

TOXICITY EVALUATION OF SELECTED HEAVY METALS
ON TWO AQUATIC DIPTERIAN LARVAE
(*Chironomus plumosus* & *Culticoides furens*)

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2007

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*This thesis is dedicated to my mother who stood by me as I went
along in this journey and still continues to do so.*

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**Thesis Submitted in Fulfillment of the Requirements for the
Degree of Doctor of Philosophy in the Institute of Oceanography
Universiti Malaysia Terengganu**

June 2007

1100053953

Abstract of thesis presented to the Senate of Universiti Malaysia Terengganu
in fulfillment of requirements for the degree of Doctor of Philosophy

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A study was conducted to determine the suitability of using selected aquatic dipterian larvae for biomonitoring bioassays. The organisms included a member of the biting midge family that was identified as *Culicoides furens* and a member of the non-biting midge family, identified as *Chironomus plumosus*. Median lethal toxicity tests were conducted to observe the variation between metal sensitivities between the two larval forms and how variations in temperature could affect the experimental setup. Nine heavy metals were used in the study. It was observed that the 96 hour LC50 (in mg/L) for the different metals was found to be Zn-16.21(18.55±13.87); Cr-0.96(1.08±0.84); Ag-4.72(6.87±1.57); Ni-0.42(0.59±0.25); Hg-0.42(0.59±0.25); Pb-16.21(18.31±14.11); Cu-42.24 (45.18±39.30); Mn-4.22 (7.19±1.25); Cd-0.42(0.59±0.25). for the *Chironomus plumosus* and Zn-4.22(6.56±1.88; Cr-0.42(0.54±0.30); Ag-0.42(0.54±0.30); Ni-0.42(0.54±0.30); Hg-0.04(0.07±0.01); Pb-0.42(0.54±0.30); Cu-42.24 (45.18±39.30); Mn-4.22(6.56±1.88; Cd-0.42(0.54±0.30) in the case of the *Culicoides furens*. With temperature as a variable the LC50 values were observed to increase from 2.51 mg/L at 10°C to 4.22ppm at 30°C and to reduce slightly to 3.72 mg/L at 35°C as seen in the case of Zn. It was also observed

that at 40°C thermal toxicity and chemical toxicity overlapped as 100% mortality was observed in the controls. This trend was observed in all metals for both *C. plumosus* and *C. furens*. Studies were also conducted on the effects of long term exposure of heavy metals on the two dipterian larvae. The results indicated that long term exposure to all metals improved tolerance within the two organisms. The tolerance to all metals studied increased for both dipterian larva as a result of the long term exposure. The tolerance towards all metals increased at each generation until a maximum at generations 4 to 6. Studies also noted that the tolerance was lost when the organisms were subsequently bred in clean water for 2 generations.

Experiments using artificial substrate, silica gel powder, were conducted to observe the effect of larval tubes in reducing the toxicity of heavy metals to the organisms. As the *Culicoides furens* does not build nests, the availability of substrate is of no account, unlike in the case of the *Chironomus plumosus* where the larval tube insulates, to some extent, the organism from environmental changes. The LC50 values obtained in this experiment were found to be 10 times greater than the LC50 values obtained without the silica gel. This would indicate that the larval tubes protect the larvae to some extent. Studies using three sediment samples – clay loam, silty-clay loam and loam – were conducted to see if sediment type affects the toxicity of the heavy metals to the two dipterian larvae. The LC50 of the different heavy metals was observed to vary between the three sediment types. In the case of zinc, the LC50 value was observed to be 38.53 for silt loam, 42.22 for silty-clay loam and 70.99 for loam, in the case of *C. plumosus*. This trend was observed for all nine heavy metals tested as well as for both dipterian larvae.

Investigations conducted into the effects of dual metal mixtures on the two dipterian and to see the modifying behavior of the mixture on individual metal toxicity levels. It was observed that heavy metal toxicity varied according to the metals it was combined with. In the case of zinc (LC50= 9.50 mg/L), the toxicity of the metal was observed to have reduced when placed in combination with chromium (10.20 mg/L), silver (23.80 mg/L), nickel (12.40 mg/L), mercury (7.5 mg/L), lead (36.7 mg/L), copper (5 mg/L) manganese (46.5 mg/L) and cadmium (3.9 mg/L). Different metals had different reactions, some becoming more toxic and some becoming less toxic. Comparisons between test methods showed that the PPM+PPM method provided better data than the Toxic Unit method due to the ease at which LC50 values could be calculated for both metals within the mixture. Studies into heavy metal levels in the sediment, pore water and metal content in the organisms was measured to observe if a metal threshold level could be identified it was observed that metal values in the two dipterian larvae increased as the concentration of metal in the surrounding media increased, however, no threshold values could be identified.

In the final step of this investigation morphological deformity of the mentum, mandibles and antennae were observed to see if a correlation existed between metal type, concentration and frequency of deformities. From the observations of the mouth parts it was observed that there was a strong correlation between concentration and the frequency of mouth part deformities but an insignificant relationship between metal type and mouth part deformity.

This study has, for the first time in Malaysia, studied the effects of heavy metals on the *Chironomus plumosus* and *Culicoides furens* and has thus served as a pathfinder in this branch of ecotoxicology.