Environmentally-Friendly Dyeing of Cotton Yarn with Natural Indigo Dye

Noman Junejo[1], Dr. Awais Khatri[2], Dr. Shamshad Ali[2] and Farah Ishaque[2]

[1]*Institute of Environmental Engineering & Management, Mehran UET-Jamshoro, Sindh, Pakistan*
[2]*Color Research Lab, Department of Textile Engineering, Mehran UET-Jamshoro, Sindh, Pakistan*

***Abstract*: At present, the textile industrialists of Pakistan are increasing their capital investments into denim manufacturing and processing due to increasing export value for international brands. Denims are usually dyed with synthetic indigo dyes which are harmful to the environment. This is because the wastewater discharge from denim indigo dyeing contains a high amount of biochemical oxygen demand (BOD), chemical oxygen demand (COD), total dissolved and suspended solids contents (TDS and TSS respectively). Therefore, the wastewater must be treated before it is drained to main streams to reduce the pollutants to the level which can be tolerated by the environment itself. Due to negative effects of synthetic dyes on human health and environment, the use of natural dyes has been reviving from cottage to mass industrial scale. This is because natural dyes are biocompatible with the environment, non-toxic, and particularly non-carcinogenic. The aim of the work presented in this paper was to dye cotton yarn lea with natural indigo dyes using indigenous indigo plants for denim dyeing industry of Pakistan in collaboration with Archroma Pakistan Ltd. The dyeing of cotton warp yarn for denim was carried out by existing dyeing methods adopted by denim manufacturing industry. Dyeing and wastewater characterization results of natural dye were compared with the synthetic indigo dye. It was found that natural indigo dye shows better color strength (*K/S* value) as ever before; and the COD and BOD contents were reduced from 1410 mg/L to 783 mg/L (44% reduction) and from 384 mg/L to 176 mg/L (54% reduction) respectively, which makes the natural indigo dye more environmentally-friendly.**

***Keywords*: environmentally-friendly dyeing, Cotton Yarn, Dyeing, Natural Indigo dye, Wastewater pollution**

I. INTRODUCTION

Textile industry utilizes large amounts of water and is one of the top most water polluting industries in terms of color as well as chemical composition of residual wastewater due to various residues. Textile wastewater contain considerable amount of organic as well as inorganic compounds [1, 2]. Especially in dyeing processes, the dyes that are applied to the yarn are not fixed on yarn some remains unfixed to the yarn and gets washed out. Table 1 shows the amount of unfixed dye of different dyes. In case of indigo dye as a type of vat dye around 5-20% remains unfixed [3]. The wastewater coming from dyeing industries contain the unfixed dye and chemicals which are non-biodegradable and carcinogenic and have an adverse effect to health and the environment. This can affect the aesthetic merit, light penetration through water reduces photosynthetic activity, causing oxygen insufficiency and de-regulating the biological cycles of aquatic biota [4]. Various primary, secondary and tertiary treatment processes such as flocculation, trickling filters and electro dialysis have been used to treat wastewater. But the dyeing wastewater generated from the textile dyeing industry is one of the most difficult wastewater to be treated [5, 6]. Unfortunately, the conventional waste treatment techniques do not clean and decolorize the wastewater completely [7].

Table 1: Percentage of unfixed dyes in dyeing of different fibrous materials[3]

Fiber	Dye Type	Unfixed Dye %	Fiber
Cotton and Viscose	Azoic dyes	5-10	Cotton and Viscose
	Direct dyes	5-20	
	Vat dyes	5-20	
	Reactive dyes	20-50	
	Sulphur dyes	30-40	
Wool and Nylon	Acid dyes	7-20	Wool and Nylon
	Premetallized dyes	2-7	
Polyester	Disperse	8-20	Polyester

The world consumption of synthetic dyes is increasing significantly year by year. Vat textile dyes, especially indigo, play an important role in today's dyeing industry because of increasing demand for denim production [8]. Indigo dye is one of the most numerous marketed organic colorants used for coloration of textiles, leather, and paper, plastic and for other applications such as food, cosmetic, drug [9]. The annual consumption of indigo and other vat dyes extents up to the 33 million kg [10, 11]. Synthetic dyes generally have a complex chemical structure. Some of dyes or the product of dyes are proven to be carcinogens and mutagens containing high coloration, chemical oxygen demand (COD), biological oxygen demand (BOD) and salinity and if discharge of this industry is directly feed into the streams without appropriate treatments then it makes the water toxic and unfit for human and animal consumption, and can also cause imbalance within different aquatic ecosystem food chains [12-14].

