



## Research article

# Integrated effect of inorganic and organic nitrogen sources on nutrient uptake and crop quality of broccoli

Md. Jamal Hussain<sup>a,\*</sup>, AJM Sirajul Karim<sup>b</sup>, ARM Solaiman<sup>b</sup>, Md. Shafiqul Islam<sup>b</sup>, Mizanur Rahman<sup>c</sup><sup>a</sup> Soil and Water Management Section, Horticulture Research Centre, Bangladesh Agricultural Research Institute, Gazipur 1701, Bangladesh<sup>b</sup> Department of Soil Science, Bangabandhu Sheikh Mujibur Rahman Agricultural University, Gazipur 1706, Bangladesh<sup>c</sup> Department of Horticulture, Bangabandhu Sheikh Mujibur Rahman Agricultural University, Gazipur 1706, Bangladesh

## Article Info

### Article history:

Received 29 October 2019

Revised 23 August 2020

Accepted 9 October 2020

Available online 26 February 2021

### Keywords:

Broccoli,  
Head quality,  
Nutrient uptake,  
Organic manure,  
Urea super granules (USG)

## Abstract

The integrated effect was investigated of inorganic and organic nitrogen sources on nutrient uptake and crop quality of broccoli at Bangabandhu Sheikh Mujibur Rahman Agricultural University, Gazipur, Bangladesh. The experiment was designed based on 24 treatment combinations (integrated plant nutrition system [IPNS]-based) with four doses of inorganic nitrogen sources at 140, 160 and 180 kg N/ha as urea super granules (USG) and 180 kg N/ha as prilled urea (PU) in combination with two levels of three organic nitrogen sources being 1) 1 t/ha and 2 t/ha mustard oil cake (OC); 2) 2 t/ha and 3 t/ha poultry manure (PM) and 3) 3 t/ha and 5 t/ha cow dung (CD). The results indicated that the single-photon avalanche diode (SPAD) value, nutrient uptake, nitrogen use efficiency and the head quality (compactness co-efficient, vitamin C,  $\beta$ -carotene and chlorophyll content) of broccoli were superior following USG-organic manure integration compared to PU-organic manure. Integration of USG-organic manure increased the head compactness with the highest compactness coefficient (21.92) from USG-N<sub>160</sub>×OC<sub>2</sub>. Almost all the parameters such as nutrient (N, P, K, S) uptake and head quality had increased values with increasing rates of inorganic and organic fertilizer; however, N use efficiency and vitamin C contents were slightly decreased with increasing levels of inorganic and organic fertilizer. Maximum N uptake (208.33 kg/ha) was produced with USG-N<sub>180</sub>×OC<sub>2</sub> followed by USG-N<sub>160</sub>×OC<sub>2</sub> (204.30 kg/ha), but the maximum N use efficiency (92.89%) was obtained with USG-N<sub>140</sub>×OC<sub>2</sub> followed by USG-N<sub>160</sub>×OC<sub>2</sub> (89.06%). The highest vitamin C of 87.28 mg/100 g fresh weight (FW) and  $\beta$ -carotene content of 0.393 mg/100 g FW were produced with both USG-N<sub>160</sub>×PM<sub>3</sub> and USG-N<sub>180</sub>×PM<sub>3</sub>.

## Introduction

Broccoli is one of the nutritious vegetables containing substantial quantities of protein, carbohydrates, phosphorus, calcium, iron, thiamine, riboflavin and niacin with very high levels of ascorbic acid and carotene (Thomson and Kelly, 1985). Generally, the nutrient

content and post-harvest quality of broccoli are influenced by the application of different levels of N fertilizer. For example, Yoldas et al. (2008) reported that the application of N fertilizer increased N, P, K and Fe concentrations in broccoli heads. Everaarts and Willigen (1999) found that deep placement of N positively influenced N uptake. Chao-Jionget al. (2010) reported that the concentrations of ascorbic acid and glucoraphanin in the broccoli floret and stem decreased with an incremental rate of N application. Nitrogen also influences

\* Corresponding author.

E-mail address: [hussainmdjamal@gmail.com](mailto:hussainmdjamal@gmail.com) (M.J. Hussain)

online 2452-316X print 2468-1458/Copyright © 2021. This is an open access article, production and hosting by Kasetsart University of Research and Development Institute on behalf of Kasetsart University.

<https://doi.org/10.34044/j.anres.2021.55.1.10>

the chlorophyll content and single-photon avalanche diode (SPAD) value of broccoli. Kapotis et al. (2003) reported that leaf greenness is closely related to leaf chlorophyll (leaf N content) and there was a proportional relationship between SPAD values and leaf chlorophyll content. Wang et al. (2004) also observed a good correlation of SPAD values with both the chlorophyll content and N status of the ornamental plant peace lily. Thus, the assessment of plant nutrient and quality for selection of appropriate N doses is important in quality broccoli production. Therefore, the present study was undertaken to assess the combined effect of urea (super granules [USG]) and 1 dose of prilled urea (PU)] along with different organic sources of N [OC, PM and CD] on the nutrient uptake and N use efficiency of broccoli and the crop quality and to select the best combined dose of USG or PU along with different organic sources of N for quality broccoli production.

