

Response of snap bean to biogas slurry combined with either rock Phosphate or super phosphate fertilizers

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Abstract

This research was carried out to study the potential use of by-products of biogas production, i.e. biogas slurry (BS) generated from cattle dung, as nutrient sources in agriculture. The pot trials were conducted under green house conditions on the growth, yield of snap bean and their chemical composition. Some soil enzymes, i.e. nitrogenase and dehydrogenase were determined, also available P. Snap bean was fertilized with various sources of phosphate fertilizers, calcium superphosphate or rock phosphate at the rate of 30 kg P₂O₅/fed combined with BS.

Results showed that vegetative growth parameters significantly increased by using BS and RP or SP for two used varieties, applying of 10 m³BS + 30kg P₂O₅/fed as SP gave the maximum values of vegetative growth parameters for Paulista variety. For other variety, Xera, 10 m³BS + 30kg P₂O₅/fed as SP led to the highest plant height. While 2.5 m³BS + 30kg P₂O₅/fed as RP gave the highest values of plant dry weight and No of branches. With respect to yield and its components, pods weight /plant and pod length of Xera cultivar significantly increased over control, also pods weight /plant for another cultivar significantly increased over control as a result of using BS and various sources of P fertilizer

Regarding the leaves chemical composition, using Bs combined with RP or SP caused significant increase of N, P, K as macronutrients, where 10 m³BS + 30kg P₂O₅/fed as RP gave its highest values for two used varieties. Micronutrients Fe, Mn, Zn and Cu significantly increased over control. Applying of rock phosphate (RP) with BS was more effective on most of these nutrients than super phosphate (SP) + BS for the two cultivars used.

For the pod chemical composition which significantly increased, N, K and total protein of Xera variety reached to the maximum values by using SP + 10 m³BS. While the highest value of T.P% obtained by RP +10 m³BS. Fe, Mn and Zn concentration (ppm) enhanced by using RP +10 m³BS. For Paulista cultivar, T.N, T. protein and Fe increased up to the greatest values by RP +10 m³BS, while T.P and Mn reached to the maximum values by using SP +10 m³BS.

With regard to biological activity and available phosphorus, data showed significant increases by the treatment under this study. Clearly applying RP along with 10 m³BS gave the highest values of dehydrogenase, nitrogenase enzyme activity and total count of bacterial more than those obtained by SP +10 m³BS, while the best results for available P were obtained by SP +10 m³BS. Accordingly, this study ensured that the use of rock phosphate with biogas slurry may be recommended as a substitute for super phosphate with biogas slurry in snap bean production.

Key words: biogas slurry, rock phosphate, super phosphate, nitrogenase enzyme, dehydrogenase enzyme, snap bean, N, P, K, Fe, Mn, Zn, Cu.

Introduction

Snap bean is one of important vegetable in Egypt, which has a great weight for both local consumption and export. Egyptian Ministry of Agriculture aimed to extend the cultivation area of snap bean especially in newly reclaimed land because of its economic value. Snap bean is the second exportable vegetable in Egypt.

Lack of adequate nutrient supply and poor soil structure are the main constraints to agricultural production systems in low-input agriculture. Chemical fertilizers are not the most appropriate solution to overcome the seconstraints because use of chemical fertilizers is expensive and a threat to human health and the environment [Weltzein (1990)]. Integration of organic with inorganic fertilizers improves the physiological system of the crop, provides adequate growth regulating substances and modifies soil physico-chemical behavior and results in augmented crop yield (Hukkeri et al, 1977). Hence,

it is suggested that there should be an emphasis on finding alternatives to chemical fertilizers such as biogas slurry, compost which are cheaper than other sources of nutrients and relatively safe. Nutrients can be added to the soil through inorganic fertilizers or organic input including biogas slurry, farm yard manure (FYM), animal waste and crop residues. Biogas slurry is a by-product obtains from the biogas plant after the digestion of dung or other biomass for generation of methane rich gas. Biogas slurry supplies essential nutrients enhance water holding capacity, soil aeration, accelerates root growth and inhibit weed seed germination. It contains appreciable amounts of organic matter (20 to 30%). Cattle dung, crop residues and other aquatic biomass are available in large amounts in many of the developing countries. It can be used in liquid, compost, and dry form and is a very good fertilizer/composting substance for agricultural crops. It is a more easily available form of compost than traditional compost (Gurung1998). Regarding increased yields, Gurung1998 stated that the most

responsive crops to bio slurry and bio slurry compost are vegetables; root crops, potatoes, fruit trees, maize, and rice. Furthermore, bio slurry has the potential not only to improve soil fertility and soil structure, but also to act as a plague repellent. Finally, seeds treated with bio slurry have given better germination rates. Application of digested bio slurry increases the crop yield, quality of vegetables like size and shapes. It also helps in reduction of dependence on mineral fertilizer (Karki, 1996). Yield increase due to bio-slurry application has been reported in many crops. Vegetable crops produced with bio-slurry have better quality as compared to those produced with chemical fertilizer (Krishna, 2001).

