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Prevalence of major parasites of Nile tilapia (*Oreochromis niloticus*) in south west Showa zone selected fish farms, Oromia region, Ethiopia

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Abstract
A cross-sectional study was conducted from November, 2016 to April, 2017 to determine the prevalence, parasite genera and associated risk factors of fingerling fish parasites in small scale fish farms found in South West Shewa Zone, Oromia. Out of the total Nile tilapia fish sampled for the study, 77.60% were found infected with different parasites. Higher prevalence of external parasites (32.29%) than internal parasites (20.31%) were encountered in the current study. Different parasites belonging to different taxonomic groups namely *Trichodina* (8.85%), *Euclinostomum* spp (7.81%), *Dactylogyrids* spp (4.95%), Plerocercoid *spp*. (2.34%), *Acanthocephala* spp. (2.08%), *Contracaecum* spp. (2.08%), *Clinostomum* spp (1.30%) and Black spot (0.52%) were recovered from the positive sampled fish. Besides, higher proportion of mixed infection (47.66%) was found. Distribution of parasitic genera was found to be statistically significant ($P<0.05$) across sexes, body weights, total lengths and collection months of the sampled fish in that male fish (79.71%), fish with body weight of 50-100g (81.00%), fish with total length of >20cm (92.00%) were found to be highly infected with parasites. Temporal variation also seen with collection month that the higher prevalence was recorded in fish samples collected from January-February (93.91%). In this study, higher prevalence of fingerling fish parasite infestation (both external and internal parasites) was recorded. Therefore, due attention should be given to prevent and control the parasite burden of the study area.

Keywords: Nile tilapia, parasites, prevalence, south west shewa zone

1. Introduction
One of the rapidly increasing food producing area is aquaculture globally [1]. Like other live stocks, fish is crucial in contributing for trade and economy to human beings. In countries where there is serious under nutrition especially in the tropics and subtropics, fish is of paramount importance in improving the diet of citizens [2]. Freshwater fish accounts for more than 80% of aquaculture products in the world [1]. For more than 1.5 billion people of the world, fish supply 15% of the their entire average per capital intake of animal protein [3]. In developing and low income deficit countries, a large proportion of fish products come from small-scale producers. Fish farming is attributing to the increased need of affordable animal protein in the tropics and there has been huge improvement of aquaculture [4].
Fish are prone to different health conditions due to environmental disorders and pathogens like bacteria, virus, fungus and commonly parasites. The usual effect of fish parasites on fish is causing illness, destroy the aesthetic value of fish by spoiling the look of fish and consequently causes refusal of the fish product by consumers [5]. This causes significant economic blow through morbidity, loss of productivity and treatment costs. Some fish borne parasites are becoming an important public health issues in recent years. Widely increased fish born human parasites is speeded up due to poor food hygiene and traditional methods of food preparation [6]. To apply an effective curative or prevention strategy, understanding the causes and taxa of parasites at least to the genus level is very important of crucial importance [7]. However, the presence of a massive number of parasites on each fish might constitute a real threat to the fish population and should require immediate action [8].
The absence of good facilities like laboratory equipment, skilled man power, lack of public awareness on the fish production and health, library facilities are the main factors for inadequate research commitments to the study of parasites fishes of the African [7].

In Ethiopia, fish disease research is limited but only for few researches which have not been well documented [9] and hence researches are the base to undertake intervention measures for fish disease associated problems in fish farms and many studies have to be implemented in this area for the future. Despite the facts, information on fish diseases are very few in the country [10, 11]. Moreover, there is no any research conducted on fish parasites from the South West Shewa Zone particularly from small scale fish farms. Therefore, the objectives of the current study were to identify the parasite genera in the study area, to determine the prevalence of fish parasites in the study area and to identify the associated risk factors of the disease.