## Materials and Methods

### Experimental site

The experiment was carried out at Bangabandhu Sheikh Mujibur Rahman Agricultural University, Gazipur, Bangladesh from October 2014 to June 2015. The soil texture of the experimental field was a silty clay loam with poor physical properties, representing the agro-ecological zone Madhupur Tract (AEZ 28). Before starting the experiment, soil samples were collected from different experimental plots and their physico-chemical properties were analyzed in the laboratory and these are presented in Table 1. Samples were collected from decomposed cow dung (CD), poultry manure (PM) and mustard oil cake (OC) for chemical analysis and their analytical values are presented in Table 2.

### Experimental design

The experiment was designed based on 24 treatment combinations (integrated plant nutrition system [IPNS]-based) with three replications.

The treatments were randomly assigned based on four doses of inorganic nitrogen sources [USG at 140, 160 and 180 kg N/ha, and PU at 180 kg N/ha] with two levels of three organic nitrogen sources [1 t/ha and 2 t/ha mustard oil cake (OC); 2 t/ha and 3 t/ha poultry manure (PM), and 3 t/ha and 5 t/ha cow dung (CD)] comprising 24 (integrated plant nutrition system (IPNS)-based) treatment combinations. Specifically, the combinations were as- T<sub>1</sub>: USG-N<sub>140</sub>×OC<sub>1</sub>; T<sub>2</sub>: USG-N<sub>140</sub>×OC<sub>2</sub>; T<sub>3</sub>: USG-N<sub>140</sub>×PM<sub>2</sub>; T<sub>4</sub>: USG-N<sub>140</sub>×PM<sub>3</sub>; T<sub>5</sub>: USG-N<sub>140</sub>×CD<sub>3</sub>; T<sub>6</sub>: USG-N<sub>140</sub>×CD<sub>5</sub>; T<sub>7</sub>: USG-N<sub>160</sub>×OC<sub>1</sub>; T<sub>8</sub>: USG-N<sub>160</sub>×OC<sub>2</sub>; T<sub>9</sub>: USG-N<sub>160</sub>×PM<sub>2</sub>; T<sub>10</sub>: USG-N<sub>160</sub>×PM<sub>3</sub>; T<sub>11</sub>: USG-N<sub>160</sub>×CD<sub>3</sub>; T<sub>12</sub>: USG-N<sub>160</sub>×CD<sub>5</sub>; T<sub>13</sub>: USG-N<sub>180</sub>×OC<sub>1</sub>; T<sub>14</sub>: USG-N<sub>180</sub>×OC<sub>2</sub>; T<sub>15</sub>: USG-N<sub>180</sub>×PM<sub>2</sub>; T<sub>16</sub>: USG-N<sub>180</sub>×PM<sub>3</sub>; T<sub>17</sub>: USG-N<sub>180</sub>×CD<sub>3</sub>; T<sub>18</sub>: USG-N<sub>180</sub>×CD<sub>5</sub>; T<sub>19</sub>: PU-N<sub>180</sub>×OC<sub>1</sub>; T<sub>20</sub>: PU-N<sub>180</sub>×OC<sub>2</sub>; T<sub>21</sub>: PU-N<sub>180</sub>×PM<sub>2</sub>; T<sub>22</sub>: PU-N<sub>180</sub>×PM<sub>3</sub>; T<sub>23</sub>: PU-N<sub>180</sub>×CD<sub>3</sub> and T<sub>24</sub>: PU-N<sub>180</sub>×CD<sub>5</sub>. Other fertilizers were applied as a blanket dose at 53 kg/ha P, 83 kg/ha K, 20 kg/ha S, 2.0 kg/ha Zn, 1 kg/ha B and 0.8 kg/ha Mo, respectively in the form of triple super phosphate, muriate of potash, gypsum, boric acid, zinc oxide and sodium molybdate, respectively. Additional nutrients after obtaining nutrients from these organic sources were adjusted from inorganic sources.

