DECOLORIZATION OF REACTIVE BLACK 5 BY PURE CULTURE OF IE1, C1 AND THEIR MIXTURE

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ABSTRACT

Release of colored textile effluents is undesirable in the aquatic environment as they reduce light penetration, thereby affecting aquatic life and limits utilization of the water media. Microbial bioremediation is an alternative treatment option available other than the commonly employed physicochemical and biological methods to treat these toxic effluents. Two unidentified bacterial cultures were selected for investigation of their ability to degrade RB5 dye in liquid medium. The cultures showed best decolorization under aerobic and shaking conditions at pH 7 and room temperature (28±2°C). The individual cultures of IE1 and C1 exhibited 71.74% and 73.34% respectively whereas mixture of both showed 89.56% decolorization within 6 h at a dye concentration of 200mg/l, while at higher dye concentration (1000mg/l and 5000mg/l) the decolorization was decreased. The mixture of two culture exhibited decolorization ability (34.83% - 95.24%) over a wide pH range (pH 4.0 to 10.0) compared to individual culture. However, the removal of peptone from the medium inhibited the decolorization activity of the individual cultures whereas their mixture exhibited 15% decolorization of Reactive Black 5 dye. The microbial decolorization of dyes in an indigenously designed fixed film bioreactor showed almost 88% decolorization for individual cultures while 98.34% for their mixture within 48h.

Key Words: Azo dye, Bacteria, Decolorization, Bioreactor, Temperature

INTRODUCTION

Azo dyes are most widely used synthetic colorants. More than 2000 structurally different azo dyes are currently in use1. They have been increasingly used (more than 1 million tonnes worldwide) because of the ease and cost effectiveness of their synthesis, their stability and the variety of colors available in comparison to natural dyes2. Azo dyes are extensively used in the textile industry, but due to inefficiencies of the industrial dyeing process, 10-15% of the used dyes are lost in the effluent of textile units, rendering them highly colored3. It is estimated that 280,000 forms of textile dyes are discharged in industrial effluents every year worldwide4. Several abiotic techniques have been proposed for textile wastewater treatment. They include adsorption, oxidation, precipitation, sonochemical decomposition, photodegradation, bleaching with oxidizing agents and even membrane filtration.5 But when these treatments are compared with that of the biological methods, it is evidenced that though these physical treatments are faster, they are less effective and costly. As an alternative and an eco-friendly technique, azo dyes can be reduced by using special bacteria that have the ability of breaking the azo bonds under low redox potential under oxygen deficient conditions thus forming the corresponding aromatic amines. Successful work in the past decade on biological methods has included the use of single strains, mixed cultures and microbial consortia in the decolorization.6

AIMS AND OBJECTIVES

To find out the capability of previously isolated cultures IE1 and C1 in their pure form and mixture to decolorize the sulfonated azo dye Reactive Black 5.

The present study reports that previously isolated two cultures IE1 and C1 from textile industry effluent were found to be highly effective for decolorization of RB5 dye individually and also in a mixture (consortium)
giving more than 90% decolorization within a span of 6 to 48h.

**MATERIAL AND METHODS**

**Chemicals and culture media**

The commercial textile dye Reactive Black 5 used during this study was obtained from local market Mumbai. All other chemicals used were of analytical grade procured from HiMedia Pvt. Ltd., Mumbai, India.

**Bacterial cultures IE1 and C1**

In the present investigation, two previously screened bacterial cultures (IE1 and C1) from textile industry effluent giving best decolorization activity were maintained on NA slants incorporated with dye at 4°C in Refrigerator.

**Determination of color reduction**

The color reduction was determined by means of a Schimadzu UV/Visible spectrophotometer (Model 1800) by optical density measurement at the wavelength of 620 nm ($\lambda_{max}$ of RB5). The decolorization efficiency was calculated according to the equation:

$$% \text{ decolorization} = \frac{(A-B)}{A} \times 100$$

Where,  

$A = A_{620}$ of culture filtrate  
$B = A_{620}$ of control

The statistical analysis of decolorization of RB5 was done.

**Decolorization assay under static and shaking conditions**

For decolorization assay RB5 dye 200 mg/l was incorporated in Erlenmeyer flasks (three sets), each containing 50 ml sterile nutrient broth. Actively growing culture 1% (v/v) ($10^8$ cells ml$^{-1}$) was added in two sets. Among these two sets, one set was incubated under static condition and other on shaker (100 rpm) at room temperature (28±2°C) for 24h. The third set served as a control without adding inoculum. Aliquots were withdrawn after every 24h from both the sets and centrifuged separately at 3000rpm for 30min. The cell free supernatant was then used for measurement of % decolorization as stated above. This experiment was continued till maximum decolorization was achieved.

