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EFFECTS OF FERTILIZATION WITH LIQUID EXTRACTS OF BIOGAS RESIDUES ON THE GROWTH AND FORAGE YIELD OF ALFALFA (*MEDICAGO SATIVA* L.) UNDER ARID ZONE CONDITIONS

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Abstract

In this study liquid extract of biogas residue (LEBR) from date palm waste was used to fertilize soil for alfalfa (*Medicago sativa* L.) cultivation. The results revealed that LEBR application significantly increased plant height, number of branches per meter and the contents of crude protein and ash over the control. The largest plant height and number of branches per meter and the highest ash and Phosphorus contents in the plant dry matter were obtained at the application of 3 L/m^2 of LEBR solution containing 15% extract, to the soil while a solution containing 10% extract gave the highest fresh and dry biomass yields. Soil treated with LEBR had higher EC and higher contents of NH_4, NO_3, Na, K, Ca, Mg, Fe, Mn, Zn and Cu than the untreated soil. The pH of the soil decreased with increased amount of LEBR applied.

Introduction

The coarse and arid soils of Saudi Arabia are very poor in plant nutrients. They have low organic matter content, high calcium carbonate content and high pH (Anon., 2005). In general, soil fertility is related to the physical, chemical and biological properties of that particular soil (Bulluck *et al*., 2002; Knoepp *et al*., 2000). Organic matter is a key component of soils which affects their physical and chemical properties such as water retention, accretion erodibility, cation exchange capacity and nutrient availability (Rice, 2002). The maintenance of soil organic matter levels and the optimization of nutrient cycling are essential to the sustained productivity of agricultural systems (Khan *et al*., 2013). Both are related closely to the bioturbating activities of macrofauna and the microbially-driven mobilization and immobilization processes, which also encourage the activities of large invertebrates. Recycling of organic residues to arable soils proved to be an efficient way of improving the physical, chemical and biological properties of the soil (Odlare *et al.*, 2008). Many researchers reported the use of biogas residues as soil fertilizer with success (Maunuksela *et al*., 2012; Diacono & Montemurro, 2011; Haraldsen *et al*., 2011). The residues generated from biogas processes have a higher concentration of NH_4^+ compared with conventional animal manure and compost, hence their potential fertilization value is also higher (Zhong *et al*., 2010). In addition, most elemental nutrients such as P, K, Mg and a number of other essential trace elements from the raw material fed to the biogas process remain in the biogas residue (Abubaker, 2012). The organic compounds not broken down during the biogas process increase the carbon content of farmland and improve its biological activity as they break down in the soil (Bessam & Mrabet, 2003).

Saudi Arabia is one of the largest growers of date palm trees in the world (Anon., 2013). Lots of wastes are produced annually from these trees constituting potential environment pollutants. A new trend in the country is the removal of these wastes through biogas producing anaerobic digestion processes. The residues that remain after biogas digestion can be recycled by using them as soil fertilizers. In this study we used liquid extracts from residues of biogas produced from date palm tree wastes to fertilize soils for alfalfa crop cultivation. The use of extracts of biogas residue is not a common practice in soil fertilization with biogas sludge. Its use reduces bulks applied, also reduces bad odors and it is expected that the plants may utilize the nutrients present in the liquid extract quicker than utilizing the nutrients present in the residue containing the solids.

Materials and Methods

The study was conducted in a complete randomized block design with 3 replications. Alfalfa of the Siriver cultivar was used as a test crop. Plot sizes were 4×3 m and rate of seeding was 20 kg h^{-1}. Three levels of liquid extract of biogas residues, viz solutions containing 5, 10 and 15% extract, were applied as spray on the soil before irrigation in addition to the control (no organic fertilizer). The liquid extracts, made by cloth straining, were obtained from biogas residues produced by the authors from anaerobic digestion of date palm tree wastes. Three liters were applied from each extract concentration per square meter soil area. In addition, all plots received 25:100:25 kg NPK fertilizer per hectare before cultivation. The crop was planted in 15 Feb. 2012 and the treatments applied after 21 days from planting date and after every cut. Weeds were removed mechanically two times and plants irrigated every week by furrow surface irrigation. Data were collected from 5 cuts starting from the second one. In each cutting, two outer rows of each plot and 50cm space from each end of the plots were left as borders and the middle 3 meters of the central rows were harvested.

