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Biosorption of heavy metals from sewage effluent using prickly pear cactus

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Abstract

The practice of environmentally benign agents in the treatment of water is rapidly gaining interest due to their naturally renewable and available character and low harmfulness. Common Mexican cactus produces a gum-like substance, cactus mucilage, which shows excellent flocculating abilities and is an economically viable alternative for low-income communities. Mucilage of *Opuntia ficus indica* was used as a flocculating agent for heavy metals like Cadmium (Cd), Lead (Pb), Zinc (Zn), Manganese (Mn), Nickel (Ni), Copper (Cu) and Chromium (Cr). Biosorption experiment was conducted using two types of treatments i.e. Dice-Cut-Pressed (DCP) cactus treatment and Extracted Mucilage Treatment (EMT). Results showed the mucilage efficiency for adsorbing heavy metals like Pb, Cd, Cu and Ni from wastewater as determined by Atomic Absorption Spectroscopy. Nickel and Zn concentration was reduced by 72-77% with both DCP and EMT treatments. The Reduction (%) in Pb was higher than Ni and Zn, i.e., 90% with both extractants. Cd and Cu sorption was almost 100% by both the treatments. When quality of treated and untreated waste water effluent was compared with WHO (World Health Organization) standards for irrigation, treated waste water was found to be nearly fit for the irrigation. Results indicate that both the materials can potentially be used as sorbent for chemical pollutants.

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Introduction

We are living in a period of history where the communal heritage of humanity and the earth is under systemic obstruction. Everything is for sale according to the current model of globalization. Fresh water dramatizes the crisis of the commons more clearly than nothing. Freshwater shortages combined with deeply inequitable access and increased waste water production poses the greatest environmental and human rights threats of our time. Such scenario makes the reuse of wastewater a needy application. Currently, about 80% of urban wastewater is used for irrigation in developing countries that contributes to the 70-80% food security and the livelihoods of urban and peri-urban communities (Mara and Cairncross, 1989 and Cooper, 1991). The untreated wastewater is a rich source of pollutants particularly heavy metals. Continuous application contaminates the soil and hence crop produced. Prolong exposure of men and animals to contaminated products can cause serious health hazards. Pakistan is basically an agriculture country, where water consumption for irrigation is 90% of the total water. Unfortunately, while Pakistan is blessed with groundwater and surface water resources, with the passage of time hasty population growth, urbanization and unmaintainable water consumption practices in the industrial and agricultural sectors have placed immense stress on the quality as well as quantity of water resources in the country (WWF, 2007). The economy of entire population depends on the water of river Indus system, mostly derived from the snowmelt of western Himalayas.

Bioremediation is an umbrella term for technologies based on living (or once-living) organisms. One way to of treatment is to use plant extractants that have coagulating properties. These extractants are natural coagulants that can clean water by acting as a flocculating agent. Prickly pear cactus is already tested for its coagulating properties in drinking water samples by adding bacteria and other contaminants. It was found that cactus produces gum-like mucilage that can adhere with certain metals like Arsenic, Nickle and Chromium present in drinking water samples. Similarly bacteria (*Bacillus cereus*) can also adsorb to the mucilage resulting in water purification. So to meet the idea of waste water reuse, this study was designed for industrial and municipal sewage effluent biotreatment.

In this study, industrial/ municipal sewage effluent was given mucilage treatment to check its remedial efficiency. Treated and untreated waste water samples were periodically analyzed for a period of 9 days for heavy metals like Manganese (Mn), Cadmium (Cd), Chromium (Cr), Iron (Fe), Copper (Cu), Lead (Pb), Zinc (Zn) and Nickle (Ni). Heavy metals analysis was used to determine the quality of treated wastewater to see its suitability for irrigation purpose and to check the percentage remediation by Prickly pear cactus mucilage. Moreover the degree of treatment was determined and treated wastewater quality parameters were compared with WHO (World Health Organization) standards for agriculture.

The present study was performed with the objectives to bio-remediate the industrial and municipal sewage effluent for irrigation purpose. Moreover to compare remedial efficiency of Dice-Cut-Pressed (DCP) cactus treatment and Extracted Mucilage treatment (EMT), and to check the efficacy of DCP and EMT treatments at different time period.

