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# Bioremediation for the Decolorization of Textile Dyes by Bacterial Strains Isolated from Dyeing Wastewater

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# Abstract

Background: The major concern to meet environmental regulations is related with the decolorization and detoxification of industrial dyes contaminated wastewater. So, this study was undertaken to examine the use of bacteria isolated from wastewater of textile factories in the removal of the synthetic textile dyes (Sudan Black, Methyl Red, Malachite Green, Rhodamine B and Brilliant Cresyl Blue). Methods: Dye contaminated wastewater was collected from some synthetic textile factories in Gorgan and Gonbad, Iran, and evaluated for the screening and isolation of bacteria capable of decolorizing textile dyes. The effect of function of operational parameters includes temperature (25, 37 and 50 °C), pH (4, 6 and 8) and initial dye concentration (100, 200 and 300 mg/mL) on the efficiency and rate of discoloration was assessed. Results: Totally, out of the 19 bacterial isolates from textile wastewater: Five bacterial isolates showed dye discoloration ability and the most efficient bacterial isolates were Enterococcus faecium and Pantoea spp. that decolorized Methyl Red, Sudan Black and Malachite Green dyes at 25-37°C, concentration of 200-300 mg/mL and slightly acidic to neutral pH. Enterococcus bacterium was able to decolorize Sudan Black to the 19.79% in the concentration of 100 mg/ml and pH=8 and temperature of 50°C. The highest amount of decolorizing was observed by Pantoea on Malachite Green to the amount of 73%. Enterococcus had the highest decolorizing on Methyl Red to the 65.7%. The amount of decolorizing on Sudan Black by Enterococcus (49.9%) was also higher than Pantoea (39.7%). Conclusion: Isolated bacteria had a significant reduction in toxicity and cationic malachite green dye and azo dye- methyl red. Thus, bacteria can be used in full-scale industrial wastewater treatment with the bio-synergy and its application in discoloration.

Key words: Textile Effluent, Dye, Bio-synergy, Pantoea, Enterococcus faecium

## 1. Introduction

Since 1856 when the first synthetic dye Mauveine was obtained, 100000 synthetic dyes in the world with an annual volume of a 0.7 million tons have been produced so far (Zollinger, 2005). Textile industry consumes two-thirds of this amount of dye (Gita et al., 2017; Lakshmanan and Raghavendran, 2017; Singleton, 2013). Synthetic dyes are macromolecules that are not easily degraded, and some of them are toxic to plants and animals (Varjani et al., 2020). Synthetic azo dyes are known with nitrogen-nitrogen double bond attached to the aromatic groups, and are as the most widely used dyes in the textile industry (Kannan et al., 2013; Chaturvedi et al., 2019). Inefficiency in the dyeing processes makes the 10-15% of the used dyes enters the wastewater (Nikhil et al., 2012; Natarajan et al., 2018; Samsami et al., 2020). Declining water quality, lack of penetration into the lower layers of water, the impact on the gases solubility and toxicity of the dyes or materials resulting from their degradation are such factors threatening the environment (Boyd, 2019). Therefore, elimination of dyes from textile dyeing effluents currently represents a major ecological concern. Chemical methods of wastewater treatment are generally expensive and have limited effectiveness with creating a lot of waste materials, eliminating of which is a new problem (Wu et al., 2011; Crini and Lichtfouse, 2019). Microorganisms are a good alternative for treating contaminants such as synthetic dyes due to genetic and metabolic diversity because many of them, such as a variety of gram positive and negative bacteria, fungi, etc., are capable to decolorize synthetic dyes (Dangi et al., 2019; Lellis et al., 2019). Thus, bioremediation is very important because of low cost, no damage to the nature and small amount of waste products (Alalewi and Jiang, 2012; Vikrant et al., 2018). Effluents of many textile mills of Iran are discarded to the deserts and saline lands because of their geographical location. The structure of many dyes contains toxic metals such as chromium that are released to the wastewater during dyeing and soluble salts such as sodium chloride and potassium chloride are also used in the process of dyeing. Thus, bioremediation in saline soils is associated with many problems due to the harsh conditions in these areas.

