

**Effect of Different Sources of Nitrogen on Yield and Quality of Broccoli (*Brassica oleracea* L. Var. *Italica*) Varieties under Gaindakot, Nawalparashi, Nepal Condition**Samjhana Lamichhane<sup>1</sup>, M. D. Sharma<sup>1</sup>, S.S. Panta<sup>1</sup> and S.C. Shah<sup>1</sup>**ABSTRACT**

The experiment was conducted at farmer's field of Gaindakot, Nawalparasi during October 2012 to February 2013 to evaluate the influence of different sources of nitrogen and varieties on yield and quality of broccoli. The experiment was laid out in two-factorial RCBD with three replications. A total of fifteen treatments with combination of five different sources of nitrogen and three varieties were taken in each replication. The result showed that the variety Premium Crop produced the highest (23.70 t/ha) terminal curd yield in  $N_{50\%Urea}+N_{50\%Vermi}$  (20.30 t/ha) whereas Calabrese produced the highest (9.34 t/ha) auxiliary curd yield in  $N_{50\%Urea}+N_{50\%Vermi}$  (8.25 t/ha). Green Sprouting produced the highest total yield (25.10 t/ha) in  $N_{50\%Urea}+N_{50\%Vermi}$  (28.55 t/ha). Calabrese exhibited higher (13.27%) dry matter and the lowest (18.44%) physiological weight loss of curd in  $N_{100\%FYM}..N_{50\%Urea}+N_{50\%Vermi}$  produced more compact curd while  $N_{100\%Vermi}$  produced the most appreciable taste, color and overall acceptability of curd. Premium Crop performed better regarding compactness, color and acceptability of curd while Calabrese performed better on taste. Thus, Calabrese along with  $N_{100\%Vermi}$  or  $N_{100\%FYM}$  would be suggested for better quality regarding physiological weight loss and taste while Premium crop along with  $N_{100\%Vermi}$  would be suggested for attractive color and overall acceptability of curd.

**Keywords:** curd quality, FYM, physiological weight loss, Vermi-compost

**INTRODUCTION**

Broccoli is an important vegetable crop which has high nutritional and commercial value (Brahma *et al.*, 2002). It belongs to family *Brassicaceae*. It is a rapidly developing compact floral vegetable that is harvested at compact head and immature bud stage (Gray, 1982). Nutritionally, broccoli is an excellent source of antioxidants, vitamin C, fibre and folate. It contains good levels of iron, calcium, potassium and vitamin A and E (Lister and Bradstock, 2003). It possesses cancer fighting properties related to the high levels of active plant chemicals called glucosinolates (Zhao *et al.*, 2007). Broccoli is comparatively a newer winter vegetable in Nepal (Ghimire *et al.*, 1993). MoAD (2013) reported that the total area under broccoli in Nepal in the year 2012/13 was 2138 ha with the production of 26769 mt and productivity 12.5 mt ha<sup>-1</sup>.

Being a heavy feeder crop, broccoli demands constant supply of large amount of both macro and micro nutrients for its luxuriant growth. Among the essential nutrients, nitrogen plays significant role in metabolism, growth, reproduction and hereditary characters of plant (Dutta, 1998). The indiscriminate use of chemical fertilizers degrades the soil quality, increases susceptibility to pests and diseases and also creates micronutrient deficiencies. Thus, an alternative source of low cost plant nutrient is necessary to search for the maintenance of soil fertility and productivity over a longer period of time minimizing the adverse impact of chemical fertilizer on soil.

---

<sup>1</sup> Institute of Agriculture and Animal Science Rampur, Chitwan, Nepal  
Mailing author: remember2samjhana@gmail.com,

Organic manure is the traditional source of plant nutrient which is most readily available to the farmers (Gaur *et al.*, 1995). The vermin-compost improves the soil physics, chemical structure, and promotes biological properties of it (Suthar, 2008). It contains the plant hormone like substance which may be due to the presence of higher microbial population (Krishnamoorthy and Vajranabhaian, 1986). Application of vermicompost significantly increases the micronutrient in the field soil than with animal manure (Reddy and Reddy, 1999). It acts as a better source of plant nutrient to substitute or complement the chemical fertilizer and may also reduce the need for synthetic pesticides as it also provide resistance to disease and hence help to produce quality curd.