2. Materials and Methods

2.1 Study Area

The study was conducted in South West Shewa Zone, Oromia Regional State, Ethiopia. South West Shewa Zone is found to the West of Addis Ababa and South of Ambo. The coordinates of the zone are 8°32′N latitude and 37°58′E longitude. It is characterized by temperate type of climate with daily temperature ranging from 18 ^{0}C – 27 ^{0}C with average temperature of 22.5 ^{0}C, and is located 1900m above sea level, characterized by warm temperature weather which is locally called "weynadega" (Temperate) [12]. The farming activity in the study area is mostly crop farming and animal husbandry. Fish farming is also one of the emerging agricultural activity in the region. The Zone is one of the priority area for fish farming and there are many ponds constructed for the purpose of aquaculture supported by the National Fishery Research center in Sebeta. In the study area, farmers are organized by group and the ponds are constructed with 15m by 20m fish farming purpose with inlet and outlet for water entrance and exit. The source of water for the ponds is the river passing across the district. The source of fingerlings to stock these small scale fish farmers were from Koftu Lake. The fish species introduced include *O. niloticus* and *T. zilli.* Six ponds were selected randomly and sampled for the present study.

2.2. Study Population

The study animals were *Oreocromis niloticus* fish species of sexes, different body weight and total length collected from ponds in S/ W/ Shewa zone.

2.3. Study Design and sampling method

A cross sectional study design was conducted to determine the prevalence of fingerling fish parasites, parasitic genera and associated risk factors in the study area. The prevalence of the parasites was determined in relation to risk factors; sex, different body weight, total length and collection months. The study animals were collected randomly from the ponds of the study area. The collected sample was transported to the National Fishery and Aquatic Life Research Center laboratory by using plastic container in which enough water and dissolved oxygen was applied.

2.4. Postmortem Examination

Total length and weight of all collected fish specimens was measured using board and balance, respectively. Each fish was examined externally and internally for the presence of any lesions and parasites following standard measurements. The codes of each fish, species, sex, total length, weight, name of the site, date of sampling, organ of fish sampled, and parasites observed were recorded. After thorough examination of sampled fish for external parasite by visualization and skin scraping, each fish was dissected and postmortem examination was done using appropriate postmortem kits and standard evisceration technique previously described by Zhokhov *et al*., [13]. Briefly, the whole body cavity was opened and examined for parasites. First, the skin scrap was taken using blade and placed on the microscope slide then cover slip was applied and examined under microscope. Then the gill filaments and various organs including eyes, stomach, intestine, liver and gonads were removed and placed separately in petri-dishes containing 0.75% saline solution. The external surface of each organ was examined and then opened for further examination for the presence of parasites. Presence of parasites in each fish organs and cavities was examined visually by naked eye, using stereomicroscope and compound microscopes. Parasites collected from each fish organs were examined under the microscope to identify each parasite to the lowest possible taxa using standard literatures and published papers [7].

2.5. Data Analysis

The collected raw data was entered in to Microsoft excel data sheets and analyzed using SPSS 20statistical software. Descriptive statistics was employed and expressed in terms of frequency and percentage. Chi-square (x^2) test statistics was used to determine the association between the risk factors and the prevalence of the parasites. For all analysis P-value<0.05 was used as cut-off point for significance difference.

3. Results

In the current study a total of 384 fish were sampled and examined for the presence of internal or external parasites. Out of the total sampled fishes *O. niloticus*, 77.60% of fishes were positive with both internal and external parasitic genera, of which 20.31% and 32.29% were infested by internal and external parasites, respectively. Among them 25.00% harbored multiple parasites belonging to the several genera (Table 1).