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and the only microorganisms are capable of solving it that can withstand the harsh conditions and grow and operate in which. Therefore, the use of halophilic and halotolerant bacteria is considered as a good choice for treating such effluents because halophilic and halotolerant bacteria show high tolerance in harsh environmental conditions such as high content of salt and pollutants such as heavy metals and oxyanions. Halophilic and halotolerant bacteria have shown great success in bioremediation of contaminants from oil and toxic oxyanions (Solís *et al.*, 2012; Ghodake *et al.*, 2011; Romano-Armada *et al.*, 2020). Thus, the aim of this study was to evaluate the use of microorganisms isolated from industrial textile effluents in the Gorgan and Gonbad cities in Golestan province, Iran for the removal of different synthetic dyes.

## 2. Materials and methods

#### 2.1. Bacteria isolation and identification

In this study, 86 samples from different devices and final effluent of textile factories in Gorgan and Gonbad, Iran were collected and transported to the laboratory under sterile conditions. Then, 10 mL of each sample was added to 100 mL of Tryptic Soy Broth medium (Merck, Germany) and were incubated for 24 h at 37°C. Then, the bacteria were identified as conventional microbiological methods (Mahon *et al.*, 2018; Murray *et al.*, 2020).

### 2.2. PCR-Based-Sequencing

After the extracted purified genomic DNA, PCR was performed to amplify *tuf* gene using forward primer of 5'-GCCAGTTGAGGACGTATTCT-3' and reverse primer of 5'-CCATTTCAGTACCTTCTGGTAA-3'.

The amplification reaction was done in 25  $\mu$ L containing 10 pmol of each primer, dNTP in 2.5 mM, 0.6 U of Taq polymerase, 2.5  $\mu$ L of the DNA template and 15 mM MgCl2 buffer. The time and temperature cycle of thermocycler was as 15 minutes at 95 °C, then 35 cycles as 30 seconds at 95 °C, 30 seconds at 56 °C and 45 seconds at 72 °C; and finally, 72 °C for 10 minutes (MyCycler Thermal Cycler, Bio-Rad, Munich, Germany) (Hwang *et al.*, 2011). Amplified products were pooled and purified were purified from agarose gel, and then they sequenced by a national instrumentation center for environmental management, South Korea (NICEM). Sequenced alignments were analyzed by the Basic Local Alignment Search Tool (BLAST) program and submitted to National Center Biotechnology Information (NCBI).

#### 2.3. Dye discoloration

Dyes were sterilized by 0.45 micro-filter and then were added to Tryptic Soy Broth medium. Then, the purified strains were added to the culture medium tubes containing 10 mL of decolorizing medium (containing 0.5 g of glucose, 2.5 g yeast extract and 5 mg dye in 100 mL). The tubes were inoculated with 1% bacteria (opacity of 0.5 McFarland) and incubated at 37°C. Five different dyes (Sudan Black, Methyl Red, Malachite Green, Rhodamine B and Brilliant Cresyl Blue) were used (R Rohban et al., 2009). Sudan Black (C29H24N6) is a non-fluorescent, fatsoluble and thermostable azo dye that is used for staining neutral triglycerides, lipids and some lipoproteins. It seems as a dark brown to black powder with the maximum absorption at 596-605 nm and melting point of 120-124 °C. Methyl Red (2-(N,N-dimethyl-4-aminophenyl) azobenzenecarboxylic acid) that is also called as C.I. Acid Red 2 is an indicator dye that turns red in acidic solutions. Methyl Red is an azo dye as dark red crystalline powder. Methyl Red is a pH indicator. Malachite Green is an organic compound that is used as a pigment and is used as an antimicrobial agent in aquaculture controversially. It is commercially used for staining materials such as silk, leather and paper. Among the 19 isolated strains, the 5 strains were able to decolorize that were selected based on the degree of decolorizing. All strains were investigated separately at the effect of different temperature (25, 37 and 50°C), different initial dye concentration (100, 200 and 300 mg/L) and different pH (4, 6 and 8) on dye discoloration was assessed. All experiments were performed two times and mediums with dyes and noninoculated mediums with dyes used as a blank. Due to the lack of decolorizing on Rhodamine B and Brilliant Cresyl Blue, these two dyes were deleted, and the process was continued with three other dyes (Rohban et al., 2009).

