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COMPARATIVE ANALYSIS OF PHYSICO-CHEMICAL PARAMETERS OF SOIL CONTAMINATED WITH PETROLEUM HYDROCARBONS COLLECTED FROM SEMI-ARID (JAIPUR-AJMER) AND ARID (BARMER) REGIONS OF RAJASTHAN WITH REFERENCE TO BIOREMEDIATION

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Environment contaminated with petroleum or its product is the major global concern today and leads to a serious hazards to living bodies therefore some remedial methods are required to cure it. Bioremediation is considered as one of the best technology for the treatment of petroleum contaminated soil. In order to make Bioremediation technology successful and efficient it is necessary to assess various physico-chemical parameters of soil contaminated with petroleum hydrocarbon. All the parameters play important role in bioremediation of petroleum contaminated soil. Present study was conducted to determine various physico- chemical parameters such as soil colour, soil texture, pH, Moisture content, Electrical conductivity, Total Dissolved Solids (TDS), Carbonate and Bicarbonate content, Chloride content, Sodium and Potassium content, Organic carbon, Organic matter, Nitrogen and Phosphorous content of the soil samples collected from Jaipur-Ajmer (Semi-arid) and Barmer (arid) regions of Rajasthan (India). Physico-chemical analysis of contaminated soil was performed by using standard analytical methods. The physico-chemical properties of soils alter due to petroleum hydrocarbon contamination. The petroleum hydrocarbon contaminated soil samples exhibited relatively lower pH value, moisture content, carbonate content, bicarbonate content and higher electrical conductivity, TDS value, available sodium concentration, available potassium concentration, organic carbon, organic matter content, available nitrogen content, available phosphorous content, chloride content as compared to uncontaminated soil sample (control).

Keywords: Bioremediation; Contaminated soil; Petroleum hydrocarbon; Physico-chemical parameters.

Introduction

Soil is the thin layer of earth surface where we live. Soil nourishes and supports growing plants and provides us food, fiber and forest products. It consists of mineral matter, organic matter, water and air. The soil stores and purifies water. Soil contamination of petroleum hydrocarbon results due to transportation, accidental spills, storing, drilling, manufacturing, industries etc. the petroleum hydrocarbon contamination of soil leads to affect plants adversely by producing hazardous substances in the soil easily available to plants[1]. Petroleum hydrocarbons are toxic, carcinogenic and mutagenic in nature[2,3,4]. Its contamination in soil causes depletion of organic matter content, soil mineral nutrients, deterioration of soil structure, inhibit enzymes activities, leads to leaching, soil erosion, contaminate surface and ground water and also affect aquatic life[5,6,7]. A technique called Bioremediation used to remediate the petroleum contaminated soil. Bioremediation involves the interaction of microorganisms with the pollutants at the contaminated site under the influence of environmental factors, which affect the growth and activity of microorganisms. In order to make bioremediation more effective and successful the proper knowledge about the environmental factors/ physico-chemical parameters are necessary[8]. This paper deals with the assessment of various physico-chemical properties of petroleum contaminated soil such as pH, Moisture content, Electrical conductivity, Total Dissolved Solids, Available Nitrogen, Potassium content, Carbonate and Bicarbonate content etc in order to make bioremediation technology more efficient.

Methodology

Sample collection

Soil samples were collected from Arid and Semi-Arid regions of Rajasthan which includes- Jaipur- Ajmer (Semi-Arid) and Barmer (Arid). Total 21 soil samples were collected from contaminated site. There were Seven soil samples (A1, A2, A3, A4, A5, A6, A7) collected from Ajmer, seven soil samples (J1, J2, J4, J5, J6, J7, J8) collected from Jaipur, seven soil samples (B1, B2, B3, B6, B7, B8, B9) were collected from Barmer. All these soil samples were collected from petrol pumps, garages, filling stations, depot and generator sites. Soil samples were collected at a depth of 0-5cm using a soil auger in pre- sterilized plastic bags. All plastic bags were tightly packed to maintain the original moisture. The date, time and site of collection were labeled in each plastic bag and brought immediately to research lab for further processing.

