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Structure and population dynamics of Myxosporeans (Myxozoa:Myxosporea), parasites of *Barbus callipterus* Boulenger, 1907(Cyprinidae) in the Soudano-guinean zone of Cameroon

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Abstract

In order to contribute to a better mastery and understanding of fish pathologies mainly Myxosporidiosis, so as to develop control strategies, 305 specimens of Barbus callipterus were sampled from May 2016 to May 2017 in Mapé River (Sanaga basin, Adamawa-Cameroon). Fish sampling and conservation were classical while Myxosporeans species were identified morphologically. A total of 13 species belonging to 4 genera (Myxobolus, Myxidium, Henneguya, Thelohanellus) were identified. Out of 305 specimens examined, 140 were significantly more infested by the genus Myxobolus (Prevalence = 45.90%). Irrespective of the parasite species, 147 fishes were infested (Prevalence = 48.20%). Three parasite species were secondary (10% ≤ Prevalence ≤ 50%) namely Myxobolus pseudodispar, M. sp_{10}, and M. umidus whereas the 10 others were scarce (Prevalence < 10%). The prevalence of species varied significantly from 0.65 to 14.10 % in M. muelleri, M. pharyngeus and M. pseudodispar respectively. The host's sex, class size and season did not significantly influence the prevalence and intensity of infestation. However, M. sp_2 was significantly more prevalent during the dry season. Kidneys of 209 fishes were infested (Prevalence = 68.20%) making it the most parasitized organs beside the 6 others target organs. Moreover, kidney harbored all parasite species. A broad spectrum of target organs was noticed for Myxobolus sp_{10} (infested 5 organs). Fishes were significantly more monoinfested than polyinfested. The awareness of the effects of endogenous and exogenous factors on Myxosporeans infestations is helpful to develop control strategies before the domestication of Barbus callipterus in order to boost its production.

Keywords: *Myxosporeans, Prevalence, Intensity, Barbus callipterus, Mapé River, Cameroon*

Introduction

According to FAO [1], fish represents nearly 51% of animal proteins intake in Africa. Climate change, rapid population growth and overfishing are some factors responsible for the decrease in fish's production [2, 3]. In addition to those constraints hindering fish's production, there are pathogens among which are Myxosporeans [4]. Myxosporeans affect fish's growth [5], their reproduction [6], and are involved in epizooties responsible for massive fish death [7, 8]. *Barbus callipterus* is a tropical endemic fish [9]. It is delicious and highly appreciated by many households and therefore, its pathologies should be taken into account in order to boost the production whether in natural or artificial environment. In Africa, researches emphasize more on Myxosporeans taxonomic than dynamics [3, 10]. In Cameroon particularly, few studies are focused on population dynamics of Myxosporeans apart from those of Tombi and Bilong Bilong [11], Lekeufack and Fomena [12], Nchoutpouen et *al.* [13]. Bilong Bilong and Tombi [14] claimed that, the host / parasite equilibrium is dynamic in natural environment and that anthropogenic activities can modify the physico-chemical characteristics of water leading to fishes' stress. Furthermore, water becomes more conducible to epizooties that can result in massive fish's deaths and important economic losses. Effective drugs against Myxosporeans being unavailable [12], a better knowledge and adequate monitoring of both endogenous and exogenous factors affecting Myxosporeans can help interrupting their life cycle. This study aims at increasing the yield of fish's production via the better understanding of their pathologies / Myxosporidiosis. Particularly, it intends assessing the effects of exogenous factors (seasons) and endogenous factors (host's sex, size, organs) over the prevalence and intensities of

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Myxosporeans infestations in *Barbus callipterus* in Cameroon.

Materials and methods

Study site

Fishes were sampled in MAPE River (tributary of Mbam River) in a village named MGBADJI (6°00'- 6°20'NL and 11°20'-11°40' EL, Bankim subdivision , Adamawa – Cameroon Region , Central Africa). The average altitude is about 724m. The soil is a mixture of clay and sand. The climate is of tropical Soudano-Guinean type with two seasons: a long rainy season running from March to November and a short dry season from November to March. The annual average temperature is about 23°C and the rainfall varies between 1500 and 2000 mm [15].

Fish sampling and conservation

Fishes were bought monthly from fishermen during the study period i.e., May 2016 to May 2017. They were captured both at the day and night using fish nets and fishing canes. On the field, specimens were immediately stored at 10% formalin solution and transported to the laboratory for examination.

Identification of myxosporeans

In the laboratory, fishes were identified according to Stiassny et *al*. [9] and examined according to the method used by Abakar [3]. So, standard and total lengths were measured to the closest millimeter using a slide caliper of stainless brand. Fishes were weighed using Sartorius electronic scale of 0.01g accuracy and were sex determined after dissection. External organs (fins, skin, scales and eyes) and internal organs (gills, spleen, kidneys, intestines, gall bladder, stomach and gonads) were examined with naked eyes, then with Motic stereoscopic microscope at 10X to look for the macroscopic cysts. As for kidneys, spleen and gonads, three smears were made per organ (anterior, medium and posterior regions) and examined at a total magnification of 1000X with a light microscope in order to look for spores. Spores were counted in 40 microscope fields for each smear [16]. Cysts were crushed between slide and cover glass in a drop of distilled water and their contents were identified with the light microscope at 1000X. Spores were fixed using methanol, stained with May-Grünwald-Giemsa and snapped with digital camera, Canon Ixus brand. Species were identified according to Lom and Arthur [17].

