

**Ecotoxicological and Risk Assessment of Hydroquinone Cream Residue on Duckweed Plants**

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**ABSTRACT**

Residues from personal care products have been reportedly found in soils and are harmful to plants and animals. This research studied the ecotoxicology effects and risk assessment of hydroquinone cream residue on duckweed plant. The effects of different concentrations (50, 100, 150 and 250 ppm) on the duckweed (*Lemna minor*) were used for the study. The duckweed was collected from the Dam area of University of Ilorin, Ilorin, Nigeria and cultured for a period of one week before using it for the study. The plants were exposed to the different concentrations of hydroquinone cream for a period of 8 days. The results revealed that all the concentration shows adverse effect on the plant pigment content as well as biomass with the effect increasing as the concentration increases. The study has shown that the residue of hydroquinone cream could be dangerous to the ecosystem at concentration as low as 50 ppm which is well below the amount of the residue expected in the environment.

**Key words:** Ecotoxicology, Cosmetics, Hydroquinone, Duckweed.

**INTRODUCTION**

Plants remain indispensable base of all food sources in the ecosystem. Either directly or indirectly both animals and humans depend on plants for food. Unfortunately, due to high industrialization and improper disposal of organic based products such as pharmaceuticals and personal care products, organic chemical contaminants can directly contact and accumulate in aboveground plant tissues through vapor and particle deposition, or in belowground tissues via the roots (Wu *et al.*, 2013). When such organics have accumulated in high amounts within the plants, such plants become diseased and may die eventually (Phetsombat *et al.*, 2006; Su and Liang, 2015). Plants and associated microorganisms can also transform organic chemicals, impacting their environmental fate and transfer to higher trophic levels. Quantifying and predicting the transfer of chemicals from the physical environment into terrestrial plants are important for assessing human and ecological risks, evaluating the use of plants as biomonitors of environmental contamination, and predicting the effectiveness of phytoremediation (Rufli *et al.*, 1998; Doucette *et al.*, 2018).

The quest of quantifying and assessing the impact of these contaminants on the ecosystem resulted to what is known as ecotoxicology. The term ecotoxicology was first coined by Truhaut in 1969 as a natural extension from toxicology, the science of the effects of poisons on individual organisms, to the ecological effects of pollutants (Moriarity *et al.*, 1988). It attempts to combine two very different subjects: ecology ("the scientific study of interactions that determine the distribution and abundance of organisms") and toxicology ("the study of injurious effects of substances on living

organisms", usually man). In toxicology the organisms set the limit of the investigation whereas ecotoxicology aspires to assess the impact of chemicals not only on individuals but also on populations and whole ecosystems. In the broadest sense ecotoxicology has been described as toxicity testing on one or more components of any ecosystem (Cairns, 1989).

Paracelsus (1493-1541) was very clear when he stated that all things are toxic in too high concentrations. He was not perfectly right but toxicants must be defined both quantitatively and qualitatively since toxicity is dose-responsive. Therefore, a chemical might be a contaminant at one concentration and a toxicant at a different concentration because dosage makes a big difference (Tomas, 2005)

During the early years, the major tools of Environmental Toxicology were detection of toxic residues in the environment or in individual organisms and testing for the toxicity of chemicals on animals other than man. It was however, a very big jump in understanding from an experimental animal to a complex, multivariate environment and the subject of ecotoxicology developed from the need to measure and predict the impact of pollutants on populations, communities and whole ecosystems rather than on individuals. There is an on-going debate as to the exact scope and definition of ecotoxicology. The simplest definition found to date is that ecotoxicology is "the study of the harmful effects of chemicals upon ecosystems" (Walker *et al.*, 1996). Suter (1993) defines risk assessment as the process of assigning magnitudes and probabilities to the adverse effects of human activities or natural catastrophes.