The adjusted treatment combinations (IPNS-based) were: T<sub>1</sub>: N<sub>115</sub>P<sub>49</sub>K<sub>78</sub>S<sub>17</sub>Zn<sub>2</sub>B<sub>1</sub>M<sub>0.8</sub>+1 t/ha OC; T<sub>2</sub>: N<sub>89</sub>P<sub>45</sub>K<sub>73</sub>S<sub>13</sub>Zn<sub>2</sub>B<sub>1</sub>M<sub>0.8</sub>+2 t/ha OC; T<sub>3</sub>: N<sub>117</sub>P<sub>32</sub>K<sub>69</sub>S<sub>16</sub>Zn<sub>2</sub>B<sub>1</sub>M<sub>0.8</sub>+2 t/ha PM; T<sub>4</sub>: N<sub>106</sub>P<sub>22</sub>K<sub>62</sub>S<sub>14</sub>Zn<sub>2</sub>B<sub>1</sub>M<sub>0.8</sub>+3 t/ha PM; T<sub>5</sub>: N<sub>127</sub>P<sub>49</sub>K<sub>68</sub>S<sub>16</sub>Zn<sub>2</sub>B<sub>1</sub>M<sub>0.8</sub>+3 t/ha CD; T<sub>6</sub>: N<sub>118</sub>P<sub>46</sub>K<sub>58</sub>S<sub>14</sub>Zn<sub>2</sub>B<sub>1</sub>M<sub>0.8</sub>+5 t/ha CD; T<sub>7</sub>: N<sub>135</sub>P<sub>49</sub>K<sub>78</sub>S<sub>17</sub>Zn<sub>2</sub>B<sub>1</sub>M<sub>0.8</sub>+1 t/ha OC; T<sub>8</sub>: N<sub>109</sub>P<sub>45</sub>K<sub>73</sub>S<sub>13</sub>Zn<sub>2</sub>B<sub>1</sub>M<sub>0.8</sub>+2 t/ha OC; T<sub>9</sub>: N<sub>137</sub>P<sub>32</sub>K<sub>69</sub>S<sub>16</sub>Zn<sub>2</sub>B<sub>1</sub>M<sub>0.8</sub>+2 t/ha PM; T<sub>10</sub>: N<sub>126</sub>P<sub>22</sub>K<sub>62</sub>S<sub>14</sub>Zn<sub>2</sub>B<sub>1</sub>M<sub>0.8</sub>+3 t/ha PM; T<sub>11</sub>: N<sub>147</sub>P<sub>49</sub>K<sub>68</sub>S<sub>16</sub>Zn<sub>2</sub>B<sub>1</sub>M<sub>0.8</sub>+3 t/ha CD; T<sub>12</sub>: N<sub>138</sub>P<sub>46</sub>K<sub>58</sub>S<sub>14</sub>Zn<sub>2</sub>B<sub>1</sub>M<sub>0.8</sub>+5 t/ha CD; T<sub>13</sub>: N<sub>155</sub>P<sub>49</sub>K<sub>78</sub>S<sub>17</sub>Zn<sub>2</sub>B<sub>1</sub>M<sub>0.8</sub>+1 t/ha OC; T<sub>14</sub>: N<sub>129</sub>P<sub>45</sub>K<sub>73</sub>S<sub>13</sub>Zn<sub>2</sub>B<sub>1</sub>M<sub>0.8</sub>+2 t/ha OC; T<sub>15</sub>: N<sub>157</sub>P<sub>32</sub>K<sub>69</sub>S<sub>16</sub>Zn<sub>2</sub>B<sub>1</sub>M<sub>0.8</sub>+2 t/ha PM; T<sub>16</sub>: N<sub>146</sub>P<sub>22</sub>K<sub>62</sub>S<sub>14</sub>Zn<sub>2</sub>B<sub>1</sub>M<sub>0.8</sub>+3 t/ha PM; T<sub>17</sub>: N<sub>167</sub>P<sub>49</sub>K<sub>68</sub>S<sub>16</sub>Zn<sub>2</sub>B<sub>1</sub>M<sub>0.8</sub>+3 t/ha CD; T<sub>18</sub>: N<sub>158</sub>P<sub>46</sub>K<sub>58</sub>S<sub>14</sub>Zn<sub>2</sub>B<sub>1</sub>M<sub>0.8</sub>+5 t/ha CD; T<sub>19</sub>: N<sub>155</sub>P<sub>49</sub>K<sub>78</sub>S<sub>17</sub>Zn<sub>2</sub>B<sub>1</sub>M<sub>0.8</sub>+1 t/ha OC; T<sub>20</sub>: N<sub>129</sub>P<sub>45</sub>K<sub>73</sub>S<sub>13</sub>Zn<sub>2</sub>B<sub>1</sub>M<sub>0.8</sub>+2 t/ha OC; T<sub>21</sub>: N<sub>157</sub>P<sub>32</sub>K<sub>69</sub>S<sub>16</sub>Zn<sub>2</sub>B<sub>1</sub>M<sub>0.8</sub>+2 t/ha PM; T<sub>22</sub>: N<sub>146</sub>P<sub>22</sub>K<sub>62</sub>S<sub>14</sub>Zn<sub>2</sub>B<sub>1</sub>M<sub>0.8</sub>+3 t/ha PM; T<sub>23</sub>:

**Table 1** Physicochemical properties of field soil in experiment

Physical soil properties		Chemical soil properties	
Soil property	Analytical value	Soil property	Analytical value
Particle size distribution			
Sand (%)	17.8	Soil pH	5.97
Silt (%)	45.6	Organic carbon (%)	0.96
Clay (%)	36.6	Total N (%)	0.083
Soil texture	Silty clay loam	Available P (µg/g)	15.14
Bulk density (g/cc)	1.34	Exchangeable K (meq/100g soil)	0.298
Particle density (g/cc)	2.61	Available S (µg/g)	11.878
Porosity (%)	47.47	Available B (µg/g)	0.182
Field capacity (%)	28.67	CEC (meq/100g soil)	12.67

