RESPONSE OF CAULIFLOWER (*Brassica oleraceae* var. *botrytis* L.) TO DIFFERENT LEVELS OF BORON UNDER MAIZE BASED CROPPING SYSTEM

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ABSTRACT
To assess the effect of Boron (B) application on cauliflower, we conducted a greenhouse pot experiment that consisted of soil application of five levels of nutrient i) no input of fertilizers, ii) $N:P_2O_5:K_2O$ at the rate of 200, 28.6, and 80 kg ha^{-1} respectively (recommended dose of NPK), iii) recommended dose of NPK plus 1.1 kg ha^{-1} B, iv) recommended dose of NPK plus 2.2 kg ha^{-1} B, and v) recommended dose of NPK plus 4.4 kg ha^{-1} B. Total plant biomass was 106 g $plant^{-1}$ at 4.4 kg B ha^{-1} compared to 79 g $plant^{-1}$ at the control of no nutrient inputs. Relative to the control treatment, there was a two-fold increase in fresh curd yield at 1.1 Kg B ha^{-1} application. Boron application improved the shoot B content as well as its partitioning towards curd and was associated with higher B concentration in the curd. While the soil residual B was 0.26 mg Kg^{-1} when no B was applied, it was 0.3 mg Kg^{-1} at 4.4 Kg B ha^{-1} application. It is concluded that soil B application tends to improve the yield and quality of cauliflower cultivated at the upland maize-based system in mid-hill areas of Nepal.

Keywords: *biomass, boron partitioning, curd yield, residual boron, upland*

INTRODUCTION

Boron (B) is an essential micronutrient for plant growth and development. It is required for several physiological functioning in plants: translocation of sugars, root extension, growth of meristematic tissues, synthesis of amino acid, protein and carbohydrate metabolism, pollen enlargement, fertilization, and flowering processes of plants (Singh, 2003). Being immobile in plants, B deficiency affects the development of terminal buds of shoots and growing root tips (Albert & Wilson, 1961; Dell & Huang, 1997; Kouchi & Kumazawa, 1975). Therefore, its deficiency adversely affects the yield and quality of the crop. In cauliflower, B deficiency first appears as water-soaked patches on the developing head of cauliflower, which soon turn brown and rots. The leaves curl down and become brittle. Blistering may occur on the petiole and along the midrib. The pith stem below the curd breaks down and develops the cracks, which later on form an elongated cavity (Adhikary et al., 2004; Raja, 2007; Sarkar et al., 2012). B deficiency in the crops is mostly associated with soil characteristics. Soil type and cropping system may influence the concentration of B in soil. B deficiency is commonly observed in light-textured acidic soils, soils with a high amount of calcium carbonate ($CaCO_3$) or oxides and hydrous oxides of iron (Fe) and aluminum (Al), and soils with low organic matter content (Keren, 1985; Mandal et al., 2004). B has been reported to be deficient in most of the field-grown crops including vegetables (Shorrocks, 1997). Nepalese soils, particularly in uplands, are deficient in B (Andersen, 2007; Shrestha et al., 2020), and poor B fertility is mainly attributed to the coarse sandy soils which are poor in B minerals, acidic in reaction, and has a high rate of the B leaching from soil (Ahmad et al., 2012; Bajracharya et al., 2007; Carson, 1992; Dawadi & Thapa, 2015; Dear & Weir, 2005; Shrestha, 2015). In a maize-based upland cropping system, B is found to be inherently deficient due to dry soil conditions that limit B mobility (Fleming, 1980; Hobbs & Bertramson, 1950; Wilcox, 1960).

Investigations in different parts of the world have revealed that B application can increase as high as 90% curd yield in cauliflower (Batabyal et al., 2015; Kotur, 1991). However, there has been a dearth of adequate information regarding cauliflower response to the external inputs of B when cultivated in maize-based upland areas in midhill of Nepal. This study hypothesized that cauliflower can respond better to the added B in a maize-based system for its poor B fertility. We compared the effects of five levels of B on growth, yield, B sharing between curd and remaining parts of the shoot in cauliflower, and soil residual B. This study aimed to contribute to the site-specific B management for cauliflower production in maize-based upland soils.

