

## RESPONSE OF BROCCOLI TO ORGANIC AMENDMENTS AND ACCUMULATION OF HEAVY METALS IN IT

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### Abstract

A field experiment was conducted to see the effect of cow-dung and sewage sludge as organic amendments to broccoli along with chemical fertilizer and heavy metal accumulation due to these organic amendments. The curd yield was significantly increased with the application of both cow-dung ( $p = 0.039$ ) and sewage sludge ( $p = 0.047$ ). The highest curd yield for cow-dung and for sewage sludge amendments were 22.18 and 21.73 t/ha, respectively. These yields were obtained from the treatments where 5 t/ha of both cow-dung and sewage sludge were applied along with the recommended dose of chemical fertilizer. Higher amounts of Cd and Pb were found to have accumulated in plants at higher rates (20 t/ha) of organic amendments.

**Key words:** Broccoli, cow-dung, sewage sludge, growth, heavy metal

### Introduction

Bangladesh is a developing country where agriculture plays a vital role in her economy. About 85% of the total population in the country depend on agriculture directly or indirectly for their livelihood (BBS 2004). Bangladesh is an agricultural country with a per capita land of 0.06 hectare, it possesses one of the lowest agricultural growth rates in the world (FAO 1986). Fertilizer is a major modern farming input and about 50% of the world's crop production being attributed to fertilizer use (Pradhan 1992). Fertile soil is the fundamental resource for higher production so its maintenance is a pre-requisite for long term sustainable crop productivity. However, it is true that the sustainable production of crops cannot be maintained by using chemical fertilizers alone. Similarly, it is not possible to obtain higher crop yield by using only organic manure (Bair 1990). Integrated use of organic and chemical fertilizers would be quite promising not only for sustainable production, but also for maintaining higher soil fertility status (Nambiar 1990) as well as preventing the soil resource from degradation (Bhuiyan 1994). The

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organic matter content of most of the Bangladesh soils are very low, the majority being below the critical level of 1.5% (BARC 2005). Soil organic matter has been depleted by 5-36% during the period 1967-1995 in Bangladesh (Ali *et al.* 1997). It is therefore, needed to use organic and chemical fertilizers in an integrated way to obtain economically profitable crop yields without experiencing unacceptable degradation of soil's quality.

The total cultivable land is around 9.17 million ha, with a cropping intensity of 154.5% (BBS 2004). Vegetables occupy only 1.7% of the cultivable land. Vegetables can play a vital role in elevating the nutritional status of the Bangladesh people. Vegetable production in Bangladesh is far below requirements. Scientists of different research institutes are trying to acclimatize different exotic vegetables like broccoli, baby corn, capsicum, etc. All these are rich in vitamin content and expected to be available in common people's diet in near future. Currently, there is about 1,55,000 ha land under vegetable production in the country of which 60,000 ha is under summer vegetables (FAO 1986). The winter vegetables contribute to about 70% of the total vegetable production of one million ton. More than 40 different kinds of vegetables are grown in Bangladesh. Broccoli (*Brassica oleracea* L., Family Brassicaceae) is a high quality vegetable for use and is one of the more popular frozen vegetables. It is not only one of the tastiest vegetables in the world, but also an integral part of a very healthy diet. It is an excellent source of the vitamins K, C, A and D, beta carotene, folate and fiber. Broccoli is also a very good source of phosphorus, potassium, magnesium, calcium and the vitamins B6, riboflavin (or vitamin B2). The prospect for growing broccoli in our country is good. Its cultivation should be increased. Bangladesh Agricultural Research Institute (BARI) has conducted many experiments with broccoli regarding its fertilizer requirement. In the backdrop of these considerations, the present research has been carried out with the objectives to study the effect of different sources of organic matter on the production and yield of broccoli and to study the accumulation of heavy metals in broccoli as contributed from different sources of organic matter application.

#### **Materials and Methods**

A field experiment was conducted at the central farm of Bangladesh Agricultural Research Institute (BARI), Joydebpur, Gazipur. The experimental plots selected were well-drained high land. The soil is Grey Terrace belonging to Chhiata series (AEZ-28). The geographic location is 23<sup>o</sup>50' N and 82<sup>o</sup>20' E.