Materials and methods

The research was conducted at Soil Environment and Soil Fertility laboratory at Land Resources Institute, National Agriculture Research Centre, NARC, Islamabad and Microbiology laboratory at Pakistan Council of Research in Water Resources (PCRWR), Islamabad. Bioremediation of heavy metal rich industrial/municipal sewage effluent was carried out using mucilage of Prickly pear cactus (*Opuntia ficus indica)*.

Prickly pear cactus collection from field

Pads of Prickly pear cactus were collected from the field located in NARC.

The extraction of mucilage of Opuntia ficus indica for biosorption

Cactus pads were first washed with water and dried. It was next followed by removing spines and peeling of pads. Half of the pads were used for dicing purpose and half of them were used for mucilage extraction by boiling in water.

Mucilage extraction by Dice-Cut-Press (DCP) method

Peeled pads of Prickly pear cactus were cut into small pieces of about 5cm^2 for better mucilage extraction. It was then diced using knife in order to expose the parenchymatous cells which secrete mucilage. Further cactus pieces were pressed with spoon to expose mucilage. The material was then stored at 4^{o}C in refrigerator until use (Fox, 2011).

Mucilage extraction in water (Extracted Mucilage Treatment)

In EMT method, 6kg of cactus pads were cut into small pieces followed by their cooking in 5 liters of water for 30 minutes. The soluble sugars of mucilage get release into water. Material was then decanted and stored at 4^{o}C in refrigerator until use.

Sewage effluent sample collection

100 liters of industrial/municipal sewage effluent sample was brought from Gujranwala (industrial estate) for biotreatment purpose, using mucilage of *Opuntia ficus indica*. Industries included cottage, steel, marble, confectionary and sanitary industries.

Bioremediation of sewage effluent

An in-vitro experiment was conducted to evaluate the bio-remedial efficiency of Prickly pear cactus mucilage. The collected effluent was poured in three sets of plastic containers (8L capacity).

All three sets were in triplicate. One set of containers was treated with mucilage extracted by DCP method. Second set of containers was treated with water extracted mucilage. While third set of containers was kept control i.e. it was plain sewage water.

Each container was filled with 7 liters of sewage effluent sample. In the first set of container, 1.5kg each of diced cactus was added, while in the second set of container, 1.5L each of water extracted mucilage was added for bio-treatment purpose. The third set of containers was kept control. After the addition of biomaterial, the system was left for 10-15 minutes to settle. It was then followed by the withdrawal of 200ml of sample in autoclaved bottles from the entire 9 container for day 0 (D0) analysis, followed by analysis at day1 (D1), day 2 (D2), day 6 (D6) and day 9 (D9).

Heavy metals analysis

Samples were analyzed for heavy metals like Iron, Nickle, Cadmium, Lead, Zinc, Manganese, Copper and Chromium. Vacuum filtration of all the samples drawn from control containers and containers given DCP treatment was performed using nitrocellulose membrane of 0.42nm pore size. On the other hand, samples drawn from containers receiving liquid mucilage treatment was centrifuged at 10,000 rpm for 5 minutes in order to separate the mucilage part from liquid sample (Mane *et al.,* 2011). It was next followed by the addition of 2-3 drops of nitric acid in samples to stop any microbial activity. Metals detection was performed through Flame Atomic Absorption Spectrometer (Perkin Elmer, Analyst 700), attached with graphite furnace (Richards, 1954). For every metal, the instrument was first standardized using different standards and doubled distilled water. Readings were then noted down for all the samples taken from plastic containers.

Physiochemical analysis

All 9 samples were analyzed for the following physiochemical parameters. Dissolved oxygen, EC and DO were determined for the samples using Lutron WA-2015 instrument. The instrument was first calibrated at pH 4 and 7 for pH and at 1.143mScm^{-1} for EC determination. Probe for Dissolved oxygen was calibrated by dipping probe in sodium sulphite solution. When the reading was stabilized, the reading was adjusted to zero. Similarly mercury thermometer was used for the recording of temperature of the samples.

