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# ECOTOXICOLOGICAL EFFECTS OF HYDROQUINONE COSMETIC ON GROWTH AND PIGMENT CONTENT OF *SALVINIA CUCULLATTA* (FERN PLANT)

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**Abstract**
*Study on the effects of different concentrations of (10ppm, 20ppm and 50ppm) hydroquinone solutions has been carried out on Salvinia Cucullata growth. The plants were cultured in a control with no hydroquinone and at different concentrations of hydroquinone solutions in the laboratory for a period of 8days. The toxicity symptoms, total chlorophyll content and biomass were determined at 2,4,6 and 8days.The results revealed significant ($P \leq 0.05$) effects on chlorophyll contents. Plants in the control having the highest chlorophyll content) and biomass as the concentration increases (from 11.45g which is control to 5.79g which is 50 ppm). The result has shown that cosmetic products that contain 2% hydroquinone (20,000ppm) are likely to produce residue of about 50ppm and hence can be detrimental to the ecosystem.*

***Keywords***: *Cosmetics residue, ecotoxicity, salvinia cucullatta, pigment, hydroquinone*

## Introduction

The term “ecotoxicology” has been defined as the branch of toxicology concerned with the toxic effects, caused by natural or synthetic pollutants, to the constituent of ecosystems, animal (including human), vegetable and microbial, in an integral context (Truhant, 1977). We are all connected between the communities of living things. Plants can absorb toxins through their roots and leaves. Animals and humans are always exposed to chemicals by the air we breathe, things we touch, and what we put in our mouth (Harley et al, 2006). Animals and humans also eat other animals or plants that are already poisoned, which will continue the spread of chemicals, which is referred to as secondary poisoning (Oregon State University, 2011).

Cosmetics products since the dawn of civilization are considered a part of routine body care (Brown, 2013). During the last few decades these products have had a big boost and applied to the human body for beautification (AL-Dayel et al., 2011). These cosmetic products include, care creams, talcum and face powders, lipsticks, eye makeup, etc. (Chauhan et al, 2010). Some cosmetics are benign; (things we cannot do without) others can cause harmful effects such as cancer, allergic reaction, mutations, respiratory problems as well as development and reproductive problems in humans (CDC, 2003). An increase level of cadmium has been reported to cause inhibition of DNA mismatches.

Cosmetics have often been considered by many dermatologists as more harmful products (Groot et al., 1994). These contain more than 10, 000 ingredients which are linked to many diseases like cancer, birth defects, developmental and reproductive harm. A variety of chemicals are used in cosmetics as ingredient and some are used as preservatives. These chemicals have different health effects (Groot et al., 1994).

Plants are what make up the most vital tropic level of the biomass pyramids, known as the primary producers. Because they are at the bottom of the pyramid, every other organism in an ecosystem relies on the health and abundance of the primary producers in order to survive. Ferns are vascular plants that reproduce via spores and have neither seeds nor flowers. They have stems, roots and leaves (McCausland., 2009). In botany, chlorosis is a condition in which leaves produce insufficient chlorophyll, as it is responsible for the green colour of leaves, chlorotic leaves are pale, yellow, or yellow-white. The affected plant has little or no ability to manufacture carbohydrates through photosynthesis and may die unless the cause of its chlorophyll insufficiency is treated (Kubis et al., 2004). Hydroquinone is one of the components of cosmetics used as a topical application in skin whitening to reduce the color of skin. The Food and Drug Administration stated that hydroquinone cannot be ruled out as a potential carcinogen. This conclusion was reached based on the extent of absorption in humans (Olumide et al., 2008).

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This study aim at investigating the ecotoxicological and risk assessment of hydroquinone cosmetics residue using the effect on growth and pigment contents of Fern (*Salvinia Cucullata*).

