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First Report of Infection with Lernea (Linnaeus, 1758) in Rainbow Trout Cage Culture in Iran

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Abstract
Aqueous Lazanea parasite was one of the most devastating parasitic infections in freshwater fishes that creates Lernazis disease. This study for the first time examined the contamination rate to this parasite in farmed rainbow trout in the cage system in aqueous sources of northwestern of Iran from epidemiological perspective. Totally, among 240 examined samples (average weight 175 g), 33 numbers i.e. 13.8% of the fishes were infected with the parasite. Also, the biological characteristics of the Lazanea parasite such as season, water temperature, water resources of farms and health and managerial issues were studied to specify causes of pollution in the studied cages. In various seasons' comparison, the most pollution was seen in summer with 40%, while in winter, no infestation to parasite was found. This represents a direct impact of the water temperature and season in the life cycle of the parasite. This is the first report of the Lazanea parasite on rainbow trout in farmed cage system in Iran.
Keywords: parasite, Lazanea, rainbow trout, Iran

Introduction

Nowadays, with the advancement of technology and modern methods of fishing, countless changes are obtained in aquatic products fishing and global statistics indicate that breeding and production of aquatics in inland water has taken considerable growing process (Azadikhah, 2008). The importance and value of aquaculture and fish farming is increasing day by day in our country and in the meantime, West Azerbaijan province is one of the 5 top provinces in terms of proliferation and cold-water fish farming and in terms of fishing has the first place among non-coastal provinces (Rezavandi, Peyravi, et al, 2009). Generally, this province was the water filled province and with high rainfall and abundant rivers. According to this issue and the importance of the role of fish in terms of supplying a part of protein needs, the importance of investment and development of fisheries in this province and creating new methods of breeding including fish breeding in cage are determined. Fish breeding in cage is a method that was common in dams' reservoirs, Gulfs, lakes, estuaries, swamps, lakes, seas, and rivers which including breeding of one or more species of fishes in floating and constant net cages. On the other hand, with the development of the fisheries sector, paying enough attention to risks that can threaten aquatic animals, especially fishes either in the natural environment or in the breeding environment is necessary. One of these risks refers to parasites and diseases caused by them (Jalali, Barzegar. & Nezamabadi. 2008). In general, parasites are proposed as a large group of invertebrate from the perspective of Zoology and health. Especially, with the development of proliferative activities and fish farming and continuous exploitation of water resources, it is necessary to emphasis on health care and fish diseases to protect them and the importance of this issue can be realized by observing annual irreparable economic damage to water resources in this way on reproduction and breeding activities (especially fishes). Parasites lead to the

growth reduction, mortality, and delay in puberty or infertility and often provide ground for bacterial, viral and fungal diseases. Also, in some cases, severe fish mortality has been caused by parasites. Following the increase in the fish population and diversity of the parasite and prevalence of parasites with no host specificity or in some conditions with that specificity, severe injuries are occurred in introduced or native fishes. Now, different sex species of lernea are the most dangerous parasites of freshwater farmed fishes of the worlds and Iran and their importance increased with the intensive expansion of fish farming. In this regard, it can be pointed to the report of Jazebi Zadeh (1983) on severe pollution of introduced fish species to the lernea parasite in Zarivar Lake (Jalali, 1998). The importance of lernea parasite and its role in creating disease and casualties and economic losses to fish farmers, proposing questions regarding the existence or lack of disease and the mortality rate of this disease as well as the absence of disease study in province especially with applying cage farming industry are important cases examined in this study.

Materials and methods

In this study the fish samples prepared from cage breeding farms of Hasanlu dam and Salmâs which were totally about 240 samples of live fishes were transferred to the Department of Parasitology by using fiberglass tanks connected to the oxygen and in stress-free conditions. After transmission of sample to the laboratory and performing biometry, samples were examined in terms of parasitic contamination. Because most crustacean fish parasites firmly attached to the outer surface of the fish, their separation was performed by using a forceps gently. When the head of parasite (lernea) was mired deeply in the body, at first, it is necessary to gap gently the surrounding tissue of the penetration with a scalpel to isolate the parasite completely. Then, sample was cleaned in solution of sodium chloride through an anatomy needle to eliminate the tissues and the organic materials sticking to parasites. To maintain the parasite until the final diagnosis and coloring, it was kept in 70% ethanol or 5-10% formalin. Solutions such as sodium hypochlorite was used to make the parasite transparent and the sample fixed in formalin for 1 to 3 minutes was put in 5.25% sodium hypochlorite and then was washed with distilled water. Fixed sample was first put in water then for 12 to 24 hours was placed in 10% KOH and after this period, and after clearing parasites, sample was washed with water and in the next stage it was put in 70% ethanol acid to remove its excess water, and it became completely clear. Now, sample is prepared for coloring and mounted on glass slides and materials such as polyvinyl laktol phenol or CMC-S were used for this purpose (Rezavand, Peyravi, et al. 2009).

