BIOREMEDIATION OF POLLUTED WATERS USING NANOPARTICLES

M.Meenakshi, K.Nishanthini

Abstract: Water pollution is an issue of great concern worldwide, contamination by organic compounds, inorganic compounds and microorganisms. Bioremediation using microorganisms helps in the removal of toxic metals from the environment. The focus is on the heavy metals associated with environmental contamination, lead (Pb), cadmium (Cd), and chromium (Cr) which are potentially hazardous to ecosystems. In the present study textile effluent was collected, and subjected to Physicochemical treatment methods , Herbal-Metal nanocomposite was prepared and used to treat textile effluents. As a bioremediation study, the plant growth potential of treated effluents was evaluated using pot studies of an aquatic plant .Laboratory and field test results confirmed superior bioremediation efficiency and long-term effect. When compared to today's most-efficient bioremediation technologies there is an efficient, fast, safe, and inexpensive way to clean up polluted waters through acceleration of natural bioremediation process. Nanotechnology provides an economical, convenient and ecofriendly means of wastewater remediation. The results obtained in this study shall be carried out as future studies using different types and concentrations of nanoparticles for the treatment of any types of effluents causing land and water pollution. There is a growing need for the development of novel, efficient, eco-friendly, and cost-effective approach for the remediation of inorganic metals released into the environment and to safeguard the ecosystem. In this regard, recent advances in microbes-base heavy metal have propelled bioremediation as a prospective alternative to conventional techniques.

Keywords : Bioremediation, Heavy Metals, Nanotechnology, Polluted waters.

1.INTRODUCTION

Environmental contamination by heavy metals from anthropogenic and industrial activities has caused considerable irreparable damage to aquatic ecosystems. Sources include the mining and smelting of ores, effluent from storage batteries and automobile exhaust, and the manufacturing and inadequate use of fertilizers, pesticides, and many others. The metals and metalloids that contaminate waters and are most commonly found in the environment include lead, chromium, mercury, uranium, selenium, zinc, arsenic, cadmium, silver, gold, and nickel. These metals are the subject of concern due to their high toxicity. Apart from being hazardous to human health, they also have an adverse effect on the fauna and flora, and they are not biodegradable in nature.

The products of sugar cane industry are crystalline sugar and bio-ethanol. The latter is obtained from the fermentation and distillation of sugar cane juice and molasses. Quantitatively, the most significant by-product is bagasse, the solid residue from sugar cane after juice extraction and stillage (also called vinnasse or dunder), the liquid waste effluent after the distillation process of sugar cane juice (Tewari et al., 2007).

In terms of wastewater treatment, nanotechnology is applicable in detection and removal of various pollutants. Heavy metal pollution poses as a serious threat to environment because it is toxic to living organisms, including humans, and not biodegradable (Mamadou and N. Savage, 2005).

BIOREMEDIATION

The Physicochemical treatment methods are more preferable due to their cost-effective and environmental friendly nature. These are considered the most effective tool for removal of wastes from wastewater loaded with organic constituents. In the mineralization of multifaceted organic molecules and recalcitrant nature of molecules, microorganisms play an important role [Anjaneyulu et al., 2015]. Biological treatment has advantage over physicochemical treatments methods about 70% of the organic materials are degraded by the biological treatment. To biodegrade synthetic dyes, bioremediation is a smart and easy operational way but found to be complex. Many microbial types have been evaluated for their aptitude to either decolorize or mineralize a range of dyes [Mahmoud, 2016]. Various fungal strains have been well reported to effect the dye concentration on rate of color removal. Elevated amounts of dyes may hinder cell development leading to a decrease in the speed of decolorization. Reports showed that degradation speed of dyes decreased owing to manufacture and gathering of poisonous products in the growth media [Sawant et al., 2017].

2. MATERIALS AND METHODS

The present research work was carried out to apply Bioremediation of polluted waters. The textile mill waste-water effluent was collected from a local cotton dyeing and processing mill, Tamil Nadu, India.

Collection and processing of medicinal herb – *Hemigraphiscolorata*

Hemigraphiscolorata, (Moorikooti) leaves were collected from a Nursery garden, Coimbatore, Tamil Nadu, India (Fig. A). The leaves were pre-washed in deionised water and dried with dry towel and then cut into smaller pieces. The leaves were pre-treated with blanching. The leaves were blanched in hot water (96–98°C) for 90s.

Fig. A: *Hemigraphiscolorata*, (Moorikooti) leaves

Sample preparation – Drying and milling of leaves (Lin et al., 2012)

The blanched leaves were separated and arranged in aluminium trays in each drying method. The leaves were dried using oven drying (40 °C for 7h, 50 °C for 6h, 60 °C for 4h and 120°C for 15min) using oven dryer until both the blanched leaves reached a moisture content of below 10%. The moisture content of 10% and below is the recommended value for drying of leaves and for powder production (Fig. B). The dried leaves were grounded to powder and the powder was then sieved manually by using sieve with size 250mm. Sieved particles were stored at room temperature prior testing.

Fig. B: Dried and Powdered leaves

Soxhlet extraction of *Hemigraphiscolorata* leaves using solvent (Saohin et al., 2007)

Extraction is the separation of medicinally active portions of plant using selective solvents through standard procedures. The purpose of all extraction is to separate the soluble plant metabolites, leaving behind the insoluble cellular residue. The initial crude extracts contain complex mixture of many plant metabolites, such as alkaloids, glycosides, phenolics, terpenoids and flavonoids. The initial stage in studying medicinal plants is the preparation of plant samples to preserve the biomolecules in the plants prior to extraction. Plants samples such as leaves, barks, leaves, fruits and flowers can be extracted from fresh or dried plants material. Other pre-preparation of plant materials such as grinding and drying also influences the preservation of phytochemicals in the final extracts. Some of the initially obtained extracts could be used as medicinal agents however most of plant extracts need further processing.

For the present study, soxhlet method which follows the principle of infusion method was chosen to extract the content from the given herbs. In the Soxhlet extraction method, finely ground sample – *Hemigraphiscolorata* leaf herbal powder was placed in a porous bag or “thimble” made from a strong filter paper or cellulose, which is placed, is in thimble chamber of the Soxhlet apparatus. Extraction solvent (acetone) is heated in the bottom flask, vaporizes into the sample thimble, and condenses in the condenser and drip back. When the liquid content reaches the siphon arm, the liquid contents is emptied into the bottom flask again and the process is continued. For the study, infusion method of Soxhlet Extraction had been adopted. The powdered herbs of *Hemigraphiscolorata* leaves were filled in the thimble and placed in the soxhlet extractor. The extractor had been filled with solvent solution of ethanol and the temperature of 60ºC was set and left for 6 hours. Slowly and steadily the temperature was increased upto 100ºC. The extract from the thimble was collected in the round botton flask kept in the heating mantle below by passing through a side arm tube (Fig. C, D). Thus collected extract was taken in a separating funnel and stored at room temperature prior to testing.

Fig. C: Soxhlet extraction of *Hemigraphiscolorata* leaves

Fig. D: Leaf extracts of *Hemigraphiscolorata*

Synthesis of Copper nanoparticles using chemical reduction method (Shende et al., 2016)

For the synthesis of copper nanoparticles, 50 ml of *Hemigraphiscolorata* leaf extract was mixed with 50ml aqueous solution of 1mM copper sulphate (1:1 ratio of plant extract and copper solution) and stirred continuously for 2min at 30^0C. The reduction takes place rapidly which is indicated by the change in colour of the solution. The mixture was incubated at room temperature overnight. The mixture was centrifuged at 3500rpm for 10min to get copper nanoparticles. The green synthesis of copper nanoparticles was achieved in aqueous solution using plant extract as reducing agent. The plant extract was mixed with copper sulphate solution, the colour of aqueous solution was changed immediately within 10 min, which turns dark brown within 24 hours (Fig) indicated the formation of copper nanoparticles. The nanoparticles were washed and dried at room temperature (Fig. E, F).