Parasitological parameters studied

The prevalence (Pr) of infestations expressed in percentage was defined as the number of host species infested by a given parasite species divided by the number examined [18]. The status of each parasite species was determined according to Valtonen et *al.* [19], therefore, parasites were qualified as frequent or principal (Pr > 50 %); secondary or intermediate (10 % ≤ Pr ≤ 50 %) ; scarce or satellite (Pr< 10 %)**.** The intensity (I) of infestation was the sum total of cysts (cysts load) or spores (spores load) of a given parasite species divided by the number of host harboring at least one cyst or one spore of that parasite [6]. The intensities were classified according to Bilong Bilong and Njiné [20]. So, intensities were very low (I < 10), low (10 ≤ I ≤ 50), average (50 < I ≤ 100) and high (I >100).

Statistical analysis

The Chi-square (X^2) test was used to compare prevalences. The H test of Kruskal-Wallis helped to compare several intensities while the U test of Mann Whitney was used to separate intensities. Spearman correlation coefficient "r" was calculated to search the relationships between parasitological parameters and variables. The error probability was P < 0.05 and the Graph Pad Prism 5 software was used for analysis.

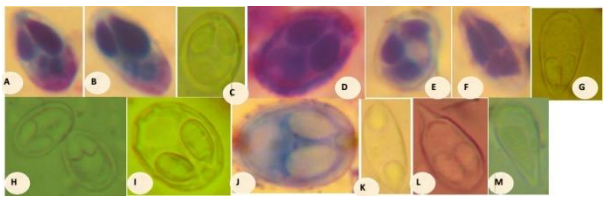
Results

Structure of the hosts' population

A total size of 305 fishes was captured. The sex ratio was skewed toward males (159 males against 146 females i.e. 1.09/1).The standard lengths ranged from 52 to 110 mm with an average of 94.66 mm. Based on these sizes, fishes were grouped into 3 classes of 20 mm amplitude each. The modal class i.e.] 70 - 90] represented 64.92% of sampled fishes. The average weight was 28. 89 g and varied between 3.89g and 42.95g.

Myxosporeans fauna of Barbus callipterus

The myxosporeans fauna recorded as shown in figure 1 was composed of 13 species belonging to 4 genera: *Myxobolus* (10 species), *Myxidium* (1 species), *Henneguya* (1 species) and *Thelohanellus* (1 species)



A : M*yxobolus tchadanayei* Abakar et *al*., 2006 *(*x 1500)
B : *Myxobolus* sp$_2$ *(x 1200)*
C : *Myxobolus muelleri* Bütschli ,1882 (x 1500)
D : *Myxobolus ellipsoides* Thélohan, 1982 (x 1500*)*
E : *Myxobolus pseudodispar* Gorbunova, 1936 (x 1500)
F : *Myxobolus pharyngeus* Parker et *al*., 1971 (x 1500)
G : *Myxobolus sp$_{10}$* (x 1000)
H : *Myxobolus umidus* Carriero MM et *al*., 2013 (x 1500)
I : *Myxobolus sessabai* Lekeufack et *al*., 2017 (x 1500)
J : *Myxobolus ngassami* Lekeufack et *al*., 2017 (x 1500)
K : *Myxidium barbatulae* Cépède, 1906 (x 1000)
L : *Henneguya ntemensis* Fomena and Bouix, 1996 (x 1500)
M :*Thelohanellus valeti* Fomena and Bouix, 1987 (x 1500)

Figure 1: Spores micrographs of myxosporeans studied

Prevalence of infestations

Prevalence of the genera and myxosporeans species

The prevalence of the genera and myxosporeans species (Figure 2) shows that the genus *Myxobolus* was significantly and highly the most prevalent (X^2 = 370.40; P < 0.001). Moreover, its prevalence (45. 90%) was about 46 times greater than the lowest prevalence (0.98%) recorded in *Henneguya*. When the prevalence of myxosporeans species is taken into account, it appears that, regardless of the parasite species, the overall prevalence was 48.20%. Three parasite species were secondary namely *Myxobolus pseudodispar* (14.10%), *M. sp$_{10}$* (11.15%) and *M. umidus* (12.79%) whereas the 10 others were scarce (Pr<10%). The prevalence of species varied very considerably (X^2 = 145.90; P < 0.001) from 0.65 % in *M. muelleri* and *M. pharyngeus* to 14.10% in *M. pseudodispar.*

Prevalence of parasite species as a function of class size

The prevalence of parasite species as a function of class size (Table 1) illustrates that fishes were infested in all class sizes. Irrespective of the parasite species, the infestation rates increased not significantly (X^2 = 4.21; P = 0.122) with the host's length. Hence 43.82; 51.01; and 77.78% were respectively the prevalence in the classes [50 - 70],] 70 - 90] and] 90 - 110]. Comparing the classes in term of species richness shows that, all the 13 parasite species recorded were present in the class] 70 - 90], followed by [50 - 70] with 11 species. Only 7 species were noticed in the oldest class. No matter the parasite species and the class size, the prevalence did not vary remarkably (P > 0.05) with the parasite species except in *M. pseudodispar* whose prevalence fluctuated significantly (X^2 = 7.95; P < 0.05) from 10.11 to 44.44% respectively in the classes [50 - 70] and] 90 - 110]. The infestation rates varied significantly (P < 0.05) between parasite species in all classes.