Physico-chemical parameters

In the laboratory, the soil samples were air dried and sieved to remove the unwanted particles. The sieve used was of 2mm pore size[9] and samples were stored at 10^0C for physico-chemical characterization. The various Physical properties of soil samples like soil colour, soil texture, soil moisture content, and total dissolved solid were determined for all 21 soil samples. The various chemical properties like electrical conductivity, pH, organic carbon and organic matter, available phosphorous, carbonate and bicarbonate content in soil, available potassium and sodium content in soil were also determined. A paste of air dried soil samples were prepared in distilled water in the ratio of 1:10 (soil: water). This paste was stirred to make homogenous slurry and then allowed to stand for at least 2-4 hours, after that the pH was determined by the pH Analyzer (L1612, ELICO), electrical conductivity and total dissolved solid was determined by EC-TDS Analyzer (CM183, ELICO). Moisture content of soil samples were determined by weighing 10g of soil (W_1) and drying it in hot air oven at 105^0C for 24 hrs. After 24 hrs the samples were again weighted (W_2). The moisture content was determined by subtracting the final weight (dried soil) from the initial weight (post dried) of soil (W_1-W_2). Available Sodium and Potassium content in soils was determined by the flame photometer. The Organic Carbon and Organic Matter in soils were determined by the Walkley and Black[10] rapid titration method. The available Nitrogen in soils was determined by the procedure followed by Subbiah and Asija[11]. The available Phosphorus in soils was determined by the method used by Olsen et al.[12]. Carbonate and bicarbonate in a solution can be determined by titrating the solution against standard acid using phenolphthalein and methyl red respectively. Chloride content in the soil can be determined by titrating the soil extract against standard $AgNO_3$, solution using potassium chromate as indicator.

Result and Discussion

The soil microorganisms utilize contaminant present in the soil such as petroleum hydrocarbon as a source of carbon andenergy and eliminate it. For successful bioremediation, environmental conditions are among the most important limiting factors. Environmental factors include various physico-chemical factors such as temperature, pH, nutrients, electrical conductivity etc and biological factor includes microbial degradation of contaminant (petroleum hydrocarbon). These environmental factors are used to access the capability of microorganisms to degrade petroleum hydrocarbon.

Table 1: Petroleum contaminated soil samples collected from various sites of Jaipur, Ajmer and Barmer (Rajasthan, India).

S. No.	Sample code	Colour of Soil	Site	Address
1	J1	Dark brown	Generator	Jayoti Vidyapeeth Women's University, Jharna, Jaipur
2	J2	Grey	Petrol pump	Pushapraj petrol pump, Bhakrota,Jaipur
3	J4	Brown	HPCL Depot	Chittroli, near bagru, Jaipur
4	J5	Black	Garage	Mahipal auto repair centre, Jaalpura, Jaipur
5	J6	Brown	Garage	Mahipal auto repair centre, Jaalpura, Jaipur
6	J7	Black	Garage	Prem Jodhpur,
7	J8	Black	Garage	Jagdamba automobile, Jaipur
8	A1	Brown	Petrol pump	Shahid suber petrol pump, Ajmer
9	A2	Brown	Petrol pump	Near Indian oil petrol pump depot, Nasirabad bypass, Ajmer
10	A3	Black	Garage	Chauhan autocare, Nagra, Ajmer
11	A4	Black	Garage	Puja tempo repairs, parvatpura, Ajmer
12	A5	Black	Garage	T.S Tractor garage, Makhupura, Ajmer
13	A6	Brown	Petrol pump	Carnal filling center, Ajmer
14	A7	Grey	Petrol pump	Bharat petroleum, yadav petrol pump, Ajmer
15	B1	Grey	Petrol pump	Near new bus stand, Balotra (Barmer)
16	B2	Brown	Petrol pump	Jagdamba petrol pump, Baytu (Barmer)
17	B3	Brown	Mangala Processing Terminal	Kavas, Nagana (Barmer)
18	B6	Black	Garage	Barmer
19	B7	Black - brown	Garage	Barmer
20	B8	Brown	Petrol pump	Barmer filling station, Indian oil (Barmer)
21	B9	Brown	Petrol pump	Agarwal petrol pump, Nagana (Barmer)

The present study analyzes the various physico-chemical properties of petroleum contaminated soil which would be useful for standardization of bioremediation protocols. These environmental factors play a vital role in the bioremediation of soil.

