International Research Journal of Environmental Sciences ISSN 2319–1414
Vol. **7(8),** 1-7, August **(2018)** *Int. Res. J. Environmental Sci.*

Microbial degradation of textile effluent and in genotoxic effect on *Allium cepa*

Maghdu Nainamohamed Abubacker*, Thirugnanam Mehala, Sadasivam Senthilkumar and Ramu Selvam
Department of Biotechnology, National College, Tiruchirappalli-620 001, Tamil Nadu, India
abubacker_nct@yahoo.com

Available online at: www.isca.in, www.isca.me
Received 27th November 2017, revised 3rd July 2018, accepted 14th August 2018

Abstract

Textile effluent discharged from the small scale industries was assessed. There was a total difference in the physico chemical characteristics of effluent sample and bacterial and fungal consortia degraded samples. The untreated effluent samples were highly colored, fishy odour, slight alkaline. The other physic chemical properties like total solids (TS) total suspended solids (TSS), total dissolved solids (TDS), biological oxygen demand (BOD), chemical oxygen demand (COD), dissolved oxygen (DO), hardness as $CaCO_3$ alkalinity, bicarbonate (HCO_3^-) alkalinity, chloride, calcium, magnesium, sodium, sulphate, zinc, chromium, copper and lead were found to be above to the permissible level of WHO standards, which ensure the presence of pollutants in the textile effluent. From the polluted sites, four bacterial strains which are identified as indicator bacterial strains able to degrade the effluent and dye were used for the decolourization and degradation in vitro experiments. They are Bacillus subtilis (NCBT 012), Clostridium butyricum (NCBT 017), Enterobacter aerogens (NCBT 024) and Pseudomonas fluorescens (NCBT 046). Four fungal strains which are identified as indicator fungi, able to decolourize effectively were selected for the decolourization and degradation in vitro experimental work. They are Aspergillus erythrocephalus (NCBT 124), Aspergillus fumigates (NCBT 126), Cladosporium herbarum (NCBT 142) and Fusarium oxysporum (NCBT 156). The physico-chemical analysis of bacterial and fungal consortia mediated textile effluent degradation process have shown reduction in all these parameters tested for the untreated textile effluent. Between bacterial consortium and fungal consortium mediated degradation process, the fungal consortium mediated degradation process has shown much reduction in all the parameters than the bacterial consortium mediated degradation. The genotoxicity studies in relation to mitotic index and chromosomal abnormalities have shown gradual increase in active mitotic index and reduction in the chromosomal abnormalities which ensures the reduction in toxicity of textile effluent by bacterial and fungal consortia degradation.

Keywords: Physico-chemical parameters, textile effluent, decolourization and degradation, genotoxicity.

Introduction

The untreated textile effluent contains chemicals like dyes, dispersant substances, leveling agents, acids and alkali were discharged into the nearby water bodies from textile industries without any treatment. This alters the physic chemical properties and makes the water bodies (rivers) intense colorations[1,4]. The presence of very small amounts of dyes as colouring agent in water is highly visible as intense colour and affects the aesthetic sense which alter water transparency and gas solubility in water bodies[15]. The treatment of textile effluents is still a major environmental problem because of synthetic dyes[8,11]. The synthetic dyes have varied structural properties that are not readely degradable and are not removed from contaminated source by conventional physical and chemical technologies such as coagulation, membrane filtration, precipitation, flotation, absorption, ion exchange and advanced chemical oxidation waste water treatment systems[1,13]. The main aim of this work was to study aerobic degradation of textile effluent and its major dye components containing Congo red, Crystal violet, Direct orange-102 and Malachite Green, used for dying thread fibres. These four dyes were used in the dying house during the course of the research work. The decolourization and degradation study with indicator bacterial and fungal consortia and the results were presented.

