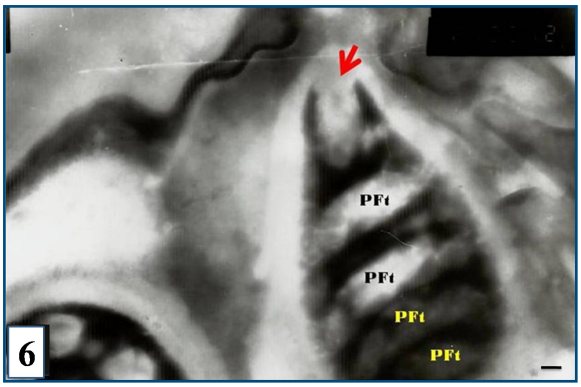

Figure 6. *Transmission electron microscopy of longitudinal section through* M. dermatobius *parasite from the eye of* Oreochromis niloticus, showing well developed polar capsule, four turns of polar filament (PFt) and apical cap (arrow). Scale bar = 500 nm.

and, sometimes, exhibits dense matrices known as sporoplasmosomes. A small area of sporoplasm is occupied by a glycogen body (Figure 4).

Two polar capsules are pyriform in shape, equal size, located side by side at the same level and occupy approximately half of the total spore length. Each polar capsule has a homogenous core of medium electron-density containing polar filaments, surrounded by an electron-lucent layer and an outer layer of medium density (Figure 5). Four turns of polar filament coils are probably in each capsule. The apical portion of each mature polar capsule is plugged by a cap-like cover (Figure 6).

Discussion

Myxobolus Bütschli, 1882, contains over 450 of the 1,700 species described within phylum *Myxospora* (*Myxozoa*). These parasites primarily infect fishes, but a small number of species have been found parasitizing amphibians and reptiles (Lom and Dyková 1992). The infected fishes showed slight unilateral exophthalmia with whitish cyst in the eye. Similar lesion was observed in the eye of tilapia nilotica by Abd El-Aal (Abd El-Aal 2002) and Mohamed and colleagues (Mohamed *et al.* 2004). Numerous species of *Myxobolus* have been reported among different African tilapia species; *Myxobolus agolus, M. brachysporus, M. clarii, M. cichlidarum, M. heterosporus, M. tilapiae* and *M. camerounensis* and *M. kainjiae* appeared to be common in gills, fins, eyes and teguments of *Oreochromis niloticus* (Ousman *et al.* 2007).

In Egypt, several species of *Myxobolus* were isolated from *O. niloticus*. Faisal and Shalaby (Faisal and Shalaby 1987) identified *M. nilei* (syn. *Myxosoma tilapiae*) from eyes, skin, gills, kidney, spleen and pancreas while, Abed (Abed 1987) described *M. heterosporus* from eye, muscle and kidney. Abu El-wafa (Abu El-wafa 1988) identified *M. spheroidalis* and *M. ocularis* from eye. Mazen (Mazen 1994) described *M. heterosporus* from eye and gills. Abdel-Ghaffar and colleagues (Abdel-Ghaffar *et al.* 1995) described *Myxobolus* sp. from the inner wall of cornea, the base of the gill arch, and roof of the mouth. Hegazy (Hegazy 1999) isolated *M. cornealis* from the eye while, Abd El-Aal (Abd El-Aal 2002) and El-Mansy (El-Mansy 2005) described *M. dermatobia* and *M. heterosporus* from eye and cornea, respectively. *M. dermatobia* was isolated from eye of *Tilapia zilli* at Giza province (Mohamed *et al.* 2004).

Most of the Egyptian *Myxobolus* species morphological descriptions have been mainly based on light microscopy and diagrammatic drawings and just few ultrastructural descriptions are available. In accordance with Abd El-Aal (Abd El-Aal 2002) and Abdel-Ghaffar (Abdel-Ghaffar *et al.* 2005), the ultrastructural characteristics of *M. dermatobius* revealed that the spore developed from five cells. The capsulogenic cell of the present species showed capsular primordial to that described in *M. stomum* and *Myxobolus* sp. by Ali and colleagues (Ali *et al.* 2003) and Abdel-Ghaffar and colleagues (Abdel-Ghaffar *et al.* 2005), respectively. The valvogenic cells gave rise to shell valve surrounding each spore and sutural ridge joining the valves were similar to *M. dermatobia* described by Abd El-Aal (Abd El-Aal 2002).

The sporoplasm of the investigated species was composed of single mono-nucleated cell as in *M. dermatobia* described by Abd El-Aal (Abd El-Aal 2002) but different from bi-nucleated sporoplasm observed in other *Myxobolus* species (Ali *et al.* 2003, Casal *et al.* 1996, Abdel-Ghaffar *et al.* 1994, Abdel-Ghaffar *et al.* 2005). The sporoplasmosomes of the present species complied with a similar dense body found in *M. cotti* reported by El-Matbouli and colleagues (El-Matbouli *et al.*

1990); *M. dermatobia* (Abdel-Aal 2002), *M. stomum* (Ali *et al.* 2003) and *Myxobolus* sp. (Abdel-Ghaffar *et al.* 2005). The glycogen body noticed in the sporoplasm is essential in the myxosporean spore as it provides the energy necessary for further developmental stages in the life cycle. It was similar to that reported in *M. dermatobia* (Abd El-Aal 2002), *Myxobolus* sp. (Abdel-Ghaffar *et al.* 1994) and *M. cotti* (El-Matbouli *et al.* 1990).

