Screening of Plant Species for Phytoremediation of Synthetic Textile Dye Wastewater

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Research

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Abstract

Most of the dyes are carcinogenic and mutagenic in nature. Plants are potential candidates to remEDIATE textile dye wastewater from contaminated sites. The present study aimed to design an efficient hydroponic system to screen potential ornamental plant species for removal of synthetic dye solution of triarylmethane dye Methylene Blue (MB) and diazo dye Congo Red (CR). The six plants selected for screening are Trachyspermum ammi, Tagetes erecta, Hibiscus rosa-sinensis, Chrysanthemum indicum, Bryophyllum fedtschenkoi, and Catharanthus roseus. The phytotreatment of dyes was done up to 40 h for two different concentrations of dyes, i.e., 10 and 20 mg L\(^{-1}\). Among screened plant species, the maximum decolorization was obtained from T. ammi followed by B. fedtschenkoi. Both plant species showed active growth in indigenous designed hydroponic system even after the phytoremediation process. T. ammi decolorized the MB dye 99% (10 mg L\(^{-1}\)) and 86% (20 mg L\(^{-1}\)) while the decolorization of the CR dye solution was up to 95% (10 mg L\(^{-1}\)) and 84% (20 mg L\(^{-1}\)). T. ammi was found to have maximum potential among screened plants for the removal of MB and CR dye from synthetic dye solution when kept in designed hydroponic system and can be used for phytoremediation of wastewater contaminated with synthetic dyes.

Introduction

Due to the increasing world population, there is a tremendous growth of various industries, which uses many harmful chemicals for the generation of different commodities for public demands but the side byproducts such as contaminants not only affect water bodies but also the air and soil. Dyes have a major demand and application in the textile industries for the dyeing process. About 10–15% of the azo dyes get lost in the effluent during the dyeing process (Stolz 2001) and 50% of other reactive dyes are reported for use in the textile industry which is discharged into water (Chen 2002). Azo dyes are extensively used in the dyeing process. The effluent containing dyes released into the surrounding seriously affects the environment by destroying the ecosystem, causing water pollution, and reducing light penetration for aquatic life (Imron et al. 2019). Due to textile dye wastewater, the biological oxygen demand, chemical oxygen demand, and suspended solids increase in the nearest river located beside the textile industry (Ekanayake et al. 2021). So, there is a big challenge to treat textile dyes effluent before released into water bodies.

