

PRODUCTIVITY OF GINGER (*ZINGIBER OFFICINALE*) BY AMENDMENT OF VERMICOMPOST AND BIOGAS SLURRY IN SALINE SOILS

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Abstract

Ginger is a crop of economical importance. Though its productivity is generally reduced under saline conditions, it could still be improved by various physiological treatments which offset toxicity of excessive sodium of rooting medium. Amendments of Vermicompost and Biogas slurry have shown some reduction of Sodium induced inhibitory effects. Analyses of leaves chlorophyll, carbohydrate, protein etc., have been undertaken with reference to various above-mentioned treatments. Vermicompost amendments improved net yield, fresh and dry biomass of shoot and rhizome yield of plants. Their chlorophyll, carbohydrate and protein contents were increased under Vermicompost amendment both in the saline and non-saline conditions. Keeping in mind economical feasibility, one can still obtain permissible economic returns by this method from moderately saline soil, which was so far considered not suitable for ginger production.

Introduction

Ginger (*Zingiber officinale* Roscoe) of the family Zingiberaceae is indigenous to warm tropical climates, particularly southeastern Asia and is propagated through rhizome. About half of its world's production comes from India, whereas, its cultivation is extended to China, Australia, Malaysia, Nigeria, Fiji, Brazil and Mexico (Sanderson *et al.*, 2002). It is an important spice used as a condiment in vegetable preparations. It is also an important ingredient in herbal medicines for the treatment of rheumatism, nervous diseases, gingivitis, toothache, asthma, stroke, constipation and diabetes (Awang, 1992; Wang & Wang, 2005; Tapsell *et al.*, 2006). The oil and oleoresins obtained from ginger are also used in many food items, soft drinks and beverages (Singh *et al.*, 2008).

Ginger is basically considered salt sensitive crop and its productivity is generally reduced under saline conditions. It could be improved by various physiological treatments, which offset toxicity of excessive sodium of rooting medium. Since now a days with the recent advancement in science, it has become possible to increase agricultural productivity and extend threshold values of salt tolerance by different cultivation practices.

In recent years, Vermicompost and Biogas slurry are used as organic fertilizers to increase the productivity of crops. Vermicomposts are products of organic matter degradation through interactions between earthworms and microorganisms. Addition of different Vermicomposts, produced from different sources, like cattle manure, food and agricultural waste etc., increases the rate of germination and growth, and yield of many high value crops (Atiyeh *et al.*, 2000). Vermicompost contains plant-growth promoters (i.e., auxins, gibberellins, cytokinins) and also represents an excellent soil conditioners due to its higher porosity, aeration, drainage, water-holding capacity and microbial activity (Singh *et al.*, 2008). Like Vermicompost, Biogas slurry is also a good supplement of nutrients. It is a by-product obtained from the Biogas plant after the digestion of dung or other biomass for the generation of methane rich gas.

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Due to the increasing consumption of ginger in medicines and in food ingredients it was considered important to assess the effects of Vermicompost and Biogas slurry on the growth and yields of ginger under saline conditions, which are existing either due to soil salinity or as a result of saline water irrigation in the country.

Material and Methods

Plant material and growth conditions: Healthy rhizomes of ginger (*Zingiber officinale* Roscoe) were taken from local market and sown in earthen pots filled with sandy loam soil, having basal holes for leaching irrigation water.

Experiment 1: The threshold value for growing ginger was not found in literature, though, it is considered as salt sensitive plant. This experiment was conducted to determine the extent of its salt tolerance. Ginger rhizome weighing 15 g approximately, having at least one well-differentiated bud was sown in earthen pot containing about 5 kg sandy loam soil mixed with cow dung (9:1). Pots were irrigated with non-saline control to 0.7% sea salt solution ($EC_{iw}^1 = 0.7 - 10.2 \text{ dS m}^{-1}$, $EC_e^2 = 1.2 - 11.2 \text{ dS m}^{-1}$) keeping leaching percentage about 40%. Since the plants started degenerating at higher levels of saline water irrigation, they were harvested after four months.