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The sources of many environmental contaminants are relatively easy to identify. While short lived contaminants are most readily identified close to the source, the more persistent substances, such as heavy metals and polychlorinated biphenyls (PCBs), may achieve a truly global distribution due to atmospheric transport and deposition to soils and surface waters. The interim period between emission or discharge of an environmental contaminant and ultimate contact with a specific ecosystem or representative species often contains many varied and interesting processes. (Harrison et al., 1992) describe some of the more important processes involved in pollutant transport and removal from the environment and discuss how such processes influence the distribution of pollutants. Included are processes leading to the transfer of chemical substances between environmental compartments such as water to air and air to soil.

Millions of consumers use cosmetic/personal care products (PCP) and their ingredients on a daily basis. Natural and synthetic substances may produce local effects in human skin, such as irritation, sensitization or photoreactions. Given the significant and relatively uncontrolled human exposure to PCP, these products must be thoroughly evaluated for their safety prior to their marketing. (Malkey et al., 1993). Generally, cosmetics are synthetically prepared chemical substances (powdered, liquid or sprays) applied externally to change or enhance the beauty and appearance of skin, hair, nails, lips, and eyes or odour etc. However, some of the chemical substances can be derived from natural sources such as coconut oil (Schneider et al., 1995; Günther et al., 2005).

Hydroquinone is an organic compound of the Phenol family used in body care products for its therapeutic value as a topical agent for the treatment of certain skin conditions. Hydroquinone ( $C_6H_6O_2$ ) is a white, odourless, crystalline solid with an extremely low vapour pressure; it is moderately soluble in water and highly soluble in alcohol. In the presence of water, hydroquinone can slowly oxidize to Quinone which is more volatile (O'Donoghue et al., 2001). Hydroquinone occurs naturally as a glucose ether (commonly known as arbutin) in the leaves of many plants and in fruit, coffee, and wheat products. It is also referred to as 1,4-Benzenediol, para-Dihydroxybenzene and para-Benzenediol. Hydroquinone has been used for more than 100 years as a developer for black and white film (including X-ray film). It is also used as raw materials in the production of antioxidants for rubber, food grade antioxidants, and liquid-crystal polymers; as a polymerization inhibitor for vinyl acetate and acrylic monomers; and as a topical skin lightening agent (O'Donoghue et al., 2001).

Hydroquinone based products have however been observed to be potential carcinogens as most of the benzene metabolites and derivatives are health hazards (Joseph et al., 1998; Olumide, 2008). Aqueous solutions containing hydroquinone permeated mouse and rat skin in vitro with permeability constants ( $K_p$ ) of  $28 \times 10^{-6}$  and  $23 \times 10^{-6}$  cm/hr, respectively, and in vitro human skin  $K_p$  values of  $4 \times 10^{-6}$  and  $9.3 \times 10^{-6}$  cm/h have been reported (Barber et al., 1995; DeCaprio, 1999). In vitro human skin absorbed 43.3 % of hydroquinone from a 2 % cream at  $2.85 \mu\text{g}/\text{cm}^2/\text{h}$  (Wester et al., 1998). Inhibition of metabolic enzymes with sodium azide did not affect in vitro absorption (Wester et al., 1998), suggesting that the penetration was by passive and not active transport. Some percentages of hydroquinone which have not been absorbed by the body are being washed away in to the environment mostly the aquatic system. It has been reported that hydroquinone is very toxic to aquatic life (fish, plants, algae and invertebrates) and therefore the presence of it in the ecosystem may be hazardous to both plant and animal in the system and consequently have devastating effect on them, however this toxic effect have not been quantitatively studied in recent time. This study therefore aimed at assessing the effect of different concentrations of hydroquinone cream on the ecosystem using the effect on Duckweed as a study organism.

#### **MATERIALS AND METHODS**

The duckweed (*lemna minor*) used in this experiment were collected at the Dam area of University of Ilorin, Ilorin, Nigeria. The plant samples were authenticated at the herbarium of the Department of Plant Biology, Faculty of Life Sciences, University of Ilorin, Nigeria and a specimen copy (UIL001/1014) was deposited. Miss Caroline cream was used which contains 2 % hydroquinone. This percentage translates to 2 g of hydroquinone in every 100 g of the cream. A 1.00 g of the cream in 1000 ml of solution is equivalent to 20 mg/L (20 ppm). Therefore 2.5 g, 5 g, 7.5 g, 12.5 g of hydroquinone cream equals 50 ppm, 100 ppm, 150 ppm, 250 ppm respectively. All experiments were carried out in triplicates. Other concentrations 50 ppm, 100 ppm, 150 ppm and 250 ppm were also prepared using the same procedure (Odumosu et al., 2010; Adebayo et al., 2016).