Biogas is an important component for energy production from renewable resources (Møller *et al.*, 2009) and for this reason biogas slurry, the secondary product of the anaerobic digestion process, is increasingly abundant and used as a fertilizer (Møller, 2009). To investigate the effects of biogas and raw slurry on Italian ryegrass yield, N uptake (two harvests), soil microbial biomass, soil fungi, and root-colonizing microorganisms, Wentzel and Joergensen (2016) found that Biogas slurries increased the mean total above-ground plant biomass by 66% and raw slurries by 35% in comparison to the control. The mean plant N-uptake increased under biogas and raw slurry application by 166% and 65%, respectively, compared with the unfertilized pots.

Various trials have compared the yields from inorganic fertilizer, organic fertilizers and or combination of both. In many cases, combination of organic and inorganic fertilizers have produced higher yields (Sajjad and Shad, 2014), also Zebider (2011) stated that there is a need to apply both organic and inorganic fertilizers in a balanced way to get expected output or the crop yield.

Compost and bio slurry improves the physical, chemical and biological quality of soil besides providing both macro and micro nutrients to crops. The improvement in qualities include improvement in soil structure, water holding capacity, electrical conductivity, bulk density, lesser soil erosion, preventing the leaching of nutrients and provide nutrients to soil micro flora (Fentaw, 2010). Bio-slurry, the residual manure generated through anaerobic decomposition of various organic materials is considered a quality organic fertilizer. About 25 to 30% of organic matter is converted into biogas during the anaerobic fermentation process, while the rest becomes available as manure (bio-slurry). This residual manure is normally rich in macro and micro nutrients (Islam, 2006; Thu, 2007). Yield responses of vegetable crops to bio-slurry manure application have been reported in different crops including okra (Shahbaz, 2011) maize and cabbage (Karki, 2001). Garg *et al.*, 2005 reported that fertilization of soil with biogas slurry generated from cattle dung improved the yield of wheat over non-modified controls. Grain yield increased with the application of biogas residue,

which was attributed to the lower bulk density of soil, increased hydraulic conductivity, and greater moisture retention. Consequently, the improved status of nutrients through amendment of the physical properties of soil contributed to the higher yield of wheat.

Phosphorus (P) is the second important key plant nutrient which affects the overall growth of plants by influencing the key metabolic processes such as cell division and development, energy transport (ATP, ADP), signal transduction, macromolecular biosynthesis, photosynthesis and respiration (Shenoy and Kalagudi, 2005; Khan *et al.*, 2009, 2014). Soils contain very little total P (0.02–0.5% (w=w); Fernandez *et al.*, 2007), of which only 0.1% is available to plants (Zou *et al.*, 1992). Thus, P needs to be applied to soils as soluble P fertilizers; a small part (1 %) is utilized by plants and the remainder (99 %) is rapidly converted into insoluble complexes (Mehta *et al.*, 2014). Hence, the recovery efficiency of P is not more than 20% of applied P in the world soils (Qureshi *et al.*, 2012). Considering the low recovery of applied and native P and the high cost of chemical phosphatic fertilizers in addition to an increasing concern about environmental degradation (Aziz *et al.*, 2006; Khan *et al.*, 2014), it is important to find viable solutions to increase P fertilizer use efficiency. Two management options can be effective: (i) increasing the recovery and solubility of applied P fertilizers and (ii) replacing the expensive chemical P fertilizers with novel, cheaper, more ecologically friendly but nevertheless efficient P sources, such as indigenous rock phosphates (RPs). The application of RPs directly to the soils has yielded some positive results in acidic soils, but the efficacy of such material is almost negligible in neutral and alkaline soils (Begum *et al.*, 2004). Therefore, efforts have been made to find suitable ways to improve the solubility and efficiency of indigenous RPs. Numerous studies have been conducted to evaluate the efficiency of different amendments to increase the availability and solubility of P from native and applied sources including RP. Among these, organic amendments, including animal manure, plant residues and green manure (Alloush, 2003; Toor, 2009; Aria *et al.*, 2010; Adesanwo *et al.*, 2012), composts (Nishanth and Biswas, 2008; Wickramatilake *et al.*, 2010; Saleem *et al.*, 2013), and bacterial inoculation (Panhwar *et al.*, 2011; Gupta *et al.*, 2011) are considered beneficial for improving the P efficiency. In addition, the combined application of water-soluble P fertilizers with RP is another option to increase the efficiency of RP.

Soil microorganisms have generally been found effective in making P available to the plants from both inorganic and organic sources by solubilizing and mineralizing complex P compounds (Khan *et al.*, 2014). Similarly, the application of organic manure with phosphate fertilizers is considered another possible means of mobilizing P because of the acidic environment generated during the decomposition of

the manure (Nishanth and Biswas, 2008). The different types of organic manure increase the microorganisms, release acids in the root rhizosphere and may help to solubilize P and to increase P availability to the plants (Hu et al., 2006).

Keeping in view the considerable expense involved in importing raw material for manufacturing P fertilizers or in P fertilizers directly imported, it is imperative to explore the possibility of the utilization of indigenous RPs and the ways to increase the efficiency of other P fertilizers. The effects of biogas slurry (BS) on the efficiency of both soluble and insoluble P fertilizers with regard to plant growth and yield have been studied. However, the effect of these combinations on P release capacity (mineralization) of both soluble and insoluble P sources, especially RPs, has been given little attention. Therefore, the present study was conducted to examine the effect of biogas slurry (BS) with soluble P fertilizers (SP) and insoluble rock phosphate (RP) on P availability and their subsequent effect on the growth, yields, chemical composition of leaves and pods, also on biological activity of snap bean (*Phaseolus vulgaris* L.) grown in a greenhouse condition.