The fully developed polar capsule was surrounded by an electron-lucent layer and an outer layer of medium density and covered by a cap-like structure at its apical end. A similar finding was reported in many species of *Myxobolus* as *M. dermatobia* (Abd El-Aal 2002), *Myxobolus* sp. (Abdel-Ghaffar *et al.* 2005), *M. stomum* (Ali *et al.* 2003), *M. cotti* (El-Matbouli *et al.* 1990) and *M. sciades* (Azevedo *et al.* 2010). The homogenous core of each polar capsule that contains four turns of polar filaments was identical to that of *M. dermatobia* described by Abd El-Aal (Abd El-Aal 2002). The same number of polar filament coils was reported in *M. hetersporus* by El Mansy (El Mansy 2005) while different numbers of polar filament turns were mentioned in *M. maculatus* (14-15), in *Myxobolus* sp. (5), in *M. stomum* (5-6), in *M. sciades* (9-10), in *M. sclerii* (4-5), in *M. brachysporus* (6-7) and in *M. cuneus* (7-8) (Casal *et al.* 2002, Abdel-Ghaffar *et al.* 2005, Ali *et al.* 2003, Azevedo *et al.* 2010, Kaur and Singh 2010, Abdel-Baki *et al.* 2015, Manrique *et al.* 2016).

References

Abd El-Aal A.M.I. 2002. Studies on tissue parasites in fish. Ph.D. Thesis, Parasitology Dept., Fac. Vet. Med., Tanta Univ. (Kafr El-Sheikh branch), Egypt.

Abdel Ghaffar F., Abdel-Aziz A., El-Shahawi G. & Naas S. 1994. Light and electron microscopic studies on *Myxobolus* sp. infecting *Oreochromis* (Syn: Tilapia) *niloticus* and *O. aureus* in the River Nile. *J Union Arab Biologists*, **2**, 241-261.

Abdel Ghaffar F., Abdel-Baki A.A. & El Garhy E.M. 2005. Ultrastructural characteristics of the sporogenesis of genus *Myxobolus* infecting some Nile fishes in Egypt. *Parasitol Res*, **95**, 167-171.

Abdel Ghaffar F., El-Shahawi G. & Naas S. 1995. Myxosporidia infecting some Nile fishes in Egypt. *Parasitol Res*, **81**, 163-166.

Abdel Ghaffar F., Morsy K., Mehlhorn H., Bashtar A.R., Shazly M.A., Saad A.H. & Abdel-Gaber R. 2012. First report of *Kudoa* species (Myxozoa: Kudoidae) infecting the spotted coral grouper *Plectropomus maculatus* from the Red Sea. A light and ultrastructural study. *Parasitol Res*, **111**, 1579-1585.

Abdel-Baki A.A., Zayed E., Sakran T. & Al-Quraishy S. 2015. A new record of *Myxobolus brachysporus* and *M. israelensis* in the tilapia (*Oreochromis niloticus*) collected from the Nile River. *Egypt Saudi J Biol Sci*, **22** (5), 539-542.

Abed G.P. 1987. Studies on *Myxosporidia* of some Nile fishes in Assuit province. MSc thesis, Assuit University, Egypt.

Abu El- Wafa S.A.D. 1988. Protozoal parasites of some fresh water fish in Behera governorate, Egypt. M.V.Sc.Thesis, Fac. Vet. Med., Alexandria Univ., Egypt.

Adriano E.A., Arana S. & Cordeiro N.S. 2005. Histology, ultrastructure and prevalence of *Henneguya piaractus* (Myxosporea) infecting the gills of *Piaractus mesopotamicus* (Characidae) cultivated in Brazil. *Dis Aquat Organ*, **64**, 229-235.

Adriano E.A., Arana S., Alves A.L., Silva M.R., Ceccarelli P.S., Henrique-Silva F. & Maia A.A. 2009. *Myxobolus cordeiroi* n. sp., a parasite of *Zungaro jahu* (*Siluriformes: Pimelodiade*) from Brazilian Pantanal: morphology, phylogeny and histopathology. *Vet Parasitol*, **162**, 221-229.

Ali M.A., Abdel-Baki A.S., Sakran T., Entzeroth R. & Abdel-Ghaffar F. 2003. Light, electron microscopic studies of *Myxobolus stomum* n. sp. (*Myxosporea: Myxobolidae*) infecting the black spotted grunt *Plectorhynicus gaterinus* (Forsskal, 1775) in the Red Sea, Egypt. *Parasitol Res*, **9**, 390-397.