**Materials and Methods**

**Culture of plants**: Salvina Cucullata were collected from a natural pond and cultured in the laboratory under controlled conditions (12hr/12hr. light and dark cycle, under the temperature of 25$^0$C. The solution of 100 ppm hydroquinone was prepared by dissolving 0.1 g of hydroquinone in a 1000 ml volumetric flask and tap water was added to make up to the makeup to the mark in the volumetric flask. 10 ml of hydroquinone was measured and makeup to the mark in a 100 ml volumetric flask to give 10 ppm solution. The same was procedure was repeated for the preparation of 20 ppm and 50 ppm. About 10g fresh weight of plant was placed in each experimental jar each contained 500ml of a control and 10ppm, 20ppm, 50ppm of hydroquinone solution. The pH of the solutions was adjusted to 5.58. Plant samples from each container were separately harvested after 2, 4, 6, and 8days to analyze for toxicity symptoms, total chlorophyll content and biomass determination. The experiments were set up in triplicate for each concentration and test duration (Siriwan et al., 2006).

**Total Chlorophyll Determination**

The chlorophyll content of treated and control plants were measured by the absorption spectra of frond extracts in UV spectrophotometer. The absorbance of pigment extracts were measured at 663nm, 645 nm and 470nm.The extraction was carried out by weighing 1g of each plant sample, placed in a mortal and crushed with 50ml acetone. It was filtered through a sieve into test-tubes, and was centrifuged using Model PLC-012 E Universal Centrifuge at 7000rpm for five minutes. The chlorophyll content of treated and control plants were measured by DU 730 Life Science UV/Vis Spectrophotometer at 663 nm, 645nm and 470 nm and the amount of these pigments were calculated using formula: Acetone

$C_a$= 11.75$A_{663}$ – 2.350$A_{645;}$ $C_b$ = 18.61$A_{645}$ – 3.960$A_{663;}$ $Cx_{+c}$ = 1000$A_{470}$ – 2.2.70$C_a$- 81.4$C_b$/227 (Lichtentaller et al., 1985) where $C_a$, $C_b$ and $C_{x+c}$ are Chlorophyll a, Chlorophyll b and Carotenoid contents respectively.

**Measurement of Biomass**

Plant samples were cut and weighed before drying in a pre-labeled weighing dish in a drying oven set at 100$^0$C for 24hrs. The dry weight of plants for each concentration was expressed as decrease of biomass relative to controls (Siriwan et al., 2006).

**Results and Discussions**

**Toxicity symptoms**

The toxicity symptoms of *Salvinia Cucullata* treated with Hydroquinone at different concentrations and exposure times were investigated. Chlorosis was observed in the leaves starting from the margin of leaves and extending towards the inner portion of the blades. They later turned brown and separated from other parts on day 8. The toxicity symptoms increased with increasing concentration and exposure time. The roots of *Salvinia Cucullata* were still brown in color and healthy looking. The results indicated that roots of *Salvinia Cucullata* were more tolerant to hydroquinone concentration than leaves.

**Total Chlorophyll content**:

The effect of Hydroquinone concentrations on total chlorophyll content of *Salvinia* Cucullata is shown in Table 1. The total chlorophyll content of control increased with increasing exposure time. The total chlorophyll contents in treated plants were significantly P ≤ 0.5 decreased from that of control as shown in the table below

**Table 1.1: The result after calculating the chlorophyll a and chlorophyll b and total carotene levels of Day 2**

| | $C_a$ | $C_b$ | $C_{x+c}$ |
|---|---|---|---|
| **Control** | **11.0** | **17.6** | **6.8** |
| **10ppm** | **10.8** | **17.2** | **2.1** |
| **20ppm** | **7.4** | **12.7** | **1.8** |
| **50ppm** | **3.3** | **6.2** | **1.3** |

**Table 1.2: The result after calculating the chlorophyll a and chlorophyll b and total carotene levels of Day 4**

| | $C_a$ | $C_b$ | $C_{x+c}$ |
|---|---|---|---|
| **Control** | **9.5** | **19.4** | **6.3** |
| **10ppm** | **5.9** | **13.9** | **5.6** |
| **20ppm** | **3.1** | **13.2** | **2.6** |
| **50ppm** | **0.7** | **6.8** | **1.7** |

**Table 1.3: The result after calculating the chlorophyll a and chlorophyll b and total carotene levels of Day 6**

| | $C_a$ | $C_b$ | $C_{x+c}$ |
|---|---|---|---|
| **Control** | **12.9** | **24.6** | **5.3** |
| **10ppm** | **3.1** | **12.0** | **1.8** |
| **20ppm** | **1.8** | **11.1** | **1.6** |
| **50ppm** | **0.5** | **2.6** | **1.1** |

