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Influence of Garlic (*Allium sativum*) and Mother Worth (*Matricaria chamomilla)* Extract effects on *Ichthyophtirius multifilus* Parasite Treatment in Sail Fin Molly *(Poecilia latipinna)* Ornamental Fish

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Abstract: The present study was carried out to evaluate the effect of *Allium sativum* and *Matricaria chamomilla* extract on *Ichthyophtirius multifilus* parasite. Group A bathed with 0.1 g/L garlic and group B bathed with 0.4 g/L mother worth extract. After microscopic examination in both treatments *Ichthyophtirius multifilus* parasites were treated after 5 days. However Ich parasites were observed on control group. The lowest mortality percentage was observed in group A. The highest mortality was related to group B which was bathed with mother worth extract. Both A and B groups fish treated after 5 days perfectly. Herbal extract can be noted as safe treatment instead of chemical medicine.

Key word: *M. chamomilla* · *A. sativum* · Extract · *P. latipinna* and Garlic

INTRODUCTION

Fish are an indispensable source of proteins for humans, not withstanding their importance as an object of sport fishery and pets in the case of ornamental fish. Besides direct losses caused by mortality, parasites may have considerable impact on growth and behavior of fish, their resistance to other stressing factors, susceptibility to predation, etc.; their presence may also reduce marketability of fish [1-6]. The ciliate protozoan *Ichthyophthirius multifilus* Fouquet, 1876, "Ich" or white spot disease is recognized to be one of the most pathogenic diseases of wild and cultured freshwater fish. This disease is a major problem to aquarists and commercial fish producer's worldwide. Ichthyophtirius is an important disease of ornamental and food fish. While many protozoans reproduce by simple division, a single Ich organism can multiply into hundreds of new parasites. This organism is an obligate parasite which means that it cannot survive unless live fish are present. It is capable of causing massive mortality within a short time. Outbreak of Ich is an emergency situation which requires immediate treatment if left untreated; this disease may result in 100% mortality. Adult organisms are oval to round and measure 0.5 to 1.0 mm in size. The adult is uniformly ciliated and contains a horseshoe-shaped nucleus which can be seen in older individuals. Concentration of the parasite in wild populations is low and massive mortalities from the infection are rare.

Herbs ability to inhibit activity of bacteria having potential interest as fish pathogens has been documented [7-11]. However, there is limited knowledge about anti-parasitical activity of herbs from Iran as a natural treatment for fish parasites, use of herbs in aquaculture industry was improved in recent 10 years, because of chemical pollution that cause by use of commercial medicines. Mother worth and garlic are members of herbs that in present study were used for Ich disease treatment. The aim of this study is to evaluate the effect of *Allium sativum* and *Matricaria chamomilla* extraction on *Ichthyophthirius multifilus*.

MATERIALS AND METHODS

Experimental Fish: A total number of 54 parasites free Sail fin Molly fish weighting 2.92 ± 21 g were obtained from private ornamental fish farm (Tehran-Iran). They were kept under usual situation on 26.14°C [12] with proper aeration. This species (*Poecilia latipinna*) is member of Poeciliidae family originated to Southern Virginia, Carolina, Florida and Texas [12]. Fish fed according to 10% of body weight

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Collection of *Ichthyophthirius multifilus* Tomonts: Fish with natural heavy parasitic infection (5 days post-infection) were anesthetized with extraction of *Eugenia caryophlyllata* then washed with water and the skin was scraped to dislodge the tomonts. The isolated tomonts were concentrated with 70µm mesh. The collected tomonts with 70µm mesh were transferred into 1 liter glass containing 1000 ml water as a modification of the method of Noe and Dickerson [13].