Table 1: The prevalence of parasite in the examined fingerlings

No. of fish examined	Type of parasite found			Total
	Internal	External	Mixed	
384	78 (20.31%)	124 (32.29%)	96 (25.00%)	298 (77.60%)

In this investigation, different parasites namely Plerocercoid (*cestoda*), *Dactylogyrids species*, *Euclinostomum* spp., *Clinostomum* spp, *Acantocephala* spp, *Contracaecum* spp, *Trichodina* spp and Black Spot belonging to different taxonomic groups were recovered from 298 positive sampled fish. From the encountered parasites, mixed infection (47.66%) took higher proportion. The most commonly prevalent parasites include *Trichodina* spp. (8.85%) followed by *Euclinostomum* spp. (7.81%) and *Dactylogyrids* spp. (4.95%) (Table 2).

Table 2: Distribution of parasites genera in the examined fingerlings

Parasites found	Frequency	Percentage
Plerocercoids	9	2.34%
Dactylogyrids	19	4.95%
Euclinostomum	30	7.81%
Clinostomum	5	1.30%
Acanthocephala	8	2.08%
Contracaecum	8	2.08%
Trichodina	34	8.85%
Black spot	2	0.52%
Mixed Infection	183	47.66%
Total	298	100%

In this study sex, total length, body weight and collection month were taken as a risk factor and their association with the prevalence of parasite infestation was measured. Males had higher prevalence of parasites infestation 165 (79.71%) than their Females counterparts 133 (75.14%). Higher prevalence of both internal (25.60%) and external (36.23%) parasites were found among male fish as compared to female fish, internal (14.12%) and external (27.68%). However, mixed infections were higher among female fish (33.33%) than male fish (17.87%). The variation in the prevalence of fish parasites between sexes was statistically significant (P<0.05) (Table 3).

Statistically significant (P<0.05) difference in the prevalence of parasites was found among the different body weights. Higher prevalence of parasites was among fish having weight of 50-100g (81.00%) followed by fish with body weight of <50g (77.15%) and >100g (74.71%). Higher prevalence of internal parasite was found among fish with body weight >100g (44.83%). On the other hand, prevalence of external parasite infestation was higher among fish with body weight of <50g (37.56%) and 50-100g (36.00%). Similarly mixed infestation was higher in fish with body weights of 50-100g (29.00%) and <50g (27.92%) (Table 3).

Higher prevalence of parasite was found in fish with total body length of >20cm (92.00%), followed by 10-20cm (76.98%) and <10cm (75.00%). Higher proportion of internal and external parasites were found in fish with total body length of >20cm (48.00) and <10cm (41.18), respectively. Likewise, the difference was found statistically significant (P<0.05) (Table 3).

Higher parasite burden was seen between January-February (93.91%) followed by parasite collecting months March-April (77.55%) and November-December (52.10%). Higher internal parasite burden was found from November to December (39.73%), whereas, higher prevalence of external parasite (46.09%) and mixed infestation (37.39%) was observed from January-February. The difference in prevalence among the different parasite collecting months were found statistically significant (P<0.05) (Table 3).

Table 3: Distributions of parasite in relation to sex, weight, length and collection month

Variables	No. fish examined	Parasite type				X^2 (P-value)
		Internal (%)	External (%)	Mixed (%)	Total (%)	
Sex						
Female	177	25 (14.12)	49 (27.68)	59 (33.33)	133(75.14)	18.3594 (0.000)
Male	207	53 (25.60)	75 (36.23)	37 (17.87)	165(79.71)	
Weight (g)						
<50	197	23 (11.68)	74 (37.56)	55 (27.92)	152(77.15)	49.6229 (0.000)
50-100	100	16 (16.00)	36 (36.00)	29 (29.00)	81(81.00)	
>100	87	39 (44.83)	14 (16.09)	12 (13.79)	65(74.71)	
Total length						
<10 cm	68	10 (14.71)	28 (41.18)	13 (19.12)	51(75.00)	17.5773 (0.007)
10-20cm	291	56 (19.24)	89 (30.58)	79 (27.15)	224(76.97)	
>20cm	25	12 (48.00)	7 (28.00)	4 (16.00)	23(92.00)	
Collection month						
November-December	73	29 (39.73)	4 (5.48)	5 (6.85)	38(52.10)	94.1523 (0.000)
January-February	115	12 (10.88)	53 (46.09)	43 (37.39)	108(93.91)	
March-April	196	37 (18.88)	67 (34.18)	48 (24.49)	152(77.55)	