Table 2: Various physico-chemical properties of soil contaminated with petroleum hydrocarbon collected from Semi-Arid (Jaipur-Ajmer) and Arid (Barmer) regions of Rajasthan

Sample	pH	EC (dsm^{-1})	Moisture content %	TDS (ppm)	CO_3^{2-} (meL^{-1})	HCO_3^- (meL^{-1})	Cl^- (meL^{-1})	Na^+ (meL^{-1})	K^+ (meL^{-1})	Organic carbon %	Organic matter %	N %	P %
Control	7.2 ± 0.017	0.15± 0.021	15.00 ± 0.019	88.5± 0.025	6.89 ± 0.030	6.10 ± 0.027	0.5 ± 0.023	2.0 ± 0.018	0.48 ± 0.015	0.15± 0.032	0.25 ± 0.031	0.019± 0.022	0.015± 0.020
J1	6.2 ± 0.023	0.241± 0.011	11.08± 0.016	97.25± 0.013	0.0± 0.002	3.0± 0.019	0.9± 0.029	3.5± 0.020	0.80± 0.019	1.02± 0.027	1.75± 0.033	0.025± 0.018	0.021± 0.017
J2	5.0 ± 0.021	1.76± 0.020	11.15± 0.031	846.0± 0.026	0.0± 0.009	2.0± 0.024	8.4± 0.020	4.0± 0.027	0.76± 0.021	1.31± 0.023	2.26± 0.027	0.029± 0.026	0.025± 0.010
J4	4.5 ± 0.011	0.186± 0.023	10.05± 0.011	101.4± 0.021	0.0± 0.002	0.5± 0.011	2.0± 0.013	2.5± 0.033	0.72± 0.013	0.37± 0.019	0.64± 0.018	0.021± 0.014	0.022± 0.022
J5	4.8 ± 0.018	1.19 ± 0.019	13.12± 0.025	485.1± 0.015	0.0± 0.005	3.0± 0.028	4.4± 0.024	3.2± 0.016	0.50± 0.018	2.07± 0.021	3.56± 0.013	0.042± 0.017	0.041± 0.028
J6	4.2 ± 0.020	2.07± 0.027	11.20± 0.027	937.6± 0.013	0.0± 0.008	5.0± 0.022	8.2± 0.016	6.5± 0.021	0.72± 0.020	1.69± 0.013	2.92± 0.025	0.038± 0.020	0.035± 0.021
J7	4. 9 ± 0.028	2.45± 0.015	13.25± 0.023	91.13± 0.028	0.0± 0.014	1.0± 0.029	9.0± 0.021	7.0± 0.011	0.54± 0.016	2.85± 0.033	4.91± 0.009	0.048± 0.023	0.045± 0.013
J8	6.1 ± 0.024	1.61± 0.013	13.50± 0.012	671.0± 0.023	0.0± 0.018	1.5± 0.017	7.1± 0.028	7.6± 0.018	0.56± 0.027	2.89± 0.031	4.99± 0.021	0.051± 0.030	0.047± 0.017
A1	5.3 ± 0.019	0.515± 0.017	10.70± 0.021	223.1± 0.013	0.0± 0.010	1.5± 0.031	2.5± 0.030	5.5± 0.024	0.58± 0.013	1.43± 0.022	2.46± 0.017	0.039± 0.028	0.035± 0.019
A2	5.0 ± 0.029	0.696± 0.026	10.50± 0.018	288.8± 0.025	0.0± 0.004	0.5± 0.013	4.0± 0.011	4.5± 0.019	0.60± 0.031	1.35± 0.014	2.32± 0.032	0.041± 0.021	0.037± 0.008