Experimental Tank and Protocol: After parasite exposure, fish were transferred to treatment aquarium without theronts. The treatments with 3 replicates were prepared. There were 9 tanks with 15 liters capacity with good aeration. The water temperature was 26°C. Fish got the natural day light and the environment was kept calm to reduce any stress. Thirty percent of water was exchanged daily. Treatments were labeled with A, B and C. Treatment (A) bathed with garlic (*Allium sativum*) with density of 0.1 g/L, treatment (B) bathed with *Matricaria chamomilla* with density of 0.4 g/L [14] and treatment C kept as control (no herbal treatment). During the study fish disease progress monitored, mortality rate and density of parasitic infection were recorded [15, 16].

Statistical Analysis: Treatments were compared by One-way Analysis of Variance (ANOVA) and Nonparametric tests (Chi-square). In completely randomize design; comparisons of means were made using Duncan's multiple range tests using SPSS (Version 9.0). The significant level was set at $P < 0.05$.

RESULTS

Growth: The performance of weight gain is present in figure 1. There was no significant difference in weight between treatments (P>0.05), Showing that use of herbs didn't have any negative effect on growth. During study different appetite was observed. It was controlled with feed weight measuring, which was fed every day (Figure 2).

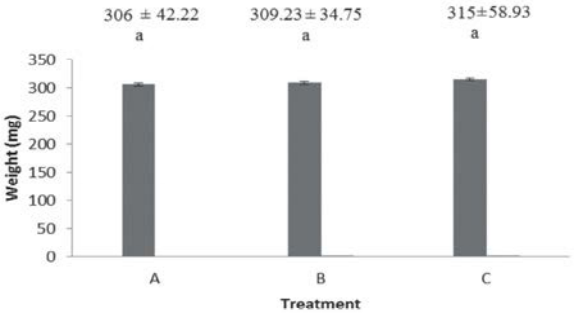

Fig. 1: Growth indices of Sail fin Molly in different herbal treatments

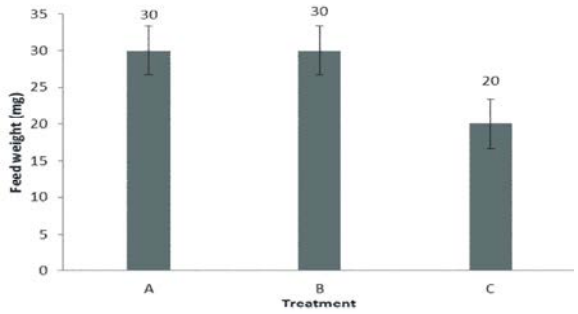

Fig. 2: Feed weight of different treatments

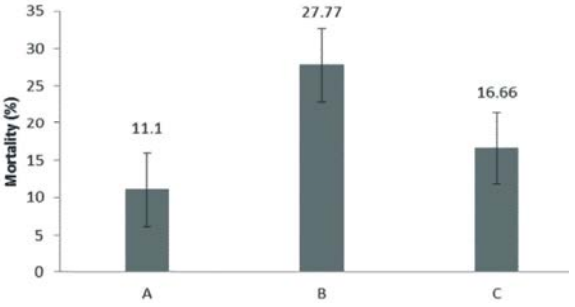

Fig. 3: The performance of mortality percentage

Parasite Treatment: Use of garlic and mother worth extract treated Ich parasite after 5 days. But "Ich" parasites were present in control group. In our attempt, mean temperature was 26.14°C (the optimum temperature for Ich parasite). During the study fish were monitored and treatment effect was recorded which is presented in Table 1.

Table 1: The performance of *Ichthyophthirius multifilus* presence in different times

	Experiment period									
Treatment	1st day	2nd day	3rd day	4th day	5th day	6th day	7th day	8th day	9th day	10th day
A	+	+	+	+	+	-	-	-	-	-
B	+	+	+	+	+	-	-	-	-	-
C	+	+	+	+	+	+	+	+	+	+

+ show the presence of parasites,-show the absence of parasites,

A: treated with garlic, B: treated with mother worth, C: control group

Fish Survival: Fish mortality percentage is presented in figure 3. Group B that treated with mother worth extract (0.4 g/L) showed the highest mortality percentage ($P<0.05$). The lowest mortality percentage was related to group A.

