

***Myxobolus franciscoi* sp. nov. (Myxozoa: Myxosporea: Myxobolidae), a parasite of *Prochilodus argenteus* (Actinopterygii: Prochilodontidae) from the Upper São Francisco River, Brazil, with a revision of *Myxobolus* spp. from South America**

Jorge C. Eiras^{1,4}; Cassandra M. Monteiro² & Marilia C. Brasil-Sato³

¹ Departamento de Zoologia e Antropologia, Faculdade de Ciências, Universidade do Porto. Rua do Campo Alegre, Edifício FC4, 4169-007 Porto, Portugal. E-mail: jceiras@fc.up.pt

² Curso de Pós-Graduação em Ciências Veterinárias, Universidade Federal Rural do Rio de Janeiro. 23890-000 Seropédica, Rio de Janeiro, Brasil.

³ Departamento de Biologia Animal, Universidade Federal Rural do Rio de Janeiro. Rodovia BR 465, km 7, Caixa Postal 74539, 23851-970 Seropédica, Rio de Janeiro, Brasil.

⁴ Corresponding author.

ABSTRACT. *Myxobolus franciscoi* sp. nov. (Myxozoa: Myxosporea: Myxobolidae) is described from the “curimatá-pacú” fish, *Prochilodus argenteus* Spix & Agassiz, 1829 (Actinopterygii: Prochilodontidae) from the Upper São Francisco River, Brazil. This parasite forms whitish plasmodia (about 1 x 1 mm) that develop in the connective tissue of fins. The spores are more or less round in frontal view and ellipsoidal in lateral view, measuring 6.4 (6.0-6.9) µm in length, by 6.0 (5.8-6.4) µm in width and 3.2 µm in thickness. The polar capsules are very small, measuring about 2 µm in length by 1.5 µm in width and ending in a tapered anterior neck. The polar filament makes three turns in a plane at right angles with the longitudinal axis of the spore. Thorough comparisons with the remaining species of *Myxobolus* Bütschli, 1882 described from South American fish, as well as with almost all species of *Myxobolus* described so far, are provided. This paper also includes a revision of *Myxobolus* species from South American fish hosts.

KEY WORDS. “Curimatá-pacu”; freshwater fish; taxonomy.

The Myxosporea are common parasites of marine and freshwater fish. The biology of this important group was recently revised by FEIST (2008). Several species are very important because they can infect economically important fish species and cause high mortality rates in farmed fish (FEIST 2008). *Myxobolus* Bütschli, 1882, the most common genus, is relatively small (30 nominal species described for South America) when contrasted with the overall freshwater fish diversity in this continent, with over 8,000 species representing ca 24% of all fish species (CELLERE *et al.* 2002). GIOIA & CORDEIRO (1996) provided a list of all myxosporeans infecting Brazilian fishes, and EIRAS *et al.* (2005a) included the South American species within a synopsis of the *Myxobolus* species.

Since then, several new Brazilian species have been described (ADRIANO *et al.* 2002, 2006, 2009a,b, TAJDARI *et al.* 2005, EIRAS *et al.* 2005b, 2007, CASAL *et al.* 2006, MARTINS & ONAKA 2006).

Fish are the main protein source of several local human populations in South America. This high demand for local fish has resulted in a need for extensive studies on the subject of native fish farming, as for example those that improve our

knowledge of the parasites that infect local fish populations. In this paper we describe *Myxobolus franciscoi* sp. nov., infecting *Prochilodus argenteus* Spix & Agassiz, 1829 (Actinopterygii, Prochilodontidae). Also known as “curimatá-pacu”, this species is native to the São Francisco River, being economically important as a fishery resource (SATO *et al.* 2003). Additionally, we present a list of the species of *Myxobolus* infecting South American freshwater hosts.