Table 1: Prevalence of parasite species as a function of class size

Parasite species	Class size (mm) [50 - 70] N = 96	] 70 - 90] N = 198	] 90 - 110] N = 11	X^2	P
M. tchadanayei	8.99	6.06	11.11	1.03	0.599
M. sp$_2$	6.74	10.10	22.22	2.57	0.276
M. muelleri	1.12	0.51	0.00	0.41	0.813
M. ellipsoides	3.37	3.54	11.11	1.42	0.491
M. pseudodispar	10.11	15.15	44.44	7.95	< 0.05
M. pharyngeus	0.00	1.01	0.00	0.10	0.608
M. sp$_{10}$	10.11	12.12	11.11	0.25	0.884
M. umidus	11.24	13.13	33.33	3.49	0.175
M. sessabai	1.12	4.55	0.00	2.53	0.283
M. ngassami	7.87	11.62	0.00	1.20	0.369
Myxidium barbatulae	5.62	5.05	0.00	0.54	0.765
H. ntemensis	1.12	1.01	0.00	0.10	0.350
T. valeti	1.12	2.53	0.00	0.80	0.671
Total	43.82	51.01	77.78	4.20	0.122
X^2	36.62	101.00	14.14		
P	< 0.001	< 0.001	< 0.05		

N: number of examined fishes

Prevalence as a function of host's sex

The prevalence as a function of host's sex illustrated in figure 3 reveals that both males and females were infested. Irrespective of the parasite species, male fishes were more infested (81.76%) than females (78.08%), however no significant difference (X^2 = 0.64; P = 0.422) was observed between both prevalences. When parasite species are considered, it reveals that *M. muelleri* and *M. pharyngeus* infested only a single sex. Whether in males (X^2 = 86.86; P < 0.001) or in females (X^2 = 62.71; P < 0.001), the infestation rates differed highly and considerably between parasite species. In males, prevalence ranged from 0.33 to 7.87% respectively for *Henneguya ntemensis* and *Myxobolus pseudodispar*. On the contrary, in females, the lowest (0.66%) prevalence was observed in *M. muelleri*, *H. ntemensis* and *Thelohanellus valeti* while *M. pseudodispar* exhibited the highest infestation rate (6.23%).

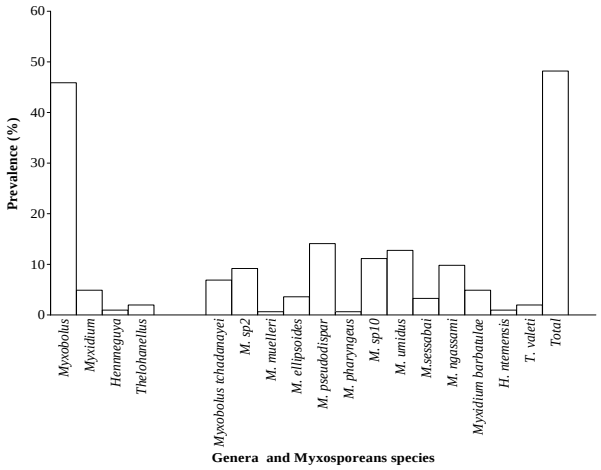


Figure 2: Prevalence of the genera and myxosporeans Species

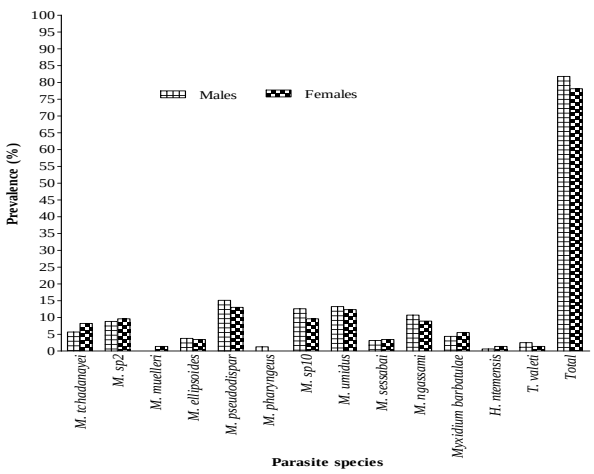


Figure 3: Prevalence as a function of host's sex

Prevalence of parasite species as a function of organs

The prevalence of parasite species as a function of organs illustrated in table 2 shows 7 parasitized organs. The comparison of species richness of target organs indicates that kidneys, operculum and liver were the most colonized with 13; 6 and 5 parasite species respectively. On the contrary, fins, gills and gall bladder were infested by only one species. Out of 7 infested organs, the prevalence of parasites varied significantly in operculum (X^2 = 11.43; P < 0.05) and highly remarkably in kidneys (X^2 = 164.50; P < 0.001) and gall bladder (X^2 = 156.12; P < 0.001). In the kidney, *M. pseudodispar* (13.77%) followed by *M. umidus* (12.46%) exhibited the highest infestation rates contrary to *M. muelleri* (0.66%). The comparison of parasite species according to the spectrum of target organs shows a broad spectrum for *M. sp$_{10}$* (5 organs) and *M. ngassami* (4 organs) whereas *M. muelleri, M. ellipsoides, M. pharygeus* and *H. ntemensis* appeared to be specific to only one organ precisely the kidneys.