***Sample collection***

The soil samples were collected from the experimental site by composite soil sampling method as suggested by the soil survey staff of the USDA (1951) for laboratory analysis. Sewage sludge were collected from Pagla sewage treatment plant, Dhaka and cow-dung were collected from the Milkway dairy farm, Bangladesh Agricultural Research Institute (BARI), Joydebpur, Gazipur.

***Sample preparation***

Soil samples were air dried and visible roots and debris were removed. After air-drying, a portion of the larger and massive aggregates were broken by gently crushing them by a wooden hammer. The collected sewage sludge and cow-dung were also air-dried and ground. The ground samples were then screened to pass through a 0.5 mm stainless steel sieve and mixed thoroughly. The samples were preserved in plastic containers and labeled properly which were used for various laboratory analyses.

***Collection of seedlings and fertilizers***

Seedling of broccoli (*Brassica oleracea* L.) were collected from the Bangladesh Agricultural Research Institute (BARI), Joydebpur, Gazipur and fertilizers (urea, TSP, MP, gypsum, zinc, boric acid and sodium molybdate) were collected from certified fertilizer dealer.

***Experimental set-up***

The field experiment was conducted at the central farm of Bangladesh Agricultural Research Institute (BARI), Joydebpur, Gazipur. The experiment was set up in a completely randomized block design with 5 treatments replicated three times. There were 24 plots of 3 m × 2.4 m size each. Twenty five days old healthy seedlings of broccoli (cv. Green sprout) were transplanted with spacing of 60 cm from row to row and 40 cm from plant to plant accommodating 30 plants in each plot. Nutrient combinations in each treatment, along with sewage sludge and cow-dung are shown in Table 1. Urea, TSP, MP, gypsum, zinc, boric acid and sodium molybdate were used as the source of N, P, K, S, Zn, B and Mo, respectively. The recommended doses of P, K, S, Zn, B and Mo, cow-dung and sewage sludge and  $\frac{1}{3}$  N were applied at the time of final land preparation and the remaining  $\frac{2}{3}$  N was applied in two equal installments, each at 3rd and 5th week after transplanting (bulking and curd formation stage), respectively (BARC 2005). All intercultural operations such as weeding, irrigation etc. were done as and when necessary.

**Table 1. Treatment combination for broccoli.**

Treatments	Treatment combination								
	N	P	K	S	Zn	B	Mo	Sewage sludge	Cow-dung
	kg/ha							t/ha	
T <sub>1</sub>	150	45	80	30	5	2	1	0	0
T <sub>2</sub>	100% recommended fertilizer							0	5
T <sub>3</sub>	100%		"		"		0	10	
T <sub>4</sub>	100%		"		"		0	20	
T <sub>5</sub>	100%		"		"		5	0	
T <sub>6</sub>	100%		"		"		10	0	
T <sub>7</sub>	100%		"		"		20	0	
T <sub>8</sub>	Native nutrient (control)							-	-

Harvesting was done when the heads attained proper size and were at the right stage of maturity. It was continued up to the branching of new curd. Five plants from each plot were tagged at random to take records on plant height, number of leaves per plant, circumference of curd and curd weight (t/ha). Then the plants were washed with water. The plants were separated into curd, leaves and stem and the fresh weight was taken. The samples were dried in an oven at  $80 \pm 5^{\circ}\text{C}$  and dry weight was taken. The oven-dried samples were ground and passed through a 0.2 mm sieve for analyses. After harvesting, soil samples from each plot were also collected and processed for further laboratory analyses.

#### *Laboratory analysis*

The various properties of the soil, cow-dung, and sewage sludge were determined following the procedures described in Imamul Huq and Alam (2005). Soil, cow-dung, sewage sludge and plants were analyzed for total Pb, Cd, Zn, Fe and Mn by atomic absorption spectrometer (AAS). Soil, cow-dung and sewage sludge were extracted with aqua-regia solution (Portman and Riley 1964). The plant samples were extracted with ternary acid mixture solution  $\text{HNO}_3 : \text{H}_2\text{SO}_4 : \text{HClO}_4$  at 5 : 1 : 2. Certified reference materials (CRMs) were used through the digestion and analyzed as part of the quality assurance/quality control protocol. Each batch of 20 samples was accompanied with reference standard samples to ensure strict QA/QC procedures.