Materials and Methods

I-Materials:

1.1. Seeds:

Snap bean (*Phaseolus vulgaris* L.) varieties Xera and Paulista were kindly provided from Horticulture Research Institute, Agriculture Research Center (ARC), Giza, Egypt.

1.2. Soil:

A clay soil sample was collected from Moshtohor, Kalubia, Egypt. The soil was slightly alkaline. Soil was then air dried, pulverized and sieved to 2 mm diameter size to be prepared for snap bean. Some physical and chemical properties of the experimental soil are shown in Table (1) Mechanical and physical analyses were estimated according to Jackson (1973).

1.3. Biogas slurry and rock phosphate:

Chemical analysis of the rock phosphate (Table 2) and biogas slurry was estimated according to Black *et al.*, (1982). Chemical analysis of biogas slurry showed 2.5% N, 0.87% P, 1.5% K, 10% total solids, 58.2% O.M., 7.5pH and 90% moisture content.

1-4. Rhizobium:

Rhizobium phaseoli was supplied kindly provided by Bio fertilizers Production Unit, Soils, Water Environ. Res. Inst., Agric. Res. Center (ARC), Giza, Egypt.

Table 1. Some physical and chemical properties of the experimental soil

Soil characters	Value	Soil characters	Value	
Coarse sand	7.43	<i>Soluble cations (meq. L⁻¹):</i>		
Fine sand	16.77			
Silt	34.88		Ca ⁺⁺	1.28
Clay	40.92		Mg ⁺⁺	0.70
			Na ⁺	1.70
Textural class	Clay loam	K ⁺	0.40	
Organic matter content %	1.50	<i>Soluble anions (meq. L⁻¹):</i>		
E.C (dsm ⁻¹)	2.80		CO ₃ ⁻	0.00
T. N %	0.40		HCO ₃ ⁻	1.99
T. P%	1.20		Cl ⁻	1.42
pH (1: 2.5 ,soil suspension)	7.50	SO ₄ ⁻	0.67	

Table 2. chemical characteristics of rock phosphate used in study.

Chemical character	Value	Chemical character	Value	
Soluble ions (meqL ⁻¹)		Available macro and micronutrients (mg kg ⁻¹)		
	Ca ⁺⁺	8.5	N	32.32
	Mg ⁺⁺	3.75	P	0.38
	Na ⁺	16.9	K	40.5
	K ⁺	0.85	Fe	0.94
	CO ₃ ⁼	--	Zn	0.90
	H CO ₃ ⁻	1.9	Mn	0.38
	Cl ⁻	25.0	Total macro and micronutrients	
	SO ₄ ⁼	3.1	N (%)	0.17
pH (1:2.5)	7.43	P ₂ O ₅ (%)	12.0	
ECedS m ⁻¹	15.99	K ₂ O (%)	0.37	
SP%	28	Fe (mg kg ⁻¹)	2.93	
CEC (meq 100g ⁻¹)	4.27	Zn (mg kg ⁻¹)	106.10	
		Mn(mg kg ⁻¹)	285	

1-5. Fertilizers used:

The fertilizers used for snap bean were calcium super phosphate (15.5% P₂O₅) as a source of P, slurry of biogas as an organic manure, Ammonium sulphate as a source of N and potassium sulphate as a source of potassium. .

II-Methods:

A pot experiment was conducted under the green house conditions at Training Center for Recycling of Agricultural Residues (TCRAR) Moshtohor, Kalubia, to evaluate the efficiency of slurry of biogas with rock phosphate and super phosphate, at recommended dose, on the growth and yield of snap bean. Pots of 30 cm diameter were filled with 6Kg soil. Each pot was fertilized with biogas slurry (cattle dung) at rate of 10m³, 5m³ and 2.5m³ /fed (60ml, 30ml and 15ml / pot) and with calcium superphosphate (SP) or rock phosphate at recommended dose, 200Kg / fed (1.2gm of superphosphate or 1.5g of rock phosphate (RP) /pot. These fertilizers were applied with three doses. Recommended N and K at a rate of 60 Kg and 50 Kg / fed, respectively according to Ministry of Agriculture, were applied to all treatments, 200 Kg/fed of super phosphate was used for the control. Five seeds were sown in the pot at the depth 6 cm. The experiment was arranged in complete randomized design with 4replicates.

Experiment treatments:

The experiment included 7 treatments for each snap bean variety used

- 1) 10 m³ biogas slurry + 30Kg P₂O₅/ fed as rock phosphate.
- 2) 5 m³ biogas slurry + 30Kg P₂O₅/ fed as rock phosphate.
- 3) 2.5 m³ biogas slurry + 30Kg P₂O₅/ fed as rock phosphate.
- 4) 10 m³ biogas slurry + 30Kg P₂O₅/ fed as calcium super phosphate.
- 5) 5 m³ biogas slurry + 30Kg P₂O₅/ fed as calcium super phosphate.
- 6) 2.5 m³ biogas slurry + 30Kg P₂O₅/ fed as calcium super phosphate. 1)
- 7) Control (recommended dose of N, P and K of ministry of agriculture).