**Table 2** Nutrient status of cow dung, poultry manure and mustard oil cake

Source	Nutrient content (oven-dry basis)				
	Moisture (%)	N (%)	P (%)	K (%)	S (%)
Cow dung	41.53	1.35	1.01	0.68	0.24
Poultry litter	48.57	1.72	1.29	0.82	0.38
Mustard oil cake	18.88	5.32	0.83	0.71	0.66

$N_{167}P_{49}K_{68}S_{16}Zn_2B_1M_{0.8}+3$  t/ha CD and  $T_{24}: N_{158}P_{46}K_{58}S_{14}Zn_2B_1M_{0.8}+5$  t/ha CD. Broccoli seedlings (*Brassica oleracea* cv. Premium Crop) were transplanted in lines maintaining row-to-row and plant-to-plant distances of 0.60 m and 0.45 m, respectively.

#### Data collection

Data on different parameters were recorded at defined intervals throughout the cropping season. The leaf chlorophyll content was measured by the SPAD value calculated for each plot averaged data recorded from five randomly selected mature leaves of each plant according to Costa et al. (2003). SPAD values are directly proportional to the leaf chlorophyll content which is closely related to the leaf N content (Yamamoto et al., 2002). The instrument used to determine the SPAD value was a Minolta SPAD-502. Measurements were done only on the abaxial lateral part of the leaf in the same position at an interval of 5 days after commencing the treatment.

Leaf, stem and head samples were collected for analysis of their N, P, K and S contents to determine the nutrient contents at commercial maturity. Samples for chemical analysis were collected from five plants, selected randomly, after oven-drying at 65–70°C for 72 hr and machine grinding followed by passing through a 20-mesh sieve.

To estimate the head quality, fresh samples were collected from five randomly selected plants of each treatment for the chlorophyll content, vitamin C and  $\beta$ -carotene analyses. From the plant sample, the nitrogen content was determined following the micro-Kjeldahl method (Bremner, 1965). Phosphorus was determined using a spectrophotometric method (Fox et al., 1964) and the potassium content in the leaf, stem and head samples of broccoli were determined directly using a flame photometer (Jackson, 1973). The sulfur content of broccoli head samples was determined by adding 6N HCl to plant extract with  $BaCl_2$  as suggested by Black (1965).

#### Calculations of parameters

The biomass per plant and biomass per plot (both measured in grams) were calculated using Equations 1 and 2, respectively:

$$\text{Biomass per plant} = (\text{Total above ground biological yield from 10 plants}) / 10 \quad (1)$$

$$\text{Biomass per plot} = \text{Biomass per plant} \times \text{Number of plants at final harvest in a plot} \quad (2)$$

Where all amounts are measured in grams.

The biomass per hectare measured in kilograms per hectare was calculated using Equation 3:

$$\text{Biomass per hectare} = (\text{Biomass yield per plot}) / (\text{Plot area}) \times 10,000 / 1,000 \quad (3)$$

Where the biomass per hectare is measured in kilograms, the biomass yield per plot is measured in grams and the plot area is measured in square meters.

The nutrient uptake from the soil in kilograms per hectare was calculated using Equations 4 and 5:

$$\text{Nutrient uptake} = (\% \text{Nutrient} \times Y) / 100 \quad (4)$$

Where % Nutrient is the average percentage of nutrient content, (%) of the plant or head biomass and Y is the total dry matter production of the plant or head biomass measured in kilograms per hectare.

$$\text{Nutrient uptake} = (\text{Nutrient} \times Y) / (100 \times 10,000) \quad (5)$$

Where Nutrient is the average nutrient content of the plant or head biomass measured in micrograms per gram and Y is the total dry matter production of the plant or head biomass measured in kilograms per hectare.

The nitrogen use efficiency (NUE) was determined by the ratio of N in the crop at harvest compared to the N applied by subtracting the uptake made by the control plot. The efficiency of applied fertilizer N was quantified using Equation 6 (Craswell and Godwin, 1984):

$$\text{Nitrogen use efficiency} = (\text{N uptake F} - \text{N uptake C}) / (\text{N fertilizer applied}) \times 100 \quad (6)$$

Where F and C denote the fertilized crop and unfertilized control, respectively.

The head compactness coefficient (CC) was estimated using Equation 7:

$$\text{Compactness coefficient} = \text{Head yield} / \text{Head diameter} \quad (7)$$

Where the head yield is measured in grams per plant and the head diameter is measured in centimeters.

The ascorbic acid (vitamin C) content of the fresh head sample was determined in milligrams per 100 grams fresh weight using a centrifuge technique and the  $KIO_3$  titration method using Equation 8 (Samotus et al., 1982):

$$\text{Ascorbic acid (mg/100 g Fresh weight)} = (f \times V_1 \times V_2 \times 100) / (W \times V_3) \quad (8)$$

Where f is the dye factor

$V_1$  is the titrated volume of  $KIO_3$  (ml),  $V_2$  is total volume of the blended sample (100 ml),  $V_3$  is the volume of sample extract taken (5 ml), W is the weight of fresh head sample (20 g).