Materials and methods

Collection of samples: The area under the study for this research work was identified based on the need, diversity and extend of pollutants produced by the small scale textile dying houses located on the banks of the Amaravathy River, Karur, state of Tamil Nadu, and peppered with a number of large and small scale textile dying houses and industries. Textile effluent samples from the field receiving discharge from small scale cottage dying houses were collected. The samples were filled in properly cleaned, sterilized, wide mouth plastic bottles and stored in the dark at a temperature of 4°C, and utilized for physico-chemical decolourization and degradation studies. The soil samples for microbial isolation were collected at three sampling sites in the longitudinal section – Site-1: sample sources collected at the discharge site of the dying house; Site-2:

the samples collected 100 meters away from the discharge point; Site-3: 200 meters away from the discharge point. After thorough mixing, 200g representative samples from each point was collected and stored in polythene bags, transported to the microbiology research lab, PG and Research Department of Biotechnology, National College, Tiruchirappalli, in sealed, labeled containers and maintained a temperature of 4°C, to ensure minimal biological activity. Soil samples are processed in 24h for isolation, enumeration, identification and characterization of microbial flora. Another portion of the effluent and soil samples were acidified to pH ≤ 2 with Conc. HNO_3, refrigerated to prevent the volume change due to evaporation and the concentration of various heavy metal present in all the samples. A portion of moist soil samples were dried and sieved to remove the particles greater than 2 mm, homogenized and stored in a plastic bag for further analysis. The textile effluent and dyes Congo Red, Crystal Violet, Direct Orange-102, Malachite Green samples were obtained from small scale cottage dying house situated near the banks of Amaravathi River, Karur District, Tamil Nadu.

Physico-chemical Analysis of Textile Dying House Wastewater: Standard methods[29] were used for analyses of various physicochemical parameters of the textile effluent. The physico-chemical parameters like colour, odour, pH (pH meter Model EC1O), were recorded at the spot.

Results and discussion

The results of physico-chemical analysis of untreated textile effluent and degraded by bacterial and fungal consortia were presented in Table-1.

The bacterial consortium consists of Bacillus subtilis, NCTBT012 Clostridium butyricum, NCTBT017 Enterobacter aerogens NCTBT024 and Pseudomonas fluorescens NCTBT046. The fungal consortium consists of Aspergillus erythrocephalus NCTBT124, Aspergillus fumigatu NCTBT126, Cladosporium herbarumand NCTBT142, Fusarium oxysporium NCTBT156.

Physico-chemical effluent samples: Textile effluent discharged from the small scale industries was assessed. In the present investigation, there was a wide variation in the physico chemical characteristics of effluent sample and bacterial and fungal consortia degraded samples. The untreated effluent samples were highly colored, fishy in odour, alkaline and having moderate temperature, well above the limits[1]. Colour is regarded as the first contaminant to be recognized due to the presence of unused dye stuff in water, which affects the aesthetic as well as the biological activity of the ecosystem[2]. Photosynthesis active was affected due to dark coloration and disagreeable odor in water may be because of the presence of decaying vegetation, inorganic and organic materials in the effluent[3]. Temperature is an important ecological factor and an environmental variable, as it also fluctuates on daily or even hourly basis and it affects the other properties of waste water. It greatly influences on vital activities like metabolism, behavior, reproduction and development of microorganisms. Temperature was noted at the site prior to its collection and recorded to be 40°C at site1, where the effluent is discharged directly and recorded to be 37°C at the termination (site-III) and the same was also reported by Arun Prasad and Bhaskara Rao[4]. The pH was alkaline, TSS 1750mg/l and TDS 5875mg/l. Different industries have recorded different amount of solid particulate matter either as suspended solid or total dissolved solids in the effluent samples. The TS, TSS and TDS affect the light intensity of water, influencing turbidity and transparency. In the present study, TS was 2700 ± 0.84 for the untreated effluent and 1800 ± 0.21 for bacterial consortium bioremediated sample whereas it was 1700 ± 0.64mg/l for fungal consortium remediated sample. The TSS was 1780 ± 0.75mg/l for the untreated effluent, reduced to 1320 ± 0.63 and 950 ± 0.84mg/l for bacterial and fungal consortia mediated bioremediation samples respectively. The TDS 3420 ± 0.65mg/l for the untreated effluent, reduced to 2150 ± 0.84 and 1850 ± 0.41mg/l for bacterial and fungal consortia mediated bioremediation process respectively. The results are in line with Arun Prasad and Bhaskara Rao[4] as mentioned earlier and also with Furaha et al.[5], but the study results are opposing to Aklilu Asfaw[6], who has reported TDS 88.6 to 334.1mg/l. Avasan Maruthi and Ramakrishna[7] observed the TSS content of sugar mill effluent was 220 to 790mg/l. Senthil Kumar et al.[8] observed the discharged mill effluent showed total solids of 4485-1520 mg/l. The possible reason for the differences of total solids and subsequent dissolved solids are due to collision of the colliding particles, influenced by pH of the effluents. The EC is reduced from 4200 ± 0.80 in the untreated effluent to 2720 ± 1.21 for bacterial consortium bioremediated effluent and 2150 ± 0.63mhos/cm for fungal consortium remediated effluent. Jamaluddin Ahmed and Nizamuddin[9] reported EC range from 487 to 6225mhos/cm, falls in line with the present investigation.