#### *Statistical analysis*

Statistical analyses of all collected data of the experiment were made by using commonly used statistical software Minitab 14.2.

#### **Results and Discussion**

The collected soil, cow-dung and sewage sludge samples were analyzed in the laboratory before set up of the experiment to see the nutrient as well as some elemental status of the soil. The analytical values are shown in the Table 2.

**Table 2. Properties of the collected soil, cow-dung and sewage sludge samples.**

Properties	Soil	Cow-dung	Sewage sludge
pH	6.52	-	-
Particle size analyses		-	-
Sand	18.8 %		
Silt	40.72 "		
Clay	40.48 "		
Texture	Silty clay	-	-
Moisture content	5.20 %	-	-
Organic matter	0.82 "	6.29 %	11.19 %
Total nitrogen	0.36 "	1.60 "	2.56 "
Total phosphorus	5.22 ppm	31.9 ppm	58.84 ppm
Total potassium	0.02 %	0.025 %	0.015 %
Available phosphorus	4.04 ppm	20.75 ppm	52.15 ppm
Available potassium	65 "	1001 "	233 "
Cadmium	0.58 "	0.95 "	0.82 "
Lead	9 "	14.53 "	288.8 "
Zinc	37.72 "	92.80 "	268.54 "
Iron	26.78 "	19.38 "	20.56 "
Manganese	468.06 "	652.93 "	196.86 "

***Effects of various rates of organic matter application on the production of broccoli***

The growth of broccoli as affected by different rates of cow-dung was observed during the experimental period. Results presented in Table 3 shows that the highest curd yield ( $22.12 \pm 0.30$  t/ha) was obtained from the treatment T<sub>2</sub> (RD + 5 t/ha CD) whereas the lowest curd yield ( $11.35 \pm 0.15$  t/ha) was obtained from the treatment T<sub>8</sub> (control). The yield showed an increase of more than 94.89% over control as well as slightly higher than T<sub>1</sub> (RD). The yield increase could have happened because of addition of organic matter that might have improved the soil physical environment thus contributing to better plant growth leading to higher yield. The lowest yield from the control implies that the native nutrients failed to generate substantial yield. The effect of all the treatments on the growth of broccoli was significant ( $p = 0.039$ ). The growth of broccoli as affected by different rates of sewage sludge was also investigated during the experiment. Results presented in Table 3 shows that the effect of sewage sludge along with the recommended dose of fertilizer followed the same trend as was observed for cow-dung. The highest curd yield (21.73 t/ha) was obtained from the treatment T<sub>5</sub> (RD + 5 t/ha SS) whereas the lowest curd yield ( $11.35 \pm 0.15$  t/ha) was obtained from the treatment T<sub>8</sub> (control) with an increase of more than 91.45% over control. The effect of all the treatment on the growth of broccoli was significant ( $p = 0.047$ ).

**Table 3. Effect of different rates of cow-dung and sewage sludge on the yield of broccoli.**

Treatment	Yield (t/ha)	Treatment	Yield (t/ha)
T <sub>1</sub> (RD)	21.51 ± 0.72	T <sub>1</sub> (RD)	21.51 ± 0.72
T <sub>2</sub> (RD + 5 t/ha CD)	22.12 ± 0.30	T <sub>5</sub> (RD + 5 t/ha SS)	21.73 ± 0.08
T <sub>3</sub> (RD + 10 t/ha CD)	17.43 ± 0.43	T <sub>6</sub> (RD + 10 t/ha SS)	19.28 ± 0.18
T <sub>4</sub> (RD + 20 t/ha CD)	18.83 ± 0.35	T <sub>7</sub> (RD + 20 t/ha SS)	20.84 ± 0.61
T <sub>8</sub> (Control)	11.35 ± 0.15	T <sub>8</sub> (Control)	11.35 ± 0.15

RD = Recommended dose of chemical fertilizer, CD = Cow-dung, SS = Sewage sludge.