In the current study parasitic distribution of fish was also studied in relation to the sex, weight, total length and collection month. Accordingly, *Trichodina* (12.03%), *Euclinostomum* (7.52%) and *Dactylogyrids* (6.77%) were the popular parasites identified from female fish of the study area with *Contracaecum* (0.00%) parasite infestation. Similarly, in male fish the commonest parasite investigated are *Euclinostomum* (12.12%), *Trichodina* (10.91%), *Dactylogyrids* (6.06%), *Contracaecum* (4.85%) and Plerocercoid (4.24%). However, Black spot were absent from male fish (0.00%). Comparable higher mixed infestation was observed in both female (65.41%) and male fish (58.18%) (Table 4).

Table 4: Distribution of parasite genera in relation to sex

Genera of Parasite	Sex	
	Female	Male
Plerocercoid	2 (1.50%)	7 (4.24%)
Dactylogyrids	9 (6.77%)	10 (6.06%)
Euclinostomum	10 (7.52%)	20 (12.12%)
Clinostomum	3 (2.26%)	2 (1.21%)
Acanthocephala	4 (3.01%)	4 (2.42%)
Contracaecum	0 (0.00%)	8 (4.85%)
Trichodina	16 (12.03%)	18 (10.91%)
Black spot	2 (1.50%)	0 (0.00%)
Mixed Infection	87 (65.41%)	96 (58.18%)
X^2 (P-value)	13.6451(0.091)	

Trichodina (17.11%) *and Euclinostomum* (10.53%) were the major parasites found among fish having<50g body weight. Among fish with body weight of 50-100g, higher proportion of *Trichodina* (7.41%), *Dactylogyrids* (8.64%), Plerocercoid (4.94%) *and Euclinostomum* (4.94%) were found. Whereas, among fish with body weight >100g, *Euclinostomum* (15.38%), *Contracaecum* (9.23%), *Dactylogyrids* (7.69%) and Plerocercoid (7.69%) were the popular parasites identified. The difference in the distribution of parasite among the different body weights were found statistically significant (P<0.05) (Table 5).

Table 5: Distribution of parasite genera in relation to weight of fingerlings

Genera of Parasite	Weight (gm)		
	<50	**50-100**	**>100**
Plerocercoid	0 (0.00%)	4 (4.94%)	5 (7.69%)
Dactylogyrids	7 (4.61%)	7 (8.64%)	5 (7.69%)
Euclinostomum	16 (10.53%)	4 (4.94%)	10 (15.38%)
Clinostomum	2 (1.32%)	1 (1.23%)	2 (3.08%)
Acanthocephala	2 (1.32%)	2 (2.47%)	4 (6.15%)
Contracaecum	0(0.00)	2 (2.47%)	6 (9.23%)
Trichodina	26 (17.11%)	6 (7.41%)	2 (3.08%)
Black spot	0 (0.00%)	2 (2.47%)	0 (0.00%)
Mixed Infection	99 (65.13%)	53 (65.43%)	31 (47.69%)
X^2 (P-value)	52.5778 (0.000)		

Only three parasites; *Euclinostomum* (21.57%), *Trichodina* (17.65%) and *Dactylogyrids* (3.92%) were identified from fish with total length of <10cm. *Trichodina* (11.16%), *Dactylogyrids* (6.70%) and *Euclinostomum* (6.70%) were the popular parasites identified from fish of 10-20cm total length. Similarly, fish with total length of >20cm found to have higher proportion of *Euclinostomum* (17.39%), Plerocercoid (8.70%) and *Dactylogyrids* (8.70%). The difference in the distribution of parasite among the different total body lengths were found statistically significant (P<0.05) (Table 6).

