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Biochemical Properties of Vertisols and Performance of Fenugreek (*Trigonella foenum-graecum* L.) with Treated Industrial Effluent Irrigation

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Use of treated industrial effluent for irrigating agricultural crops has great significance to crop yield, soil fertility and soil biological activity. A field experiment was conducted on a Vertisol with fenugreek as test crop to compare treated industrial effluent from NCPL, Bharuch, Gujarat with best available tubewell water (BAW) as irrigation water. Plant height increased by 22.9 per cent with treated effluent irrigation as compared to only BAW. Highest fenugreek seed and straw yields were 42.8 and 74.0 g m^{-2}, respectively in 50% diluted effluent with BAW, and differed significantly from other options. Total N, P and K uptake by the plant parts was also improved when irrigated with treated effluent. The soil properties after harvest were influenced when treated effluent with or without dilution was used for irrigation. Soil organic carbon content increased due to irrigation with treated effluent as compared to BAW alone. Similarly, build-up of native N and P was observed in plots receiving treated effluent for irrigating the crop.

Key words: Industrial effluent, fenugreek, wastewater, soil properties

Current FAO estimates show that 1.2 million people in the world live in water scarce areas, which is expected to increase by 1.5 times in the coming decade. This is worsened further by the fact that most of the countries lack dependable water supply or policies for usage and distribution to the population. Growth of population, massive urbanization and rapid rate of industrialization has lead to greater demand for freshwater.

At present, 70% of surface and groundwater is used for agriculture. However, with increasing competition among agriculture, industry and domestic demand, agriculture would receive less water. In India, the per capita average annual freshwater availability has reduced from 5177 m^3 in 1951 to about 1869 m^3 in 2001 and is estimated to further come down to 1341 m^3 in 2025 and 1140 m^3 in 2050 (MOWR 2003). Despite improvement in existing water use techniques, increasing reliance of water scarce countries on alternate water sources for irrigation had been projected. One alternative source is the reuse of wastewater. A significant part of the wastewater is already used for crop production in response to limited availability of freshwater for agriculture. The total wastewater generated from all major industrial sources of India is about 83048 MLD (Biswas 2003).

Treated wastewater is considered as a potential water resource, because it contains considerable amount of nutrients that may prove beneficial for plant growth (Mishra and Behera 1991) and may curtail fertilizers input (Marecos do Monte *et al.* 1989). The quality of discharged effluent differs from industry to industry, and requires assessing its suitability for irrigation. Application of wastewater has been reported to improve crop yields, and impart other beneficial effects (Hati *et al.* 2007). A wide range of soil properties *viz*., NO_3-N, pH, available nutrient content increased significantly in wastewater treatment compared with the control *i.e.* groundwater (Yaryan 2000).

Fenugreek (*Trigonella foenum-graecum* L.), an important condiment belonging to the family *Papilionaceae*, is an annual herbaceous seed spice crop of India. This plant is cultivated in semi-arid

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region during winter (rabi) season and can be grown on sandy as well as heavy black cotton soils. Its seeds are used as spice and leaves as leafy vegetables which are rich in vitamins and minerals (Grower *et al*. 2002). The protein rich seeds have medicinal values and are also rich in sugars, mucilage, volatile oil, fixed oil, vitamins, enzymes and essential amino acids. Besides, it is also a good cattle fodder (Ahmadiani *et al*. 2001). In spite of great utility of fenugreek, little attention has been paid to enhance its productivity for remunerative and sustainable cultivation. Water requirement of fenugreek is 300-350 mm (Mehta *et al.* 2014), that is applied at frequent intervals of 20 to 25 days in 4 to 6 irrigations (Anonymous 2015). This offers an ample scope to reuse treated effluent for irrigation in fenugreek, especially in the regions where irrigation water availability is limited and rainfall is also scanty.