**Table 1.4:The result after calculating the chlorophyll a and chlorophyll b and total carotene levels of Day 8**

| | $C_a$ | $C_b$ | $C_{x+c}$ |
|---|---|---|---|
| **Control** | **14.2** | **26.9** | **5.3** |
| **10ppm** | **3.1** | **10.1** | **4.3** |
| **20ppm** | **1.9** | **8.1** | **3.7** |
| **50ppm** | **0.2** | **2.7** | **2.6** |

**Biomass Determination:**
The effects of Hydroquinone solution after day 8 is shown in Table 2. There were significant $P \leq 0.5$ decreases of biomass both before the plants were dried and after they were oven dried. The highest biomass was found in the control, while the lowest was found in 50ppm.

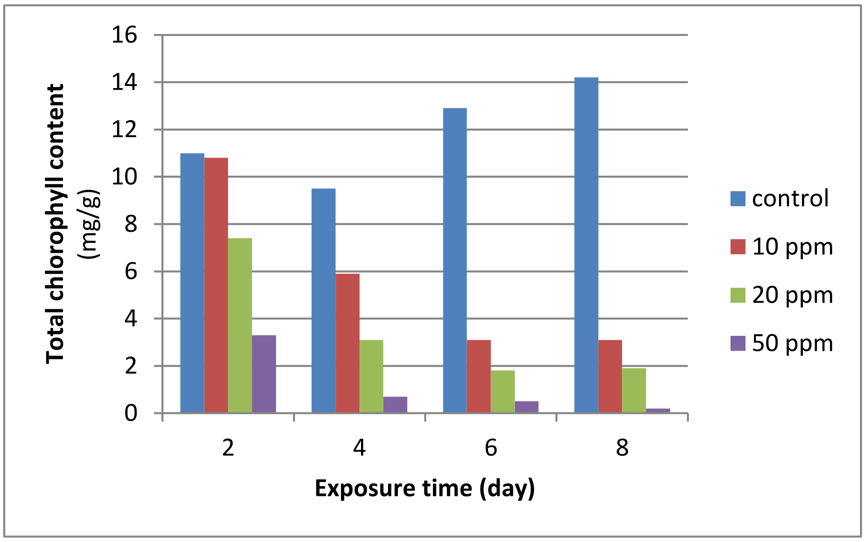



**Fig. 1.0 The graphical representation of total chlorophyll content a (mg/g) against exposure time (day)**

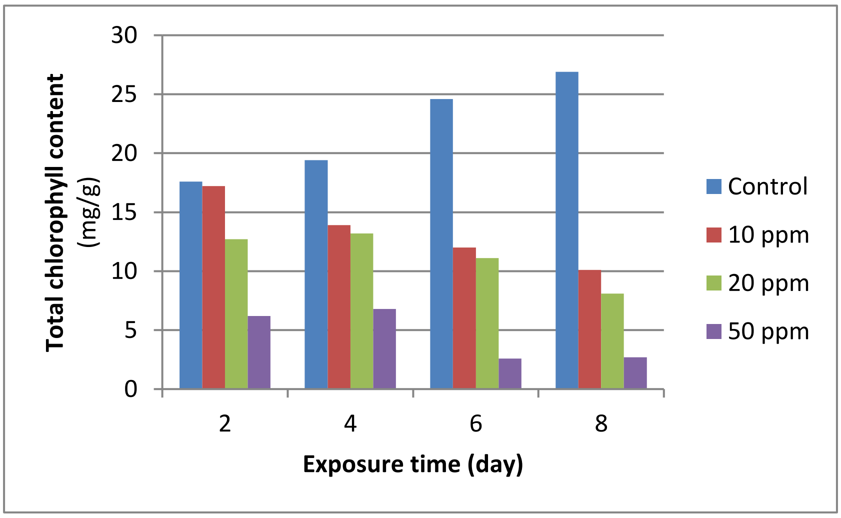
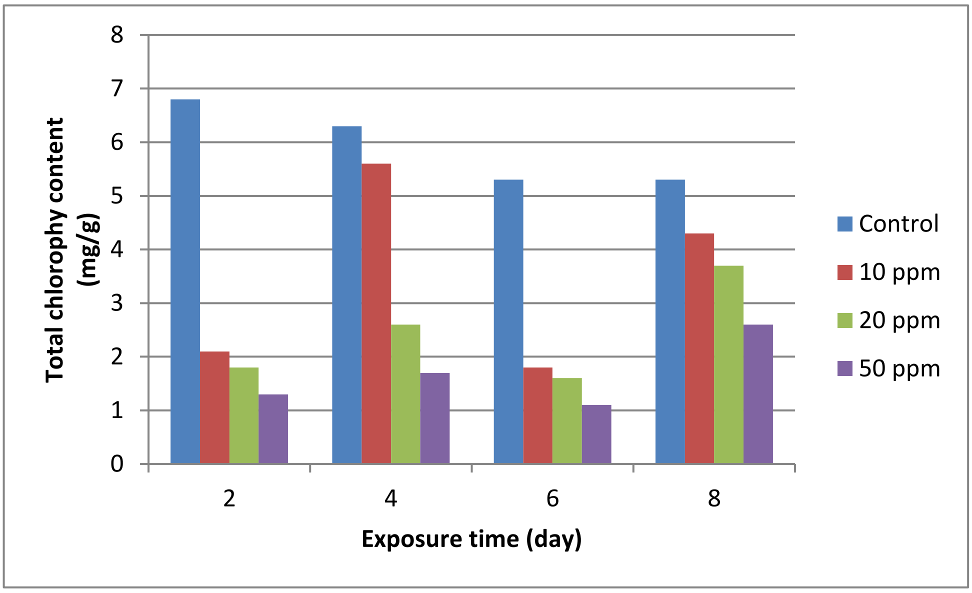