**Decolorization under aerobic/anaerobic conditions**

RB5 dye (at a concentration of 200mg/l) was incorporated in 50 ml sterile nutrient broth in tube. For anaerobic process the content of the tube was boiled and liquid paraffin was layered on it. Then the tubes were inoculated with 1% (v/v) inoculum and sealed with wax whereas for aerobic process flask was inoculated and incubated on shaker maintained at room temperature (28±2°C) for 24h. No inoculum was added in control tubes and flasks. Aliquots were withdrawn after 24h, centrifuged at 3000rpm for 30 min. and the amount of dye decolorization was calculated.

**Influence of different physico-chemical factors on decolorization**

The effect of various physico-chemical parameters such as temperature [20°C, room temperature (28±2°C), 37°C and 40°C], pH (4, 5, 6, 7, 8, 9 and 10), peptone concentration in nutrient broth medium (0, 2, 4, 6, 8, 10 g/l) on dye decolorization were examined. The uninoculated controls were also incubated under the same conditions. After 24h the % decolorization was calculated using standard method as stated above.

**Correlation between growth and dye decolorization**

Nutrient broth (50ml) in side-arm flasks was inoculated with all three cultures (IE1, C1 and mixture) separately and incubated on shaker at 28±2°C. Growth and reduction in color was measured respectively at 600nm and 620nm after every hour.

**Decolorization of dyes by growing cells**

This decolorization experiments were performed in triplicate. The microbial cultures under investigation (IE1, C1 and mixture) were first cultivated aerobically for 24h in Erlenmeyer flasks containing 100ml NB devoid of dye. After 24h RB5 dye was added at a concentration of 200, 1000 and 5000mg/l in three different flasks respectively for each culture and consortium. The inoculated flasks were incubated on rotary shaker at room temperature. After defined time period, aliquot (5ml) of the culture media was withdrawn, centrifuged at 3000xg for 30min. to
separate the bacterial cell mass and supernatant. The % decolorization was measured spectrophotometrically and the rate of decolorization was calculated.

**Microbial decolorization of dyes in indigenously fabricated fixed film bioreactor**

To improve decolorization of dye a bioreactor was designed in glass column. The 3/4th of bioreactor was filled with gravels. Thus, it is known gravel bed reactor. The bioreactor was closed using a rubber stopper at one end with a hole through which a pipe was inserted for the transfer of media in the reactor. The other end (bottom) of the bioreactor was attached to a reservoir for the collection of treated sample. The bioreactor and gravels were dry heat sterilized. Then, it was filled with NB and culture (IE1, C1 and mixture) and allowed to stand for 48h for formation of biofilm on gravels. After 48h the media was drained off and the inflow of dye (RB5) solution (200mg/l) was continued till complete decolorization was obtained. Absorbance of both inflow and outflow were noted as mentioned in previous section. Results are average mean of three replicates for each experiment.

### RESULT AND DISCUSSION

#### Decolorization under static and shaking conditions

The pattern for dye decolorization in static as well as in shaking conditions is elucidated in Table 1. In that culture C1 decolorized dye at minimum in static condition as compared to shaking (73.63 and 78.41%), whereas the culture IE1 and mixture showed no significant difference in dye decolorization under static as well as shaking conditions (73.63 and 73.63% and 93.17 and 93.60%).

<table>
<thead>
<tr>
<th>Condition</th>
<th>IE1</th>
<th>C1</th>
<th>Mixture</th>
</tr>
</thead>
<tbody>
<tr>
<td>Static</td>
<td>73.63</td>
<td>73.36</td>
<td>93.17</td>
</tr>
<tr>
<td>Shaking</td>
<td>73.63</td>
<td>78.41</td>
<td>93.6</td>
</tr>
</tbody>
</table>

Results are indicative of the fact that C1 individually and in mixture with IE1 could decolorize dye more efficiently under shaking conditions which contradicts the findings of 8. Some of the laboratories9-13 reported less or no decolorization under shaking condition, exactly opposite to the results obtained in present study. 14 stated that the color removal of textile dye effluent was more effective under shaking conditions than under static condition. 15Kodam et al., reported that the color removal of textile dye effluent was more effective under shaking conditions than under static condition. According to 16 maximum color removal was observed in shaking cultures because of better oxygen transfer and nutrient distribution as compared to the stationary cultures. 17 Investigated the effect of static and shaking conditions and found that the decolorization activity remains unaltered under both conditions, similar to the results obtained in our laboratory for culture IE1 and the mixture of IE1 and C1 during this investigation.