Majority of industries that lie along the heavy industrial belt of Gujarat state, do produce huge quantities of effluents. These effluents are subjected to various treatments and the final discharge has chemical parameters like, pH, electrical conductivity (EC), biological oxygen demand (BOD), chemical oxygen demand (COD), and concentrations of chloride, sulphate, free ammonia, ammonical nitrogen (N) and heavy metals well within threshold limit. Non-availability of sufficient fresh water and also the prevalence of poor quality ground waters in the salt-affected areas of the state compel the farmers to use industrial wastewater to meet the irrigation needs of the crops. Present study was, therefore, conducted to ascertain the efficiency of treated industrial effluent in fenugreek through a field experiment on Vertisol.

Materials and Methods

A field experiment was conducted at Bharuch (21.72° N and 73.01° E) with fenugreek as test crop, irrigated with treated industrial effluent from Aniline-TDI industrial unit of the Gujarat Narmada Valley Fertilizer Company Ltd. (GNFC), Bharuch, Gujarat and compared with irrigations through best available water (BAW). The experiment was laid in randomized block design with three replications and the treatments comprised treated effluent, 50% diluted treated effluent with BAW, and BAW.

Fenugreek seeds were sown in lines with row-to-row spacing of 30 cm and plant-to-plant spacing of 15 cm. Recommended rates of N, P and K were applied to the crop through urea, diammonium phosphate (DAP) and muriate of potash (MOP), respectively. The crop was irrigated through surface irrigation method as per the treatments and total four irrigations of about 3.5 cm each were applied.

The observations on plant growth parameters like height, leaf area and root length were recorded in fresh plant samples collected from respective plots. The plant samples from each plot were collected near maturity and were successively and thoroughly washed with tap water, 0.1 *N* HCl solution, and two washings with distilled water. After washing, the plant samples were air-dried and then dried in hot air oven at 65 °C to a constant weight. After recording the dry matter yield, plant samples were powdered in grinder. The powdered plant samples were digested in di-acid mixture for estimation of P, K, Na, Ca and Mg. The total N content was estimated through sulphuric acid digestion and micro-Kjeldahl distillation.

After harvest of the crop, the soil samples were collected from respective plots and analyzed for physicochemical properties and biological properties using standard analytical methods (Jackson 1973; Singh *et al*. 1999). Soil bacterial and fungal load were also assessed on nutrient agar and potato dextrose agar, respectively as per standard spread plate techniques in freshly collected soils at 60 days after sowing (DAS).

Results and Discussion

Physicochemical properties of soil

The physical and chemical analysis of the initial soil sample from the experimental site is presented in table 1. Surface soil pH was 7.31 and EC 0.34 dS m^{-1}. Soil organic C content was 4.8 g kg^{-1} and available N content was 142 kg ha^{-1}. In saturation extract, Ca and Mg content were 27.7 and 13.6 meq L^{-1}, respectively while Na and K were 33.7 and 8.25 meq L^{-1}. Bulk density of the soil was 1.55 Mg m^{-3}.

Chemical properties of treated effluent and BAW

The chemical parameters including heavy metal content of the treated industrial effluent were well within the permissible limits as per the pollution control board norms (Table 2). There was considerable reduction in NH_4-N, NO_3-N (about 90%), Na (9%), S (14%) and elevation in Mg (20%) and HCO_3 (33%) content due to dilution of treated effluent with BAW. Dilute aqueous ammonia can be converted to harmless N and water by wet air oxidation over Ni supported on Al_2O_3 catalysts (Kaewpuang-Ngam *et al*. 2004), which might have accounted for decrease in NH_4-N in treated effluent during dilution. Boufekane and Saighi (2013) recorded similar decline in NO_3-N

