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## Description of Four New Myxosporean Species (Myxozoa: Myxosporae) from Genus *Myxobolus*, Fish Parasites of Burkina Faso, West Africa

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**Abstract:** The Myxosporean met in freshwater fishes of Burkina Faso, form all of them some whitish cysts on the fins for *Myxobolus labeoi* sp. n. and *Myxobolus tingrelaensis* sp. n. and on the branchial for *Myxobolus heterotisi* sp. n. and *Myxobolus sourouensis* sp. n. The spores of *M. heterotisi* sp. n. are ootid, they measure  $2.13 \pm 0.46$  out of  $9.51 \pm 0.45$   $\mu\text{m}$  and have their anterior extremity slightly contracted. Their pyriform polar capsule measure  $6.41 \pm 0.41$  out of  $3.53 \pm 0.51$   $\mu\text{m}$ . The polar filament is rolled in 10 turns of spires. *M. labeoi* sp. n. Presents some lengthened spores of  $16.43 \pm 0.50$  out of  $10.73 \pm 0.52$   $\mu\text{m}$ . The dissymetrical polar capsules measure  $8.37 \pm 0.56$  out of  $6.53 \pm 0.68$   $\mu\text{m}$  and  $1.57 \pm 0.17$  out of  $0.35 \pm 0.09$   $\mu\text{m}$ . On *M. sourouensis* sp. n., the spores measure  $11.33 \pm 0.33$  out of  $8.84 \pm 0.72$   $\mu\text{m}$ . The polar capsules are ootid measuring  $5.77 \pm 0.73$  out of  $2.32 \pm 0.46$   $\mu\text{m}$ . *M. tingrelaensis* sp. n. form some ootid spores of  $11.6 \pm 0.62$  out of  $9.37 \pm 0.67$   $\mu\text{m}$ . The pyriform polar capsules are  $4.80 \pm 0.55$  out of  $2.40 \pm 0.38$   $\mu\text{m}$  and have a small inter-capsular appendix.

**Key words:** Fish parasite, *Myxobolus*, *Myxobolus labeoi* sp. n., *Myxobolus tingrelaensis* sp. n., *Myxobolus sourouensis* sp. n. and *Myxobolus heterotisi* sp. n. Burkina Faso, West Africa

### INTRODUCTION

Many African Myxosporean known today are cystical. They present a vegetative form which can have different implantations on the tissues or organs under the form of cysts. These African Myxosporean represent more than 135 species of parasites, subjected to the fishes from freshwater, brackish water and sea water (Obiekezie and Okaeme, 1990; Sakiti *et al.*, 1991, 1999; Sakiti, 1997; Kostoingue *et al.*, 1998, 1999, 2001; Kostoingue and Toduebaye (1994) Kabré *et al.*, 1995, 1997, 2002; Kpatcha *et al.*, 1996a, b, 1997, 1999; Fomena and Bouix, 1997, 2000; Fomena *et al.*, 2004; Diebakate *et al.*, 1999; Cecilé *et al.*, 2002, 2003). However, the biodiversity of Myxosporean in African is still under estimated in comparison to the diversity of the environment and the stretch of the continent. Because of the pathogenic risk that these organisms sometimes present, it is important to register the different forms of this group.

In present study four new species presenting under the form of cysts are described on fishes gathered in Burkina Faso. They are *Myxobolus labeoi* sp. n., *Myxobolus heterotisi* sp. n., *Myxobolus tingrelaensis* sp. n. and *Myxobolus sourouensis* sp. n.

### MATERIALS AND METHODS

The study has been realised between February 1997 and November 2004.

The examined host fishes come from traditional fishing on different stretches of water of Burkina Faso which are Diarbakoko, Lemourdougou and Tounoura's dams and the lake of Tingrela in the Comoé, the hand of Mouhoun river in the localities of Di and Lanfiera in the Sourou. These fishes belong to three different species which are: *Heterotis niloticus* (Cuvier, 1829), *Labeo coubie* (Rüppel, 1832) and *Sarotherodon galilaeus* (Linné, 1758).