Table 6: Distribution of parasite genera in relation to total length of fingerlings

Genera of Parasite	Length (cm)		
	<10	**10-20**	**>20**
Plerocercoid	0(0.00%)	7 (3.13%)	2 (8.70%)
Dactylogyrids	2 (3.92%)	15 (6.70%)	2 (8.70%)
Euclinostomum	11 (21.57%)	15 (6.70)	4 (17.39%)
Clinostomum	0 (0.00%)	5 (2.23%)	0 (0.00%)
Acanthocephala	0 (0.00%)	6 (2.68%)	2 (8.70%)
Contracecum	0 (0.00%)	8 (3.57%)	0 (0.00%)
Trichodina	9 (17.65%)	25 (11.16%)	0 (0.00%)
Black spot	0 (0.00%)	2 (0.89%)	0 (0.00%)
Mixed Infection	29 (56.86%)	141 (62.95%)	13 (56.52%)
X^2 (P-value)	29.3118 (0.022)		

Except *Trichodina* (0.00%) and Black spot (0.00%) all other parasites were investigated from fish collection months of November-December. Higher proportion of *Trichodina* (16.67%) *and Euclinostomum* (9.26%) parasites were collected from January to February. All type of parasites was collected from fish collecting month of March to April. Higher distribution of mixed infestation of parasite was observed from January-February (71.30%) followed by fish collecting months March-April (60.53%) and November-December (36.84%). The difference in the distribution of parasite among the different fish collection months were found statistically significant (P<0.05) (Table 7).

Table 7: Distribution parasite genera in relation to collection months of fingerlings

Genera of Parasite	Collection Month		
	November-December	**January-February**	**March-April**
Plerocercoid	5 (13.16%)	0 (0.00%)	4 (2.63%)
Dactylogyrids	4 (10.53%)	2 (1.85%)	13 (8.55%)
Euclinostomum	5 (13.16%)	10 (9.26%)	15 (9.87%)
Clinostomum	2 (5.26%)	1 (0.93%)	2 (1.32%)
Acanthocephala	4 (10.53%)	0 (0.00%)	4 (2.63%)
Contracaecum	4 (10.53%)	0 (0.00%)	4 (2.63%)
Trichodina	0 (0.00%)	18 (16.67%)	16 (10.53%)
Black spot	0 (0.00%)	0 (0.00%)	2 (1.32%)
Mixed Infection	14 (36.84%)	77 (71.30%)	92 (60.53%)
X^2 (P-value)	63.3832 (0.000)		

4. Discussion

In the current study, an overall prevalence of 77.60% fish parasite was found. This finding is in line with previous reports of 75.67% Zekaryas [14] at Lake Hawassa, 73.24% Teferra [15] at Lake Tana. On the other hand, the current finding is higher than the study conducted by Amare *et al.* [9]

who found prevalence rate of 47.8% at Lake Lugo. The variation in prevalence might be due to parasitic infections can be devastating in farmed organisms than in wild populations because of stressful conditions linked to crowding (high rate of fertility) and frequent water quality deterioration [16] or absence of proper waste disposal and management system.

In the current study the overall prevalence of external parasite was found to be 32.29%. *Trichodina* spp. (8.85%) and *Dactylogyrids* spp. (4.95%) were the major external parasites identified in this study. Similarly, Tadesse [17] also reported higher prevalence of *Trichodina* spp. in cultured system in Yemlo and Wonji ponds with a prevalence of 56.67% and 46.70%, respectively. Correspondingly 34.6% of prevalence was recorded for *Trichodina* in Uganda [18]. The higher prevalence of *trichodina* spp. could be resulted from the territorial behavior of *O. niloticus*. *Oreochromis niloticus* forms and defends territories along the shores. This territorial behavior increases the proximity to and maintains continuous exposure to free swimming stages of protozoans, crustaceans and digenetic trematodes metacercariae [19].