Experiment 2: Healthy rhizome weighing about 30-35 g having at least two well differentiated buds were sown at a depth of about four inches in earthen pots containing 20 kg sandy loam soil mixed with cow dung (9:1). Treatments of soil amendments are described below:

1. Control, without any soil amendments (C)
2. Upper layer of soil around rhizome covered with Vermicompost (VC)
3. Biogas slurry poured on rhizome embedded in soil (BS)
4. Both the above-mentioned treatments (2 and 3) were given to rhizome embedded in soil (VC+BS).

Amendments of 1/2 kg Vermicompost or 500 ml of Biogas slurry or both were given thrice in experiment. The doses of soil amendments were given at sowing, after 3 months of growth and two months prior to harvest (after 7 months of growth). The above mentioned doses were given in two sets, without salinity and with salinity ($EC_{iw} = 2.9 \text{ dS m}^{-1}$ or 0.2% sea salt dilution). Five replicates were kept for each treatment. Plants were harvested after 9 months of sowing for determining growth and yield of rhizome.

Noticing the results of experiment 1 (Table 1), growth of ginger start decreasing at EC values higher than that of non-saline control and at onward increase up to 0.2% sea salt irrigation ($EC_{iw} = 2.9 \text{ dS m}^{-1}$, $EC_e = 3.6 \text{ dS m}^{-1}$), the reduction in shoot biomass becomes 43.1% and that of rhizome 13.03%. Hence, further work for improving growth in experiment 2 through application of Vermicompost and Biogas slurry was undertaken at $EC_{iw} 2.9 \text{ dS m}^{-1}$.

Vermicompost and biogas slurry: Vermicompost and Biogas slurry was provided by Agrotool (Pvt) Ltd. Sohana farm, Karachi. The compost was prepared in rack type earthworm bin made of perforated cement slabs filled with a mixture of moist *Leucaena leucocephala* leaves compost and cow dung. Biogas slurry was a by-product of Biogas production from well rolled cow dung by conventional methods. There analysis is presented in Table 2.

¹Electrical conductivity of irrigation water, ²Electrical conductivity of soil

Table 1. Growth of Ginger (*Zingiber officinale*) affected by saline water irrigation.

Electrical conductivity of irrigation water and soil (EC=dS m ⁻¹)	Fresh shoot biomass (at harvest) (g)	% Reduction of shoot biomass	Fresh weight of rhizome (at harvest) (g)	%Reduction of rhizome
Control (EC _{iw} =0.7, EC _e =1.2)	10.96 ± 0.62	-	35.26 ± 0.94	-
0.1% Sea salt (EC _{iw} =1.8, EC _e =2.4)	10.85 ± 0.78	1.0	31.56 ± 0.61	10.76
0.2% Sea salt (EC _{iw} =2.9, EC _e =3.1)	6.24 ± 0.26	43.1	30.7 ± 0.85	13.03
0.3% Sea salt (EC _{iw} =4.1, EC _e =4.8)	4.1 ± 0.25	62.6	28.1 ± 0.47	20.39
0.4% Sea salt (EC _{iw} =6.1, EC _e =6.8)	2.10 ± 0.29	80.8	24.63 ± 1.22	30.31
0.5% Sea salt (EC _{iw} =7.6, EC _e =8.3)	2.0 ± 0.32	81.8	22.6 ± 0.92	35.97
0.6% Sea salt (EC _{iw} =9.8, EC _e =10.6)	2.20 ± 0.3	79.9	19.3 ± 0.7	45.32
0.7% Sea salt (EC _{iw} =10.2, EC _e =11.2)	1.96 ± 0.33	82.1	18.2 ± 0.58	48.44

Table 2. Chemical properties of Vermicompost and Biogas Slurry on oven dry basis.

Chemical properties	Vermicompost	Biogas slurry
EC (dS m ⁻¹)	1.8	6.7
pH	6.91	8.36
N (%)	1.8	1.6
P (%)	0.58	0.65
K (%)	0.71	0.60
Na (%)	0.09	0.23

Growth analysis and yield: The experiment 2 was continued for nine months, the shoot length, number of shoots, fresh and dry weight of aerial biomass and fresh and dry weight of rhizome were recorded after harvesting the plants.

