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DECOLORIZATION OF TEXTILE DYES BY NEWLY ISOLATED TRAMETES VERSICOLOR STRAIN

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ABSTRACT

In this century, the amount of industrial produces and their consumption has increased tremendously. Along with this increase, accumulation of industrial waste and its effect on nature has caused serious problems. Because of including various chemicals and especially dye, textile waste water is one of the most hazardous industrial wastes. Color is the most important pollutant in waste water and it should be decolorized. Decolorization is more important than degradation of organic substances from waste water. Even a small amount of dye found changes the color of rivers, lakes and other water resources, and reduces the penetration of light and solubility of gases.White rot fungus are used as a biological system in degradation and decolorizing of textile dyes.

In this study, parameters for decolorization of several textile dyes (Blue 49, Orange 12, Orange 13, Red 31, Black 5, RBBR) by newly isolated *Trametes versicolor* M96was studied.

It has been determined that pH, amount of inoculum, shaking speed (rpm), dye concentration and temperature are important factors in decolorization of the studied dyes. The maximum decolorization was found pH 4.5, amount of inoculums 50 ml, shaking speed 200 rpm, dye concentration 50 mg/l and heat 30 °C.

Keywords: White rot fungi, *Trametes versicolor*, Decolorization, Textile dyes.

YENİ İZOLAT *TRAMETES VERSİCOLOR* İLE TEKSTİL BOYARMADDELERİN RENGİNİN GİDERİMİ

ÖZ

Çağımızda endüstri ürünlerinin üretimi ve tüketimi hızla artmıştır. Bunun yanında oluşan endüstriyel atıkların birikimi ve bunların doğa üzerine olan etkileri ciddi problemlere yol açmaktadır. Tekstil atık suları içerdikleri çok değişik kimyasallardan ve özellikle de boyar maddelerden dolayı arıtılması zor olan endüstriyel atık sulardan birisidir. Renk atıksu içerisindeki en önemli kirleticidir ve bu ortamlara ulaşmadan önce mutlaksuretle renginin giderilmesi gerekir. Atık sudan rengin giderimi çözünmüş organik maddelerin gideriminden daha fazla önemlidir. Çünkü suda çok az miktarda bile boya bulunması rengi arttırır ve nehirlerin, göllerin ve diğer su kaynaklarının ışık geçirgenliğini ve gazların çözünürlüğünü etkiler. Beyaz çürükçül funguslar boyarmaddelerin yıkımı ve renginin giderimi çalışmalarında biyolojik sistem olarak kullanılmaktadır.

Bu çalışmada yeni izole edilen *Trametes versicolor* M96 suşu kullanılarak çeşitli tekstil boyalarının (Blue 49, Orange 12, Orange 13, Red 31, Black 5, RBBR) renk giderimini etkileyen parametreler araştırılmıştır. Çalışılan boyaların renk giderimi için pH, inokulum miktarı, çalkalama hızı,

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boya konsantrasyonu ve sıcaklık gibi faktörlerin önemli olduğu belirlenmiştir. Maksimum renk giderimi; pH 4.5 , inokulum miktarı 50 ml, çalkalama hızı 200 rpm, boya konsantrasyonu 50 mg/l ve sıcaklık 30 °C'bulunmuştur.

Anahtar Kelimeler: Beyaz çürükçül fungus, *Trametes versicolor,* Renk giderimi, Tekstil boyası.

1. INTRODUCTION

Dye is used widely in colorization and printing processes in industry. Although colors extracted from various herbal roots were used to textile and dye fabrics in the past, nowadays synthetic chemical dyes are used in the textile industry by economical reasons. Approximately $7x10^5$ tons, and about 10.000 different colors and dyestuff are produced throughout the world, and 10 % of them are discharged (Talarposhti et al. 2001; Sponza et al. 2000). Commercial colors and dyes are resistant to light, heat, and microbial degradation, in general.

Reactive dyestuff among textile dyes plays an important role in the dying process, especially in coloring cotton. 80 % of reactive dyestuff is based on azo chromagen (Zollinger, 1991). With the increase of the use of cotton throughout the world, the use of azo dye has increased as well. Azo reactive dyes are synthetic groups of dyes which have a variety of colors and different types of structures (Sumathi and Manju, 2000). Azo dyes can be characterized by including one or more azo correlation (-N=N-) and comprise the greatest amount of annual dye production (Zollinger, 1991). Intense utilization of these synthetic chemicals has caused serious problems, such as contamination, environmental pollution, and toxicity. Moreover, azo group coloring agents are degraded into carsinogenic amines under anaerobic conditions.

Waste water of textile industries can be refined by applying biological methods as well as physical and chemical approaches. However, there are disadvantages as high costs, toxic side produces, excessive energy consumption, concentrated mud formation and difficulty in adapting to different waste water resources. Yet biological methods are more economical and have no destructive effects to the environment. Therefore, applying biological methods in purifying textile waste water is more advantageous. There have been many studies on the fission and decolorization of dye by the use of microorganisms, and it was found out there are many microorganisms having this potential. Although the results have been publicized, still no purification plants based on biological methods have been constructed. In contrast to physical and chemical systems, because of the use of living organisms in biological systems, they make them more affectable by the environmental conditions. Since there are many factors affecting the development of microorganisms which are very delicate causes biological systems to be inefficacious. Recent studies have shown that using white-rot fungus in the decolorization process is very effective.