About 10 g fresh weight of plant (duckweed) was placed in each experimental jar, which contained the control (hydroquinone free) and 50, 100, 150 and 250 ppm of cosmetic solution. Plant samples from each container were separately harvested after 2, 4, 6 and 8 days to analyse for toxicity symptoms, biomass productivity and total chlorophyll content (Siriwan et al., 2006).

The biomass productivity was analyzed by drying the plant samples to a constant weight in an oven at 100 °C for 24 hr. The weight of the plants and exposure time was expressed as percentage decrease of biomass relative to controls (Siriwan et al., 2006).

The chlorophyll content of treated and control plants were measured by the absorption spectra of frond extracts in UV spectrophotometer (Agilent Cary, 60 Malaysia) (Bahrs et al., 2013). The absorbance of pigment extract in 80 % Acetone was measured at wavelengths; 663 nm (A<sub>663</sub>), 645 nm (A<sub>645</sub>) and 470 nm (A<sub>470</sub>). The Chlorophyll a, b and Carotenoid contents of extract were calculated using the formulae:

$$\text{Chlorophyll a; Ch-a} = 12.25A_{663} - 2.79A_{645}$$

$$\text{Chlorophyll b; Ch-b} = 21.5A_{645} - 5.1A_{663}$$

$$\text{Carotenoid; Cx+c} = (1000A_{470} - 1.82Ca - 85.02Cb)/198$$

### RESULTS AND DISCUSSIONS

Variations in leaf pigments (chlorophylls a, b and carotenoids) and its relation can be due to internal factors and environmental conditions. Shaikh and Dongare (2008) reported that chlorophyll and carotenoids content varied with microclimatic conditions in *Adiantum* species. The ratio of chlorophyll-a and chlorophyll-b in terrestrial plants has been used as an indicator of response to light shade conditions (Porra, 1991).

The effect of hydroquinone on the chlorophyll contents of duckweed is shown in Figures 1 (a-c). The total chlorophyll content of the control increased with increasing exposure time. The

chlorophyll contents in the treated plants were significantly decreased from that of the control. The highest chlorophyll contents were observed in the control and the lowest was observed at 250 ppm hydroquinone on day 8. An increase in chlorophyll b above chlorophyll a was observed from day 4 upwards. This may be due to microclimatic conditions or light shade conditions and effect of the concentrations (Shaikh and Dongare 2008). By the day 8 the chlorophyll contents of the 250 ppm was almost 0; meaning that the exposure of these plants to high concentrations of hydroquinone lead to the reduced growth and eventually the death of the plants (especially at higher concentrations; 150 ppm and 250 ppm). Hydroquinone has been reported to caused loss of cellular membrane integrity and loss of macromolecules such as carotenoids and this was further confirmed by the decrease in carotenoid contents as the day went by (Uarrotta et al., 2018)

The toxicity symptoms of duckweed treated with cream containing 2% hydroquinone at different concentrations and exposure times were quite different. From day 4 upwards the plants in 100 - 250 ppm began to look pale when compared to the control. Chlorosis was observed in the leaves starting from the margin of leaves and extending towards the inner portion of the blades. The toxicity symptoms increased with increasing concentration and exposure time. The results indicated that the plants were intolerant to hydroquinone. The plant parts began to dry up compared to the control (Figure 3).

