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Biological color stripping: A novel technology for removal of dye from cellulose fibers

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ABSTRACT

This research work was carried out to compare the color stripping efficiency of optimized biological method with the chemical stripping, commonly employed in the textile industries. Knitted fabric dyed with Reactive black B dye in 2, 4 and 6% shades strengths was subjected to chemical and biological stripping processes individually. Biological stripping process was found many fold superior to chemical one. It was noted that shade strength does not showed any pronounced effect on the bursting strength of fabric but biological and chemical treatment affect the quality of fabrics in terms of bursting strength/durability of fabric. White rot fungus *Ganoderma lucidum IBL-05* showed good potential for decolorization/color stripping of cotton fabric dyed with Reactive black B under optimized set of conditions. The chemical stripping technology is inferior to biological stripping process regarding the quality of fabric and percent color removal from cotton fabric dyed with Reactive black B dye.

1. Introduction

Almost one million tons of synthetic dyes are annually produced in the world and used by the industries for coloring various materials to make them colorful and attractive to cope the challenges in the competitive atmosphere of marketing (Santos, Bisschops, Cervantes, & Lier, 2004). It is often enviable to remove the color from the dyed fabrics in order to correct faulty dyeing or re-dye the surplus fabric in different shades and colors for reuse. Stripping is the process which is commonly employed to remove dye from dyed fabrics and the process is either designated "back stripping" or "destructive stripping". In back stripping, only the depth of shade is changed while in destructive stripping the dye is chemically altered (Fono & Montclair, 1980).

Textile fabrics are commonly stripped using various stripping liquids, consisting of a chemical reducing agent and a stripping assistant; chemicals used to enhance the stripping efficiency of a reducing agent. Various chemical combinations of reducing agents and stripping assistants are being used in stripping liquids to strip dye from fabrics (Chavan, 1969). Direct dyes are stripped off by boiling the fabric in alkaline sodium hydrosulfite, bleaching the fabrics with sodium hypochlorite or boiling the fabrics in 1–2% sodium chlorite solution at pH 3–4, adjusted by formic/acetic acid. Vat dyes are removed from fabrics by treating it in high temperature reducing bath containing caustic soda, sodium dithionite and a substance that is responsible to combine with the dye molecule so that it does not re-attach with the textile material once it has been stripped out. Hot solution of soda ash and sodium sulfide is employed for the removal of sulfur dyes while disperse dyes can be stripped from fabrics with hot solution of sodium hydrosulfite and ammonium hydroxide (Chavan, 1969).

None of the reported stripping liquids has universal acceptance in the dyeing industries as none of them could be used on various blends of fabrics and dyes; rather it has to be individually designed depending on the nature of dye to be removed from and type of fabrics. Many of the reported stripping liquids have pH greater than 9 that could be harmful to fabric material and most often the stripped fabric is very difficult to re-dye (Fono & Montclair, 1980). Moreover, all of these techniques are purely chemical based and reported to be associated with a number of drawbacks due to the chemical system involved that may restrict the recycle ability of polymeric material. In general, processes that utilize harsh stripping agents destroy the usefulness of the colorant, thus generating a chemical waste stream that must be treated or disposed off in an environmental conscious manner (James, 1992; Wilhelm, Walles, & Axelon, 1963).

The attention in the biodegradation/decolorizarion of synthetic dyes has principally been encouraged by the apprehension over their possible toxicity and carcinogenicity (Maas & Chaudhari, 2005; Revankar & Lele, 2007). WRF are considered to be the better dye-degraders than prokaryotes because of their extracellular non-specific lignin mineralization enzyme system that make them enable to degrade a wide range of dyestuffs (Christian et al., 2005).

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Most of the earlier studies for dye decolorization were based on *Phanerochaete chrysosporium* and *Trametes versicolor* (Asgher, Bhatti, Ashraf, & Legge, 2008, Asgher, Kousar, Bhatti, Shah, & Ali, 2008; Toh, Jia, Yen, Obbard, & Ting, 2003). However, a number of other species of WRF like *Phellinus gilvus*, *Pleurotus sajor-caju*, *Pycnoporussanguineus*, *Dichomitus squalens*, *Irpex flavus*, *Daedalea flavida*, *Polyporus sanguineus*, *Funalia trogii*, *Ischnoderma resinosum* and *Ganoderma sp.* have been recognized for better dye decolorization rates than *P. chrysosporium* and *T. versicolor* (Balan & Monteiro, 2001; Chander & Arora, 2007; Chander, Arora, & Bath, 2004; Eichlerova, Homolka, & Nerud, 2006; Ozsoy, Unyayar, & Mazmanci, 2005; Revankar & Lele, 2007).