Due to the cacogenic, toxic and non-biodegradable nature of synthetic dyes and drawbacks associated with conventional treatment methods and poor removal efficiency of pollutants from wastewater, the use of natural dye has been reviving. Among the natural dye, indigo is one of the oldest known natural dyes [15]. This research reveals that through Natural Indigo dyeing better color yield (*K/S*) value can be achieved and other valuable advantage of using natural indigo dye is that the dyeing wastewater contains low chemical oxygen demand (COD) and biological oxygen demand (BOD) as compare to the synthetic indigo dyeing wastewater. This is because of the biocompatible and nontoxic nature of natural dye with environment which makes it more environmentally friendly. Waste produced during production of natural indigo is mostly plant polymers and can serve as a large source of biomass energy, which are easily biodegradable and if cultivated in larger area the plants also provides green cover which is good for the environment.

II. MATERIAL & METHODS

A. Material

Industrial 100% raw cotton yarn were provided by Archroma Pakistan Limited (APL), Karachi. Fresh natural Indigofera Tinctoria plants were cultivated at the Textile Engineering department of Mehran University of Engineering and Technology (MUET) Jamshoro, and natural indigo dye was developed in Color Research Lab of Textile Engineering department of MUET. Reagents like sodium hydroxide (48˚Be), sodium dithionite and Leonel-EHC were obtained from the Wet Processing Lab of the Textile Engineering department. Synthetic Indigo dye (powder form) was obtained from APL and used for comparison.

B. Methods Dye extraction

Conventional dye extraction technique was followed and further optimization was successfully done, in terms of liquor ratio, soaking time and temperature in order to increase the dye yield and color yield on cotton warp yarn. Due to protecting the intellectual property and confidential disclosure agreement with APL, the procedure of dye extraction is not discussed here.

Dyeing of Cotton Yarn

Before dyeing, yarn lea was pre-wetted in order to increase the dye penetration. This was carried out on lab horizontal padder (Mathis HF, Switzerland) by continuous method. Pre-wet solution was prepared in a 500mL glass beaker in which a lea of de-sized yarn lea was dipped for 30 seconds. The lea was then squeezed via padder nip (at 70% pickup) and then rinsed.

To carryout dyeing of cellulose based textile with indigo, it needs to be converted into water soluble (leuco-form)[16]. This was done by dye reduction using reducing agent (sodium dithionite) in presence of alkali (sodium hydroxide). Optimized dyeing procedure was adopted for dyeing of yarn lea that was developed in the lab. Table 2 shows the dyeing recipe for 10% shade which was adopted/followed.

Table 2. Dyeing recipe of 10% Shade

Dye	10g/L
Caustic Soda (48%)	6g/L
Sodium Hydrosulphite	13g/L

The dyebath was prepared (containing 10g, 13g and 6 of natural indigo dye, reducing agent and alkali respectively) and was kept at 50 ºC for 1 hour. The yarn lea was immersed into the dyebath for 30 seconds and padded through nip of a padder (at 75% pick-up). Padded yarn lea was then oxidized in air for 90 sec. The process was repeated for 4 times in the same dyebath in order to achieve the desired color yield.

C. Rinsing of dyed yarn lea

After dyeing, rinsing is necessary in order to remove the unfixed dyes from yarn lea. Dyed yarn lea was gently rinsed at room temperature for 2 min and dried. Filtered water was used for rinsing purpose.

D. Testing Color measurement

The *K/S* value of both i.e. synthetic and natural dyed yarn lea were was measured on a Datacolor Spectrophotometer 850. Each measurement was made at five different locations of the sample and averaged.