Table 2: Prevalence (%) of parasite species as a function of organs

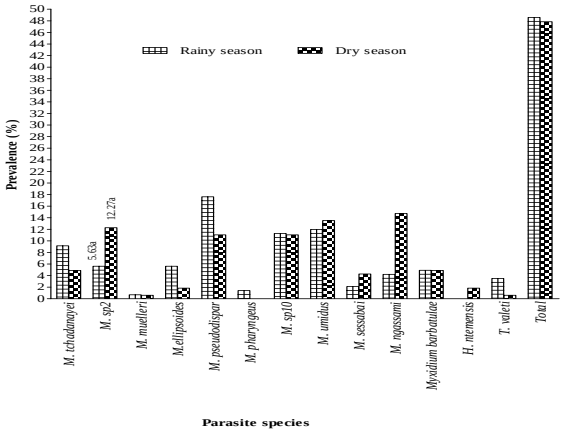
Parasite species	Infested organs						
	Operculum	Skin	Fins	Gills	Kidneys	Liver	Gall bladder
M. tchadanayei	0.33	0.00	0.00	0.33	6.23	0.00	0.00
M. sp$_2$	0.00	0.00	0.00	0.00	8.52	0.66	0.00
M. muelleri	0.00	0.00	0.00	0.00	0.66	0.00	0.00
M. ellipsoides	0.00	0.00	0.00	0.00	3.61	0.00	0.00
M. pseudodispar	0.33	0.00	0.00	0.00	13.77	0.00	0.00
M. pharyngeus	0.00	0.00	0.00	0.00	0.66	0.00	0.00
M. sp$_{10}$	1.97	0.66	0.33	0.00	8.52	0.33	0.00
M.umidus	0.00	0.00	0.00	0.00	12.46	0.66	0.00
M. sessabai	0.33	0.33	0.00	0.00	2.62	0.00	0.00
M. ngassami	0.33	0.33	0.00	0.00	8.85	0.33	0.00
Myxidium barbatulae	0.00	0.00	0.00	0.00	0.66	0.00	4.26
H. ntemensis	0.00	0.00	0.00	0.00	0.98	0.00	0.00
T. valeti	0.33	0.00	0.00	0.00	0.98	0.66	0.00
Total	3.61	1.31	0.33	0.33	68.52	2.62	4.26

Effects of seasons on the prevalence of parasite species

The effects of seasons on the prevalence of parasite species exhibited in figure 4 reveals that hosts were infested during the dry and rainy seasons. Irrespective of the parasite species, fishes were more infested in rainy season than in dry season, however without significant difference (X^2 = 0.02; P = 0.898). When the occurrence of parasites is taken into consideration, it appears that *M. pharyngeus* and *H. ntemensis* appeared only during a single season. On the one hand, during the rainy season, prevalence was significantly higher (X^2 = 80.15; P < 0.001; Pr = 17.61%) with *M. pseudodispar* and lower (0%) in *H. ntemensis.* On the other hand, *M. ngassami* was the most present during dry season (X^2 = 98.48; P < 0.001; Pr = 14.72%) and *M. pharyngeus* absent (0%). *Myxobolus* sp$_2$ was the only parasite species that exhibited a significant variation of prevalence with season. Furthermore, it was more prevalent (X^2 = 4.00; P < 0.05) in the dry season than in the rainy season.

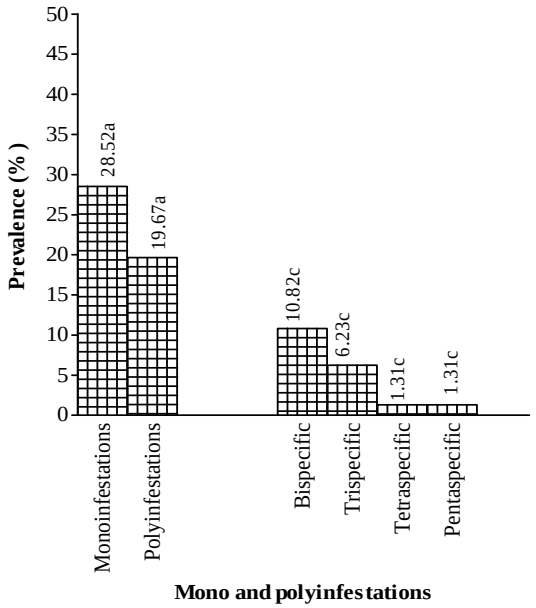
Prevalence of mono and polyinfestations

The prevalence of mono and polyinfestations is summarized in figure 5. It appears that fishes were infested by a single (monoinfestations) and more than one (polyinfestation) parasite species. Overall, 28.52% of examined fishes were monoinfested against 19.67% polyinfested (P < 0.001). As shown in figure 5, there were 4 categories of mixed infesfations (species combinations) namely bi, tri, tetra and pentaspecific corresponding respectively to 2; 3; 4; and 5 parasite species. Their frequencies dropped very significantly (P < 0.001) with the increasing number of combined parasite species.



