Isolation and Screening of Azo Dyes Tolerant Bacteria in Semi-Scale Industrial Effluents

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1. Introduction

The poly-aromatic molecules give colour to materials (fabrics, paper, leather, painting and medical industries known as dye. Dyes from industrial effluents are hazards because of mutagenic, carcinogenic and biodegradation resistant properties which effect to the environmental entities in the nature [5, 9]. Several methods are exist to eliminate the Azo dyes from the effluents such as electrochemical treatment, evaporation, reverse osmosis, chemical precipitation, and adsorption process. However these methods are not economic and eco-friendly methods [17]. Biological methods, microbes have ability to decolourize dyes effluents through the process of transformation in this process several metabolites degrade complex wastes in to non toxic simpler by products [16,19]. This ability of microorganism is due to their resistance mechanism of survival such as extracellular precipitation, sorption, enzymatic oxidation and reduction and accumulation [8,12,14]. Biological methods of dye decolourization are efficient, economical and eco-friendly method in which microbes includes bacteria, fungi and yeast.

In present work there is two different bacteria have been isolated which play a significant role among several isolated colonies. These two isolates are further tested over two different azo dyes, which show high potency of decolourization along with good survival resistance.

2. Materials and Methods

2.1 Sampling of Effluent

Samples of industrial effluent were procured from different semi skilled industries (dyer) which are using different dyes for colouring of fabrics in Jaunpur Uttar Pradesh, India. The water samples were collected in sterilized screw cap container during the peak time of dyeing and aseptically transported in laboratory with maintained the temperature of 4ºC [6,10].

2.2 Physicochemical analysis

In this analysis different parameters pH, temperature, turbidity, conductivity, dissolved oxygen (DO), biological oxygen demand (BOD), chemical oxygen demand (COD) and total suspended solid (TSS) of samples were also determined [2].

2.3 Sterilization

Steam sterilization process was perform for disinfection of nutrient agar media using autoclave treatment at 121 ºC, 15 psi for 20 minutes and same process was adopted for all glass wares after proper cleaning [15].

2.4 Isolation and Screening

Procured effluent samples were undergone a serial dilution from 10⁻¹ to 10⁻¹⁰ using double distilled water. The 20 µL of
diluted sample were spread over the surface of nutrient agar media and incubated at 28±2 ºC for 48 hours [4,7,14]. The bacterial colonies were isolated on the basis of morphological appearance and transferred on same media for further purification. Different isolated colony were screened over nutrient agar media supplemented with methyl orange (MO) and trypan blue (TB) Azo dye resistant bacteria [13, 15].

3. Result and Discussion

Table 1 contains the values determined for all physical and chemical parameters of the effluent samples. The sample was bluish black in colour, other parameters like pH, temperature, turbidity, conductivity, DO, BOD, COD and TSS of samples were measured along with dye concentration. pH value of the effluent was 5.4 at 32.6 ºC temperature which indicates slight acidic nature. Very acidic condition pH lower than 4.0 or alkaline condition (above 9.5) are not favourable for microorganism [16]. Generally pH range 6.5 - 7.5 preferred for bacterial growth. Turbidity of effluent sample was observed 7.32 NTU (Nephelometric turbidity unit). DO was measured to be 6.3 mg/L which an important parameter for water quality assessment [4]. However, the TDS and the TSS of the sample were recorded as 21.1mg/L and 25.3 mg/L, respectively. Other parameter like BOD and COD shows relative oxygen depletion. Oxygen demand of microorganisms for biodegradable pollutants is known as BOD, however, COD is the measurement of the oxidisable pollutants in presence of a strong oxidizing chemical agent [1, 3, 11]. The values for BOD and COD were observed as be 40.2 mg/L and 65.30 mg/L, respectively during present study.

| Parameters | Effluent |
|------------|----------|
| Temperature | 32.6 ºC |
| pH | 5.4 |
| Turbidity | 7.32 ntu |
| TDS | 21.1 mg/L |
| TSS | 25.3 mg/L |
| DO | 6.3 mg/L |
| COD | 65.30 mg/L |
| BOD | 40.2 mg/L |

3.1 Isolation of Bacteria

The strain for bacterial community which is present in the sample varied from 200 to 300 cfu. All bacterial strain were isolated, are able to resist 25 mg/L of MO and TB at nutrient agar plate where two best isolate VN1 and VN2 were selected on the basis of resistance Figure- 2.

Figure 1. Isolated and purified bacteria A (VN1) and B (VN2) from dying sites

Figure 2. Methyl orange and Typan Blue (100ppm) completely decolorized using native isolates

3.2 Colony morphology

Highly resistant strain (VN1) was observed as in round shape, green colour and convex, whereas the elevation is convex. The isolate VN2 also shows tolerant nature against MO and TB exhibit similar morphological characteristics. However isolates (VN1 and VN2) have noticeable characteristics because of its releasing legends as greenish in the case of VN1 and brownish in the case of VN2 as shown in Figure 1 and Table 2.

| Isolates | Shape | Colour | Elevation |
|----------|-------|--------|-----------|
| VN1      | Round | Green  | Convex    |
| VN2      | Round | Brown  | Convex    |

3.3 Bacterial adaptation against dye

The eleven isolates obtained from screening, only two are adapted under increasing concentration of azo dyes due to significant potency of microbes. Such kind of potency are found due to suitable ecological interaction under natural condition [18, 19], which is isolated from the dye effluent.
samples by pour plate method based on preliminary tests, plating on selective media.

4. Conclusion

This bioremediation technique is beneficial tool for the removal of contaminants from the environment. Bacterial resistant mechanism against hazards in the environment, both bacterial strain VN1 and VN2 indicates the natural adaptation against chemical dyes. The rate of decolourization is differing due to ecological interaction. This study suggested that dyes contamination impact on human being is directly related to the pollution in his aspects both strain are able to tolerate 500 ppm of MO and TB. In future studies, isolated strain these bacteria can be applied for the bioremediation of industrial effluents.

Acknowledgements

I would like to thanks unseen power which blessings us and my supervisor Prof. M P Singh who encourage me for writing the paper.

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