MATERIALS AND METHODS

Description of the site for soil sampling and analysis

We collected soil samples (0-20 cm depth) from a maize-based cauliflower production site in Kuntabeshi, Kavre (27° 31’ 33.24” N and 85° 33’ 40.32” E) during the first week of September 2017. Collected soil samples were taken into Aastha Scientific Research Service Private Limited, Kathmandu, and analyzed for particle size distribution, organic matter, and soil pH. Similarly, total nitrogen (N), plant-available phosphorus (P_2O_5), plant-available potassium (K_2O), and plant-available B were analyzed (Table 1). The analysis showed that soils collected were rich in plant-available P and K, moderate in soil organic matter and total N but poor in B fertility.

Table 1. Physical and chemical properties of soil used in the experiment.

Soil properties	Analysis methods	Value
Soil textural class	Hydrometer method (Gee & Bauder, 1986)	Sandy clay loam
Soil organic matter (%)	Walkey and Blacks’ titration method (Walkley & Black, 1934)	2.28
Total N (mg Kg^{-1} soil)	Kjeldhal distillation (Bremner & Mulvaney, 1983)	1100
Plant available P (mg Kg^{-1} soil)	Modified Olsen method (Olsen et al., 1954)	849.36
Plant available K (mg Kg^{-1} soil)	Flame photometry (Knudsen et al., 1983)	288.96
Plant available B (mg Kg^{-1} soil)	Atomic Absorption Spectroscopy (AAS) method (Wolf, 1971)	1.40
Soil pH	Beckman Glass Electrode pH meter (Cottenie et al., 1982)	4.9

Experimental plants were grown in a glasshouse at Khumaltar, Lalitpur, Nepal (27.6644° N, 85.3188° E) from August 2017 to January 2018 (the mean daily temperature and relative humidity during the growing period were 21^{o}C and 70% respectively, Figure 1). From the cauliflower production site, an adequate quantity of soil was collected for the pot experiment. After air-drying, the soil was passed through 4 mm sieve.

Treatment details

In this experiment, we had five fertilizer treatments (Table 2) with four replications. Plastic pots (Inner diameter- 28 cm at the top, 8 cm at base; height- 25 cm) with two drainage holes were filled with 8.5 kg of air-dried soil. Different ratios of fertilizers were mixed with soil except for control to attain desired treatments. The treatment details have been presented in table 2. N, P and K were applied as urea, single super phosphate and murate of potash respectively. Full amounts of P and K were applied before seed sowing while N was applied in three equal splits – before sowing, 20, and 35 days after sowing (DAS). B levels were applied at the rate of 1.1, 2.2, and 4.4 kg ha^{-1} through Borax ($Na_2B_4O_7 \cdot 10H_2O$, 11% B) before sowing.

Table 2. Different levels of nutrient inputs used in the experiment

Treatments	Explanation
Control	No inputs of nutrients
NPK	Inputs of $N:P_2O_5:K_2O$ (200:28.6:80 kg ha^{-1})
NPK + $B_{1.1}$	Inputs of $N:P_2O_5:K_2O$ (200:28.6:80 kg ha^{-1}) + B (1.1 kg ha^{-1})
NPK + $B_{2.2}$	Inputs of $N:P_2O_5:K_2O$ (200:28.6:80 kg ha^{-1}) + B (2.2 kg ha^{-1})
NPK + $B_{4.4}$	Inputs of $N:P_2O_5:K_2O$ (200:28.6:80 kg ha^{-1}) + B (4.4 kg ha^{-1})

Nine seeds of cauliflower (Var. *Rami*) at 8 cm spacing were directly sown at the depth of 4 cm from the soil surface in pots (pot size- 28 cm at the rim, 8 cm at base and height 25 cm, dry soil pot^{-1}- 8.5 Kg, field capacity (FC) of soil by weighing method 44.3%) on September 25, 2017, and the pots were completely randomized. Then, soil moisture was increased to 70% of the FC. Soil moisture was monitored frequently by weighing the plants with pot, and water loss due to evapotranspiration was replaced. Manual weeding was done at 40, 60, and 90 DAS. Only one healthy plant pot^{-1} was maintained at 21DAS. Rogar @2ml/ltr was applied once at 73 DAS against aphids. *Earthing up* was done to provide support to the plant before the curd initiation stage at 70 DAS. Manual harvesting of the crop was done by uprooting the crop at a fully mature stage at 123 DAS.