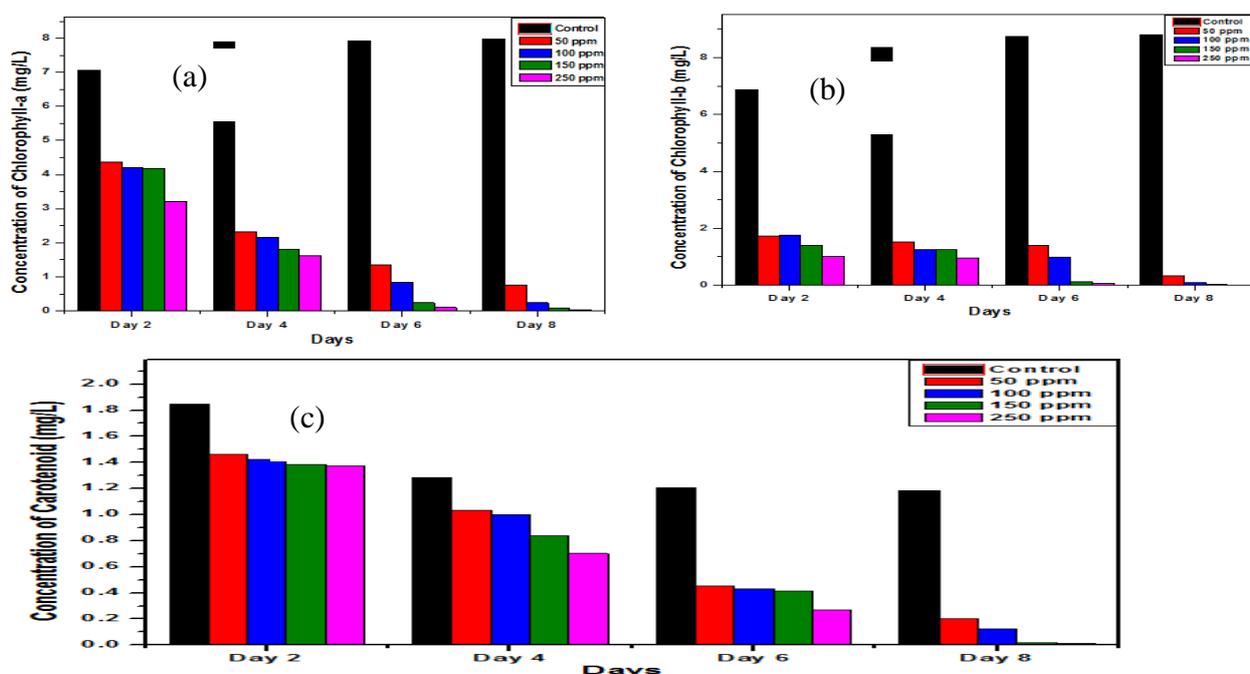


Figure 1: The effects of hydroquinone on (a) chlorophyll a; (b) chlorophyll b and (c) carotenoids contents of Duck weed at different concentrations after eight days.

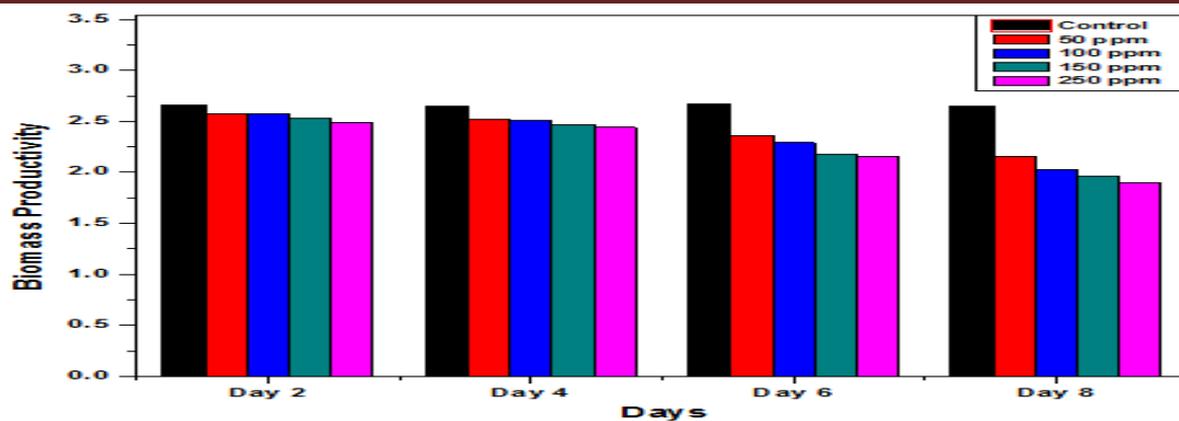


Figure 2: The effects of hydroquinone on biomass productivity of Duck weed at different concentrations after days 2-8.