Chemical analysis of soil, biogas slurry and rock phosphate:

Chemical analysis for the soil samples (Table 1) were done as described by **Jackson (1973)**. Chemical analysis of the rock phosphate (Table 2) and biogas slurry was estimated according to Black *et al.*, (1982). **Fe, Mn, Cu and Zn were determined by using**

Atomic Absorption (model GBC 932), according to(Cotteniet al., 1961).

Growth and yield parameters:

Random samples of ten plants from each replicates were chosen after 45 days of sowing then some plant growth parameters were estimated, plant height (cm), No. of leaves, Plant dry wt. (g) and No. of branches/ plant. After 60 days (the end of experiment), Pods weight /plant (g), Pod length (cm) and Pod diameter (cm) were estimated.

Chemical analysis of Plant:

- Total nitrogen in the plant samples was determined using the method of microkjeldahel described by **Jackson (1973)**.
- Total phosphorus and available phosphorus determined according to **ApHA, (1989)**.
- Zn and Fe were determined in plant as described by **Jackson (1973)**.

Microbiological analysis:

After 45 days, nitrogenase enzyme (N₂-ase) activity of rhizosphere soil was assayed by the acetylene reduction assay (**Hardy et al., 1973**). Dehydrogenase (DHA) enzyme activity of rhizosphere soil was determined by the method described by **Thalman (1967)**. Total bacteria were estimated in the rhizosphere by plate count method (**Allan, 1959**).

Statistical analysis:

The obtained data were subjected to the proper statistical analysis for the complete randomized block design according to **Snedecor and Cochran (1980)**. Least significant difference (L.S.D.) at 5% level of significance was used for treatment means comparison. All statistical analysis was performed using analysis of variance technique by means of (M STAT) computer software package.

Results and Discussion

Vegetative growth and yield and its components:

Data in Table (3) showed the response of snap bean vegetative growth and yield components to biogas slurry (BS) 10, 5 and 2.5 m³ /fed with rock phosphate (RP) or super phosphate (SP) at 30Kg of P₂O₅ / fed. Vegetative growth i.e., plant height, No of leaves, plant dry weight and No of branches / plant for two used varieties resulted in significant differences as compared to the control. The highest values of plant height, plant dry weight and No of branches / plant were obtained by 2.5m³ Bio slurry + 30Kg P₂O₅/fed as rock phosphate for Xera cultivar while the highest No. of leaves /plant for Xera cultivar (19.3) was obtained using 5m³ biogas slurry + rock phosphate.

Table 3. Effect of biogas slurry with rock phosphate and super phosphate on vegetative growth and yield of snap bean

Treatments		Plant height (cm)	No. of leaves	Plant dry wt. (g)	No. of branches/plant	Pods wt./plant (g)	Pod length (cm)	Pod diameter (cm)	
Xera	10m ³ BS	30Kg P ₂ O ₅ /fed as RP	22.0 bc	16.7 b	2.10 c	4.1 bc	7.50 b*	10.2 abc	0.55
	5m ³ BS		25.7 ab	19.3 a	2.40 bc	4.6 b	7.00 c	9.9 bc	0.63
	2.5m ³ BS		27.3 a	16.0 b	2.95 a	5.8 a	9.15 a	11.9 a	0.64
	10m ³ BS	30kg P ₂ O ₅ /fed as SP	29.0 a	12.3 bc	2.75 ab	5.5 a	4.90 e	11.2 abc	0.72
	5m ³ BS		25.0 ab	14.7 bc	2.30 c	4.4 b	6.54 c	11.4 ab	0.68
	2.5m ³ BS		25.3 ab	9.3 d	1.50 d	3.6 c	6.65 c	11.0 abc	0.52
	Control	NPK	19.7 c	14.0 bc	1.70 d	3.7 c	5.55 d	9.5 c	0.50
	LSD (0.05)		3.52	2.62	0.33	0.43	0.48	1.22	N.S.
	Paulista	10m ³ BS	30Kg P ₂ O ₅ /fed as RP	18.7 d	11.7 c	1.80 d	4.80 b	3.40 c	10.0
5m ³ BS		19.3 d		14.3 b	2.10 c	4.90 b	6.40 a	10.4	0.55
2.5m ³ BS		24.0 c		17.0 a	2.45 b	5.65 a	3.80 c	12.2	0.54
10m ³ BS		30kg P ₂ O ₅ /fed as SP	32.0 a	17.3 a	2.75 a	5.70 a	4.30 b	10.9	0.60
5m ³ BS			24.0 c	13.0 bc	2.48 b	5.40 ab	4.50 b	11.7	0.64
2.5m ³ BS			28.0b	13.7 b	2.05 c	4.90 b	2.13 d	11.0	0.63
Control		NPK	18.7 d	13.0 bc	2.20 c	5.05 ab	2.17 d	10.1	0.52
LSD (0.05)		1.04	1.19	0.34	0.49	0.44	Ns	Ns	

RP: Rock phosphate

SP: Super phosphate

BS: Biogas slurry

*Values have the same letter are not significantly different.

For Pulista cultivar, all vegetative growth parameters were obtained their maximum values by 10m³BS+30Kg P₂O₅/fed as SP, where the highest No. of leaves /plant were 17.3 and 17.0 obtained by 10m³BS+ SP and using 2.5 m³ of biogas slurry + rock phosphate, respectively. Most of the lowest vegetative growth parameters for two used varieties were obtained by the control (full dose of NPK).

