Effect of Sewage Sludge Metals on *Daphnia Similis* Acid Phosphatase

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Chemicals released into the environment are responsible for several adverse ecological effects, such as food chain bioaccumulation, toxicity to most of the freshwater organisms and corporal homeostasis damage. Modification of enzyme activities is one of the consequences of the toxicity to freshwater organisms. Acid phosphatase plays important roles in aquatic organisms metabolism and their environment, including decomposition of organic phosphates, hydrolysis of phospholipids and availability of inorganic phosphate from the extracellular medium. The aim of this work was to evaluate the *in vitro* effect of nine metals present as contaminants in sewage sludge, on the total acid phosphatase (TAP) activity extracted from the water flea *Daphnia similis*. One hundred mg of *D. similis* was homogenized in 0.1M acetate buffer pH 5.0 (1:4). After centrifugation, the clear extract was collected and utilized for protein and enzyme activity determinations. The TAP activity was determined at pH 5.0 using p-nitrophenylphosphate as substrate, at 37°C for 40 min (Km value of 0.29 mM). From the metals tested, only Al^{3+}, Se^{4+} and Mo^{6+} markedly inhibited (=50%) the TAP activity. A more detailed kinetic study of the inhibition by Al^{3+} was performed. An IC50 value was determined to be 1.0mM. The inhibition by Al^{3+} was of non-competitive type, with an inhibition constant (Ki) value of 0.9mM, determined by the Dixon method. Our *in vitro* results suggest that *D. similis* TAP does not contain cysteine residues in the active site and that the inhibition of this organism enzyme by metals is different when compared with other acid phosphatases.

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