A3	4. 9 ± 0.013	0.766± 0.022	13.42± 0.033	343.6± 0.019	0.0± 0.008	0.8± 0.021	3.0± 0.014	3.5± 0.022	0.70± 0.027	2.88± 0.025	4.96± 0.016	0.052± 0.019	0.045± 0.015
A4	4.7 ± 0.026	0.952± 0.032	13.50± 0.026	416.6± 0.022	0.0± 0.016	0.6± 0.015	3.6± 0.027	3.8± 0.028	0.90± 0.022	2.92± 0.017	5.04± 0.023	0.052± 0.012	0.045± 0.021
A5	5.6 ± 0.018	1.02± 0.016	13.48± 0.017	458.6± 0.024	0.0± 0.012	3.0± 0.027	5.6± 0.019	3.9± 0.030	0.84± 0.017	2.88± 0.035	4.96± 0.026	0.050± 0.031	0.048± 0.022
A6	5.3 ± 0.020	0.638± 0.011	11.25± 0.024	341.1± 0.011	0.0± 0.013	0.8± 0.021	2.2± 0.021	4.0± 0.017	0.88± 0.023	1.23± 0.021	2.12± 0.019	0.032± 0.016	0.047± 0.012
A7	6.4 ± 0.023	2.73± 0.024	11.42± 0.014	91.21± 0.026	0.0± 0.011	1.0± 0.014	1.7± 0.028	2.5± 0.010	0.60± 0.020	1.37± 0.013	2.36± 0.015	0.037± 0.018	0.045± 0.026
B1	5.3 ± 0.014	0.838± 0.033	9.08± 0.011	402.4± 0.028	0.0± 0.007	1.0± 0.017	4.3± 0.016	2.6± 0.031	1.30± 0.009	0.18± 0.023	0.31± 0.011	0.021± 0.011	0.022± 0.017
B2	6.3 ± 0.027	0.73± 0.027	10.90± 0.033	260.3± 0.031	0.0± 0.005	2.0± 0.023	2.5± 0.012	4.6± 0.022	1.32± 0.012	1.12± 0.012	1.93± 0.023	0.035± 0.009	0.037± 0.020
B3	4.6 ± 0.021	8.43± 0.016	11.12± 0.027	93.34± 0.021	6.0± 0.006	0.5± 0.028	10.5± 0.022	4.0± 0.029	1.20± 0.022	1.47± 0.027	2.53± 0.018	0.042± 0.014	0.049± 0.024
B6	5.1± 0.030	1.16± 0.023	13.15± 0.019	561.1± 0.013	0.0± 0.002	0.5± 0.012	4.8± 0.032	3.6± 0.014	0.76± 0.015	2.77± 0.033	4.78± 0.014	0.059± 0.025	0.029± 0.013
B7	4.4 ± 0.017	3.2± 0.014	11.14± 0.013	97.10± 0.010	0.0± 0.019	0.5± 0.020	8.5± 0.029	3.8± 0.010	0.84± 0.032	1.46± 0.011	2.52± 0.031	0.032± 0.017	0.022± 0.018
B8	5.6 ± 0.012	1.91± 0.020	11.07± 0.023	99.124± 0.020	0.0± 0.014	1.0± 0.033	9.5± 0.016	3.9± 0.008	0.84± 0.021	1.45± 0.017	2.50± 0.027	0.035± 0.033	0.025± 0.027
B9	5.7 ± 0.020	2.07± 0.018	10.85± 0.034	857.9± 0.017	0.0± 0.008	2.0± 0.013	1.0± 0.024	4.5± 0.029	0.80± 0.027	0.93± 0.020	1.61± 0.019	0.025± 0.010	0.020± 0.021