Values having the letter "a" are significantly different (P < 0.05)

Figure 4: Prevalence of parasite species as a function of seasons



Values having letters "a" and "c "differ significantly at: P < 0.05 (a) and P < 0.001 (c)

Figure 5: Prevalence of mono and polyinfestations

Intensities of infestations

Intensities of myxosporeans genera

The intensities of myxosporeans genera (Table 3) indicate that fishes were infested by both cysts and diffused spores. Cysts were found only in the genera *Myxobolus* and *Thelohanellus* while all the 4 genera harbored diffused spores. Whatever be the form of parasites, the intensities were very low. The intensities of infestations by diffused spores varied significantly between genera (H = 15.99; P < 0.05). In addition, fishes were more infested by *Myxobolus* than *Myxidium* spores (U = 421.50; P < 0.05).

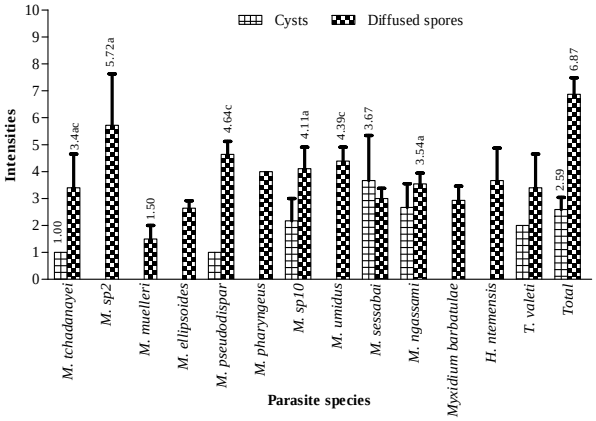
Table 3: Intensities of myxosporeans genera

Genera	Cysts	C_k / N	Spores	C_k / N
Myxobolus	2.53 ± 0.51	38 / 15	6.72^a ± 0.61	867 / 129
Myxidium	-	-	2.80^b ± 0.50	42 / 15
Henneguya	-	-	3.07 ± 1.20	11 / 3
Thelohanellus	2.00 ± 0.00	2 / 1	3.40 ± 1.25	17 / 5

Intensities are followed by the standard deviation; -: not infested ; C_k: cysts load; C_k : spores load; Values having different letters are significantly different (P < 0.05); N: number of infested fishes

Intensities of myxosporeans species

The intensities of myxosporeans species (Figure 6) were very low. Independently on the parasite species, the overall intensities were 2.59 and 6.87 respectively for cysts and diffused spores. The comparison of cysts intensities between species shows that they varied but not remarkably (H = 4.28; P = 0.510) from 1.0 (*M. tchadanayei* and *M. pseudodispar*) to 3.67 in *M. sessabai*. Meanwhile, the intensities of diffused spores fluctuated highly and significantly (H = 38.50; P < 0.001) from 1.50 to 5.72 in *M. muelleri* and *M*. sp_2 respectively.

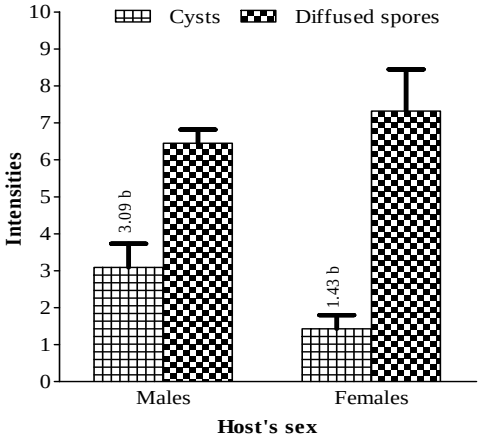


Values having the same letter are significantly different at: P < 0.05 (a) and P < 0.001 (c)

Figure 6: Intensities of myxosporeans species

Intensities as a function of host's sex

The intensities were low in both sexes (Figure 7) and very significantly (U= 20.50; P < 0. 01) higher in males when infested by cysts. Although spore intensities were higher in females than males, no significant difference was observed (U = 2243; P = 0.912).



Values having the same letter are significantly different (P < 0.01)

Figure 7: Intensities as a function of host's sex

Intensities of parasite species as a function of host's sex

The intensities of parasite species as a function of host's sex (Table 4) show that, males and / or females were

infested by cysts and / or diffused spores depending on the parasite species. Whatever the parasite species, the intensity did not vary significantly (P > 0.05) between males and females except *M. umidus* in which female fishes harbored more spores than males (U = 95.00; P < 0.05).

Intensities as a function of class size

The intensities as a function of class size illustrated in table 5 did not vary significantly between classes. Moreover, no significant correlation was found between cyst or spore loads and fishes' length (cysts: r = + 0.03; P = 0.237; spores: r = -0.05; P = 0.561)

Table 4: Intensities of parasite species as a function of host's sex

Parasite species	Hosts sex			
	Males		Females	
	Cysts	Spores	Cysts	Spores
M. tchadanayei	1.00 ± 0.00	2.29 ± 0.18	-	2.50 ± 0.28
M. sp₂	-	-	2.29 ± 0.18	2.50 ± 0,28
M. muelleri	-	-	-	1.50 ± 0.50
M. ellipsoides	-	2.50 ± 0.29	-	2.71 ± 0.42
M. pharyngeus	-	3.00 ± 1.00	-	-
M. sp₁₀	3.60 ± 0.81	4.13 ± 0.64	1.00 ± 0.00	4.09 ± 1.80
M. umidus	-	3.55[a]± 0.58	-	5.56[a]± 0.90
M. sessabai	4.50 ± 2.50	2.33 ± 0.33	2.00 ± 0.00	3.50 ± 0.50
M. ngassami	1.00 ± 0.00	3.67 ± 0.61	3.00 ± 0.00	3.42 ± 0.38
Myxidium barbatulae	-	3.29 ± 0.97	-	2.38 ± 0.46
H. ntemenis	-	2.00 ± 0.00	-	4.50 ± 1.50
T. valeti	-	3.25 ± 1.60	2.00 ± 0.00	4.00 ± 0.00