Carbonates and bicarbonates content was determined by titrating 5ml sample against 0.009N H_2SO_4 solution, using phenolphthalein as an indicator in case of CO_3, while for HCO_3, methyl orange was used as an indicator (Richards, 1954). 5ml sample was taken in a flask and 1 drop of phenolphthalein indicator was added. If solution turns pink, it shows presence of CO3 but there were no CO_3 in solution and solution remained colorless. Next 2 drops of methyl orange was added for HCO_3 detection. It was then titrated against 0.009N H_2SO_4. End point was determined by color change from colorless to red.

Calcium and Magnesium content were determined for samples by taking 5ml sample followed by the addition of 10 drops of Ca+Mg buffer. Next 4 drops of Black T indicator was added and sample was titrated against 0.01N EDTA. Sample was titrated until maroon color changes to purple. This indicates end point (Richards, 1954).

Turbiditric method was used for turbidity analysis by using Microprocessor turbidity meter, HANNA instrument (HI 93703). Instrument was calibrated at 0 NTU and 10 NTU by using hydrazine sulphate and hexamethylene tetra amine respectively. 10ml sample was loaded in a cuvette and placed in cavity made in instrument. Reading was noted for all the samples. Flame photometer (420 Sherwood) was used for Na detection followed by SAR determination by following formula as described in Richards, 1954;

$SAR = Na / (Ca + Mg/2)^{1/2}$

1ml sample was poured into a test tube followed by the addition of 4ml $LiCl_2$ and 5ml distilled water. A 10ml solution was prepared. Next Flame photometer was standardized at 20 using Na+K 20 standard solution. Samples were then analyzed for Na content in ppm for SAR determination. EC meter, JENWAY 4320 was used to record TDS of the control samples and treated samples.

Statistical Analysis

Statistix 8.1 software was used for the statistical analysis of the data. A 2Factor factorial-design was applied during experiment. Moreover two factors were under observation, i.e.

1. Mucilage type (diced cactus and extracted mucilage)
2. Time

Results and discussions

Efficiency of Prickly pear cactus mucilage for wastewater treatment was measured by different parameters in terms of heavy metals and other physiochemical parameters. Results presented in the preceding section shows concentration of heavy metals and other physiochemical parameters in treated and untreated waste water samples.

Pre-analysis of the industrial/municipal sewage effluent sample was performed in order to record the water quality before giving treatment. Figure 1 and 2 shows reduction (%) over control in Cu and Pb concentrations respectively. Both treatments equally biosorbed the metals and 100% sorption took place by Day(2) for copper and 90% sorption took place for Pb by Day(9). Figure 5 shows 100% biosorption of Cd by Day1 indicating 100% adsorption for both treatments. No active biosorption of Mn took place for both the treatments. Percent reduction in Mn concentration over control was very less as compared to other heavy metals as shown in figure 4. EMT treatment achieved about 8% of removal rates while DCP achieved only 6% removal rates. Percentage reduction over control in Zn concentration for a 9-Days experiment is shown in figure 5. Both treatments equally sorbed the metal and by Day (9), almost 70% of the metal biosorption took place. Percent reduction in Ni concentration increases with time and by Day (9), 77% of the metal biosorption took place.

Few studies have been done to see cactus effectiveness for biosorbing metals like Ni, Cr, As and bacteria like *Bacillus cereus* but present study is the first report to evaluate mucilage ability to treat wastewater. Idea was to treat raw wastewater and to check its quality in terms of heavy metals. Two types of treatments were given to the wastewater; Dice-Cut-Pressed cactus treatment (DCP) and extracted mucilage treatment (EMT). 7L of wastewater sample was given 20% treatment.

Wastewater sample was treated for a period of 9 days and water was periodically analyzed for its quality assessment. Pre-analysis of the wastewater was performed soon after the sample collection. Similarly, metals were detected in samples and were found in variable concentration. Ni was present in highest concentration (0.724mgl^{-1}) as against other heavy metals. Mn, Cu, Cd and Pb were also comparatively high in concentration.

Table 1. Comparison of Experimental Results (Means) with World Health Organization standards for Irrigation.