The collected and identified fishes are brought into laboratories in a cool box. After an eyesight check to locate the parasites of surface, they are dissected and the different parts host external parasites are taken in order to look for some cysts by the mean of binocular magnifying glass. Once located, the cysts are taken and measured. For the observation of the spores resulting from cysts, some fresh smears are realised and mounted between blades and lamella. Then these smears are fixed in methanol and coloured in M.G.G. or kept on the blue of lactophénol.

The observation, the numbering and the drawing of the spores have been possible owing to a microscope of the kind of Leitz Orthoplan equipped for the photography and tube of drawing. The measures are realised on at list a thirty of spores on fresh materials.

For scanning electron microscopy, fresh smears are fixed with 2% osmic acid buffered to palade pH 7, 4. After rinsing in distilled water, the pieces are dehydrated by alcohol and dried at the critical point. The samples dressed on some blocks serving to carry objects are metallised in palladium gold and observation with a cleaning JEOL 6300F electronic microscope from the electronic microscopy service of Montpellier II University.

## RESULTS AND DISCUSSION

The characteristic of *Myxobolus* species found in freshwater are as follows:

*Myxobolus labeoi* sp. n. (Fig. 1- 4, 15)

Host: *Labeo coubie* (Rüppel, 1832)

Type locality: Comoé (Diarbakoko).

Site of infection: implanted among the rays of the fins

Prevalence: 17.4% (88/504)

### Description

Vegetative form: Whitish cysts, ootid, with small height (150-350)  $\mu\text{m}$  and most often numerous on the fins.

Spores: Lengthened with rounded off anterior extremity and back extremity. The wall is little thick. The polar capsules are strongly dissymmetrical. The big polar capsule's polar filament is rolled over about ten turns. The evagination holes of the filaments are face to face. The suture is clear. Very reduced sporoplasm occupying less than the third of the intrasporal cavity.

### Mensurations

Spores:

length  $16.43 \pm 0.50$  (16-17)  $\mu\text{m}$

width  $10.73 \pm 0.52$  (10-12)  $\mu\text{m}$

ratio length out of the width of the spores 1.53.

Polar capsules:

Big capsules

length  $8.37 \pm 0.56$  (7-9)  $\mu\text{m}$

width  $6.53 \pm 0.68$  (5-7)  $\mu\text{m}$

Small capsules

length  $1.57 \pm 0.17$  (1, 6-2)  $\mu\text{m}$

width  $0.35 \pm 0.09$  (0, 3-0, 5)  $\mu\text{m}$

ratio length out of the width of the big capsules out of the width of the small capsule is 5.28

**Remarks:** Many of the myxosporean having unequal polar capsules have been pointed out by different writers for examples *Myxobolus dispar* (Thélohan, 1895), *Myxobolus diversicapsularis* (Shulman, 1966) described on *Rutilus rutilus*, *Myxobolus type 1* (Abolarin, 1971) observed in the *Labeo coubie*, *Myxobolus dossoui* (Sakiti *et al.*, 1991) in the *Tilapia zillii*, *Hemichromis fasciatus* and hybrid *Tilapia*, *Myxobolus burkinei* (Kabré *et al.*, 1995) found in *Labeo coubie* in Burkina Faso and *Myxobolus bilongi* (Fomena, 1995) in *Tilapia zillii* in Cameroon. *Myxobolus dossoui*, *Myxobolus burkinei* and *Myxobolus bilongi* are distinguished from the species we've described on the one hand by reducing height of their big polar capsule and the relative importance of their small capsule and on the other by the absence of an inter-capsular appendix. *Myxobolus diversicapsularis* and *Myxobolus dispar*, in spite of the dissymetry of their polar capsules are different by the existence of an inter-capsular appendix. The *Myxobolus type 1* (Abolarin, 1971) which presents some similarities is different because of the important height of the big capsules.

Considering these arguments, we think that the species we've just described is new and we propose to call it *Myxobolus labeoi* to remind the name of the host fish on which it is generally found.

*Myxobolus heterotisi* sp. n. (Fig. 5 - 8, 16)

Host: *Heterotis niloticus* (Cuvier, 1829)

Type locality: Tounoura and Lemouroudougou dams in the Comoé.