DISCUSSION

Growth: Fish growth were affected by many factors same as feeding period, kind of diet and it compositions and environmental factors. In this study that lasted for 15 days we first challenge sail fin Molly fish in the first 5 days with Ich theronts which was collected from sick fish. This period of study doesn't have effective influence on growth parameters. Herbal extracts can use as prebiotic. Khalil *et al.* [17] reported that garlic contains allicin, which improved the performance of the intestinal flora, by improving digestion and enhancing the utilization of energy, leading to improve growth. Several herbs have been used for growth-promoting activities in aquatic animals [18-20]. Other studies have shown that garlic did not affect growth performance of broilers [21] or growing lambs [22]. Aly *et al.* [23] reported that 2 months of feeding with garlic didn't show significant effect on growth parameters of *Oreochromis niloticus* but significant growth was observed after 8 months. The same results were reported by Metwally [24], on *Oreochromis niloticus* after 2 months feeding with garlic.

Parasite Treatment: The development of disinfection agents to treat parasites infestation is one of the most fascinating stories in the history of aquatic animal health. Garlic can help control of pathogens, especially bacteria and fungi [25-27]. It has long been considered that garlic (*Allium sativum*) has several beneficial effects for human and animals, exhibiting antimicrobial, antioxidant and antihypertensive properties [28, 29]. Many studies on garlic product have been published; however, there are very few reports on its effects on ectoparasite infestation. The other herb that used in this study was mother worth. Kazemipour *et al*. [14], reported the effect of mother worth extracts due to treat ulcers in common carp (*Cyprinus carpio*). The current results showed the stimulatory effect of garlic on the fish appetite. There are some studies about antibacterial activity of garlic but all of them were not successful. Colorni *et al*. [30] reported that garlic did not have any significant effect of *Mycobacterium marinum* infection in European sea bass culture. There are other studies that showed positive effect of garlic on bacterial and fungal diseases [31, 32]. Aqueous and methanolic extracts of *A. sativum* showed a measurable inhibition zone against *Phaeoisariopsis griseola* [33]. William [32] reported that sprays made from aqueous garlic extracts have antibiotic and antifungal properties and will suppress a number of plant diseases, including powdery mildew on cucumbers and to some extent black spot on roses. Similar results were reported by Slusarenko *et al.* [31] who tested the effectiveness of garlic juice against a range of plant pathogenic bacteria, fungi and Oomycetes *in vitro*.

Fish Survival: Reduced mortalities, following pathogenic challenge in the presence of low herbal dose have been reported by Kim *et al*. [34] and Jain and Wu [35]. In our study the survival rate was significantly greater in garlic treatment compared with control at the end of the experiment. However, survival rate was significantly higher in group A than the other groups. Using a combination of five herbs developed an *Artemia* enriched herbal diet for *Penaeus monodon*, which significantly increased survival rate during stress conditions [19].

CONCLUSION

Allium sativum and *Matricaria chamomilla* which used in this study treated the *Ichthyophthirius multifilus* parasite. Mortality rate was compared with each other and the results showed, mother worth influence on fish survival rate. Herbs are the most accessible medicine which can be used in aquaculture industry to reduce chemical material. According to this study garlic has effective influence on parasites in addition of bacterial and fungal disease. Garlic reduce mortality rate and improved sail fin Molly health.

REFERENCES

1. Crowden, A.E. and D.M. Boom, 1980. Effects of the eyefluke, *Diplostomum spathaceum*, on the behavior of dace (*Leuciscus leuciscus*). Anim. Behav., 28: 287-294.
2. Brassard, P., M.E. Rau and M.A. Curtis, 1982. Parasite-induced susceptibility to predation in diplostomiasis. Parasito, 85: 495-501.
3. Lom, J. and I. Dyková, 1992. Protozoan parasites of fishes. Developments in Aquac. and Fish Sci., 26: 315.
4. Williams, H.H. and A. Jones, 1994. Parasitic worms of fish. Taylor and Francis, London, pp: 593.