There are many physical and chemical methods, for example, adsorption, coagulation, sedimentation, flocculation, filtration, photo degradation, and chemical oxidation, for managing contamination produced by textile dyes (Robinson et al. 2001; Khandare and Govindwar 2015). These methods relate to the high expense, low productivity, require huge space and undependable to work. Because of these issues, there is a requirement for the advancement of productive and cost-effective methods for the treatment of textile dyes. Biological methods are more effective than physical and chemical methods to treat the textile dye wastewater. Biological methods involve different enzymes, microorganisms, and plants for the removal of dyes from wastewater (McMullan et al. 2001; Chacko and Subramaniam 2011; de alkimin et al. 2020). From the different biological methods, plant-based phytoremediation is an energy-efficient,
solar-driven process to remove contaminants from soil, air, and water (Khandare and Govindwar 2015; de alkimin et al. 2020). Phytoremediation is also used to remove pollutants from textile dye wastewater. There are various phytoremediation mechanisms as phytoextraction, phytodegradation, rhizofiltration, phytostabilization, phytovolatilization for dye removal (Jeevanantham et al. 2019). Due to these different processes, plants are used for the treatment of textile dye wastewater. There are many studies reported in the literature on the use of aquatic plant species for the phytoremediation of dye wastewater such as *Ipomoea aquatic* (Rane et al. 2011), *Salvinia molesta* (Al-Baldawi et al. 2020; Imron et al. 2019; Kaushal and Mahajan 2015), *Typha angustifolia* (Chandanshive et al. 2017), *Chara vulgaris* (Mahajan and Kaushal 2013; Mahajan et al. 2019; Mahajan and Kaushal 2020), *Eichhornia crassipes* (Tan et al. 2016), *Lemna minor* (Imron et al. 2019; Reema et al. 2011), *Azolla pinnata* (Al-Baldawi et al. 2020) and *Pistia stratiotes* (Mahajan and Kaushal 2019), but very few reports are available on phytoremediation textile dye wastewater using ornamental plants. The *Petunia grandiflora* which is a flowering ornamental plant species reported for its potential to remove the triphenylmethane textile dye Brilliant Blue G (Watharkar et al. 2013). *Aster amellus*, a herbaceous plant species used to decolorize a sulfonated azo dye Remazol Red and a mixture of dyes and a textile effluent (Khandare et al. 2011). *Glandularia pulchella* has been explored to decolorize the dye Green HE4B (Kabra et al. 2011) and *Ipomoea hederfolia* ornamental plant able to decolorize the dye mixtures and Scarlet Red dye (Rane et al. 2014). *Alcea rosea* plant has the potential to remove Disperse Red 60 and Reactive Blue 19 dye (Mahmoudabadi et al. 2019). The researchers also explored the phytoremediation potential of *Portulaca grandiflora* (Khandare et al. 2013), *Blumea malcolmii* (Kagalkar et al. 2009), *Typhonium flagelliforme* (Kagalkar et al. 2010), among others for dye degradation in aqueous form. There is no research work based on textile dye removal by *Trachyspermum ammi*, *Tagetes erecta*, *Hibiscus rosa-sinensis*, *Chrysanthemum indicum*, *Bryophyllum fedtschenkoi*, and *Catharanthus roseus*. In literature, these plant species have been reported for their potential remediation of heavy metal in different studies (Li et al. 2020; Bardiya-Bhurat et al. 2017; Bada and Raji 2010; Ahmad and Misra 2014; Kumar and Dwivedi 2013; Richwagen et al. 2019; Chukki et al. 2018; Missoum 2018) where these plants have an efficient root system and plants do not affect the food chain. The researchers have also exploited the potential of phytoremediation potential for removal of heavy metals using hydroponic system (Bello et al. 2018) but there is no report to our knowledge where ornamental plants are exposed in remediation of synthetic dyes using hydroponic system so in the present research study a hydroponic system is designed to engrossed on the ability of terrestrial ornamental plants for decolorization of Methylene Blue (MB) and Congo Red (CR) dyes.

**Materials And Methods**

**Chemicals and plant material**

The triarylmethane dye, MB and a diazo dye, CR dye were used for experimentation. MB is a heterocyclic aromatic chemical compound with molecular formula \( \text{C}_{16}\text{H}_{18}\text{N}_{3}\text{SCl} \). The molecular weight of MB dye is 320 g mol\(^{-1}\). CR dye is a diazo dye that can be synthesized by a coupling reaction containing hydroxyl, amino or other groups with an aromatic diazotized base. The chemical formula of CR dye is
C\textsubscript{32}H\textsubscript{22}N\textsubscript{6}Na\textsubscript{2}O\textsubscript{6}S\textsubscript{2} and its molecular weight is 696 g mol\textsuperscript{-1}. The chemical structure of MB and CR is given in Fig. 1. MB dye and CR dyes were purchased from Sanjay lab Amritsar, India. All the chemicals used were of the highest purity and of an analytical grade. The synthetic dye wastewater was prepared at two different concentrations of 10 and 20 mg L\textsuperscript{-1}. The entire apparatus was sterilized before experimentation. Screened ornamental plants \textit{T. ammi}, \textit{T. erecta}, \textit{H. rosa- sinensis}, \textit{C. indicum}, \textit{B. fedtschenkoi}, \textit{C. roseus} were harvested from the Botanical garden of Guru Nanak Dev University campus, Sathiala and Government High School, Sathiala (Punjab), India. The plants were washed completely to remove mud, dirt, and particulate matter and acclimatized for three days in distilled water. Table 1 shows the description of screened plants used for the research study.