The overall prevalence of fish internal parasite in the present study was 20.31%. In the present study the most common internal parasite genera encountered were *Euclinostomum spp.* (7.81%) followed by Plerocercoid (2.34%) and *Acanthocephalans* spp. (2.08%) and *Contracaecum* spp (2.08%). This result is not in line with the previous study, Amare *et al.* [9] encountered *Contracaecum* spp. (42.6%) most frequently followed by *Clinostomum* spp. (38.6%) and *Eustrongylide* spp. (2.7%) in Lake Lugo. This result suggest that the distribution of parasites can be varied from one habitat to the other due to host parasite relationship; abiotic factors like dissolved oxygen, temperature and pH.

In this study the prevalence of *contracaecum* spp. (2.08%) was much lower than the finding of Moa and Anwar [20] who recorded 39.3% at northern Lake Tana and 39.67% at Lake Awassa [21]. This variation could be resulted from the difference in number of piscivorous birds in the study areas.

The current study showed that the difference in prevalence of parasites between sexes was statistically significant with higher prevalence was found in males (79.71%) than females (75.14%). This finding is in agreement with the study conducted by Akinsanya *et al*. [22] and Allumma and Idowu [23], found that male specimens presented a higher rate of internal parasite infestation. However, findings of the present study disagree with the findings of Imam and Dewu [24], Bichi and Ibrahim [25] and Mhaisen *et al.* [26] that stated female fishes were generally more liable than males to infestations with parasites. The variation might be due to the difference of their physiological condition of the females especially gravid ones which could have had reduced resistance to infection by the parasites [9], but, in this study the difference might be due to the specimens taken, this physiological condition doesn't exist.

The prevalence of parasites was also evaluated based on different length categories. This study revealed a higher parasite infestation in fish with longest (> 20 cm) size classes. This result is in agreement with Olurin and Samorin [27] and Ray [28] who observed that the larger the fish, the greater the susceptibility to parasite infection, as adult fish consumes a great variety of foods and exhibit a great variety of feeding styles, hence the correlation of prevalence of parasitic infections with fish length which in turn corresponds to fish age [29], but contradicts with Akinsanya *et al.* [22] that stated the low level of immunity in the smaller sized fish could explain the high prevalence of helminthiosis.

In this study the higher parasitic infestation was found in fish with medium weight groups (50g-100g) (81.00%) followed by fish with body weight of <50g (77.15%) and fish with body weight of >100g (74.71%). The current finding was higher than the study conducted by Emaminew *et al*. [39] who found the prevalence of 40.46 %, 25.50% and 13.33% in small, medium and large body weight fishes, respectively.

In the current study the prevalence of parasites was also evaluated based on collection month and it was found that the higher parasitic infestation was recorded from January-February (93.91%). This result is in agreement with the report of Ajala and Fawole [31] which says more fishes are affected by parasite in dry season.

In this study multiple infestations were the predominant cases. Similarly, Amare *et al.* [9] also found multiple infestations of parasites at Lake Lugo. Multiple infestations were common due to the fact that the environment supports several parasites species thereby exposing the host to simultaneous infection with many of them. The presence of one parasite and its activity within the host weaken the resistance which makes concurrent infection feasible [23].

5. Conclusion and recommendations

In the current study, the overall prevalence of parasite was found to be 77.60%. The prevalence of external parasite (32.29%) was higher than internal parasites (20.31%). The study recovered nine genera of parasites *Tricodina* spp. and *Dactylogyrids* spp. being the popular external parasites identified. Similarly, *Euclinostomum* spp. was the major internal parasite identified in the study area. Sex, weight, total length and collection month the fish were found important determinant factors for the parasite distribution in fingerling fish. And hence emphasis should be given to basic fish culture, pond management and related issues to decrease risk of parasite infestation. Further studies should be conducted toward the life cycle and source of the identified parasites in order to prevent and control these parasites.

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