With respect to yield and its components, results indicated that both of Pods weight/plant and Pod length of Xera cultivar significantly increased by applying biogas slurry and used different phosphate forms. The highest values of Pods weight/plant and Pod length were 9.15 g and 11.9 cm, respectively by using 2.5m³BS + rock phosphate, while Pod diameter insignificantly increased over control. The Pods weight/plant of Paulista variety significantly increased when snap bean received 5m³BS + rock phosphate which presented 6.4 g followed by 4.5 g by applying 5m³BS+ SP. Both of Pod length and Pod diameter insignificantly increased in this variety as a result of using bio slurry and different forms of phosphate fertilizers used in this study. The control treatment caused the minimum values of yield and its components under this study..Sajjad and Shad, 2014 improved that combination of organic and inorganic fertilizers have produced higher yields of wheat. Also, Yield responses of vegetable crops to bio-slurry manure application have been reported in different crops including okra (Shahbaz, 2011), maize and cabbage (Karki, 2001). Krishna, 2001 declared that vegetable crops produced with bio-slurry have better quality as compared to those produced with chemical fertilizer. Gurung (1998) stated that the bio slurry contains nitrogen, phosphor and potassium as well as

zinc, iron, manganese and copper, the last of which has become a limited factor in many soils. So, it can be used to build healthy fertile soil for crop production. Also, Sterrett et al., 1988 showed that the addition of organic matter improved the physical, chemical and biological properties of soils and in turn improved the ability of the plant to absorb nutrients which led to increasing in crop production. Jeptoo et al., 2013 emphasize the effect of bio-slurry in carrot when they reported that higher plant heights and leaf numbers obtained could also be attributed to better levels of major nutrients (N, P, and K) in the bio-slurry manure, as reflected by higher nutrient levels in the growing medium. They added that these findings could be attributed to improved nutrient availability and better soil structure that could have favored shoot and root growth. Garg et al., (2005) reported that wheat grain yield increased with the application of biogas slurry generated from cattle dung, which was attributed to the lower bulk density of soil, increased hydraulic conductivity, and greater moisture retention. Consequently, the improved status of nutrients through amendment of the physical properties of soil contributed to the higher yield of wheat

Leaves chemical composition of snap bean:

Data presented in Table (4) show the values of NPK % and content of Fe, Mn, Zn and cu in snap bean leaves as affected by biogas slurry (BS) 10, 5 and 2.5 m³/fed with rock phosphate (RP) or super phosphate (SP) at 30Kg of P₂O₅ / fed. Results revealed that these factors led to significant increases in these parameters. Regarding macronutrients %, using of rock phosphate (30 Kg P₂O₅/fed) caused maximum increases in total N, P and K % in leaves of Xera cultivar when RP was

applied along with 10m³BS which recorded 4.7, 0.258 and 2.9 %, respectively. The highest values of N and K % (3.5 and 2.88 %, respectively) in leaves of Pulista cultivar were obtained by using 10m³BS + RP while

the maximum P% (0.330%) was obtained by 10m³BS + SP. The lowest values of these macronutrients under this study for two cultivars were caused by using the recommended dose of NPK fertilizer (control).

Table 4. Effect of biogas slurry with rock phosphate and super phosphate on leaves chemical composition of snap bean

Cultivar	Treatments	T.N	T.P	T.K	Fe	Mn	Zn	Cu	
		%		Ppm					
Xera	10m ³ BS	30Kg P ₂ O ₅ /fed as RP	4.700 a	0.258 a	2.90 a	390.0a	53.89a	36.89b	9.89ab
	5m ³ BS		4.330 a	0.257 a	2.02c	242.4b	50.33a	35.00bc	10.67a
	2.5m ³ BS		4.000 b	0.240 ab	2.14c	295.0 b	48.14 ab	30.70 c	8.07c
	10m ³ BS	30kg P ₂ O ₅ /fed as SP	3.540 c	0.197 c	2.83a	284.2 b	55.94a	39.40b	9.20b
	5m ³ BS		3.920 b	0.210 c	2.45b	146.3c	54.77 a	38.56b	9.25b
	2.5m ³ BS		4.490 a	0.230 b	2.60ab	253.8 b	42.40b	46.12a	10.56a
	Control	NPK	3.490 c	0.250 ab	2.75ab	237.0 b	49.30ab	30.27c	9.50 ab
	LSD (5%)		0.087	0.002	0.156	41.86	2.54	0.95	0.55
	Paulista	10m ³ BS	30Kg P ₂ O ₅ /fed as RP	3.50 a	0.252 b	2.88a	277.3ab	47.10cd	38.96ab
5m ³ BS		3.18 ab		0.260 b	2.78ab	237.0bc	46.90cd	35.86b	9.76 c
2.5m ³ BS		3.44 a		0.271 b	2.65bc	345.0a	54.39a	24.50c	11.80b
10m ³ BS		30kg P ₂ O ₅ /fed as SP	2.75 b	0.330 a	2.55cd	300.0ab	50.37bc	40.50 a	11.50 b
5m ³ BS			3.00 ab	0.262 b	2.40de	198.0 c	43.70d	2.440c	13.16a
2.5m ³ BS			3.15 ab	0.250 b	2.30e	320.0a	51.77ab	24.02c	10.39c
Control		NPK	2.73 b	0.245 b	2.70 abc	280.0 ab	48.60 bc	28.70 c	8.33 d
LSD (5%)		0.095	.009	.034	50.84	1.64	1.30	0.44	

RP: Rock phosphate

SP: Super phosphate

BS: Biogas slurry

*Values have the same letter are not significantly different.