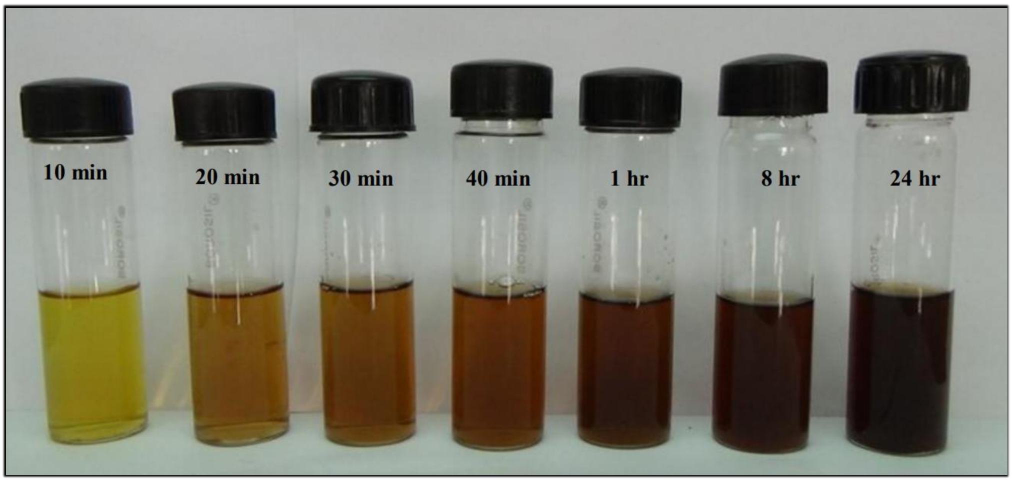


Fig. E: Copper nanoparticles synthesis step wise during different incubation period

Fig.F: Copper nanoparticles (Dried, powdered and sieved)

Development of Herbal-Metal nanocomposite (HM_{NC}) using standard stirrer method

Using the herbal extracts and copper nanoparticles, Herbal-Metal nanocomposite (HM_{NC}) was developed. The herbal extracts was kept under stirring conditions using a magnetic stirrer (110 rpm, 40ºC) in a beaker. Followed by copper nanoparticle solution was added drop wise onto the herbal extract at the rate of 20ml per minute. The magnetic stirring condition was kept constant for 2 to 3h till complete development of Herbal-Metal nanocomposite. The developed nanocomposites were stored in brown amber bottle at refrigeration temperature prior to antibacterial activity and other tests (Fig. G, H).

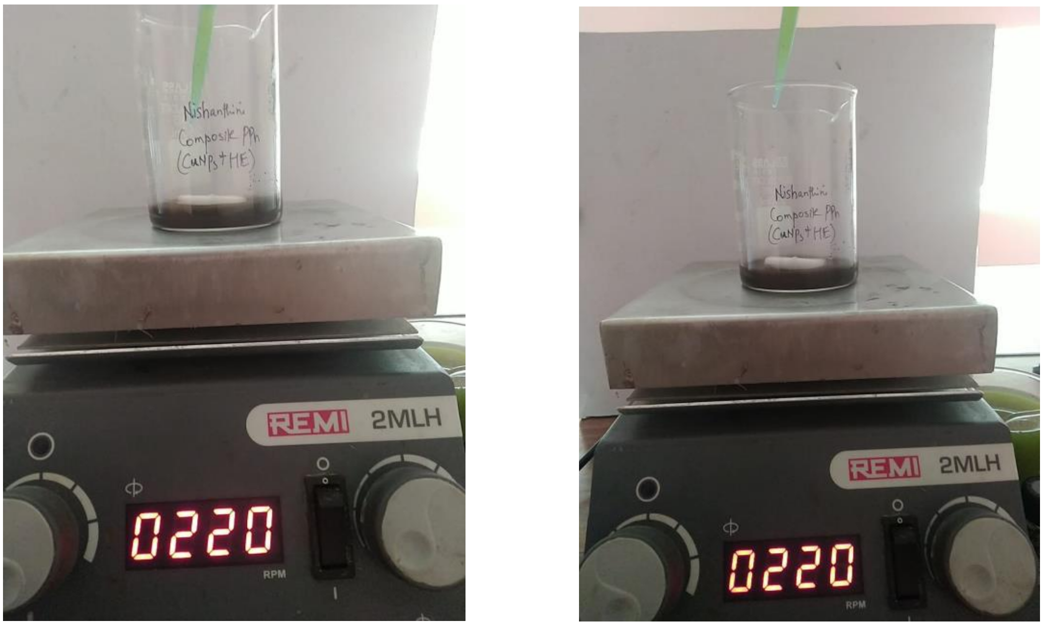


Fig. G: Herbal-Copper nanocomposite preparation

Fig. H: Synthesized Herbal-Copper nanocomposite

Textile mill Waste-Water effluent collection

Sample collection

About 2L of textile mill waste-water effluent was collected from a local cotton dyeing and processing mill, Tamil Nadu, India. The samples were observed for the presence of grease, oil etc as naked eye examination (Fig. I).

Fig. I: Textile mill Waste-water effluent

Bioremediation of Textile mill Waste-Water effluent using developed Nano-composites

After that the samples were collected in a plastic container closed tightly and brought to the laboratory for remediation process. The samples were separately added as 1L each in screw cap glass bottles (1L capacity). The first set of waste water (Fig. J) was added with Herbal-Metal nanocomposite (HM_{NC}); the second set was used as control.

Fig. J: Effluent added with Herbal-Metal nanocomposite (HM_{NC})

The setup was operated continuously for 30 days. Every five days, one flask of each sample was analyzed using standard procedures, described by APHA (2005), for the following; pH, turbidity, biochemical oxygen demand (BOD), chemical oxygen demand (COD) and colour removal. Each tests protocol was described below in brief.

Determination of pH

Twenty-five ml of wastewater was added to a 100 ml beaker and placed in magnetic stirrer and stirred well; pH of sample were recorded using a digital pH meter.

Determination of turbidity

Turbidity was measured using a digital Nephelometer (turbidity meter). The sample was mixed well and solid dispersed. The samples was kept until air bubbles disappeared and poured into the turbidimeter tube (10ml). The value was read based on the turbidity present in the sample by the instrument scale as Nephelometric Turbidity Unit (NTU).

Biochemical oxygen demand (BOD)

Biochemical oxygen demand of samples was determined by Dilution method using DO meter. Dissolved oxygen (DO) concentrations in a sample was measured before (Day 1 - D1) and after the 5 days incubation period (Day 5 - D2). The analysis was performed using 300ml incubation bottles in which buffered dilution water is dosed with waste water and stored for 5 days in the dark room (Incubator) at 20°C. The dilution water blank is used to confirm the quality of the dilution water that is used to dilute the other samples. BOD was calculated from following equation.

BOD (mg/l) = D1-D2/P

where : D1 = DO of diluted sample immediately after preparation, mg/l
D2 = DO of diluted sample after 5 days (incubation time), mg/l
P = decimal volumetric fraction of the sample used.

Determination of Chemical oxygen demand (COD)

Chemical oxygen demand was determined as described by Zupanc and Ros (2012). About 20ml of the sample was transferred into the reaction flask and 20mL of potassium dichromate solution [$K_2Cr_2O_7$] = 0.20 mol/L) was added. Silver sulphate-sulphuric acid (30mL) was added and the flask was attached to the condenser. The reaction mixture was heated on a heating mantle at 100ºC for 2 hours and then leaved to cool at room temperature. The condenser was rinsed with a small volume of deionised water and then removed; the reaction mixture diluted to 150mL. The excess dichromate is then titrated with ferrous ammonium sulphate (FAS) of concentration 0.50 mol/L. The blank test is carried out with distilled or deionised water (20 mL) in place of the sample. The chemical oxygen demand, COD, expressed in milligrams of oxygen per litre was calculated by the following expressions:

COD as mg O_2/L. = (A – B) x M x 800/ML sample

where: A = mL FAS used for blank,
B = mL FAS used for sample,
M = molarity of FAS, and
8000 = milliequivalent weight of oxygen × 1000 mL/L.

