

**EFFICACY AND TOXICITY OF COMMON
DISINFECTANTS ON SEABASS (*Lates calcarifer*)
LARVAE**

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**MASTER OF SCIENCE
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Efficacy and toxicity of common disinfectants on seabass (*Lateolabrax niloticus*) larvae / Nik Haiha Nik Yusoff.



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EFFICACY AND TOXICITY OF COMMON DISINFECTANTS
ON SEABASS (*Lates calcarifer*) LARVAE

NIK HAIHA BT NIK YUSOFF

April 2006

Chairperson : Professor Noor Azhar Mohd Shauki, Ph.D.

Member :
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Faculty : Science and Technology

The efficacy of four chemicals as eggs disinfectant was evaluated on seabass eggs following a toxicological approach. The focus of this study is to establish disinfection protocol that will facilitate manipulation of the microflora of marine fish eggs to obtain healthy, bacteria-free larvae. The four chemicals used were formalin, hydrogen peroxide, iodine and sodium hypochlorite. The chemicals were administered to seabass eggs at different concentrations and were evaluated on the basis of their bactericidal and potential toxic effects on various fish eggs under hatchery conditions. The effects of exposure of seabass eggs to different disinfectants on 24-h larval survival were evaluated.

**Thesis Submitted in Fulfillment of the Requirements for the
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ON SEABASS (*Lates calcarifer*) LARVAE**

NIK HAIHA BT NIK YUSOFF

April 2006

Chairperson : Professor Noor Azhar Mohd Shazili, Ph.D.

**Member : Professor Faizah Mohd Shaharom, Ph.D.
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Faculty : Science and Technology

The efficacy of four chemicals as eggs disinfectant was evaluated on seabass eggs following a toxicological approach. The focus of this study is to establish disinfection protocol that will facilitate manipulation of the microflora of marine fish eggs to obtain healthy, bacteria-free larvae. The four chemicals used were formalin, hydrogen peroxide, iodine and sodium hypochlorite. The chemicals were administered to seabass eggs at different concentrations and were evaluated on the basis of their bactericidal and potential toxic effects on marine fish eggs under hatchery conditions. The effects of exposure of seabass eggs to different disinfectants on 24-h larval survival were evaluated.

The results suggested that hydrogen peroxide (H₂O₂) was the most promising candidate of the four chemicals tested. The effective and safe concentration of H₂O₂ that will reduce bacterial load but will not decrease 24-h survival rate for the disinfection of seabass egg is 1600 ppm for 10 min. Results of the study also showed that the best stage to disinfect seabass egg is at the embryonic stage. Total bacterial count of eggs disinfected with 1600 ppm H₂O₂ for 10 min (2.50cfu/egg) is significantly lower than the undisinfected eggs (3.21 x 10³ cfu/egg). H₂O₂ did not have any negative effect on survival, growth and occurrence of deformity. No adverse long-term effects on either larval survival or growth resulted from the egg disinfection treatment.

A study on the quantitative and qualitative analyses of aerobic bacterial flora associated with eggs of seabass, *Lates calcarifer*, along with important water quality parameters, were conducted over five cycles of spawning. Results showed that aerobic bacterial flora of seabass egg was largely influenced by the bacterial flora in the incubating water. Total viable count varied between 3.92 x 10⁵ and 1.13 x 10⁶ cfu/mL in water, 5.57 x 10⁴ and 8.88 x 10⁴ cfu/egg in eggs. Physico-chemical characteristics of water were within the optimum range. *Pseudomonas sp*, *Vibrio sp* and *Enterobacteriaceae* were frequently encountered genera in both the eggs and in water. *Pseudomonas sp* were found to be the dominant bacteria in both in eggs and incubating water. The susceptibility of eggs and early life stages of seabass to *Pseudomonas sp* and *Vibrio sp* was also investigated in infection experiments. Eggs of seabass were exposed to the bacterium prior to hatching. Eggs exposed to three different *Vibrio sp* and

Pseudomonas sp showed different mortality patterns, with low mortality at hatching, followed by a continuous fairly high mortality throughout the yolk sac stage. Mortality in the uninfected control group was low throughout the experiment. The results suggest that the hatchability of eggs and survival of larvae are influenced by bacterial infection and that seabass larvae exposed to *Pseudomonas sp* and *Vibrio sp* may become infected and that the bacterium may persist in larvae.

Thus, from the study it can be suggested that a routinely performed surface disinfection of seabass eggs with hydrogen peroxide at 1600 ppm for 10 to 15 minutes would be a good management practice not only to maintain high quality seeds but also to prevent the occurrence of disease and to increase the survival rate of seabass larvae.