Chlorophyll-a, chlorophyll-b and  $\beta$ -carotene were determined according to the acetone-hexane method of Nagata and Yamashita (1992). All pigments in samples were extracted immediately with acetone-hexane (4:6). Then the optical density of the supernatant was measured at 663 nm, 645 nm, 505 nm and 453 nm using a spectrophotometer. From these values, the contents of chlorophyll-a, chlorophyll-b and  $\beta$ -carotene (all measured in milligrams per 100 milliliters) were estimated using Equations 9, 10 and 11:

$$\text{Chlorophyll a} = 0.999A_{663} - 0.999A_{645} \quad (9)$$

$$\text{Chlorophyll b} = 0.328A_{663} - 1.77A_{645} \quad (10)$$

$$\beta - \text{Carotene} = 0.216A_{663} - 1.22A_{645} - 0.304A_{505} + 0.452A_{453} \quad (11)$$

Where  $A_{663}$ ,  $A_{645}$ ,  $A_{505}$  and  $A_{453}$  are the absorbances at 663 nm, 645 nm, 505 nm and 453 nm, respectively.

#### Statistical analysis

The collected data were analyzed based on analysis of variance using the statistical package MSTAT-C (Gomez and Gomez, 1984). Means of the 24 combined treatments were calculated and compared using Duncan's multiple range test (DMRT) at 95% confidence interval.

## Results and Discussion

### *Integrated effect of inorganic and organic sources of N on single-photon avalanche diode value of broccoli*

The SPAD value was significantly affected from 30 days after transplanting (DAT) by the different levels of USG and PU with organic sources of N (Table 3). At 30 DAT, the highest SPAD value (62.30) was recorded in the PU-N<sub>180</sub>×OC<sub>2</sub> treatment followed by USG-N<sub>180</sub>×OC<sub>2</sub> (60.50) and at 35 DAT the highest SPAD value (63.97) was recorded in PU-N<sub>180</sub>×OC<sub>2</sub> which was followed by USG-N<sub>180</sub>×OC<sub>2</sub> (62.57). At 40 DAT, the highest SPAD value (66.17) was recorded in both the treatments PU-N<sub>180</sub>×OC<sub>2</sub> and USG-N<sub>180</sub>×OC<sub>2</sub> which was followed by USG-N<sub>140</sub>×OC<sub>2</sub> (65.77) and at 45 DAT, it was highest (69.83) in USG-N<sub>180</sub>×OC<sub>2</sub> which was followed by PU-N<sub>180</sub>×OC<sub>2</sub> (69.73). An almost similar trend was observed in the SPAD readings from 50 DAT to 70 DAT (Table 4). At 70 DAT, the highest SPAD value (76.97) was recorded with USG-N<sub>180</sub>×OC<sub>2</sub> which was followed by USG-N<sub>160</sub>×OC<sub>2</sub> (76.90). It was observed that SPAD values were increased with increasing levels of nitrogenous fertilizer and the highest value was obtained with the USG-N<sub>180</sub>×OC<sub>2</sub> treatment, which indicated the superiority of USG to PU. This might have been due to sufficient available N supply from the organic matter and USG. The higher supply of N might have increased the leaf chlorophyll content which might have increased the SPAD value. This result was supported by the findings of Yamamoto et al. (2002) and Kapotis

et al. (2003) as they reported that SPAD values are proportional to the leaf chlorophyll content. Varvel et al. (1997) also demonstrated that N fertilizer significantly increased both corn grain yield and SPAD readings.

### *Nitrogen uptake*

The nitrogen uptake by the broccoli plants was significantly influenced by the different organic and inorganic sources of N. The highest N uptake was recorded with treatment USG-N<sub>180</sub>×OC<sub>2</sub> (208.33 kg/ha) followed by USG-N<sub>160</sub>×OC<sub>2</sub> (204.30 kg/ha), as shown in Fig. 1. However, USG-N<sub>180</sub>×PM<sub>3</sub> (202.84 kg/ha), PU-N<sub>180</sub>×OC<sub>2</sub> (199.83 kg/ha), USG-N<sub>180</sub>×CD<sub>5</sub> (198.08 kg/ha) and PU-N<sub>180</sub>×PM<sub>3</sub> (193.26 kg/ha) had higher levels of N uptake. It was observed that the N uptake increased with increasing levels of N applied and that this was higher in the USG-applied plots with mustard oil cake than for PU. This might have been due to the high availability and continuous supply of N with the deep-placed USG in combination with mustard oil cake. Organic manure might act as a slow-release N source which can supply N for a long time to the plant, resulting the plant having a higher N uptake. This result was supported by the findings of Rickard (2008) who reported that the plant N uptake was strongly affected by N application. Tremblay et al. (2001) also showed that the N uptake was approximately 260 kg N/ha for an average yield of field vegetables. Everaarts and Willigen (1999) reported a maximum N uptake of 300 kg N/ha for broccoli.