#### Decolorization under aerobic / anaerobic conditions

Many microorganisms are able to decolorize azo dyes aerobically as well as anaerobically. Bacteria usually degrade azo dyes under anaerobic conditions to colorless amines of which, some are readily metabolized under aerobic conditions17. The aerobic microorganisms further oxidize the reduced products via deamination or hydroxylation18. The results depicted in Table 2, reveals that both cultures alone and mixture found to degrade dye efficiently under aerobic conditions but showed decolorization upto 40% of RB5 even under anaerobic condition. In that mixture of IE1 and C1 decolorized dye more than 90% under aerobic condition. Most of the research work on azo dyes reveals that biological degradations of azo dyes are efficiently carried out under anaerobic condition. Generally, azo dyes are resistant to attack by bacteria under aerobic conditions. Only few studies reported that bacteria could also degrade azo dyes under aerobic conditions.
Table 2: Decolorization of reactive black 5 (200 mg l\(^{-1}\)) under aerobic/anaerobic conditions

<table>
<thead>
<tr>
<th>Incubation condition</th>
<th>% dye decolorization</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>IE1</td>
</tr>
<tr>
<td>Aerobic</td>
<td>73.63</td>
</tr>
<tr>
<td>Anaerobic</td>
<td>37.27</td>
</tr>
</tbody>
</table>

It is revealed from our results that both the cultures in pure form and in mixture is able to decolorize RB5 in range of 73.63 to 93.6\% under aerobic conditions as compared to 27.66 to 39.92\% in anaerobic condition. It is clear from literature that in both the cases (aerobic and anaerobic conditions) the initial step in the biodegradation is the cleavage of the azo bond\(^{19}\). Under aerobic conditions, the initial step of azo bond cleavage is typically followed by hydroxylation and ring opening of the aromatic intermediates\(^{20}\). Azo dyes are not readily metabolized under aerobic conditions although Kulla reported the ability of *Pseudomonas* strains to aerobically degrade certain azo dyes which is similar to our findings\(^{22}\). The permeability to the dye and presence of azoreductase affected reduction of azo dye by pure culture of *P. stutzeri*, *B. subtilis* and *P. cepacia*, under aerobic condition. Thus, the decolorization obtained under aerobic conditions, which is in contrast to other reports\(^{23,24}\).

**Influence of different physico-chemical factors on decolorization**

Industrial effluent is never stable and it varies often in a wide range of pH and temperature depending upon the process practiced. The concentration of dye varies in the effluent of textile industry and that again depends upon the class of dye used for dyeing. Adaptation of microorganisms to wide range of pH, temperatures and varying dye concentration makes them more suitable for the degradation of industrial dye containing effluents.

**pH**

For any biological process pH is an important parameter hence, it needs to be maintained within a range to keep biological system active. The results obtained for effect of pH on dye decolorization are as per Fig. 1.

![Fig. 1: Influence of pH on decolorization of RB5 (200 mg l\(^{-1}\))](image)

As there was increase in pH from 4 to 7 by 3.0 units there was 84\% increase in decolorization of RB5 dye for culture C1 whereas for culture IE1 decolorization was increased by 75\% and for mixture the increase was 55\%. Further, rise in pH from 7 to 10 the marked reduction in % decolorization was observed. A gradual decrease in decolorization activity was observed before and after pH 7 hence, pH 7 is considered as an optimum pH giving maximum decolorization of RB5 for both organisms separately as well as for their
mixture. Similar results were recorded earlier with *Enterobacter* sp. azol, where decolorization was found to be highest at pH 7 after which (pH 8-9) the activity decreased. Mali et al., found that pH between 6.0 - 8.0 as optimum for decolorization for triphenylmethane and azo dyes by *Pseudomonas* sp. Saratale et al., reported that over the pH range of 5.0–8.0, the consortium GR achieved the highest decolorization activity for Green HE4BD at pH 8.0, while the decolorization rate decreased at lower pH (5.0–7.0) and higher pH (10–12). It is thought that the pH effect may be more likely related to the transport of dye molecules across the cell membrane, which was considered as the rate limiting step for decolorization. In the same way, the mixture of IE1 and C1 was able to decolorize RB5 over a wide range of pH from 4 to 10 with % decolorization between 34.83 to 86.33% and showing highest decolorization (95.24%) at pH 7. Wang et al., studied decolorization of Reactive Black 5 by a bacterial strain *Enterobacter* sp. EC3. According to their results, *Enterobacter* sp. EC3 showed a high decolorization rate at pH 7.0 after 108 h of incubation. Similar decolorization efficiency was observed from pH 8.0–12.0 in 120 h, whereas the rate of color removal was much lower at acidic conditions (pH 4.0 and 6.0). Kumara et al., found the maximum removal (96 %) of Remazol Black B dye at pH 7.0 and 8.0 after 30 hours of incubation. Further increase in pH beyond 8.0 and decrease in pH below 7.0 resulted in decreased removal of dye. On either side of pH optima, percentage decolourisation decreased as the pH increased to acidic or alkaline range.