Collection of soil samples

Petroleum contaminated soil samples were collected from various sites as listed in Table 1. All the collected soil samples had peculiar odors characteristic of petrol and diesel.

Physical and chemical parameters

Physico-chemical parameters determined in this study are useful for the standardization of bioremediation process for the treatment of petroleum hydrocarbon contaminated soil. Environmental factors (physico-chemical factors) play vital role in the bioremediation of soil. The various physico-chemical parameters of soil samples collected from Jaipur, Ajmer and Barmer regions were summarized in Table 2**.**

pH is a very important factor in the biodegradation of petroleum hydrocarbon. Soil samples collected from various sites showed variations in the pH value. The highest pH value observed was 6.4 obtained from A7 sample of Ajmer region and the lowest pH value was observed 4.2 obtained from J6 sample of Jaipur region. A great diversity was observed in the pH values for the soil samples collected from Barmer region and ranged from 4.6 to 6.3. The control soil sample (uncontaminated) had pH value 7.2. All the contaminated soil samples had a relatively low pH value as compared to uncontaminated soil sample (control) (Table: 2).

The average pH value calculated for Jaipur, Ajmer and Barmer regions were 5.1, 5.31 and 5.2 respectively. Present study was supported by Atlas[13]. He reported that a pH near neutral was preferred by most microorganisms. The study done by Vidali[8] also suggested a pH range 5-8 was required for microbial activity and thus favoured present research work. US-EPA[14] proposed the most appropriate range for bioremediation was in the range of pH 6-8 which complements present study. These findings were in agreement with the pH range obtained from our results.

For the growth and proper functioning, microorganisms require moisture thus moisture content play important role in the bioremediation. The highest Moisture Content was 13.50%, observed in two soil samples J8 of Jaipur and A4 of Ajmer region and the lowest moisture content found was 9.08% from B1sample of Barmer region. The moisture content observed for uncontaminated (control) soil was 15.0% which was higher than the contaminated soil. The average moisture content calculated for Jaipur, Ajmer and Barmer regions were 11.91, 12.04 and 11.04% respectively. The soil samples of Ajmer region had highest average moisture content as compared to Jaipur and Barmer regions. Barmer region had lowest average moisture content (Table: 2). The present study was supported by Rowell[15]. He reported the presence of petroleum hydrocarbon contamination in the soil samples decreased the moisture content of soil because hydrocarbons increase the hydrophobicity of soil thus reduced the moisture content. Dibble and Bartha[16] reported the optimal rates of biodegradation of oily sludge was to be 30-90% water saturation. In oil contaminated soil, Amund et al.[17] and Dibble and Bartha[16] reported a significant decrease in the moisture content due to oil rendered the soil hydrophobicity and hence reduced its water holding capacity thus complements present study . These findings supported the present research where the moisture content was also decreased.

Information about the electrical conductivity in the petroleum contaminated soil samples is not very much as compared to the uncontaminated soil. The highest

Electrical Conductivity (EC) recorded was 8.4 dsm^{-1} from the B3 sample of Barmer region and the lowest electrical conductivity recorded was 0.186 dsm^{-1} from J4 sample of Jaipur region. The electrical conductivity recorded for uncontaminated soil (control) was 0.15dsm^{-1} and found to be lower as compared to contaminated soil samples. The average electrical conductivity calculated for Barmer, Jaipur and Ajmer regions were 2.62, 1.54 and 1.05 dsm^{-1} respectively. In present research the electrical conductivity reported was higher in contaminated soils as compared to uncontaminated soil. Present result was favoured by Odu et al.[18] and Osuji[19], they reported the high electrical conductivity in the oil contaminated soil and suggested the reason for the greater electrical conductivity was due to the presence of high concentration of metal ion introduced from hydrocarbon contamination in the soil.

The maximum Total Dissolved Solids (TDS) value recorded was 937 ppm in J6 sample of the Jaipur region and the minimum total dissolved solids was reported 91.13 ppm in J7 sample of Jaipur region. The total dissolved solids reported for the soil samples collected from Jaipur region were in the range of 91.13-937 ppm and for Ajmer region were 91.21-458 ppm while the total dissolved solids reported for Barmer region was in the range of 93.34-857.9 ppm. The total dissolved solid reported for uncontaminated soil sample (control) was 88.5 ppm. Control sample had low TDS value as compared to contaminated soil samples (Table: 2). The average TDS value calculated for the soil samples of Jaipur, Ajmer and Barmer region were 461.3, 308.9 and 338.7 ppm respectively. In present study the high TDS value obtained in the contaminated soil was agreed with the results of Pathak et. al.[20] .

The highest Available Sodium (Na^+) concentration was recorded 7.6 meL^{-1} in J8 sample of Jaipur region and the lowest available sodium concentration was 2.5 meL^{-1} observed in J4 and A7 sample of Jaipur and Ajmer respectively. The available sodium concentration of the soil samples collected from Jaipur region varied from 2.5-7.6 meL^{-1} and the samples collected from Ajmer region were reported in the range of 2.5-5.5 meL^{-1} while Barmer regions soil samples were found to have available sodium concentration in the range of 2.6-4.6 meL^{-1} (Table 2). The available sodium concentration reported for the uncontaminated (control) soil was 2.0 meL^{-1}. The highest available sodium concentration was recorded in contaminated soil samples as compared to control soil. The average value of available sodium content calculated for the Jaipur, Ajmer and Barmer regions soil samples recorded were 4.9, 3.95 and 3.85 meL^{-1} respectively. In Present study the highest sodium concentration was obtained in contaminated samples as compared to umcontaminates sample and complemented by the results of Pathak et al.[20]. A contradictory result was obtained in a study by Kayode et al.[21], he reported reduced sodium concentration with the presence of spent lubricating oil in the polluted soils.