Germination Rate

The germination rate of a particular seed lot is a key indicator as to how that seed will perform in the field [17]. Simple germination test method prescribed in the methods and procedures manual published by CFIA was performed on two different seeds, one obtained from APL Jamshoro and second was from WWF. The experiment was carried out in the Color Research Lab.

Toxicological Testing

The major toxicity issue with synthetic indigo dye is the presence of carcinogenic compound in the dye, which is highly toxic to inhaling and skin [18]. Presence and contents of aniline and N-methylaniline compounds (potentially carcinogenic to human health), and other unwanted compounds, were tested at APL Karachi that included presence of aniline and N-methylaniline compounds, analysis of banned amines (test method: EN14362-1/3:2012), analysis of chlorinated organic carriers (test method:

In House Method), analysis of chlorinated phenols (test method: BS EN ISO 17070:2015) and ecological analysis for COD contents of dyes. Banned amines were tested by reductive cleavage followed by extraction and subsequent determination of released aryl amines using GC-MS. Chlorinated organic carriers and chlorinated phenols were tested by extraction/sonication, with acetone then subsequent determination of chlorinated organic carriers using GC-MS and acetylation, and with acetic anhydride then subsequent determination of chlorinated phenols using GC-MS, respectively.

Wastewater Testing

After rinsing wastewater of dyeing were collected from the rinsing bath in a 500mL bottle for wastewater analysis within 30 min. The analysis of wastewater was carried out mainly in Advance Water Quality Labs of Center of Advance studies in water (CAS-W) and of Institute of Environmental Engineering and Management (IEEM), MUET.The pH is the measure of acidity or basicity of an aqueous solution. The pH of dyeing wastewater was measured by using Digital pH Meter, at Color Research Lab, Textile Engineering Department, MUET Jamshoro. Total dissolved solids (TDS) content was measured by TDS measuring probe on conductivity/TDS meter (HACH Company) at Water Quality Lab of IEEM by using TDS meter. Whereas, total suspended solids (TSS) content was measured by photometric method on a Spectrophotometer DR 2000 (HACH Company) at same lab.

Calorimetric method was used for determining the COD of wastewater [19] at Advance Water Quality Lab of CAS-W. COD vials, heating block (RD125), colorimeter (HACH model: DR 900), and micro pipette were used for the test. Two vails was prepared for each wastewater and averaged. First, COD vials were prepared that contain about 2.5mL dyeing wastewater sample, 3.5mL sulphuric acid and 1.5mL potassium dichromate. After proper mixing the vials were kept in heating block for 2 hours at 150 ºC and then cooled for testing. Once sample was cooled the COD of each wastewater sample was determined by using Calorimeter. Winkler method was followed for determining the BOD of wastewater. Reagents such as ferric chloride, magnesium chloride, phosphate buffer solution and magnesium sulphate and 300mL BOD bottles, 1000mL glass beaker, 10mL measuring pipettes, DO probe (Multi Parameter 9630), incubator (capable of maintaining 20±°C of Lovibond) were used for the test at CAS-W. The initial DO Content was determined and recorded on a DO meter through probe and incubated in the BOD bottle in the dark for five days at 20℃. Then, final DO content was determined. The BOD of wastewater was calculated as per Winkler equation (Equation 1).

$$BOD = \frac{L_o - L_5}{P} \, (mg / L) \qquad (1)$$

where, L_o is the initial DO, L_5 is the fifth day DO and P is the volumetric fraction of sample.

III. RESULTS

A. Germination Rate

Fig. 1 shows the germination rate test results of two different seeds samples i.e. WWF seeds and Archroma seeds. Fig.1 (a) shows the first day germination whereas fig. (b) shows the seventh day germination. The germination rate of WWF seeds was about 100% which showed that almost all seeds were grown i.e. 10 seeds were germinate out of 10, whereas the germination rate of Archroma seeds was around 90% which means that 9 out of 10 seeds were germinate.

(a) First Day (b) Seventh Day

Fig. 1: Germination rate test result of WWF and Archroma seeds

B. Optimized parameters for dye extraction process

The extraction parameters of the process were studied and optimized (i.e. liquor ratio, soaking time and temperature) to produce information on their effect on the dye yield as well as on color yield. The optimized extraction parameters which obtain high dye yield and good color strength (*K/S* value) of cotton yarn lea dyed with 10 g/L natural indigo of about 0.6% and 26.895 respectively, is shown in Table 3.