Azevedo C., Casal G., Mendonica I., Carvalho E., Matos P. & Matos E. 2010. Light and electron microscopy of *Myxobolus sciades* n. sp. (*Myxozoa*), parasite of the gills of the Brazilian fish *Sciades herzbergii* (Block, 1794) (Teleostei, Ariidae). *Mem Instit Oswaldo Cruz*, **105**, 203-207.

Casal C., Matos E. & Azevedo C. 1996. Ultrastructure data on the life cycle stages of *Myxobolus braziliensis* n. sp., parasite of an Amazonian fish. *Eur J Parasitol*, **32**, 123-127.

Casal G., Matos E. & Azevedo C. 2002. Ultrastructure data on the spore of *Myxobolus maculatus* n. sp. (Phylum: Myxozoa), parasite from the Amazonian fish; *Metynnis maculatus*; (Teleostei). *Dis Aquat Org*, **51**, 107-111.

Eiras J.C., Zhang J. & Molnar K. 2014. Synopsis of the species of *Myxobolus* Buetschli, 1882 (Myxozoa: Myxosporea: Myxobolidae) described between 2005-2013. *Syst Parasitol*, **88**, 11-3.

El-Mansy A. 2005. Revision of *Myxobolus heterosporus* Baker, 1963 (syn. *Myxosoma heterspora*) (Myxozoa: Myxosporea) in African Records. *Dis Aquat Organ*, **63**, 205-214.

El-Matbouli M., Fischer-Scherl T. & Hoffmann R.W. 1990. Light and electron microscopic studies on *Myxobolus cotti* El-Matbouli and Hoffmann, 1987 infecting the central nervous system of the bullhead (*Cottus gobio*). *Parasitol Res*, **76**, 219-227.

Faisal M. & Shalaby S.I. 1987. *Myxosoma tilapiae* as a new species (Myxozoa: Myxosporea) in wild *Oreochromis niloticus* in lower Egypt. *Egyp J Vet Sci*, **24**, 73-86.

Hegazy A.1999. Light and electron microscopic studies on Myxozoan infecting some Egyptian fishes in Bahr Shebin Canal. MSc thesis, Menoufia University, Egypt.

Kaur H. & Singh R. 2010. A new myxosporean species *Myxobolus sclerii* sp. nov. and one known species *M. stomum* Ali *et al.* 2003 from two Indian major carp fishes. *J Parasit Dis*, **34** (1), 33-39.

Lee W.R. 2002. Ophthalmic histopathology. 2nd edition. Springer-Verlag, London, U.K .

Lom J. & Dyková I. 1992. *Myxosporidia* (Phylum Myxozoa). 7. Protozoan parasites of fishes. *Dev Aquat Fish Sci*, **26**, 159-235.

Manrique W.G., Figueiredo M.A.P., Belo M.A.A., Martins M.L. & Moraes F.R. 2015. First report of *Myxobolus* sp. infection in the skeletal muscle of Neotropical freshwater fish *Piaractus mesopotamicus. Parasitol Res*, **114**, 2041-2044.

Manrique W.G., Figueiredo M.A.P., Belo M.A.A., Martins M.L. & Azevedo C. 2016. Ultrastructural description of *Myxobolus cuneus* (Myxosporea) in the skeletal muscle and kidney of tropical farmed fish *Piaractus mesopotamicus* (Characiformes: Characidae). *Parasitol Res*, **115**, 2505-2510.

Mazen A.M. 1987. Effect of the protozoan parasite, *Myxosoma heterospora* on the eye of fish *Tilapia nilotica*. *Assiut Vet Med J*, **31**, 121-129.

Mohamed M.A., Ezz El Dien & Elsayed E.E. 2004. Tissue protozoa (*Myxobolus dermatobia*) from the eye in *Tilapia zillii* in Egypt. First International Conference Veterinary Research Division, NRC, Egypt, 307-319.

Morsy K.S. 2010. Studies on myxosporidian and microsporidian parasites infecting some economic fishes from the Red Sea in Egypt. Ph.D. Thesis, Zoology Department, Faculty of Science, Cairo University, Egypt.

Morsy K., Abdel-Ghaffar F., Bashtar A.R., Mehlhorn H., Al Quraishy S. & Abdel-Gaber R. 2012. Morphology and small subunit ribosomal DNA sequence of *Henneguya suprabranchiae* (Myxozoa), a parasite of the catfish *Clarias gariepinus* (*Clariidae*) from the River Nile, Egypt. *Parasitol Res*, **11** (4), 1423-1435.

Ousman A., Filex B.B., Thomas N. & Abraham F. 2007. Structure and dynamics of myxosporean parasites component communities in two freshwater Cichlids in the Chari River (Republic of Chad). *Pak J Biol Sci*, **10** (5), 692-700.

Vital P., Corral L., Matos E. & Azevedo C. 2003. Ultrastructural aspects of the myxosporean *Henneguya astyanax* n. sp. (*Myxozoa: Myxobolidae*), a parasite of the Amazonian teleost *Astyanax keithi* (Characidae). *Dis Aquat Org*, **53**, 55-60.