Parameters	Control	DCP Treatment	Extracted mucilage treatment	WHO Standards
pH	7.35	7.49	7.47	6-10
Temperature	12.5	12.4	12.5	20-26
Dissolved Oxygen (mg/l)	1.54	0.1	0.9	6-8
Conductivity (uS/cm)	1354	1325	1330	1500
Turbidity (NTU)	56	51.6	129	25-50
Total dissolved solids (mg/l)	902	843	897	1000
SAR (mmol/L)0.5	56	40	45	10
Fecal coliforms (MPN/100 ml)	686	230	284	<250
Total coliforms (MPN/100 ml)	1600	1233	1366	<250
Fe (mg/l)	0.04	0.03	0.032	5
Mn (mg/l)	0.562	0.4	0.4	0.2
Ni (mg/l)	0.728	0.44	0.38	0.2
Cu (mg/l)	0.541	0.006	0.005	0.02
Cr (mg/l)	-	-	-	0.10
Pb (mg/l)	0.144	0.09	0.10	0.065
Cd (mg/l)	0.025	0.003	0.003	0.01
Zn (mg/l)	0.368	0.25	0.22	2

Initial pH was 7.9 and thus medium was alkaline. Initial turbidity and TDS of the sample was high and was recorded to be 92 NTU and 908 mgl^{-1} respectively. Day (0) readings were recorded for all the samples after 10-15 minutes of the addition of cactus mucilage.

Metals biosorption took place with variable degree for cactus mucilage. Ni concentration remarkably decreased for both treatments. In this study, the concentration of Ni decreased from 0.72mgl^{-1} to 0.101mgl^{-1} for DCP treatment and 0.72mgl^{-1} to 0.102mgl^{-1} for EMT treatment indicating 71% and 76% removal rate. Correspondingly, as demonstrated by Mane *et al.* (2011), Ni biosorption (88.4%) through cactus mucilage was found to be effective at 150 rpm on using 10% (v/v) Opuntia natural polyelectrolyte. While in this study, both treatments showed better biosorption with no agitation provided.

Moreover their percent Ni removal was high as compared to present study and here possible explanation is the presence of different metals in same medium that increased competition among metals for biosorption sites.

Both treatments showed increased biosorption for Cu by decreasing initial concentration of 0.54mgl^{-1} to 0.00mgl^{-1} for both DCP and EMT treated samples by Day (2). By Day (2) the, concentration of Cu in all treated samples were zero indicating 100% removal rate. In present study, no agitation was provided and experiment was conducted at room temperature. For Cd, both treatments showed similar adsorption and Cd was completely biosorbed by Day (2) with 0 concentrations in the treated samples showing 100% removal. Fe was present in permissible range.