Table 3: Optimized Extraction Parameters

S. No	Parameters	Optimized Parameters	Dye Yield (%)	Color Strength (*K/S*)
1	Liquor Ratio	1:6		
2	Soaking Time	1 hr.	O.66	26.895
3	Temperature	50ºC		

C. Toxicological Testing of Indigo Dyes Toxicity analysis

Table 4 shows the test results of contents of aniline and N-methylaniline compounds present in synthetic and natural indigo dyes. These compounds were not detected in the natural indigo dye, thus, making the dye nontoxic and health friendly.

Table 4: Presence of aniline and N-methylaniline in the indigo dye

S. No	Sample	Result (mg/Kg)	
		Aniline	Aniline
1	Natural Indigo Powder	n.d.	n.d.
2	Synthetic Indigo Powder	6100.58	3378.75

Other Toxic Components in Indigo Dye

The optimized natural indigo dye was analyzed further in order to detect other toxic components. Tables 5, 6 and 7 (obtained from APL) show test results of analysis of banned amines, chlorinated organic carriers and chlorinated phenols respectively. It can be seen from table 5 that natural indigo dye does not contain any banned amines which makes the dye more environmental friendly. It can be seen from table 6 and 7 that natural indigo dye does not contain any Chlorinated organic carriers which is toxic.

Table 5: Analysis of Banned Amines

S. No	Cleavable/Carcinogenic Aryl amines	CAS no.	Results (mg/Kg)
1	2.4.5-Trimethyl aniline	137-17-7	n.d.
2	2,4-Diaminoanisole	615-05-4	n.d.
3	2,4Toluylenediamine	95-80-7	n.d.
4	2,4 Xylidine	95-68-1	n.d.
5	2,6 Xylidine	87-62-7	n.d.
6	o-aminoazotoulene	97-56-3	n.d.
7	2-Naphthylamine	91-59-8	n.d.
8	3,3'- Dichlorobenzidine	91-94-1	n.d.
9	3,3'- Dimethyloxibenzidine	119-90-4	n.d.
10	3,3'- Dimethyl-4,4'-Diamainodiphenylmethane	838-88-0	n.d.
11	3,3'- Dimethylbenzidine	119-93-7	n.d.
12	4,4 – Diamainodiphenylmethane	101-77-9	n.d.
13	4,4 – Methylene-bis(2-chloraniline)	101-14-4	n.d.
14	4,4 – Oxydianiline	101-80-4	n.d.
15	4,4 – Thiodianiline	139-65-1	n.d.
16	– Aminodiphenyl	92-67-1	n.d.
17	– Chloro-o-toluidine	95-69-2	n.d.
18	Benzidine	92-87-5	n.d.
19	o- Anisidine	90-04-0	n.d.
20	o- Toluidine	95-53-4	n.d.
21	– Chloroaniline	106-47-8	n.d.
22	p – Kersidine	120-71-8	n.d.
23	–aminoazobenzene	60-09-3	n.d.
24	– nitro-o-toluidine/2-Amino-4nitrotoulene	99-55-8	n.d.

25	Aniline	62-53-3	n.d.
26	P – phenylenediamine	16-50-3	n.d.

Table 6: Analysis of Chlorinated Organic Carriers

S.No.	Chlorinated organic carriers	Results (mg/Kg)
1	Chlorotoluene	n.d.
2	Chlorotoluene	n.d.
3	Chlorotoluene	n.d.
4	1,3-dichlorobenzene	n.d.
5	1,4-dichlorobenzene	n.d.
6	1,2-dichlorobenzene	n.d.
7	2,4-dichlorotoulene	n.d.
8	2,5-dichlorotoulene	n.d.
9	2,6-dichlorotoulene	n.d.
10	1,3,5-trichlorobenzene	n.d.
11	Sum of 2,3-dichlorotoulene & 3,4-dichlorotoulene	n.d.
12	1,2,4-trichlorobenzene	n.d.
13	1,2,3-trichlorobenzene	n.d.
14	2,4,5-trichlorobenzene	n.d.
15	2,3,6-trichlorobenzene	n.d.
16	1,2,3,5-tetrachlorobenzene	n.d.
17	1,2,4,5-tetrachlorobenzene	n.d.
18	1,2,3,4-tetrachlorobenzene	n.d.
19	Pentachlorobenzene	n.d.
20	Pentachlorotoluene	n.d.
21	Hexachlorobenzene	n.d.