Table 1. Physicochemical properties of initial soil

Soil properties	Values	
	Soil depth (cm)	
	0-15	15-30
pH (1:2 w/v)	7.3	7.5
EC (dS m^{-1})	0.34	0.38
Org C (g kg^{-1})	4.8	4.3
Available N (kg ha^{-1})	142	138
Available S (mg kg^{-1})	12.6	16.7
Available P (kg ha^{-1})	45.9	63.1
Boron (mg kg^{-1})	0.66	0.82
Total N (mg kg^{-1})	0.010	0.012
Na (meq L^{-1})*	33.7	36.9
K (meq L^{-1})*	8.25	6.45
Ca (meq L^{-1})*	27.7	32.2
Mg (meq L^{-1})*	13.6	8.5
HCO_3 (meq L^{-1})*	220	207
CO_3 (meq L^{-1})*	-	-
Cl^{-1} (meq L^{-1})*	36.7	36.7
Bulk density (Mg m^{-3})	1.55	1.56

*saturation paste extract

Table 2. Chemical analysis of treated effluent, diluted effluent and BAW

Properties	Treated effluent (T_1)	50% diluted effluent with BAW (1:1) (T_2)	Best available water (BAW) (T_3)
pH	8.5	7.5	7.1
EC (dS m^{-1})	12.70	7.10	0.30
NH_4-N (mg L^{-1})	5.9	0.3	0.2
NO_3-N (mg L^{-1})	3.7	0.6	0.2
Sulphur (mg L^{-1})	27.6	23.7	21.2
Boron (mg L^{-1})	0.46	0.91	1.23
Na (meq L^{-1})	340	163	14.5
K (meq L^{-1})	6.8	5	2.6
Ca (meq L^{-1})	10.5	6	2
Mg (meq L^{-1})	2	2.5	2
HCO_3 (meq L^{-1})	10	20	20
CO_3 (meq L^{-1})	0	0	0
Cl^{-1} (meq L^{-1})	40	30	30

content of groundwater due to dilution during recharge.

Effect of treated industrial effluent on plant growth

The dry weight per plant was maximum (9.05 g) in 50% dilution of effluent with BAW followed by BAW, and treated effluent (Fig 1a). The increase in dry biomass observed with 50% dilution of effluent with BAW was 22.4 per cent over BAW, and the difference was statistically significant. Increase in drymatter yield of fenugreek with increasing concentrations of effluent was also observed by Kumar and Chopra (2012).

The plant growth parameters such as plant height, leaf area and root length differed among the treatments. The plant height was influenced significantly when irrigated with treated effluent compared to its application after dilution with BAW or BAW alone. It was 22.9 per cent more in sole effluent treatment as compared to BAW (Fig 1b). Highest root length of 17.3 cm was recorded with BAW and decreased with presence of effluent in irrigation water. No significant difference was observed for number of nodules per plant among the treatments, and average maximum number was recorded in BAW followed by diluted effluent and raw effluent. However, Sharma *et al*. (2011) reported positive impact of effluent on nodulation in fenugreek grown in soils of U.P. Irrigation with effluent hastened