**Temperature**

Temperature is an important environmental factor and the biodegradation activities of microorganisms are affected by changes in temperature. Beyond the optimum temperature, the degradation activities of the microorganisms decrease because of slower growth, reproduction and deactivation of enzymes responsible for degradation. Thus, the biodegradation performance of microorganisms will be best at the optimum temperature needed for their growth, reproduction and biochemical activities. Many dye-house effluents are discharged from plants at a temperature well in excess of ambient temperature. Therefore, the ability of IE1, C1 and their mixture to decolorize RB5 at different temperatures [20°C, room temperature (28±2°C) 37°C and 40°C] was studied and results are depicted in Fig. 2.

![Fig. 2: Influence of temperature on decolorization of RB5 (200 mg l⁻¹)](image)

From the results it can be said that the optimum temperature for decolorization is room temperature (28±2°C) for both the cultures in pure form (IE1- 94%, C1-79.26%) and in mixture (77.48%). The culture IE1 was able to decolorize RB5 33.8% and 19.2%, culture C1 from 46.03 to 18.69% and their mixture from 37.98 to 21.65% at 20°C and
A decline in decolorization activity beyond the optimum temperature can be attributed to the thermal deactivation of the decolorizing enzymes and the low biomass\textsuperscript{18,29}. A halophilic organism \textit{Halomonas} sp. strain GTW showed highest decolorization at 30°C (room temperature) as reported by\textsuperscript{30}. These results were similar to our observations. However, in contrast to our finding\textsuperscript{31} Wong and Yuen reported that \textit{Klebsiella pneumoniae} RS-13 exhibited decolorization of methyl red at temperatures varying from 23 to 37°C, whereas, at 45°C decolorization was completely inhibited. Maximum potential of \textit{Pseudomonas} sp. to decolorize Malachite green, Fast green, Brilliant green, Congo red and Methylene blue was noticed at 37°C.\textsuperscript{32} Wang et al., found that, with an increase in temperature from 22°C to 37°C, the decolorization rate increased and a further increase in temperature to 42°C drastically affected decolorization activity of \textit{Enterobacter} sp. EC3. They found 37°C as an optimum temperature for decolorization. The results of present study are in contrast to this because the optimum temperature for Reactive Black 5 decolorization was found to be 28±2°C (room temperature). The reason may be that the optimum temperature required for the growth and decolorization activity may vary with the type of organism. Similarly, Kumara et al., reported that the percent removal of Remazol Black B dye by mixed culture increased with an increase in temperature from 20 to 30°C. The percent removal of dye decreased with further increase in temperature up to 45°C. Decolorizing activity was significantly suppressed at 45°C this might be due to the loss of cell viability or deactivation of the enzymes responsible for decolorization at 45°C. Above results indicate that decolorization is influenced by temperature as it affects both growth and metabolic activities of the organism.

**Peptone concentration**

Peptone is used as a complex component of bacteriological media, used as a source of carbon and nitrogen. Normally, in nutrient broth 1% of peptone is added. During this research work the decolorization activity has been tested with varying concentrations of peptone (0, 0.2, 0.4, 0.6, 0.8 and 1%). The results showed in Fig. 3 which states that for decolorization activity of both the organisms the presence of peptone is essential. This indicates that for decolorization activity organisms require co-substrate, they cannot use dye as the sole source of carbon and nitrogen.