Plant height and dry matter contents: Data about plant height and number of branches per meter was recorded by selecting ten plants randomly from each plot. The plant height was measured using a measuring tape from ground level to highest leaf tip. For dry matter content, plant samples were dried in shade then shifted to electric oven at 70°C till a constant weight was reached.

Chemical analysis: For chemical analysis, plant samples were dried and ground to pass through a sieve with 0.50 mm pore size. A composite sample of 30 cm deep soil was used for physical and chemical analysis before preparing the soil for planting and after harvest. Crude protein and total ash contents in plants were determined using Anon. (1984) methods, while P, Ca, K, Mn, Fe, Zn and Cu contents were determined using the method proposed by Westerman (1990).

Statistical analysis: Data reported were means of determinations made on three sample replicates and 5 cuts. Less significant difference test (5% level) was used to determine significant differences between treatments (Steel & Torrie, 1980).

Results and discussion

Soil characteristics: The soil used to grow the crop in this work was loamy sand with salinity level of 2.28 mS cm^{-1} (Table 1). According to reports of Anon., (1954), such a soil is suitable for normal plant growth. The pH of the soil was 7.4, which lies within the range reported by Zucconi & Berttoldi (1987) for optimum plant growth (between 6.5 and 8.5). The organic matter (0.57%) and total nitrogen (0.08%) contents and the cation exchange capacity (8.6 $cmol_c$ kg^{-1}) of the soil were low. On the other hand, the soil was characterized by high levels of available P (8.5 ppm) and K (362 ppm), while the contents of available Fe, Mn, Zn and Cu were 6.8, 3.4, 4.1, 4.7 ppm, respectively (Table 1).

Characteristics of liquid extract of biogas residue (LEBR): The properties of LEBR are shown in Tables 2a and 2b. The EC of the liquid did not exceed the limit of 3 mS cm^{-1} as recommended by Anon., (1995). The pH was slightly alkaline (8.2) but was within the normal range suitable for plant growth (Zucconi & Berttoldi, 1987). The total heavy metal contents (Table 2b) were below the limits set for bio-solids intended for agricultural use (Anon., 1986). This implies that this material can be used in agriculture.

Table 1. Some chemical properties of the soil used in the study.

Property												
Texture	EC mS cm^{-1}	$CaCO_3$ %	pH	Organic matter %	CEC $cmol_c$ kg^{-1}	Total nitrogen %	Available nutrients (ppm)					
							K	P	Mn	Fe	Zn	Cu
Loamy sand	2.39	10.80	7.4	0.57	8.6	0.08	362	8.5	3.4	6.8	4.1	4.7

The pH and EC were determined in saturated soil paste extraction. $CaCO_3$ and texture estimated according to Black (1965)

Available K extracted using 0.1 N NH_4OAC buffered at pH 7.0, Mn, Fe and Zn using DTPA (Lindsay & Norvell, 1978), phosphorus determined according to Olsen *et al*., (1954) and total nitrogen determined according to Bremner (1965)

Table 2a. Some chemical properties of liquid extract of biogas residue.

Parameters						
EC mS cm^{-1}	pH	OM %	C/N ratio	NO_3^- mg kg^{-1}	P-$NaHCO_3$ mg kg^{-1}	K-NH_4OAC mg kg^{-1}
2.07	8.2	25.2	30.2	14.7	25.7	142

Table 2b. Some chemical properties of liquid extract of biogas residue.

Parameters						
Ca-NH_4OAC mg kg^{-1}	Mg-NH_4OAC mg kg^{-1}	Cu-DTPA mg kg^{-1}	Pb-DTPA mg kg^{-1}	Zn-DTPA mg kg^{-1}	Fe-DTPA mg kg^{-1}	Mn-DTPA mg kg^{-1}
2108.9	3145.2	4.9	10.8	32.1	228.6	12.9