Plant growth potential of treated effluents – Pot studies of Aquatic plants (*Ludwigiarepens*)

In this experiment, the plant growth potential of treated effluents was evaluated using pot studies of an aquatic plant called *Ludwigiarepens*. Small leaflet plant with two leaves and a stem was placed in a garden soil filled in plastic disposable glasses. Normal water was added to the plantlets daily and recorded as Control (positive control). Similar set up was made and the soil was poured with treated effluents. In the third set of experimental cups, untreated effluent was added to the plantlets (Table-A). To calculate average mean ± standard deviation values, three plants for each experiment was performed. Parameters like number of leaves per plant, average shoot length, average root length and overall growth was selected for evaluation.

Table-A: Different parameters measured during plant growth potential studies

S. No.	Parameters analysed during the pot studies
1	No of leafs / plant
2	Average shoot length (cm)
3	Average root length (cm)
4	Overall Growth

3. RESULT AND DISCUSSION

In this present study, Herbal-Metal Nanocomposite (HM_{NC}) formulation treated textile mill waste-water showed distinct reduction in the tested parameters.

Sample processing:

Samples were blanched and dried. The extract was obtained using infusion method.

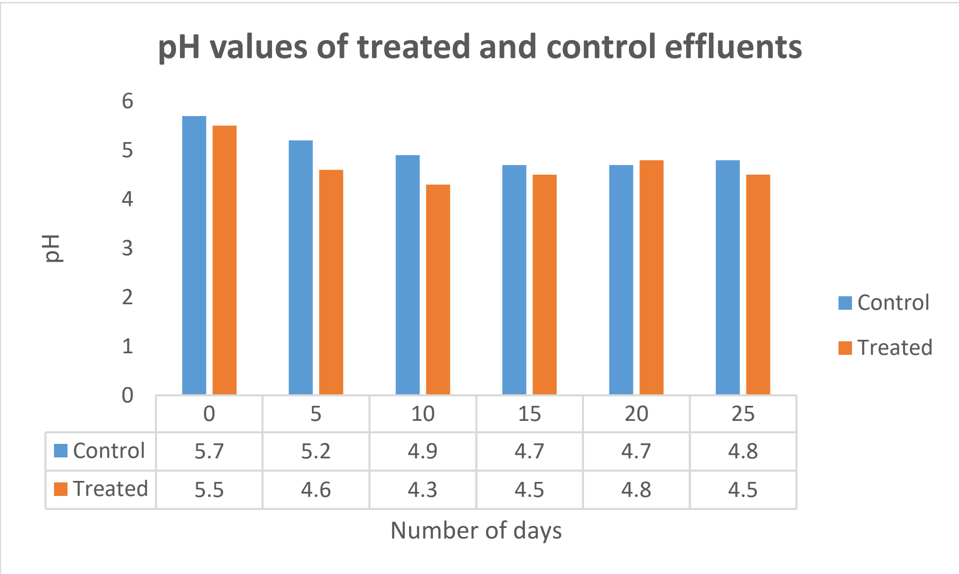
pH changes

The results of the pH analysis in industrial wastewater of a sugar factory when treated with Herbal-Metal Nanocomposite (HM_{NC}) were presented in Table-1 and Fig. 1.

Table-1: pH analysis of waste water treated with Herbal-Metal Nanocomposite (HM_{NC})

Duration of Treatment		0 days	5 days	10 days	15 days	20 days	25 days
pH	Control	5.7	5.2	4.9	4.7	4.7	4.8
	Nanocomposite treated	5.5	4.6	4.3	4.5	4.8	4.5

Fig. 1: pH analysis of waste water treated with Herbal-Metal Nanocomposite (HM_{NC})



The nanocomposite treated waste-water resulted in decreased level of pH values. This finding is compatible with Szymanski et al., (2003) experiment, which compared between 5 septic tanks treated with effective microbes and found that; after completion of the trial, the pH tended to drop to a lower level.

According to Higa and Chinen, (1998), we have to take into consideration that the pH of nanocomposite treated effluent was about 3.5 (below 4). So, these results indicate that the pH results of samples before the treatment of nanocomposite showed that the pH was in acidic condition. Betty and Winiarti, (1990) stated that, the lack of diluted oxygen in the wastewater due to the high content of organic matter can create malodor and muddy water. High content of protein, sulfur and phosphate will result in the formation of hydrogen sulfide, which can cause malodor and make the surrounding building black. Most of malodor arises from the degradation of nitrogen, sulfur, phosphate, protein and organic matter in the wastewater.

The bad odor smell was like mainly fermentation smell mixed with H_2S odor. As expected the odor removal was very efficient. Odor compounds e.g. derivatives of ammonia, hydrogen sulfide, methyl mercaptan and methyl sulfide, etc are usually produced through the metabolic action of putrefying microorganisms. Using the nanocomposite can suppress the action of these microbes and achieve breakdown of organic matter without producing odorous compounds.

Sanjay (2005) posted that effective microbes reduced the foul odor, and the treated effluent does not contain any odor. It suppresses the total coliforms and *Escherichia coli*. The nanotechnology based treatment method of industrial effluent treatment thus proved to neutralize the waste water.

Turbidity Changes

The turbidity value of the raw wastewater effluent collected from the textile mill processing industry. By time and addition of herbal-metal nanocomposite, the turbidity decreased dramatically during the first 15 days, and then increased again to reach double of the initial value. The reason for this turbidity increase is due to the organic matter in the waste water. The natural microbial load present in the effluent decomposed the organic materials in the wastewater and thus variation in turbidity was recorded. The results were given in Table-2 and Fig. 2a, b.

Table-2: Turbidity test of the nanocomposite treated waste-water

Duration of Treatment		0 days	5 days	10 days	15 days	20 days	25 days
Turbidity (NTU)	Control	425	390	345	350	355	440
	Nanocomposite treated	420	355	320	310	435	490