Intensities are followed by the standard deviation; : -: not infested; values having letter "a" are significantly different (P < 0.05)

Table 5: Intensities as a function of class size

Intensities	Class size (mm)			H	P
	[50 - 70]	] 70 - 90]	] 90 - 110]		
Cysts	1.50 ± 0.50	3.20 ± 0.65	3.00 ± 0.00	4.45	0.110
Spores	7.18 ± 1.34	6.76 ± 0.72	7.00 ± 1.86	0.32	0.851

Intensities are followed by the standard deviation; P: error probability H: Kruskal –Wallis value

Intensities as a function of organs

The intensities as a function of organs assigned in figure 8 show that cysts were found in the kidneys, liver and gall bladder contrary to operculum, skin and fins. Cyst intensities were not only very low in kidneys and gall bladder and low in liver, but they did not fluctuate considerably between those organs (H = 6.02; P = 0.221). The spores' intensities were maximum (3.08) and minimum (1.00) respectively in operculum and gills (P > 0.05).

Intensities of parasite species as a function of organs

The intensities of parasite species as a function of organs (Figure 9) show that *Myxobolus umidus* recorded higher cyst (I = 13.00) and spore (I=7.00) intensities in the liver and operculum respectively. Although the spores of all parasite species were encountered in the kidneys, their intensities varied not significantly (P > 0.05) from 1.5 to 6.04 respectively for *M. muelleri* and *Myxobolus* sp_2.

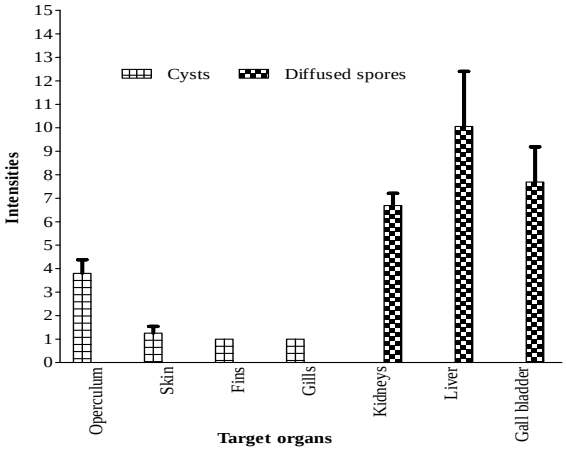
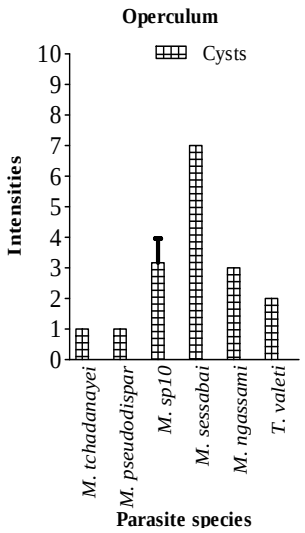