| Plant species                   | Common name          | Family      | References                  |
|---------------------------------|----------------------|-------------|-----------------------------|
| \textit{Trachyspermum ammi}     | Ajwain               | Apiaceae    | (Kumar and Dwivedi 2013)    |
| \textit{Bryophyllum fedtschenkoi}| Lavender scallops    | Crassulaceae| (Richwagen et al. 2019)     |
| \textit{Chrysanthemum indicum} | Guldaudi             | Asteraceae  | (Chukki et al. 2018)        |
| \textit{Tagetes erecta}        | Marigold             | Asteraceae  | (Bardiya-Bhurat et al. 2017)|
| \textit{Hibiscus rosa-sinensis}| Chiana rose          | Malvaceae   | (Missoum 2018)              |
| \textit{Catharanthus roseus}   | Periwinkle           | Apocynaceae | (Ahmad and Misra 2014)      |

**Experimental design**

A two-stage hydroponic system was designed in which two PVC pipes of 0.30 m diameter and 1m long was horizontally connected via movable stopper as shown in Fig. 2. Both the pipes were attached at an angle of 45°. The bigger holes of 0.05 m diameter were made for keeping the plants in vertical position in each pipe and small sized holes of 0.10 diameter were kept for aeration. The inlet of wastewater was kept above the top of first pipe and final outlet at the lower end of second pipe (Fig. 2).

Initial experiments were performed to identify the plants having the potential to decolorize the textile dyes, for which \textit{T. ammi}, \textit{T. erecta}, \textit{H. rosa- sinensis}, \textit{C. indicum}, \textit{B. fedtschenkoi}, and \textit{C. roseus} plants were selected. Firstly, the roots of these plants were washed with running tap water to remove adherent soil after which plants were entirely washed with distilled water. Plants were put into distilled water for hydroponic treatment (without soil) and the growth of the plants is checked up to three days. The treatment of selected plants was done in the upper pipe of hydroponic system after the acclimatization period of three days with 10 and 20 mg L\textsuperscript{-1} concentrations of both MB and CR dye solution. Each pipe of the system is having two types of holes – one is bigger in which plant is kept and in smaller hole, atmospheric oxygen can enter freely. Synthetic solution was added from the inlet of container attached to
upper pipe. The acclimatized plants were transferred to prepared dye solutions of different concentrations. Both biotic and abiotic controls were also maintained. The abiotic controls contained the MB and CR dye solution without plants whereas plants in distilled water were kept as biotic controls. The decolorization was noticed up to 40 h (0, 8, 16, 24, 32, and 40). The absorbance of each solution was determined with UV-Visible spectroscopy at its respective absorption maxima ($\lambda_{\text{max}}$ (MB) – 668 nm; $\lambda_{\text{max}}$ (CR) – 498 nm) using Systronic-2202 UV-Vis double beam spectrophotometer. The percentage decolorization was calculated as per equation (Al-Baldawi et al. 2018):

$$\text{Decolorization (\%)} = \frac{A_0 - A_1}{A_0} \times 100$$

Where $A_0$ is an initial concentration of dye and $A_1$ is the final concentration of dye. After 40 h of treatment, treated water was drained to lower pipe and same plants were placed on the bigger holes of lower pipe. The lower pipe was to observe the growth of plant roots and it was perceived that after 7 days, there were small growth of newly developed roots. To optimize the efficiency of the hydroponic system, similar set of plants were kept in upper pipe for treatment of synthetic water. Each batch of dye concentration and screened plants had triplicates for each biological sample for obtaining the concordant results. The data was analyzed by using MS-excel 2007 windows.

### Results And Discussion

The utility of hydroponic system was optimized by using two sets of plants- one set in upper pipe where treatment of synthetic water was screened by different plants and in lower pipe, growth of roots of screened plant were observed in treated water after treatment (Fig. 3). The decolorization results of each plant were compared with the abiotic and biotic control dye solution. The roots of plants were found to have dye pigmentation in comparison to biotic control via physical examination (Fig. 4). The results of different batch experiments for decolorization of dyes with respect to time are shown in Fig. 5. It has been observed from Fig. 5 that the decolorization percentage of dye increases with increase in time. The same pattern of dye decolorization has been reported by various researchers (Al-Baldawi et al. 2020; Kaushal and Mahajan 2015; Reema et al. 2011). For instance, the different Green HE4B dye concentrations were reduced to varying extent during 48 h of contact by *Glandularia pulchella* and maximum decolorization was observed at 48 h in each concentration (Kabra et al. 2011). All these decolorization results and the impact of synthetic dye wastewater on the growth of plant used for screening are summarized in Table 2.
Table 2
Decolorization pattern of Methylene Blue and Congo Red dyes and their impact on plant growth