5. Kumaraguru, A.K., F.W.H. Beamish and P.T.K. Woo 1995. Impact of a pathogenic haemoflagellate, *Cryptobia salmositica* on the metabolism and swimming performance of rainbow trout, *Oncorhynchus mykiss* (Walbaum). J. Fish Dis., 18: 297-305.
6. Woo, P.T.K., 1995. Fish diseases and disorders. Vol. I. Protozoan and metazoan infections. CAB International, Oxon, UK, pp: 808.
7. Direkbusarakom, S., 2004. Application of medicinal herbs to aquaculture in Asia. Walailak J. Sci. and Technol., (1): 7-14.
8. Muniruzzaman, M. and M.B.R. Chowdhury, 2004. Sensitivity of fish pathogenic bacteria to various medicinal herbs. Bangladesh J. Vet. Med., 2(1): 75-82.
9. Abutbul, S., A. Golan-Goldhirsh, O. Barazani, R. Ofir and D. Zilberg, 2005. Screening of desert plants for use against bacterial pathogens in fish. Isr. J. Aquac. Bamid., 57(2): 71-80.
10. Borisutpeth, P., P. Kanbutra, S. Weerakhun, K. Sarachoo and S. Porntrakulpipat, 2005. Antibacterial activity of Thai medicinal plant extracts on *Aeromonas hydrophila* and *Streptococcus agalactiaee* isolated from diseased tilapia. In: Abstracts of 31st Congress on Science and Technology of Thailand at Suranaree University of Technology, Thailand, 18-20 October 2005.
11. Dubber, D. and T. Harder, 2008. Extracts of *Ceramium rubrum*, *Mastocarpus stellatus* and *Laminaria digitata* inhibit growth of marine and fish pathogenic bacteria at ecologically realistic concentrations. Aquac., 274: 196-200.
12. Sandford, G., 2007. All illustrated encyclopedia of Aquarium fish. Quantom, London, pp: 251.
13. Noe, J.G. and H. Dickerson, 1995. Sustained growth of *Ichthyophthirius multifiliis* at low temperature in the laboratory. J. Parasitol., 81(6): 1022-1024.
14. Kazemipour, Y., M. Rezaei and Y. Keyvani, 2005. Qualitative comparison of effect of garlic and mallow and motherworth extracts in healing of superfacial wounds in the common carp (*Cyprinus carpio*). Pajohesh and Sazandegi bulletin, 66: 93-97 (In Farsi)
15. Hai, X., P.H. Klesius and C.A. Shoemaker, 2008. Protective immunity of Nile tilapia against *Ichthyophthirius multifiliis* post-immunization with live theronts and sonicated trophonts. Fish and Shel. Immunol. In press.
16. Osman, H.A., F. El-Bana, A.M. Layla, A. Noor El Deen and O.K. AbdEl-Hady, 2009. Investigations on White Spots Disease (Ichthyophthriasis) in Catfish (*Clarias gariepinus*) with Special Reference to the Immune Response, Global Veterinaria, 3(2): 113-119.
17. Khalil, R.H., B.M. Nadia and M.K. Soliman, 2001. Effects of Biogen and Levamisol HCl on the immune response of cultured *Oreochromis niloticus* to *Aeromonas hydrophila* vaccine. Beni-Suef Vet. Med. J. Egypt, (2): 381-392.
18. Jayaprakas, V. and J. Eupharsia, 1996. Growth performance of *Labeo rohita* (Ham.) Livol (IHF-1000), an herbal product. Proc. Indian Natl. Sci. Acad, 63: 1-10.
19. Citarasu, T., M.M. Babu, R. Raja Jeya Sekar and M.P. Marian, 2002. Developing artemia enriched herbal diet for producing quality larvae in *Penaeus monodon* Fabricius. Asian Fish Sci., 15: 21-32.
20. Sivaram, V., M.M. Babu, G. Immanuel, S. Murugadass, T. Citarasu and M.P. Marian, 2004. Growth and immune response of juvenile greasy groupers (Epinephelus tauvina) fed with herbal antibacterial active principle supplemented diets against Vibrio harveyi infections. Aquac, 237: 9-20.
21. Freitas, R., J.B. Fonseca, R.T. Soares, H.S. Rostagn and P.R. Soares, 2001. Utilization of garlic (*Allium sativum* L.) as growth promoter of broilers. Rev. Bras. Zootec, 30: 761-765.
22. Bampidis, V.A., V. Christodoulou, E. Christaki, P. Florou-Paneri and A.B. Spais, 2005. Effect of dietary garlic bulb and garlic husk supplementation on performance and carcass characteristics of growing lambs. Anim. Feed Sci. Techno., 121: 273-283.
23. Metwally, M.A.A., 2009. Effects of Garlic (*Allium sativum*) on Some Antioxidant Activities in Tilapia Nilotica (*Oreochromis niloticus*), World J. Fish and Mari. Sci., 1(1): 56-64.
24. Aly, S.M., N.M. Abdel-Atti and F.M. Mohamed, 2008. Effect of garlic on the survival, growth, resistance and quality of *Orechromis niloticus*, 8th International Symposium on Tilapia in Aquaculture, pp: 277-296.
25. Corzo-Martinez, M., N. Corzo and M. Villamiel, 2007. Biological properties of onions and garlic, Trends in Food Sci. and Techno., 18: 609-625.
26. Adetumbi, M., G.T. Javor and B.H. Lau, 1986. *Allium sativum* (garlic) inhibits lipid synthesis by *Candida albicans*. Antimicrobial Agents and Chemotherapy, 30: 499-501.
27. Ress, L.P., S.F. Minney, N.J. Plummer, J.H. Slatter and D.A. Skyrme, 1993. A quantitative assessment of the antimicrobial activity of garlic (*Allium sativum*). World J. Microbio. and Biotechno., 9: 303-307.