### 2.4. Determination of dye discoloration

The maximum absorption wavelength of each dye determines by the spectrophotometer in the range of 200 to 800 nm. Then it was plotted the chart of each dye according to the amount of absorption in the wavelength of maximum absorbance and concentration of dye in the medium. Absorption of each dye in the wavelength of maximum absorption was measured, and the percentage of discoloration was calculated in accordance with the following formula (Pathak *et al.*, 2014; Rahimi *et al.*, 2016).

Percentage of discoloration: (The obtained OD - The initial OD)/ initial OD  $\times$  100

#### 3. Results

Based on biochemical tests and molecular methods, some species of *Pantoea* genus and Gram-positive cocci, *Enteroccus faecium* were identified (Table 1).

The percentages of discoloration in samples of Methyl Red, Sudan Black and Malachite Green dyes based on the absorption at 340 nm for different initial dye concentrations, temperatures and pHs are shown in Tables 2, 3 and 4, respectively. The highest discoloration percentages for all the tested dyes were attained for acidic to neutral pH values and room temperature (25°C) and 37°C.

Table 1. Identification of species of Pantoea genus and Gram-positive cocci, Enteroccus faecium

No.	Sequencing Result	BLAST
1	CCATCCAAACCCCACGCACAAAAAGCACCTTGGCCGCGAGTATAAGAAAGCCTT CGGGTTGTAAAGTACTTTCACCGGGGAGGAAAGCGATGGGGTTAATAGCCCCTTT TATTGACGTGACCCGCAAAATAATCCACGCTAACTCCGCTGCCTACAACCTCGGT AACTTTTATAAGGTGCCGGCTGTTCTTGACAACTTTCGGCGGGGGTCGTGCACGCTC GTGCTCCTGCTTCGATTCAATCTACACTTCCCCAATCTAAACGTAAGTTTACCTTTT ACTTTAGACTAGGGTGGACCTCCACCCCATTCTTCAATACTACCGCCCACACTCCT ATCTTCCATACTACTCACGTAACTCGTGTTTTTAACTTACAGGACTTGATGAT GGATCTTTAACTTACTAGACCTAATCGCAACCTCAAGTTTAACTTACATCATCAT ATCCTTCTCTTTAAAATTTCTTCTCTGTCTCTCAATTTCCACCGTCATTCCTTCT TCA	Pantoea sp.
2	AAATTGGCGGGACTGTTGCTACAGGTCGTGTTGACGTGGACAAGTTCGCGTTGGT GACGAAGTTGAAGTTGTTGGTATTGCTGAAGAAACTTCAAAAACAACAGTTACTG GTGTTGAAATGTTCCGTAAATTGTTAGACTACGCTGAAGCTGGAGACAACATTGG TGCTTTACTACGTGGTGTTGCACGTGAAGACATCCAACGTGGACAAGTTTAGCTA AACCAGGTACAATCACACCTCATACAAAATTCTCTGCAGAAGTATACGTGTTGAC AAAAGAAGAAGGTGGACGTCATACTCCATTCTTCACTAACTA	Enterococcus faecium
3	AATTGGGGGGGGTCTGTTGCTACGGTCGTGTTGACGTGGACAAGTTCGCGTTGGTG ACGAAGTTGAAGTTGTTGGTATTGCTGAAGAAACTTCAAAAACAACAGTTACTGG TGTTGAAATGTTCCGTAAATTGTTAGACTACGCTGAAGCTGGAGACAACATTGGT GCTTTACTACGTGGTGTTGCACGTGAAGACATCCAACGTGGACAAGTTTTAGCTA AACCAGGTACAATCACACCTCATACAAAATTCTCTGCAGAAGTATACGTGTTGAC AAAAGAAGAAGGTGGACGTCATACTCCATTCTTCACTAACTA	Enterococcus faecium

Table 2. Discoloration percentage of Methyl Red at different initial dye concentrations, temperatures and pH values by *E. faecium* and *Pantoea* sp.