Site of infection: primary gill lamellae

Prevalence: 16.20% (29/179)

### Description

Vegetative form: presents itself under the form of whitish cysts which height varies from 350 to 500  $\mu\text{m}$ . Very numerous cysts (from 10 to 20 or more) by gill ark.

Spores: Ootid with slightly retracted anterior extremity. Little thick wall. Pyriform polar capsules with equal height. Polar filament rolled in about ten turns of spires. At maturity before evagination the capsule is sealed with a stopper. Hole of evagination of the polar filaments are face to face. Granular sporoplasm, occupying more than the middle of the intrasporal cavity. Absence of inter capsular appendix. Presence of teratological forms.

### Mensurations

Spores:

length  $12.12 \pm 0.46$  (11, 5-13)  $\mu\text{m}$

width  $9.51 \pm 0.45$  (9, 5-10)  $\mu\text{m}$

ratio length out of the width of the spores 1.27.

Polar capsules:

length  $6.41 \pm 0.41$  (6-7)  $\mu\text{m}$

width  $3.53 \pm 0.51$  (3-4, 5)  $\mu\text{m}$   
ratio length out of the width of the polar  
capsules 1.81

**Remarks:** Among the *Myxobolus* that have been described earlier in Africa as well as in the world, many forms shore *Heterotis niloticus* species described here. They are *Myxobolus camerounensis* (Fomena *et al.*, 1993), *Myxobolus tilapiae* (Abolarin, 1974), *Myxobolus clarii* (Mandour *et al.*, 1993), *Myxobolus dahomeyensis* (Sakiti *et al.*, 1991) and *Myxobolus comoei* (Kabré *et al.*, 1995). If we compare our specimen to these species, we notice that *Myxobolus tilapiae* and *Myxobolus camerounensis* with their important height (length of the superior spore: 14  $\mu\text{m}$ ) are strongly distinguished. Moreover both of them present some locations which are different from the Myxosporean we are describing here. *Myxobolus clarii* and *Myxobolus comoei* are different respectively by the smallness of their spores and the strongest height of the polar capsules.

We conclude that the species which we have described is new and propose it to be called *Myxobolus heterotisi* to remind the host on which it has previously been found for the first time.

*Myxobolus tingrelaensi* sp. n. (Fig. 9-11, 17)  
Host: *Sarotherodon galilaeus* (Linné, 1758)  
Type locality: Comoé (Diarbakoko), the lake of Tingrela and the dam of Lemouroudougou.  
Site of infection: among the rays of the fins.  
Prevalence: 23.74% (161/678)

#### Description

Vegetative form: Presented under the shape of small whitish cysts from 150 to 200  $\mu\text{m}$  of diameters, ootid and sometimes lengthened. The number of cysts on the fins can be important.

Spores: Ootid with very slightly narrow and rounded off anterior extremity and back extremity. The wall of the spores is little thick. The polar capsules are pyriform and of equal dimension. Polar filament from 15 to 20  $\mu\text{m}$  is rolled in 4 or even 5 turns of spires. In evagination process it shows in its middle part a hollow light. It appears at a strong tubular magnifying. Presence of small inter-capsular appendix. Sporoplasm occupying less than half of the intrasporal cavity.

#### Mensurations

Spores

length  $11.6 \pm 0.62$  (11-13)  $\mu\text{m}$   
width  $9.37 \pm 0.67$  (8-10)  $\mu\text{m}$   
ratio length out of the width of the spores 1.28

Polar capsules

length  $4.80 \pm 0.55$  (4-6)  $\mu\text{m}$   
width  $2.40 \pm 0.38$  (2-3)  $\mu\text{m}$   
ratio length out of the width of the polar capsules 2