With respect to Fe, Mn, Zn and Cu in leaves, data indicated that different P source fertilizers with biogas slurry (BS) caused significant increases in these micronutrients. For Xera cultivar, Fe content in leaves reached to the maximum contents by 10m³BS + RP. Both of Mn and Cu were responded to RP or to SP where Mn content represented the highest value, 55.94 ppm by using 10m³BS + SP and represented 53.89 ppm by 10m³BS + RP without significant differences. Also, Cu content in leaves recorded 10.67 and 10.56 ppm when seeds received 5m³BS + RP and 2.5 m³BS + SP, respectively. Zn content represented the maximum value, 46.12 ppm by 2.5 m³BS + SP followed by 39.40 ppm by applying 10 m³BS + SP.

Regarding Pulista cultivar, using SP led to significant increase in Zn and Cu which recorded 40.50 and 13.16 ppm by 10 m³BS and 5 m³BS, respectively. Applying 2.5 m³BS with RP gave the highest content of Fe, 345.0 ppm followed by 320.0 ppm by using 2.5 m³BS + SP. The maximum Mn content was obtained by 2.5 m³BS + RP.

The increase of nutrient uptake by snap bean in treatment fertilized by organic manure can be attributed to their role in plant growth as a source of all necessary macro and micronutrients in available forms. These results are in accordance with those reported by Abou-zeid and Bakry (2011).

Pod chemical composition of snap bean:

The influence of biogas slurry and various sources of phosphate fertilizers on chemical composition of snap bean pod is shown in Table (5). Data showed

significant increases in N, P, K%, total protein and some micronutrients, Fe, Mn, Zn and Cu in pod as a result of the above mentioned treatments for Xera cultivar. The maximum values of N, K and total protein % were obtained by using calcium superphosphate (SP), where applying 10m³ of bio slurry (BS) gave 3.98, 3.80 and 24.88 % of N, K and total protein % respectively. Using rock phosphate (RP) with 10 m³bioslurry led to the highest value (0.479%) of P%. Regarding some micronutrients, results from the Table showed that Fe and Zn reached the maximum values, 444 and 50.7 ppm when snap bean fertilized with RP +10 m³. 102.9 and 101.75 ppm were the highest values of Mn when applying 10 m³ of BS with RP and SP, respectively. Cu was more effective with SP where it gave 19.6 and 17.5 ppm with using 10 m³ and 2.5 m³bioslurry, respectively.

For Paulista cultivar, data in Table (5) indicated that N, P, total protein, Fe and Mn significantly increased over control as a result of BS and RP or SP. In more details, both of N% and total protein gave the highest percentage, 4.23 and 36.44% by using 10m³ and RP. 10 m³ of biogas slurry with SP led to the maximum value of P% (0.553%) followed by 0.547% by applying 5m³ of BS + SP.

With regard to Fe and Mn concentration (ppm), results revealed that 10m³ of BS + RP led to the maximum percentage of Fe while, the same rate of BS + SP gave the highest Mn (97.4 ppm). K%, Zn and Cu ppm insignificantly increased by using BS and RP or SP.

Table 5. Effect of biogas slurry with rock phosphate and super phosphate on pod chemical composition of snap bean

Treatments	T.N		T.P		T.K		T.protein		Fe		Mn		Zn		Cu	
	%															
ppm																
Xera	10m ³ BS	30Kg	3.75 ab	0.479 a	2.59 c	23.44 bc	444 a	102.90 a	50.70 a	15.7 ab						
	5m ³ BS	P ₂ O ₅ /fed	3.01 c	0.458 a	2.89 bc	18.80 e	424 a	75.49 c	41.24 ab	8.76 c						
	2.5m ³ BS	as RP	3.87 ab	0.474 a	2.03 d	24.19 ab	211 d	49.90 d	29.47b	13.39 abc						
	10m ³ BS	30kG	3.98 a	0.320 c	3.80 a	24.88 a	269 c	101.75 a	24.50 b	19.60 a						
	5m ³ BS	P ₂ O ₅ /fed	3.60 ab	0.382 b	3.55 a	22.50 c	346 b	90.75 ab	23.00 b	14.75 abc						
	2.5m ³ BS	as SP	2.75 c	0.391 b	2.52 c	17.19 f	137 e	100.20 a	28.40 b	17.50 a						
	Control	NPK	3.40 b	0.388 b	3.02 b	21.00 d	360 b	82.70 bc	42.00 ab	10.15 bc						
LSD (5%)		0.38	0.03	0.30	1.01	55.70	11.58	14.93	4.61							
Paulista	10m ³ BS	30Kg	4.23 a	0.444 ab	3.40	36.44 a	473 a	74.00 c	37.00	15.57						
	5m ³ BS	P ₂ O ₅ /fed	3.85 ab	0.367 b	2.80	24.06 b	328 c	41.30 e	38.50	12.25						
	2.5m ³ BS	as RP	3.90 ab	0.390 b	3.08	24.38 b	459 a	85.80 b	39.17	12.50						
	10m ³ BS	30kG	2.90 b	0.553 a	3.38	18.13 c	363 bc	97.40 a	42.17	11.90						
	5m ³ BS	P ₂ O ₅ /fed	4.00 a	0.547 a	3.24	25.00 bc	240 d	52.27 de	42.80	8.90						
	2.5m ³ BS	as SP	3.38 ab	0.400 b	2.77	21.13 b	193 d	42.50 e	47.60	11.16						
	Control	NPK	3.50 ab	0.464 ab	2.90	22.00	424 ab	61.00d	40.50	12.68						
LSD (5%)		0.72	0.08	N.S	3.10	61.17	9.27	ns.	Ns							

RP: Rock phosphate

SP: Super phosphate

BS: Biogas slurry

*Values have the same letter are not significantly different.