Dye	The initial concentration (mg/L)	pН	25 °C		37 °C		50 °C	
2		-	Enterococcus	Pantoea	Enterococcus	Pantoea	Enterococcus	Pantoea
Methyl	100	4	4.3%	12.06%	4.3%	9.04%	0.00%	2.51%
Red		6	9.1%	26.02%	7.3%	28.31%	0.00%	0.00%
		8	43.99%	9.02%	1.34%	11.45%	0.00%	0.00%
	200	4	14.33%	16.57%	13.16%	26.28%	0.00	18.85%
		6	54.43%	49.22%	12.11%	45.87%	0.00	15.93%
		8	65.78%	49.74%	28.62%	42.21%	28.22%	0.00%
	300	4	0.00%	14.09%	0.00%	14.09%	0.00%	0.00%
		6	0.00%	9.55%	0.00%	9.55%	0.00%	0.00%
		8	0.00%	0.00%	0.00%	0.00%	0.00%	0.00%

Table 3. Discoloration percentage of Sudan Black at different initial dye concentrations, temperatures and pH values by *E. faecium* and *Pantoea* sp.

*								
Dye	The initial concentration (mg/L)	pН	25 °C		37 °C		50 °C	
			Enterococcus	Pantoea	Enterococcus	Pantoea	Enterococcus	Pantoea
Sudan	100	4	26.08%	0.00%	29.34%	0.00%	5.34%	0.00%
Black		6	28.88%	18.85%	37.03%	22.13%	0.00%	0.00%
		8	44.67%	35.29%	46.19%	39.70 %	19.79%	8.08%
	200	4	24.21%	0.00%	0.00%	0.00%	24.21%	0.00%
		6	49.96%	28.99%	0.00%	34.31%	32.10%	0.00%
		8	43.60%	13.18%	0.00%	18.13%	43.12%	0.00%
	300	4	7.3%	0.00%	7.3%	0.00%	7.1%	0.00%
		6	0.00%	0.00%	0.00%	0.00%	0.00%	0.00%
		8	43.25%	0.00%	23.41%	0.00%	0.00%	0.00%

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Table 4. Discoloration percentage of Malachite Green at different initial dye concentrations, temperatures and pH values by E. faecium a	ind
Pantoea sp.	

The initial concentration (mg/L)	pН	25 °C		37 °C		50 °C	
		Enterococcus	Pantoea	Enterococcus	Pantoea	Enterococcus	Pantoea
100	4	5.6%	0.00%	0.00%	0.00%	0.00%	0.00%
	6	22.33%	0.00%	26.21%	0.00%	2.9%	0.00%
	8	41.84%	0.00%	42.55%	0.00%	41.84%	0.00%
200	4	2.81%	8.6%	0.00%	10.75%	0.00%	7.52%
	6	0.00%	8.8%	0.00%	12%	0.00%	8.8%
	8	25.73%	71.53%	44.85%	18.13%	25%	0.00%
300	4	0.00%	7.05%	0.00%	9.4%	0.00%	9.4%
	6	0.00%	0.00%	0.00%	0.00%	0.00%	0.00%
	8	28.08%	73.87%	32.87%	73.87%	32.19%	0.00%
	The initial concentration (mg/L) 100 200 300	The initial concentration (mg/L)pH1004682004683004683004	$ \begin{array}{c} \mbox{The initial concentration} (mg/L) & \mbox{pH} & 25 \ \mbox{°C} \\ \hline \mbox{Enterococcus} \\ \mbox{Interococcus} \\ \mbox{100} & 4 & 5.6\% \\ \mbox{6} & 22.33\% \\ \mbox{8} & 41.84\% \\ \mbox{200} & 4 & 2.81\% \\ \mbox{6} & 0.00\% \\ \mbox{8} & 25.73\% \\ \mbox{300} & 4 & 0.00\% \\ \mbox{6} & 0.00\% \\ \mbox{8} & 28.08\% \\ \end{array} $	$ \begin{array}{c c} \mbox{The initial concentration} \\ \mbox{(mg/L)} \end{array} \begin{tabular}{c} \mbox{Pantoea} \\ \hline \mbox{Enterococcus} \\ \mbox{Pantoea} \\ \hline \mbox{Enterococcus} \\ \mbox{Pantoea} \\ \end{tabular} \\ \mbox{formula} $	$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	$ \begin{array}{c c c c c c c c c c c c c c c c c c c $	$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$