The aquatic ecosystem completely depends on dissolved oxygen, various biochemical changes and its effects are due to the metabolic activities of microorganism. Many literature collectively indicated the importance of the dissolved oxygen concentration. The recommended DO in normal drinking water standard was 6mg/l. DO is an index of physical and biological process occur in water and the most important parameter is to assess the water quality, in measuring the molecular oxygen utilized during specified incubation period in which degradation of organic material and oxygen occur, which narmally used to oxidize as organic material. In general, BOD reflects the high concentration of substances that can be biologically degradable and consumption of oxygen by microbes, potentially results in dissolved oxygen. The results of the present study, i.e. 160 ± 1.23 for the untreated textile effluent, 42 ± 1.04 and 18 ± 0.54mg/l respectively for the bacterial and fungal consortia remediated samples show much less values when compare with the work of Okunade and Adekalu[10] who has reported BOD of Cassava industry varied between 70 to 290mg l^{-1}. Ameer Basha and Rajaganesh[11] reported BOD ranging between 243 to 1842mg/l in textile dye effluent.

Table-1: Physico-chemical analysis of textile dying house effluent using microbial consortium mediated degradation process.

Parameters	Unit	Untreated effluent sample*	Bacterial consortium degraded sample*	Fungal consortium degraded sample*	WHO standards
Colour	-	Tann	Light Tann	Light Tann	-
Odour	-	Unpleasant	No odour	No odour	-
pH	-	7.8 ± 0.21	7.2 ± 0.21	7.2 ± 0.15	7.0–8.5
Total Solids	Mg/l	2700 ± 0.84	1800 ± 0.21	1700 ± 0.64	500–1500
Total Suspended Solids	Mg/l	1780 ± 0.75	1320 ± 0.63	950 ± 0.84	100–600
Total Dissolve Solids	Mg/l	3420 ± 0.65	2150 ± 0.84	1850 ± 0.41	850–1500
Electrical Conductivity	µmhos/cm	4200 ± 0.80	2720 ± 1.21	2150 ± 0.63	500–1500
Biological Oxygen Demand	Mg/l	160 ± 1.23	42 ± 1.04	18 ± 0.54	≥ 5.0
Chemical Oxygen Demand	Mg/l	570 ± 1.41	260 ± 1.63	180 ± 0.48	250
Dissolved Oxygen	Mg/l	20 ± 0.23	12 ± 0.34	8.0 ± 0.72	≤ 6.0
Total Hardness as $CaCO_3$	Mg/l	1870 ± 0.64	1070 ± 0.81	780 ± 0.51	500
Carbonate alkalinity	Mg/l	36 ± 2.02	28 ± 1.02	28 ± 0.64	200
Bicarbonate alkalinity	Mg/l	370 ± 1.56	190 ± 1.02	170 ± 0.44	50
Chloride	Mg/l	980 ± 2.08	620 ± 1.06	580 ± 0.21	200–600
Calcium	Mg/l	330 ± 0.21	220 ± 0.42	210 ± 0.32	75–200
Magnesium	Mg/l	180 ± 0.48	140 ± 0.21	130 ± 0.46	50–150
Sodium	Mg/l	980 ± 0.84	520 ± 0.61	500 ± 0.81	1000–1500
Sulphate	Mg/l	520 ± 0.61	380 ± 0.61	320 ± 0.81	200–400
Nickel	Mg/l	1.06 ± 0.73	1.02 ± 0.43	1.00 ± 0.61	–
Zinc	Mg/l	1.12 ± 0.21	1.00 ± 0.21	1.00 ± 0.21	5.0
Chromium	Mg/l	4.38 ± 0.05	2.16 ± 0.31	1.80 ± 0.85	0.1
Manganese	Mg/l	1.24 ± 0.05	1.12 ± 0.21	1.10 ± 0.42	2.0
Copper	Mg/l	1.26 ± 0.02	0.10 ± 0.15	0.10 ± 0.01	0.2
Lead	Mg/l	0.80 ± 0.02	0.15 ± 0.22	0.10 ± 0.85	0.10
Iron	Mg/l	2.14 ± 0.15	1.82 ± 0.30	1.80 ± 0.45	4.5