In the field of waste and environmental biotechnology, white rot fungi are widely used (Yeşilada, 1995). This group of fungi can be used in several industrial applications, such as pulp delignification, textile dye bleaching, effluent detoxification, biopolymer modification and bioremediation (Leonowicz et al., 2001).

Minussi et al. (2001), used the cultures of four rot fungus (*T. versicolor*, *T. villosa, Phanerochaete chrysosporium,* and *Lentinus edodes*) which are prepared on agar medium to decolorize widely used reactive dye (Reactive Blue 19, Reactive Red 195, Reactive Yellow 145 and Reactive Black 5) in the textile industry. In the study, it was observed that all the dyes were decolorized; especially the decolorization process on *L. edodes* petri dishes was more swiftly. After a seven-day period, the color of these dye petri dishes decolorized totally.

In a study done by Amaral et al. (2004), the decolorization process of real textile waste water and a synthetic waste water composed of a mixture of three synthetic textile dyes of *T. versicolor* (R.Orange 4, R.Red 23 and R.Black 5) have been investigated. Various dye concentrations (0, 50, 100 and 300 mg/l.) with/without presence of glucose were tested. After a ten-day treatment with glucose, when the pH value was set to 4.5, at 50-100 mg/l dye concentration 97% of the color, and at 300 mg/l dye concentration 87% was decolorized. In a textile wastewater, which was diluted 42 times, the decolorizing percentage was 92% at 50 mg/l dye concentration.

Machado et. al. (2006), investigated the decolorizing process of reactive textile dye used in cotton industry by the use of *T. villosa,* which was isolated in Brazilian eco-system. When fungus growing and decolorizing studies were done

at malt extract agar containing 0.002g L-1, *P.sanquineus* could decolorize totally in nine days whereas *T. villosa* achieved the same result in 28 days. During the process of growing fungus at a synthetic agar medium, the effects of culture conditions (rinsing, dye and nitrogen concentrations) in decolorizing Drimaren Brilliant Blue was evaluated. It was found out that *T. villosa* was able to decolorize in rinsed cultures 85% of the dye in seven days. Pure cultures of *T. villosa* and *P. sanquineus* were able to decolorize a synthetic wastewater, which was a mixture of ten-textile dye, at high rates. It was found out that using a mixed culture of these fungus results better in the decolorization process than using pure cultures.

Ramsay et al. (2005) studied the effects of *T. versicolor,* which is grown in seaweed-based environment, in decolorizing textile dye in recurrent batch culture. Three modified Kirk's agar medium containing dye was used in this study. As the results revealed, 75% of Amaranth, Reactive Black 5, Reaktive Blue 19 and Direct Black 22 were decolorized. There withal, the mixture of these dye were decolorized totally.

In the present study, to decolorize some reactive dyestuff (Blue 49, Orange 12, Orange13, Red 31, Black 5, RBBR) used in textile industries in Turkey, a newly isolated white-rot fungus strain, *T. versicolor* , in agitated culture was attempted. Moreover, the best environmental conditions (pH, heat, amount of inoculation, shaking speed (rpm), dye concentration) for the maximum decolorization have been determined.

2. MATERIALS AND METHODS

2.1 Microorganism

Basidiomata samples of the *T. versicolor* were collected from timberland area in Eskişehir, Turkey. The specimen of native mushroom was identified in agreement with relevant literature (Breitenbach and Kränzlin, 1986; Ellis and Ellis 1990; Phillips, 2006). *T. versicolor* M96 culture was maintained on Potato Dextrose Agar (PDA) at 4 ºC in the Basidiomycetes Culture Collection of Biology Department, Eskişehir Osmangazi University and was transferred periodically to fresh media.

2.2 Dyes

The dyestuff used in this study are brand named and Color Index (C.I.) enumerated Cibacron Blue 3R (C.I. Reactive Blue 49), Reactive Golden Yellow HR (C.I. Reactive Orange 12), Reddish Orange (C.I. Reactive Orange 13), Reactive Red H8B (C.I. Reactive Red 31), Remazol Black (C.I. Reactive Black 5), Remazol Brilant Blue R (C.I.61200, Reactive Blue19). All the dyes used in this study are kindly supplied by Eskişehir Sarar Textile A.Ş.

2.3 Decolorization Studies

Decolorization studies were conducted using rinsed cultures. For the shaking incubation, 45ml Kirk medium, 5ml inoculums, and sterilized dyestuff solutions of 50 mg/l concentration was added to 250 ml erlenmayer flasks. The flaks were put in a shaking incubator, which was set at 30 °C and 150 rpm with 4.5 pH level, for a one-week incubation. The incubation has been done in a dark environment regarding that the dyestuff might disintegrate due to light.