Table 7: Analysis of Chlorinated Phenols

S. No.	**Chlorinated Phenols**	**CAS no.**	**Results (mg/Kg)**
1	2-CP	95-57-8	n.d.
2	3-CP	108-43-0	n.d.
3	4-CP	106-48-9	n.d.
4	2,6-DiCP	87-65-0	n.d.
5	3,5-DiCP	591-35-5	n.d.
6	Sum of 2,4 & 2,5-DCIP	120-83-2 583-78-8	n.d.
7	2,3-DiCP	576-24-9	n.d.
8	3,4-DiCP	95-77-2	n.d.
9	2,4,6-TCP	88-06-2	n.d.
10	2,3,5-TCP	933-78-8	n.d.
11	Sum 2,3,6 & 2,4,5-TCP	933-75-5 95-95-4	n.d.
12	Sum 2,3,4 & 3,4,5-TCP	15950-66-0 609-9-8	n.d.
13	2,3,5,6-TeCP	935-95-5	n.d.
14	2,3,4,6-TeCP	58-90-2	n.d.
15	OPP	90-43-7	n.d.
16	2,3,4,5-TeCP	4901-51-3	n.d.
17	PCP	87-86-5	n.d.

Ecological analysis

The comparative COD contents of the synthetic and natural indigo dyes is shown in Table 8. The results show that the COD content of natural indigo dye was reduced up to 46% as compare to that of the synthetic indigo dye.

Table 8: COD Content of Indigo Dyes

S. No.	Sample	COD (mg/g of O_2)
1	Synthetic Indigo	1158
2	Natural Indigo	624

D. Optimized parameters for dyeing

The effect of sodium hydrosulphite and sodium hydroxide in dyeing of natural indigo dye for obtaining high color yield on cotton yarn lea was optimized. As the natural dye is 100% concentrated whereas the synthetic dye was 30% diluted and pre-watted (pre-reduced form). Therefore, concentrations of sodium hydrosulphite and sodium hydroxide were optimized for better color yield.

Effect of Sodium Hydrosulphite Concentration on Color Yield

Fig. 2 shows the effect of sodium hydrosulphite (SHS) concentration on the color yield (*K/S* value) of yarn lea dyed with 10 g/L natural indigo dye. It can be seen from fig. 2 that the color yield is increased by increasing the concentration, maximum color yield was obtained at 13 g/L of sodium hydrosulphite, and the color yield decreased afterward by increasing sodium hydrosulphite concentration. The required concentration of sodium hydrosulphite varies with variation in the dye concentration, which can be practiced by maintaining the ratio of both concentrations (i.e. 13:10) obtained during this experimentation.

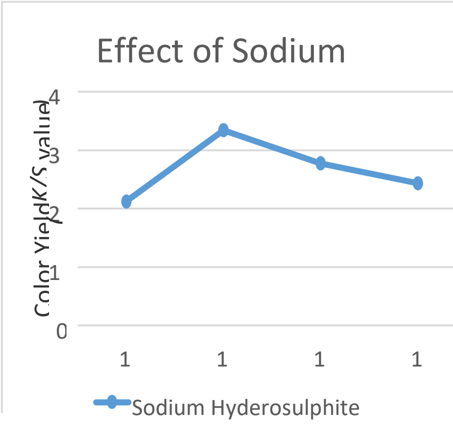

Fig. 2: Effect of SHS concentration on color yield

Effect of Sodium Hydroxide Concentration on Color Yield

Fig. 3 shows the effect of sodium hydroxide concentration on color yield of yarn leas dyed with 10 g/L natural indigo dye, whereas the SHS concentration was kept optimum (i.e. 13 g/L). It can be seen in the fig. 3 that with increasing concentration of sodium hydroxide the color yield increased up to 6 g/L then the yield decreased by further increase in the concentration. Therefore, 6 g/L was the optimum concentration, where pH of 11 – 11.5 (required for the process) was obtained. Further, it was concluded that the concentration of sodium hydroxide should always be around half of the concentration of SHS.