The highest Available Potassium (K^+) concentration recorded was 1.32 meL^{-1} found in B2 soil sample collected from Barmer and lowest potassium concentration observed was 0.50 meL^{-1} found in J5 soil sample collected from Jaipur region (Table: 2).. The available potassium concentration recorded for uncontaminated (control) soil was 0.48 meL^{-1} and was found less than contaminated soil. The average available potassium concentration calculated for the soil samples of Jaipur, Ajmer and Barmer region were

0.65, 0.72 and 1.0 meL^{-1} respectively. In present study, the petroleum hydrocarbon contaminated soil samples showed high potassium content as compared to uncontaminated soil sample. Present result was supported by Baruah[22] and Deka and Devi[23]. They reported the high rate of potassium content in the oil contaminated site due to the deposition of potassium in the upper layer of soil due to drilling process which caused leakage of saline effluent along with oil and resulted in the buildup of ionic concentration which resulted in the high concentration of potassium in the oil contaminated soil.

The highest value for the Organic Carbon content was 2.92 % recorded in A4 sample of Ajmer region and lowest value recorded was 0.10 % in B1sample of Barmer region. The organic carbon value recorded for uncontaminated soil (control) was 0.15 %. It was concluded from the readings that contaminated soil samples had higher organic carbon value as compared to uncontaminated soil. The average value of organic carbon content calculated for Jaipur, Ajmer and Barmer regions soil samples were found be 1.74, 2.0 and 1.32 % respectively (Table: 2). The present study concluded that contaminated soil samples had higher organic carbon value as compared to uncontaminated soil sample. Present result was supported by Baruah[22]. He reported that the soil heavily contaminated with crude oil represented high organic carbon content as compared to soil less contaminated with crude oil. The organic carbon is directly proportional to oil contamination. Udo and Fayemi[24] also reported increase in the organic carbon which indicated the increase of crude oil contamination in the soil. These findings were in agreement with present research work where the high organic carbon content was observed in petroleum hydrocarbon contaminated soil samples as compared to uncontaminated soil.

The highest Organic Matter reported was 5.04 % in A4 sample of Ajmer region and the lowest organic content reported was 0.31 % from B1 sample of the Barmer region. The organic matter recorded for uncontaminated (control) soil was 0.25%. The contaminated soil samples had higher organic matter content as compared to uncontaminated sample. The average organic content calculated for Jaipur, Ajmer and Barmer regions were 3.00, 3.46 and 2.31% respectively (Table: 2). The result obtained from present study for the organic matter content in the petroleum hydrocarbon contaminated soil showed high organic matter content. The contaminated soil samples had high organic matter content as compared to uncontaminated sample. Present result was supported by Clayden et al.[25] and McMurry[26]. They also reported high amount of organic matter from oil contaminated soil and suggested that high organic matter was due to the contamination of soil by automobile fuel consisted of hydrocarbons.

The highest Available Nitrogen (N) content observed was 0.059 % in B6 sample of Barmer region and lowest nitrogen content was 0.021% in J4 and B1 sample of Jaipur and Barmer region respectively. The nitrogen content observed for the uncontaminated sample (control) was 0.019%. The available nitrogen content was higher in contaminated soil as compared to control soil sample (Table: 2). The average nitrogen content calculated for Jaipur, Ajmer and Barmer region were 0.036, 0.043 and 0.035 % respectively. In present study the available nitrogen content was higher in contaminated soil samples as compared to uncontaminated sample. It was found in an experiment that soil contaminated with 10.6% oil attributed to increase the nitrogen content to 23% which leads to increase in total nitrogen content to 62% in the soil[24]. The study performed by Schwendinger[27] complements present work and suggested the reason of presence of high amount of nitrogen content in the crude oil contaminated area might be because of the atmospheric nitrogen fixation by the microorganisms which assimilate hydrocarbons. Present study was also favoured by Amund and Nwokaye[28] and Odu[29]. The findings of present work contradict with the study performed by Akoachere et al.[30]. They found no significant difference in nitrate and phosphate content between contaminated and uncontaminated soils. They does not confirmed the reason for their result but suggested that might be due to extent of contamination as well as some soil and microbial properties.