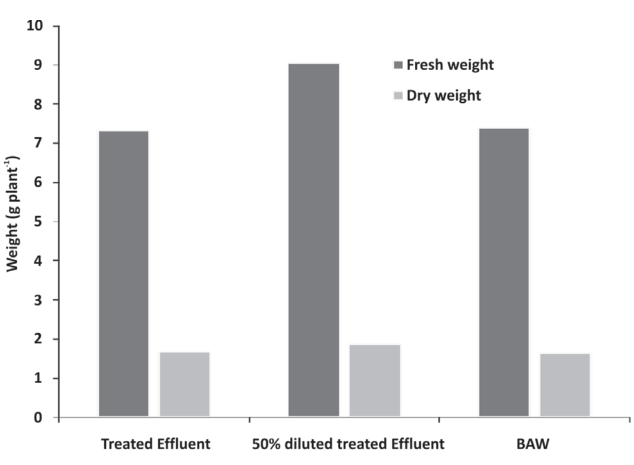

Fig 1a. Effect of treated effluent irrigation on growth of fenugreek

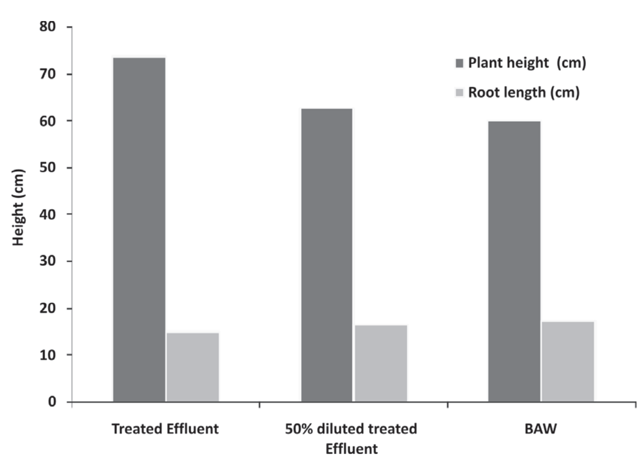

Fig. 1b. Effect of treated effluent irrigation on plant height and root length of fenugreek

the flowering by 5 days in fenugreek, suggesting a possible early maturity that may prove an added advantage in water scarce areas.

Effect of treated effluent on yield attributes and yields of fenugreek

Number of pods/plant was higher in diluted effluent (5.3) as compared to BAW (4.7). Irrigation with diluted effluent produced 30.3 per cent increase in leaf area followed by undiluted treated effluent (19.4 per cent) over BAW. Significant increase in pod length was observed where treated effluent was applied (22.9 per cent) or diluted effluent was applied (19.6 per cent) for irrigation compared to BAW (Fig 2a). Seeds per plant and 1000 seed weight were also significantly higher in diluted effluent by 20.0 and 22.5 per cent, respectively over BAW. Fenugreek seed and straw yields were maximum of 42.8 g m^{-2} and 74.0 g m^{-2}, respectively in diluted treated effluent and differed significantly to others (Fig. 2b).

Diluted effluents enhanced the growth of plant and this was reasoned out to the decrease in the concentration of various chemicals in the effluent and presence of root promoting phenolic compounds. The

Fig. 2a. Effect of treated effluent irrigation on yield-attributes of fenugreek

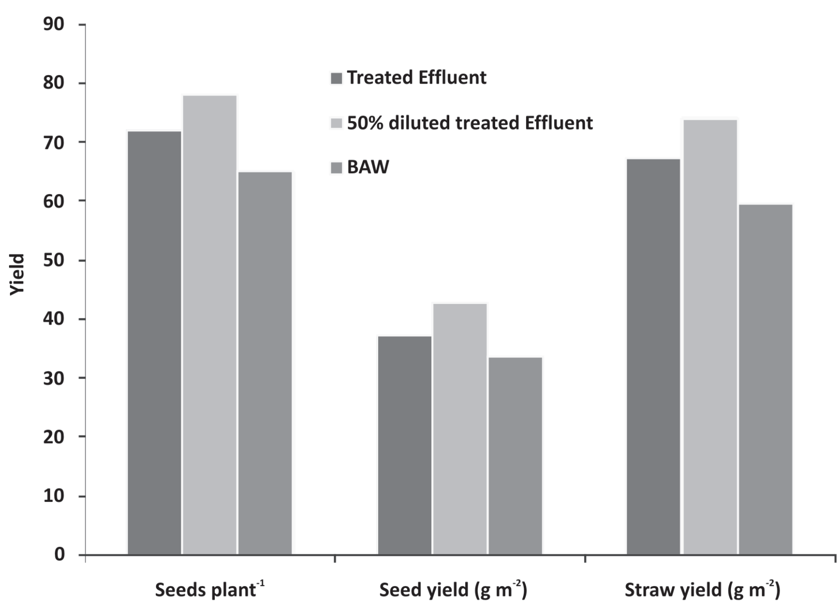

Fig. 2b. Effect of treated effluent irrigation on seed and straw yield of Fenugreek

growth of maize in terms of plant height, leaf area per plant and leaf area index increased due to irrigation with ten times diluted distillery effluent compared to the control *i.e.* good water (Singh and Bahadur 1997). The presence of nutrients in the effluents, may promote the growth through enhanced cell division, expansion and differentiation (Kumar *et al.* 2003). Use of industrial effluent continuously for three years has been found beneficial on the crop growth and yield of sunflower, and dill crops as no deleterious effect on soils and crop growth has been observed with use of effluent. Moreover, the bioavailability of micronutrient has been enhanced (Chinchmalatpure *et al*. 2014).