MATERIAL AND METHODS

Forty specimens of *P. argenteus* were net-fished from the Upper São Francisco River in July, 2007, in the municipality of Três Marias, state of Minas Gerais, by fishermen of Estação de Hidrobiologia e Piscicultura da Companhia de Desenvolvimento dos Vales do São Francisco e Parnaíba (EPT/CODEVASF). In the laboratory, specimens were necropsied and thoroughly inspected for the presence of parasites. Measurements were made from fresh spores (30 specimens) under Alphaphot-2, Nikon, according to LOM & ARTHUR (1989), and spores were observed under Nomarski differential interference-contrast. For

checking the presence of an iodophilous vacuole, a drop of Lugol solution was added to the spores. Infected fins were routinely processed for histology and stained with Haematoxylin and Eosin, and Masson's Trichrome. Syntypes of *Myxobolus franciscoi* sp. nov. were sent to deposit in the collection of the Museu de Zoologia of the Universidade Estadual de Campinas (UNICAMP), State of São Paulo, Brazil. Voucher specimens of *P. argenteus* were deposited in the collection of the Museu de Zoologia of the Universidade de São Paulo (MZUSP 95167). Scientific names and their authorship followed the database Fishbase (FROESE & PAULY 2009).

TAXONOMY

Myxobolus franciscoi sp. nov.

Figs 1-4

Description. This parasite forms whitish plasmodia in the connective tissue of fins. The plasmodia were more or less rectangular or slightly elongated, with rounded ends, measuring about 1 x 1 mm (Fig. 1). The plasmodia occurred singly or in small groups, and were clearly visible to the naked eye. Histologically, the plasmodia were located in the connective tissue, immediately under the dermis, and were surrounded by a dense ± 5 μ m thick layer of fibres from the host connective tissue. No membranous wall of parasitic origin was observed. Most of the central part of the plasmodium was occupied by fully mature spores, while in the periphery initial stages of development were observed (Fig. 2). These stages were composed by round, elliptical or slightly irregular cells, with a prominent nucleus, measuring about 8-10 μ m in diameter. The spores (Figs 3 and 4) were almost round in frontal view, and ellipsoidal in lateral view, very small in size, measuring 6.4 (6.0-6.9) μ m in length by 6.0 (5.8-6.4) μ m in width and 3.2 μ m in thickness. The spore length/width ratio varied between 1.03 and 1.06, and the spore length/polar capsule length ratio varied between 3.0 and 3.45. The spore walls were smooth, the valves symmetrical, and an iodophilous vacuole was not present. The polar capsules were very small and equal in size, not reaching the middle part of the spore, more or less rounded, measuring about 2 μ m in length by 1.5 μ m in width and ending in a tapered anterior neck. The polar filament formed three turns in a plane at right angles to the longitudinal axis of the spore.

In some specimens the gills were infected with an unidentified species of *Henneguya* Thélohan, 1892.

Type Host: *Prochilodus argenteus* Spix & Agassiz, 1829.