The highest Available Phosphorous (P) content was 0.04% recorded in B3 sample of Barmer region and the lowest available phosphorous content was 0.020% recorded in B9 sample of Barmer region. The available phosphorous content recorded for the uncontaminated (control) soil samples was 0.015%. The contaminated soil samples had higher available phosphorous than control soil sample (Table 2). The average phosphorous content calculated for the Jaipur, Ajmer and Barmer regions were 0.03, 0.04 and 0.02 % respectively. The present study was supported by Amund and Nwokaye[28] and Odu[29]. They also reported that the soil contaminated with petroleum hydrocarbon products showed large increase in phosphate content. Adams and Ellis[31] also favoured present study as they reported from their study that available phosphorous content increased with increased in the oil contamination in the soil. These studies compliments the observations made in present research work where high nitrogen and phosphorous content was found in petroleum contaminated soil samples. The petroleum contaminated soil samples exhibited high nitrogen and phosphate content as compared to uncontaminated soil sample. The findings of present work contradict with the study performed by Akoachere et al.[30] They found no significant difference in nitrate and phosphate content between contaminated and uncontaminated soils. They does not confirmed the reason for their result but suggested that might be due to extent of contamination as well as some soil and microbial properties.

Only the B3 soil sample collected from Barmer region showed Carbonate (CO_3^{2-}) content of 6.0 meL^{-1} while Jaipur and Barmer regions soil samples had no carbonate content. The carbonate content for the uncontaminated soil was 6.89 meL^{-1} (Table 2). The carbonate content in contaminated soil samples were less than uncontaminated sample (control). The average carbonate content was higher in Barmer region as compared to Jaipur and Ajmer regions (Figure 1.(k)). In present research work negligible or low carbonate content was obtained which was supported by the results obtained by Pathak et al.[20].

The highest Bicarbonate Content (HCO_3^-) reported was 5.0 meL^{-1} in J6 soil sample of Jaipur region and the lowest bicarbonate content was 0.5 meL^{-1} reported in J4, A2, B3, B6 and B7 soil samples collected from Jaipur, Ajmer and Barmer regions respectively (Table 2). The bicarbonate content reported for uncontaminated soil sample was 6.10 meL^{-1}. The bicarbonate content in contaminated soil samples were less than uncontaminated sample (control). The average bicarbonate content calculated for Jaipur, Ajmer and Barmer regions soil samples were 2.28, 1.17 and 1.07 meL^{-1} respectively. The Jaipur soil samples had highest average bicarbonate content value than Ajmer and Barmer soil samples. The Ajmer and Barmer regions soil samples had almost the similar average bicarbonate content. The present study was supported by Pathak et al.[20]. The low bicarbonate content in contaminated samples as compared to uncontaminated sample was also obtained in their study which complements present research work.

The highest Chloride Content (Cl^-) observed was 10.5 meL^{-1} from B3 soil sample of Barmer region and the lowest chloride content was 0.9 meL^{-1} recorded in J1 soil sample of Jaipur region (Table 2). The chloride content reported for uncontaminated soil sample was 0.5meL^{-1}. The chloride content in contaminated soil samples was higher than uncontaminated sample (control). The average chloride content calculated for Jaipur, Ajmer and Barmer regions were 5.71, 3.22 and 5.87 meL^{-1} respectively. From the result it was concluded that the chloride content in contaminated soil samples was higher than uncontaminated sample. In present study the high chloride content obtained in the contaminated soil was agreed with the results of Pathak et al.[20] and Onojake and Osuji[32].

Conclusion

From this study, it can be concluded that physico-chemical parameters play important role in the bioremediation technology. Bioremediation process or study can't be made successful without the prior knowledge of soil properties. These physico-chemical properties influence the microbial degradation of petroleum hydrocarbon. Contamination of petroleum hydrocarbon in soil cause alteration in the physico-chemical properties which leads to hazardous effect on environment, affect the vegetation, fauna and flora, microbial count etc. therefore it is a necessary step to assess the physico-chemical properties for the standardization of bioremediation protocols.

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