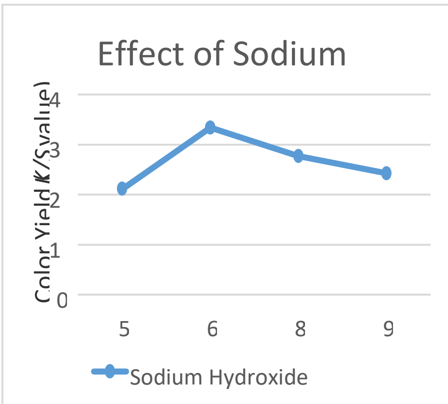

Fig. 3: Effect of sodium hydroxide on color yield

E. Color Yield (K/S value) of Indigo dyed yarns

The color yields of cotton yarn lea dyed of 10% shade with conventional natural indigo dye, optimized natural indigo dye and synthetic indigo dye is shown in Table 9. Higher *K/S* values indicate higher dyeing yield and deeper colors. Results reveals that the *K/S* value natural indigo is significantly increased up to 35% i.e. from 20.798 to 32.032.

Table 9: Color strength value

S. No	10% Shade	Max. *K/S*	*K/S* at 640 nm
1	Conventional Natural Indigo Dye	20.798	20.489
2	Optimized Natural Indigo Dye	32.032	29.146
3	Synthetic Indigo Dye	44.050	37.646

F. Wastewater Analysis

Table 10 shows similar and slightly alkaline pH (around 8) of the wastewaters from natural and synthetic indigo dyeings. The table also shows the TDS, TSS, COD and BOD contents of the dyeing wastewater of natural and synthetic indigo dyes. TDS content of both dyeing wastewaters were similar (around 200) whereas the TSS content of natural indigo dye was 88% lower than that of the synthetic indigo dye. The COD and BOD contents of wastewater from natural indigo dyeing were 44% and 54% lower than those of the synthetic dyeing respectively. The high levels of COD in synthetic dyeing wastewater indicates the high toxicity and the presence of massive amounts of biologically resistant organic substances in wastewater. Additionally, the excessive levels of BOD in synthetic dyeing wastewater indicate the pollutants strength in the water which also shows that very much less oxygen is available for the living organisms within the wastewaters [20-23].

Table 10 *Wastewater Analysis of Indigo dyeings*

S. No.	Wastewater	pH	TDS (mg/L)	TSS (mg/L)	COD (mg/L)	BOD (mg/L)
1	Natural Indigo Dye	8.37	240	93	783	176
2	Synthetic Indigo Dye	8.42	200	800	1410	384

IV. CONCLUSIONS

Dyeing of denim yarn lea can be successfully carried out with natural indigo dye with the procedure adopted for synthetic indigo dye. The natural indigo dye was found non-toxic in comparison to the synthetic dye, especially in terms of aniline compound and COD content. Through optimization of extraction and dyeing the color yield (*K/S* value) was significantly increased up to 35%. Additionally, it can be concluded from results that COD and BOD content was reduced up to 44% and 54% respectively. Results also concluded that TSS content in natural indigo dyeing wastewater was reduced up to 88% which makes natural indigo dye more environmentally friendly.

Furthermore, the test results it can also be concluded that the contents of aniline and N-methylaniline compounds is not present in natural indigo dye, thus, making the natural indigo dye nontoxic and health friendly.
It can also be concluded from Germination rate test that the WWF's seed were germinate 100% where as Archroma's germinate 90%.

V. RECOMENDATION

This study provides an opportunity to utilize the left over mass of plants for biomass energy.