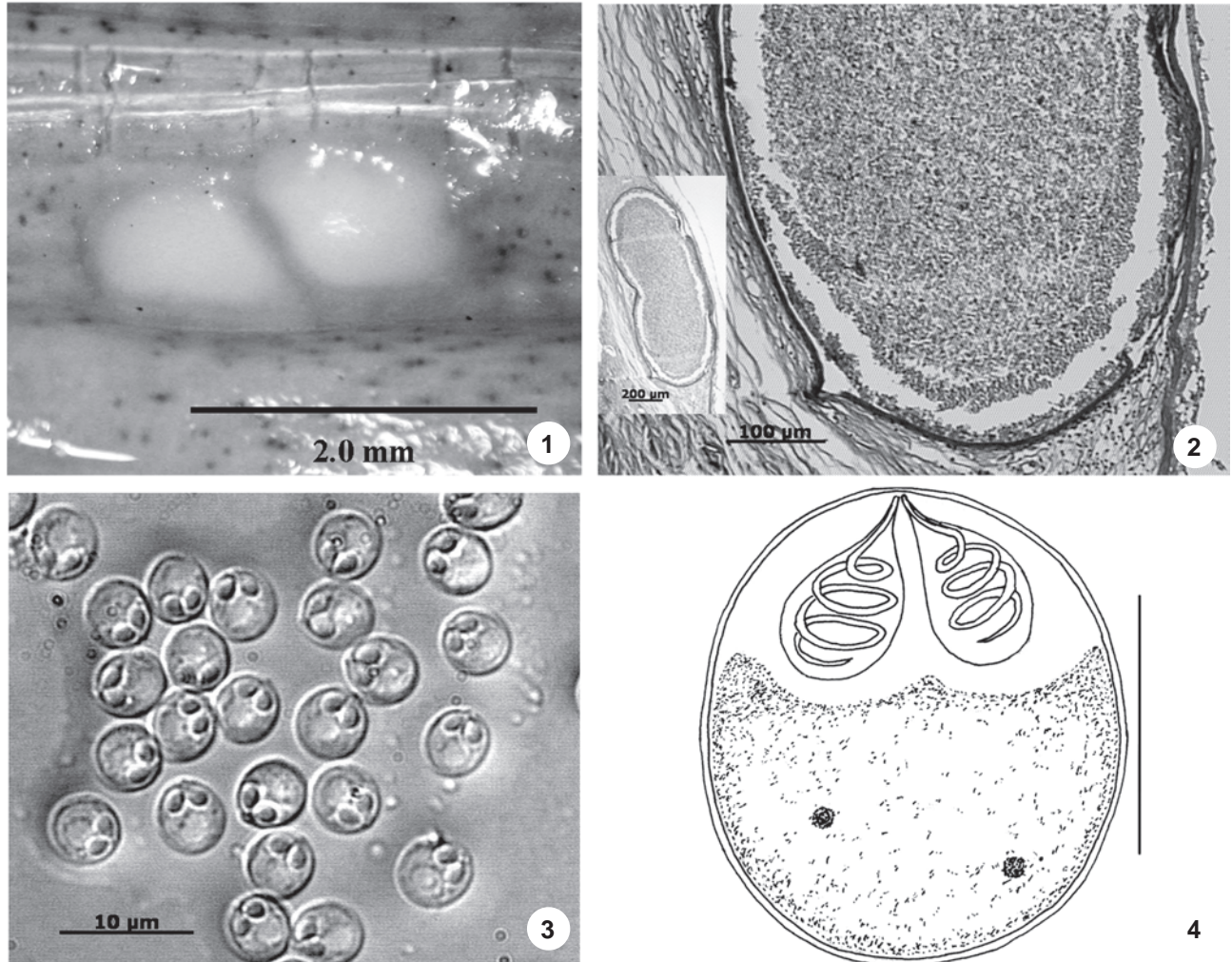
Type Locality: Brazil, State of Minas Gerais, municipality of Três Marias, Upper São Francisco River (18°12'59"S, 45°15'41"W).

Etymology: the specific name derives from the São Francisco River.

Type specimens: The syntypes were deposited in the Museu de Zoologia, Universidade Estadual de Campinas, São Paulo. Prevalence: 4 of 40: 10%.

Remarks. The most striking feature of *M. franciscoi* in our sampling is the unusually small size of the spores. Our species is morphologically and morphometrically distinct from all other species of *Myxobolus* described parasitizing fish from South America (Tabs I and II), with two exceptions: *M. chondrophilus* Nemeček, 1926 and *M. porofilus* Adriano, Arana, Ceccarelli & Cordeiro, 2002. The dimensions of the spores are similar in *M. franciscoi* and *M. chondrophilus*, but specimens of the latter are thinner (4.5 μ m thick) and have bigger polar capsules (3 μ m in length). Also, the host of *M. chondrophilus* is a marine fish (*Sardinella anchovina* (sic) (= *Sardinella aurita* Valenciennes, 1847) and the parasites occur only in the gills (NEMEČEK 1926). These differences are sufficient to justify the separation of these two species. *Myxobolus porofilus*, infecting a host, *Prochilodus lineatus* (Valenciennes, 1837), that is closely related to the host of *M. franciscoi*, has even smaller spores (5.7 by 4.8 μ m) and smaller polar capsules than our species. Besides, unlike *M. franciscoi*, *M. porofilus* infects the visceral cavity of the host (ADRIANO *et al.* 2002). Therefore we cannot consider the two forms to be identical.