| Plant species                  | % Decolorization | Plant growth (after removal of dye) |
|-------------------------------|------------------|-------------------------------------|
|                               | Methylene Blue   | Congo Red                           |
|                               | 10               | 20                                  |
|                               | 10               | 20                                  |
| (mg L$^{-1}$)                 |                  |                                     |
| Trachyspermum ammi            | 99 ± 6           | 86 ± 7                              |
|                               | 95 ± 6           | 84 ± 7                              |
|                               | Active           |                                     |
| Bryophyllum fedtschenkoi      | 85 ± 6           | 69 ± 7                              |
|                               | 77 ± 6           | 70 ± 6                              |
|                               | Active           |                                     |
| Chrysanthemum indicum         | 87 ± 9           | 70 ± 10                             |
|                               | 60 ± 9           | 52 ± 9                              |
|                               | Inactive         |                                     |
| Tagetes erecta                | 84 ± 8           | 68 ± 8                              |
|                               | 67 ± 8           | 66 ± 7                              |
|                               | Inactive         |                                     |
| Hibiscus rosa-sinensis        | 86 ± 6           | 71 ± 7                              |
|                               | 59 ± 7           | 47 ± 6                              |
|                               | Inactive         |                                     |
| Catharanthus roseus           | 35 ± 6           | 34 ± 3                              |
|                               | 48 ± 5           | 43 ± 3                              |
|                               | Active           |                                     |

Remark: All data values are median ± S.D., n = 3

Figure 5a and 5b show the decolorization pattern of MB and CR dye by utilizing *T. ammi* plant. Out of six screened plants, excellent decolorization of MB was observed in the case of *T. ammi* plant. The decolorization of 10 and 20 mg L$^{-1}$ MB was 99 and 86%, respectively. Plant growth was normal after adsorption of the dye into the roots. The decolorization (%) of CR dye by *T. ammi* shown in Fig. 5b clearly indicates the admirable efficiency of *T. ammi* to decolorize the CR. The percentage decolorization of 10 and 20 mg L$^{-1}$ of CR dye was 95 and 84%, respectively. The plant remains survived after the adsorption of dye into the roots. However, percentage decolorization decreases with an increase in concentration. These outcomes show that *T. ammi* plant is an outstanding plant to decolorize the azo dye CR and triarylmethane dye MB at a lower concentration. The decolorization of MB and CR dye by using *B. fedtschenkoi* plant is shown in Fig. 5c and 5d respectively. The plant *B. fedtschenkoi* shows significant decolorization of triarylmethane dye MB having a percentage decolorization of 85% (10 mg L$^{-1}$) and 69% (20 mg L$^{-1}$). The response of *B. fedtschenkoi* plant towards the removal of a toxic azo dye, CR was also observed as significant for textile wastewater treatment. The *B. fedtschenkoi* decolorized the CR dye 77 and 70% for 10 and 20 mg L$^{-1}$ dye concentrations, respectively. It was observed that plant parts remained active after adsorption of the dye and were able to remove more dye concentration than 20 mg L$^{-1}$. These results proved that *B. fedtschenkoi* plant has a good tendency to decolorize synthetic wastewater of CR azo dye as well as triarylmethane dye MB.
Figure 5e and 5f show the decolorization of MB and CR respectively by using *C. indicum*. The percentage decolorization obtained for 10 and 20 mg L\(^{-1}\) MB dye concentrations were 87 and 70\% respectively. Initially plant leaves became dried, later stems and roots of the plant also showed dryness after the removal of dyes. The plant becomes died after treatment with higher dye concentrations. However, the MB color removal by this plant was acceptable yet plant endurance was not significant for the treatment of triarylmethane dye, MB. The results with CR dye synthetic wastewater revealed only 44 and 42\% decolorization at 10 and 20 mg L\(^{-1}\) concentrations, respectively. Wilting of the plant takes place after treatment of CR dye. The plant was not able to treat dye concentration higher than 20 mg L\(^{-1}\). Hence, *C. indicum* is not suitable for the phytotreatment of CR synthetic dye wastewater.