Biochemical analysis: The chlorophyll contents were recorded in 80% cold acetone according to the method described by MacLachlan & Zalik (1963). Soluble carbohydrates were determined in hot water extract of leaves by Anthron's reagent method (Yemm & Willis, 1954). Soluble proteins were extracted in fresh leaves by using 0.1 M Potassium phosphate buffer (pH, 7.0) and recorded by the method of Bradford (1976) against Bovine serum albumin (BSA) standards. Nitrogen contents of rhizome were determined by Kjeldahal's nitrogen method describe by AOAC official method 955.04 (AOAC, 2005).

Mineral analysis: The estimation of Na⁺ and K⁺ ions in leaves were determined as described by Ahmad & Jabeen (2009). Dry leaves (1 g) were ashed at 550°C for 6 h and then it was digested in 2M HCl. The estimation of minerals was done by using flame photometer (PetraCourt PFP I).

Statistical analysis: Data sets were subjected to two-way analysis of variance (ANOVA). Fisher's least significant difference tests (LSD) and standard error of means were also performed.

Results and Discussion

The effects of amending Vermicompost and Biogas slurry at soil on different growth parameters in the plants irrigated with irrigation water of 0.2% sea salt ($EC_{iw} = 2.9 \text{ dS m}^{-1}$, $EC_e = 3.6 \text{ dS m}^{-1}$) is presented in Fig. 1. It was confirmed at harvest that only two shoots arise from seed rhizome and rest of the shoots arise from newly formed rhizome. The seed rhizome was rather disintegrated darker in colour and the new rhizome was comparatively turgid and lighter colour. Supplement of above mentioned organic fertilizers have shown promotion in growth vigor under non-saline condition as well which persisted even after encountering toxicity due to saline water irrigation at various growth parameters. Inhibitory effect of saline water irrigation on number of shoots and their height (Fig. 1A and 1B), fresh and dry biomass of shoot (Fig. 1C and 1D) and fresh and dry weight of rhizome (Fig. 1E and 1F) appears to be removed under amendment of Vermicompost up to greater extent and with mixture of Biogas slurry up to lesser extent, whereas amendment of only Biogas slurry showed growth promotive effects only on rhizome production. Dry biomass of shoot display little increase under saline condition over non-saline control only under Vermicompost and Biogas slurry amendment. Growth inhibition due to salinity of root zone is consequence of several physiological responses including modifications of ion balance, photosynthetic efficiency, carbon allocation and utilization (Ahmad & Jabeen, 2005, Munns & Tester, 2008). The salt sensitive plants have poor ability to exclude salts. These salts accumulate in transpiring leaves to excessive levels, exceeding the ability of the cells to compartmentalize these ions in the vacuole. Ions then build up rapidly in the cytoplasm and inhibit enzyme activity or they build in the cell walls and dehydrate the cell (Munns, 2002; Tester & Davenport, 2003).

The amount of chlorophyll in leaves was significantly increased ($p < 0.001$) only in plants under non-saline condition provided by Vermicompost as fertilizer, its concentration was reduced under saline water irrigation and remained more or less equal even in those plants provided with organic fertilizers (Fig. 2A). The decrease in chlorophyll contents in leaves under salt stress is also reported by others (LeDily *et al.*, 1991; Kahn, 2003). This depletion of chlorophyll content in leaves may be due to a decrease in chlorophyll synthesis or to an increase in chlorophyll degradation (Santos, 2004). Abdelkader *et al.*, (2007) also reported that salt stress inhibits chlorophyll accumulation partly by reducing the rate of porphyrin formation and also by a possible reduction in the formation of chlorophyll-binding proteins.