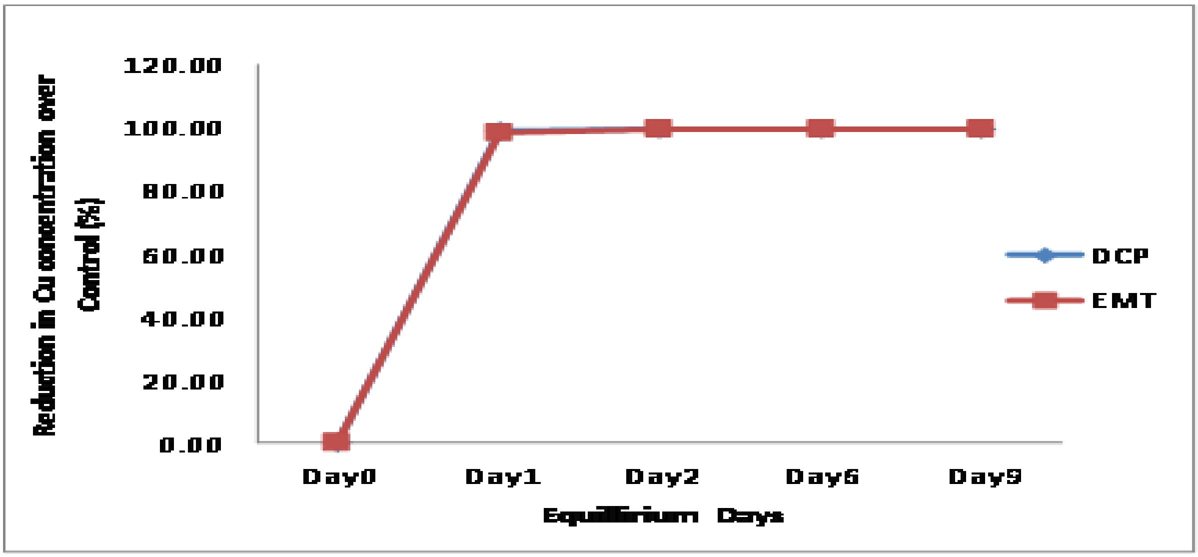

Fig. 1. Reduction (%) in Copper concentration over control from Day0 to Day9 in industrial/municipal sewage effluent for Dice-Cut-Pressed cactus treatment (DCP) and Extracted Mucilage treatment (EMT).

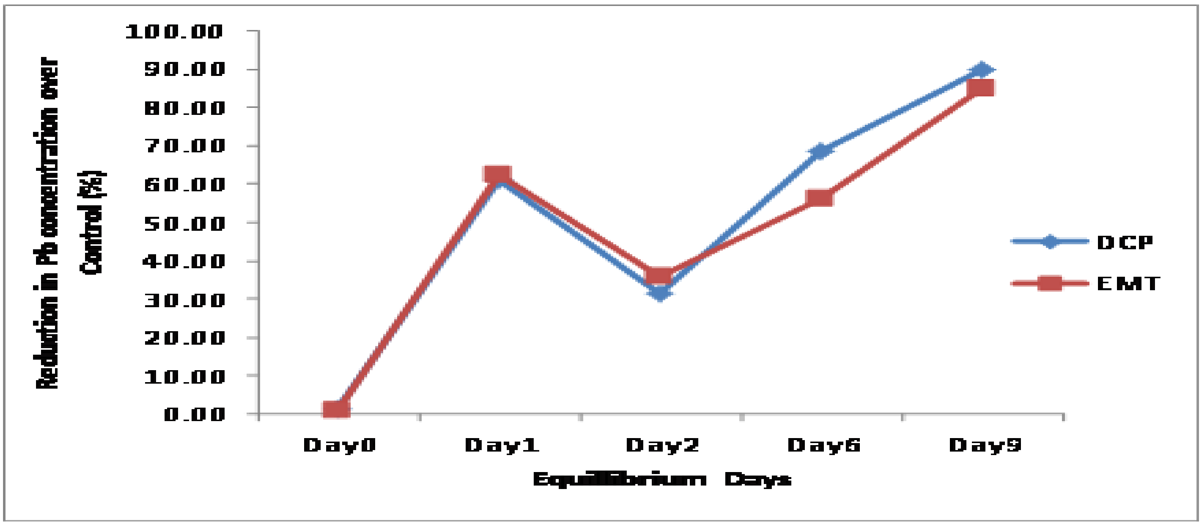

Fig. 2. Reduction (%) in Lead concentration over control from Day0 to Day9 in industrial/municipal sewage effluent for Dice-Cut-Pressed cactus treatment (DCP) and Extracted Mucilage treatment (EMT).

It showed adsorption for cactus mucilage decreasing the initial concentration of 0.044mgl^{-1} to 0.001mgl^{-1} by Day(1) for both DCP and EMT treatments. For both the treatments, the % removal of Fe was 97.7%. No data is available to show efficacy of mucilage for Zn and Pb adsorption. But present study indicated following description for both metals. Biosorption of Zn for both treatments was effective, decreasing initial concentration of 0.376mgl^{-1} to 0.121mgl^{-1} and 0.126mgl^{-1} for DCP and EMT treatments respectively indicating 68% and 67% removal rate.

Pb biosorption took place form initial concentration of 0.149mgl^{-1} to the final concentration of 0.014mgl^{-1} and 0.021mgl^{-1} at Day (9) for DCP and EMT treatment respectively indicating 90% and 85% removal rate. Contrarily, for Mn no effective biosorption was observed for both the treatments. Possible explanation for this is the presence of different metals in same medium that increased metals competition for biosorption sites. Ni, Cu, Cd, Pb, and Mn were present initially in comparatively high concentration. After adsorbing to the biomaterial, their concentration decreased and placed them in safe range.