Fig. 2a: Turbidity test of the nanocomposite treated waste-water

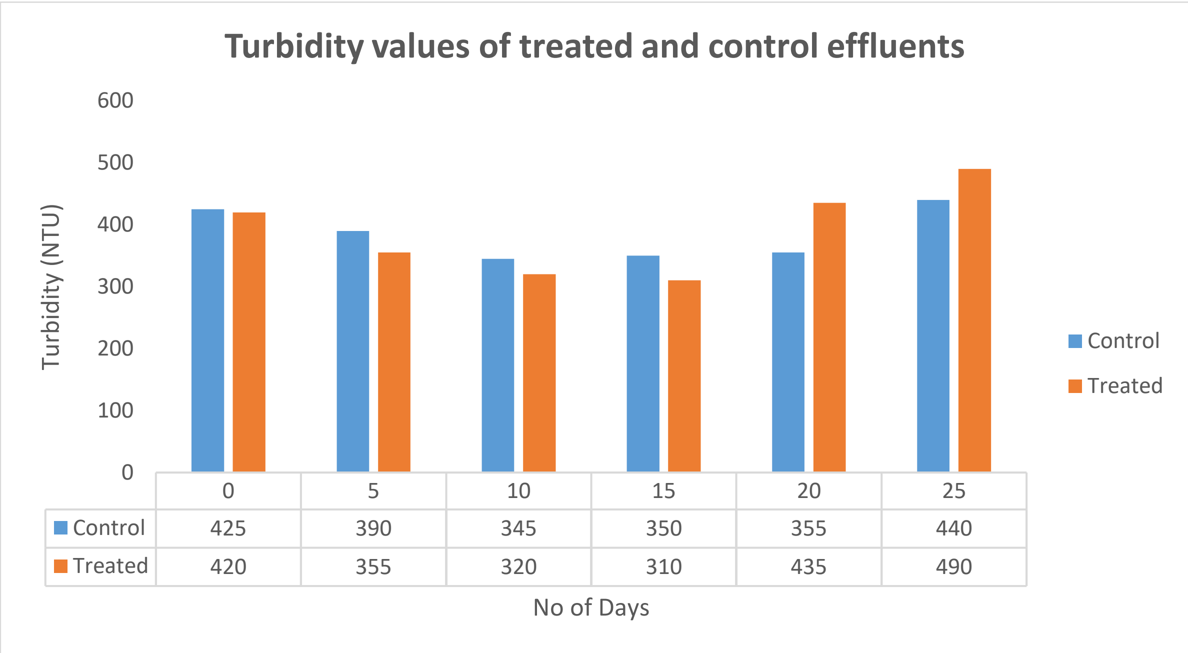


Fig. 2b: Turbidity test of the nanocomposite treated waste-water

Turbidity measurements from Day-0 to Day-25

Biochemical Oxygen Demand (BOD) and Chemical Oxygen Demand (COD) changes

The content of high organic matter in the wastewater can be used as a source of energy for the growth of microorganisms. The results of our study showed that, the Nanocomposite treatment affecting BOD and COD of the wastewater. During 25 days of treatment using nanocomposite and continuous application every 5 days, BOD and COD decreased between 5% and 70%, respectively.

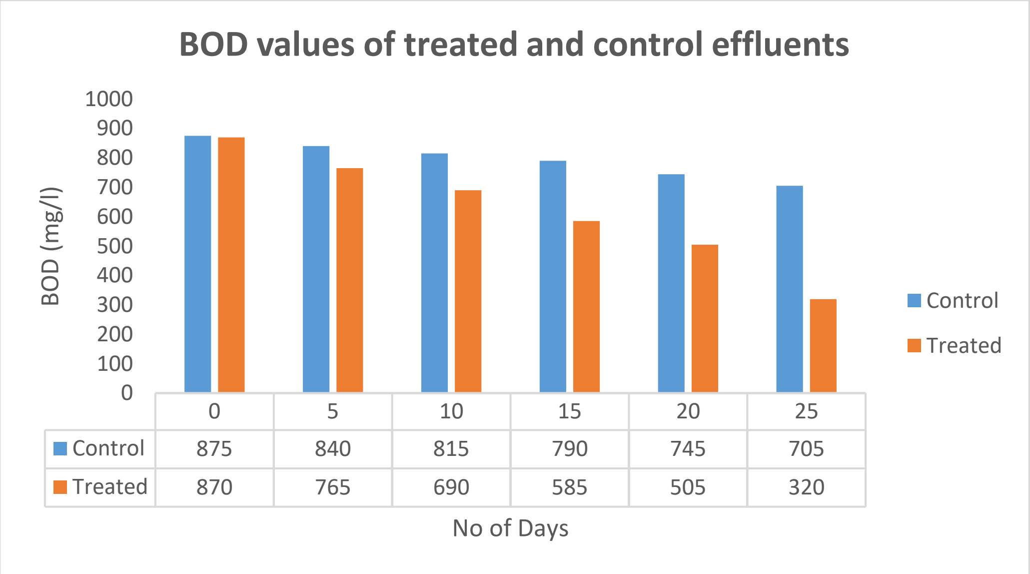
Effect of Herbal-Metal Nanocomposite on Biochemical Oxygen Demand (BOD) of treated effluents

The BOD value of the raw wastewater effluent collected from the textile mill processing industry was 780mg/l as an average. By addition of Herbal-Metal Nanocomposite, the BOD decreased dramatically throughout the time of study. The experiment showed that the inoculation of nanocomposite to the wastewater tend to decrease the BOD during the 25 days of treatment from 875 to 320 mg/l (about 75% reduction). The results were given in Table-3 and presented in Fig-3.

Table-3: Effect of Herbal-Metal Nanocomposite on Biochemical Oxygen Demand (BOD)

Duration of Treatment		0 days	5 days	10 days	15 days	20 days	25 days
BOD (mg/l)	Control	875	840	815	790	745	705
	Nanocomposite treated	870	765	690	585	505	320

Fig. 3: Effect of Herbal-Metal Nanocomposite on Biochemical Oxygen Demand (BOD)



From Table-3, It was evident that the values of BOD was decreased due to the action of Herbal-Metal Nanocomposite in the degradation of the organic compounds present in the effluent water. The present findings were found to be supportive with the work done by Higa (29). The researcher found that effective microbes was applied successfully for recycling of organic compounds of sewage and kitchen garbage. The result of fermentation by microbes was the formation of simpler organic compounds, such as amino acids, alcohol, sugars, organic acids and esters. It was also assumed that the fermentation process released active oxygen diluted in the wastewater that consequently activates the biochemical reactions.

Effect of Herbal-Metal Nanocomposite on Chemical Oxygen Demand (COD) of treated effluents

The COD value of the raw wastewater effluent collected from the textile mill processing industry was 945mg/l as an average. By addition of Herbal-Metal Nanocomposite, the COD decreased dramatically throughout the time of study. The experiment showed that the inoculation of nanocomposite to the wastewater tend to decrease the COD during the 25 days of treatment from 945 to 335 mg/l (about 70 % reduction). The results were given in Table-4 and presented in Fig.5. Before and after titration, the change in colour for the samples tested on 0^{th} day, 10^{th} day and 25^{th} day was presented in Fig. 6a, b, c. From the obtained values, it was evident that the values of COD was decreased due to the action of Herbal-Metal Nanocomposite in the degradation of the organic compounds present in the effluent water.

Table-4: Effect of Herbal-Metal Nanocomposite on Chemical Oxygen Demand (COD)

Duration of Treatment		**0 days**	**5 days**	**10 days**	**15 days**	**20 days**	**25 days**
COD (mg/l)	Control	945	970	1055	950	895	855
	Nanocomposite treated	950	890	790	720	425	335

Fig. 5: Effect of Herbal-Metal Nanocomposite on Chemical Oxygen Demand (COD)

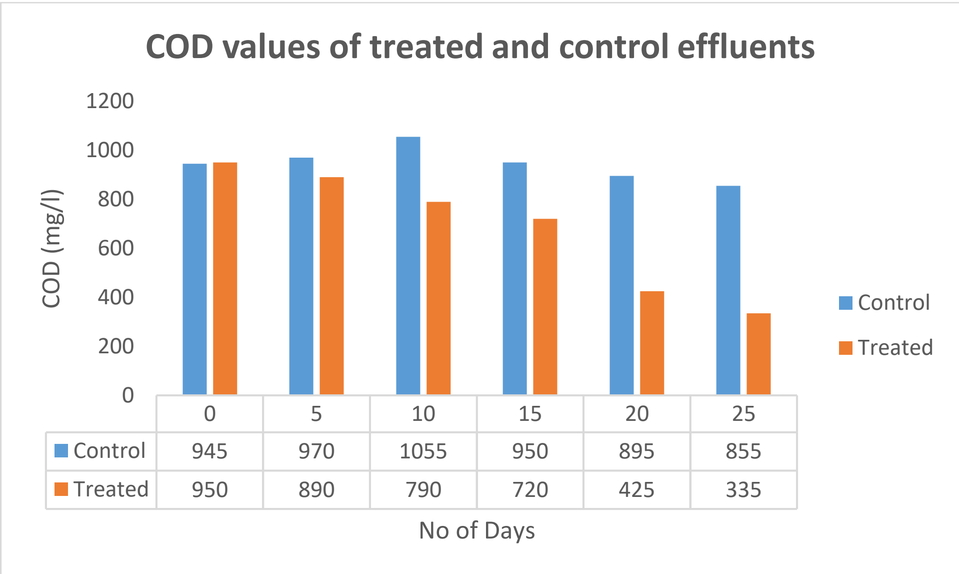


Fig. 6: Change in colour for the samples before and after titration

Fig. 6a: Before and after titration, the change in colour for the samples tested on 0^{th} day