*The mean difference is significant at 0.01 level.

In textile effluent, the presence of free chlorine and toxic heavy metals leading to an oxygen sag in receiving water. In other words, a high degree of pollution is associated with high COD[12]. In present study the COD ranges from 570 ± 1.41 in untreated textile effluent to 260 ± 1.63 mg/l for bacterial consortium bioremediared effluent and 250 ± 0.48 mg/l for fungal consortium remediated samples in a decreasing order due to bioremediation. However, high and low COD levels, 584 mg l^{-1} and 160mg l^{-1} have also been reported to textile effluents. Mohabansi et al.[2] reported that the COD levels in textile effluent released from textile mill are within the WHO standard limits, though BOD and other parameters were found to be high. Rajeswari et al.[13] reported high COD of 2865-997mg l^{-1} from textile dyeing effluent. If the Ratio of BOD: COD is above 0.5, the waste water is considered to be highly biodegradeble. Azbar et al.[14] reported textile waste waters have shown low BOD: COD ratios (<0.1), indicating non-biodegradable nature of dyes which is opposing to the present study.

The chloride content in the effluent also contributes to the increased TDS and found to be positively correlated with increased EC, TDS, TSS, %NaCl, alkalinity and sulfate content. The present study also agrees with the report and found to vary between 980 ± 2.08 for untreated sample, 620 ± 1.06 and 580 ± 0.21 mg/l for bacterial and fungal consortia remediated samples respectively. Pratibha Mahawar and Azra Akhtar[15] reported that physico-chemical properties like EC (1470-2200µS/cm), BOD (488-1090 mg l^{-1}) and COD (3120-6864mg l^{-1}) TDS (980-1440mg l^{-1}), Total hardness (230-2250mg l^{-1}) of the textile dye effluent were found to be higher than the WHO standards[1]. The Total Hardness (TH) of waste water which is a combined effect of Calcium (Ca^{2+}) and Magnesium (Mg^{2+}) metal cations present in effluent. The present study revealed 1870 ± 0.64 in the case of untreated effluent, whereas 1070 ± 0.81 for bacterial consortium and 780 ± 0.51mg/l for fungal consortium bioremediation of effluent. Okunade and Adekalu[10] reported a range of 195 to 376mg/l in different location of samples tested. According to Deepali Gangwar[16] the TH value was 676.95mg/l in Textile industry effluent. These results show much lesser value than the present investigation.