**Fig. 1.1 The graphical representation of total chlorophyll content b (mg/g) against exposure time (day)**




**1.2 The graphical representation of total chlorophyll content b (mg/g) against exposure time (day)**

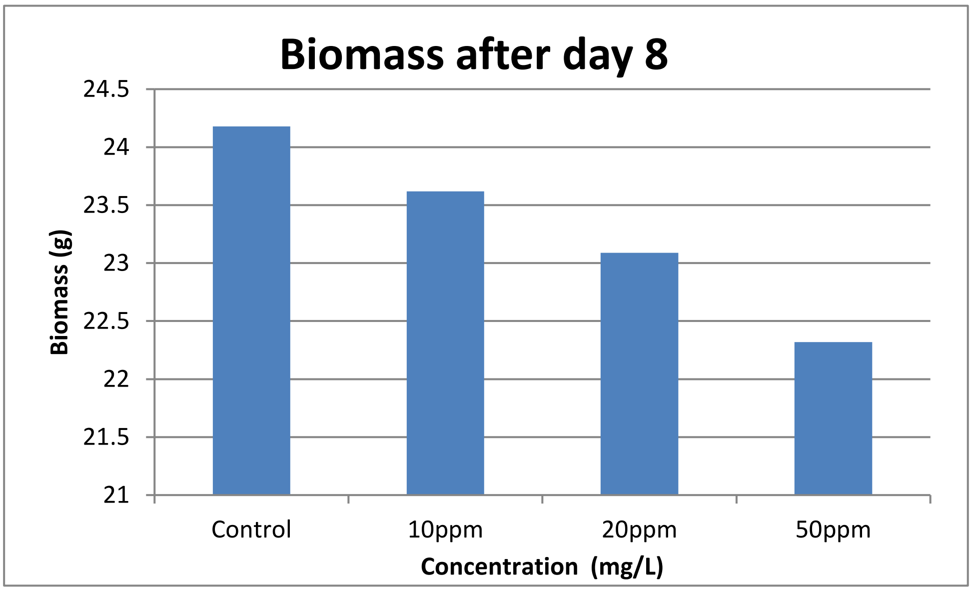



**Fig. 1.3 The graphical representation of biomass (g) against exposure time (mg/L)**

**Discussion**

This study shows that exposure to higher concentration of hydroquinone for longer time has adverse effect on growth, pigment content and biomass of *Salvinia Cucullata*. The present study shows that hydroquinone are toxic to *Salvinia cucullata* as shown by the toxicity symptoms such as chlorosis, decrease in the biomass and total chlorophyll content. It was reported that in botany, chlorosis is a condition in which leaves produce insufficient chlorophyll. As chlorophyll is responsible for the green color of leaves, chlorotic leaves are pale, yellow, or yellow-white. The affected plant has little or no ability to manufacture carbohydrates through photosynthesis and may die unless the cause of its chlorophyll insufficiency is treated (Kubis et al., 2004).

Therefore, this result has shown chlorosis in the leaves of *Salvinia Cucullata* because of the hydroquinone concentration introduced. Other studies have shown that cadmium and lead are toxic to *Salvinia Cucullata* (Mohan et al.,1997) and ,*E.Crassipes* (Zhu et al.,1999), Similar symptoms were reported in other plants exposed to concentrations of mercury resulting in the poisoning of the cell cytoplasm in the plants decreasing the total chlorophyll content (Overnell,1975).The content of chlorophyll a decreased more than the total carotenoids when the plants were treated with 20ppm and 50ppm of hydroquinone solution from day 2 to day 8 as shown in the

table above as total carotenoid decreases at different days. Chlorophyll a of 10ppm did not show significant $P \leq 0.5$ difference to that of the control at day 2 which shows that the adverse effect might be insignificant.
The low levels of cadmium and lead as ingredients in cosmetics is prohibited in many advanced countries, (Al-Saleh et al., 2011). Metallic impurities cannot be avoided, even under good manufacturing conditions. This suggests that some hazardous impurities cannot be avoided, even under control, because some exist naturally in the environment (Al-Saleh et al., 2011). Although carotenoids are more tolerable than chlorophyll a, the content of total carotenoids has been reported to decrease significantly under high concentrations of Antimony Sb (V) (50 and 100mg/l) after a long term exposure (72h). The protection mechanism was damaged or reduced to long term exposure to high concentration of Sb (V) (Pan et al., 2008), this is similar to the result obtained in this study (fig 1.0). The decrease of chlorophyll b in the plants may be due to more carotene pigments that have short wavelength and can absorb the light. It has been pointed out that in the studies with *Chlorella* Beyerinck that carotene pigment levels might be different even with different plants (Moshe et al., 1994).

The highest amount of chlorophyll values was found in control. Another toxicity symptom observed as a result of hydroquinone exposure was decrease in biomass of *Salvinia Cucullata* after day 8 as shown in fig 1.2 in when the exposure time and concentration were increased. It was reported that exposure of this plants to cadmium and lead concentration also shows a decrease in biomass of the plants. Both cadmium and lead were found to inhibit growth in many other species of aquatic plants such as *Lemna Species*. (Wang et al., 1986).

Comparing this result with the assumption of it being the residue after wash off with the concentration of the hydroquinone (20,000ppm) present in a skin toning cream (Miss Caroline) bought in the market, the effect of lower concentration of hydroquinone (10ppm, 20ppm, and 50ppm) is a clear testimony that the 2%hydroquinone (20,000ppm) in the toning cream (Miss Caroline) can be hazardous and injurious to the ecosystem and overall environment.

**Conclusions**

This study has revealed that continuous use of cosmetics that contain hazardous chemicals could result in an increase in the toxic effect to the ecosystem beyond acceptable limits. Cosmetics are benign i.e. we cannot do without using them on our skin most especially after bathe, due to this reason there is a growing concern that some of the cosmetics may contain hazardous substances which are injurious to the ecosystem because of their high durability. They can enter into plants; through wash off since plants have the tendency to absorb these hazardous chemicals, water via drainage, and industrial activities. Efforts should be made to enlightening the users and the general public on the dangers involved.

**Reference**


Al- Saleh and S. Al-Enazi (2011): “Trace metals, “Toxicological & Environmental Chemistry, 93. (6), 1149-1165

Annegrete Kuschel, Xemapetsitis, wolfgangpape, Halimaat /ppenandwalterDiembeck(2005). “Skin cosmetics in allmann’s Encyclopedia of Industrial Chemistry /Willey – Vch Weiner .