Throughout the incubation process, samples from the extracellular culture fluid were taken every day, and they were diluted with some amount of distilled water when needed. Later, the concentration of the dye was determined by measuring the UV visible spectrophotometers absorbance. Accordingly, calibration graphics were constituted primarily. In order to form a calibration curve for each dyestuff, agar mediums with 1, 5, 10, 20, 30, 40, 50 mg/l dye concentrations were prepared. Afterward, the absorbance rate of each agar was determined by reading the values on the spectrophotometer (Blue 49 (600 nm), Orange 12 (430 nm), Orange 13 (490 nm), Red 31 (545 nm), Black 5 (600 nm), RBBR (600 nm). The decolorization was determined as follows:

Decolorization %: $(C_0^{\sim} \; C_1) \; / \; C_{,0}$).100 (*Co*; initial colour concentrations and C_1 final concentrations)

The used dyes Blue 49, Orange 12, Orange 13, Red 31, Black 5 and Remazol Brilant Blue R (RBBR), were diluted 1% with distilled water and were sterilized in autoclave at 121 °C for 15 minutes. These solutions were preserved in colored bottles at 4 °C until use. To obtain the required concentration, the dye solutions were added under sterile conditions to the medium. The absorbance rate of each dyestuff was determined at their maximum wavelength. In order to determine the absorbance rate of each dye solution, absorbance was measured by UV visible spectrophotometer (JascoV-530 UV/VIS Spectrophotometer). The maximum absorbance values were determined by scanning the absorbance

values between 300 - 700 nm. These values were determined for each dye solution and all the absorbance measures were performed considering these wavelengths.

2.4 Inoculum Preparation

The studied mushroom strain, *T. versicolor* M96, was grown on Potato Dextrose Agar and was incubated for one week at 30 °C. After the incubation period, 10 mycelial discs (10 mm diam.) were transferred to 250 ml Erlenmeyer flasks involving 50 ml modified Kirk's medium (Tien and Kirk, 1988). The Kirk's media were then incubated at 30 °C 150 rpm four days. After the incubation period, the pellets were filtered under sterile conditions and were homogenized and used as inoculums in decolorization studies.

2.5 Statistical Analysis

The decolorization percentages and results of white-rot fungus were calculated and analyzed using the SPSS program considering pH, inoculums concentration, shaking speed (rpm), dye concentration and temperature parameters.

3. RESULTS

3.1 Effect of pH on Decolorization

To determine the optimum pH, shaking speed 150 rpm, temperature 30 °C, inoculums concentration 5 ml, dye concentration Blue 49, Red 31, Black 5 and RBBR 50 mg/l, Orange 12 and Orange 13 25 mg/l.

To determine the optimum pH value, the decolorization time of the dyestuff with *T. versicolor* culture medium were studied in media adjusted to pH 3.5, 4.5, 5.5 and 6.5. The decolorization rate was the maximum in pH 4.5 when compared to the others ($p<0.05$) When the pH value for the *T. versicolor* culture medium is set to 4.5 the decolonization percentage after three days for Blue 49 and RBBR is determined as 66 % and 62 % respectively. After seven days the percentages are determined as 89 % for Blue 49 and 94 % for RBBR. The decolonization percentages after seven days for Orange 12 was 73 %, for Orange 13 75 %, for Black 5 and Red 31 80 % as shown in Figure1.

3.2 Effect of Inoculum Amount on Decolorization

To determine the optimum inoculum concentration, pH 4,5, shaking speed 150 rpm, temperature 30 °C, dye concentration Blue 49, Red 31, Black 5 and RBBR 50 mg/l, Orange 12 and Orange 13 25 mg/l.

To determine effect of inoculum amount on the decolorization ratio, culture media were inoculated with different amount of inoculum (5, 25 and 50 ml). As in the other dye, the decolorization impetus increased by increasing the inoculums concentration, however, there was a slight increase in the total decolorization percentage.

In the experiments with the *T. versicolor* fungi at different inoculums concentrations, the worst decolorization were taken 25 ml. inoculum concentration. The highest decolorization percentages were recorded for Blue 49 and RBBR. Following the seven-day period the changes in decolorization at different inoculums concentration in Orange 12 and Orange 13 were observed as follows: at 5ml inoculums concentration 73-75 %, at 25 ml. inoculums concentration 84-82 % and at 50 ml. inoculums 60-62'te The decolorization changes in Red 31 and Black 5 were as the following: at 5 ml. 79-78 %, at 25ml. 85-87 % and at 50ml. 64-66 %. Considering these results, it can be said that there was an increase in the decolorization process depending on the increase in the inoculums concentration. However, as a final effect very slight changes could be observed in the total decolorization of the dye. As can be shown in Figure 2, in the experiments performed with different inoculum amounts, 25 ml has been a significant difference.

3.3 Effect of Shaking Speed on Decolorization

To determine the optimum shaking speed concentration, pH 4,5, inoculum concentration 25 ml., temperature 30 °C, dye concentration Blue 49, Red 31, Black 5 and RBBR 50 mg/l, Orange 12 ve Orange 13 25 mg/l.

Different shaking levels were tested to find out whether increasing the shaking speed will increase the decolorization percentage. In order to find out the correlation three different shaking speed levels were tested; namely, 100, 150 and 200 rpm. We have been recorded a significant difference at the speed of 100 rpm, where as there was significant difference at speed levels 150 and 200 rpm ($p<0.05$) After a seven-day incubation of Blue 49 textile dye with 50 mg/l dye concentration at 100 rpm 65 % of decolorization was achieved, while at 150 rpm 92 % and at 200 rpm 94 % decolorization was achieved.

