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Effect of Dyes on the Storage Metabolites and Growth of *Hibiscus rosa - sinensis* under Field Conditions

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Abstract: The objective of the present study was to investigate the physico-chemical properties and the impact of dyes on the biochemical properties of *Hibiscus rosa- sinensis.* The physico-chemical properties of dye effluent was determined by following the standard methods of APHA 2005 and the biochemical methods were analyzed with the help of UV spectrophotometer. The physico-chemical parameters showed great variation in the different parameters analyzed and the concentration of different storage metabolites decreased due to the influence of different dyes used in this study. However, the cumulative impact may be tolerated by the plants of *Hibiscus rosa- sinensis* and can be grown near the industrial areas manufacturing different dyes.

Key words: Dyes · Effluent · *Hibiscus rosa - sinensis* · Storage metabolites

INTRODUCTION

The different parts of *Hibiscus rosa-sinensis* are reported to have beneficial effects for various ailments. The dried flower powder of *H. rosa-sinesis* has significant protective effects in ischemic heart disease. The leaves and flowers besides having healing properties are effective in the treatment of arterial hypertension. Few studies claim that *Hibiscus rosa- sinensis* promotes growth of hair and checks bleeding, relaxes spasms and have significantly antifertility effects [1,2]. The buds of *Hibiscus rosa- sinensis* are reported to have cooling and astringent effect and it removes burning sensation of the body as well [3]. Soil pollution by industrial effluent is one of the vital issues on the environmental concern. The dyes effluent from almost all dyeing industries of the country is directly or indirectly discharge into the soil. The continuous disposal of dye effluent into soil deteriorates the quality of the soil due to the mixing of various chemical pollutants of the effluent. Effluents from industries are normally considered as the main industrial pollutants containing organic and inorganic compounds. On the other hand, irrigating with waste water may reduce purification level and fertilization costs, as it may serve as fertilizers, due to its nutrients value. Most of the time use of plants offers an advantage over other organisms, because they can be more sensitive to environmental stresses. Use of dye industrial effluents for irrigation purpose is a highly warranted utility of dye effluent in the present world. However, the continuous supply of dye effluent in the soil ecosystem will decline the soil fertility and may cause different changes in the physiology and biochemistry of the plant at the same time.

It is expected that dye industries in Gwalior may change the properties of soil and water as most of the dyes may dissociate in the water and soil. This is turn, may cause various changes in the biochemistry of the *Hibiscus rosa- sinensis* if grown around these industries. Keeping the above facts in view, the present study was undertaken to know the physicochemical properties of dye effluents generated from dye industries in Gwalior and to investigate the biochemical changes like carbohydrates protein fats and carotenoids in *Hibiscus rosa- sinensis* under temperate condition.

MATERIALS AND METHODS

Physico- Chemical Analysis: The color of the dye samples was determined by platinum cobalt method, odour was analyzed by dilution factor, pH of the dye

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waste water were determined by portable (Digital pH meter (Modal No-112) Electrical conductivity was also determined by (EI Digital conductivity meter 641) and turbidity was measured with the help of Jyoti 1028 digital turbidity meter following the procedure of Quirk & Schofield. Dissolved oxygen (DO) was measured by dilution and Winkler's method. Biochemical oxygen demand (BOD) was measured by dilution and Winkler's method [4]. Chloride was measured by Mohr's method and Total Suspended Solid (TSS) and Total Dissolved Solid (TDS) were measured by physical and gravimetrically methods, while Suspended Solids (SS) was obtained by subtracting the TDS from TS. Other parameter such as hardness was measured by titration method, alkalinity/acidity by dilution titration method. The concentration of sodium and potassium of the sample were determined by flame photometer (Model No.1034).

Biochemical Parameters: The storage metabolites were determined with help of the UV spectrophotometer (λ -25) by following methods.

The determination of total carbohydrate was estimated by Duboi's *et al*., [5] method and the lipid content was determined by the method of Folch and Lees [6]. The chlorophyll content of the cells was estimated by the method of Mackinny [7], Myers and Kratz [8] and protein was estimated by Lowry's method [9].

RESULTS AND DISCUSSION

Untreated textile dye effluent that is discharged into various drains contains a number of azo dyes used in textile printing industries of Gwalior. The effluent is discharged directly into drains may also percolate into the soil ecosystem. During the last few years, most of the plants have been tested for dye tolerance and change in storage metabolites due to influx of pollutants. It is strongly advised to treat dye effluent before draining the effluent into the agriculture or specific plants. However, treating effluent may remove the organic nutrient as well, as dye effluent is considered as nutrient resources is industrial waste water, i.e. about 80% water and the rest as organic and inorganic nutrients. Since, its disposal is a big problem in urban areas, applying the textile waste water to *Hibiscus rosa-sinensis* instead of disposing off in lakes is an alternative, if it may not interfere with its chemical composition. It is argued that there can be both beneficial and damaging effects of irrigation with waste water on various crops including vegetables [10] especially the textile industry effluent water that contains much of undesirable qualities like opacity due to suspended particles, alkaline pH and toxic synthetic chemicals such as the dyes. The need is to assess waste water quality and plant species requirements before using treated waste water for crop production [11]. The presence of acids, alkalis, salts, or metal ions as impurities may interfere in enzyme action by replacing metal ions from metal enzymes and inhibit different physiological processes of plants [12,13].The decrease in pH is due to addition of various soluble dyes in industrial effluent. Accordingly, crop growth neither needs a high pH (above 6.25) nor low pH (below 6.05) for maximum production. Electrical conductivity is commonly used as a measure of salinity of samples. Electrical conductivity ranged between 7.7 to 150.8μs/cm in dye effluent industrial samples. In the contaminated samples, electrical conductivity may have increased due to presence of total dissolved solids, particularly Na^+ and K^+ that may have significantly increased the electrical conductivity in the effluent. Increase in the sodium ion concentration of samples can be attributed to minerals in the dye effluent. High amounts of sodium ions can result in precipitation of calcium and magnesium ions from the samples thus affecting their effectiveness in enhancing internal drainage.

