Dye Effluent: Challenges And Opportunities

Ann Maxton, Sam A. Masih*

Molecular and Cellular Engineering, Sam Higginbottom University of Agriculture, Technology and Sciences, Prayagraj, India-211007

***Corresponding author:** Sam A. Masih
*Molecular and Cellular Engineering, Sam Higginbottom University of Agriculture, Technology and Sciences, Prayagraj, India-211007

Treatment of industrial effluents rich in toxic dye, phenolic compounds, heavy metals and toxic level of biological/chemical oxygen demand (BOD/COD) is a challenge for water bodies including aquatic fauna as currently available physio-chemical methods are questionable due to their by-product formation. Microbial based approach coupled with Bio-electrochemical system could be a game changer in this case as heavy metals and organic matters can be removed using this approach and toxic BOD/COD level can also be neutralized along with power output. Novel dye reduction-based electron-transfer activity monitoring (DREAM) assay, if coupled with bio-electrochemical system could be a game changer in this scenario.

Keywords: Azo dye, heavy metal, bio-electrochemical system, DREAM assay.

Introduction:

Rapid industrialization uplifts our comfort but made significant negative impact on environmental comfort. Textile industries release huge amount of waste water after their dyeing operations and it is estimated that around 20% dyes are lost in effluent during dyeing process (Baban et al., 2003). In general, dyes are made up of organic compounds with saturated/ unsaturated functional groups that are covalently attached to this including auxochrome and chromophore groups. The unsaturated groups/linkages (N=N, C=C or C≡C) are mainly accountable to absorb light in the visible range *i.e.* 350–750 nm, and show color. Chromophore groups are known as electron acceptor, responsible for color whereas auxochrome group (–SH, –OH, or NH_2) works as electron donor, mainly liable to enhance dye color. Microbial Fuel Cell (MFC) based on electron proton movement has emerged out as renewable and economical wastewater treatment based on microbial degradation approach that shows its ability in the treatment of toxic dye, organic matter and even several heavy metals as well along with improved COD removal rate presenting a solution in the current scenario. MFC operations with dye reduction based assay are closely related to unveil the process of electron liberation and microbial pili nanowire mediated electron transfer potential of industrial waste water sample

Problems: This contains some extensively toxic chemicals like azo dye and reactive dyes that makes serious impact on flora and fauna, results in ecosystem disturbance. The effluents released by the industries pose a major threat to the ecosystems as many azo dyes as well as their degradation products, for instance aromatic amines, are toxic, mutagenic and carcinogenic. Apart from this, dyes block sunlight to penetrate into water bodies results in dissolved oxygen reduction, a necessity for aquatic life (Garg and Tripathi, 2013) results in decreased aquatic fauna. Among the various dye classifications, azo dyes are the most commonly used industrially accounting up to 70% of total dyes consumption. In addition to their health hazards, the presence of the dyes in aquatic environment leads to poor aesthetic water quality and prevents sunlight from reaching into deeper water layers, which in turn reduces respiration and photosynthesis activities, consequently causing toxicity to aquatic life. Azo dye rich in several specific saturated (-SH, -OH) and unsaturated (N=N, C=C) functional groups that are responsible for absorbing light and acts ac electron donor/acceptor. Being highly soluble in water, Azo dye have high absorbing ability also even through skin touch and inhalation that may lead to mutagenic and carcinogenic effect (Mani and Bharagava, 2016). Chemical functional groups such as amide, carbonate, carboxyl, carbonyl, hydroxyl and phosphate, with dye binding capability, were found in the scales of certain fish species (Vieira et al., 2012). Additionally, it was reported that fish scales and their derivatives were effectively used to adsorb dyes. Para-phenylene diamine (PPD), an aromatic amine and a leading component in azo dyes have shown contact dermatitis skin irritation, chemosis and even permanent blindness (Aiyer et al., 2020).

Due to this nature of Azo dye, a suitable approach of treatment is necessary as currently in trends approaches (Physio-Chemical) utilize major amount of toxic materials results in toxic sludge production (Sudha et al., 2014). Therefore, a natural/biological treatment is necessary, several bacterial strains naturally secrets numerous metabolites that are able to convert toxic effluent into non/less toxic compound. Numerous researchers have shown aerobic and anaerobic bacterial species *viz., Bacilus subtilis, Escherichia coli, Xenophilus* sp*, Corneybaterium* sp*, Acinetobacter* sp*, Geobacillus,* *Rhizobium*, as degraders for azo dyes. Aerobic strains use azo dyes as major carbon and nitrogen source whereas another class reduce azo group with the help of special oxygen-tolerant azo reductases enzyme.

Table 1: Industrial waste water effluent with dyes

Industry	Dye	Effluent characteristics
Textile	Azo	Solid, phenols and sulfur with intense color due to various dyes including multiple toxic heavy metals like Cadmium, Chromium, Lead Nickel *etc.* amended with Phosphorus and Potassium minerals, higher level of BOD and COD.
Paper	Vat dyes, cationic direct dyes and sulfur dyes	Dark brown color, increased BOD, COD, TDS level because of lignin, tannins, resin acids, chlorinated compounds, sulfur or its compounds.
Leather	Azo	Phenols, tannins, Chromium and toxic level of COD and BOD
Tannery	Acid/Basic dyes, methyl orange	Azo, Chlorophenols, Phenols, tannins with several heavy metals like Cadmium, Chromium, Lead, High level of BOD and COD.