We also compared the present material with all the 744 nominal species of *Myxobolus* included in the synopsis by EIRAS *et al.* (2005a). The first conclusion to be drawn from this comparison is that our specimens can be compared with only a few species due to the small size of the spores. In fact, from the 744 species referred to in that contribution, five have similar spore dimensions: *Myxobolus artus* Akhmerov, 1960 described infecting the kidneys of *Carassius auratus gibelio* (= *Carassius gibelio* (Bloch, 1782)) from the Amur Basin (AKHMEROV 1960), *M. calcariferum* Basu & Haldar 2003 described from the gills of *Lates calcarifer* (Bloch, 1790) from India (BASU & HALDAR 2003), *M. dermatobius* (Ishii, 1915) parasitizing the integument of *Anguilla japonica* Temminck & Schlegel, 1846 from Japan (ISHII 1915, LANDSBERG & LOM 1991), *M. minutus* Nemeček, 1911 located in the gills of *Leuciscus leuciscus* (Linnaeus, 1758) and *L. cephalus* (= *Squalius cephalus* (Linnaeus, 1758) from Germany (NEMEČEK 1911), and *M. saranai* Landsberg & Lom, 1991 occurring in the gills of *Barbus sarana* (= *Punctius sarana* (Hamilton, 1822) from India (TRIPATHI 1952, LANDSBERG & LOM 1991). In general, our specimens are very different from all of these species in several morphometrical and morphological features. *Myxobolus artus* has considerably wider spores (9 μ m) and larger polar capsules (4 x 1.8-2 μ m); *M. calcariferum* has slightly larger spores and considerably larger polar capsules (3.8-4.5 x 2.0-2.7 μ m); *M. dermatobius* has clearly narrower smaller spores in width (4.2-4.9 μ m) and larger polar capsules; *M. minutus* presents smaller spore width values (4.2-5 μ m) and larger polar capsules. Finally, *M. saranai* has spores not so wide (4.5-5.0 μ m) and larger polar capsules. Furthermore, these species infect different organs other than the fins, and their hosts are phylogenetically and geographically unrelated to *P. argenteus*. For these reasons our material cannot be definitely be identified with any of these previously described species.



Figures 1-4. *Myxobolus franciscoi* sp. nov. of *Prochilodus argenteus*: (1) plasmodia in the fin; (2) histological section of the plasmodium, parcial view; the centre of the plasmodium is filled with mature spores while at the periphery there are developmental stages. Insert: total view of the plasmodium. Masson's Trichrome; (3) spores; (4) schematic drawing of the spores in frontal view. Bar = 3 µm.

Therefore we believe that the present material represents an undescribed species and propose to name it *Myxobolus franciscoi* sp. nov., a name relating to the São Francisco River.

There are 30 nominal species of *Myxobolus* described for South American hosts. With the exception of two species, *Myxobolus magellanicus* Szidat, 1893 and *Myxobolus paranensis* Bonetto & Pignalberi, 1965, described from Argentinean hosts – *Galaxias maculatus* (Jennyns, 1842) and *Salminus maxillosus* (= *Salminus brasiliensis* (Cuvier, 1816) –, widespread in hosts distributed in other South American countries, all species were described from Brazilian fishes. While most species have freshwater hosts,, two species, – *M. chondrophilus* (NEMECZEK 1911) and *M. platanus* (EIRAS *et al.* 2007) – were described from the saltwater fishes *S. anchovina* (*sic*) and *Mugil platanus* (Günther, 1880), respectively.