The major problem associated is the presence of heavy metal ions in Textile dyeing industries, which arise from material used in the dyeing process or in considerably higher amount from metal containing dyes. Metal based complex dyes release heavy metals are one of the source of pollutant added to nearby water bodies and the surrounding soil. The presence of heavy metals in the effluent samples were found to be above the recommended CPCB limits and guideline for irrigation. Metals like calcium, magnesium, nickel, sodium, zinc, chromium, manganese, copper, lead and iron have been detected in the untreated textile effluent. The concentration of these metals are much reduced during the biodegradation by both bacterial and fungal consortia. Calcium level 330 ± 0.21mg/l was reduced to 220 ± 0.42 and 210 ± 0.32mg/l, magnesium 180 ± 0.48mg/l to 140 ± 0.21, 130 ± 0.46mg/l, Nickel 1.06 ± 0.73mg/l to 1.02 ± 0.43, 1.00 ± 0.61mg/l, sodium 980 ± 0.84mg/l to 520 ± 0.61, 500 ± 0.89mg/l, zinc 1.12 ± 0.21mg/l to 1.00 ± 0.21, 1.00 ± 0.21 mg/l, chromium 4.38 ± 0.05mg/l to 2.16 ± 0.31, 1.80 ± 0.85 mg/l, manganese 1.24 ± 0.05mg/l to 1.12 ± 0.21, 1.10 ± 0.42 mg/l, copper 1.26 ± 0.02mg/l to 0.10 ± 0.15, 0.10 ± 0.01mg/l respectively for untreated effluent and treated effluent by bacterial and fungal consortia. Lead level 0.80 ± 0.017mg/l was reduced to 0.15 ± 0.22 and 0.10 ± 0.85mg/l, iron 2.14 ± 0.15mg/l to 1.80 ± 0.30 and 1.80 ± 0.45mg/l[17,18].

Genotoxic Studies of Textile Effluent, Bacterial and Fungal Consortia Degraded Effluent grown Allium cepa: The impact of Textile effluent as well as bacterial and fungal consortia mediated degraded effluent was tested for genotoxic effects. The results of mitotic index and chromosomal abnormalities due to genotoxic effect in Allium cepa was shown in Table-2 and Figure-1. A total of 500 cells were examined in all treatments, the control (deionised water) have shown 220 dividing cells and its Active Mitotic Index (AMI) percentage was 44.0 ± 1.50, the chromosomal abnormalities have shown vagrant and stickiness of chromosome and the Total Abnormality Percentage (TAP) was 1.2 ± 1.00. The textile effluent (20%) treatment have shown AMI percentage was 28.0 ± 0.45, the chromosomal abnormalities were chromosome break, chromosome bridge, C-mitosis, vagrant, chromosome loss and micro nuclei. The TAP was 7.2 ± 1.50. In the case of bacterial consortium mediated treatment, the AMI percentage was 34.0 ± 0.20, the chromosomal abnormalities were chromosome loss, localated nuclei, stickiness and binucleate cells. The TAP was 4.4 ± 0.45. In fungal consortium treated, the AMI percentage was 38.0 ± 1.50. The chromosomal abnormalities were chromosome break, C-mitosis, loculated nuclei and stickiness, the TAP was 2.8 ± 0.20. Results of Sabour et al.[19], Malik et al.[20] and Jaishree and Khan[21] are similar to the present study. They reported high amount of Cd, Cr, Cu, Ni and Pb and Zn from the fabric industry effluent. Similarly Joshi and Kumar[22] reported Cr (0-0.1126mg l^{-1}), Zinc (1.05 to 8.77mg l^{-1}), Pb (0.0029 to 0.1394), Co (0 to 0.0290mg l^{-1}), Ni (0 to 0.3721mg l^{-1}), Cu (0 to 0.3517mg l^{-1}), Ameer Basha and Rajaganesh[11] reported lead 11.12 to 32.10mg/l and Zn 13.03 to 36.13mg/l in the textile effluents are complied within the permissible limit in the present study. Studies conducted by Pratibha Mahawar and Azra Akhtar[15] reported the metal concentrations Zn (0.3-6.8 mg g^{-1}), Cu (0.8-48.65mg g^{-1}), Mn(4.3-11.5mgg^{-1}) and Fe(0.2-9.0mgg^{-1}) were found to be higher than the guidelines for irrigation. The active mitotic index refers to the frequency of cell division which is a vital parameters for determining the rate of root growth[23]. The results of genotoxic studies of the present study show that there is an increase in the active mitotic index percentage by the bacterial and fungal consortia degraded effluent showed 34.0 ± 0.20 and 38.0 ± 1.50 respectively when compare with the textile effluent, it was 28.0 ± 0.45%. The untreated dose of toxic substance reduce the active mitotic index whereas the degraded effluent show increase percentage of mitotic index seen in the present study are in line with the studies of Bianchi et al.[24], Kuchyet al.[25] and Bruna de Campose Ventura-Camargo et al.[26]. The total abnormality percentage was high (7.2 ± 1.50) for textile effluent, whereas the bacterial and fungal consortia degraded textile effluent has shown the reduction in the total abnormality percentage (4.4 ± 0.45 and 2.8 ± 0.20) respectively. The reduction in the abnormality percentage ensure the degradation and less toxic as reported by Bruna de Campose Ventura-Camargo et al.[26]. The chromosomal abnormalities like chromosome break, chromosome bridge, c-mitosis, vagrant, chromosome loss and micronuclei were recorded for untreated textile effluent. Whereas the bacterial consortium degraded sample has shown chromosome loss, loculated nuclei, stickiness and binucleate cells and the fungal consortium degraded sample has shown chromosome bridge, c-mitosis, loculated nuclei and stickiness. Such occurrence was confirmed by the earlier findings by Gul et al.[27] and Mustafa and Arikan[28].