28. Konjufca, V.H., G.M. Pesti and R.I. Bakalli, 1997. Modulation of cholesterol levels in broiler meat by dietary garlic and copper. Poul. Sci., 76: 1264-1271.
29. Sivam, G.P., 2001. Recent advances on the nutritional effects associated with the use of garlic as supplement. Am. Soc. Nutr. Sci., pp: 1106-1108.
30. Colorni, A., R. Avtalion, W. Knibb, E. Berger, B. Colorni and B. Timan, 1998. Histopathology of sea bass *Dicentrarchus labrax* experimentally infected with *Mycobacterium marinum* and treated with streptomycin and garlic (*Allium sativum*) extract. Aquac., 190: 1-17.
31. Slusarenko, A.J., A. Patel and D. Portz, 2008. Control of plant diseases by natural products: Allicin from garlic as a case study. European J. Plant Pathol., 12: 3.
32. William, Q., 2008. Least toxic controls of plant diseases. Brooklyn Botanic garden. Natural Disease Control, 11: 225.
33. Charimbu, M.K., I.N. Wagara and D.O. Otaye, 2009. Antifungal activity of various crude plant extracts against *Phaeoisariopsis griseola* pathogenic on common bean, African Crop Science Conference Proceedings, pp: 683-685.
34. Kim, K.M., S.B. Chun, M.S. Koo, W.J. Choi, T.W. Kim, Y.G. Kwon, H.T. Chung, T.R. Billiar and Y.M. Kim, 2001. Differential regulation of NO availability from macrophages and endothelial cells by the garlic component S-allyl cysteine. Free Radic. Biol. Med., 30: 747-756.
35. Jain, J. and Z. Wu, 2003. Effect of traditional Chinese medicine on nonspecific immunity and disease resistance of large yellow croaker *Psedcosciaena crocea* (Richardson). Aquac., 218: 1-9.