#### 3.1. Effect of pH on dye discoloration

The highest decolorizing percentage of Methyl Red and Malachite Green for both bacteria (i.e. *Pantoae* sp. and *E. faecium*) was attained at pH 8. As for Sudan Black, the highest discoloration was achieved at pH 8 for *Pantoea* sp. and at pH 6 for *E. faecium*.

#### 3.2. Effect of temperature on dye discoloration

The highest decolorizing percentage of Methyl Red for both bacteria was attained at 25°C, whereas for Sudan Black it occurred at 37°C for *Pantoea* sp. and at 25°C for *E. faecium*. As for Malachite Green, highest discoloration was achieved at 37 °C for *E. faecium* and at 25 °C and 37 °C for *Pantoea* sp.

# *3.3. Effect of initial dye concentration on dye discoloration*

Both bacteria showed the highest Methyl Red discoloration at a concentration of 200 mg/L. The highest Sudan Black discoloration was attained at 100 mg/L for *Pantoea* sp and at 200 mg/L for *E. faecium* whereas Malachite Green was highest decolorized at the concentrations of 200 mg/L and 300 mg/L for *E. faecium* and *Pantoea* sp., respectively.

## 3.4. Effect of bacteria type on dye discoloration

According to the results obtained, both *E. faecium* and *Pantoea* spp. showed high ability in dye discoloration at 25 and 37 °C, initial dye concentrations of 200-300 mg/L, and pH values from slightly acidic to neutral. But *E. faecium* decolorized Sudan Black by 19.79% at an initial dye concentration of 100 mg/L, pH=8 and 50 °C. It also had discoloration ability in the concentration of 200 mg/L at 50 °C and pH of 4, 6 and 8 to the extent of 24.21%, 32.10% and 43.12%, respectively. Methyl Red was decolorized at a temperature of 50 °C and pH=8 by 28.22% and Malachite Green was decolorized at three initial dye concentrations (100, 200 and 300 mg/L) at pH=8 and a temperature of 50 °C by 41.84%, 25% and 32.19%.

## 4. Discussion

Azo dyes are the largest group of synthetic dyes that are about 60-70% of all organic dyes in the world. These dyes have a great performance and are used in various industries such as textile, pharmaceutical and cosmetics industries, as well as food, paper, leather and painting industries. However, the negative aspects of these dyes and the risks may have for human health and the environment are not considered more and this may cause the disorder in nature in the not-too-distant future. Based on the findings of this study, the highest amount of discoloration was observed by *Pantoea* sp. bacterium on Malachite Green (73.87%) followed by *E. faecium* on Methyl Red (65.78%). The amount of discoloration on Sudan Black by *E. faecium* (49.96%) was higher than by *Pantoea* sp. (39.7%).