It appears thus that addition of either CD or SS can improve the yield of broccoli. However, the best result is obtained either with 5 t/ha of CD or 5 t/ha of SS along with the RD. Higher rates (10 and 20 t/ha) of either CD or SS could have led to fixation of some nutrient elements through chelation that might be one of the reasons for lower yield (Table 3) compared to 5 t/ha (Molla and Imamul Huq 2004). Besides, the higher rates of either CD or SS could have created oxygen depletion in the rhizosphere that contributed to lesser yield compared to 5 t/ha application.

#### ***Effects of cow-dung on the accumulation of Cd, Pb, Fe, Zn and Mn in broccoli***

It was observed that the whole plant of broccoli accumulated the highest amount of Cd and Pb (0.71 and 7 mg/kg d.w., respectively) from T<sub>4</sub> (the recommended dose of chemical fertilizer plus 20 t/ha cow-dung treated soil) and accumulated the least amount of Cd and Pb (0.11 and 2.75 mg/kg d.w., respectively) from T<sub>8</sub> (no chemical fertilizer and no organic matter). Though increased rates of cow-dung application in combination with the recommended dose of chemical fertilizer increased the accumulation of Cd and Pb in broccoli but the difference in uptake of Cd and Pb by the total plants of broccoli due to the application of different rates of cow-dung (5, 10 and 20 t/ha) was not significant. The presence of Cd and Pb in T<sub>1</sub> (where only recommended rate of chemical fertilizer was applied) indicates the presence of these elements in chemical fertilizer. So, the presence of Cd and Pb in broccoli could be due to the presence of Cd and Pb in chemical fertilizer as well as cow-dung. Both the Cd and Pb contents were higher in stem compared to curds and leaves. The Cd uptake in different parts of broccoli followed the order: stems > leaves > curds from maximum to minimum whereas the Pb uptake order was stems > curds > leaves from maximum to minimum. The whole plants of broccoli accumulated the highest amount of Fe (1830 mg/kg d.w.) from the recommended dose of chemical fertilizer plus 20 t/ha cow-dung treated soil and the least amount of Fe (847 mg/kg d.w.) from the recommended dose of chemical fertilizer plus 5 t/ha cow-dung treated soil. It was also observed that the increasing rates of cow-dung application in soil increased the uptake of Fe by curds, leaves, and stems of broccoli. The Fe content was higher in leaves

compared to curds and stems. Therefore the Fe uptake order of different parts of broccoli could be arranged as leaves > curds > stems from maximum to minimum.

**Table 4. Accumulation of Cd, Pb, Fe, Zn and Mn in broccoli due to cow-dung application.**

Treatment	Plant (mg/kg)				
	Cd	Pb	Fe	Zn	Mn
T <sub>8</sub> (Control)	0.11	2.75	1148	52.75	43.75
T <sub>1</sub> (RD)	0.28	2.86	1340	133	40.43
T <sub>2</sub> (RD + 5 t/ha CD)	0.58	5.37	847	37.67	55.50
T <sub>3</sub> (RD + 10 t/ha CD)	0.63	6.57	1021	59.12	53.50
T <sub>4</sub> (RD + 20 t/ha CD)	0.71	7.00	1830	106	44.14

RD = Recommended dose of chemical fertilizer, CD = Cow-dung.

Table 4 shows that the whole plants of broccoli accumulated the highest amount of Zn (133 mg/kg d.w.) from the recommended dose of chemical fertilizer treated soil and the least amount of Zn (37.67 mg/kg d.w.) from the recommended dose of chemical fertilizer plus 5 t/ha cow-dung treated soil. These results are in consistent with the findings of Molla and Huq (2004), where they showed that the uptake of Zn by *Amaranthus gangeticus* L. decreased due to application of compost in soil. The probable cause of the decrease in the accumulation of Zn may be that Zn<sup>2+</sup> forms stable complex with cow-dung. The humic and fulvic acid fractions are prominent in Zn<sup>2+</sup> adsorption or immobilization by high molecular weight organic substances such as lignin or complexation by initially soluble organic substances that forms insoluble salts (Tisdale *et al.* 1993). The Zn content was higher in curds compared to stems and leaves. The Zn uptake order of different parts of broccoli could be arranged as curds > stems > leaves from maximum to minimum. The whole plants of broccoli accumulated the highest amount of Mn (55.5 mg/kg d.w.) from the recommended dose of chemical fertilizer plus 5 t/ha cow-dung treated soil and the least amount of Mn (40.43 mg/kg d.w.) from the recommended dose of chemical fertilizer treated soil. In the present experiment, it was observed that the increasing rates of cow-dung application in soil decreased the uptake of Mn by curds, leaves and stems of broccoli. The reason for low accumulation of Mn in curds, leaves and stems of broccoli could be attributed to the formation of organo-metal complex. The Mn content was higher in leaves compared to curds and stems. The Mn uptake order of different parts of broccoli could be arranged as leaves > curds > stems from maximum to minimum. The difference in uptake of Fe, Zn and Mn by the total plants of broccoli due to the application of different rates of cow-dung (5, 10 and 20 t/ha) was not significant.