Abstrak tesis yang dikemukakan kepada Senat Kolej Universiti Sains dan Teknologi Malaysia sebagai memenuhi keperluan Ijazah Master Sains.

**KEBERKESANAN DAN KETOKSIKAN KIMIA
PENYAHBASMIKUMAN YANG BIASA TERHADAP BENIH IKAN
SIAKAP (*Lates calcarifer*)**

NIK HAIHA BT NIK YUSOFF

April 2006

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Keberkesanan empat bahan kimia sebagai penyahbasmikuman telur dinilai ke atas telur menggunakan pendekatan toksikologi. Fokus kajian adalah untuk mewujudkan protokol penyahbasmikuman yang akan membantu memanipulasikan mikroflora pada telur bagi mendapatkan benih yang sihat dan bebas bakteria. Ke empat-empat bahan kimia yang digunakan adalah formalin, hidrogen peroksida, iodin dan natrium hipoklorit. Bahan-bahan kimia itu didedahkan kepada telur ikan pada pelbagai kepekatan dan penilaian dibuat berasaskan kesan bacterisidal dan ketoksikan terhadap telur ikan siakap di bawah persekitaran hatcheri. Kesan pendedahan pelbagai disinfektan pada telur siakap terhadap kadar kehidupan 24 jam benih dinilai.

Keputusan yang diperolehi mendapati bahawa hidrogen peroksida (H_2O_2) merupakan bahan kimia yang terbaik. Kepekatan H_2O_2 yang berkesan dan selamat untuk menyahbasmikuman telur ikan yang dapat mengurangkan kadar bakteria tanpa mengurangkan kadar kehidupan 24-jam benih adalah 1600 ppm selama 10 min. Keputusan kajian juga menunjukkan bahawa peringkat terbaik untuk menyahbasmikuman pada telur siakap adalah pada peringkat embrionik. Bilangan keseluruhan bakteria pada telur yang dirawat dengan 1600 ppm H_2O_2 selama 10 min (2.50cfu/telur) adalah signifikan lebih rendah dari telur yang tidak dirawat (3.21×10^3 cfu/telur). H_2O_2 tidak mempunyai kesan negatif terhadap kehidupan, tumbesaran dan kejadian kecacatan. Tiada kesan jangka panjang kepada kehidupan atau tumbesaran hasil dari telur yang dirawat.

Kajian analisis kuantitatif dan kualitatif bakteria aerobik yang berkaitan telur ikan siakap, *Lates calcarifer*, bersama-sama dengan parameter-paramter air yang penting dijalankan meliputi lima jangka kitaran peneluran. Keputusan menunjukkan bahawa bakteria aerobik telur siakap sebahagian besarnya dipengaruhi oleh bakteria di dalam air ternakan. Jumlah bilangan bakteria berubah di antara 3.92×10^5 dan 1.13×10^6 cfu/mL dalam air, 5.57×10^4 and 8.88×10^4 cfu/telur dalam telur. Ciri-ciri fisiokimia air berada di dalam julat yang optimum. *Pseudomonas sp*, *Vibrio sp* dan *Enterobacteriaceae* merupakan genera yang kerap ditemui di dalam telur dan air. Spesies *Pseudomonas* merupakan bakteria dominan di dalam telur dan air. Kepekaan telur dan peringkat awal kehidupan siakap kepada *Pseudomonas sp* dan *Vibrio sp* juga diselidiki di dalam kajian infeksi. Telur-telur siakap didedahkan kepada

bakteria-bakteria tersebut sebelum menetas. Telur-telur yang didedahkan kepada 3 jenis *Vibrio sp* dan *Pseudomonas sp* menunjukkan corak mortaliti yang berbeza, mortaliti yang rendah semasa menetas, diikuti dengan mortaliti yang agak tinggi pada peringkat kuning telur. Mortaliti kumpulan kawalan adalah rendah sepanjang tempoh kajian. Keputusan ini menunjukkan penetasan telur dan kehidupan benih ikan dipengaruhi oleh jangkitan bakteria dan benih ikan yang didedahkan kepada *Pseudomonas sp* dan *Vibrio sp* mungkin akan dijangkiti penyakit.

Sebagai kesimpulan, amalan rutin menyahbasmikuman pada telur siakap menggunakan hidrogen peroksida pada kepekatan 1600 ppm selama 10 ke 15 minit merupakan amalan pengurusan yang baik. Ia bukan saja untuk mengekalkan benih berkualiti tetapi juga untuk mencegah penyakit dan meningkatkan kadar kemandiran benih ikan siakap.