**Remarks:** Some species with an inter-capsular appendix are known in the world. By its morphometrical characteristics, the species we are describing, can't be brought closer to *Myxobolus comoei* (Kabré *et al.*, 1995) and *Myxobolus nokouenensis* (Sakiti *et al.*, 1991), which are met in the same biotope. It's different from the first one, by its equal height capsule. It is different from the second one by the length of its spore  $10.0 \times 5.9$   $\mu\text{m}$  smaller. Other forms in the world can be compared to it. They are mainly *Myxobolus galilae* (Landsberg, 1985) and *Myxobolus clarii* (Mandour *et al.*, 1993). The comparison to *Myxobolus galilaea* reveals some differences essentially on the mensurations of the spore. In fact, with its spores of 12.9  $\mu\text{m}$  long and polar capsules of 7.8  $\mu\text{m}$  long, *Myxobolus galilaea* is relatively bigger. If *Myxobolus clarii* observed on *Clarias lazera* in Egypt, reveals some similarities to our specimen (the height of the spore is 10.61 long out of 8.7  $\mu\text{m}$  wide) and some comparable height polar capsules (4.14  $\mu\text{m}$  long out of 2.45  $\mu\text{m}$  wide). However it is distinguished by the absence of the inter capsular appendix.

It's surely about a new species that we name *Myxobolus tingrelaensis* to remind the spot where it has been found the first time.

*Myxobolus sourouensi* sp. n. (Fig. 12-14, 18)

Host: *Heterotis niloticus* (Cuvier, 1829)

Type locality: Di, Lanfiera (Sourou).

Site of infection: primary gill lamellae

Prevalence: 25.58% (154/602)

#### Description

Vegetative form: presents under the form of whitish cysts, polymorphs, lobular, important height (600 to 700  $\mu\text{m}$ ), on the terminal extremity of the gills filaments. They have been formed by the accumulation of microcysts.

Spores: subspherical with the front extremity slightly projected. Walls of the spores little thick. Ootid polar capsules with equal dimension. Polar filament rolled in 7 turns of spires. Suture line is preeminent. Sporoplasm occupying nearly half of the intrasporal cavity.

#### Mensurations

Spores:

length  $11.33 \pm 0.33$  (11-14)  $\mu\text{m}$   
width  $8.84 \pm 0.72$  (8-10)  $\mu\text{m}$   
ratio length out of the width of the spores 1.28.

Polar capsules

length  $5.77 \pm 0.73$  (5-7)  $\mu\text{m}$   
width  $2.32 \pm 0.46$  (2-35)  $\mu\text{m}$   
ratio length out of the width of the polar  
capsules 2.48

**Remarks:** Several species of *Myxobolus* with equal height polar capsules with shape comparable to the species we've been describing have been pointed out by different writers in Africa as well as in all over the world.

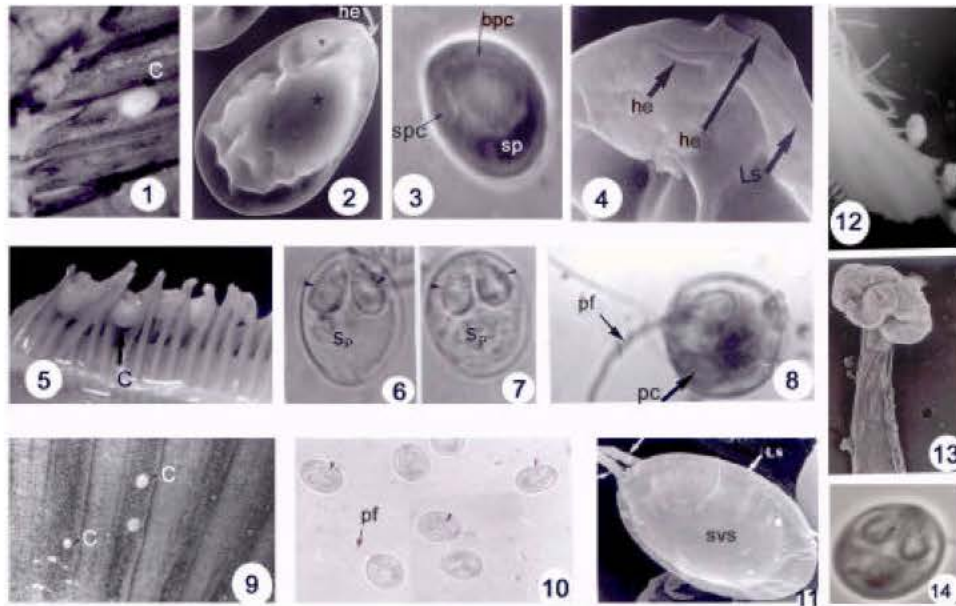


Fig. 1-14: Microphotography of the living spores of the studied species of Myxosporeans

1-4: *Myxobolus labeoi* sp. n.