Table-2: Genotoxic Studies: Mitotic Index and Chromosomal Abnormalities in Allium cepa.

Treatment	Total No. of cells Examined	No. of Dividing Cells	Active Mitotic Index % (AMI)	Percentage of Decolourization									Total no. of cells showing abnormal-ities	Total abnormali-ties (%) (TAP)
				CB	CBr	C-M	V	CL	MN	LN	S	BC		
Control (deionised water)	500	220	44.0±1.50*	0	0	0	3	0	0	0	3	0	6	1.2±1.00*
Textile effluent (20%)	500	140	28.0±0.45*	8	10	4	6	4	4	0	0	0	36	7.2±1.50*
Bacterial Consortium Degrated Effluent (20%)	500	170	34.0±0.20*	0	0	0	0	4	0	8	6	4	22	4.4±0.45*
Fungal Consortium Degrated Effluent (20%)	500	190	38.0±1.50*	0	4	2	0	0	0	4	4	0	14	2.8±0.20*

Note: CB: Chromosome break, CBr: Chromosome bridge, C-M: C-Mitosis, V: Vagrant, CL: Chromosome Loss, MN: Micro nuclei, LN: Loculated nuclei, S: Stickness , BC: Binnuclate cells * : The mean difference is significant at 0.01 level.

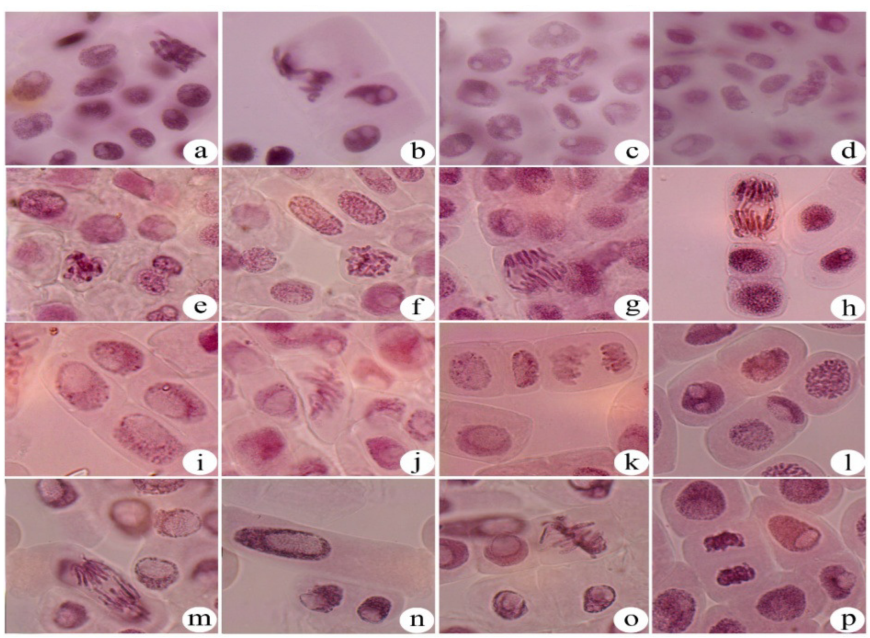

Figure-1: Genotoxic study of textile effluent Bacterial and Fungal degradation effluent (1000X).
a-h) Genotoxic effect of textile effluent: a) Chromosome break, b) Chromosome bridge, c) Mitosis, d) Vagrant, e) Chromosome loss, f) Micro nuclei, g) Chromosome break & bridge, h) Chromosome bridge. i-l) Genotoxic effect of Bacterial degraded Textile effluent: i) Loculated nuclei, j) Chromosome loss, k) Stickiness, l) Binucleate cells, m-p) Genotoxic effect of Fungal degraded Textile effluent: m) Chromosome bridge, n) Loculated nuclei, o) C-Mitosis, p) Stickiness

Conclusion

Disposal of dye effluents without proper treatment into the environment poses major hazards, through biomagnifications. The textile effluent used in the present study was ecotoxic since it extended hazardous effect to plant source namely Allium cepa. Monitoring of biodegradation by bacterial and fungal consortium rendered it an eco-friendly approach in reducing toxicity point of view.

Acknowledgements

The authors wishes to thank Padmavibhushan Dr. V. Krishnamurthy, President, Sri. K. Ragunathan, Secretary, Dr. K. Anburasu, Director and Dr. R. Sundararaman, Principal, National College, for providing the instrumentation facilities and supports to PG and Research Department of Biotechnology to complete this research work.

References

1. WHO (World Health Organization) (2001). Guideline for drinking water quality. *World Health Organization Press*, Geneva.
2. Mohabansi N.P., Tekade P.V. and Bawankar S.V. (2011). Physico-chemical parameters of textile mill effluent, Hinganghat, Dist. Wardha (MS). *Current World Environment*, 6(1).
3. Rohilla S.K., Salar R.K. and Kumar J. (2012). Optimization of physiochemical parameters for decolourization of reactive Black HFGR using soil fungus. *Aspergillus allhabadii* MTCC 9988, *Journal of Bioremediation and Biodegradation*, 3, 1-5.
4. Prasad A. and Rao K.V.B. (2011). Physicochemical analysis of textile effluent and decolorization of textile azo dye by Bacillus Endophyticus strain VITABR13. *Environ. Biotechnol*, 2(2), 55-62.