FYM plays an important role in maintaining and improving the soil as it contains all plant nutrients, humus and organic substances. Additionally, it helps in solubilization of plant nutrients and increases the uptake of N, P, K, Ca and Mg during crop growth (Subbiah *et al.*, 1982). It is readily available in the most part of the country.

Neither the chemical fertilizers alone nor organic sources exclusively can achieve the production sustainability of soil as well as crops under high intensive cropping systems (Singh and Yadhav, 1992). Quality product along with higher economic return can be obtained without deteriorating the soil condition for subsequent cropping through the judicious application of organic and inorganic fertilizer (Devi *et al.*, 2003).

Broccoli is an important vegetable having shorter shelf life, which hasten the post harvest losses as well as economic loss and make the crop unpopular even then it has greater potential to improve nutritional situation of the country. Moreover, awareness of the consumers towards quality product is increasing day by day in different parts of the country. This indicates the enough scope for its promotional efforts. Not enough work has yet been done to study the effect of nitrogen sources and varieties on yield and quality of broccoli. Every variety does not respond equally to the available nutrients at all places of different climatic condition. Therefore, the suitable variety and appropriate source of nutrient to the plant must be identified to obtain optimum and quality yield with better post harvest life.

## METHODOLOGY

The study was conducted at farmer's field of Gaindakot, Nawalparasi from October 2012 to February 2013. The experiment was laid out in a factorial randomized complete block design (RCBD) with three replications. A total of fifteen treatments with the combinations of five different sources of nitrogen ( $N_{100\% \text{Urea}}$ ,  $N_{100\% \text{Vermi}}$ ,  $N_{100\% \text{FYM}}$ ,  $N_{50\% \text{Urea}} + N_{50\% \text{Vermi}}$  and  $N_{50\% \text{Urea}} + N_{50\% \text{FYM}}$ ) and three varieties of Broccoli (Calabrese, Green Sprouting and Premium Crop) were taken in each replication. First two varieties were open pollinated while the third one was hybrid variety. Source of nitrogen was the first factor of experiment while the variety of broccoli was the second under the experiment. There were a total of 45 plots with 4.5 m<sup>2</sup> of each. Seedlings were transplanted at spacing of 45 x 40 cm in each plot. All the treatments were based on the fulfillment of the required nitrogen by the crop (100 k ha<sup>-1</sup>). Required quantity of vermicompost, FYM and chemical fertilizer (urea) was calculated on the basis of total amount of nitrogen present in them. The laboratory analysis of vermicompost, FYM and soil before transplanting of seedlings was done in the regional soil laboratory, Pokhara, Nepal. Data recording was done on various parameters such as curd yield, dry matter content of curd, physiological weight loss, sensory evaluation regarding compactness, color, taste, and over all acceptability of curd of broccoli. Collected data were subjected to analysis of variance and Duncan's Multiple Range Test (DMRT) for mean separation using MSTAT-C.

### Physiological weight loss

A hundred gm of randomly selected curd from each treatment was kept in ordinary room condition for four days until they became unmarketable. Then postharvest loss in weight was calculated to determine the post harvest life of the curd using formula:

$$\text{Physiological weight loss(\%)} = \frac{\text{initial wt. of sample} - \text{Final wt. of sample}}{\text{Initial wt. of the sample}} \times 100$$

### Sensory evaluation

Sensory evaluation was done to judge the compactness, taste, color and overall acceptability of the curd at harvest stage by a panel of 10 people using hedonic scale of 1 to 9 in which 1 being the poor and 9 being the excellent performance (Acedo and Bautista, 1999). The overall acceptability of curd of broccoli was evaluated based on consumer's preference to the product. The description of hedonic scale for quality parameter is given below:

**Table 1.** The description of hedonic scale

Scale	Color	Taste	Compactness	Acceptability
1-3	Poorest	Poorest	Poorest	
3-5	Poor	Poor	Poor	Poor
5-7	Good	Good	Good	Good
7-9	Better	Better	Better	Better
9	Best	Best	Best	Best