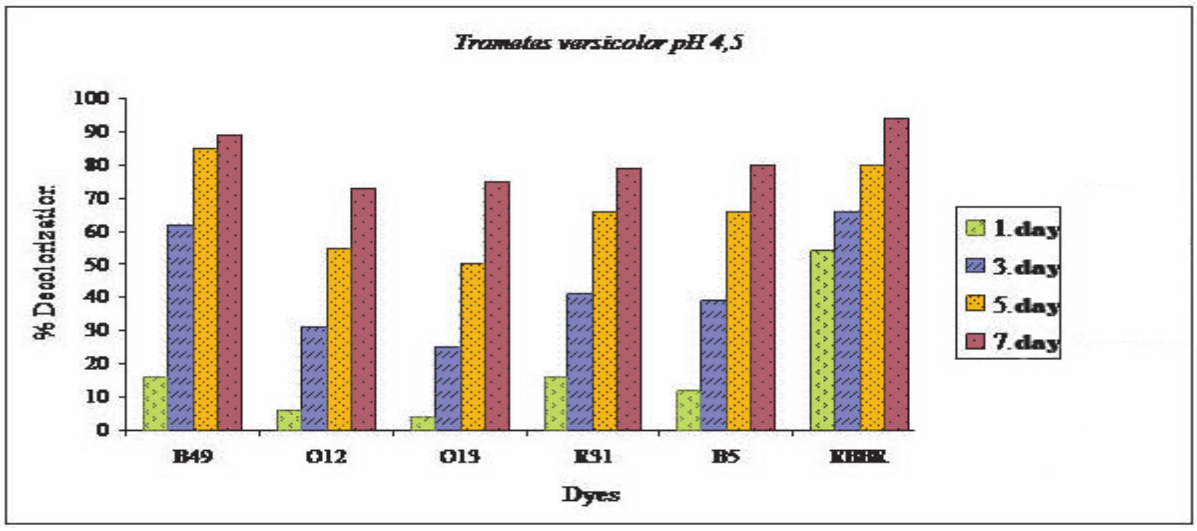

Figure 1. Time course of dye decolorization by *T. versicolor* M96 at pH 4,5.

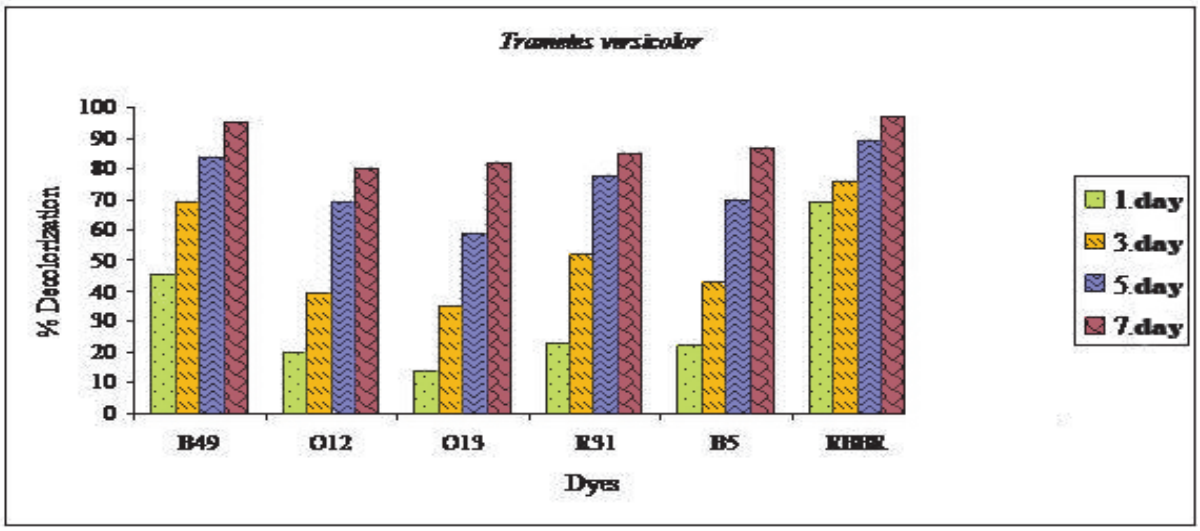

Figure 2. Time course of dye decolorization by *T. versicolor* (M96) with 25 ml inoculum amount (pH 4,5)

Following a seven-day incubation of Orange 12 and Orange 13 textile dyes at 100 rpm 40 %-42 %, at 150 rpm 80 % - 82 % , and at 200 rpm 83 % -84 % respectively, of decolorization was achieved. By increasing the shaking speed the decolorization amount increased accordingly. However, the decolorization amount was slightly faster at 200 rpm than at 150 rpm.

Black 5 textile dye discolored at a great amount at 200 rpm on the third day of incubation at 58 %. On the 7th day the decolorization percentage was at 88 %. The decolorization percentage at 150 rpm on the 7th day of incubation was 84 % which decreased significantly at 100 rpm down to 49 %.

After seven days of incubation of RBBR textile dye the decolorization percentage at 100 rpm was 58 %, 92 % at 150 rpm and the highest decolorization was achieved as 97 % at 200 rpm as shown in figure 3.

3.4 Effect of Dye Concentration on Decolorization

To determine the optimum dye concentration, pH 4,5, inoculum concentration 25 ml., temperature 30 °C, shaking speed speed 200 rpm., dye concentration Blue 49, Red 31, Black 5 and RBBR 50 mg/l, Orange 12 and Orange 13 25 mg/l.