Figure 8: Intensities as a function of organs



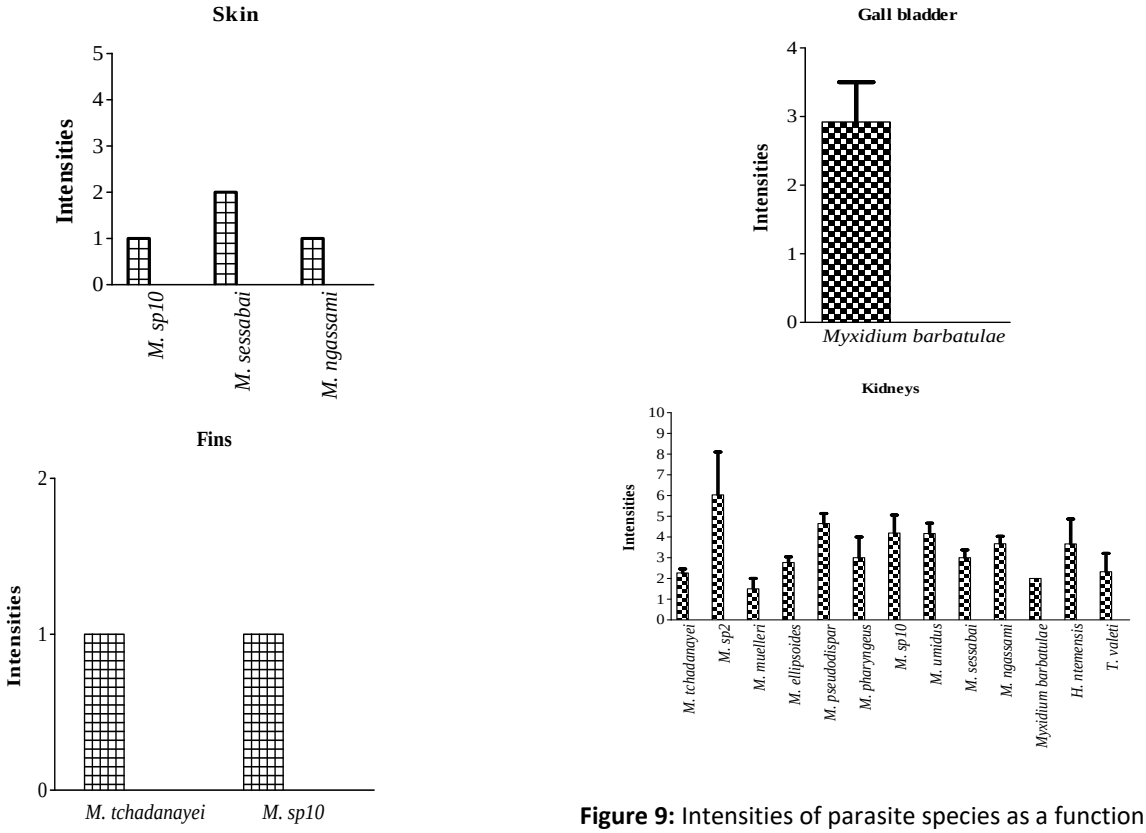


Figure 9: Intensities of parasite species as a function of organs

Effects of seasons on the intensities of infestations

The effects of seasons on the intensities of infestations are summarized in table 6. It appears that, irrespective of the parasite species, cyst (U = 27.50; P = 0.476) and spore (U = 1857; P = 0.069) intensities were higher during the dry season than the rainy season. The intensities of all species did not vary within and between seasons (P > 0.05). During the dry season, *M*. sp_2 exhibited higher spores intensity (I = 6.78) whereas *M. muelleri* and *T. valeti* spores were less encountered (I = 2.00). On the other hand, *M. muelleri* (I = 1.00) and *M. pseudodispar* (I = 4.04) spores were mostly recorded during the rainy season.

Table 6: Effects of seasons on the intensities of infestations

Parasite species	Seasons			
	Rainy season		Dry season	
	Cysts	Spores	Cysts	Spores
M. tchadanayei	1.00± 0.00	1.92 ± 0.15	-	2.85 ± 0.34
M. sp_2	-	3.00 ± 0.44	-	6.78 ± 2.62
M. muelleri	-	1.00± 0.00	-	2.00 ± 0.00
M. ellipsoides	-	2.50 ± 0.35	-	3.00 ± 0.58
M. pseudodispar	1.00 ± 0.00	4.04 ± 0.71	-	5.37± 0.60
M. pharyngeus		3.00 ± 1.00	-	-
M. sp_{10}	2.00 ± 1.00	3.86 ± 0.73	2.83± 0.87	4.38 ± 1.51
M. umidus		3.88 ± 0.75	-	4.81 ± 0.73
M. sessabai	2.00 ± 0.00	2.50 ± 0.30	-	4.50 ± 2.50
M. ngassami	-	3.75 ± 1.22	2.00± 0.35	3.48 ± 0.29
Myxidium barbatulae	-	2.71 ± 0.57	-	2.88 ± 0.85
H. ntemensis	-	-	-	3.67 ± 1.20
T. valeti	2.00 ± 0.00	3.75 ± 1.55	-	2.00 ± 0.00
Total	2.00 ± 0.43	5.65 ± 0.57	3.00± 0.70	7.94 ± 1.02

Intensities are followed by the standard deviation ; -: not infested

Intensities of mono and polyinfestations

The intensities of mono and polyinfestations (Figure 10) illustrate that polyinfestations intensity with cysts was about twice higher than that of monoinfestations, however without any significant difference (U = 116.50; P = 0.932). On the contrary, polyinfestations with spores were remarkably more represented (U = 2770; P < 0.001) than monoinfestations. The comparison of the intensities of the categories of polyinfestations reveals that whether infested by spores or cysts, intensities did not fluctuate significantly (P > 0.05).

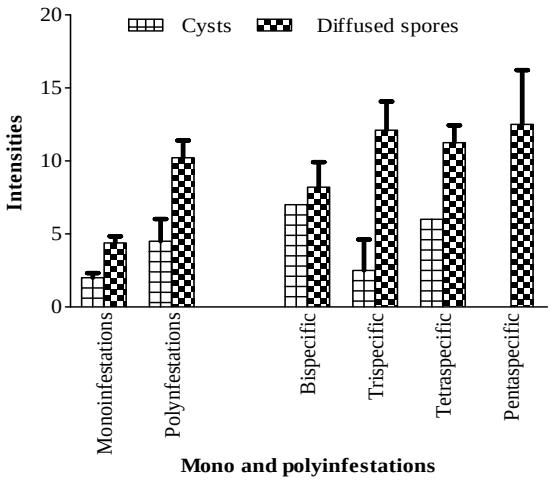


Figure 10: Intensities of mono and polyinfestations

Discussion

The diversity of the myxosporeans fauna can be buttressed by Combes [21] assertion as which, pathogenic effects are scarcely caused by a single parasite species. Among the four myxosporeans genera recorded, *Myxobolus* spp exhibited a higher prevalence. This result is not new. In fact, Lom and Diková [4] estimated that, the world of myxosporidia fauna was composed of about 2180 species gathered within 62 genera among which the genus *Myxobolus* Bütschli, 1882 represented about 36. 33% of species (792 species). This observation is in agreement with the findings of Lekeufack and Fomena [12] who recorded in the River Sangé in Cameroon 54.55% of myxosporeans belonging to the genus *Myxobolus* infesting various hosts namely *Ctenopoma petherici, Clarias pachynema* and *Hepsetus odoe.* Eiras et *al*. [22] reported that 29 *Myxobolus* species were identified from the fishes of the genus *Barbus* and related species in the Rivers of Iberian Peninsula.