A species misidentified as *Myxobolus cerebralis* Hofer, 1903 which is parasite of salmonids, was reported from *Mugil brasiliensis* (= *Mugil liza* Valenciennes, 1836) by MENDES (1980).

Myxobolus inaequalis Gurley, 1893 was described from the head integument of *Pimelodus clarias* (Bloch, 1782) from Guiana and Surinam. Only the length (11 µm) and width (7 µm) of the spores were initially described for this species, which has polar capsules of unequal size (GURLEY 1893). Later, spores of 5.2 µm in length by 3.3 µm in width were reported for this species, along with the hosts, *Piramatana blochi* (= *Corydoras blochi* Nijssen, 1971) and *Synodontis schall* (Bloch & Schneider, 1801) from the same locality (KUDO 1920). According to PINTO (1928) *P. blochi* referred as *Pimelodus clarias* (Bloch, 1782), while *S. schall* occurs only in the River Nile in Africa according to

Table I. Features (measurements in μm) of the valid *Myxobolus* species described from South American fishes. (SL) Body spore length, (SW) body spore width, (ST) body spore thickness, (LPC) length of the polar capsules, (WPC) width of the polar capsules, (NC) number of coils of the polar filament.

<i>Myxobolus</i> species	SL	SW	ST	LPC	WPC	NC
<i>M. absonus</i>	15.7 \pm 1.5	10.2 \pm 0.7		6.4 \pm 0.7	3.6 \pm 0.5	5
				4.2 \pm 0.6	2.5 \pm 0.5	3
<i>M. associatus</i>	15	10		7		
<i>M. braziliensis</i>	10.1 (9.4-10.9)	5.3 (4.7-5.9)	3.6 (3.2-4.0)	5.3 (5.0-5.4)	1.4 (1.4-1.4)	9-11
<i>M. chondrophilus</i>	6	4.5		3		
<i>M. cordeiroi</i>	10.8 \pm 0.5-11.3 \pm 0.3	7.1 \pm 0.2-7.5 \pm 0.3	5.2 \pm 0.3-5.6 \pm 0.1	5.2 \pm 0.3-5.4 \pm 0.2	1.4 \pm 0.1-1.5 \pm 0.3	5-6
<i>M. colossomatis</i> ***	11.8 (11.4-11.2)	6.9 (6.6-7.2)	3.7 (3.5-4.0)	6.0 (5.8-6.6)	2.1 (1.8-2.5)	7-8
<i>M. cunhai</i>	9-11	4-6				
<i>M. cuneus</i> **	10 \pm 0.6	5.7 \pm 0.3	1.7 \pm 0.2	5.7 \pm 0.3	1.7 \pm 0.2	8-9
<i>M. desaequalis</i>	18.3 (17.6-19.1)	11.2 (10.6-11.9)	4.4 (4.0-5.0)	11.2 (10.7-11.9)	4.9 (4.5-5.2)	11-12
				4.6 (4.1-4.8)	2.8 (2.5-3.1)	4-5
<i>M. galaxii</i>	13-15	8.8-10				
<i>M. inaequus</i>	19.8 (15.6-22)	8.6 (7.8-9.3)	8.0 (7.7-8.5)	11.8 (9.4-13)	3.6 (3.1-3.9)	
				4.8 (3.9-5.5)		
<i>M. insignis</i>	14.5 (14-15)	11.3 (11-12)	7.8 (7-8)	7.6 (7-8)	4.2 (3-5)	6
<i>M. kudoii</i>	8.5 - 8.9	6.5 - 7.3		3.5 - 4.2	1.3 - 2.0	
<i>M. lutzii</i>	10	7				
<i>M. maculatus</i>	21.0 (19.7-23.0)	8.9 (7.9-9.5)	7.5 (7.2-7.9)	12.7 (11.8-13.8)	3.2 (3.0-3.6)	14-15
<i>M. macropasmodialis</i>	11 (10.5-12)	8.5 (8-9)	5.2 (5-5.5)	4.5 (4-5)	2.8 (2-3)	6
<i>M. magellanicus</i>	10-13	8.1-8.8		3		
<i>M. metynnis</i>	13.1 (12.9-13.5)	7.8 (7.5-8.3)	3.9 (3.4-4.5)	5.2 (5.0-5.5)	3.2 (3.0-3.6)	8-9
<i>M. noguchii</i>	13.6	8.5		6.8	2.2	
<i>M. paranensis</i>	12-15	7-8		6-7	2.5	
<i>M. peculiaris</i> *	23.2 (23.0-23.2)	14.8 (14.4-15.2)		10.7 (10.5-10.9)	4.4 (4.0-4.8)	4 - 5
<i>M. platanus</i>	10.7 (10-11)	10.8 (10-11)	5.0	7.7 (7-8)	3.8 (3.5-4)	5-6
<i>M. porofilus</i>	5.7 \pm 0.3	4.8 \pm 0.2		1.6 \pm 0.1	1.1 \pm 0.1	3
<i>M. pygocentris</i>	15-16	9-11		9-11	3-4	
<i>M. salminus</i>	9.6-10.5	5.8-6.6	4.7-5.3	4.3-4.8	1.5-1.9	7-8
<i>M. serrasalmi</i> ****	14.4 (12.5-18.0)	8.6 (7.0-10.0)		7.7 (6.0-9.0)	3.1 (2.5-4.0)	
<i>M. stokesi</i>	8.5	5.3		3.1	1.7	
<i>M. testicularis</i>	8.6 (8.2-9.1)	7.2 (6.7-7.5)	2.7 (2.4-3.0)	3.5 (3.3-3.8)	1.7 (1.3-2.0)	5-6
<i>M. franciscoi</i>	6.4 (6.0-6.9)	6.0 (5.8-6.4)	3.2	2.0	1.5	3