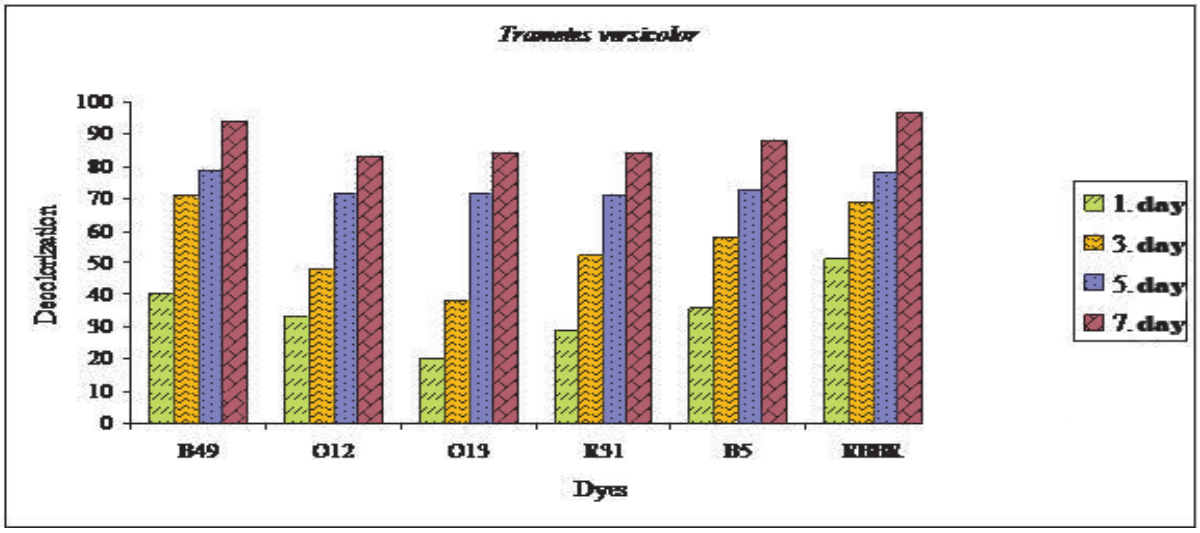

Figure 3. Time course of dye decolorization by *T. versicolor* (M96) at 200 rpm shaking speed (pH 4,5; 25 ml inoculum amount)

In the time based decolorization process of *T. versicolor*, culture medium containing 25, 50 and 100 mg/l of initial dye concentration were used. The decolorization rate decreased by gradually increasing the dye concentration in textile dye. After seven-day incubation of culture medium containing 25 mg/l Blue 49 and RBBR textile dyes the decolorization rate was 98 % (Figure 4). The decolorization percentage with 50 mg/l dye concentration the result was 96 % for Blue 49 and 97 % for RBBR, which was a similar result compared with 25 mg/l dye concentration. However, there was a slight decrease in decolorization at 100 mg/l as 90 % for Blue 49 % 90 and 86 % for RBBR (Figure 6). When the dye concentration is increased the decolorization rate decreased slightly, however, after the incubation period close results were revealed. When analyzed the decolorization percentage there was a significant difference between 25 mg/l and 100 mg/l with Blue 49 and RBBR dye concentrations.

Interestingly, there has been a great decline by the fungus in the decolorization speed by increasing the concentration of Orange 12 and Orange 13 dye concentrations. 25 mg/l dye concentration revealed 83 % for Orange 12 and 85 % for Orange 13 (Figure 4). At 50 mg/l concentration the decolorization rate decreased to 53 % for Orange 12 and 52 % for Orange (Figure 5). The decolorization rate decreased tremendously at 100 mg/l dye concentration which was 25 % for Orange 12 and 28 % for Orange 13 (Figure 6). Statistically there was a significant difference between 25 mg/l and 100 mg/l dye concentrations.

In culture medium containing 25 mg/l of Red 31 and Black 5, after seven-day incubation, there has been a decolorization percentage at 95 % and 96 %, respectively. The rate was 80 % for Red 31 and 91 % for Black 5 at 50 mg/l dye concentration, and at 100 mg/l the result was Red 31 68 % and Black 5 75 %.

These results illustrate that increase in the dye concentration results in slowing down the decolorization ratio. Statistical analyzes demonstrate that there has been a significant difference between 25 and 50 mg/l dye concentrations. In all the textile dyes, dye concentration is increased the decolorization rate decrease accordingly.

3.5 Effect of Incubation Temperature on Decolorization

To determine the optimum temperature, pH 4,5, inoculum concentration 25 ml.,shaking speed 200 rpm., dye concentration Blue 49, Red 31, Black 5 and RBBR 50 mg/l, Orange 12 and Orange 13 25 mg/l.