Effect of treated effluent on nutrient content of fenugreek

Since the treated effluent was rich in nutrients like N, P, S and Ca, uptake of important major nutrients like N, P and K by the plant parts was estimated. Higher Na content was observed under undiluted treated effluent application as compared to BAW (Fig. 3). Among the plant parts, highest Na content was recorded in stem (6.62 mg kg^{-1}) followed by leaf (6.48 mg kg^{-1}) and root (2.34 mg kg^{-1}). The highest K content in leaf and stem of fenugreek was under 50% diluted effluent irrigation, while the same in root was recorded under BAW (Fig. 3).

Highest content of Ca (120 mg kg^{-1}) and Mg (168 mg kg^{-1}) in leaf samples of fenugreek was recorded with undiluted treated effluent irrigation (Fig. 4). As compared to BAW, 154, 25 and 116 per cent increase was recorded in Mg content in leaf, stem, and root samples of fenugreek, respectively when irrigated with undiluted treated effluent.

Significantly higher N content in case of leaf and stem was observed in undiluted treated effluent, while that in root samples was under BAW. Total P content was 31.3 per cent higher in treated effluent compared to diluted treated effluent, and the same decreased significantly in all plant parts with BAW irrigation.

Effect of treated effluent on soil properties

Effect of treated effluent on soil properties and available nutrient status was observed with or without its dilution (Table 3). The pH of the surface soil increased from 7.1 in BAW irrigated plot to 7.7 and 7.8 in diluted and undiluted treated effluent, respectively. Similar effect due to treated effluent irrigation was also noticed in case of EC of the soils.

The Na content of soil increased by 14.3 per cent with treated effluent as compared to that with BAW (Table 3). The K content varied from 240-340 mg kg^{-1} and a build-up of 10.6 mg kg^{-1} in surface soil with undiluted treated effluent was observed over BAW. Calcium content in saturation extract of surface soil decreased in diluted treated effluent (21.5 meq L^{-1}) compared to initial value of 27.7 meq L^{-1}. Magnesium content of surface soil increased irrespective of the treatments. There was gradual increase (26.7%) in the chloride content under effluent