The prevalence was relatively low. This is in accordance with Euzet and Pariselle [23] who asserted that, the low prevalence in natural milieu is due to the equilibrium established during the evolution of host / parasite system. El-Tantawi [24] thought that for a given parasite, the host infestation rate and the status of the parasite species vary geographically. The prevalence of infestations did not vary with the size of fish. This result is not in accordance with some findings which showed that younger fishes were more infested than the older ones or vice versa. This may be due to the small sample size of older fishes. In fact the fish's population sampled was essentially young. Nchoutpouen et *al*. [13] pointed out that, in farming situation, older *Oreochromis niloticus* were more infested than the younger ones. The most common tendency is the decreasing of the infestation rate with the size (age) of fish. So, Tombi and Bilong Bilong [11] , Viozzi and Flores [25], Abakar [3] found that young fishes were more vulnerable to myxosporeans infestations than older ones. The same observation was made by Brummer – Korvenkontio et *al*. [26] in Finland where the prevalence of infestation of *Rutilus rutilus* by *Myxobolus rhodei* and *M. pseudodispar* decreased with the fish age. These authors explained their observation by the increase of the immune system response with the size of fish.

The fact that host's sex did not influence the parasitism of fishes is in agreement with the claims of Abakar [3], Lekeufack and Fomena [12]. Fomena [27] didn't find any difference between the infestation rates of males and females *Oreochromis niloticus* at Mélen fish ponds (Yaoundé –Cameroon) by kidneys and livers myxosporeans. Likewise, Viozzi and Flores [25] noticed that the prevalence of *Myxobolus biliare in Galaxias maculatus* was sex independent and claimed to be the global situation with myxosporeans infestations. However, males harbored more cysts than females. Gbankoto et *al*. [8] declared that male fishes harbored more cysts than female. Poulin [28] thought that this observation might be due to the loss of huge amount of energy by males for testosterone synthesis thus weakens the efficiency of fish immune system.

The seasons did not influence the infestation rates of all parasite species (except *M*. sp_2). Irrespective of the parasite species, cysts and spores intensities were higher during the dry season than the rainy season. Only *Myxobolus* sp_2 exhibited significant high prevalence during the dry season; this is in accordance with the observation of Abakar [3] who reported that *Myxobolus brachysporus* and *M. camerounensis* appeared in *Oreochromis niloticus* mainly during the dry season. In addition, Gbankoto et *al*. [8] showed that *Myxobolus sp* and *Myxobolus zillii* which are gills' parasites of *Tilapia zillii* and *Sarotherodon melanotheron melanotheron* in Bénin were more frequent during the dry season. This observation was explained by Obiekezie and Okaeme [6] who thought that during the dry season, the high temperature of water and mud might encourage the infestation with myxosporeans. Uspenkaya [29] opined that myxospores sink in water where they get aging and become mature in mud or sludge so as to infest the new host. Oligochaetes being intermediate hosts in myxosporeans life cycle [30], the seasonal variation of parasitism by myxosporeans could be due to the seasonal supplying of actinospores by oligochaetes [31]. The fact

that kidneys were the most infested organs may be because, since they filter blood and secrete many solutes [32], parasites converge there for metabolites need. The specificity of some parasites to a particular organ may be explained by the fact that, the organ provides suitable microbiotope conditions for optimal life and the exclusion of the parasites by competition. Parasites infesting the same organ perhaps do not compete [33]. The broad spectrum of target organs observed with *Myxobolus* sp_{10} may be due to the flexibility or versatility of its metabolic pathways. Ibrahim and Soliman [34] opined that the heterogeneity of biotopes generates different infestation sites which are also habitat options for parasites. The drop in polyinfetations' prevalence with the increasing number of associated parasite species may be explained by the interspecific competition. The higher the number of associated species, the higher the intensity of interspecific competition because of the shortage of resources thus the lower the prevalence.

Conclusion

At the end of our study which was aimed at evaluating the effect of endogenous factors (host's size, sex, organs) and exogenous factors (seasons) on the prevalence and intensities of myxosporeans infestations, the following conclusions can be drawn: The myxosporeans fauna was composed of 13 species belonging to 4 genera (*Myxobolus, Myxidium, Henneguya and Thelohanellus*); the parasitological parameters were affected by host's size, sex, organs and seasons. The infestations of our fishes may lead to several and severe pathologies. Ellis et *al*. [32] claimed that fish's kidneys are complex organs having hematopoietic, reticulo-endothelial, endocrine and excretory functions. So, its infestation can induce severe dysfunctions. Gills are used not only for breathing but also as osmoregulatory organ; its damage can lead to fish death. Ectomyxosporeans can cause skin damage facilitating access of secondary pathogens into the body. The awareness of the effect of these parasites is useful to develop control strategies before the domestication of *Barbus callipterus.*

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