* The measurements indicated relate spores stained in smears; values for fresh spores: SL: 25.2 (25.0-25.3); SW: 15.4 (15.0-15.5). ** The parasites were observed in the connective tissue of several organs: gall bladder, urinary bladder, gills, spleen, fins, head surface, liver and heart. *** A small portion of the spores were oval in shape (the majority were elongated ellipsoidal) and measured 10.5 (10.3-10.9) in length and 8.0 (7.2-8.5) in width. The parasites were found in the connective tissue in gills, heart, serous membranes of the intestine, liver parenchyma and skeletal muscle. **** Different spores in shape (pyriform against oval) and size were described: 8.3 (7.0-9.5) in length and 4.0 (3.5-5.0) in width, with polar capsules being 5.8 (5.0-7.5) in length and 1.5 (1.0-2.0) in width.

Table II. References for features listed in table I.

Myxobolus species	Host	Site of infection	Reference
<i>M. absonus</i>	<i>Pimelodus maculatus</i> Lac., 1803	Opercular cavity	CELLERE <i>et al.</i> (2002)
<i>M. associatus</i>	<i>Leporinus mormyrops</i> Stein., 1875	Kidney	NEMECZEK (1926)
<i>M. braziliensis</i>	<i>Bunocephalus coracoideus</i> Stein., 1882	Gills (interlamellar)	CASAL <i>et al.</i> (1996)
<i>M. chondrophilus</i>	<i>Sardinella anchovina</i> (= <i>S. aurita</i> Val., 1847)	Gills	NEMECZEK (1926)
<i>M. cordeiroi</i>	<i>Zungaro jahu</i> (Ihering, 1898)	Skin, gill arch, eyes, urin. bladder	ADRIANO <i>et al.</i> (2009a)
<i>M. colossomatis</i>	<i>Colossoma macropomum</i> (Cuv., 1816)	Connective tissue	MOLNÁR & BÉKÉSI (1993)
<i>M. cunhai</i>	<i>Pygocentrus piraya</i> (Cuv., 1819), <i>Pimelodus clarias</i> (Bloch, 1782)	Intestinal content	PENIDO (1927)
<i>M. cuneus</i>	<i>Piaractus mesopotamicus</i> (Holmberg, 1887)	Connective tissue	ADRIANO <i>et al.</i> (2006)
<i>M. desaequalis</i>	<i>Apteronotus albifrons</i> (L., 1766)	Gill lamellae	AZEVEDO <i>et al.</i> (2002)
<i>M. galaxii</i>	<i>Galaxias maculatus</i> (Jenyns, 1842)	All organs except gills	SZIDAT (1953)
<i>M. inaequus</i>	<i>Eigenmannia virescens</i> (Val., 1836)	Brain	KENT & HOFFMAN (1984)
<i>M. insignis</i>	<i>Semaprochilodus insignis</i> (Jardine, 1841)	Gills (intralamellar)	EIRAS <i>et al.</i> (2005b)