Effects of liquid extract of biogas residue (LEBR) on alfalfa growth and yield parameters: It is clear from the data presented in Table 3 that LEBR application significantly affected the plant morphological traits such as height, number of branches per meter and fresh/dry weight yields. The largest plant height (75.9 cm) and number of branches per meter (73.2 branches m^{-1}) were obtained at LEBR application rate of 15%, while the highest fresh weight yield (1.90 kg m^{-2}) and dry hay yield (5.75 ton ha^{-1}) were obtained at the rate of 10%. Unlike the effect of 15% LEBG application, no significant differences in plant height were observed between the control and the plants that received 5 and 10% LEBR, though the height of the 15% plants was not significantly larger than those of the 5 and 15% LEBR (Table 3). With respect to the number of branches per meter, application of 10 and 15% LEBR yielded values significantly higher than the control but not significantly different from each other. The lowest values for all recorded parameters were achieved when no LEBG was applied. It could be concluded that applying LEBR increased soil fertility and reduced its pH hence leading to improved plant growth and increased yields (Munir *et al*., 2005). It might also have caused better root development which in turn provided a better habitat for the activity of biological nitrogen fixing bacteria. In addition, the higher root mass exploit the soil from the surrounding more effectively and improves the nutrient availability for plants. Grewal *et al.*, (2000) and Naagar *et al.*, (2004) have also reported a significant increase in yield of cluster bean by fertilizer application. Significant differences in fresh and dry matter yields at various biogas residue application rates have also been reported by Abubaker (2012). Therefore, application of biogas residues can be expected to lead to different effects in arable soil compared with the use of regular organic or inorganic fertilizers (Levén & Schnurer, 2005; Engwall & Schnürer, 2002; Angelidaki *et al.*, 2000; Grossi *et al*., 1998; Abbas *et al*., 2013).

Effects of liquid extract of biogas residue (LEBR) on alfalfa quality parameters: Application of LEBR did not cause significant changes in the plant contents of dry matter, crude protein and calcium. The contents of total ash and phosphorus were significantly higher at the 15% LEBR than the control and the 5% LEBR; however they were not significantly higher than those obtained by 10% application (Table 4). Still, the application of LEBR at 15% rate produced the highest contents of total ash (9.7%), Ca (2.31%) and phosphorus (0.59%) while the highest protein content (21.1%) was achieved with the 10% treatment and the highest dry matter content (31.9%) at the zero treatment. It can therefore be said that application of LEBR improved most quality parameters of alfalfa crop. This can again be attributed to improved soil physical, chemical and biological activity properties such as water retention, accretion erodibility, cation exchange capacity and nutrient availability which resulted in more absorption of minerals. These findings are in agreement with findings reported by Diacono & Montemurro, (2011); Chantigny *et al.*, (2008); Kocar, (2008); Jabeen & Ahmad, (2013).

Effects of liquid extract of biogas residue (LEBR) on some soil properties: As shown in Table 5, the pH of the soils treated with LEBR was less than that of the soil treated with inorganic fertilizer only with significant differences at the 10 and 15% application rates. Difference in the pH values of LEBR and inorganic fertilizers probably contributed to their different effect on soil pH. LEBR can also lower the pH of soils because of the nitrification reaction and microbial production of CO_2 in the soil and around root systems (Pocknee & Summer, 1997). Concentration of soluble salts, expressed as EC value, was higher in the soil treated with LEBR than in the conventionally fertilized soil, the differences being significant at the 10 and 15% LEBR application rates (Table 5). At 0-2 mS cm^{-1} salinity effects are mostly negligible and at 2-4 mS cm^{-1}, yields of very sensitive crops may be restricted (Katerji *et al*., 2005). The alfalfa grown in this experiment has medium salt tolerance (Anon., 1982) and no salt damage to the crop was observed.

Table 3. Growth and forage yield parameters of alfalfa as affected by various liquid extract of biogas residue (LEBR) levels.

Treatment (% LEBR)	Plant height (cm)	No. of Branches (m^{-1})	Fresh weight yield (kg m^{-2})	Dry hay yield (t ha^{-1})
Zero	60.8^{b}	50.7^{c}	1.26^{c}	4.03^{b}
5	66.9^{ab}	55.1^{bc}	1.52^{b}	4.75^{b}
10	72.5^{ab}	65.9^{ab}	1.90^{a}	5.75a
15	75.9^{a}	73.2^{a}	1.38^{cb}	4.29^{b}
$LSD_{0.05}$	14.30	14.70	0.25	0.88
CV%	10.37	12	8.23	9.31

Zero = inorganic fertilizer at 25:100:25 kg NPK ha^{-1}

Means in each column for each parameter followed by the same letter are not significantly different according to least significant differences at 0.05 level of significant ($LSD_{0.05}$)

Table 4. Forage quality parameters of alfalfa as affected by various liquid extract of biogas residue (LEBR) levels.