Fig. 6b: Before and after titration, the change in colour for the samples tested on 10^{th} day

Fig. 6c: Before and after titration, the change in colour for the samples tested on 25^{th} day

Colour removal

The color removal from textile waste water seems to be one of the most challenging tasks to the environmental Engineers (Sirianuntapiboon and Srisornsak, 2007). Many techniques are available for removing the color. In the present research, the developed nanocomposite were found to be very efficient in removing the color from 60% to 95% during the period of studies. For untreated control samples, colour reduction was not evident; in contrast the nanocomposite treated effluents showed colour removal significantly from 0th day to 25th day with reduction of 93%. The effect of herbal-metal nanocomposite on the removal of effluent colour was depicted in Table-5. In Fig. 7a and b the colour removal was presented as graphical representation and as lab work test results respectively.

Table-5: Effect of Herbal-Metal Nanocomposite on colour removal of effluent

Duration of Treatment		0 days	5 days	10 days	15 days	20 days	25 days
Colour removal (%)	Control	60	55	55	60	55	60
	Nanocomposite treated	55	65	75	75	85	95

Fig. 7a: Graphical representation of Herbal-Metal Nanocomposite treated effluent and colour removal

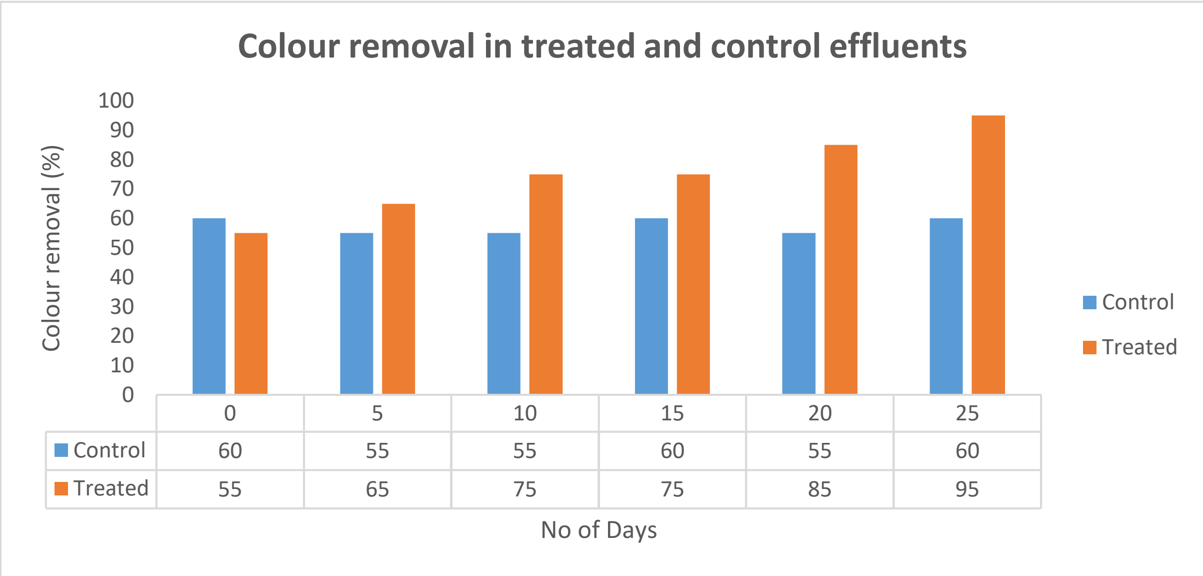


Fig. 7a: Effect of Herbal-Metal Nanocomposite on colour removal of effluents

Colour reduction in the effluent from 0th Day to 30th Day was evident from the above image

Plant growth potential of treated effluents – Pot studies of Aquatic plants (*Ludwigiarepens*)

In this experiment, the plant growth potential of treated effluents was evaluated using pot studies of an aquatic plant called *Ludwigiarepens*. Small leaflet plant with two leaves and a stem was placed in a garden soil filled in plastic disposable glasses. Normal water was added to the plantlets daily and recorded as Control (positive control). Similar set up was made and the soil was poured with treated effluents. In the third set of experimental cups, untreated effluent was added to the plantlets. To calculate average mean ± standard deviation values, three plants for each experiment was performed. Parameters like number of leaves per plant, average shoot length, average root length and overall growth was selected for evaluation.

Table-4: Plant growth potential of treated effluents – Evaluating number of leaves, shoots and roots

Plant type (Aquatic plant)	**Water and Effluent types (Before and after Treatments)**	**Number of leaves, shoots and roots**		
		No of leaves / plant	**Average shoot length (cm)**	**Average root length (cm)**
	Water treated (Control)	10.9 ± 0.57	7.3 ± 1.03	3.6 ± 1.03

Ludwigiarepens	Herbal-metal nanocomposite treated	9.6 ± 1.25	6.6 ± 0.76	3.3 ±0.76
	Untreated waste water effluent	0.0 ± 0.01	0.0 ± 0.01	0.0 ± 0.01

The plant growth promoting abilities in the nanocomposite treated water was compared with normal water and untreated waste-water. Among them, the pots with plants poured using normal water (control) showed more number of leaves (10.9 ± 0.57). In comparison to the control samples, the pots with plants poured using Herbal-metal nanocomposite treated water also showed significant numbers of leaves per plants (9.6 ± 1.25). But the pots poured with untreated effluent restricted the growth of plants and finally made all plants die (Fig. 8a, b, c). The average shoot length of the plants grown in control pots (water) showed increased number of shoots (7.3 ± 1.03); also significant increase in average shoot length of the plants grown in pots (Herbal-metal nanocomposite treated water) was found evident (6.6 ± 0.76). The average root length of the plants grown in control pots (water) showed almost similar lengths (3.6 ± 1.03); when compared to the average shoot length of the plants (3.3 ±0.76) treated with Herbal-metal nanocomposite.

Fig. 8: Plant growth potential of treated effluents under Green-house – Evaluating number of leaves, shoots and roots

Fig. 8a: *Ludwigiarepens* grown in the pot poured with water (control)

Fig. 8b: *Ludwigiarepens* grown in the pot poured with Nanocomposite treated water

Fig. 8c: *Ludwigiarepens* grown in the pot poured with untreated waste-water

4. SUMMARY AND CONCLUSION

. In terms of wastewater treatment, nanotechnology is applicable in detection and removal of various pollutants. Herbal-Metal Nanocomposite (HM_{NC}) was developed in the present research using *Hemigraphis colorata* and copper nanoparticles with the aim of treating waste-water effluent collected from a textile mill industry . It was very interesting to note that the turbidity which averaged 425 NTU at the beginning of the tests, decreased dramatically during the first 15 days by the addition of Herbal-Metal Nanocomposite and then increased again to reach double of the initial value. The BOD was reduced from 875 to 320 mg/l. The COD was decreased from 945 to 335 mg/l. In the present research, the developed nanocomposite were found to be very efficient in removing the color from 60% to 95% during the period of studies. For untreated control samples, colour reduction was not evident; in contrast the nanocomposite treated effluents showed colour removal significantly from 0^{th} day to 25^{th} day with reduction of 95%. As a bioremediation study, the plant growth potential of treated effluents was evaluated using pot studies of an aquatic plant called *Ludwigia repens*. The number of leaves, shoots and roots per plant was calculated after the growth period of each test plants in green house. Herbal-metal nanocomposite treated water showed significant numbers of leaves per plants (9.6 ± 1.25). The average shoot length of the plants grown in pots (Herbal-metal nanocomposite treated water) was found evident (6.6 ± 0.76). The average root length of the plants grown in control pots (water) showed almost similar lengths (3.6 ± 1.03); when compared to the average shoot length of the plants (3.3 ±0.76) treated with Herbal-metal nanocomposite. The results obtained in this study shall be carried out as future studies using different types and concentrations of nanoparticles for the treatment of any types of effluents causing land and water pollution.

