EFFECT OF HYDROGEN PEROXIDE TREATMENT ON GOLDFISH (CARASSIUS AURATUS AURATUS, L.) EGGS

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Abstract
In this study, the effectiveness of hydrogen peroxide as a treatment for disinfecting goldfish (Carassius auratus auratus, L.) eggs were investigated, considering hatching success and embryo survival in various hydrogen peroxide treatment regimes ranging in concentration from 300 ppm at a water temperature of 26°C. We searched 5, 10, 20-min bath in hydrogen peroxide (H2O2) at this dose that hatching success and embryo survival.

Keywords: Aquaculture, Izmir Bay, Bacteria, Fishes

Introduction
Many researchers have investigated the effects of various disinfectants of intensive egg incubation of marine and freshwater fish for prevent to the growth of bacteria and reduce the transfer a disease with them [1,2,3]. However, while goldfish having an important place within the ornamental fish sector, few studies on the effect and mode of application are some type of disinfectant which used during breeding time. After hatched during goldfish larvae period are seen high mortality problems caused by bacteria, such as Flexibacter columnaris [4], Flavobacterium columnare [5]. Aeromonas hydrophila [6]. Hydrogen peroxide has been investigated as an alternative treatment to control bacterial or fungal infections of eggs of several additional fish species [3,7]. Although hydrogen peroxide has long been used for external treatment of fish [8,9] its use for treatment of fish eggs has only recently been examined. It is considered to be a therapeutant of Low Regulatory Priority (LRP) by the U.S. Food and Drug Administration’s (FDA’s) Center for Veterinary Medicine (CVM) to control fungal on fish and fish eggs (R. E. Geyer, CVM, personal communication). Although hydrogen peroxide is not an approved drug, the LRP status allows hatchery personnel to administer treatments on fish and fish eggs to control or prevent fungus at 500 ppm or less with little concern for regulatory action. At the same time, evaluating the efficacy of hydrogen peroxide to treat ectoparasites of Atlantic salmon (Salmo salar, sable) found a linear relationship between hydrogen peroxide efficacy and temperature [9].

In the present study, goldfish eggs incubated at 26 °C were treated with increasing bath time in same concentrations of hydrogen peroxide to determine what time yielded the greatest hatching success.

Materials & Methods
Hydrogen peroxide (30% active ingredient) was obtained from Sigma-Aldrich and its test solution were prepared from a stock solution of 30% hydrogen peroxide. Egg-full kakabans taken from the tanks with 10 brood fish that mated randomly. The eggs were disinfected in 5 min, 10 min and 20 min with 300 ppm dose of hydrogen peroxide. Any treatment were not done in control group. Three replicates were conducted for each treatment; For each replicate, as an average 1g eggs were removed from the kakabans, and transferred to 500 mL of the test disinfectant solution in a beaker. After trial of disinfection, the solution was poured off and the eggs were rinsed three times with sterile water (about 103 cfu/ml). The bacteriological examination were done after disinfection treatment. Trypticase soy agar (TSA) were used to test for bacterial growth. The trypicase soy agar (TSA) medium was used to discern prevalence and abundance for all other culturable bacteria and fungi. For each treatment were inoculated per replicate to infect three agar plates. Plates were incubated at 25°C and examined 24,32 and 48 hours after inoculation. Counts were made of all colony-forming units (CFU) when possible, but many plates were classified as too numerous to count. Then the eggs were placed into 10 glass containers of 5 liter volume with the same water temperature with breeding tank (which was 26 °C). An airstone was put in the middle of the each experiment container. Every day, the dead eggs were counted in their containers during hatching time. This study were made in triplicate.

Results
After surface disinfection of goldfish eggs with hydrogen peroxide (30%, Sigma-Aldrich), bacterial growth in trypicase soy agar (TSA) culture media studied at 5, 10, and 20 minutes; after cultivation on the groups were counted respectively 4.94±(0.02)×103 cfu/ml, 1.55±(0.34)×103 cfu/ml, 9.89±(0.54)×102 cfu/ml and 5.51±(0.21)×102 cfu/ml were determined in the control. Accordingly, 5, 10, 20-minute disinfections and in the control group results were not found statistical important in the formation of colonies of those stored (p> 0.05). The survival rate of goldfish egg groups in the incubators which disinfected with 300 ppm hydrogen peroxide in 5, 10, 20 minute applications was found respectively 76 ± 17.2%, 79.11 ± 4.12% and 72.88 ± 6.99%. The survival rate was determined 84.66±3.33% in the control group.

Discussion
Treatment of goldfish eggs with hydrogen peroxide (H2O2) result that the 300 ppm is not effective on bacteriological control and survival rate when we compared with the control group. They have not statistically importance between the time treatments.

References