There appears some increase in carbohydrate content of leaves in case of only Vermicompost amendment and its mixture with Biogas slurry at non-saline soil, but reduction is evident under saline water irrigation over their non-saline control even under amendment of above mentioned organic fertilizers. However, its amount still remained more than others under Vermicompost amendment even under saline condition (Fig. 2B). Carbohydrates accumulation under salt stress is reported in number of plants (Parida *et al.*, 2002), but on the other hand a decrease has been observed by some workers (Alamgir & Ali, 1999; Gadallah, 1999). This difference could be due to different salinity regimes and the range of salt tolerance of experimental plants. Since ginger is considered salt sensitive plant the amount of carbohydrates was decreased in leaves under saline soils but improvement is shown only under the amendments of Vermicompost only.

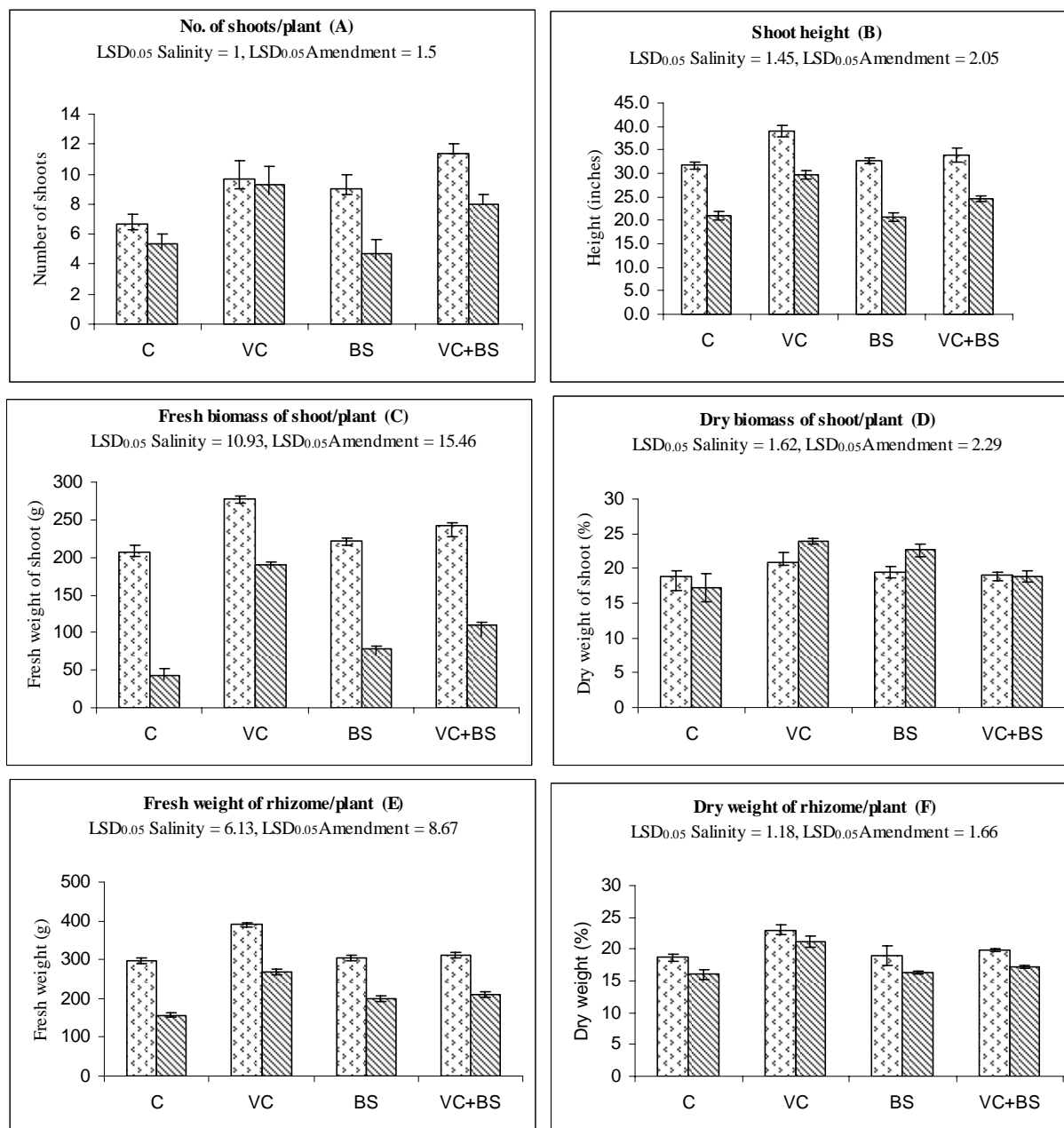
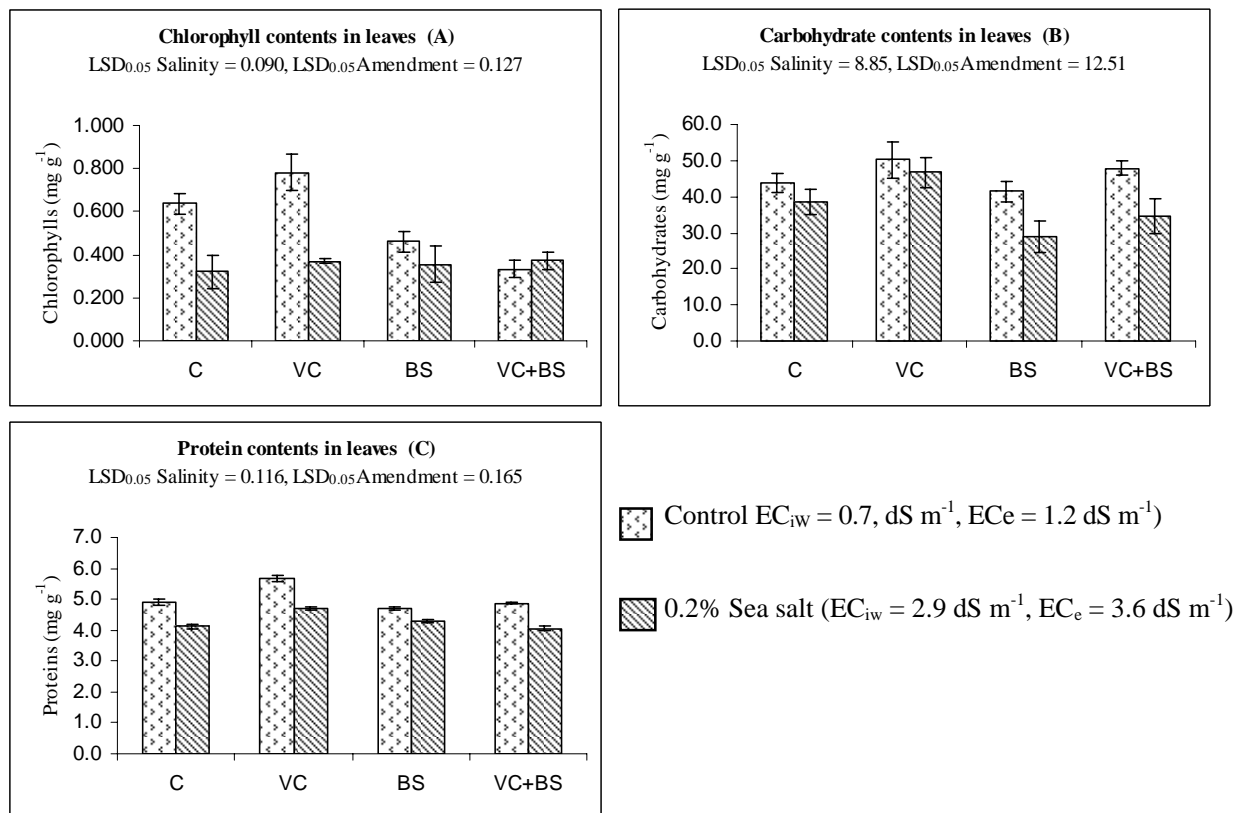


Fig. 1. Effects of amending Vermicompost and Biogas slurry in soil on different growth parameters and yield of Ginger (A – F) irrigated with 0.2% sea salt dilution ($EC_{iw} = 2.9 \text{ dS m}^{-1}$, $EC_e = 3.6 \text{ dS m}^{-1}$). C=Non-saline control, VC=Vermicompost, BS=Biogas slurry, VC+BS=Vermicompost + Biogas slurry