The mechanisms of biodegradation of different dyes have also been investigated (Gavril & Hodson, 2006; Zhao & Hardin, 2007; Zhao, Hardin, & Hwang, 2006). *P. chrysosporium* decolorized Direct Blue-15 following the first-order kinetics with respect to initial concentration of dye and manganese peroxidase (MnP) plays the key role in the decolorization as the minor mechanism (Pazarlioglu, Sariisik, & Telefoncu, 2005). MnP performed as a major enzyme with slight activities of laccase and lignin peroxidase (LiP) for the decolorization of Solar golden yellow R using *Schyzophyllum commune*. It has been by some researchers that low molecular weight mediators are essential for decolorization of most of the dyes by laccase (Lu, Shen, & Xia, 2005; Lu, Zhao, & Wang, 2007). However, *Lentinula edodes* and *Pleurotus pulmonarius* have been reported to produce only extracellular laccase for the decolorization of various dyes without any mediators (Nagai et al., 2002). MnPs mediated decolorization of azo dyes, Direct Blue-15, Direct Green-6 and Congo red with *P. chrysosporium* can be improved by adding Tween-80 and copper (Tychanowicz, De-Souza, Souza, Kadowaki, & Peralta, 2006; Urek & Pazarlioglu, 2005). Christian et al. (2005) investigated that LiP isolated from *T. versicolor* decolorized Remazol Brilliant Blue R in the presence and also in the absence of veratryl alcohol (VA). Literature also revealed that by optimizing the operational conditions like initial dye concentration, nutrients contents of the media, carbon and nitrogen sources, the decolorization ability of WRF can be enhanced further (Nilsson, Moller, Mattiasson, Rubindamayugi, & Welander, 2006; Ozsoy et al., 2005; Sanghi, Dixit, & Guha, 2006). It was also investigated that by the addition of glucose as carbon source showed the dramatic boosting in decolorization of Solar golden yellow R with *S. commune*, whereas, the additional nitrogen sources inhibited the formation of MnP and subsequently decline the dye decolorization efficiency (Asgher, Bhatti et al., 2008; Asgher, Kousar et al., 2008).

Faisalabad is famous all over the world as Manchester of Pakistan because of huge number of textile units in and around the municipal city area that has gained the status of quality in the world of fabrics. A lucrative and environment friendly method for the removal of dyes from fabrics without any treatment and disposal problems is an immense stipulate among the industrialists.

2. Experimental

2.1. Collection of material

Knitted cotton fabric was procured from Jinnah colony, Faisalabad, Pakistan. Reactive black B (hetero bifunctional) dye was provided by Sandal Dyestuffs Industries Limited, Faisalabad, Pakistan for research purpose. All the chemicals and reagents used in this research were of analytical grade and were purchased mainly from Sigma–Aldrich–Fluka (Amherst, NY, USA) and used without further purification.

2.2. Dyeing of fabric

Before dyeing the grey fabric was bleached using 50% H_2O_2 (20 g/L) and 50% NaOH (15 g/L) for half an hour at 50 °C. Fabric was dyed with Reactive black B in 2% shade strength on the basis of weight of the fabric (WOF), following the method of Pearce, Lloyb, and Guthrie (2003) with some modifications. Fabric to liquid solution ratio was kept at 1:15. The liquid solution was made by taking 30 g/L each of salt (Na_2SO_4) and alkali (Na_2CO_3). Dyeing was carried out using exhaust dyeing method at 45 °C for 15 min, followed by at 65 °C for 60 min. The dyed fabric was rinsed with tap water thoroughly.

2.3. Biological stripping

Pure cultures of *Ganoderma lucidum* IBL-05 was obtained from fungal culture collection of Industrial Biotechnology Laboratory (IBL), Department of Chemistry and Biochemistry, University of Agriculture, Faisalabad. The flasks containing inoculum media (100 mL) were adjusted at pH 4.5 using 1 M NaOH/1 M HCl and autoclaved (Sanyo MLS-3020U, Japan) at 121 °C for 15 min. The flasks were inoculated with loopful fungal spores from slant cultures of *G. lucidum* and placed in orbital shaking incubator (120 rpm) at 30 °C for 3 days to get homogenous inoculum The number of spores in the inoculum were counted using heamocytometer in Department of Microbiology, University of Agriculture, Faisalabad to get 1×10^7 to 1×10^8 spores/mL. Fresh inoculum was prepared for each experiment (Asgher, Shah, Ali, & Legge, 2006).