Source: Hedonic scale (Acedo and Bautista, 1999)

**Table 2.** Laboratory analysis of vermicompost, FYM and soil before seedling transplanting

Manure	Nutrient content percentage			Organic matter	pH	Soil texture
	Nitrogen	Phosphorous	Potash			
Vermicompost	1.4(%)	0.71 %	1.98 (%)	-	-	-
FYM	0.67(%)	0.38 %	1.14 (%)	-	-	-
Soil	0.23(%) (high)	967kg/ha (high)	648kg/ha (high)	4.6 % (medium)	5.4 (acidic)	Sandy loam

## RESULTS AND DISCUSSION

### Curd yield

Nitrogen source, N<sub>50%</sub>Urea + N<sub>50%</sub>Vermi recorded significantly (p< 0.01) the higher terminal curd yield (20.30 t ha<sup>-1</sup>), auxiliary curd yield (8.25 t ha<sup>-1</sup>) and total curd yield (28.55 t ha<sup>-1</sup>) (Table 3). The highest terminal curd yield from N<sub>50%</sub>Urea + N<sub>50%</sub>Vermi was similar to N<sub>50%</sub>Urea + N<sub>50%</sub>FYM. However, nitrogen source, N<sub>100%</sub>FYM recorded significantly lower terminal curd yield (13.28 t ha<sup>-1</sup>), auxiliary curd yield (2.81 t ha<sup>-1</sup>) and total curd yield (16.09 t ha<sup>-1</sup>). These results were in agreement with the findings of Bhatrai and Mishra (2012) that the maximum yield was recorded by the application of 1/2NPK+ 2t ha<sup>-1</sup>vermicompost whereas the minimum yield was from control in broccoli.

There were significant (p<0.01) effects of different varieties on curd yield of broccoli (Table 3). No auxiliary curd was harvested from variety Premium Crop. Premium Crop produced the highest (23.70 t ha<sup>-1</sup>) terminal curd yield whereas Calabrese produced the lowest (10.53 t ha<sup>-1</sup>) terminal curd yield. Calabrese produced significantly higher (9.34 t ha<sup>-1</sup>) auxiliary curd yield than Green Sprouting (7.09 t ha<sup>-1</sup>). Likewise, Green Sprouting produced the highest (25.10 t ha<sup>-1</sup>) total curd yield which was similar to Premium Crop (23.70 t ha<sup>-1</sup>) whereas Calabrese produced the lowest (19.88 t ha<sup>-1</sup>) total curd yield. Although the highest terminal curd yield was recorded from Premium Crop, the highest total curd yield was from Green Sprouting. It

may be attributed to the multiple harvesting in Green Sprouting over a longer period of time but no auxiliary curd harvesting was held from Premium Crop. These results were also similar to the finding of Giri (2007) who found that Green Sprouting produced higher total yield than Calabrese.

**Table 3:** Effect of nitrogen sources and varieties on curd yield and yield attributing factors of broccoli at Gaidakot, Nawalparasi, Nepal during 2012/13.

Treatments	Curd yield (t ha <sup>-1</sup> )		
	Terminal	Auxiliary	Total
<b>Source of nitrogen (N)</b>			
N <sub>100%</sub> Urea	17.83 <sup>bc</sup>	2.20 <sup>c</sup> (5.54 <sup>c</sup> )	23.36 <sup>c</sup>
N <sub>100%</sub> Vermi	16.06 <sup>c</sup>	1.94 <sup>d</sup> (4.15 <sup>d</sup> )	20.21 <sup>d</sup>
N <sub>100%</sub> FYM	13.28 <sup>d</sup>	1.68 <sup>e</sup> (2.81 <sup>e</sup> )	16.09 <sup>e</sup>
N <sub>50%</sub> Urea + N <sub>50%</sub> Vermi	20.30 <sup>a</sup>	2.61 <sup>a</sup> (8.25 <sup>a</sup> )	28.55 <sup>a</sup>
N <sub>50%</sub> Urea+ N <sub>50%</sub> FY M	19.60 <sup>ab</sup>	2.38 <sup>b</sup> (6.64 <sup>b</sup> )	26.23 <sup>b</sup>
SEM±	0.77	0.04 (0.23)	0.57
LSD <sub>0.05</sub>	2.22**	0.12 (0.65)**	1.64**
<b>Varieties</b>			
Calabrese	10.53 <sup>c</sup>	3.05 <sup>a</sup> (9.34 <sup>a</sup> )	19.88 <sup>b</sup>
Green Sprouting	18.01 <sup>b</sup>	2.73 <sup>b</sup> (7.09 <sup>b</sup> )	25.10 <sup>a</sup>
Premium Crop	23.70 <sup>a</sup>	0.71 <sup>c</sup> (0.00 <sup>c</sup> )	23.70 <sup>a</sup>
SEM±	0.595	0.03 (0.18)	0.59
LSD <sub>0.05</sub>	1.72**	0.10 (0.51)**	1.70**
Grand mean	17.41	2.16 (5.48)	22.76
CV (%)	12.88	5.86 (12.38)	7.45