To determine the optimum temperature for dye decolorization, *T. versicolor* was incubated at different incubation temperature such as 25, 30, 35 and 40 °C. The maximum decolorization rate was observed in the group of incubated at 30 °C ($p<0.05$) (Figure 7). After the third day of incubation with *T. versicolor* fungus at 25 °C Blue 49 textile dyes discolored at 46 %, and on the seventh day the decolorization rate was 67 %. The result on the third day at 30 °C was 71

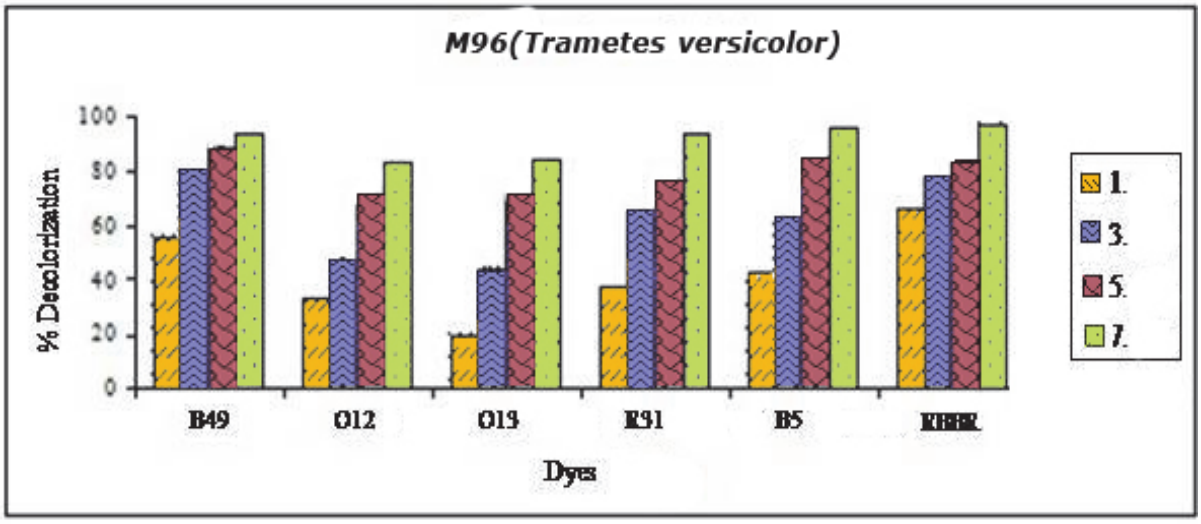

Figure 4. Time course of dye decolorization by *T. versicolor* (M96) at 25 mg/l dye concentration (pH 4,5; 25 ml inoculum amount).

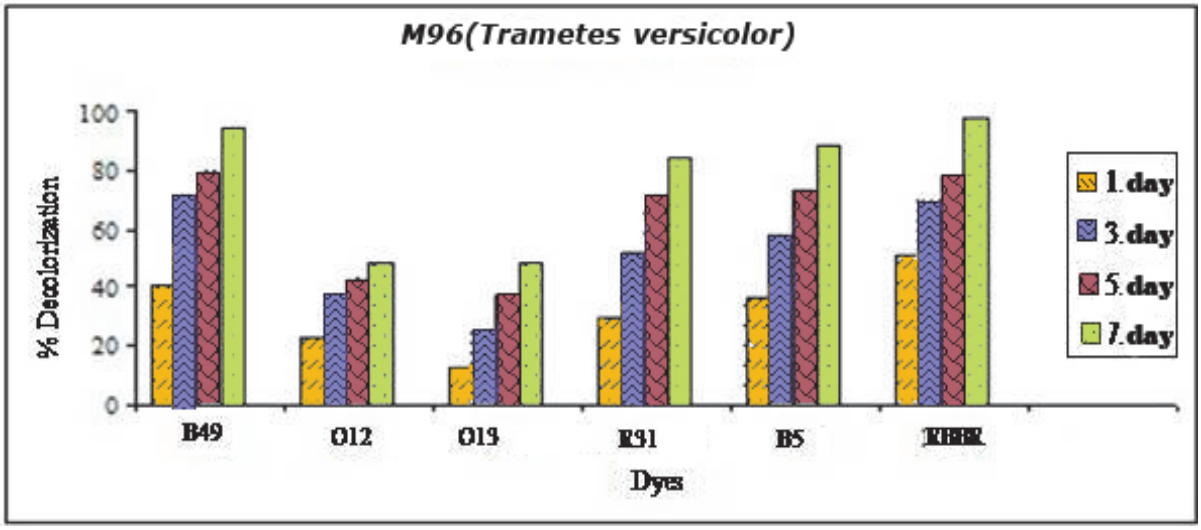

Figure 5. Time course of dye decolorization by *T. versicolor* (M96) at 50 mg/l dye concentration (pH 4,5; 25 ml inoculum amount).

%. However, decolorization on the seventh day at 35 °C was 45 %, and at 40 °C it was 31 %, which was lower when compared to the result at 30 °C.

The optimum decolorization rate for Orange 12 textile dye was at 30 °C with 83 %. Bye increasing the temperature a decrease in the decolorization percentage was observed; namely, 39 % at 35 °C, 23 % at 40 °C. The rate achieved at 25 °C very close to the one at 30 °C' but it was slower, and after seven-day incubation the rate was 67 %.

50% of the Orange 13 textile dye was decolorized at 25 °C on the fifth day. 72 % of the dye discolored at 30 °C on the fifth day, however, after the third day decolorization became faster. The rate at 35 °C was lower than at 25 °C and 30 °C, which reached to 35 % on the seventh day. The lowest, 27 %, decolorization rate was at 40 °C.

The textile dyes Red 31 and Black 5 discolored at 69 % and 72 % at 25 °C, respectively, and at 30 °C 84 % of Red 31 and 88 % of Black discolored. The rates at 35 °C was 39 % for Red 31 and 45 % for Black 5 %, and for both dyes the rate was 28 %.It could seen that the decolorization percentages decreased with higher temperatures.