![Fig. 3](image-url) : Influence of peptone concentration on decolorization of RB5 (200 mg.l\textsuperscript{-1})

The results obtained were similar to the observations reported by Jirasripongpun et al., that \textit{Enterobacter} sp. azol could not metabolize the dye as sole carbon and energy sources and require co-substrate for decolorization. It was able to decolorize dye only in the presence of either glucose or peptone. Similarly, omitting both carbon and nitrogen sources resulted in
57.6% decrease in decolorization efficiency compared to control. According to Jang et al, the addition of peptone had no significant effect on color removal of dye effluent by Citrobacter sp. strain KCTC 18061P. These results were in contrast with our findings.

**Influence of repeated addition of inoculum**

The influence of increased addition of inoculum was tested to check the efficacy of cultures to degrade dyes over a long period of time. An increase in the decolorization activity was observed after every 24h addition of inoculum. The decolorization activity of all the cultures (IE1, C1 and their mixture) reached more than 90% in 48h. The results obtained are depicted in Fig. 4. Physiological differences among the cultures and the decoloration enzymes may account for differences in discoloration ability,

Fig. 4: Influence of repeated addition of inoculum on decolorization of RB5 (200 mg/l)

These results are indicative of the fact that live cells are decolorizing the dye efficiently and with an increasing cell number, decolorization was also found get increased.

**Influence of repeated addition of dye**

The intention of this study was to check the ability of organisms for the repeated decolorization of dye at 200 mg/l concentration of Reactive Black 5. The results obtained are shown in Fig. 5. The decolorization ability of the cultures IE1, C1 and consortium decreased with repeated addition of dye. The eventual cessation of decolorization activity may be due to nutrient depletion. The other possible reason could be that, the saturation of the cells with dyes will result in inhibition of azoreductase activity and accumulation of toxic metabolites. This may lead to low activity of azoreductase and as a result it may cause toxicity to bacterial cells and there is low or no decolorization of azo dyes. In a study carried out by Telke et al., on decolorization of dye by Rhizobium radiobacter MTCC 8161 decolorized the repeated addition of Reactive Red 141 dye up to six cycles with variable decolorization rate (74–90%), whereas the mixed bacterial culture decolorized the azo dye up to three cycles. In first cycle 90% decolorization of first dye aliquot required 48h, 79% decolorization within 12 h in second and third cycles, the percent decolorization (74%) and time (6 h) remains constant up to last cycle. In another study, upon 93% decolorization of first dye aliquot within 40h, a second aliquot of dye was added, which was decolorized (94%) within next 24h. The culture caused 68% decolorization of a third aliquot, which was subsequently added, within next 24h of addition Moosvi et al. The pattern of results obtained in the present study corroborates with the results of Moosvi et al., which showed decrease in ability of decolorization after every 24h. The repeated addition of dyes is an important aspect to be observed and studied from industrial point of view.
Correlation between growth and dye decolorization

Growth of the organism was compared with the amount of decolorization in growth curve experiment after every hour. This experiment draws a relationship between growth of organism and its effect on dye decolorization. The results are shown in Table 3. The maximum decolorization activity was observed in exponential phase of growth. At the beginning of log phase the decolorization activity is very low thereafter it got slowly increased till the end of exponential phase. This phenomenon of low decolorization activity may be reasoned with the explanation that the cells need some time to get adapted to the dye environment, once the dividing cells are adapted they show increase in decolorization with growth. The pattern of decolorization observed with culture IE1 when studied with Reactive Black 5 was different from C1.

Table 3: Correlation between growth and decolorization

<table>
<thead>
<tr>
<th>Time in hour</th>
<th>Growth at 600 nm</th>
<th>% decolorization</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>IE1</td>
<td>C1</td>
</tr>
<tr>
<td>0</td>
<td>0.04</td>
<td>0.05</td>
</tr>
<tr>
<td>1</td>
<td>0.065</td>
<td>0.04</td>
</tr>
<tr>
<td>2</td>
<td>0.09</td>
<td>0.05</td>
</tr>
<tr>
<td>3</td>
<td>0.11</td>
<td>0.06</td>
</tr>
<tr>
<td>4</td>
<td>0.23</td>
<td>0.2</td>
</tr>
<tr>
<td>5</td>
<td>0.39</td>
<td>0.4</td>
</tr>
</tbody>
</table>

Bacillus subtilis HM showed color removal within the first 6h, during the exponential growth of the bacterium, whereas the remaining color was removed during the stationary phase of growth. However, in this study maximum dye removal was observed in exponential phase of growth. The decolorization was faster during the exponential growth phase of the microorganism slowed down in the stationary phase. This is not unexpected, since the process must be dependent on cell biomass and on actively growing cells. A study on Enterobacter sp. showed an initial rise in decolorization activity followed by fall before reaching stationary phase.