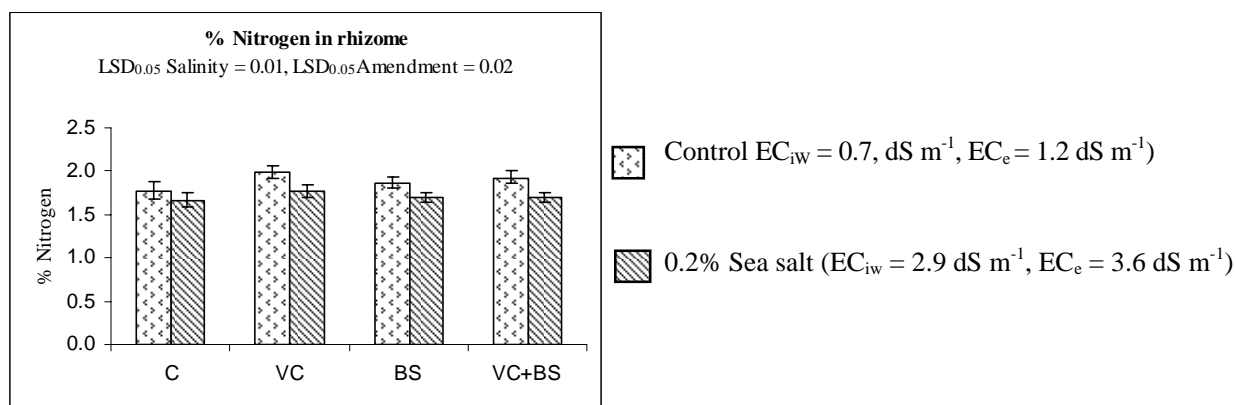
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The soluble proteins of leaves show some increase ($p < 0.001$) only under Vermicompost amendment both under non-saline and saline conditions. The application of Biogas slurry only and its mixture do not show any improvement in protein content (Fig. 2C). Decrease in soluble protein of leaves has been reported in many plants under salt stress irrespective of their salt tolerance (Ashraf & Waheed, 1993; Parida & Das, 2005). On other hand higher content of soluble proteins has been reported in salt tolerant cultivars of barley, sunflower, rice and finger millet under salt stress condition (Amini & Ehsanpour, 2005). These findings suggest that reduction in soluble proteins of ginger is due to their sensitivity to salt stress.



C=Non-saline control, V=Vermicompost, BS=Biogas slurry, VC+BS=Vermicompost + Biogas slurry

Fig. 2. Effects of amending Vermicompost and Biogas slurry in soil on chlorophyll (A), carbohydrates (B) and soluble protein contents (C) of Ginger leaves irrigated with 0.2% sea salt dilution ($EC_{iw} = 2.9 \text{ dS m}^{-1}$, $EC_e = 3.6 \text{ dS m}^{-1}$).



C=Non-saline control, VC=Vermicompost, BS=Biogas slurry, VC+BS=Vermicompost + Biogas slurry

Fig. 3. Effects of amending Vermicompost and Biogas slurry in soil on nitrogen of Ginger rhizome irrigated with 0.2% sea salt dilution ($EC_{iw} = 2.9 \text{ dS m}^{-1}$, $EC_e = 3.6 \text{ dS m}^{-1}$).

Concentration of nitrogen in rhizome shows slight increase under application of above mentioned fertilizers under non-saline conditions but remained almost equal to its control under saline water irrigation irrespective of any organic amendment (Fig. 3).