2.3.1. Basal nutrient medium

Kriks basal salt medium (Tein & Krik, 1988) had the following composition (g/L): ammonium tartrate, 0.22; KH_2PO_4, 0.21; $MgSO_4 \cdot 5H_2O$, 0.05; $CaCl_2 \cdot 2H_2O$, 0.01 and thiamine, 0.001 with 10% tween 80 solution, 10 mL; 100 mM veratryl alcohol, 1 mL and trace elements solution, 1 mL. Trace element solution was coposed of (g/L): $CuSO_4$, 0.08; H_2MoM_4, 0.05; $MnSO_4 \cdot 4H_2O$, 0.07; $ZnSO_4 \cdot 7H_2O$, 0.043 and $Fe_2(SO_4)_3$, 0.05.

2.3.2. Stripping procedure

Triplicate flasks containing 100 mL basal medium, 1 g wheat bran as carbon source, 0.2 g peptone as nitrogen source, 1 mL $MnSO_4$ (1 mM) as meditor, 1 mL $ZnSO_4$ (1 mM) as metal ion optimized in our prevous paper (Chatha, Asgher, Ali, & Bhatti, 2011) and 5 × 5 in. dyed fabric pieces of three shades (2, 4 and 6%), were adjusted to pH 4.0 with M NaOH/M HCl and autoclaved. The triplicate stripping flasks were inoculated with 2 mL inoculum of *G. lucidum* IBL-05 and incubated at 35 °C in shaking incubator at 120 rpm for 15 days. After 15 days incubation the triplicate sample and control flasks were harvested and fabric pieces were washed, dried, labeled and evaluated for color stripping.

2.4. Chemical stripping

Chemical stripping of dyed fabric was carried out according to the conventional method employed in textile industries as reported by Ogulata and Balci (2007) with some modifications. Dyed fabric was boiled in aqueous stripping liquid containing NaOH (20 g/L) and $Na_2S_2O_4$ (15 g/L), maintained at a temperature about 80 °C for 30 min with 1:20 materials to liquid ratio. Finally stripped fabric samples were washed with tap water, dried and labeled.

2.5. Analytical

The value of color strength of stripped and un-stripped cotton fabrics was determined by Spectraflash Spectrophotometer SF650 at Sandal Dyestuffs Quality Control Laboratory, Faisalabad, Pakistan. The color strength of dyed fabrics was measured in terms of *K*/*S* values which represent the ratio of light absorbed/reflected

Table 1
Comparison of chemical[a] and biological[b] stripping of cotton fabric dyed with Reactive black B.

Shade strength (%)	Un-stripped	Biological stripping			Chemical stripping		
	Color strength (*K*/*S* value)	Color strength (*K*/*S* value)		Stripping (%)	Color strength (*K*/*S* value)		Stripping (%)
		Control	Treated		Control	Treated	
2	8.4 ± 0.21	7.82 ± 0.13	0.29 ± 0.01	89.64 ± 3.17[a]	6.9 ± 0.23	1.6 ± 0.01	63.09 ± 2.05[cd]
4	20.9 ± 1.20	17.5 ± 1.31	2.1 ± 0.03	73.68 ± 2.97[b]	16.1 ± 0.27	3.1 ± 0.02	55.50 ± 2.44[d]
6	37 ± 1.43	32.7 ± 1.59	6.9 ± 0.20	69.72 ± 2.58[bc]	33.9 ± 1.52	19.7 ± 0.94	32.97 ± 1.64[e]

[a] Stripping mixture; NaOH (20 g/L) and sodium hydrosulfite (15 g/L); temperature, 70 °C ± 5, execution of process 30 min.
[b] pH 4.0; temperature, 35 °C; carbon source, wheat bran (1.0%); nitrogen source, peptone (0.2%); mediator, $MnSO_4$ (1 mM).

by the fabric. Percent stripping was calculated by using following formula:

$$\%\text{age stripping} = \frac{\left[K/S \text{ value of control sample} - K/S \text{ value of stripping sample}\right]}{K/S \text{ value of un-stripped fabric}} \times 100$$

2.6. Quality of fabric

The quality of stripped and un-stripped fabric was evaluated by following tests.