Treatment (%LEBR)	Dry matter (%)	Crude protein (%)	Total ash (%)	Ca (%)	P (%)
Zero	31.9[a]	19.7[a]	6.5[c]	1.90[a]	0.32[c]
5%	31.3[a]	20.2[a]	7.2[bc]	1.95[a]	0.45[b]
10%	30.2[a]	21.1[a]	8.9[ab]	2.20[a]	0.53[ab]
15%	31.1[a]	20.5[a]	9.7[a]	2.31[a]	0.59[a]
LSD0.05	1.76	3.23	2.41	0.46	0.113
CV%	2.82	7.94	15	11	12

Zero = inorganic fertilizer at 25:100:25 kg NPK ha^{-1}

Means in each column for each parameter followed by the same letter are not significantly different at the 0.05 level according to least significant differences ($LSD_{0.05}$).

Table 5. Effect of treatment with liquid extract of biogas residue (LEBR) on EC, pH and elemental composition (in ppm) of the soil after alfalfa harvest of fifth cut.

Parameters	Treatments				LSD 0.05	CV%
	0%	5%	10%	15%		
EC, mS cm^{-1}	2.28[b]	2.35[b]	3.05[a]	3.22[a]	0.48	2.71
pH	7.8[a]	7.5[ab]	7.3[b]	7.2[b]	0.40	8.82
NH_4-N	6.9[b]	7.3[ab]	7.5[ab]	7.9[a]	0.66	4.48
NO_3-N	12.2[d]	25.2[c]	30.8[b]	38.8[a]	1.78	3.32
P-$NaHCO_3$	2.75[d]	7.58[c]	14.0[b]	17.47[a]	3.31	15.87
K-HN_4OAC	205[c]	297[b]	325[ab]	375[a]	58.45	9.80
Ca-HN_4OAC	2000[b]	2050[ab]	2130[ab]	2205[a]	186.08	4.44
Mg-HN_4OAC	94[b]	104[ab]	108[a]	112[a]	13.20	6.32
Fe-DTPA	21.3[b]	27.9[ab]	35.6[ab]	40.1[a]	17.07	27.35
Mn-DTPA	4.3[d]	6.7[c]	7.9[b]	9.5[a]	0.97	6.85
Zn-DTPA	2.2[b]	2.3[b]	2.5[b]	3.7[a]	0.69	12.99
Cu-DTPA	0.9[d]	1.5[c]	2.4[b]	3.1[a]	0.51	12.67

Means in each row for each parameter followed by the same letter are not significantly different according to least significant differences at 0.05 level of significant ($LSD_{0.05}$)

The NH_4-N concentration in the soil with LEBR treatment was generally higher than that in the soil without LEBR treatment, with significant difference at the 15% rate. Increasing the application rate of LEBR significantly increased the content of NO_3-N at all levels of application (Table 5). The high NO_3-N content of LEBR treated soils indicate that the organic nitrogen in biogas residue extract was being mineralized to NO_3-N. Significant increases in the contents of P, K, Ca, Mg, Fe, Zn and Cu over the control were also observed at all levels of application of LEBR for some and at 15% for others (Table 5). Concentration of essential trace elements in soils is usually increased by biogas residue application, especially Fe, Mn, Zn and Cu (Sarwar *et al*., 2008; Ouedraogo *et al*., 2001).

Conclusions

Liquid extract of biogas residue produced from date palm tree seems to be a suitable soil fertilizer for alfalfa production. Its application improved soil characteristics, increased crop yield of alfalfa and improved the nutritional value of the crop. Best results were obtained at application of solutions containing 10-15% extract. Such approaches can reduce bulk application of fertilizer, are more environment friendly and it is expected that the plants may better and efficiently utilize the nutrients present in a liquid extract than in a residue containing solids. Hence, it is recommendable to use such bio-fertilization techniques for crop production.

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