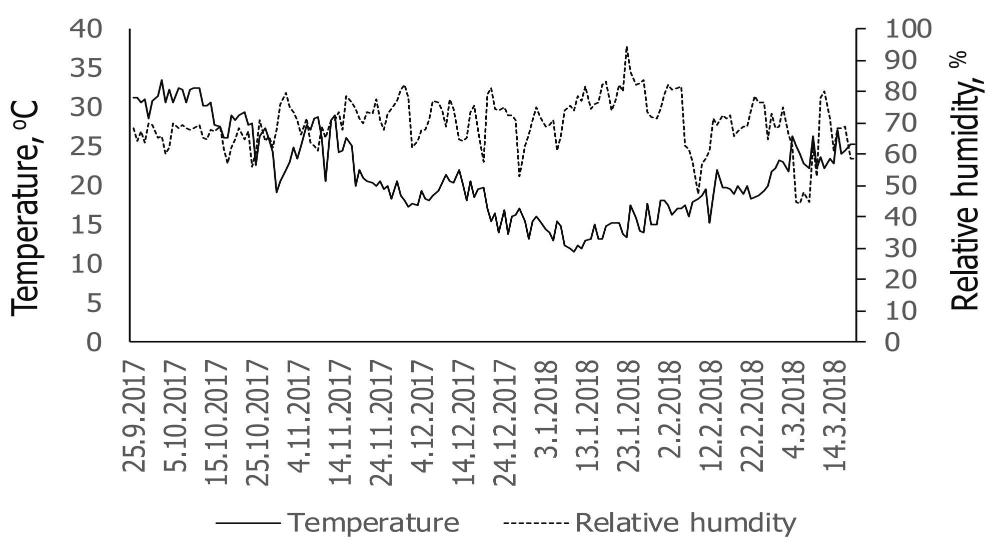

Figure 1. Average daily temperature and relative humidity during day hours inside glasshouse during the experiment at Khumaltar, Lalitpur

Measurements

Plant growth

Daily observations were done for deficiency symptoms of B in plants. After harvest, fresh curd biomass, curd diameter, and root length were recorded. Individual curd with attached leaves was separated from the shoot and was dried in the oven (48 hours at 65^0C).

Tissue boron

The finely ground (0.25 mm) leaf and stem, and curd were ashed in a muffle furnace at 550^0C for one hour and B was extracted with 0.36 N sulfuric acid (H_2SO_4) in Soil Management Directorate, Hariharbhawan, Lalitpur by using Azomethine-H method (Gaines & Mitchell, 1979), and plant available B was measured using AAS. B content of respective parts was calculated by multiplying B concentration and dry weight of the specific plant parts. From the total shoot B, the sharing between curd and the remaining parts was calculated and expressed in percentage.

Soil residual B

Similarly, plant-available B concentration in the soil after crop harvest was extracted by using hot-water calcium chloride ($CaCl_2$) extraction method (Parker & Gardner, 1981) and B level was measured in AAS.

Statistical analysis

The collected data were entered into the spreadsheets in Microsoft Excel program Windows version 2016. Simple correlations were performed between the yield and yield attributes and B uptake with B inputs. One-way analysis of variance for the effects of added B on curd yield and associated traits, shoot B content and its sharing between curd and vegetative parts in cauliflower plant was performed using STAR. The treatment effects were compared by using Duncan's multiple range test (DMRT) at a 5% level of significance.

RESULTS AND DISCUSSION

Effect on growth and curd yield

Soil application of B did not change the root length, but it significantly increased the curd and associated traits in cauliflower ($p<0.05$) (Table 3). The average length of the root appeared to be 12.2 cm. The value for the total dry matter of cauliflower was highest (18.48 g plant^{-1}) when it was grown with soil application of 1.1 kg B ha^{-1} and it did not significantly differ with B levels of 2.2 and 4.4 kg ha^{-1}. The total dry matter was strongly related to the B input (R^2=0.87). Thakur et al. (1991) and Mengel and Kirby (1978) have also reported similar kinds of results. In plants, though B plays an important role in cell division and elongation (Ahmad et al., 2012), high B often leads to the accumulation of a toxic amount of B that reduces plant growth (Nable et al., 1990). Cauliflower cultivated under soil application of 2.2 kg B ha^{-1} showed a higher curd diameter (78.25 cm) than other levels of nutrient inputs (Table 3). The fresh weight of curd was higher at 1.1 kg B ha^{-1} input and it did not significantly differ with the higher levels of B inputs. The curd yield appeared to be strongly related to B application (R^2=0.91). Similar findings were reported by Hassan et al. (2018) where maximum curd diameter was obtained at 2 Kg B ha^{-1} application.