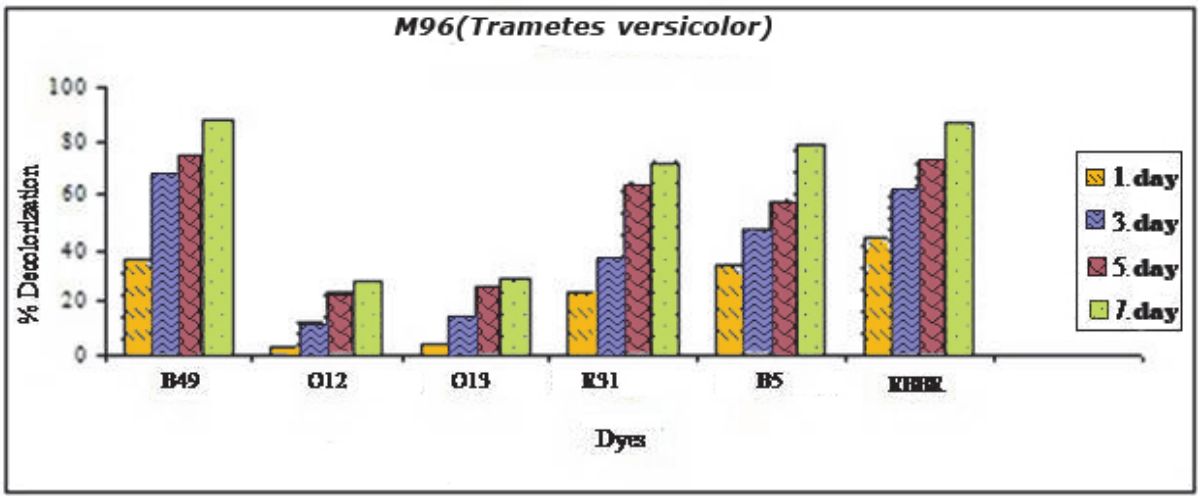

Figure 6. Time course of dye decolorization by *T. versicolor* (M96) at 100 mg/l dye concentration (pH 4,5; 25 ml inoculum amount).

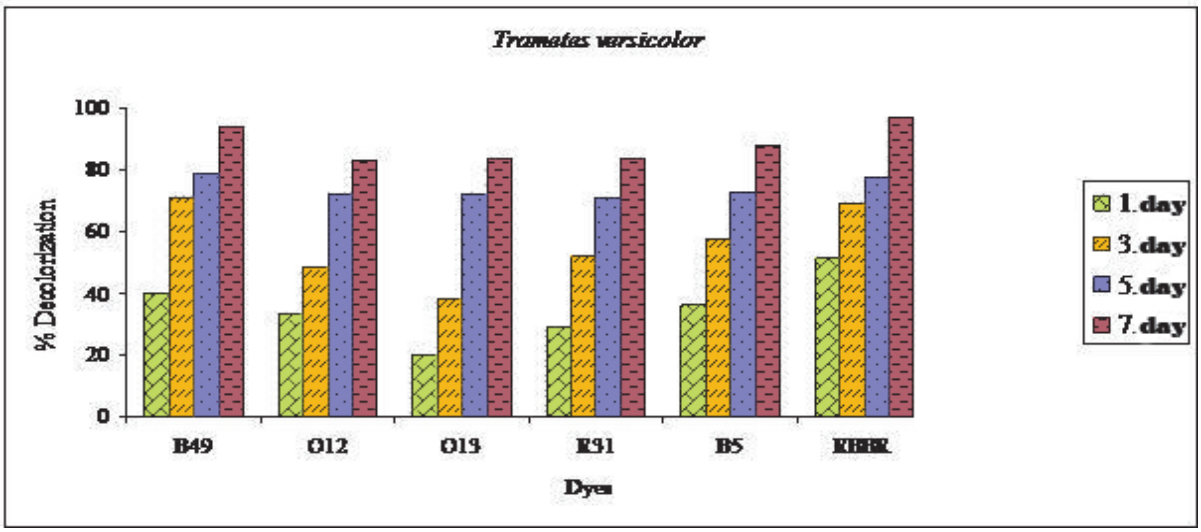

Figure 7. Time course of dye decolorization by *T. versicolor* (M96) at 30 °C incubation temperature (pH 4,5; 25 ml inoculum amount)

The decolorization percentages for the RBBR textile dye is as follows: 79 % at 25 °C, 97 % at 30 °C, which demonstrated a very high decolorization rate, 51 % at 35 °C, and 37 % at 40 °C.

4. DISCUSSION

In the last decades, control of the water pollution has gained great importance. Disposition of dye into water resources composes an important part of the pollution. Even a small amount of dye in these resources is not desired. Thus, because of ecological reasons, the process to decolorize textile waste water gains utmost importance.

Decolorization and/or detoxification of texile dyes by fungi was effected by different factors such as pH, inoculum amount, shaking speed, dye concenteration, temperature etc.

4.1 Effect of pH on Decolorization

Benito et. al. (1997) determined the optimum pH level for decolorization as 5. *T. versicolor* at pH 3.5 and 7 decolorized waste water at 60 %, 80 % and 10 %, respectively. There was only 10 % decolorization at pH 8-9 values, and there was not a significant growth of the fungus.