Results

This study which was carried out for one year and during the four seasons since spring of 2013, was studied the trout contaminated to the lernea parasite in cage breeding farms system of West Azarbaijan province. This study was conducted according to the statistic of the cage breeding farms system of West Azarbaijan province which is obtained from the Fisheries Bureau of province and accordingly the two water sources of, Hasanlu dam of Naghadeh and darik dam of Salmâs were studied. Sampling was done in four seasons and in each season 60 samples (totally 240 samples) were randomly studied in different weight groups. Generally, among 240 samples, 33 fishes namely 13.8% of the fishes were contaminated to the lernea parasite. This parasite was isolated from the skin and gills of rainbow trout samples. Table 1 shows the results of the investigation of the contamination rate to the lernea parasite in fish breeding farms of West Azarbaijan province in different seasons. The high contamination rate in summer and lack of contamination in winter season is another result of this study. The contamination mean intensity in all fishes and all contaminated fishes in each season is given in Table 1. Maximum number of isolated parasites from

contaminated samples was 6 parasites which were done in summer that is also evident in the table. The results of this study in relation to the contamination severity comparison of the total fishes in different seasons of 2013 were obtained by using Kruskal Wallis test that severe significant difference was seen (P =0.000), also there was significant difference between spring and summer in contamination severity comparison of total fishes among different seasons that was done by mann-whitny test (P<0.05). However, there was no significant difference between spring and autumn (P>0.05), and significant difference was observed between spring and winter (P<0.05), high significant difference existed between summer and autumn (P =0.000), also, sever significant was seen between summer and winter (P =0.000) and there was no significant difference between autumn and winter (P>0.05). A contamination percentage comparison of total fishes by using chi-square test in various seasons revealed severe significant difference (P =0.000). Compared with the contamination percentage of total fishes among different seasons with each other, the contamination percentage in the spring with summer season showed significant differences (P< 0.05), but spring with autumn season showed significant difference (P>0.05) and spring with winter revealed the significant difference (P<0.05). Significant differences were found in comparison of summer with the autumn and the winter (P= 0.000), also, with comparison of the autumn with winter difference was not significant (P>0.05). From the total of isolated parasite respectively 13/3% in spring, 36/7% in summer and 5% in autumn and no involvement rate was observed in winter. The highest contamination rate in summer and the lowest rate were observed in winter. Most contamination range in summer from 1 to 6 parasites and the lowest contamination range was zero in winter. The contamination mean intensity in all fishes 0.062 ± 0.3 and contamination mean intensity in contaminated fishes has been 0.278 ± 2.21.

Table 1. Results of investigation of the contamination rate to lernea parasite in fish breeding farms of West Azarbaijan Province

Season	Contamination mean intensity in contaminated fishes ±SE	Contamination mean intensity ±SE	Range	Contamination percentage
Spring	1/88±./398	./25±./97	1-4	13/3
Summer	2/45±./382	./9±./207	1-6	36/7
Autom	1/33±./333	./07±./04	1-2	5
Winter	0	0	0	0
Total	2/21±./278	./3±./062	1-6	13/8

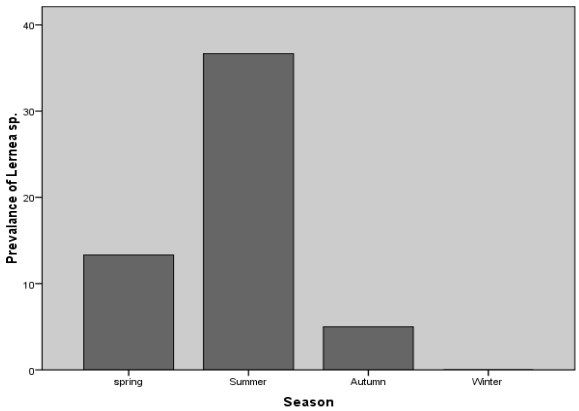

Figure 1. Contamination percentage comparison of the rainbow trout in various seasons in 2013

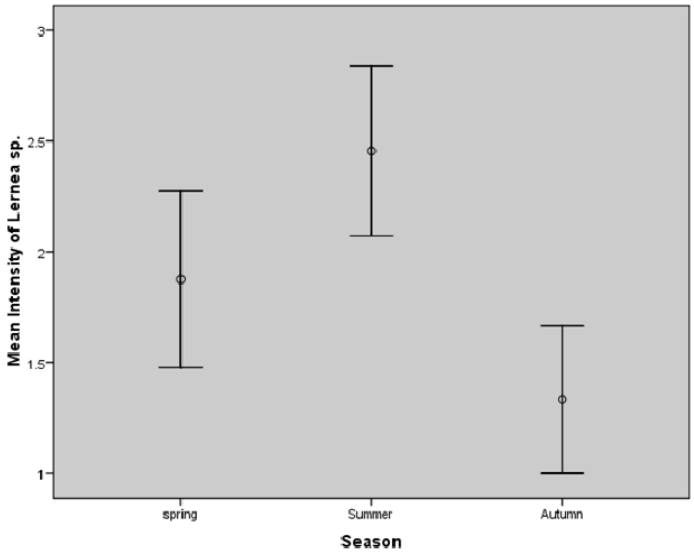

Figure 2. The comparison of the contamination mean intensity (±2 SE) of rainbow trout to lernea parasite in different seasons in 2013