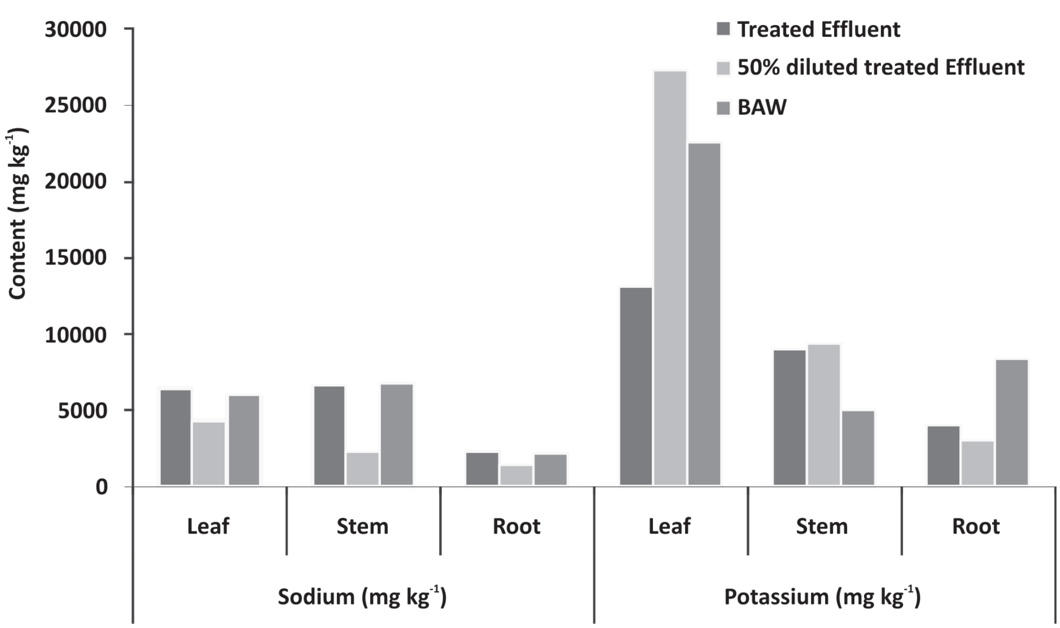

Fig 3. Effect of treated effluent irrigation on Na and K content of plants

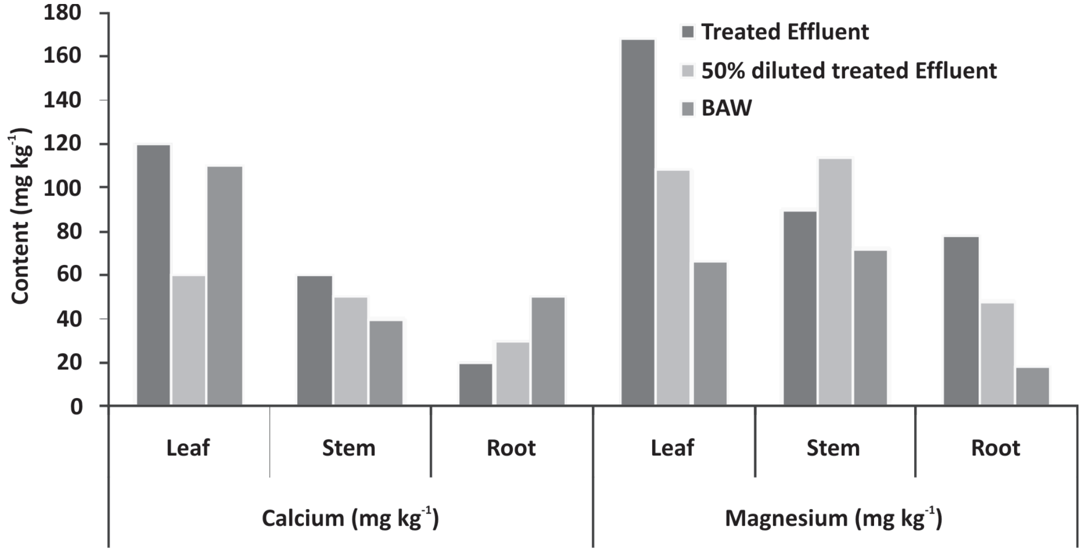

Fig. 4. Effect of treated effluent irrigation on Ca and Mg content of plants

treatment in both surface and sub-surface soil compared with that under BAW.

Irrigation with BAW or treated effluent affected the available nutrients such as N and P in surface soil. Compared with initial values, a build-up in available N and decline in available P content was noticed. However, exception was irrigation with undiluted treated effluent, where available P content increased compared to that in the initial soil. Available S content of soil decreased from the initial status in all treatments, and maximum reduction was noticed with diluted treated effluent. Hati *et al*. (2007) reported improvement in soil physical properties with the application of distillery effluent, and concluded that it could be used as a substitute for fertilizers and manure.

Irrigation with effluents and sewage water results in increasing the nutrient contents in soil and plants (Kour and Arora 2010). In wastewater treatment, NO_3-N, pH, available Mn and bulk density of soil increased, whereas EC and total porosity decreased significantly. In addition available P, K, Mn, Ni and pH increased significantly compared to control (Yaryan 2000).