5. REFERENCES

- Abdolmohammad-Zadeh, H., Ghorbani, E., Talleb, Z., Zinc–aluminum layered double hydroxide as a nano-sorbent for removal of Reactive Yellow 84 dye from textile wastewater effluents. J. Iran Chem. Soc. 10, 1103–1112, 2013.
- Adeleye, J. R. Conway, K. Garner, Y. Huang, Y. Su, and A. A. Keller, "Engineered nanomaterials for water treatment and remediation: costs, benefits, and applicability," Chemical Engineering Journal, vol. 286, pp. 640–662, 2016.
- Ali, A., Zafar, M.Z.H., Haq, I., Phull, A.R., Ali, J.S., Hussain, A., Synthesis, characterization, applications, and challenges of iron oxide nanoparticles. Nanotechnol. Sci. Appl., 9, 49, 2016.
- Alivisatos, A.P., Semiconductor clusters, nanocrystals, and quantum dots. Science, 271, 933–937, 1996.
- Amin, M.T., Alazba, A.A., Manzoor, U., A review of removal of pollutants from water/wastewater using different types of nanomaterials. Adv. Mater. Sci. Eng. 1-24, 2014.
- Anjaneyulu, Y., Chary, N.S., Raj, D.S.S., Decolourization of industrial effluents-available methods and emerging technologies. Rev. Environ. Sci. Biotechnol., 4, 245, 2005.
- Ataei-Germi, T., Nematollahzadeh, A., Bimodal porous silica microspheres decorated with polydopaminenano-particles for the adsorption of methylene blue in fixed-bed columns. J. Coll. Inter. Sci. 470, 172–182, 2016.
- Atta, A.A., Al-Lohedan, H.A., Ezzat, A.O., Tawfik, A.M., Hashem, A.I., Synthesis of zinc oxide nanocomposites using poly (ionic liquids) based on quaternary ammonium acrylamidomethyl propane sulfonate for water treatment. J. Mol. Liq. 236, 38–47, 2017.
- Azari, A.-A. Babaei, R. R. Kalantary, A. Esrafili, M. Moazzen, and B. Kakavandi, "Nitrate removal from aqueous solution using carbon nanotubes magnetized by nano zero-valent iron," Journal of Mazandaran University of Medical Sciences, vol. 23, no. 2, pp. 14–27, 2014.
- Banerjee, S., Dubey, S., Gautam, R.K., Chattopadhyaya, M.C., Sharma, Y.C., Adsorption characteristics of alumina nanoparticles for the removal of hazardous dye, Orange G from aqueous solutions. Arab. J. Chem. 2017
- Behnajady, N. Modirshahla, and R. Hamzavi, "Kinetic study on photocatalytic degradation of C.I. Acid Yellow 23 by ZnOphotocatalyst," Journal of Hazardous Materials, vol. 133, no. 1–3, pp. 226–232, 2006.
- Benhadji, A., Ahmed, M.T., Maachiv, R., Electrocoagulation and effect of cathode materials on the removal of pollutants from tannery wastewater of Rouiba. Desalination, 277, 128, 2011.

- Betty, SL and Winiarti, PR. (1990). PenangananLimbah industry Pangan. Kanisius, Yogyakarta. (148p). “Preliminary Experiment of EM Technology on Wastewater Treatment”, GedeNgurahWididana; Indonesian Kyusei Nature Farming Society, Indonesia.
- Bharathi, K.S., Ramesh, S.T., Removal of dyes using agricultural waste as low-cost adsorbents: a review. Appl. Water Sci. 3, 773–790, 2013.
- Bokare, J.-L. Jung, Y.-Y. Chang, and Y.-S. Chang, “Reductive dechlorination of octachlorodibenzo-p-dioxin by nanosized zero-valent zinc: Modeling of rate kinetics and congener profile,” Journal of Hazardous Materials, vol. 250-251, pp. 397–402, 2013.
- Bujoli, H. Roussiere, G. Montavon et al., “Novel phosphate– ` phosphonate hybrid nanomaterials applied to biology,” Progress in Solid State Chemistry, vol. 34, no. 2–4, pp. 257–266, 2006.
- Buzea, I. I. Pacheco, and K. Robbie, “Nanomaterials and nanoparticles: sources and toxicity,” Biointerphases, vol. 2, no. 4, pp. MR17–MR71, 2007.
- Chatterjee and B. L. Deopura, “Carbon nanotubes and nanofibre: an overview,” Fibers and Polymers, vol. 3, no. 4, pp. 134–139, 2002.
- Chatterjee, D., Patnam, V., Sikdar, A., Joshi, P., Misra, R., Rao, N.N., Kinetics of the decoloration of reactive dyes over visible light-irradiated TiO2 semiconductor photocatalyst. J. Hazard. Mater. 156, 435–441, 2008.
- Chen, Y. Li, M. Guo et al., “One-pot synthesis of Mn-doped TiO2 grown on graphene and the mechanism for removal of Cr(VI) and Cr(III),” Journal of Hazardous Materials, vol. 310, pp. 188–198, 2016.
- Chi, B.H.X., Yeap, S.P., Ahmad, A.L., Lim, J., Layer-by-layer assembly of iron oxide magnetic nanoparticles decorated silica colloid for water remediation. Chem. Eng. J. 243, 68–78, 2014.
- Ciardelli, G., Ranieri, N., The treatment and reuse of wastewater in the textile industry by means of ozonation and electroflocculation. Water Res., 35, 567, 2001.
- Danilczuk, A. Lund, J. Sadlo, H. Yamada, and J. Michalik, “Conduction electron spin resonance of small silver particles,” SpectrochimicaActa—Part A: Molecular and Biomolecular Spectroscopy, vol. 63, no. 1, pp. 189–191, 2006.
- Dankovich and D. G. Gray, “Bactericidal paper impregnated with silver nanoparticles for point-of-use water treatment,” Environmental Science and Technology, vol. 45, no. 5, pp. 1992–1998, 2011.
- Fujishima and K. Honda, “Electrochemical photolysis of water at a semiconductor electrode,” Nature, vol. 238, no. 5358, pp. 37–38, 1972.
- Ge, M.-M. Li, H. Ye, and B.-X. Zhao, “Effective removal of heavy metal ions Cd2+, Zn2+, Pb2+, Cu2+ from aqueous solution by polymer-modified magnetic nanoparticles,” Journal of Hazardous Materials, vol. 211-212, pp. 366–372, 2012.