In a study by Swamy and Ramsay (1999) in decolorizing Amarath, Remazol Black B,

Remazol Orange, Remazol Brillant Blue, Reactive Blue and Tropaeolin O dyes with *P. chrysosporium , T. versicolor* and *Bjerkanderara* sp. white-rot fungus has illustrated the optimum pH value which is 5.

In the study by Sam (1999) to discolor Orange II textile dye using the *T. versicolor* white-rot fungus the optimum pH value pH was found out to be pH 4.5 with the highest decolorization rate of 63 %.

In a study, Shahvali et. al. (2000), to determine the environmental factors in decolorizing textile wastes with *P. chrysosporium* the maximum decolorization was achieved at pH 3. By increasing the pH value above 5 the decolorization rate slowed down. Shahvali et al. claim this as an effect due to osmotic changes causing hydrolysis effect.

In studies with pH have illustrated that pH has an important effect on the decolorization process. Maximum decolorization rate was achieved when microorganisms have shown the optimum growth or when enzymes are activated by the optimum pH value. In this study, the utmost decolorization of *T. versicolor* (M96) was achieved at pH 4.5. However, there was a slight decolorization rate at pH 5.5, whereas the percentages were very low at pH 3.5 and 6.5.

4.2 Effect of Inoculum Amount on Decolorization

Benito et al. (1997) in their *T. versicolor* study determined that by increasing the inoculums concentration the decolorization rate increases accordingly.

When Yeşilada et al. (2002) incubated *Funalia trogii* pellets in culture medium containing Astrazon FBL; they find out that in case of high inoculums concentration the decolorization percentages increase as well. When the inoculums concentration is kept at 60 mg/50 ml the decolorization rate is about 64 %, and at 150 mg/50 ml the rate is %96.

Assadi et. al. (2001) in the study to decolorize textile waste water through *P. chrysosporium* experimented with an inoculums concentration of 2-20 % and achieved the maximum decolorization rate at 10-15 %.

As seen in these sample studies when the inoculums concentration is increased the decolorization rate increases accordingly. Since increasing the inoculums concentration means that the dyestuff has more areas to connect which, expectedly, results in high decolorizing rates.

4.3 Effect of Shaking Speed on Decolorization

Swamy and Ramsay (1999) achieved a 96 % of decolorization of *Bjerkandera* sp. BOS 55 Remazol Black B at 200 rpm shaking conditions in three days, whereas *T. versicolor* discolored 100% at 200 rpm only in two days.

In a study by Yeşilada et. al. (2003) using *F. trogii* fungus Astrazone Black FDL, Astrazone Blue FGRL and Astrazone Red FBL textile dyes the maximum decolorization was achieved at 150 rpm. They claimed that this fungus illustrates low decolorization at 100 rpm and high decolorization rate at 150 rpm.

In the optimization study by Sam (1999) of the decolorization effect of Orange II textile dye with the *C. versicolor* and *F. trogii* fungi rpm values between 50 and 200 were tested, and the maximum rate was reached at 150 rpm.

These studies demonstrate that since there is a high amount of mass and oxygen transfer between the cells and the culture medium at high shaking speeds, there is a significant increase in the decolorization rates. Similarly, the contiguity opportunity between the microorganisms and dye molecules is faster and excessive.

4.4 Effect of Dye Concentration on Decolorization

In a study by Yeşilada et. al. (2002) to research the decolorization ability of Astrazon FBL culture medium containing *F.trogii* pellets, it was found out that the decolorization percentage increased in parallel with increasing the dye concentration to 66 mg/l, and that it started to decrease exceeding 66 mg/l.

In the decolorization study with the *C. versicolor* fungus and Everzol Turguoise Blue G by Kapdan and Kargı (2000) it was stated that increasing the dye concentration might have a toxic effect on the fungus.

Myrothecium verrucaria's absorption related decolorization ability of its culture medium increases to a certain amount of dye concentration and exceeding that point it slows down (Mou et. al., 1992)

In all the textile dyes used in this study the decolorization period extended with an increase in the dye concentration.

4.5 Effect of Incubation Temperature on Decolorization

In the a study by Toh et. al. (2003) with *P. chrysosporium, T. versicolor* fungus to decolorize Remazol Red RR and Remazol Yellow RR dyes between temperatures at 30°C and 37°C, they found out that decolorization at 30°C was faster than at 37°C.

Sam (1999) experimented with *C. versicolor* and *F. trogii* fungi and Orange II textile dye, and stated that the decolorization rate was achieved at 30°C.

In the study by Assadi et. al. (2001) in which they made a research on the decolorization of textile waste water using the *P. chrysosporium* fungus between temperatures 25 – 40°C concluded that the best decolorizing results were reached between 30°C and 35°C.

In studies with *T. versicolor* (M96) 30°C has generally been determined as the optimum temperature for the decolorization of textile. This result is also in accordance with the findings of this study.

These results show that *T. versicolor* has potential for use in the treatment of dye-contaminated effluent. We believe that this fungi may play an important role in the decolorization of dyes.

In conclusion it can be said that our *T. versicolor* strain uses its enzymes in the decolorization process to dismantle dye; thus, it prevents any problem that may be caused by any dyestuff.

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