2.6.1. Bursting strength

The bursting strength of biologically and chemical stripped fabric was evaluated at National Textile University (NTU), Faisalabad following the method of American Association of Textile Chemists and Colorists (AATCC, 2001). This test method describes the measurement of the resistance of textile fabrics to bursting using a pneumatic diaphragm bursting tester. This test method is generally applicable to a wide variety of textile products. The values stated in either SI units or inch-pound units are to be regarded separately as standard.

2.6.2. Pilling

Pilling of fabric stripped in biological and chemical methods were assessed by Martindale tester at NTU, Faisalabad following the method of American Society of Testing Materials (ASTM, 1999). The observed resistance to pilling is reported on an arbitrary scale ranging from 5 to 1 (no pilling to very severe pilling).

2.6.3. Weight loss

Loss in weight of biologically and chemical treated fabric was measured by difference in weight before and after treatment following the method of American Association of Textile Chemists and Colorists (AATCC, 2001).

2.7. Statistical analysis of data

All the experimental treatments were run in three replicate and analyzed in triplicate. The data has been presented as mean ± SD (standard deviation). Furthermore, the data obtained was analyzed using statistical techniques of ANOVA, DMR and Correlation on statistical software Costat Computer Package (version 6.303, PMB 320, Monterey, CA 93940, USA) using Microsoft Windows XP 2002 (Steal, Torrie, & Dickey, 1997).

3. Results and discussion

After optimization of all the parameters for biological stripping process, cotton fabric dyed with Reactive black B dye in different shade strengths (2, 4 and 6%) was stripped under optimized set of conditions by *G. lucidum* IBL-05. To compare the efficiency of biological stripping, fabric dyed in all the three shades strengths was also stripped using chemical method as a standard which is commonly employed in the textile industries. The data regarding percent stripping of all the three shade strengths of cotton fabric stripped with biological and chemical methods have been presented in Table 1. The pictures of biologically and chemically stripped fabric samples are displayed in Fig. 1a and b. In biological process 89.64, 73.68 and 69.72% color stripping was achieved for 2, 4 and 6% shade strengths of dyed fabric, respectively. Percent stripping achieved by chemical process was 63.09, 55.50 and 32.97 for 2, 4 and 6% shade strengths of fabric, respectively. It was noticed that biological stripping was superior to chemical stripping of cotton fabric dyed in different shade strengths.

Chemical stripping showed the similar trend of color stripping/decolorization of dyed fabric as was observed in case of biological stripping for different shade strengths. The findings for the comparative study of biological and chemical stripping of cotton fabric demonstrated that the biological stripping is manifold superior to chemical stripping. Statistical analysis showed the significant ($p \leq 0.05$) differences in percent stripping among both the processes and shade strengths. Even then the biological stripping of dark shade fabric was superior to chemical stripping of light shade fabric. The work presented in this dissertation is absolutely novel approach for color stripping of fabric dyed with reactive dye using fungi and their enzyme system and no previous work has been reported in literature for comparison.

The pilling of textile fabric is a very complex property and the degree of fabric pilling is evaluated by comparing the test specimens with visual standards showing a range of pilling resistance. The resistance to pilling was evaluated on an arbitrary scale ranging from 5 to 1 (no pilling to very severe pilling) using the Martindale tester and the data have been presented in Table 2. The pilling resistance ranking of biologically stripped cotton fabric samples was found 4 for all the shade strengths whereas, the ranking of chemically stripped cotton fabric samples was 3, 3/4 and 3 for light, medium and dark shades, respectively. The pilling resistance ranking of untreated fabric (4/5) was found better than the treated ones. It was observed that the shade strength did not have any significant effect on the pilling resistance of fabric but biological and chemical treatment affect the pilling resistance. This test demonstrated the biological stripping as a superior one as compared to chemical stripping regarding this surface property of cotton fabric. It can be concluded that the enzymatic processing is gentle treatment and does not damage the surface of fabric whereas chemical treatment can damage the fabric strength and surface. This test is applicable to a wide variety of woven and knitted fabrics that vary in pilling propensity. Finishes and fabric surface changes may exert a significant influence on pilling. This test is advisable before and after refurbishing the fabric material. Pills vary appreciably in size and their appearance depends on the presence of lint and degree of color contrast. These factors are not evaluated when pilling is rated solely on the number of pills. The development