**Table 3** Integrated effect of different levels of inorganic and organic sources of N on single-photon avalanche diode values at different days after transplanting (DAT)

Treatment	25DAT	30DAT	35DAT	40DAT	45DAT
USG-N <sub>140</sub> ×OC <sub>1</sub>	55.43	57.13 <sup>bcd</sup>	58.70 <sup>figh</sup>	63.13 <sup>cdef</sup>	65.97 <sup>fg</sup>
USG-N <sub>140</sub> ×OC <sub>2</sub>	57.07	58.20 <sup>abcd</sup>	60.87 <sup>bcd</sup>	65.77 <sup>ab</sup>	67.70 <sup>bcd</sup>
USG-N <sub>140</sub> ×PM <sub>2</sub>	52.87	53.83 <sup>c</sup>	58.17 <sup>ghi</sup>	62.80 <sup>def</sup>	66.77 <sup>efg</sup>
USG-N <sub>140</sub> ×PM <sub>3</sub>	57.47	56.50 <sup>bcd</sup>	59.70 <sup>defg</sup>	63.93 <sup>bcd</sup>	68.70 <sup>abcde</sup>
USG-N <sub>140</sub> ×CD <sub>3</sub>	55.30	55.20 <sup>de</sup>	56.40 <sup>i</sup>	62.20 <sup>f</sup>	65.40 <sup>g</sup>
USG-N <sub>140</sub> ×CD <sub>5</sub>	57.07	57.53 <sup>bcd</sup>	60.20 <sup>bcd</sup>	63.10 <sup>cdef</sup>	67.70 <sup>bcd</sup>
USG-N <sub>160</sub> ×OC <sub>1</sub>	56.00	56.73 <sup>bcd</sup>	60.10 <sup>cdef</sup>	62.37 <sup>ef</sup>	68.43 <sup>abcde</sup>
USG-N <sub>160</sub> ×OC <sub>2</sub>	56.50	57.83 <sup>bcd</sup>	61.30 <sup>bcd</sup>	65.60 <sup>ab</sup>	69.17 <sup>abc</sup>
USG-N <sub>160</sub> ×PM <sub>2</sub>	57.30	55.90 <sup>de</sup>	59.07 <sup>efgh</sup>	62.90 <sup>def</sup>	66.73 <sup>efg</sup>
USG-N <sub>160</sub> ×PM <sub>3</sub>	58.17	59.33 <sup>abcd</sup>	60.90 <sup>bcd</sup>	65.07 <sup>abcd</sup>	68.60 <sup>abcde</sup>
USG-N <sub>160</sub> ×CD <sub>3</sub>	57.67	53.93 <sup>c</sup>	57.27 <sup>hi</sup>	62.77 <sup>def</sup>	67.23 <sup>cdefg</sup>
USG-N <sub>160</sub> ×CD <sub>5</sub>	54.90	59.30 <sup>abcd</sup>	60.27 <sup>bcd</sup>	64.73 <sup>abcd</sup>	67.73 <sup>bcd</sup>
USG-N <sub>180</sub> ×OC <sub>1</sub>	57.77	58.70 <sup>abcd</sup>	61.93 <sup>abcd</sup>	64.20 <sup>abcde</sup>	69.03 <sup>abcd</sup>
USG-N <sub>180</sub> ×OC <sub>2</sub>	59.60	60.50 <sup>ab</sup>	62.57 <sup>ab</sup>	66.17 <sup>a</sup>	69.83 <sup>a</sup>
USG-N <sub>180</sub> ×PM <sub>2</sub>	52.97	57.77 <sup>bcd</sup>	59.13 <sup>efgh</sup>	63.43 <sup>cdef</sup>	68.93 <sup>abcd</sup>
USG-N <sub>180</sub> ×PM <sub>3</sub>	55.47	58.20 <sup>abcd</sup>	62.43 <sup>abc</sup>	64.37 <sup>abcde</sup>	69.00 <sup>abcd</sup>
USG-N <sub>180</sub> ×CD <sub>3</sub>	54.17	56.23 <sup>bcd</sup>	58.23 <sup>ghi</sup>	62.83 <sup>def</sup>	67.07 <sup>defg</sup>
USG-N <sub>180</sub> ×CD <sub>5</sub>	57.17	57.93 <sup>bcd</sup>	59.37 <sup>efgh</sup>	64.07 <sup>abcde</sup>	68.10 <sup>abcde</sup>
PU-N <sub>180</sub> ×OC <sub>1</sub>	59.23	60.07 <sup>abc</sup>	61.17 <sup>bcd</sup>	64.70 <sup>abcd</sup>	68.27 <sup>abcde</sup>
PU-N <sub>180</sub> ×OC <sub>2</sub>	61.27	62.30 <sup>a</sup>	63.97 <sup>a</sup>	66.17 <sup>a</sup>	69.73 <sup>ab</sup>
PU-N <sub>180</sub> ×PM <sub>2</sub>	56.83	58.17 <sup>abcd</sup>	59.67 <sup>defg</sup>	63.47 <sup>cdef</sup>	68.47 <sup>abcde</sup>
PU-N <sub>180</sub> ×PM <sub>3</sub>	57.17	58.37 <sup>abcd</sup>	60.07 <sup>cdefg</sup>	64.60 <sup>abcd</sup>	69.03 <sup>abcd</sup>
PU-N <sub>180</sub> ×CD <sub>3</sub>	56.80	58.80 <sup>abcd</sup>	61.03 <sup>bcd</sup>	63.27 <sup>cdef</sup>	68.97 <sup>abcd</sup>
PU-N <sub>180</sub> ×CD <sub>5</sub>	58.43	59.13 <sup>abcd</sup>	61.27 <sup>bcd</sup>	64.83 <sup>abcd</sup>	68.67 <sup>abcde</sup>
CV (%)	4.03	3.72	2.09	2.01	1.53