<i>M. kudoii</i>	<i>Nemathognata</i> sp.	Integument	GUIMARÃES & BERGAMIN (1938)
<i>M. lutzi</i>	<i>Girardirmus januarius</i> (= <i>Phalloptychus januarius</i> (Hensel, 1868)	Testis	ARAGÃO (1919)
<i>M. maculatus</i>	<i>Metynnis maculatus</i> (Kner, 1858)	Kidney	CASAL <i>et al.</i> (2002)
<i>M. macroplasmodialis</i>	<i>Salminus maxillosus</i> (= <i>S. brasiliensis</i> (Cuv., 1816)	Abdominal cavity	MOLNÁR <i>et al.</i> (1998)
<i>M. magellanicus</i>	<i>Galaxias maculatus</i> (Jenyns, 1842)	Gills	SZIDAT (1953)
<i>M. metynnis</i>	<i>Metynnis argenteus</i> Ahl, 1923	Connect. subcutaneous tissue	CASAL <i>et al.</i> (2006)
<i>M. noguchii</i>	<i>Serrasalmus spilopleura</i> Kner, 1858	Gills (blood smear)	PINTO (1928)
<i>M. paranensis</i>	<i>Salminus maxillosus</i> (= <i>S. brasiliensis</i> (Cuv., 1816)	Testes, ovary	BONETTO & PIGNALBERI (1965)
<i>M. peculiaris</i>	<i>Cyphocharax nagelli</i> (Stein., 1881)	Gills (smears)	MARTINS & ONAKA (2006)
<i>M. platanus</i>	<i>Mugil platanus</i> Günther, 1880	Spleen	EIRAS <i>et al.</i> (2007)
<i>M. porofilus</i>	<i>Prochilodus lineatus</i> (Val., 1837)	Visceral cavity	ADRIANO <i>et al.</i> (2002)
<i>M. pygocentris</i>	<i>Pygocentrus piraya</i> (Cuv., 1819)	Intestinal content	PENIDO (1927)
<i>M. salminus</i>	<i>Salminus brasiliensis</i>	Gill filaments vessels	ADRIANO <i>et al.</i> (2009b)
<i>M. serrasalmi</i>	<i>Serrasalmus rhombeus</i> (L., 1766)	Spleen, kidney, liver	WALLIKER (1969)
<i>M. stokesi</i>	<i>Pimelodella</i> (?) sp.	Nose integument	PINTO (1928)
<i>M. testicularis</i>	<i>Hemiodopsis microlepis</i> (= <i>Hemiodus microlepis</i> Kner, 1858)	Testis	TAJDARI <i>et al.</i> (2005)
<i>M. franciscoi</i>	<i>Prochilodus argenteus</i> Spix & Agassiz, 1829	Fins	This paper

WALLIKER (1969). Clearly there are significant differences between the different descriptions reported, which are all very poorly detailed, and for these reasons *M. inaequalis* is considered a *species inquirenda*.

Therefore, we consider that there are currently 29 valid species of *Myxobolus* parasitic on South American fishes whose diagnostic features are given in tables I and II.

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