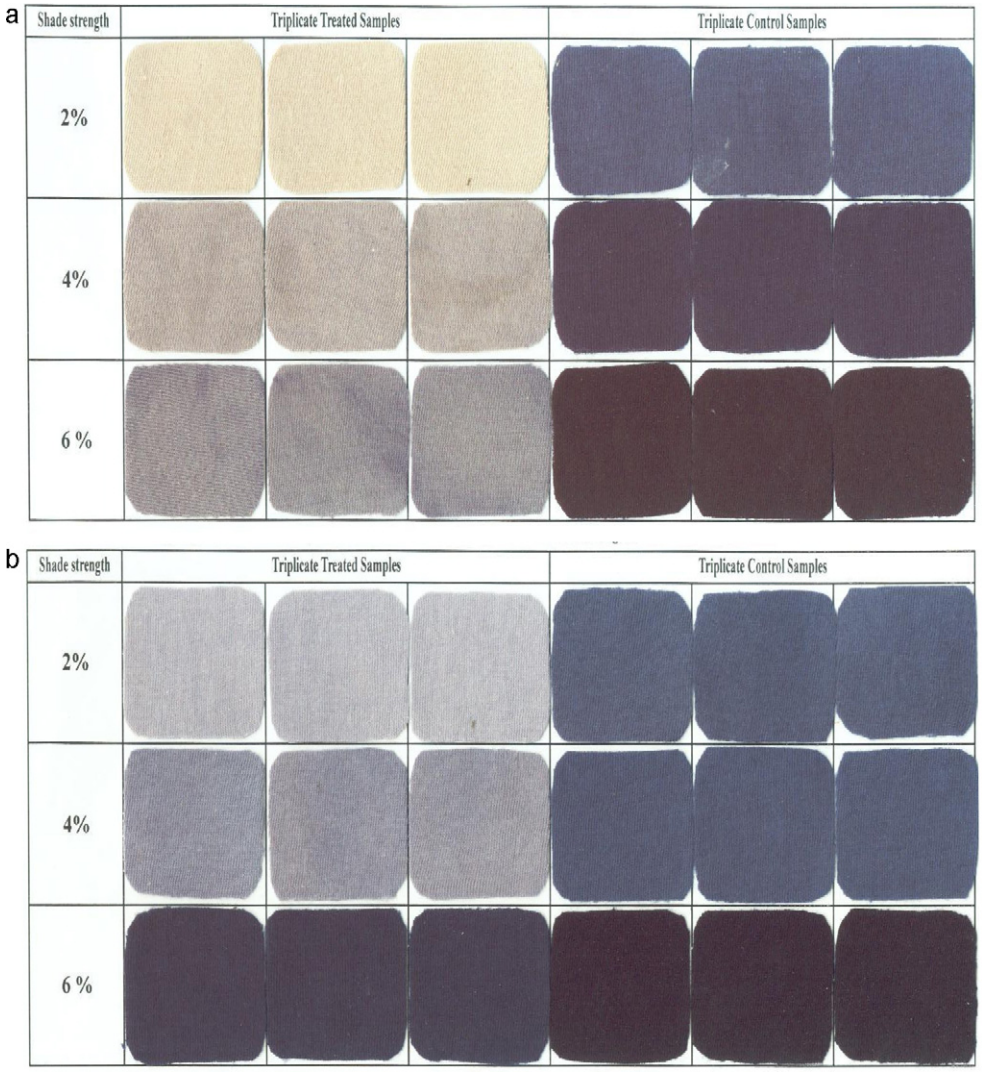

Fig. 1. (a) Samples of cotton fabric dyed with Reactive black B in different shade strengths stripped by *Ganoderma lucidum IBL-05* under optimum conditions. (b) Samples of cotton fabric dyed with Reactive black B in different shade strengths stripped by chemical process.

Table 2
Pilling resistance ranking of biological and chemical treated fabric.

Untreated	2% shade strength				4% shade strength				6% shade strength			
	BS	BC	CS	CC	BS	BC	CS	CC	BS	BC	CS	CC
4/5	4	4/5	3	3/4	4	4	3	3	4	4/5	3	3/4

BS, biologically stripped sample; BC, control for biological stripping; CS, chemically stripped sample; CC, control for chemical stripping.

of pills may be accompanied by other surface phenomena, such as loss of cover, color change, or the development of fuzz (www.astm.org).

