Evaluation of the Antifungal Activity of Zataria multiflora, Geranium herbarium, and Eucalyptus camaldolensis Essential Oils on Saprolegnia parasitica–Infected Rainbow Trout (Oncorhynchus mykiss) Eggs

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Abstract
The purpose of the present study was to evaluate and assess the capability of Zataria multiflora, Geranium herbarium, and Eucalyptus camaldolensis essential oils in treating Saprolegnia parasitica–infected rainbow trout eggs. A total of 150 infected eggs were collected and plated on glucose-pepton agar at 24°C for 2 weeks. The antifungal assay of essential oils against S. parasitica was determined by a macrodilution broth technique. The eggs were treated with essential oils at concentrations of 1, 5, 10, 25, 50, and 100 ppm daily with three repetitions until the eyed eggs stage. Of 150 eggs examined, S. parasitica (54.3%), Saprolegnia spp. (45%), and Fusarium solani (0.7%) were isolated. The minimum inhibitory concentrations of Z. multiflora, E. camaldolensis, and G. herbarium essential oils against S. parasitica were 0.9, 2.3, and 4.8 ppm, respectively. Zataria multiflora and E. camaldolensis at concentrations of 25, 50, and 100 ppm, and G. herbarium at concentration of 100 ppm had significant differences in comparison with negative control (p<0.05). The results revealed that malachite green, followed by Z. multiflora, E. camaldolensis, and G. herbarium treated eggs had remained the most number of final eyed eggs after treatment. The highest final larvae rates belonged to malachite green, E. camaldolensis, Z. multiflora, and G. herbarium, respectively. The most hatching rates were recorded with malachite green (22%), and then Z. multiflora (11%), E. camaldolensis (7%), G. herbarium (3%), and negative control (1%). Zataria multiflora and E. camaldolensis were more effective than G. herbarium for the treatment of S. parasitica-infected rainbow trout eggs in aquaculture environment.

Introduction

Fish eggs are commonly contaminated with aquatic fungi of the genus Saprolegnia in early stages of embryonic development (Perez et al., 2003). During this process, fungi can invade nonviable eggs and grow over the healthy ones, causing an important decrease of efficiency rates (Bruno and Wood, 1999). The susceptibility of fish eggs to fungal infection is often dependent on water quality, water flow rates, handling, and high density of eggs (Moreira and Barata, 2005). Periodical removal of dead eggs is effective to control fungal growth. However, this practice requires a great human effort, and handling could damage healthy eggs. These disadvantages would be more evident under culture conditions when large amount and high density of eggs are involved, representing a significant economic and welfare problem (Pottinger and Day, 1999). A number of reports have been issued on the occurrence of zoosporic fungi on incubated eggs in the basin of the Caspian Sea, referring mainly to such sturgeons as great sturgeon (Huso huso), Russian sturgeon (Acipenser gueldenstaedti), stellate sturgeon (Acipenser stellatus), and Persian sturgeon (Acipenser persicus) (Dudko et al., 1989; Ghiassi et al., 2010).

Malachite green is commonly used to solve this problem and has been widely reported as an effective fungicidal agent (Li et al., 1996). However, the use of malachite green on fish is banned in some countries because of its teratogenic effects and the finding of a residue called leucomalachite with carcinogenic activity, which can persist in fish tissues for a long

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