The distillery effluent irrigation helps to build-up the soil fertility by increasing organic carbon, and available P and K status (Singh and Bahadur 1997). Zalawadia *et al*. (1996) stated that the improvement in the soil fertility could be credited to the inadvertent addition of total N, P and K to the tune of 204, 153 and 1139 mg kg^{-1} soil, respectively through distillery water. The effluents are also good sources of

Table 3. Effect of treated effluent irrigation treatments on soil properties

Soil properties	Treated effluent (T_1)		50% diluted treated effluent (T_2)		BAW (T_3)	
	0-15 cm	15-30 cm	0-15 cm	15-30 cm	0-15 cm	15-30 cm
pH (1:2 w/v)	7.81	7.87	7.74	7.17	7.13	7.33
EC (dS m^{-1})	2.40	2.50	1.90	1.70	0.53	0.71
Organic C (g kg^{-1})	6.59	6.59	7.22	6.28	5.02	5.34
Available N (kg ha^{-1})	188	176	226	188	169	138
Available P (kg ha^{-1})	52.5	60.6	32.9	20.0	18.7	19.3
Available B (mg kg^{-1})	1.86	0.36	0.76	0.27	0.57	0.34
Available S (mg kg^{-1})	2.50	2.03	1.57	1.23	8.34	2.54
Na (meq L^{-1})*	13.5	13.3	12.9	13.1	11.8	11.7
K (meq L^{-1})*	7.65	8.68	7.66	7.50	7.38	6.35
Ca (meq L^{-1})*	46.0	42.5	21.5	45.0	50.5	42.5
Mg (meq L^{-1})*	34.5	37.0	41.5	32.5	33.5	41.0

*saturation paste extract

Table 4. Effect of treated effluent irrigation treatments on soil biochemical and microbiological properties

Treatment	Parameters					
	NH_4-N (mg kg^{-1})	NO_3-N (mg kg^{-1})	Alkaline phosphatase (µg PNP g^{-1} h^{-1})	Dehyrogenase (µg TPF g^{-1} d^{-1})	Bacteria (cfu g^{-1})	Fungi (cfu g^{-1})
Treated effluent	17.9	22.2	9.00	0.69	99×10^6	40×10^4
50% diluted effluent with BAW	15.4	16.5	8.00	0.91	50×10^6	120×10^4
BAW	15.0	15.3	13.50	0.43	41×10^6	100×10^4