Somasundaramet *al.*, (2007) reported that increased soluble protein content of maize, sunflower, and green gram (*Vignaradiata L.*) was estimated with biogas slurry. These increases as a result of applying biogas slurry, which are in accordance with those obtained by Mahdy, 2003 who stated that organic manure attains more pronounced contents of macro-(N, P and K) and micronutrients (Fe, Mn, Zn and Cu), which was consequently reflected on plant growth and yield. Also Fentaw (2010) stated that Compost and bio slurry improves the physical, chemical and biological quality of soil besides providing both macro and micro nutrients to crops. The improvement in qualities include improvement in soil structure, water holding capacity, electrical conductivity, bulk density, lesser soil erosion, preventing the leaching of nutrients and provide nutrients to soil micro flora.

Biological activity and available phosphorus:

The response of biological activity parameters i. e., dehydrogenase and nitrogenase activity, total count of bacteria, also available phosphorus to biogas slurry (BS) with various sources of phosphorus fertilizers is shown in Table (6). All the previous parameters are significantly increased as a result of using BS and super phosphate (SP) or rock phosphate (RP) for the two used varieties. Data revealed that these biological activity parameters increased concomitantly with the increase of biogas slurry (BS) doses for the two used varieties with SP or RP. Using of RP as a phosphate fertilizer led to more increase of the values in these parameters than SP. Control treatment (the recommended dose of N, P and K) gave the lowest values of above mentioned parameters. The highest

DHA, total bacteria and N₂-ase activity were 201.5 µg TPF/g rhizosphere soil/day, 17.2 X 10⁶ and 44.5 nmole C₂H₄/ g rhizosphere soil/h of the plants received 10m³BS + 30kg P₂O₅/fed as RP for Xera cultivar, while the corresponding values were 212.8 µg TPF, 28.8 X 10⁶ and 86.2 nmole C₂H₄ for the other variety with the same treatment. These results are in agreement with those by Abou-zeid and Bakry (2011) who reported that the addition of organic manures to the soil encourage proliferation of soil microorganisms which consequently increased the values of dehydrogenase enzyme activity. This may be due to the vital role of organic materials in enriching community of soil microorganisms of the rhizosphere of snap bean.

With respect to available P, results indicated that there are significant increases in available P as a result of BS and SP or RP. The fertilization with SP gave the maximum values of available P for Xera cultivar, which recorded 24.64 ppm by 10m³ BS + SP followed by 23.11 ppm by 5 m³ BS +SP then 20.52 ppm with the control while the minimum value, 5.82 ppm was presented by using 2.5 m³ BS + SP. Regarding Paulista cultivar, 10m³ BS + RP gave the highest value of available P, 34.31 ppm followed by 21.51 ppm and 20.06 ppm by applying 5 m³ BS + SP and 2.5 m³ BS + SP, respectively. Somasundaramet *al.*, (2007) reported that increased nitrogenase activity of maize, sunflower, and green gram (*Vignaradiata L.*) was estimated with biogas slurry. Hu et al., (2006) added that The different types of organic manure increase the microorganisms, release acids in the root rhizosphere and may help to solubilize P and to increase P availability to the plants.

Table 6. Effect of biogas slurry with rock phosphate or super phosphate on dehydrogenase and nitrogenase activity, total count of bacteria and available phosphor of snap bean soil

Treatments	DHA ($\mu\text{g TPF/g rhizosphere}$ soil/day)	Total bacteria $\times 10^6$	$\text{N}_2\text{-ase}$ ($\text{nmole C}_2\text{H}_4\text{/g}$ rhizosphere soil/h)	Available P (ppm)	DHA ($\mu\text{g TPF/g rhizosphere}$ soil/day)	Total bacteria $\times 10^6$	$\text{N}_2\text{-ase}$ ($\text{nmole C}_2\text{H}_4\text{/g}$ rhizosphere soil/h)	Available P (ppm)	
									Xera
10m ³ BS	30Kg P ₂ O ₅ /fed as RP	201.5 a	17.2 a	44.5 a	11.43 c	212.8 a	28.8 a	86.2 a	34.69 c
5m ³ BS		142.4 bc	14.5 b	21.7 cd	9.14 d	111.6 c	25.2 ab	72.5 a	10.41 d
2.5m ³ BS		104.2 cd	10.3 c	15.4	9.08 d	101.7 cd	11.2 c	51.1 b	14.31 a
10m ³ BS	30kg P ₂ O ₅ /fed as SP	172.8 ab	15.1 b	31.3 b	24.11 a	151.4 b	21.1 b	33.4 c	21.64 c
5m ³ BS		125.6 c	9.2 cd	25.1 c	23.64 a	110.8 c	20.5 b	15.9 d	20.51 b
2.5m ³ BS		81.3 d	8.1 de	19.0 de	5.82 e	80.0 d	9.5 c	12.4 d	16.06 b
Control	NPK	71.2 d	7.0 e	12.2 f	20.52 b	49.2 e	12.5 c	18.1 cd	15.99 c
LSD (5%)		39.16	1.16	4.39	1.89	26.53	5.92	15.61	2.65

RP: Rock phosphate

SP: Super phosphate

BS: Biogas slurry

*Values have the same letter are not significantly different.