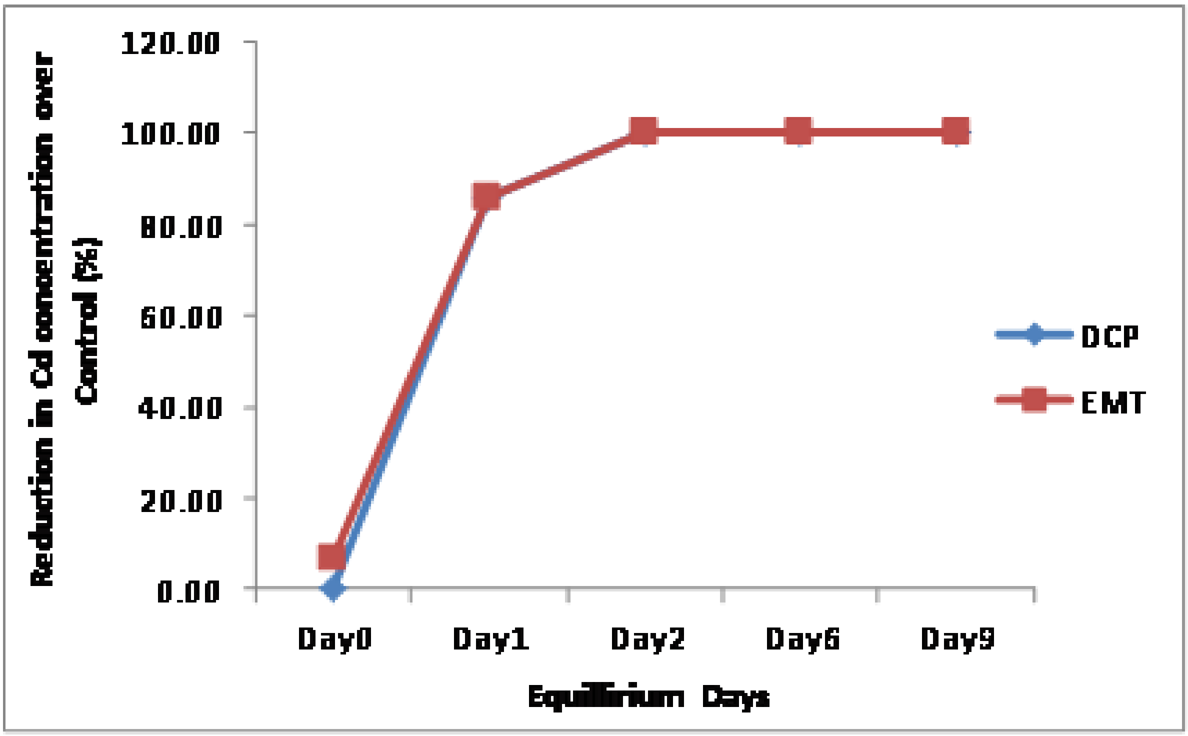

Fig. 3. Reduction (%) in Cadmium concentration over control from Day0 to Day9 in industrial/municipal sewage effluent for Dice-Cut-Pressed cactus treatment (DCP) and Extracted Mucilage treatment (EMT).