Decolorization of dyes by growing cells

Decolorization of Reactive Black 5 was observed by the bacterium, IE1, C1 and consortium. The percent decolorization of Reactive Black 5 dye at three different concentrations (200, 1000 and 5000mgl⁻¹) is shown in Fig. 6(a) to Fig. 6(c). All the cultures were grown in NB medium for 24h and then dye was added to culture medium. All the cultures showed a gradual increase in
decolorization activity with increase in time. This can be noted that dye has no toxic effect on cell growth. However, with culture IE1 at 6h a decrease in decolorization activity was observed. This may be attributed due to the release of some toxic by-products as a result of dye decolorization which affected the cell growth and hence fall in decolorization while after 24h an increase in decolorization ability was observed which implies that toxic by-products were metabolized later that resumed the decolorization activity.

Fig. 6(a) : Decolorization of Reactive Black 5dye at three different concentrations (200, 1000 and 5000mg/l) by culture IE1

Fig. 6(b) : Decolorization of Reactive Black 5dye at three different concentrations (200, 1000 and 5000mg/l) by culture C1

Fig. 6(c) : Decolorization of Reactive Black 5dye at three different concentrations (200, 1000 and 5000mg/l) by mixture of IE1 and C1
These results were compared with the previous experiment of growth curve which also proves that the rate of decolorization increases with increase in growing cells. One more phenomenon observed in this study was at 6th hour of growth the cells on centrifugation showed color, this is because of adsorption of dye on the cells whereas at 24th hour complete colorless cells were seen. It is known that biomass adsorption is effective when conditions are not always favorable for the growth and maintenance of the microbial population, because the use of biomass has its advantages, especially if the dye-containing effluent is very toxic.

In a similar experiment carried out by Kodam et al., an increase in decolorization activity with time and almost 100% decolorization was achieved in a short duration (36 h). The increase in pH towards the alkaline range with time may be attributed to the accumulation of the basic aromatic amines and/or other metabolites. Similar results were observed in this experiment which showed an increase in pH with time from pH 7 to 7.63. Thus the results obtained by both growth curve and growing cells say that biomass is directly proportional to the decolorization rate.

**Microbial decolorization of dyes in an indigenously fabricated fixed film bioreactor**

Fixed film bioreactor consisted of gravels, which were used to form biofilm formation. The gravel bioreactor used was of non-aerated. The results are shown in Fig. 7. More than 85% decolorization was observed with cultures IE1 and C1, whereas consortium exhibited 98% of decolorization within 48h. Fox et al., 1990 studied the effect of media types on the development of the biofilm and found that the media with a greater rough surface area provides better attachment to biofilm.

![Fig. 7 : Microbial decolorization of RB5 dye in an indigenously fabricated fixed film bioreactor](image)

Kumar et al., studied the decolorization rate of methyl red in fixed film bioreactor and reported that the conductivity values of mixed bed outflow were higher than gravel bed and of non-aerated than that of the aerated bioreactor. However, the COD load was equal in gravel bed and mixed bed outflow when fed with dye. In another study carried out by Sharma et al., used an immobilized cell bioreactor made up of pieces of refractory bricks. The concerted metabolic potential of the immobilized cells led to more than 94% decolorization of Acid Blue-15. The decolorization of the textile dye effluents was made easy with the help of Immobilized Packed Bed Bioreactor (IPBB). The control of pollutants was achieved in a great fashion through immobilization. The highlight of the immobilized bed is its reuse. It was stable towards reuse and can be used for both batch and semi batch process. The results in the present study showed that the developed upflow immobilized cell bioreactor can enhance the ability of the microbial mixture to decolorize and degrade industrially important textile dyes efficiently.

**CONCLUSION**

The pure culture of IE1 and C1 and mixture of it can decolorize it very efficiently under...
aerobic condition at room temperature (28±2°C) and over a wide range of pH. The dye removal was found to be growth linked process with maximum dye decolorization taking place in logarithmic phase of growth and all cultures need co-substrate for dye removal. However, in an indigenously fabricated bioreactor mixture of two cultures has given more than 98% decolorization within 48h. Hence, for removal of a textile sulfonated reactive azo dyes pure bacterial cultures IE1, C1 and their mixture can be used very effectively.

REFERENCES