5. Furaha M.C., Kelvin M.M. and Karoli N.N. (2015). Assessment of Heavy Metals in Treated Wastewater Used for the Irrigation of Vegetable Plants in Arusha City. *International Journal of Research Chemistry and Environment*, 5(1), 54-60.
6. Asfaw A. (2014). Heavy metals concentration in tannery effluents, associated surface water and soil at Ejersa area of East Shoa, Ethiopia. *Herald J Geogr Reg Plan*, 3(3), 124-130.
7. Maruthi Y.A. and Rao S.R. (2001). Effect of Sugarmill Effluent on Organic Reserves of Fish. *Pollution Research*, 20(2), 167-171.
8. Kumar R.S., Swamy R.N. and Ramakrishnan K. (2001). Pollution studies on sugar mill effluent-physico-chemical characteristics and toxic metals. *Pollution Research*, 20(1), 93-97.
9. Jamaluddin Ahmed M. and Nizamuddin M. (2012). Physico-chemical assessment of Textile Effluents in Chittagong region of Bangladesh and their possible effects on environment. *International Journal of Research in Chemistry and Environment,* 2, 220-230.
10. Okunade D. A. and Adekalu K. O. (2013). Physico-chemical analysis of contaminated water resources due to Cassava wastewater effluent disposal. *European International Journal of Science and Technology,* 2(6), 75-84.
11. Basha S.A. and Rajaganesh K. (2014). Microbial bioremediation of heavy metals from textile industry dye effluents using isolated bacterial strains. *Int. J. Curr. Microbiol. Appl. Sci*, 3, 785-794.
12. Faryal R. and Hameed A. (2005). Isolation and characterization of various fungal strains from textile effluent for their use in bioremediation. *Pak. J. Bot.,* 37(4), 1003-1008.
13. Rajeswari K., Subashkumar R. and Vijayaraman K. (2013). Physico-chemical parameters of Effluents collected from Tirupur Textile dyeing and CETP and analysis of Heterotropic bacterial population. *Journal of Microbiology and Biotechnology Research,* 3, 37-41.
14. Azbar N., Yonar T. and Kestioglu K. (2004). Comparison of various advanced oxidation processes and chemical treatment methods for COD and colourremoval from a polyester and acetate fiber dyeing effluent. *Chemosphere,* 55, 35-43.
15. Mahawar P. and Akhtar A. (2015). Physico-chemical characterization of soil and effluent of dye industries in Kaithun region of Kota Rajasthan. *Int J Pure Appl Biosci*, 3(2), 419-422.
16. Gangwar Deepali K.K. (2011). Bioremediation of chromium (VI) from textile industry's effluent and contaminated soil using *Pseudomonas putida*. *Iranica Journal of Energy and Environment,* 2, 24-31.
17. Gabr R.M., Hassan S.H.A. and Shoreit A.A.M. (2008). Biosorption of lead and nickel by living and non-living cells of Pseudomonas aeruginosa ASU 6a. *International Biodeterioration & Biodegradation*, 62(2), 195-203.
18. Joshi V.J. and Santani D.D. (2012). Physicochemical Characterization and Heavy Metal Concentration in Effluent of Textile Industry. *Universal Journal of Environmental Research and Technology*, 2(2), 93-96.
19. Sabour B., Loudiki M., Oudra B., Vasconcelos V., Martins R., Oubraim S. and Fawzi B. (2002). Toxicology of a Microcystis ichthyoblabe waterbloom from lake oued Mellah (Morocco). *Environmental Toxicology: An International Journal*, 17(1), 24-31.