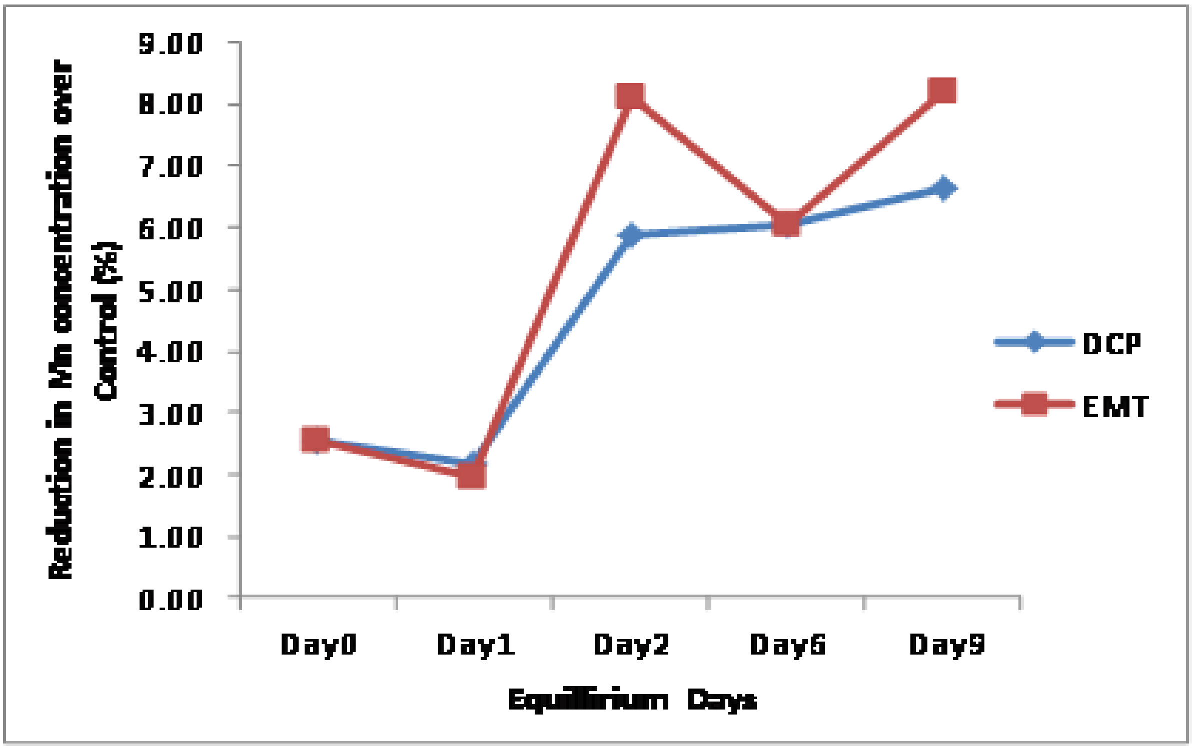

Fig. 4. Reduction (%) in Manganese concentration over control from Day0 to Day9 in industrial/municipal sewage effluent for Dice-Cut-Pressed cactus treatment (DCP) and Extracted Mucilage treatment (EMT).

Statistical analysis of the data revealed that both treatments were significant for Ni, Cu, Fe, Cd, Zn, Pb and turbidity. Moreover mean comparison test indicated that there is no significant difference in mean values of both DCP and EMT treatment which illustrates that both treatments are equally effective. While mean values of both treatments are different from the mean value of control showing treatment significance and efficiency.