- 1: Cysts (C) implanted to the base of the pectoral fin ( $\times 10$ ).
- 2: Spore seen in electronical microscopy, showing the hole of evagination (he) and the smooth valvar space. Notice the difference between the big and the small polar capsule (stars) ( $\times 7500$ ).
- 3: Fresh spore seen in photonic microscopy. It shows two piriform and unequal polar capsules. Note the difference between the small ponctiform polar capsule (spc) and the big capsule (bpc) which polar filament is sharp and clearly visible ( $\times 3500$ ).
- 4: Spore seen from the face, showing the line of suture (Ls) passing through the two holes of evagination (he). Note that the hole of evagination of the big polar capsule is bigger and baffled in relation to the one of the small capsule ( $\times 2500$ ).

5-8: *Myxobolus heterotisi* sp. n.

- 5: Cysts (C) implanted between the gill lamellae of the filaments ( $\times 10$ ).
- 6/7: Fresh spore (head of the arrows) piriform, equal in which we can see the polar filament enrolled in turns of spires. The Sporoplasm (Sp) is relatively important ( $\times 10$ ).
- 8: Tetralogical fresh spores seen in photonic microscopy. The spores showing three polar capsules spores (pc). Note the thickness of the filaments (pf) is different ( $\times 3500$ ).

9-11: *Myxobolus tingrelaensis* sp. n.

- 9: Cyste (C) implanted on the gill. Note that they are small in height and disseminates on the fins ( $\times 10$ ).
- 10: Fresh spores in photonic microscopy. The spores with small eight show some equal piriform capsules. Note that some have got an evaginated polar filament (pf) ( $\times 2500$ ).
- 11: Spore seen from the face. It shows the two evaginated polar filaments (white arrows). The line of suture (Ls) and the smooth valvar surfaces (svv) ( $\times 7500$ ).

12-14: *Myxobolus sourouensis* sp. n.

- 12: Cysts implanted at the extremity of a gill filament seen in photonic microscopy ( $\times 5$ ).
- 13: Cyst implanted at the extremity of a gill filament seen in electronical microscopy. Note that it is constituted by the assembling of microcysts ( $\times 40$ ).
- 14: Fresh spore seen in photonic microscopy. It shows two equal piriform polar capsules ( $\times 3000$ ).

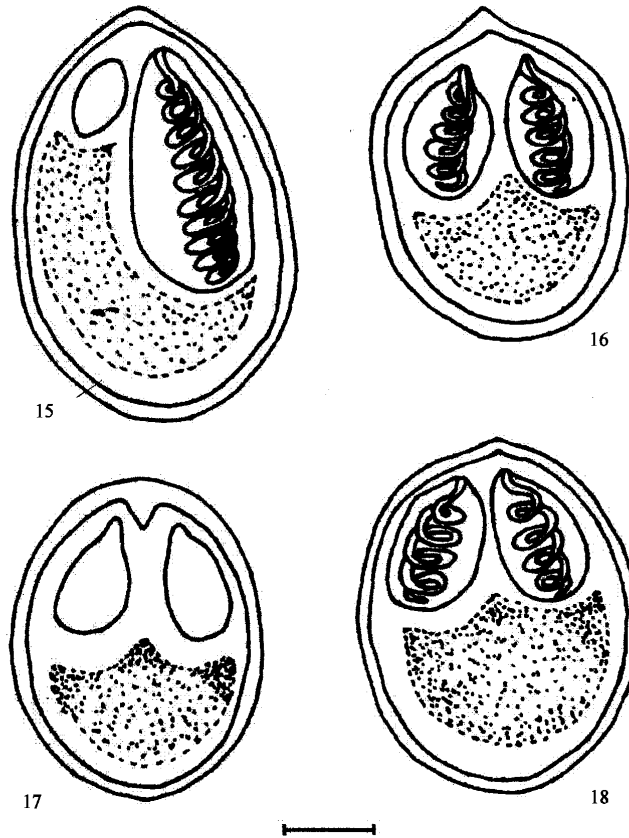


Fig. 15-18: Line drawing of mature spores: 15- *Myxobolus labeoi* sp. n., 16- *Myxobolus heterotisi* sp. n., 17- *Myxobolus tingrelaensis* sp. n., 18- *Myxobolus sourouensis* sp. n. Scale bar- 5  $\mu$ m