20. Malik A., Khan I. F. and Aleem A. (2002). Plasmid incidence in bacteria from agricultural and industrial soils. *World J Microbiol Biotechnol,* 18(9), 827-833.

21. Jaishree and Khan T.I. (2014). Physico-chemical analysis of contaminated soil collected from different areas of Sanganer textile industries Jaipur (Rajasthan). *International Journal of Geology, Earth and Environmental Sciences,* 4, 216-219.

22. Joshi N. and Kumar A. (2011). Physico-chemical Analysis of Soil and Industrial Effluents of Sanganer Region of Jaipur Rajasthan. *Res J of Agri Sci,* 2(2), 354-356.

23. Mathur N., Bhatnagar P. and Verma H. (2006). Genotoxicity of vegetables irrigated by industrial waste water. *J of Environ Sci*, 18(5), 964-968.

24. Bianchi J., Fernandes T.C.C. and Marin-Morales M.M.A. (2016). Induction of mitotic and chromosomal abnormalities on *Allium cepa* cells by pesticides imidacloprid and sulfentrazone and the mixture of them. *Chemosphere*, 144, 475-483.

25. Kuchy A.H., Waire A.A. and Kamili A.N. (2016). Cytogenetic effects of three commercially formulated pesticides on somatic and germ cells of *Allium cepa. Environ. Sci. Pollut. R.*, 23(7), 6895-6906.

26. de Campos Ventura-Camargo B., de Angelis D.D.F. and Marin-Morales M.A. (2016). Assessment of the cytotoxic, genotoxic and mutagenic effects of the commercial black dye in *Allium cepa* cells before and after bacterial biodegradation treatment. *Chemsphere,* 161, 325-332.

27. Tartar G., Kaymark F. and Murati F.D.G. (2006). Genotoxic effects of avenoxan on Allium cepa L. and Allium sativum L. *Caryologia*, 59, 241-247.

28. Mustafa Y. and Arikan E. S. (2008). Genotoxicity testing of Quizatofop-p-ethyl herbicide using the *Allium cepa* anaphase-telophase chromosome aberration assay. *Caryologia*, 61, 45-52.

29. APHA. (2005). Standard methods for the examination of water and waste water. 21st Ed. American Public Health Association, Wahington DC.