Table 3. Effect of boron application in soil on root length, total biomass, curd diameter, curd weight, total curd yield of cauliflower and residual boron in the soil

Treatments	Root length (cm)	Total biomass (g plant^{-1})	Curd diameter (cm)	curd weight (g plant^{-1})	Residual soil B (mg Kg^{-1})
Control	11a	69.97c	40.34d	34.30b	0.26b
NPK	11.62a	83.53b	53.48c	39.65b	0.21c
NPK + $B_{1.1}$	13.25a	109.07a	72.12b	68.22a	0.26b
NPK + $B_{2.2}$	13.38a	111.03a	78.25a	65.4 a	0.22b
NPK+ $B_{4.4}$	11.5a	106.05a	69.08b	65.05a	0.32a
Sig.	*ns*	*	*	*	***

*Mean values in the columns followed by the same lower case letter do not differ significantly by Duncan's multiple range test (n=4, α=0.05). ns indicates non-significant and asterisks indicates statistically significant(*P ≤ 0.05, ***P ≤ 0.001).*

Neupane et al. (1993) observed that input of 1.65 kg B ha^{-1} was optimum in producing higher curd biomass and reducing its internal browning. A higher value of weight and diameter of the curd was recorded in the plot that received 1.5 Kg B ha^{-1} than other levels (Singh et al., 2014). The increase in size and weight of cauliflower curd by B application may be due to its role in enhancing the translocation of carbohydrates from the site of its synthesis to the storage tissue in the curd (Jana, 2002; Kumar & Chaudhary, 2002; Luiz Carlos et al., 2005; Sharma, 2002; Singh, 2003).

Effects on shoot boron uptake and its partitioning

We found that an increasing amount of soil B application significantly increased the content of shoot B (p<0.001) (**Figure 2**).

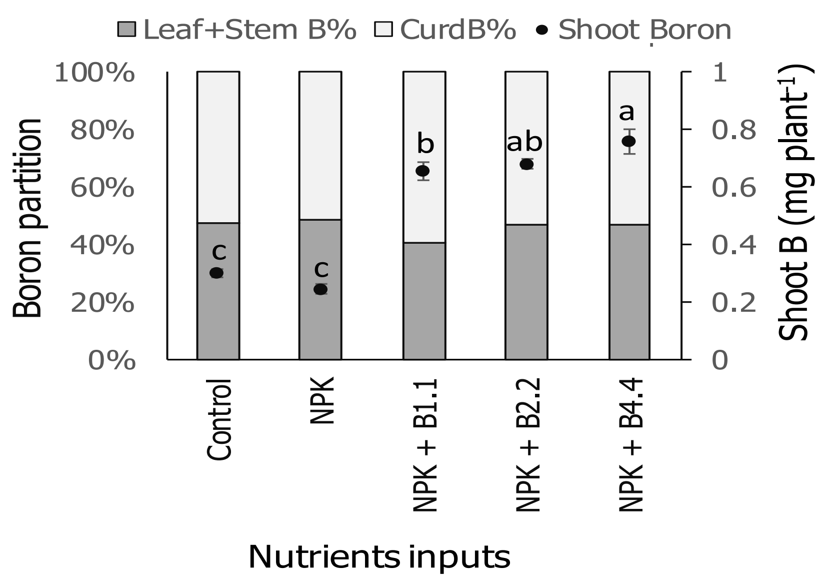

Figure 2. Effect of different levels of boron on shoot boron content and its partitioning to curd and vegetative parts in cauliflower grown under maize-based soil

Bars indicate standard error of the mean for the shoot boron content (n=4, α=0.05)

Cauliflower plant expressed a higher amount of shoot B (0.76 mg) when grown with soil application of 4.4 kg B ha^{-1} compared to 0.25 mg B per plant at the control of no B application. The shoot B content was strongly correlated with the soil B input (R^2=0.99). It indicated that the amount of B present in the soil was not enough to meet the demand for plant growth. The corresponding increase in B uptake due to soil B application has been reported by several investigators (Batabyal et al., 2015; Dhakal et al., 2014; Shorrocks, 1997). The highest level of B content in leaf tissues and curd of cauliflower was recorded when 2 kg B ha^{-1} was applied (Kumar & Chaudhary, 2002). B content of the plant was found to increase with increasing levels of hot-water-soluble B in the soil (Bucher, 1957). Soil B application also significantly influenced the sharing of B for vegetative and reproductive parts of the shoot ($p<0.001$). Soil application of 1.1 kg B ha^{-1} has shown significantly higher sharing of B towards the curd, but it tended to decline thereafter. This decline in the share of curd B at higher B input corresponded to the decline in total dry matter.