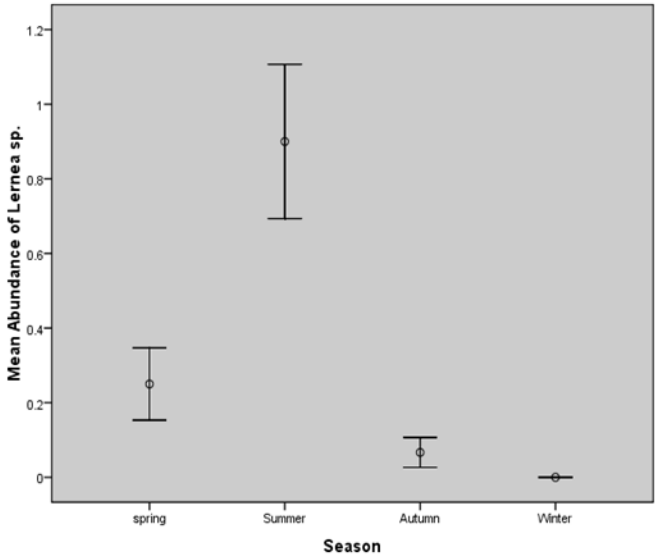

Figure 3. The comparison of the contamination mean intensity (2 SE ±) of rainbow trout in different seasons in 2013

Figure 4. Example of contaminated fish of dam of Salmâs

Figure 5. Cage farming system of Hasanlu dam

Discussion

Numerous instances of contamination of Iranian fishes to lernea parasite have been reported but there is no report regarding the contamination to this parasite in farmed trout in Iran. This study was conducted to investigate the lernea parasite in rainbow trout breeding farms in the cage system in the northwest of Iran and conducting this project is important to obtain information about the parasite fauna of native fishes in terms of systematic and ecological parasites. In this study, the contamination to lernea parasite was reported in breeding farms of the cage systems of West Azarbaijan province for the first time. The conducted survey on Lerneazis in Iranian farmed fishes by Jalali (1987) confirms this theory that temperature is the most important factor that affects life cycle and pathogenicity of the parasite (Jalali, 1997). So that at temperatures lower than 15 ° C do not metamorphosis and complete life cycle of the parasite at14 to 15 degrees needs 100 days (Barzegar and Jalali, 2009). In this study also the contamination to the lernea parasite in the cage breeding farm system was observed in three spring, summer and fall seasons that the highest amount and contamination percentage was in the summer, but in winter the parasite was not observed. Generally, it can be said that freshwater fishes are encountered to the parasite throughout the year, but severe infestation of parasite along with losses in farmed fishes are seen during the warmer months of the year (Coad, 1998). Because life cycle of the parasite is ecologically completed at high temperatures. So, spread of the disease occurs in temperate regions only during the summer. But in warm temperate regions, the possibility of epidemic occurrence exists dangerously throughout the year. Due to the weather conditions of West Azarbaijan province, the occurrence of illness was

observed in spring, summer and autumn seasons that its intensity was higher in the summer. The reason for high incidence of disease in this season refers to the existence of the suitable temperature for rapid growth and proliferation of the parasite. Especially, from late spring to late fall when the water temperature of lakes of studied dams is generally above 30 ° C which gives severe birth possibility to the parasite, so that in every 16.5 to 20 days once the parasite life cycle is completed (Behnke, 2002). Generally, it can be said that the water temperature was influenced by air temperature in large amounts and in areas that four seasons have clear approximate boundaries, variation range of temperature is to the extent that leads to the prevalence of some parasites in the summer and eliminate them in the winter. About lernea parasite which has indirect life cycle, the spread of its contamination depends on the spread of intermediate hosts and due to the reduction of intermediate hosts in cold seasons; the decrease can be seen in the contamination severity of the host fishes to this parasite.

Another important point is that the fish breeding in cage potentially leads to two-way damage. Thus, breeding fishes influence the environment and native fishes and on the other hand, native fishes can also threaten the cage breeding systems seriously by transferring different contaminations to farmed fishes as serious hazards. Crossover transmission of parasites among farmed and native fishes is one of the most important cases. Such environmental damage should be minimized to maintain the favorable conditions for breeding reserves in cages. On one hand, increasing the number of fishes per unit and the creation of high density and on the other hands, the transmission of baby fish that are contaminated at early stages are causing the epidemic occurrence by having parasite under their skin. According to the results of this study, the contaminations to lernea parasite in Darik dam of Salmâs is far higher than the dam of Hasanlu that one of the main issues in this regard refers to the relatively high density of fishes in the cage farms of Darik.

Another important point was the mesh size of nets used in cages of Gerdik dam that the size of these meshes were much larger and this causes that native fishes can easily enter into the cage of trout and native fish, however, these native fishes were small, they can easily transfer any parasitic infections such as lernea parasite to farmed trout of cages.

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