CV = coefficient of variation;

Means in a column superscripted by different lowercase letters are significantly different ( $p < 0.05$ ). Treatments are detailed in the experimental design section.

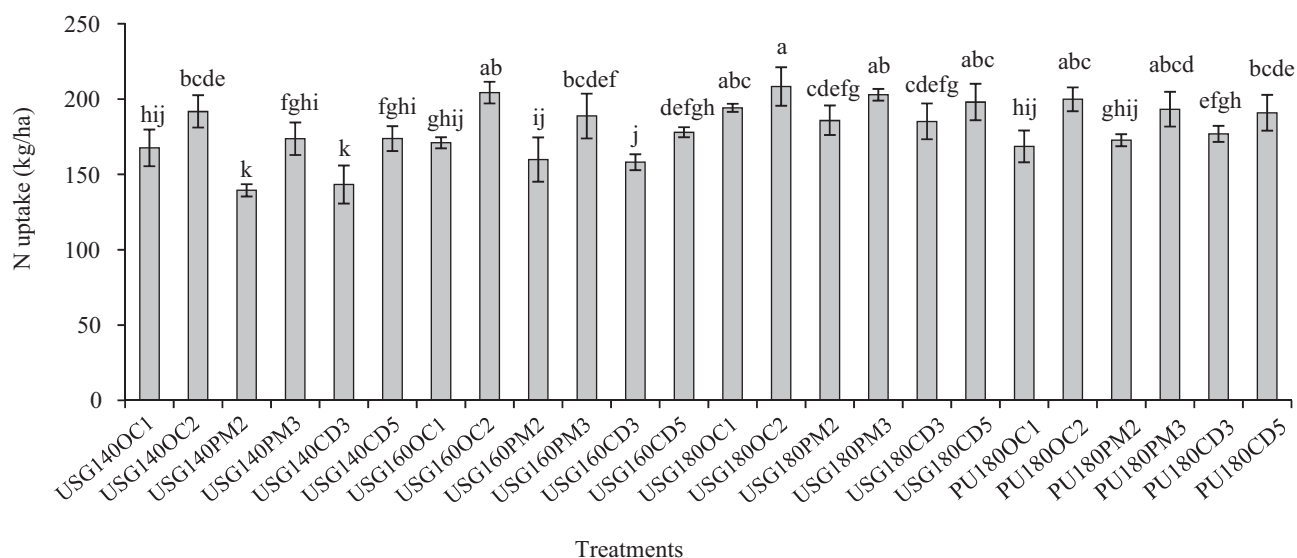
**Table 4** Integrated effect of different levels of inorganic and organic sources of N on single-photon avalanche diode values at different days after transplanting (DAT)