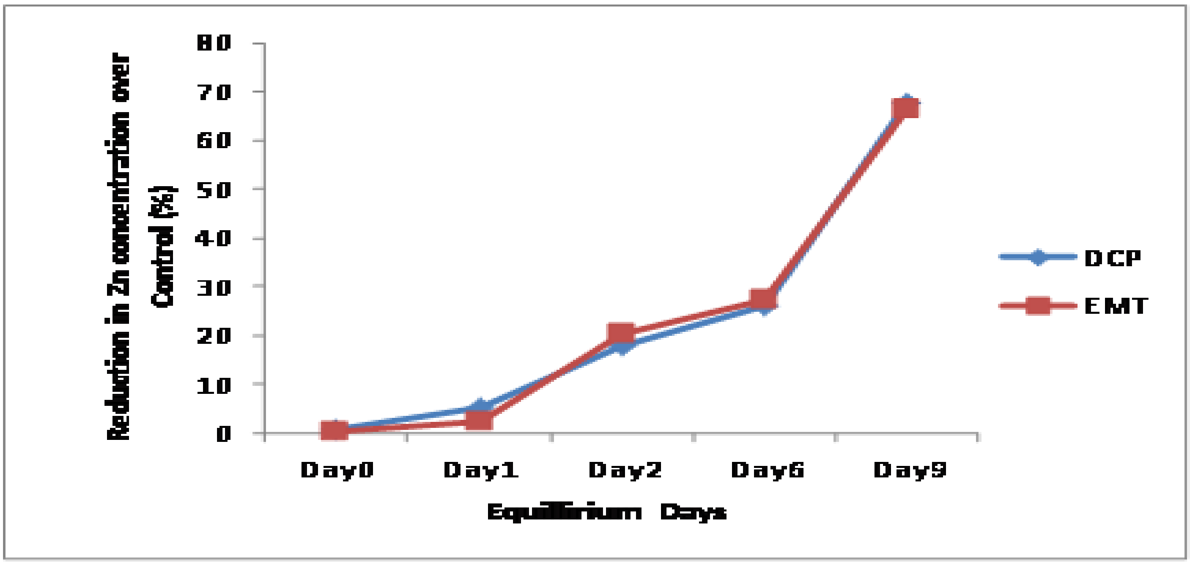

Fig. 5. Reduction (%) in Zinc concentration over control from Day0 to Day9 in industrial/municipal sewage effluent for Dice-Cut-Pressed cactus treatment (DCP) and Extracted Mucilage treatment (EMT).

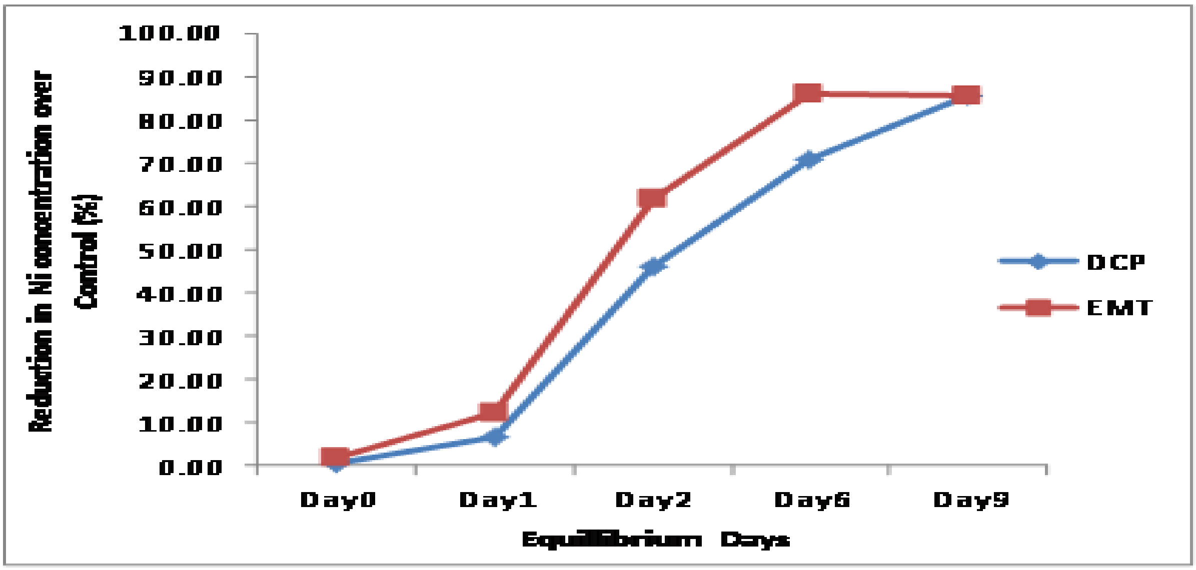

Fig. 6. Reduction (%) in Nickle concentration over control from Day0 to Day9 in industrial/municipal sewage effluent for Dice-Cut-Pressed cactus treatment (DCP) and Extracted Mucilage treatment (EMT).

Conclusion

Cactus mucilage is an efficient flocculent and both treatments i.e. DCP and EMT showed almost similar adsorption capacity for heavy metals, total coliforms and fecal coliforms and percentage reduction by both treatments was very impressing. No profound differences were observed in their treatment efficiency. Cu and Cd showed complete biosorption by Day (2), indicating 100% removal rate. Pb, Ni, Zn and Fe also showed more adsorption for mucilage and similarly fecal coliforms were completely adsorbed by Day (2) indicating 100% removal rate. Diced cactus treatment protocol is more efficient and reproducible and user friendly approach. *Opuntia ficus*-indica mucilage is a promising actor in the field of emerging technologies for metals and microbes removal.

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