Ray of hope: Azo linkages can be reduced under anaerobic environment results in colorless aromatic amines and can be degraded in aerobic conditions. So, a combined (anaerobic and aerobic) approach is necessary (Khalid et al., 2008) for the mineralization of azo compounds that may be obtained from mixed microbial culture may be due to several hidden mechanisms like electron transportation and biofilm formation. It was observed that when bacteria reacts with azo, azo reductase enzyme secrets that break azo linkage (N=N) and release aromatic amines (re-calcitrant) that further breakdown into several intermediate metabolites while release H_2O and CO_2. Bioremediation ability of microorganisms may be enhanced gradually by exposing them to increased concentrated effluents that will develop adaptation toward recalcitrant coloring /toxic compounds *i.e.* acclimatization and leads to directed/forced evolution. This type of evolution happens via expression of gene(s) encodes for enzymes that are majorly responsible towards bioremediation and alternatively development of engineered microbes with the ability to transfer genes encoding for bioremediation enzymes with improved decolorization capabilities.

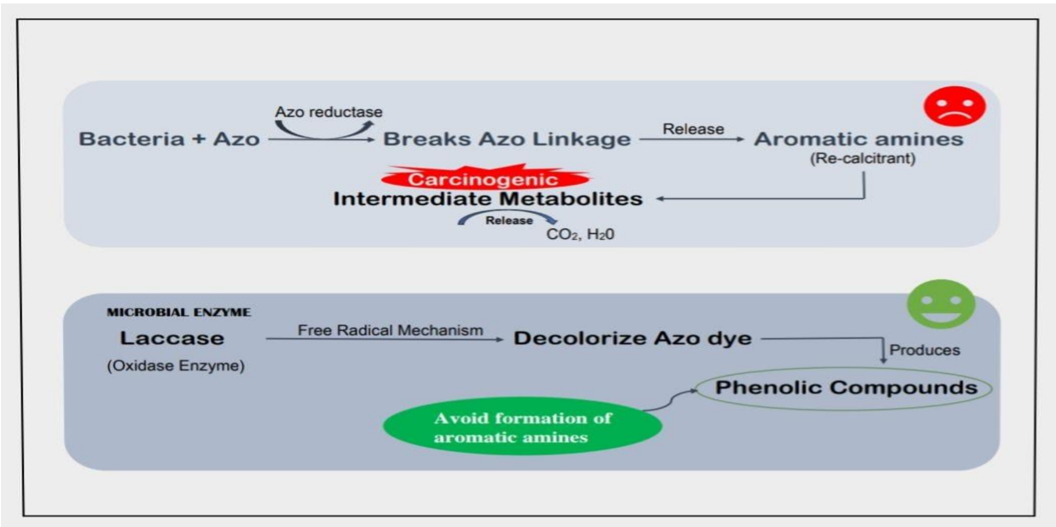

Fig. 1: Mechanism for microbial degradation of Azo dye

Thus azo reductase enzyme based approach is generally being used in microbial based waste water treatment system (Bio-electrochemical system) but produced amines are carcinogenic (fig. 1). Another approach could be usage of Laccase, a microbial enzyme that is able to decolorize azo dye involving non-specific free radical mechanism results in the formation of phenolic compound thus avoid the formation of toxic aromatic amines (Naik and Singh, 2012). Few bacterial species like *Trametes versicolor, Polyporus pinisitus, Penicillium chrysogenum* and *Phlebia radiate* (Tian et al., 2014) have been reported with this ability.

If currently used Azo reductase enzyme based approach can be coupled with Bio-electrochemical based approach *i.e.* Microbial Fuel Cell as chemophore and auxochrome work as electron acceptor and donor respectively. In our recent study, we have adopted a novel dye reduction-based electron-transfer activity monitoring (DREAM), Methylene Blue reduction method with textile waste water and noticed exciting results where textile effluent worked as analyte during MFC operation for electro liberation results in power generation with simultaneous reduction in BoD/COD and color (Masih and Devasahayam, 2015). This method relies on electrons liberating principle from available chemical bonds and transportation via electron transport chain to electron acceptor upon organic matter degradation *i.e*., mediated by microbial degradation (Aiyer et al., 2020). Methylene blue, an Azo dye take up liberated electrons and undergoes reduction reaction reactions results in leuco-methylene blue, a colorless dye. Rate of this decolorization majorly depends upon the quantity of supplied electron (that is high in industrial effluent due to involvement of mixed microbial community) in a concerned time frame that is being measured by the rate of decrease in blue color intensity that may be enhanced by improving design of MFC including separating membrane and electrode potential. We have used dye reduction based DREAM assay in our lab scale study (data unpublished) to analyze microbial electron transfer in expeditious way as we concluded from our previous researches that microbial growth rate and their viability are mainly responsible for electrons supply through breaking down of available carbon sources that helps to select proper acolyte for MFC operation. This assay will help to analyze electron transfer ability of microbial culture in waste water, results in DREAM assay coefficient that depicts ability and suitability of electron transfer ability of microbial culture by reducing Methylene Blue into colorless solution. These results strongly emphasized significance of electrogenic microbial community available in industrial waste water to be used in Microbial Fuel Cell that could be primarily checked using DREAM assay.

Conclusion:

Bioremediation, an inexpensive and eco-friendly technology for decolorization of dyes using available mix consortium/specific microbes in MFC setup along with power output is a suitable approach. DREAM coefficient directly correlates with viable cell count, microbial activity and ability of organic matter for computing the electron transfer potential of industrial waste water sample to harness optimal energy output when coupled with Microbial Fuel Cell assembly. So, if a hand held colorimeter can be taken at site of industrial waste water source, decolorization of methylene blue can give an instant idea for further usage with fuel cell that majorly depends upon electrogenic microbial population. Future research approaches in decolorization should be placed on the search of improvement and optimization of culture and conditions, complete understanding of mechanism for improved commercialization of environmental bioremediation including aquatic fauna.

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