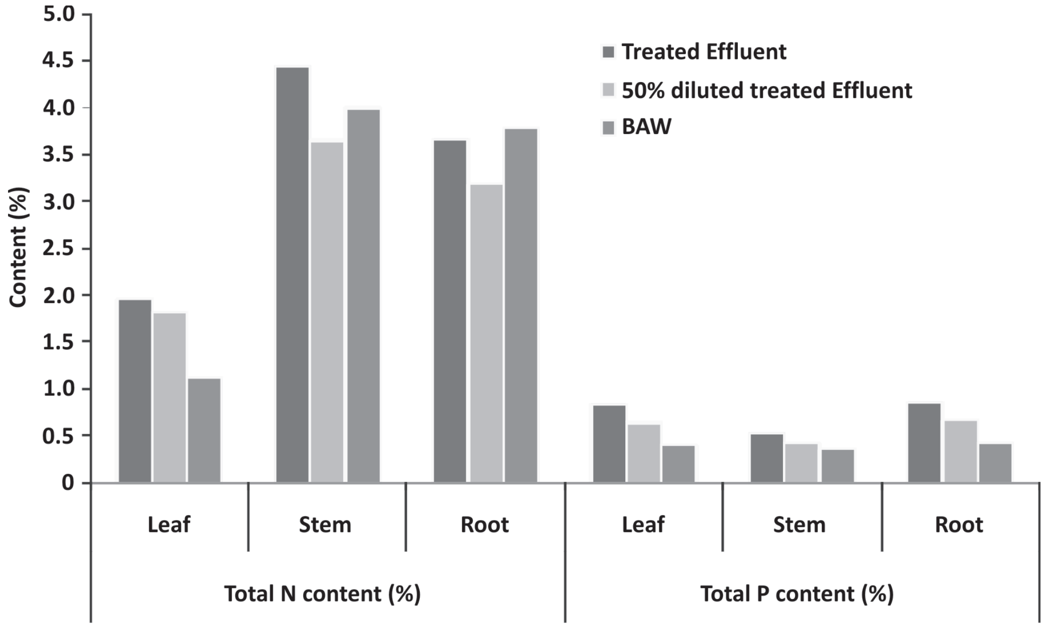

Fig. 5. Effect of treated effluent irrigation on total N and P content of plants

micronutrients as the available micronutrients (Fe, Mn, Zn and Cu) increased with distillery effluent application and availability being the highest in 1:10 dilution. Similar results were reported by Bhise *et al.* (2007).

Effect of treated effluent on soil biochemical properties

Biochemical properties were estimated in fresh soil samples, which revealed that dehydrogenase activity was to the tune of 0.91 µg TPF g^{-1} d^{-1} under diluted treated effluent compared to 0.43 µg TPF g^{-1} d^{-1} in BAW irrigation (Table 4). The alkaline phosphatase activity was 13.50 µg PNP g^{-1} h^{-1} in BAW, which was decreased considerably due to application of treated effluent with or without dilution with BAW. The mineral-N, which included NH_4-N and NO_3-N, was higher in soils irrigated with treated effluent. Higher enzyme activity was also reported by Pradeep and Narasimha (2012) in effluent treated soil over control soil (adjacent site not receiving effluent discharge). Soil with effluent discharges exhibited higher phosphatase activity as reflected by more release of *p*-nitrophenol than control soil. Relatively high dehydrogenase activity in soil samples with effluent discharges may be due to high amount of organic matter and rich microbial population in soil samples with effluents (Narasimha *et al*. 2012).

Effect of treated effluent on soil microbiological properties

Fresh rhizospheric soils of fenugreek were assayed for microbial load as influenced by the treatments at 30 and 60 days after sowing (DAS). Total population of soil bacteria at 60 DAS was higher under undiluted effluent irrigation (Table 4), but the diversity was more under diluted treated effluent irrigation (7 bacterial isolates were isolated from rhizospheric soil). However, fungal populations decreased when soils were irrigated with treated effluent either in diluted or undiluted condition. This may be due to increase in pH of the soil because of alkaline nature of the effluent (pH 8.5). Higher bacterial and fungal population was reported in soil contaminated with effluents (Rabah *et al*. 2010). Also, two-fold increase in bacterial and fungal population was observed in effluent-treated soils as compared to control (Pradeep and Narasimha 2012). The reason for more number of microbes at lower concentrations was pointed to the reduced level of toxic metabolites due to dilution factor (Dhevagi *et al*. 2000).

Bacterial colonies were characterized by observing the cultural characteristics in terms of colour, size, shape, margin and appearance of colony. Large to small sized colonies were observed in soil receiving sole effluent, while in soils of T_2 and T_3 treatment plots colonies were of intermediate size. All the bacterial isolates irrespective of treatment were cocci with no pigmentation, except 3 colonies that were yellow to orange pigmented. Among the bacterial isolates, higher frequency of *Bacillus* spp and *Staphylococcus* spp were noted. The isolated fungi were whitish to yellow in colour with filamentous margin, and appeared to be *Aspergillus*, *Penicillium* and *Mucor*.

Conclusions

It is concluded from the study that properly diluted industrial effluent can be used for crop production as it could meet water demands and partially meet nutrient requirement. Application of treated effluent as irrigation water significantly increased plant height, yield-attributes and yield of crop as compared to control (BAW). Also, soil properties and microbial population were positively influenced by the use of treated effluent. Keeping in view the variability in quality and characteristics of industrial effluents, these should be applied as irrigation water judiciously and with proper care and management.

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