- Gessner, T., Mayer, U., Ullmann'sEncyclopedia of Industrial Chemistry. Part A27. Triarylmethane and Diarylmethane Dyes. 6th edition. New York: WileyVCH, 2001.
- Giles, D.E., Mohapatra, M., Issa, T.B., Anand, S., Singh, P., Iron and aluminium based adsorption strategies for removing arsenic from water. J. Environ. Manag., 92, 3011, 2011.
- Girgis, B.S., Attia, A.A., Fathy, N.A., Potential of nano-carbon xerogels in the remediation of dye-contaminated water discharges, Desalination 265, 169–176, 2011.
- Guesh, A. Mayoral, C. M. ´ Alvarez, Y. Chebude, and I. D ´ ´ıaz, "Enhanced photocatalytic activity of TiO2 supported on zeolites tested in real wastewaters from the textile industry of Ethiopia," Microporous and Mesoporous Materials, vol. 225, pp. 88–97, 2016.
- Gugnami, A., Mishra, A., Textile & apparel compendium, Technopak, 2012.
- Guieysse, B., Norvill, Z.N., Sequential chemical–biological processes for the treatment of industrial wastewaters: review of recent progresses and critical assessment. J. Hazard. Mater., 267, 142, 2014.
- Gupta, S. Agarwal, and T. A. Saleh, "Chromium removal by combining the magnetic properties of iron oxide with adsorption properties of carbon nanotubes," Water Research, vol. 45, no. 6, pp. 2207–2212, 2011.
- Hao, O.J., Kim, H., Chiang, P.C., Decolorization of wastewater. Crit. Rev. Environ. Sci. Technol., 30, 449, 2000.
- Heidarizad, M., Sengor, S.S., Synthesis of graphene oxide/magnesium oxide nanocomposites with high-rate adsorption of methylene blue. J. Mol. Liq. 224, 607–617, 2016.
- Hossein, A.Z., Ebrahim, G., Zeynab, T., Zinc–aluminum layered double hydroxide as a nano-sorbent for removal of Reactive Yellow 84 dye from textile wastewater effluents. J. Iran Chem. Soc. 10, 1103–1112, 2013.
- Janotti and C. G. Van de Walle, "Fundamentals of zinc oxide as a semiconductor," Reports on Progress in Physics, vol. 72, no. 12, Article ID 126501, 2009.
- Javed, M., Usmani, N., Assessment of heavy metal (Cu, Ni, Fe Co, Mn, Cr, Zn) pollution in effluent dominated rivulet water and their effect on glycogen metabolism and histology of Mastacembelusarmatus. Springer Plus, 2, 390, 2013.
- Jerold, M., Joseph, D., Patra, N., Sivasubramanian, V., Fixed-bed column studies for the removal of hazardous malachite green dye from aqueous solution using novel nanozerovalent iron algal biocomposite. Nanotechnol. Environ. Eng. 1, 8, 2016.
- Kallman, V. A. Oyanedel-Craver, and J. A. Smith, "Ceramic filters impregnated with silver nanoparticles for point-of-use water treatment in rural guatemala," Journal of Environmental Engineering, vol. 137, no. 6, pp. 407–415, 2011.

- Khan, M.A., Uddin, M.K., Bushra, R., Ahmad, A., Nabi, S.A., Synthesis and characterization of polyanilineZr (IV) molybdophosphate for the adsorption of phenol from aqueous solution. React. Kinet. Mech. Catal. 113 (2), 499–517, 2014.
- Khandare, R., Kabra, A., Kadam, A., Govindwar, S., Treatment of dye containing wastewaters by a developed lab scale phytoreactor and enhancement of its efficacy by bacterial augmentation. Int. Biodeterior. Biodegrad., 78, 89, 2013.
- Khaydarov, R. R. Khaydarov, and O. Gapurova, "Water purification from metal ions using carbon nanoparticleconjugated polymer nanocomposites," Water Research, vol. 44, no. 6, pp. 1927–1933, 2010.
- Khin, A. S. Nair, V. J. Babu, R. Murugan, and S. Ramakrishna, "A review on nanomaterials for environmental remediation," Energy & Environmental Science, vol. 5, no. 8, pp. 8075–8109, 2012.
- Krishnaraj, R. Ramachandran, K. Mohan, and P. T. Kalaichelvan, "Optimization for rapid synthesis of silver nanoparticles and its effect on phytopathogenic fungi," SpectrochimicaActa— Part A: Molecular and Biomolecular Spectroscopy, vol. 93, pp. 95–99, 2012.
- Lai, T.L., Yong, K.F., Yu, J.W., Chen, J.H., Wang, C.B., High efficiency degradation of 4-nitrophenol by microwave-enhanced catalytic method. J. Hazard. Mater.,185, 366, 2011.
- Le, N.L., Nunes, S.P., Materials and membrane technologies for water and energy sustainability. Sust. Mat. Technol., 7, 1, 2016.
- Lee, C.S., Robinson, J., Chong, M.F., A review on application of flocculants in wastewater treatment. Process Saf. Environ. Prot., 92, 489, 2014.
- Lei, F. Chen, Y. Luo, and L. Zhang, "Three-dimensional magnetic graphene oxide foam/Fe3O4 nanocomposite as an efficient absorbent for Cr(VI) removal," Journal of Materials Science, vol. 49, no. 12, pp. 4236–4245, 2014.
- Li, Q., Liu, Z., Huang, L., Teng, J., Bai, Y., Characterization and mechanism elucidation of dye adsorption using cuprous selenide nanoparticles from aqueous solutions. Chem. Res. Chin. Univ., 32, 1010–1015, 2016.
- Lin, Z.Y., Zhang, Y.X., Chen, Y.L., Qian, H., Extraction and recycling utilization of metal ions (Cu2+, Co2+ and Ni2+) with magnetic polymer beads. Chem. Eng. J., 200, 104, 2012.
- Liu, J. H. Yang, J. Zuo et al., "Graphene-supported nanoscale zero-valent iron: removal of phosphorus from aqueous solution and mechanistic study," Journal of Environmental Sciences, vol. 26, no. 8, pp. 1751–1762, 2014.
- Madrakian, A. Afkhami, M. Ahmadi, and H. Bagheri, "Removal of some cationic dyes from aqueous solutions using magnetic-modified multi-walled carbon nanotubes," Journal of Hazardous Materials, vol. 196, pp. 109–114, 2011.

- Mahmoud, H.R., Ibrahim, S.M., El-Molla, S.A., Textile dye removal from aqueous solutions using cheap MgOnanomaterials: Adsorption kinetics, isotherm studies and thermodynamics. Adv. Powder Tech. 27, 223–231, 2016.
- Mahmoud, M.S., Decolorization of certain reactive dye from aqueous solution using Baker's Yeast (Saccharomyces cerevisiae) strain. HBRC J., 12(1), 88, 2016.
- Mamadou and N. Savage, "Nanoparticles and water quality". J. Nano. Res., 2005, 7: 325-330.
- Mei, L. Xuguang, D. Jinming, J. Husheng, W. Liqiao and X. Bingshe, "Antibacterial activity of chitosan coated Ag-loaded nanoSiO2 composites". Carbohydrate Polymers, 2009, 78 (1) 54-59.
- Mohan, D., Kunwar, P.S., Gurdeep S., Kundan K., Removal of dyes from wastewater using Flyash, a low-cost adsorbent. Ind. Eng. Chem. Res. 41, 3688–3695, 2002.
- Montazerozohori, M., Nasr-esfahani, M., Joohari, S., Photocatalytic degradation of an organic dye in some aqueous buffer solutions using nano titanium dioxide a kinetic study. Environ. Prot. Eng., 38(3), 45, 2012.
- Mu, J.-M. Herrmann, and P. Pichat, "Room temperature photocatalytic oxidation of liquid cyclohexane into cyclohexanone over neat and modified TiO2," Catalysis Letters, vol. 3, no. 1, pp. 73–84, 1989.
- Ohsaka, K. Shinozaki, K. Tsuruta, and K. Hirano, "Photoelectrochemical degradation of some chlorinated organic compounds on n-TiO2 electrode," Chemosphere, vol. 73, no. 8, pp. 1279–1283, 2008.
- Ong, S.A., Uchiyama, K., Inadama, D., Ishida, Y., Yamagiwa, K., Treatment of azo dye Acid Orange 7 containing wastewater using up-flow constructed wetland with and without supplementary aeration. Bioresour. Technol., 101, 9049, 2010.
- Oyanedel-Craver and J. A. Smith, "Sustainable colloidalsilver-impregnated ceramic filter for point-of-use water treatment," Environmental Science and Technology, vol. 42, no. 3, pp. 927–933, 2008.
- Parmon, "Nanomaterials in catalysis," Materials Research Innovations, vol. 12, no. 2, pp. 60–61, 2008.
- Peyravi, M. Jahanshahi, A. Rahimpour, A. Javadi, and S. Hajavi, "Novel thin film nanocomposite membranes incorporated with functionalized TiO2 nanoparticles for organic
- Qiao, J., Jiang, Z., Sun, B., Sun, Y., Wang, Q., Guan, X., Arsenate and arsenite removal by FeCl3: effects of pH, As/Fe ratio, initial As concentration and coexisting solutes. Sep. Purif. Technol., 92, 106, 2012.
- Quang, P. B. Sarawade, S. J. Jeon et al., "Effective water disinfection using silver nanoparticle containing silica beads," Applied Surface Science, vol. 266, pp. 280–287, 2013.