*Enterococcus* bacteria had the highest discoloration of Methyl Red at the concentration of 200 mg/L, the pH of 8 and 6, as the 65.78% and 54.43%, respectively. It had the highest discoloration on Sudan Black at the concentration of 200 mg/L, pH=6 to the 49.96%; and so, the discoloration of 44.85% was for Malachite Green at the concentration of 200 mg/L and pH=8. Overall, this study is consistent with other studies more closely and found that the experimental conditions of discoloration are almost identical for the isolated strains. The five dyes used in this study were isolated from the 19 strains, and two strains were not capable of decolorizing at experimental conditions including Rhodamine B and Brilliant Cresyl Blue in comparison with remaining three dyes of Methyl Red, Sudan Black and Malachite Green.

Adsorption and/or degradation are the two mechanisms responsible for dye decolorization by microorganisms (Ohadi *et al.*, 2020; Ghazvini *et al.*, 2016).

Due to the fact that the studies so far had wide variation in the type of dyes used for discoloration, the varying metabolic functions of the different bacterial isolates, and diversity of microbial species in the process of discoloration so the model described by other researchers may be different from each other but all showed a positive trend for these activities in laboratory and research scale processes. Mujtaba Ali and her colleague in 2014 revealed that the ability of discoloration on crystal violet occurred with better percentage by *Pseudomonas aeruginosa*, *Clostridium perfringens* and *Proteus vulgaris* in the presence of organic compounds compared with inorganic compounds (Ali and Akthar, 2014).

In the present study, the presence of organic compounds was considered in the wastewater by default. Despite the difference in the type of dyes used and the type of decolorizer microorganisms, the decolorizing capability of bacterial strains will not be inevitable. Aftab et al. performed a research in Pakistan in 2011 to study the ability of *Corynebacterium* sp. for discoloration and degradation of Reactive Black 5 and Yellow 15 dyes to the concentration of higher than 10 mg/ml and observed the growth of bacteria at 37 °C and pH=7. *Corynebacterium* indicated high azoreductase activity against Reactive Black 5 (68%) and Reactive Yellow 15 (80%) (Aftab *et al.*, 2011).

The amount of discoloration in any of the microorganisms tested during both studies reached 80 percent in special circumstances, which shows high degradation power on dyestuff by some microbes.

Discoloration performed at lower concentrations follows first-order kinetics and high concentrations have second-order kinetics. The calculated rate constants for lower concentrations show higher values. Evaluation of different results shows that different strains of *Pseudomonas* have various abilities for degradation of azo dyes in different concentrations which can be used based on the components and concentration of colors.

It is also the results of discoloration of azo dye of Acid Orange by Staphylococcus hominis RMLRT03 in the soil around the textile factories in Bushnell Haas medium (BHM) were showed that the bacterial strains were found by 16S rDNA sequence as Staphylococcus hominis. This strain of bacteria along with glucose supplement and yeast extract as an ancillary substrate shows good discoloration in terms of stillness. Optimal conditions for discoloration of Acid Orange by Staphylococcus hominis RMLRT03 was pH=7.0 and temperature of 35 °C in 60 hours' incubation. This bacterial strain can withstand high concentrations of Acid Orange dye to more than 600 mg/L. High discoloration activity under natural environmental conditions shows that bacterial strains have practical application in the treatment of wastewater containing dyes (Singh et al., 2014).

In the other study in India isolated *Pseudomonas* bacteria from soil contaminated quickly was decolorized the Methyl Orange azo dye solution. This bacterium has a remarkable discoloration in a wide range of concentrations of dye (50 to 200 mg/L) and pH (6-10) and temperature (30-40 °C). In addition, Pseudomonas spp. demonstrated discoloration of Methyl Orange for more than four cycles with high discoloration (10-94%) (Shah *et al.*, 2013).