For examples *Myxobolus camerounensis* (Fomena *et al.*, 1993), *Myxobolus njine* (Fomena *et al.*, 1985), *Myxobolus nkolyaensis* (Fomena and Bouix, 1994), *Myxobolus dahomeyensis* (Sakiti *et al.*, 1991), *Myxosoma tilapiae* (Faisal and Shalaby, 1987) and *Myxobolus tilapiae* (Abolarin, 1974). Three of them, *Myxobolus dahomeyensis* (height  $9.3 \times 7.1 \mu\text{m}$ ), *Myxobolus njine* (height  $16.17 \times 13.46 \mu\text{m}$ ) and *Myxobolus nkolyaensis* (height  $9.0 \times 8.3 \mu\text{m}$ ), are clearly distinguished by their shape as well as their mensurations. The comparison to *Myxosoma tilapiae* and *Myxobolus tilapiae*, reveals the following statements : *Myxobolus tilapiae* which length is 15 (12-20)  $\mu\text{m}$  and which polar capsules length is very reduced 2, 7 (2-3, 5)  $\mu\text{m}$ , can't be comparable; *Myxosoma tilapiae* met in Egypt and located on the skin, the gills, the eye, the hepatopancreas and the kidney of *Oreochromis niloticus*, is surely subspherical as the specimen we described, but have a slightly bigger height ( $12.03 \times 7.49$ ) with bigger polar capsules.

The specimen we described, with its relatively small polar capsules, can't be confused to *Myxobolus comoei*. These differences bring us to conclude that it is a new species and we propose it to be called *Myxobolus sourouensis* to remind the stream of water in which it has been found in Burkina Faso.

Totally, 4 new species Myxosporean have been described which are: *Myxobolus labeoi* sp. n., *Labeo coubie* (Rüppel, 1832); *Myxobolus tingrelaensis* sp. n., on *Sarotherodon galilaeus* (Linné, 1758); *Myxobolus heterotisi* sp. n. and *Myxobolus sourouensis* sp. n. on *Heterotis niloticus* (Cuvier, 1829) (Fig. 15-18). All these species we've met are histozoic and infest numerous organs and tissues: gills, fins. These Myxosporean form some cysts more or less numerous in the gills and in the fins. The presence of these Myxosporean can explain the care of morbidities and mortalities *S. galilaeus*, brought by fishers during some outing for studies on the ground. In effect, many works among which those of Copland

(1983), De Kinkelin *et al.* (1985), Sakiti *et al.* (1990, 1996), Fomena *et al.* (1993), Bahiri and Marques (1996), Gbankoto *et al.* (1999), revealed that these histozoic Myxosporean are often pathogenous and can cause strong mortalities among their hosts by their pathogenous actions and especially mechanics. They provoke some lyses of the infected tissues, some irritations, traumatismes and some compressions (Lom and Dykora, 1992). Because of the pathogenous effects, a particular attention must be given to these parasites during the exchanges of the fishes Cichlidae for the need of fish farming.

*Labeo coubie* and *Sarotherodon galilaeus* have been the object of several studies related to Myxosporean (Sakiti *et al.*, 1991; Kabré *et al.*, 1995). The observation of new species of Myxosporean on these host-fishes, show their parasitic richness. The presence of Myxosporean on *Heterotis niloticus* seems not to have been mentioned. These parasites constitute the first data of Myxosporean which are pledged for him. In work to come, we'll be interested in the ultrastructural study of Myxosporean. Moreover, some experimental infestations of isolated fishes insulated (and not parasitized) by different species of *Myxobolus*, will allow us to tackle the problem of the parasitic specificity.

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