*T. erecta* plant was also used for a screening test to remove MB dye from synthetic wastewater. It was observed that plant had the more capacity to decolorize the triarylmethane dye, MB in comparison to CR dye. Figure 5g and 5h show the decolorization of MB and CR dye, respectively. The decolorization for 10 and 20 mg L\(^{-1}\) MB dye wastewater was 84 and 68\% respectively. After decolorization, the MB dye plant shows withering. Initially, the leaves become dry then subsequently stems and roots. Due to these conditions, the plant was no more active for treatment with more MB dye concentrations than 20 mg L\(^{-1}\). The percentage decolorization was observed 67 and 66\% for 10 and 20 mg L\(^{-1}\) CR dye concentrations, respectively. Though the plant can decolorize the azo dye, CR and MB but *T. erecta* plant dryness after removal of the toxic dye makes it unsuitable for the treatment of synthetic dye wastewater.

Figure 5i and 5j show the decolorization of MB and CR respectively by *H. rosa-sinensis* plant. The decolorization obtained were 86 and 71\% from the 10 and 20 mg L\(^{-1}\) MB concentrations respectively and 41 and 39\% decolorization at 10 and 20 mg L\(^{-1}\) CR dye solution. It indicates the potential of *H. rosa-sinensis* for MB synthetic dye wastewater decolorization. But the toxicity of dye effects on plant growth results in its inability to remove dye concentrations than 20 mg L\(^{-1}\).

Figure 5k and 5l show the percentage decolorization of MB and CR dye respectively by *C. roseus*. The decolorization percentage obtained for MB 10 and 20 mg L\(^{-1}\) was 35 and 34\% respectively and 48 and 43\% for CR 10 and 20 mg L\(^{-1}\) respectively. In the case of *C. roseus* plant, it is found that the plant remains active after dye removal however plant removal efficiency is quite slow for both the dyes. It was observed that plant could not effectively decolorize the synthetic wastewater up to 40 h.

Hence, the results obtained from the screening experiments clearly indicate that the maximum percentage decolorization obtained from the *T. ammi* plant followed by *B. fedtschenkoi* and both plants also remain active after removal both MB and CR dyes. *C. indicum* and *T. erecta* plants also show their potential for decolorization of synthetic dye wastewater however, their survival rate makes them insignificant for phytoremediation process. *H. rosa-sinensis* plant was also not considerable for survival because flowers wither after dye removal. The plant *C. roseus* is able to bear the toxic impact of dyes but the rate of decolorization is quite slow for both MB and CR dyes.
In the literature, the removal of MB and CR was reported by a few researchers by using the phytoremediation technique as shown in Table 3. *Eichhornia crassipes* successfully removed MB dye (50 mg L\(^{-1}\)) in 20 days up to 98% (Tan et al. 2016) while *Lemna minor* (2 g) was exposed to 50 mg L\(^{-1}\) of MB dyes for 24 h decolorization of 81% (Imron et al. 2019). In another study, 98% decolorization has been reported for *Lemna minor* in 144 h at 10% concentration and authors claimed it as a phytoremediation agent to remove MB dye from wastewater (Reema et al. 2011). Another aquatic species *Azolla piñata* also reported in literature for removal of MB dye (Al-Baldawi et al. 2011). In literature, MB remediation is mostly reported by using aquatic plant species. In the present research work, ornamental plant *T. ammi* plant showed the decolorization up to 99 (10 mg L\(^{-1}\)) and 86% (20 mg L\(^{-1}\)) for MB dye in 40 h experiment only. Hence, *T. ammi* plant has been proven to be more effective than *E. crassipes* and *L. minor*. Again, for phytoremediation of CR dye, *Chara vulgaris* (Mahajan and Kaushal 2013) and *Pistia stratiotes* (Mahajan and Kaushal 2019) aquatic species are reported for maximum decolorization 95 and 90% respectively. In the present study, *T. ammi* exhibited the maximum decolorization up to 95 and 84% at 10 and 20 mg L\(^{-1}\) CR dye concentrations respectively and remained active after the decolorization process. However, it has been observed that the dye was found to be adsorbed on the roots of *T. ammi* plant possibly due to the rhizofiltration process, and hence plant could be able to provide maximum decolorization. Therefore, *T. ammi* plant acts as a potential candidate for future research where it can be used as a phytoremediator for decolorization of dye wastewater.
### Table 3
Comparison of results of present study with existing literature for phytoremediation of Methylene Blue