Means followed by the same letter (s) within a column are not significant at 5% level of significance as determined by DMRT. SEM = Standard error of mean, LSD = Least significant difference and CV = Coefficient of variance.

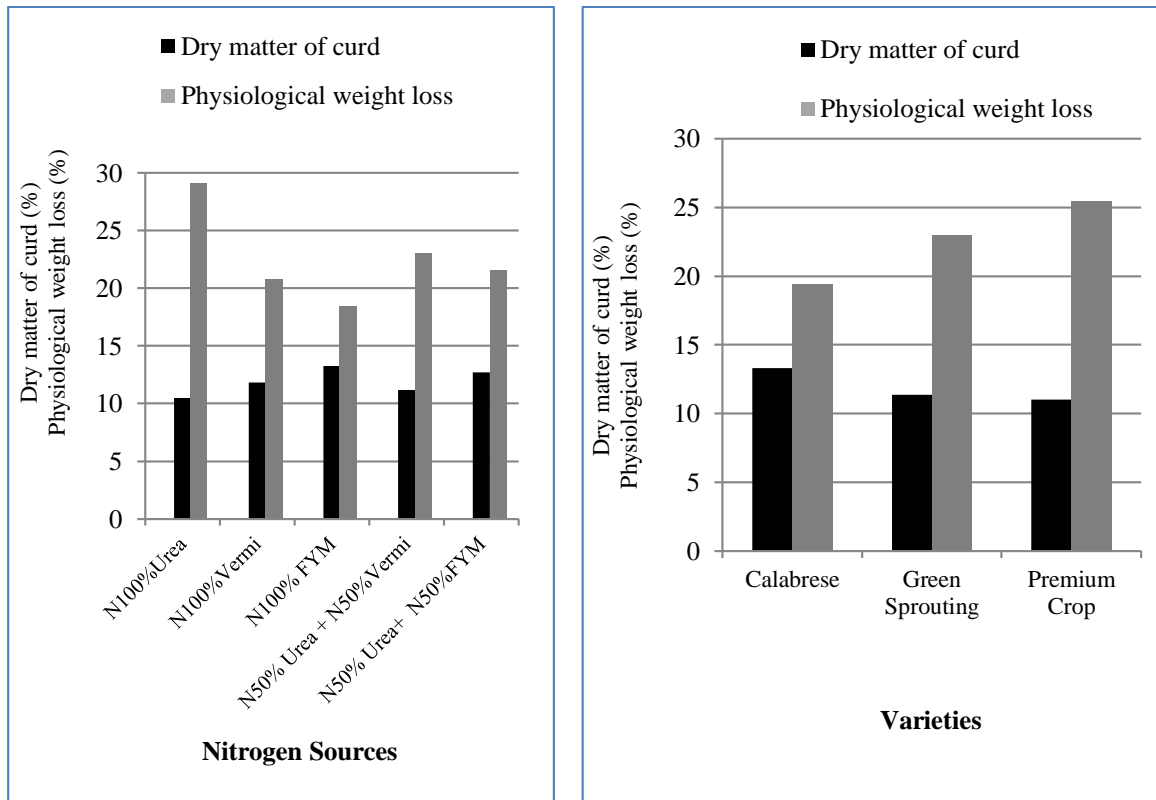
Note: Values on auxiliary curd yield were square root transformed at 0.5