REFERENCES

[1]. Elliott, A., W. Hanby, and B. Malcolm, *The near infra-red absorption spectra of natural and synthetic fibres.* British Journal of Applied Physics, 1954. **5**(11): p. 377.
[2]. Jia, J., et al., *Treatment of dyeing wastewater with ACF electrodes.* Water Research, 1999. **33**(3): p. 881-884.
[3]. O'Neill, C., et al., *Colour in textile effluents–sources, measurement, discharge consents and simulation: a review.* Journal of Chemical Technology & Biotechnology: International Research in Process, Environmental & Clean Technology, 1999. **74**(11): p. 1009-1018.
[4]. Apostol, L.C., et al., *Biological decolorization of xanthene dyes by anaerobic granular biomass.* Biodegradation, 2012. **23**(5): p. 725-737.
[5]. Kim, T.-H., et al., *Decolorization of dye solutions by a membrane bioreactor (MBR) using white-rot fungi.* Desalination, 2004. **168**: p. 287-293.
[6]. Tantak, N.P. and S. Chaudhari, *Degradation of azo dyes by sequential Fenton's oxidation and aerobic biological treatment.* Journal of Hazardous Materials, 2006. **136**(3): p. 698-705.
[7]. Puvaneswari, N., J. Muthukrishnan, and P. Gunasekaran, *Toxicity assessment and microbial degradation of azo dyes.* 2006.
[8]. Schrott, W., *Denim wieder im Blickpunkt der Textilindustrie.* Melliand Textilber, 2001. **82**: p. 331-337.
[9]. Zollinger, H., *Azo dyes and pigments.* Colour Chemistry-Synthesis, Properties and Applications of Organic Dyes and Pigments, 1987: p. 92-100. 10. Roessler, A. and X. Jin, *State of the art technologies and new electrochemical methods for the reduction of vat dyes.* Dyes and pigments, 2003. **59**(3): p. 223-235.
[10]. Roessler, A. and D. Crettenand, *Direct electrochemical reduction of vat dyes in a fixed bed of graphite granules.* Dyes and pigments, 2004. **63**(1): p. 29-37.
[11]. Banat, I.M., et al., *Microbial decolorization of textile-dye-containing effluents: A review (vol 58, pg 217, 1996).* Bioresource Technology, 1997. **61**(1): p. 103-103.
[12]. Riera-Torres, M., C. Gutiérrez-Bouzán, and M. Crespi, *Combination of coagulation–flocculation and nanofiltration techniques for dye removal and water reuse in textile effluents.* Desalination, 2010. **252**(1-3): p. 53-59.
[13]. Robinson, T., et al., *Remediation of dyes in textile effluent: a critical review on current treatment technologies with a proposed alternative.* Bioresource technology, 2001. **77**(3): p. 247-255.
[14]. Gilbert, K.G. and D.T. Cooke, *Dyes from plants: Past usage, present understanding and potential.* Plant growth regulation, 2001. **34**(1): p. 57-69.
[15]. Chakraborty, J. and R. Chavan, *Dyeing of denim with indigo.* 2004.
[16]. Soltani, E., et al., *Problems with using mean germination time to calculate rate of seed germination.* Australian Journal of Botany, 2016. **63**(8): p. 631-635.
[17]. Hassaan, M.A. and A. El Nemr, *Health and environmental impacts of dyes: mini review.* American Journal of Environmental Science and Engineering, 2017. **1**(3): p. 64-67.
[18]. Rice, E.W., et al., *Standard methods for the examination of water and wastewater*. Vol. 10. 2012: American Public Health Association Washington, DC.
[19]. Yusuff, R. and J. Sonibare, *Characterization of textile industries' effluents in Kaduna, Nigeria and pollution implications.* Global Nest: the Int. J, 2004. **6**(3): p. 212-221.
[20]. Geetha, A., et al., *Assessment of underground water contamination and effect of textile effluents on Noyyal River basin in and around Tiruppur Town, Tamilnadu.* Journal of Chemistry, 2008. **5**(4): p. 696-705.
[21]. Sawyer, C.N., P. McCarty, and G.F. Parkin, *Chemistry for environmental engineers.* New York. Mc Graw-Hill Book Company, 1978.
[22]. McMullan, G., et al., *Bioremediation and chemical analysis of textile industry waste water.* Biotechnol. Lett, 1995. **17**: p. 760-764.