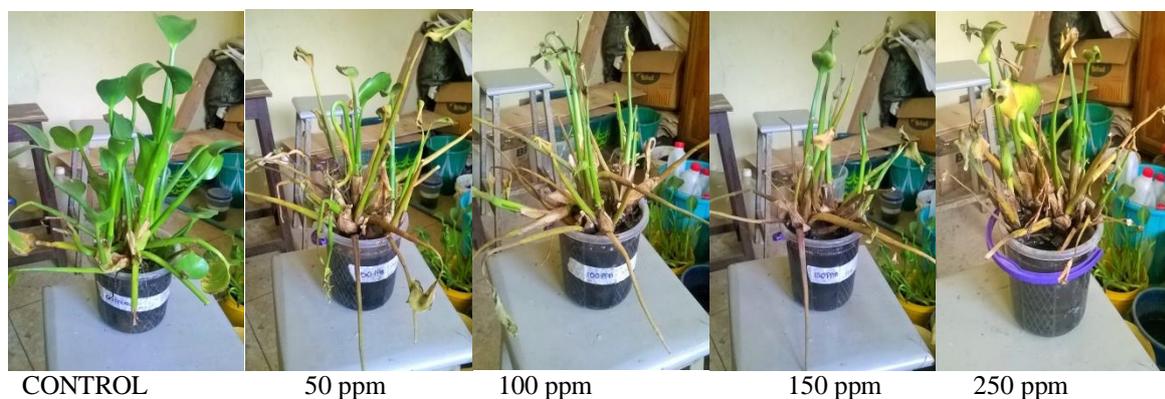


Figure 3: Physical toxicity symptoms of plants after day 8

The effects of hydroquinone on biomass productivity of duckweed are shown in fig. 3. There were significant decreases of biomass when the exposure time and hydroquinone concentrations were increased. Biomass was related to the water content of the plant sample. The water content decreased significantly with increased exposure time and concentrations. Similar observation has been reported in literatures (Bahrs et al., 2013; Adebayo et al., 2016) Highest water content was observed in the control while the least water content was observed in the plant with 250 ppm; indicating the effect of hydroquinone on the plant at increased concentrations compared to the control.

The effect of hydroquinone cream has been shown to affect the daily growth of the duckweed. The growth rate was observed to be reduced with increase in the concentrations of hydroquinone. This is attributed to the ill effect of the hydroquinone that was taken up by the duckweed which was observed to be responsible for the observed inhibition of daily growth and even the death of the plant (Uarrotta et al., 2018). The potential harmful effect of these various concentrations of hydroquinone can be seen from changes in physical appearance (toxicity symptoms), chlorophyll content, and biomass

productivity of the duckweed as the concentrations of the hydroquinone increased. These effects may as well indirectly affect the aquatic animals leading to serious ecotoxicological hazard. The presence of hydroquinone from different pollution sources at very low concentration can significant decline the growth of the duckweed and may have devastating effects on the ecosystem.

### CONCLUSIONS

This research studied the impact of different concentrations of hydroquinone on duckweed plant for a period of eight days. The results revealed that with increased concentrations of hydroquinone, the hazardous effect on duckweed became pronounced and this was evident in reduction in the pigment contents (chlorophyll-a, chlorophyll-b and carotenoid). Exposure of duckweed to higher concentrations of hydroquinone within the studied period also resulted in decrease in its biomass productivity and consequently that would result in decrease in oxygen production in the ecosystem. Overall, this study has established that exposure of plants to cosmetic residue is hazardous to the plants and the ecosystem at large and therefore there is a great need to monitor, control and enforce appropriate disposal of personal care products.

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