### Dry matter of curd, physiological weight loss and shelf life of broccoli

Nitrogen source N<sub>100%</sub>FYM showed significantly higher (13.27%) dry matter percentage of curd whereas N<sub>100%</sub>Urea showed lower (10.47%) dry matter percentage of curd (Figure 1). Calabrese recorded significantly higher (13.32%) dry matter percentage of curd, whereas Premium Crop recorded lower (11.00%) dry matter percentage of curd. The highest dry matter percentage from N<sub>100%</sub>FYM might be due to the accumulation of higher amount of dry matter during the crop growth period while the lowest dry matter percentage with the chemical fertilizer might be due to lower amount of dry matter accumulation during the crop growth period. Similar results were also observed by Bhattari (2013) in broccoli.

Application of different sources of nitrogen and varieties had shown significant effects on physiological weight loss of curd in broccoli (Figure 1)). Nitrogen source, N<sub>100%</sub>Urea exhibited significantly higher (29.11%) physiological weight loss of the curd than rest of the treatments. Similarly, Premium Crop exhibited the highest (25.40 %) physiological weight loss of curd which was similar to Green Sprouting (22.93 %) whereas Calabrese exhibited the lowest value (19.40%). The reason of the least physiological weight loss from N<sub>100%</sub>FYM may be the influence of organic manure on physiological characters of curd and other biochemical properties, reducing respiration rate during storage. These findings were in accordance with the findings of Bhattra (2013) that maximum physiological weight loss of broccoli curd was from 100%N through chemical and minimum from 100%N through FYM. Physiological weight was higher from Premium Crop than Calabrese (Bhattra, 2013).

Although there were non-significant effect of different sources of nitrogen and varieties on shelf life of curd of broccoli, nitrogen source, N<sub>100%</sub>FYM showed longer shelf life of curd of broccoli whereas N<sub>100%</sub>Urea showed shorter shelf life. Similarly, Green Sprouting showed shorter shelf life followed by Premium Crop and Calabrese.



**Figure 1.** Effect of a) nitrogen sources and b) Varieties on dry matter and physiological weight loss of curd of broccoli at Gaindakot, Nawalparasi, Nepal during 2012/13

### Curd quality characters

Nitrogen source, N<sub>50%</sub>Urea + N<sub>50%</sub>Vermi produced significantly more (7.07) compactness of the curd while N<sub>100%</sub>Urea produced very loose curd (3.64) (Table 4). More compact curd might be due to the nitrogen and other mineral elements required for plant growth supplied to plant from vermicompost. However, poor compactness of curd from N<sub>100%</sub>Urea might be due to the excessive vegetative growth with smaller curd. Likewise, variety Premium Crop produced significantly more compact curd (6.35) than Green Sprouting (5.74) and Calabrese (3.79). This finding was in harmony with the finding of Bhattra (2013) that the compactness of the curd was more from Premium Crop than Calabrese.

Nitrogen source, N<sub>100%</sub>Vermi showed more tasty curd (7.31) which was significantly higher than rest of other treatments (Table 4). However, N<sub>100%</sub>Urea showed the poorest (4.47) curd taste. Sapkota (2013) also reported that the most appreciable tasty cauliflower curd was obtained from 100% N through vermicompost. The variety Calabrese obtained the highest score (6.67) for curd taste while Premium Crop obtained the lowest score (5.08). Similar finding was also reported by Bhattra (2013). The color of the curd was significantly influenced by different sources of nitrogen and varieties (Table 4).

Nitrogen source N<sub>100%</sub>Vermi recorded the most appreciable color score (6.19) which was similar to N<sub>100%</sub>FYM. However, N<sub>100%</sub>Urea recorded poor color score (3.38) which was identical to N<sub>50%</sub>Urea + N<sub>50%</sub>FYM. Sapkota (2013) also reported that the most appreciable color of cauliflower curd

was obtained from 100% N through vermicompost and the lowest color score from 100% N through urea. Premium Crop recorded the most appreciable color score (5.50) which was significantly higher than Green Sprouting (4.11) but at par with Calabrese. Bhattra (2013) also reported better color from Premium Crop than Calabrese.