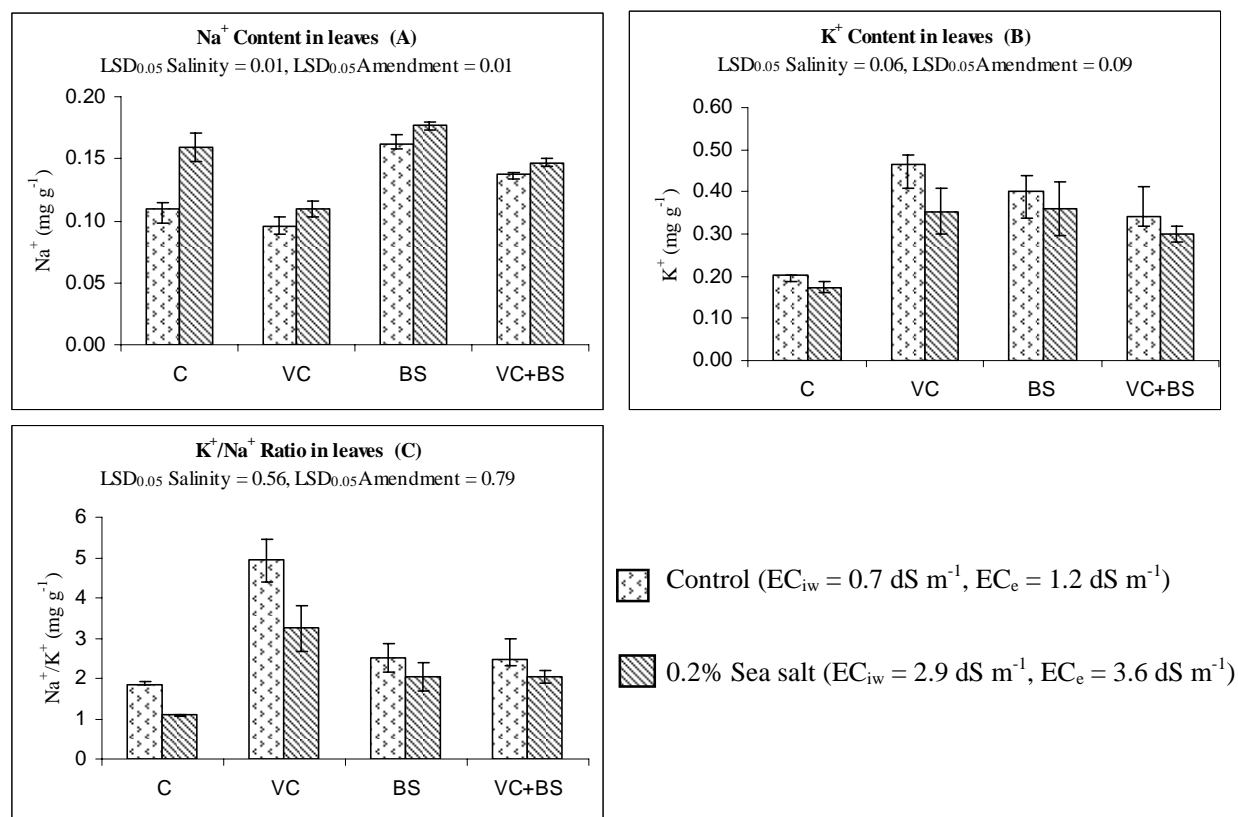
Higher amount of Na^+ is evident in the leaves of plants raised under saline water irrigation over their non-saline respective controls especially in those plants provided above mentioned fertilizers. However, its accumulation was least under application of