An J, Zhou Q, Sun Y, XuZ, (2009). “Ecotoxicological effects of typical personal care products on Seed germination and seedling development of wheat (Triticum Aestiru L)”.Chemosphere p1428-1434

Brown, V.J (2013). Metals in lip products: a cause for concern. Environ. Health prospect 121, 196.
CDC, Centres for Disease Control and Prevention, 2003 Second National Report on Human Exposure to Environmental Chemicals http://www.cdc.gov/exposurereport/2nd/.5.

Chaudan, A.S, Bhadaunia, R., Singh, A.K., Lodhi, S.S., Chaturvedi, D.K., Tomari, V.S. (2010). Determination of lead and cadmium in cosmetics products. J. Chem. Pharm. Res. 2 (6), 92 – 97.

Groot, A.C.D., Naterm J.P., Wayland, J.W., (1994). Unwanted Effects of cosmetics and Drugs used in Dermatology, third (issue 1) Elsevier, 2 -15Gunther Schrader, SVeGolda,

Harley (2006). The impacts of climate change in coastal marine systems. Ecology letters 9, (2), 228-241

Huebert, D.B and M. Shay(1993). The response of lemn triscula L. to cadmium. Environmental Pollution. 80, 247-253

Jorg. Scheber, waltiaudKaden, UweSchoridrock, Hartmet Schmidt-lewekuhrne,UNEP (2008). Mercury in products and wastes. General United Nations Environment Programme, Division of Technology, Industry and Economics, Chemicals Branch.

Lichtenthaler. H.K. Wellburn. A.R (1980). Determination of Total Carotenoids and Chlorophylls A and B of leaf in Different Solvents. Biol.Soc. Trans. 11. 591-592

Mc Clausland, Jim (2009-09-20) “Rediscover Ferns” Sunset.com. Retrieved 2013-09-07.

Mohan, B.S and B.B Hosetti (1997). Potential Phytototoxicity of lead and cadmium to lemna minor grown in the sewage stabilization ponds. Environmental Pollution.2, 133-228 (1997)
Soil Pollution. 215:517-523

Moshe Rise, Ephraim Cohen, Marina Vishkautsan, Miriam Cojocaru, Hugo E. Gottlieb, Shoshana (Malis) Arad (1994). Accumulation of Secondary Carotenoids in *Chlorella zofingiensis* Journal of plant physiology.144.3,287-292. Elsevier

Olumide YM; Akinkugbe, AO; Altrade; D; Mohamed T. Ahamefule, N; Ayanlo. S; Onyekonim,C, Session N (2008).“Complications of Chronic use of Skin Lightening Cosmetics.” International Journal of Dermatology 47(4)344-53.

Oregon State University. (2011). Ecotoxicology topic fact sheet-Retrieved December 6, 2011, from National Pesticide Information center website: http://inpic.orst.edu/factssheets/ecotox.pdf. The humane society of the United States, (2011). Ecotoxic. Retrieved December 12, 2011 from Procter and gamble website: http//altox.org/ttrc/toxicity-tests/ecotoxicity/

Overnell. J. (1975) : The effect of some heavy metal ions on photosynthesis in fresh water alga pesticides, Biochem. Physiol., 5, 19-26

Pan XL, Zhang DY, Chen X, Bao, A.M, Li H. (2010). Antimony accumulation growth Performance antioxidant defense system and photosynthesis of Zea mays in Response to antimony pollution in soil. Water Air

Siriwan Phetsombat, Maleeya Krutachue, Prayad Potethitiyook, Suchart Upatham (2006). Mahidol University, Bangkok 10400, Thailand, 3Faculty of Science, Received: 20 June, 2005; Accepted: 08 January, 2006)

Truhant, R. (1997).“Eco-Toxicology-Objectives, Principles and Perspectives”, Ecotoxicology and Environmental safety, 1 (2): 151-173

Wang, W (1986). Toxicity tests of aquatic pollutant by using common duckweed. Environ. Pollution. 11, 1-14.