Consumer's preference on the curd size, color, taste and appearance of the curd determines the overall acceptability. There were significant effects of different sources of nitrogen and varieties on acceptability of curd in broccoli (Table 4). Nitrogen source N<sub>100%Vermi</sub> obtained the highest (5.84) overall acceptability of curd while N<sub>100%Urea</sub> obtained the lowest (2.49) overall acceptability of curd.

**Table 4.** Effect of different sources of nitrogen and varieties on curd quality of broccoli at Gaidakot, Nawalparasi, during 2012/13

Treatments	Curd quality			
	Compactness	Taste	Color	Acceptability
Source of nitrogen (N)				
N <sub>100%Urea</sub>	3.64 <sup>e</sup>	4.47 <sup>e</sup>	3.38 <sup>d</sup>	2.49 <sup>e</sup>
N <sub>100%Vermi</sub>	5.27 <sup>c</sup>	7.31 <sup>a</sup>	6.19 <sup>a</sup>	5.84 <sup>a</sup>
N <sub>100% FYM</sub>	4.26 <sup>d</sup>	6.36 <sup>b</sup>	5.55 <sup>ab</sup>	5.04 <sup>b</sup>
N <sub>50%Urea</sub> + N <sub>50%vermi</sub>	7.07 <sup>a</sup>	5.78 <sup>c</sup>	4.73 <sup>bc</sup>	3.98 <sup>c</sup>
N <sub>50%Urea</sub> + N <sub>50%FYM</sub>	6.26 <sup>b</sup>	5.10 <sup>d</sup>	4.01 <sup>cd</sup>	3.12 <sup>d</sup>
SEM±	0.11	0.10	0.39	0.13
LSD <sub>0.05</sub>	0.328**	0.29**	1.13**	0.37**
Varieties				
Calabrese	3.79 <sup>c</sup>	6.67 <sup>a</sup>	4.70 <sup>ab</sup>	3.57 <sup>c</sup>
Green Sprouting	5.74 <sup>b</sup>	5.65 <sup>b</sup>	4.11 <sup>b</sup>	4.19 <sup>b</sup>
Premium Crop	6.35 <sup>a</sup>	5.08 <sup>c</sup>	5.50 <sup>a</sup>	4.53 <sup>a</sup>
SEM±	0.09	0.07	0.30	0.10
LSD <sub>0.05</sub>	0.25**	0.22**	0.87*	0.28**
Grand mean	5.29	5.83	4.77	4.10
CV (%)	6.42	5.17	24.49	9.33

Means followed by the same letter (s) within a column are not significant at 5% level of significance as determined by DMRT. SEM = Standard error of mean, LSD = Least significant difference and CV = Coefficient of variance.

## CONCLUSION

Premium crop produced the highest terminal curd yield, Calabrese produced the highest auxiliary curd yield and Green Sprouting produced the highest total yield with nitrogen source N<sub>50%Urea</sub> + N<sub>50%Vermi</sub>. Calabrese exhibited significantly higher dry matter percentage of curd and lowest physiological weight loss in N<sub>100%FYM</sub> whereas Premium Crop showed lowest dry matter and highest physiological weight loss in N<sub>100%Urea</sub>. Nitrogen source, N<sub>50%Urea</sub> + N<sub>50%Vermi</sub> produced more compact curd while N<sub>100%Vermi</sub> produced the most appreciable taste, color and overall acceptability of curd. Premium Crop performed better regarding the compactness, color and over all acceptability of curd while Calabrese performed better regarding curd taste. Thus, Calabrese along with the application of N<sub>100%Vermi</sub> or N<sub>100%FYM</sub> would be suggested for better quality regarding physiological weight loss and taste while Premium crop along with the application of N<sub>100%Vermi</sub> would be suggested for attractive color and overall consumer's acceptability of curd. Broccoli exhibited very short shelf life of only 3-4 days after harvest.