Vermicompost (Fig. 4A). The Na^+ accumulation in leaves under application of Biogas slurry both under non-saline and saline water irrigation could be due to high EC of salt in Biogas slurry itself. It may be noted that seepage of salt from the saline soil present around the well of Biogas slurry could have increased its EC (Table 2) which provided additional salts to plants. Hence due care should be taken for avoiding the presence of salt in Biogas slurry preparation. Potassium content of the leaves was decreased under saline water irrigation in comparison with its non-saline respective control (Fig. 4B) most probably due to antagonistic attitude of Na^+ against K^+ ions (Ahmad & Jabeen, 2005; Tester & Davenport, 2003). Amendment of both the above mentioned organic fertilizers helped in increasing potassium content of leaves irrespective of non-saline and saline water irrigation. Hence the K^+/Na^+ ratio was also affected (Fig. 4C). During salt stress the decrease in K^+ uptake and an increase in Na^+ influx has been reported by other workers as well (Serrano & Rodriguez-Navarro, 2001). Many plants sensitive to excessive Na^+ show adverse effects on K^+ demanding intermediary metabolism specially that of cytosolic enzyme and photosynthesis (BenKhalid *et al.*, 2003; Munns & Tester, 2008). The selective uptake of K^+ as opposed to Na^+ is considered to be one of the important physiological mechanisms contributing to salt tolerance in many plant species (Yamaguchi & Blumwald, 2005). Salt tolerant plants showed that higher K^+/Na^+ ratio is a criteria for salt tolerance (Glenn, *et al.*, 1999). This is also confirmed with the findings of Ashraf & O'Leary (1996) who found the higher salt tolerance of seven wheat genotypes was related to high K^+ and lower Na^+ accumulation in their leaves. Present findings suggest that ginger become tolerant to salt stress by the addition of above mentioned fertilizers due to making K^+ available to plants for betterment of its growth.

The improvement in growth by the addition of Vermicompost is reported due to availability of plant growth regulators and humic acid from Vermicompost (Arancon *et al.*, 2004). It is stated that earthworms by their activity produce humus or humus like substances which may promote nutrient uptake, internal metabolism and influence protein synthesis (Bachman & Metzger, 2008). Vermicompost also show hormone like activity due to generating auxin, gibberiline and cytokinin like compounds which altered morphological and physiological characteristics of plants. Beside, many growth promoting factors it brings useful microflora which helps in nitrate up take kinetics as reported by various authors (de Brito-Alvarez *et al.*, 1995; Muscolo *et al.*, 1999; Atiyeh *et al.*, 2000).

The studies on tomato and strawberries amended with Vermicompost also revealed growth and yield improvement (Arancon *et al.*, 2004, 2006; Gutiérrez-Miceli *et al.*, 2007; Zaller, 2007; Bachman & Metzger, 2008). Similarly, amendment of Biogas slurry is also reported to improve the growth and yield of wheat (Garg *et al.*, 2005).

Since the seed rhizome of approximately same weight (30-35 g) containing two well-differentiated bud initials was sown in each pot, the vegetative material per pot could be considered as single plant. The optimum spacing for planting ginger is reported 25 - 45 cm between the rows and 15 - 20 cm between plants in each row (Chadha, 1976). Hence keeping an account of the land to be left uncultivated a minimum of six plants could be grown in an area of one square meter, which amounts to 60,000 plants per hectare. Calculating mean fresh weight of ginger rhizome obtained per plant in the present investigation, reduction in yield of rhizome under saline water irrigation is 47.2%, whereas Vermicompost amendment increased it by 30.76% under non-saline condition and under saline water irrigation by 70.77% in comparison with its respective saline controls. Hence one can determine economically feasible benefit for ginger cultivation under Vermicompost application.



C=Non-saline control, VC=Vermicompost, BS=Biogas slurry, VC+BS=Vermicompost + Biogas slurry

Fig. 4. Effects of amending Vermicompost and Biogas slurry in soil on Na⁺ ion (A), K⁺ ion (B) and K⁺/Na⁺ ratio (C) of Ginger leaves irrigated with 0.2% sea salt dilution ($EC_{iw} = 2.9 \text{ dS m}^{-1}$, $EC_e = 3.6 \text{ dS m}^{-1}$).

Ginger cultivation in the light of above mentioned findings could be undertaken at non-saline sandy loam using saline water up to $EC_{iw} = 2.9 \text{ dS m}^{-1}$ in case non-saline water is not available. Soil amendment with Vermicompost has definitely given profitable growth under non-saline as well as under moderate saline conditions (up to $EC_{iw} = 2.9 \text{ dS m}^{-1}$). It appears that nitrogen from Biogas slurry was not made available for growth probably due to its higher EC. Results show that wherever biogas slurry is used alone or in mixture the growth performance was not as good as expected. Further research work could help in preventing increase of salt in Biogas slurry in order to make it a useful fertilizer for providing nitrogen.

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