

Response of *Lemna minor* L.(Duckweed) to the heavy metal copper

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Abstract

Present study aims to assess the tolerance and toxic effect of copper on common duckweed aquatic plant *Lemna minor*. Copper tolerance in *Lemna minor* was investigated under hydroponics conditions between pH of 4.5 to 6.7 and natural photoperiod. For toxicity test *lemna minor* L. were exposed to different concentration of CuSO₄ (0.5, 1.0, 2.0, 4.0 mg/l and control) for measurement of frond numbers, general appearance and chlorophyll content. CuSO₄ exposure caused chlorosis and necrosis in all test groups. Frond dissociation was noticed at 1.0 mg/l, chlorophyll content was found decreased in all concentration and was maximum at 4.0 mg/l concentration. Severity of symptoms was found toxicant concentration dependent. Possible mechanisms of copper toxicity to these valuable bioindicators have been discussed. The purpose of this study was to investigate the tolerance to copper in duckweed as a first step to determine the use of this aquatic species to remove heavy metal from polluted river water.

Key words: *Lemna minor*, Heavy metal toxicity, Copper, Chlorophyll

Introduction

Lemna minor L. (duckweed) are aquatic plants, which often form dense floating mats in ponds and rivers (Driever et.al., 2005). The plants are fast growing and adapt easily to various aquatic conditions and can tolerate a wide pH range (4.5-8.3) (Cayuela et. al.,2007). The small size, simple structure and rapid growth make duckweed very suitable for toxicity test (OECD,2002). Some studies indicate that duckweed plants are sensitive to toxicity (Khellaf and Zerdaoui,2010). Other studies however, report that duckweed plants are tolerant to environment toxicity (Wang, 1990). To assess the tolerance of the species *lemna minor* to heavy metals plants were exposed to different concentration of copper. Toxic effect of pollutant on duckweed is generally evaluated by phytotoxicity tests based on growth inhibition (Geoffroy et.al.,2004).

Copper a “ Gray listed “ heavy metal, despite working as essential metal for several biological processes, also become toxic at higher concentration. Copper is introduced in aquatic ecosystem by natural process as well as by human interferences. Copper were chosen as the metals for this study for a number of reasons. Their presence above trace levels in the environment is an indicator of industrial pollution. On the other hand, they are essential micronutrients for plants; Copper is a structural and catalytic component of many proteins and enzymes involved in metabolic pathways (Teisseire and Vernet,2000). However, when the concentration reaches a threshold value, these essential metals becomes first inhibitory and afterwards toxic. Copper is responsible for many alterations of plant cells (respiration, photosynthesis, pigment synthesis and enzyme activity (Kanoun Boule et.al.,2009). Each plant species has different resistance and tolerance level to different contaminants (Kamal et.al.,2004).

The present study investigates copper toxicity to *Lemna minor* to determine tolerance of this aquatic species to copper. The goal was to assess the possibility to use *Lemna minor* for phytoremediation of copper contamination in water and investigate the tolerance to Cu in duckweed to determine the use of this aquatic species to remove heavy metal from polluted river water.

Materials and Methods:

Plant material and culture medium: *Lemna minor* L. A common duckweed plant (Angiosperm; family *lemnaceae*) is widely distributed and found floating in local rivers and ponds with flat fronds and fine roots. The plants were collected from Gomti river and cultured in Hoagland's medium at pH 6.1 (Hoagland's, 1950). Healthy plants were used in the tests after one week of culturing (EPA, 1975). A continuous aeration system provides oxygen for the *lemna* fronds and prevent root fungal diseases (Kamal *et.al.*, 2004). Plants were cultured at $26 \pm 2^\circ\text{C}$ and natural photoperiods.

Toxicity test: The test protocol were derived from the standard draft guidelines 221 (OECD, 2002). Duckweed growth was measured after four days of exposure to different concentration of copper (0.5, 1, 2, 4, 8, 16, 32, 64, 128, 256, 512, 1024, 2048, 4096, 8192, 16384, 32768, 65536, 131072, 262144, 524288, 1048576, 2097152, 4194304, 8388608, 16777216, 33554432, 67108864, 134217728, 268435456, 536870912, 1073741824, 2147483648, 4294967296, 8589934592, 17179869184, 34359738368, 68719476736, 137438953472, 274877906944, 549755813888, 1099511627776, 2199023255552, 4398046511104, 8796093022208, 17592186044416, 35184372088832, 70368744177664, 140737488355328, 281474976710656, 562949953421312, 1125899906842624, 2251799813685248, 4503599627370496, 9007199254740992, 18014398509481984, 36028797018963968, 72057594037927936, 144115188075855872, 288230376151711744, 576460752303423488, 1152921504606846976, 2305843009213693952, 4611686018427387904, 9223372036854775808, 18446744073709551616, 36893488147419103232, 73786976294838206464, 147573952589676412928, 295147905179352825856, 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(Wang,1986; Verma *et.al.*,1999). Our finding suggest that duckweed plant very sensitive towards toxicity of copper. Heavy metal toxicity in water suppressed biomass production and metabolic activities.