**LITERATURES CITED**

- Bhattraï, B.P and S. Mishra. 2012. Effect of integrated nutrient management on growth, yield and soil nutrient status on *Brassica oleracea* L. var. *Italica*. Nepalese Horticulture, Nepal Hort. Society Vol. 8-9: 84-90.
- Bhattraï, S. 2013. Yield and quality of broccoli varieties as affected by the replacement of inorganic nitrogen with organic source of nitrogen in chitwan. M.Sc. Thesis. Institute of Agriculture and Animal Science, Rampur, Chitwan, Nepal. pp. 35-61.
- Brahma, S., D. B. Phooka and B. P. Gautam, 2002. Effect of nitrogen, phosphorus, and potassium on growth and yield of broccoli (*Brassica oleraces* L. var. *italic*) cv. Pusa Broccoli KTS-1. J. Ind. Agric. pp. 45-50.
- Devi, H. J., T. K. Maity and N. C. Paria, 2003. Effect of different sources of nitrogen on yield and economics of cabbage. *Enviro. Ecol.* 21 (4): 878-880.
- Dutta, S. C., 1998. Plant physiology. New Delhi: New Age International Private Limited, India.
- Gaur, A. C., S. Neelakantan and K. S. Dargan, 1995. Organic manure. publication and information division. Indian Council of Agriculture Research, New Delhi. 159p.
- Ghimire, A.J., M.R. Bhattarai and R. Khanal, 1993. Effect of removing terminal or axillary heads on the yield and quality of seed of broccoli cultivar Green Sprouting. PAC working paper 77: 1-11.
- Giri, R. K. Response of broccoli (*Brassica oleracea* var. *italic*) cultivars to different levels of nitrogen at Rampur, Chitwan, Nepal. M. Sc. Thesis. Institute of Agriculture and Animal Science, Rampur, Chitwan, Nepal. pp. 33-71.
- Gray, A.R., 1982. Taxonomy and evolution of broccoli. (*Brassica oleracea* var. *italica*). *Economic Botany* 36(4): 397-410. Available at: <http://ses.library.usyd.edu.au/bitstream> (Retrieved on: 25<sup>th</sup> January, 2012)
- Krishnamoorthy, R. V. and S. N. Vajranabhaian, 1986. Biological activity of earthworm casts. An assessment of plant growth promotes or levels in the casts. *Proceedings of Indian Academy of Sciences (Animal Science)*. 95(3).
- Lister, C.E. and M. Bradstock, 2003. Antioxidants: A health revolution: all you need to know about antioxidants. Christchurch, NZ. New Zealand Institute for Crop and Food Research.
- MoAD, 2013. Statistical Information on Nepalese Agriculture 2012/13. Agri-Business Promotion and Statistics Division, Singha Durbar, Kathmandu, Nepal.
- Reddy, B.G. and M.S. Reddy, 1999. Effect of integrated nutrient management on soil available micronutrients in maize-soyabean cropping system. *J. Res. ANGRAU*. 27: 24-28.
- Singh, G. B. and D. V. Yadhav, 1992. Integrated nutrient supply system in sugarcane and sugarcane based cropping system. *Fert. News* 37: 15-22.
- Sapkota, D. R. 2013. Effect of different sources of nitrogen on mid season varieties of cauliflower (*Brassica oleracea* l. var. *botrytis*) at Gaindakot, Nawalparasi. M.Sc. Thesis. Institute of Agriculture and Animal Science, Rampur, Chitwan, Nepal. pp. 61-65.
- Subbiah, K., S. Sundararajan and R. Perumal, 1982. Response of tomato and brinjal to varying levels of FYM and macronutrients under different fertility status of soil. *South Ind. Hort.* 33: 198-205.
- Suthar, S. and S. Singh, 2008. Feasibility of vermicomposting in biostabilization sludge from a distillery industry. *The Science of Total Environment* 393: 237-243.
- Zhao, H., J. Lin, H. Barton Grossman, L.M. Hernandez, C.P. Dinney and X. Wu, 2007. Dietary isothiocyanates, GSTMI, GSTTI, NAT2 polymorphisms and bladder cancer risk. *Int. J. Cancer* 120 (10): 2208-2213.