***Effects of sewage sludge on the accumulation of Cd, Pb, Fe, Zn and Mn in broccoli***

The accumulation of Cd and Pb in broccoli due to the application of sewage sludge was similar to that of cow-dung application. The highest amount of Cd (2.43 mg/kg d.w.) and Pb (7 mg/kg d.w.) was accumulated from the recommended dose of chemical fertilizer plus 20 t/ha sewage sludge treated soil whereas the least amount of Cd (0.11 mg/kg d.w.) and Pb (2.75 mg/kg d.w.) was accumulated from no chemical fertilizer and no organic matter treated soil. The differences in uptake of Cd and Pb by the whole plants of broccoli due to application of different rates of sewage sludge (5, 10 and 20 t/ha) were not significant. The Cd and Pb content was maximum in stems compared to curds and leaves and followed the order stems > leaves > curds from maximum to minimum for Cd uptake while for Pb uptake, the order being stems > curds > leaves from maximum to minimum. The whole plants of broccoli accumulated the highest amount of Fe (2359 mg/kg d.w.) from the recommended dose of chemical fertilizer plus 20 t/ha sewage sludge treated soil and the least amount (873 mg/kg d.w.) from the recommended dose of

**Table 5. Accumulation of Cd, Pb, Fe, Zn and Mn in broccoli due to sewage sludge application.**

Treatment	Plant (mg/kg)				
	Cd	Pb	Fe	Zn	Mn
T <sub>8</sub> (Control)	0.11	2.75	1148	52.75	43.75
T <sub>1</sub> (RD)	0.28	2.86	1340	133	40.43
T <sub>5</sub> (RD + 5 t/ha SS)	0.22	4.86	873	63.28	41.86
T <sub>6</sub> (RD + 10 t/ha SS)	0.57	6.43	1071	170	49.28
T <sub>7</sub> (RD + 20 t/ha SS)	2.43	7.35	2359	329	50.43

RD = Recommended dose of chemical fertilizer, CD = Cow-dung.

chemical fertilizer plus 5 t/ha sewage sludge treated soil. The Fe content was higher in leaves compared to curds and stems and followed the order leaves > curds > stems from maximum to minimum. The whole plants of broccoli accumulated the maximum amount of Zn (329.28 mg/kg d.w.) from the recommended dose of chemical fertilizer plus 20 t/ha sewage sludge treated soil and the least amount (52.75 mg/kg d.w.) from the no chemical fertilizer and no organic matter treated soil. The Zn content was maximum in curds compared to stems and leaves and followed the order curds > leaves > stems from maximum to minimum. The whole plants of broccoli accumulated the highest amount of Mn (50.43 mg/kg d.w.) from the recommended dose of chemical fertilizer plus 20 t/ha sewage sludge treated soil and the least amount (40.43 mg/kg d.w.) from the recommended dose of chemical fertilizer treated soil. The Mn content was higher in leaves compared to curds and stems and the order was similar to that of Fe. It was also observed that the difference in uptake of Fe, Zn and Mn by the whole plants of broccoli



due to application of different rates of sewage sludge (5, 10, and 20 t/ha) was not significant. So, it may be recommended that the application of both cow-dung and sewage sludge at the rate of 5 t/ha will increase the curd yield of broccoli.

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(Manuscript received on 09, May 2012; revised on 29, July 2012)