The bursting strength of biologically stripped cotton fabric samples was 889, 874 and 890 KPa for light, medium and dark shades, respectively. Whereas, the bursting of chemically stripped cotton fabric samples was 845, 839 and 853 KPa for light, medium and dark shades, respectively (Fig. 2). The bursting strength of untreated fabric (950 KPa) was however; better than fabric subjected to both the stripping processes. It was noted that shade strength does not showed any pronounced effect on the bursting strength of fabric but biological and chemical treatment affect the quality of fabrics in terms of bursting strength/the durability of fabric. This test demonstrated the biological stripping as a superior one as compared to chemical stripping regarding the durability

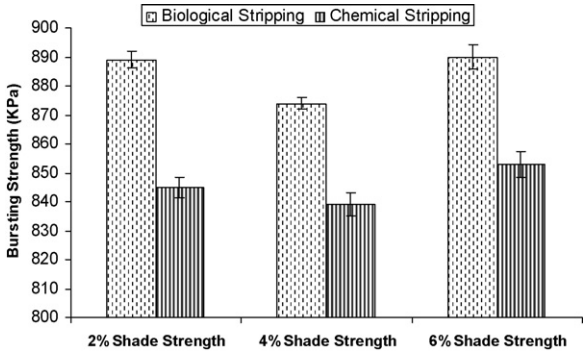

Fig. 2. Effect of biological and chemical stripping on the bursting strength of cotton fabric dyed with Reactive black B.

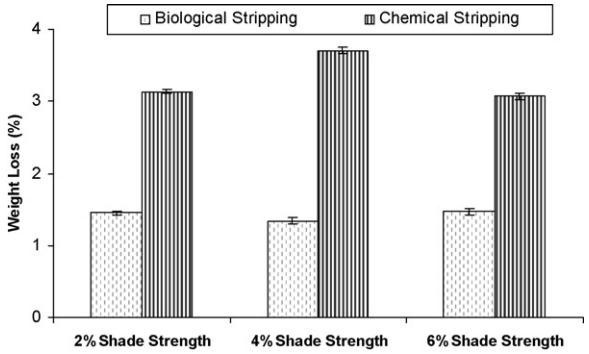

Fig. 3. Effect of biological and chemical stripping on the weight loss of cotton fabric dyed with Reactive black B.

of cotton fabric. The statistical analysis showed non significant ($p \leq 0.05$) difference among the both treatments. In contrast to the harsh chemical conditions applied in conventional treatment, the gentle enzymatic procedure, significantly lower fiber damaging and thus results in better mechanical properties of textile fabrics (Abdel-Halim, Fahmy, & Fouda, 2008; Sawada, Tokino, Ueda, & Wang, 2003).

Quality of chemically and biologically stripped fabric was also evaluated by measuring the percent weight loss. The loss in weight (%) of biologically stripped cotton fabric samples was found to be 1.45, 1.34 and 1.47 (%) for light, medium and dark shade, respectively. However, weight losses of chemically stripped cotton fabric samples were 3.13, 3.70 and 3.01 (%) for light, medium and dark shade, respectively. It was noted that the color shade strength did not show any significant effect on the weight loss of fabrics (Fig. 3). The statistical analysis showed the significantly ($p \leq 0.05$) higher weight loss in chemical treated fabric than that of biological treatment. This test demonstrated the biological stripping as a superior one as compared to chemical stripping to maintain the quality of fabric in terms of weight.

It can be concluded that the enzymatic processing is gentle treatment and does not reduce the weight of fabric during process whereas chemical treatment can damage the fabric and thus reduce the weight. The enzymatic treatment/processing of textile fabric preserve the fiber's structure and strength, and avoid the high energy consumption and severe pollution problems that are associated with conventional alkaline treatments (Abdel-Halim et al., 2008; Sawada et al., 2003). The results of this study indicate that *G. lucidum* IBL-05 has the capability to strip the color from cotton dyed with Reactive black B. This study can furnish an idea to develop an environment friendly bioprocess for color stripping to be implemented in textile industries to minimize the problem of water pollution.

4. Conclusions

The work presented in this research is absolutely new direction in the field of textile, claiming the enzymatic processing of fabric as an environment friendly approach that might be implemented in practical field by further investigating the textile aspects of this method. The results of the study are expected to form the basis for development of economical and environment friendly process for color stripping of cotton fabrics dyed with reactive dyes that could furnish an idea to replace the harsh chemical stripping with biological stripping. In addition to this it may be the initiation of biotechnological approach for the safe recycling of old fabric into low priced fabric.

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