Effects on curd boron concentration

B application significantly affected the concentration of B in the curd compared to the no B application ($p<0.001$) (Figure 3).

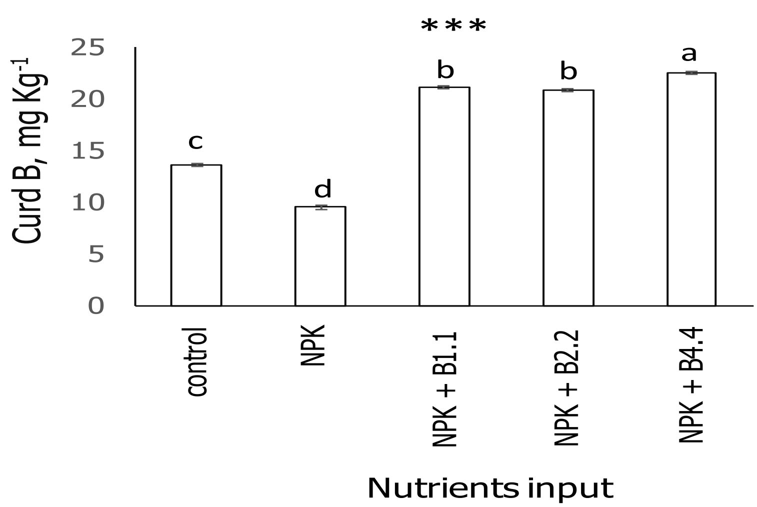

Table 3. Effect of different levels of B inputs in soil on B concentrations in curd of cauliflower. *Bars indicate the standard error of the mean. The same lower case letter above the bars indicates that the treatment means were not significant by Duncan's multiple range test (n=4). *** indicates that the treatment means were statistically significant at p<0.001).*

The results showed an increase in B concentration of curd with an increasing level of B on the soil. B concentration in curd was highest (22.51 mg kg^{-1}) for B level of 4.4 kg ha^{-1}. B application rate had a significant influence on B concentration of curd of cauliflower (Batabyal et al., 2015). Application of recommended dose of NPK reduced the B concentration in the curd compared to the control of no nutrient application. The application of these macronutrients may enhance the growth of the curd, but the rate of increase in B uptake by the plant root could be proportionally lower than the rate of the plant growth (Shrestha et al., 2021). Further, poor B fertility is likely to increase such B dilution effect in the curd.

Effects on soil residual boron

The applied B remarkably increased residual B concentration in soil ($p<0.001$) (Table 3). A significantly higher amount of soil B (0.3237 mg kg^{-1}) was extracted in B level of 4.4 kg B ha^{-1} as a residual after cauliflower harvest as compared to all other nutrient inputs. B content less than 0.5 mg kg^{-1} soil is considered critical for crop growth (Berger & Truog, 2002). Application of B to the soil for cauliflower production can increase the residual B in soil (Batabyal et al., 2015; Dhakal et al., 2014) which is important for the growth and development of follow-up crops. Compared to the control treatment, stimulation of the biomass growth by soil NPK enrichment might have enhanced the B uptake and consequently causing lower soil B.

Visual symptoms of nutrient deficiency

In control treatment of no B application, plants showed browning of curd from 90 DAS and persisted till the curd maturation stage (Figure 4). The absence of B also induces brittling of leaves with no curd initiation. B related deficiency symptoms were absent in plants grown in B enriched soil.

Figure 4. Effect of boron application on visual symptoms in cauliflower-browning of curd in control of no nutrient application (left) and without symptom at 1.1Kg B ha^{-1} input (right)

CONCLUSION

The study showed that the application of B increases the plant dry matter and curd yield of cauliflower grown at the upland condition. Soil B input increases the B contents in the above-ground parts. In our study, the application of B above 1.1 kg ha^{-1} did not increase the dry matter and curd yield but increased the B concentration. Hence, B application in upland soils of hilly regions of Nepal plays an important role to overcome the B limitation for achieving improved growth and crop quality of cauliflower as well as soil B status.

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