Conclusion

Amending soils with bio-slurry manure enhances the vegetative growth, pods yield of snap bean. This study ensured that the use of rock phosphate with biogas slurry may be recommended as a substitute for super phosphate with biogas slurry in snap bean production.

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استجابة الفاصوليا لسماذ البيوجاز السائل مخلوط مع سمدى صخر الفوسفات أو سوپر الفوسفات

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تشمل هذه الدراسة تأثير سماذ البيوجاز السائل (BS) تحت ظروف الصوبة على نمو ونتاج الفاصوليا والتركيب الكيميائى وبعض انزيمات التربة مثل انزيم النتروجينيز والديهيدروجينيز وكذلك الفوسفورالميسرفى التربة ولقد سممت الفاصوليا بأكثر من مصدر للتسميد الفوسفاتى مثل كالمسيوم سوپر فوسفات او صخر الفوسفات بمعدل 30كجم خامس اكسيد الفسفور للفدان (P_2O_5) مع BS واوضحت النتائج ان القياسات الخضرية قد زادت عند استخدام الBS سواء مع صخرالفوسفات او سوپرالفوسفات بالنسبة لكلا الصنفين من الفاصوليا وان اضافة $10\text{م}^3\text{BS}+30\text{كجم } P_2O_5$ من سوپر الفوسفات اعطت اعلى النتائج بالنسبه لصنف بوليسنا بلاضافة لذلك صنف اكزيرا المعاملة $10\text{م}^3\text{BS}+30\text{كجم } P_2O_5$ للفدان اعطت اعلى قياس لطول النبات بينما $2.5\text{م}^3+30\text{كجم } P_2O_5$ للفدان م صخرالفوسفات اعطت اعلى قيم بالنسبة للوزن الجاف للنبات وعدد الافرع بلاضافة للنتاج وقياساته مثل وزن القرون /نبات وطول القرن وذلك مقارنة بالكتنرول وكذلك وزن القرون/نبات بالنسبة لكلا الصنفين المنزرعين زادت وذلك مقارنة بالكتنرول وهذه النتائج فى حالة استخدام BSوكلا المصدرين من التسميد الفوسفاتى اما بالنسبة للتحليل الكميائى للارواق فان استخدام الBS سواء مع صخرالفوسفات او سوپرالفوسفات سببت زيادة معنوية فى محتوى الارواق من العناصر الكبرى مثل الNPK بينما المعاملة $10\text{م}^3\text{BS}+30\text{كجم } P_2O_5$ للفدان من صخر الفوسفات اعطت اعلى القياسات لكلا الصنفين والعناصر الصغرى مثل الحديد والزنك والمنجنيز والنحاس زادت زيادة معنوية مقارنة بالكتنرول ولقد وجد ان اضافة صخر الفوسفات مع الBS كانت اكثر تأثيرا بالنسبة لمعظم العناصر مقارنة بالسوبرفوسفات + الBS بالنسبة للصنفين المنزرعين اما بالنسبة للتحليل الكيماوى للقرون فان تركيز عنصرى الN,K والبروتين الكلى زادت زيادة معنوية بالنسبة لصنف اكزيرا حيث اعطى اعلى مستويات باستخدام سوپر فوسفات $10\text{م}^3\text{BS}$ بيما اعلى تركيز فى الفوسفور الكلى تم الحصول عليه بالنسبة لمعاملة صخر الفوسفات $10\text{م}^3\text{BS}$ وكذلك تركيز الزنك اما تركيز الحديد والمنجنيز والنحاس كانت اعلى تركيز مع المعاملة صخر فوسفات $10\text{م}^3\text{BS}$ بالنسبة للصنف بوليسنا اما النتروجين الكلى والبرتين الكلى والحديد سجلت اعلى معاملة صخر فوسفات $10\text{م}^3\text{BS}$ ونسبة المجنيز اعطت اعلى المسويات مع سوپر فوسفات $10\text{م}^3\text{BS}$ اما بالنسبة للنشاط الحيوى والفوسفور الميسر فقد اوضحت النتائج زيادة لكل المعاملات ولقد وضح ان اضافة صخر فوسفات $10\text{م}^3\text{BS}$ اعطت الى قياسات بالنسبة للانزيم الديهيدروجينيز و النتروجينيز والعدد الكلى للبكتيريا بالمقارنة مع سوپر فوسفات $10\text{م}^3\text{BS}$ وكانت احسن نتيجة تم الحصول عليها بالنسبة للفوسفور الميسر مع المعاملة سوپر فوسفات $10\text{م}^3\text{BS}$ وهكذا تبين هذه الداسة ان استخدام صخر فوسفات مع BS يمكن التوصيه بانها مناسبة كبديل للسوبرفوسفات لانتاج الفاصوليا.