The presence of toxic chemicals and heavy metals in aquatic bodies due to discharge of industrial effluents, accumulates in the aquatic plants and cause toxicity effects (Rai and Raizada,1989; Huebert and Shay,1993; Sental,1994).Ultimately transfer into food web, when consumed by various living organism (Sharma *et.al.*,2001; Sahu *et.al.*,2007).

Conclusion:

In this work, the tolerance of Lemna minor to copper and the potential accumulation of this metal has been investigated. The metal was tolerated by lemna at 0.5 mg/l. This result revealed high tolerance of this aquatic plant to the heavy metal copper.

Table-1: Effect of Copper on Chlorophyll contents of *Lemna minor*

Conc. Of CuSO ₄ (mg/l)	Chlorophyll-a (µg/g fresh wt.)	Chlorophyll-b (µg/g fresh wt.)	Total Chlorophyll (µg/g fresh wt.)
Control	0.07	0.330	0.4
0.5	0.039	0.313	0.352
1.0	0.055	0.307	0.362
2.0	0.301	0.068	0.369
4.0	0.069	0.056	0.125

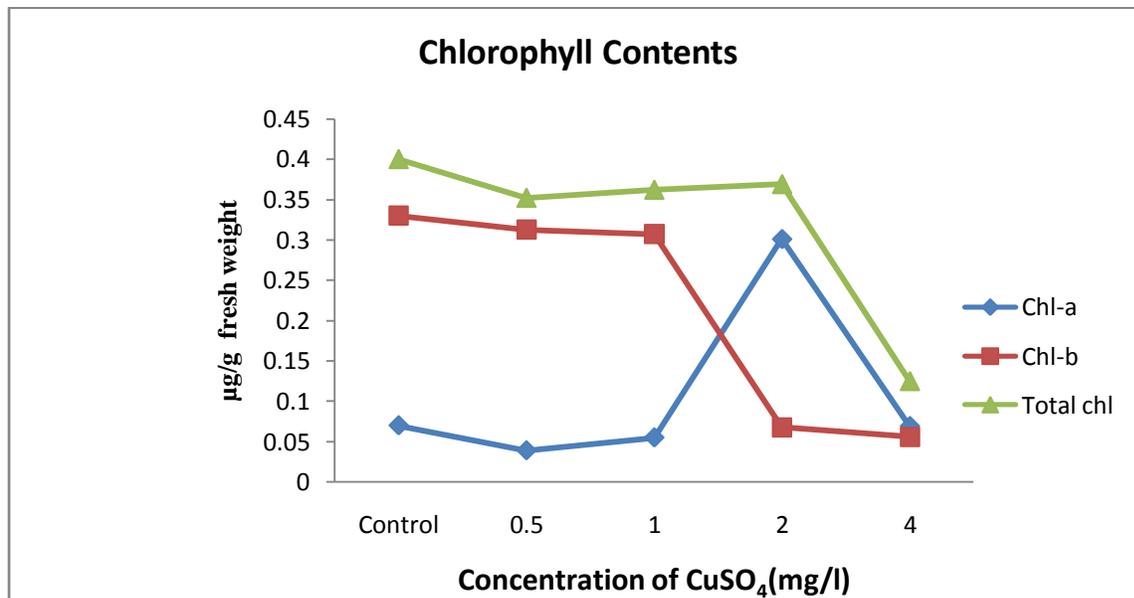


Table-2: Effect of Copper on General appearance of *Lemna minor*

Conc. Of CuSO ₄ (mg/l)	Appearance	Fronde number (Total)	Root structure
Control	No chlorosis, good healthy plants	41	Normal white colour roots of normal length
0.5	Mild chlorosis & necrosis in few number of fronds	35	Roots comparatively longer than control
1.0	Moderate chlorosis with starting of dislocation of fronds	30	Roots longer than control
2.0	Moderate chlorosis & necrosis with moderate dissociation of fronds	33	Roots shorter than control along with few black pattern at tip
4.0	Severe chlorosis and necrosis in about 80% plants, fronds were found highly dissociates and scattered	24	Roots shorter than control, black patches more pronounced in root tips upto 70%, breakage of root tips.

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