| Plant                      | Dye | Dye concentration (mg L$^{-1}$) | Time (h) | Decolorization (%) | References                          |
|----------------------------|-----|---------------------------------|----------|--------------------|-------------------------------------|
| *Lemna minor*              | MB  | 10                              | 144      | 98                 | (Reema et al. 2011)                |
|                            |     | 50                              | 24       | 80                 | (Imron et al. 2019)                |
| *Azzola pinatta*           | MB  | 25                              | 24       | 85                 | (Al-Baldawi et al. 2018)           |
| *Eichhornia crassipes*     | MB  | 50                              | 20d      | 98                 | (Tan et al. 2016)                  |
| *Chara vulgaris*           | CR  | 50                              | 24       | 95                 | (Mahajan and Kaushal 2013)         |
| *Pistia stratiotes*        | CR  | 40                              | 72       | 90                 | (Mahajan and Kaushal 2019)         |
| *Trachyspermum ammi*       | MB  | 10                              | 40       | 99                 | Present study                      |
|                            |     | 20                              |          | 86                 |                                     |
|                            | CR  | 10                              |          | 95                 |                                     |
|                            |     | 20                              |          | 84                 |                                     |
| *Bryophyllum fedtschenkoi* | MB  | 10                              | 40       | 85                 | Present study                      |
|                            |     | 20                              |          | 69                 |                                     |
|                            | CR  | 10                              |          | 77                 |                                     |
|                            |     | 20                              |          | 70                 |                                     |

### Conclusion

The design of hydroponics system shows the efficient utilization of system for screening the plants and developing the growth of the root system of plants in optimum time by using alternative two pipe system. The results from the present research support the ability of six screened plants for the removal of MB and CR dyes. *T. ammi* and *B. fedtschenkoi* are the most efficient plants for the removal of both dyes. Moreover, the survival of both plants seems to be significant. Maximum percentages of decolorization obtained from the *T. ammi* plant are 99 (10 mg L$^{-1}$) and 86% (20 mg L$^{-1}$) for MB dye, and 95 (10 mg L$^{-1}$) and 84% (20 mg L$^{-1}$) for CR dye due to its adsorption on the roots of the plant. Therefore, further research work can be focused on dye removal by using *T. ammi* plant based on the adsorption mechanism. In the future, adsorption mechanism explored by using different instrumental techniques such as Fourier Transform Infrared spectroscopy, Scanning Electron Microscopy, and statistical analysis.
can also be done with different operational parameters such as plant weights, the relative growth rate of plants, the effect of pH, etc.

**Declarations**

**Competing interests:** The authors declare that there are no competing interests.

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**Availability of data and materials**

The data used to support the findings of this study are available from corresponding authors upon request as the relevant data will be used by PhD scholar for her future works in continuation.

**Authors’ contributions**

Navjeet Kaur: Conducted the experimental studies and drafted the manuscript; Jyotsna Kaushal: Conceptualization, expert view and overall Supervision; Pooja Mahajan: Data interpretation; Arun L. Srivastva: Suggestions and interpretation on the chemical analysis. All authors read and approved the final manuscript.

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**Figures**

![Chemical structure of Methylene Blue (MB)](image1a) ![Chemical structure of Congo Red (CR)](image1b)

**Figure 1**

Chemical structure of (a) Methylene Blue (MB) (b) Congo Red (CR)
Figure 2

2-D Experimental design of hydroponic system for synthetic wastewater treatment (Inlet Chamber (1); PVC pipes (3, 7) Big Hole (4); Small holes (5)- Stoppers (2, 6, 8); Outlet tank (9); Stand (10)

Figure 3

Image depicting growth of (a) B. fedtschenkoi and (b) T. ammi before and after 2-step treatment in hydroponic system. Red circles showed the growth of roots enhanced after treatment.
Figure 4

Visual pigmentation of (a) Methylene blue (b) Congo Red dye on roots of T. ammi after 8 h
Figure 5

See the Supplemental Files section for the complete figure caption.

Supplementary Files

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