- Rajesh, S. Senthilkumar, A. Jayalakshmi, M. T. Nirmala, A. F. Ismail, and D. Mohan, "Preparation and performance evaluation of poly (amide–imide) and TiO2 nanoparticles impregnated polysulfonenanofiltration membranes in the removal of humicsubstances,"Colloids and Surfaces A: Physicochemical and Engineering Aspects, vol. 418, pp. 92–104, 2013.
- Ramalingam, R. and Thilager, M. (2000). Bioconversion of agro waste, sugarcane trash using an Indian epigeic earthworm, Perionyx excavates (PERRIER).indian. j. Environ. And Ecoplan.,3:447-452.
- Rao, R.A.K., Ikram, S., Uddin, M.K., Removal of Cr (VI) from aqueous solution on seeds of Artimisiaabsinthium (novel plant material). Desal. Water Treat. 54, 3358–3371, 2015.
- Rawal, S. Bera, D. Lee, D.-J. Jang, and W. I. Lee, "Design of visible-light photocatalysts by coupling of narrow bandgap semiconductors and TiO2: effect of their relative energy band positions on the photocatalytic efficiency," Catalysis Science and Technology, vol. 3, no. 7, pp. 1822–1830, 2013.
- Ren and J. A. Smith, "Retention and transport of silver nanoparticles in a ceramic porous medium used for point-ofuse water treatment," Environmental Science and Technology, vol. 47, no. 8, pp. 3825–3832, 2013
- Sanjay KS, 2005. Environmental pollution and sugar industry in India its management in: An appraisal. Sugar Tech.7 (1), 77-81.
- Satapathy, M.K., Banerjee, P., Das, P., Plant-mediated synthesis of silver-nanocomposite as novel effective azo dye adsorbent. Appl. Nanosci. 5, 1–9, 2015.
- Sawant, S.S., Salunke, B.K., Taylor, L.E., Kim, B.S., Enhanced agarose and xylan degradation for production of polyhydroxyalkanoates by co-culture of marine bacterium, Saccharophagusdegradans and its contaminant, Bacillus cereus. Appl. Sci., 7(3), 225, 2017.
- Sehlleier, Y.H., Hardt, S., Schulz, C., Wiggers, H., A novel magneticallyseparable porous iron-oxide nanocomposite as an adsorbent for methylene blue (MB) dye. J. Environ. Chem. Eng. 4, 3779–3787, 2016
- Siddiqui W A &Waseem M, A Comparative Study of Sugar Mill Treated and Untreated Effluent- A Case Study, Orient. J. Chem., 28(4) (2012), 1899-1904.
- Singh, H.B., Bharati, K.A., Handbook of natural dyes and pigments, Woodhead Publishing, New Delhi, 2014.
- Sirianuntapiboon, S.; Srisornsak, P. Removal of disperse dyes from textile wastewater using bio-sludge. Bioresour. Technol. 2007, 98, 1057–1066.
- Soares, S.F., Simoes, T.R., Trindade, T., Daniel-da-Silva, A.L., Highly efficient removal of dye from water using magnetic carrageenan/silica hybrid nanoadsorbents. Water Air Soil Pollut. 228, 87, 2017.

- Stoller, L. Miranda, and A. Chianese, "Optimal feed location in a spinning disc reactor for the production of TiO2 nanoparticles," Chemical Engineering Transactions, vol. 17, pp. 993–998, 2009.
- Stoyko, P.P., Pencho, A.S., Color and COD retention by reverse osmosis. Desalination, 154, 247, 2003.
- Suthar, S., (2006). Impact of vermicompost and composted farmyard manure on growth and yield of garlic (Allium stivum L.) field crop. International J. of plant prods. 3(1):25-38.
- Suthar, S., Singh, P., (2008). Effect of vermicompost& composted farmyard manure on growth and yield of garlic (Allium stivum L.) field crop.
- Szymanski, Nathan and Patterson, R.A. (2003 Effective Microorganisms (EM) and wastewater Systems in Future Directions for On-site Systems: Best Management Practice. Proceedings of On-site '03 Conference by Patterson, R.A. and Jones, M.J. (Eds). Held at University of New England, Armidale 30th September to 2nd October 2003. Published by Lanfax Laboratories Armidale. ISBN 0-9579438-1- 4 pp 347- 35.
- Tratnyek, A. J. Salter, J. T. Nurmi, and V. Sarathy, "Environmental applications of zerovalent metals: iron vs. zinc," in Nanoscale Materials in Chemistry: Environmental Applications, vol. 1045 of ACS Symposium Series, chapter 9, pp. 165–178, 2010.
- Uddin, M.K., Bushra R., Synthesis and characterization of composite cation-exchange material and its application in removing toxic pollutants, Enhancing Cleanup of Environmental Pollutants, Springer, 297–311, 2017.
- Vautier, M., Guillard, C., Herrmann, J.M., Photocatalytic degradation of dyes in water. Case study of indigo and of indigo carmine. J. Catal. 201, 46–59, 2001.
- Venkataramana, VP, Narasimha Murthy, B., Krishna Rao, J.V. and Kamble, C.R. (2010). Efficacy of foliar sprays of vermiwash and cow dung wash on biochemical and yield attributes on yield of muiberry (Morusalba L.). Karnataka J. Agric. Sci., 23(2): 358-360.
- Wong, Y.C., Szeto, Y.S., Cheung, W.H., McKay, G., Equilibrium studies for acid dye adsorption onto chitosan, Langmuir, 19, 7888–7894, 2003.
- Yu, Y., Murthy, B.N., Shapter, J.S., Constantopoulos, K.A., Voelcker, N.H., Ellis, A.V., Benzene carboxylic acid derivatizedgraphene oxide nanosheets on natural zeolites as effective adsorbents for cationic dye removal. J. Hazard. Mater. 260, 330–338, 2013.
- Zhang, F., Yang, Y., Tan, R., Song, W., Adsorption behavior and mechanism of methyl blue on zinc oxide nanoparticles. J. Nanopart. Res. 15, 2034, 2013.
- Zhou, L., Jin, J., Liu, Z., Liang, X., Shang, C., Adsorption of acid dyes from aqueoussolutions by the ethylenediamine-modified magnetic chitosan nanoparticles. J. Hazard. Mater., 185(2), 1045, 2011.

- Zhou, T. Shi, and H. Zhou, “Hydrothermal preparation of ZnO-reduced graphene oxide hybrid with high performance in photocatalytic degradation,” Applied Surface Science, vol. 258, no. 17, pp. 6204–6211, 2012.
- Zollinger, H. Colour Chemistry-Synthesis: Properties and Application of Organic Dyes and Pigments. New York: VCH, 1987.