Another study has performed to discoloration and decomposition of azo dye of Remazol Black B by new strains of Pseudomonas putida in India. Results showed that the pH=7.0 and temperature of 35 °C was optimal conditions for discoloration because the maximum discoloration has been observed only in these conditions. The amount of 5 g/L glucose in the culture medium also showed maximum discoloration. The new isolate has grown well in high concentrations of dye (300 mg/L) and has had 97.12% discoloration during 48 hours, and it also withstands concentrations over 1000 mg/L dye. P. putida colorless cells and UV-visible spectroscopy analysis suggested that discoloration activity of bio-decomposition is not just through disabling adsorption. The above results showed that this bacterial strain can be used in biological treatment of textile wastewater under optimal conditions (Kannan et al., 2013). Haidari-kashl et al. (2013) had performed a kinetic and comparative study on microbial degradation of azo dyes by P. aeruginosa and P. putida. In this study, four azo dyes including Acid Blue 113 (AB-

113), Basic Red 46 (BR-46), Direct Blue 151 (DB- 151), Direct Brown (DB-2) and a mixture of these four colors (Mix) were investigated for biodegradation by P. aeruginosa and P. putida at pH=7.2 and temperature of 30 °C. P. aeruginosa has fully decomposed AB-113 dye at all concentrations, BR-46 dye at concentrations of 0.1 and 0.2 g/L and DB-2 dye at concentrations of 0.1, 0.2 and 0.5 g/L. P. putida has completely decomposed the AB-113 dye at concentrations of 0.1 and 0.2 g/L and DB-2 dye in concentrations of 0.1 and 0.2 g/L. Also, a mixture of four dyes was degraded at a concentration of 0.1 g/L by P. putida. Discolorations performed at lower concentrations follow first-order kinetics and high concentrations follow second-order kinetics. The calculated rate constants for lower concentrations show higher values. Evaluation of different results showed that different of Pseudomonas strains have various abilities in decomposition of azo dyes in different concentrations which could be used based on components and concentrations of dyes (haidari kashl et al., 2013).

Kumar et al. (2012) have examined discoloration of Red 3BN azo dyes by *Bacillus cereus* and *Bacillus megaterium* in ZZ Medium in India. Physico-chemical parameters such as carbon sources, nitrogen sources, temperature, pH and volume of injection for discoloration process were optimized by changing one of these parameters at a time. Optimal conditions for *B. cereus* was 1% sucrose, 25% peptone, pH=7, 37 °C and 8% inoculation and for *B. megaterium* was 1% glucose, 0.25% yeast extract, pH=6, 37 °C and 10% inoculation. Enhancement of discoloration under the above conditions was 93.64% for *B. cereus* and 96.88% for *B. megaterium* (Kumar and Bhat Sumangala, 2012).

Regarding the wide range of dyes with different structures, wastewater can mainly contain highly variable combinations. Currently, conventional treatment systems to remove dyes are available which are primarily dependent on the chemical and physical principles. In these ways, a large amount of sludge is produced due to the use of high levels of chemicals, so dyes do not disappear completely, and this is administratively difficult and costly. Thus, new approaches of microbial discoloration are economically efficient; they are also very valuable in terms of contribution to the health of the environment. In the past two decades, it has been performed considerable works aimed at using microorganisms as cleaning agents for environmental pollutants from dyeing wastewater of textile mills. According to the results obtained and the ability of bacteria isolated from decomposition of dyestuff in wastewater optimization of conditions for the use of these microorganisms or the same microorganisms in biological purification in wastewater containing industrial dyes can be an effective help in this process and reuse of reversible waters. Therefore, applicable and targeted studies on strains such as Enterococcus, Pantoea or other microbial species can help in achieving this importance in the long run. Therefore, these studies highlight prospect for the authorities of economically feasible, environmentalfriendly, effective and worthwhile approaches for treatment of textile industry waste waters. The principal benefit to use of bacteria is that they are easy to culture and can grow more rapidly as compared to other microbes. Future study is expected to focus in this significant area of 224

microbial sciences to solve industry's environmental problems.

## 5. Conclusion

Our observations emphasizes that *Enterococcus and Pantoea* as bioremediation have a good efficiency to remove toxic and cationic Malachite Green and azo dye of Methyl Red. Bacteria can be used in the full-scale textile wastewater treatments as bioaugmentation, decolorization, degradation and effectively and economically treat non-biodegradable and toxic wastewaters. Nevertheless, more investigate is required to authenticate the process with particular interest.

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