The effluent water showed that pH ranged between 6.82 to 6.8 means slightly neutral in nature. Chloride concentration was maximum (mg/l) 2933 in Indigo dye as compared to other dyes (Table 1). It was 521.85, 145.55, 117.15 mg/l in Malachite, Nepthoquinone and Cochineal dyes, respectively. On the other hand, the total hardness of the dye was 240, 230, 260, 223 mg/l for Indigo dye, Cochineal dye, Malachite dye and Nepthoquinone dyes, respectively (Table 1).

Total carbohydrates, total reducing sugars, total protein and chlorophyll contents were analyzed for various treatments. The results are presented in Tables 2 and 3. Total content of protein was decreased compared to control, except for Malachite dye, where it showed a slight increase in its contents. Total protein content was 1.33, 1.24, 1.76, 0.86 and 0.98, mg/g fresh weight in control, Cochineal, Malachite, Indigo and Nepthoquinone dyes, respectively. Total carbohydrates content was increased in Cochineal and Nepthoquinone and it significantly decreased up to 14.58, 15.24 in Cochineal and Malachite dyes, respectively. At the same time, it was 6.14 mg/g fresh weight in Indigo dye and 9.18 mg/g fresh weight in Nepthoquinone dye. A significant increase in carotenoids content was observed for all the dyes investigated. There were insignificant differences among the biochemical parameters. However, the values of biological treatment

Table 1: Comparison of Physico-chemical properties of dyes

	Comparative status of dyes				
Parameter characteristics	Indigo dye	Cochineal dye	Malachite dye	Nepthoquinone dye	Irrigation standard
Temperature (°C)	17	16.5	19	18	27
pH	6.82	7.5	8	8	9.5
Electric conductivity (μs/cm)	7.76	54.1	58	150.8	200
Acidity	62.5	0	0	0	-
Total dissolve solid (gm/l)	69.46	129	139	447	2100
Total suspended solid (mg/l)	6.502	10	19	24	200
Turbidity (NTU)	13.5	4	3	2	-
Dissolve oxygen (mg/l)	9	14	15	11	4.5-8
Biological oxygen demand (mg/l)	1	2.035	4.31		250
Total hardness (ppm)	240	230	260	623	-
Chloride (mg/l)	2933	117.15	521.85	145.55	1000
Sodium (mg/l)	13	0	10	0	-
Potassium (mg/l)	0	1	12	13	-
Alkalinity	195	500	537.5	400	-

Table 2: Morphological changes in *Hibiscus rosa- sinensis* by dyes

Physical parameter	Indigo dye	Cochineal dye	Malachite dye	Nepthoquinonedye
Leaves	No change	No change	No change	No change
Stem	No change	No change	No change	No change
Flower	Light yellow	Light yellow	Light yellow	Light yellow

Table 3: Status of storage metabolites in *Hibiscus rosa-sinensis* as affected by dyes

Dyes	Protein	Carbohydrates	Carotenoids	Chlorophyll. B	Chlorophyll. a
Control	1.336	1.276	1.343	0.884	0.545
Cochineal dye	0.676	0.659	2.445	1.941	0.933
Malachite dye	1.767	1.687	2.372	0.947	0.539
Indigo dye	1.268	1.243	1.157	0.421	0.228
Nepthoquinone dye	1.225	1.177	1.947	0.599	0.367

were closer to that of the control than the values of the chemically treated effluent. However, in Indigo dye both proteins as well as carbohydrates are influenced and decreased to a large extent because of stress induced by high chloride content. However, carotenoids content was increased in the Indigo dye to overcome the stress caused by the high chloride content. It is already reported that phenolics and carotenoids helps the plants to overcome stress induced by both biotic, as well as, abiotic system [14]. Overall, the chlorophyll content was decreased by the dyes however in Cochineal there is an increase in both chlorophyll a and chlorophyll b content. Except for Cochineal dye our work follows the same trend that chlorophyll content was decreased due to dyes.

CONCLUSION

The dye effluent containing reactive azo dyes cause serious environmental hazards. The physiochemical parameters of the different dyes showed variation at different sites. Photosynthetic pigment like chlorophyll a and chlorophyll b deceased after treatment of dyes. Total amount of protein decreased in *Hibiscus rosa-sinensis* after treatment with dyes. A significant increase in the amount of Carotenoids was observed in *Hibiscus rosa- sinensis* during the study period. Change in the protein, Carbohydrate content observed significantly and there was a decrease in the chlorophyll a and chlorophyll b *Hibiscus rosa-sinensis* may be grown around textile dye industries as no marked changes were not observed in storage metabolites. However, it will be better to treat or dilute the effluent before it is discharge.

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