Treatment	50DAT	55DAT	60DAT	65DAT	70DAT
USG-N <sub>140</sub> ×OC <sub>1</sub>	70.97 <sup>efg</sup>	73.67 <sup>efgh</sup>	75.43 <sup>defghi</sup>	73.83 <sup>efgh</sup>	71.83 <sup>defg</sup>
USG-N <sub>140</sub> ×OC <sub>2</sub>	72.03 <sup>bcdef</sup>	75.23 <sup>abcdef</sup>	76.90 <sup>bcd</sup>	75.23 <sup>cde</sup>	74.27 <sup>abcd</sup>
USG-N <sub>140</sub> ×PM <sub>2</sub>	69.47 <sup>g</sup>	71.60 <sup>i</sup>	74.50 <sup>ghi</sup>	72.70 <sup>ghi</sup>	71.70 <sup>defg</sup>
USG-N <sub>140</sub> ×PM <sub>3</sub>	69.83 <sup>g</sup>	74.43 <sup>cdefgh</sup>	75.87 <sup>cdefgh</sup>	74.27 <sup>defg</sup>	73.30 <sup>bcd</sup>
USG-N <sub>140</sub> ×CD <sub>3</sub>	70.27 <sup>fg</sup>	72.53 <sup>hi</sup>	74.37 <sup>ghi</sup>	70.50 <sup>j</sup> 72.77 <sup>ghi</sup>	68.63 <sup>h</sup>
USG-N <sub>140</sub> ×CD <sub>5</sub>	70.90 <sup>efg</sup>	75.17 <sup>abcdef</sup>	74.87 <sup>fghi</sup>	73.37 <sup>fghi</sup>	73.20 <sup>bcd</sup>
USG-N <sub>160</sub> ×OC <sub>1</sub>	70.87 <sup>efg</sup>	75.00 <sup>bcdefg</sup>	76.90 <sup>bcd</sup>	77.03 <sup>ab</sup>	70.30 <sup>fgh</sup>
USG-N <sub>160</sub> ×OC <sub>2</sub>	74.10 <sup>ab</sup>	76.80 <sup>ab</sup>	78.57 <sup>ab</sup>	74.37 <sup>defg</sup>	76.90 <sup>a</sup>
USG-N <sub>160</sub> ×PM <sub>2</sub>	72.83 <sup>bcd</sup>	73.83 <sup>efgh</sup>	76.13 <sup>cdefg</sup>	74.90 <sup>def</sup>	72.43 <sup>cdef</sup>
USG-N <sub>160</sub> ×PM <sub>3</sub>	72.00 <sup>bcdef</sup>	74.17 <sup>cdefgh</sup>	76.73 <sup>cde</sup>	74.17 <sup>efgh</sup>	74.50 <sup>abcd</sup>
USG-N <sub>160</sub> ×CD <sub>3</sub>	71.13 <sup>defg</sup>	74.87 <sup>bcdefg</sup>	76.40 <sup>cdef</sup>	74.83 <sup>def</sup>	73.60 <sup>abcd</sup>
USG-N <sub>160</sub> ×CD <sub>5</sub>	72.10 <sup>bcdef</sup>	74.03 <sup>defgh</sup>	76.93 <sup>bcd</sup>	74.10 <sup>efgh</sup>	73.63 <sup>bcd</sup>
USG-N <sub>180</sub> ×OC <sub>1</sub>	72.70 <sup>bcd</sup>	75.47 <sup>abcde</sup>	75.67 <sup>cdefghi</sup>	77.73 <sup>a</sup>	72.90 <sup>cdef</sup>
USG-N <sub>180</sub> ×OC <sub>2</sub>	75.67 <sup>a</sup>	76.97 <sup>a</sup>	79.03 <sup>a</sup>	74.40 <sup>defg</sup>	76.97 <sup>a</sup>
USG-N <sub>180</sub> ×PM <sub>2</sub>	73.23 <sup>bcd</sup>	74.33 <sup>cdefgh</sup>	75.20 <sup>efghi</sup>	76.00 <sup>bcd</sup>	73.67 <sup>bcd</sup>
USG-N <sub>180</sub> ×PM <sub>3</sub>	73.37 <sup>bc</sup>	76.17 <sup>abc</sup>	77.07 <sup>bcd</sup>	72.93 <sup>gh</sup>	75.00 <sup>abc</sup>
USG-N <sub>180</sub> ×CD <sub>3</sub>	71.30 <sup>cdefg</sup>	74.37 <sup>cdefgh</sup>	74.27 <sup>hi</sup>	75.20 <sup>cdef</sup>	72.13 <sup>cdefg</sup>
USG-N <sub>180</sub> ×CD <sub>5</sub>	72.60 <sup>bcd</sup>	74.97 <sup>bcdefg</sup>	74.83 <sup>fghi</sup>	73.37 <sup>fgh</sup>	73.57 <sup>bcd</sup>
PU-N <sub>180</sub> ×OC <sub>1</sub>	72.13 <sup>bcdef</sup>	73.57 <sup>efgh</sup>	77.10 <sup>bcd</sup>	76.73 <sup>abc</sup>	72.10 <sup>cdefg</sup>
PU-N <sub>180</sub> ×OC <sub>2</sub>	73.47 <sup>b</sup>	76.10 <sup>abcd</sup>	77.27 <sup>bc</sup>	73.37 <sup>fgh</sup>	75.87 <sup>ab</sup>
PU-N <sub>180</sub> ×PM <sub>2</sub>	70.83 <sup>efg</sup>	73.27 <sup>fghi</sup>	74.63 <sup>fghi</sup>	73.47 <sup>efgh</sup>	70.93 <sup>efgh</sup>
PU-N <sub>180</sub> ×PM <sub>3</sub>	72.60 <sup>bcd</sup>	73.83 <sup>efgh</sup>	74.80 <sup>fghi</sup>	71.20 <sup>ij</sup>	72.67 <sup>cdef</sup>
PU-N <sub>180</sub> ×CD <sub>3</sub>	70.97 <sup>efg</sup>	72.97 <sup>ghi</sup>	73.87 <sup>i</sup>	72.40 <sup>hi</sup>	69.30 <sup>gh</sup>
PU-N <sub>180</sub> ×CD <sub>5</sub>	72.27 <sup>bcdef</sup>	73.60 <sup>efgh</sup>	74.60 <sup>ghi</sup>		71.23 <sup>efgh</sup>
CV (%)	1.51	1.42	1.22	1.28	2.04

CV = coefficient of variation;

Means in a column superscripted by different lowercase letters are significantly different ( $p < 0.05$ ).

Treatments are detailed in the experimental design section.

**Fig. 1** Integrated effect of inorganic and organic sources of N on N uptake by broccoli plants, where values are mean and error bars represent  $\pm$ SD; treatments are detailed in the experimental design section.

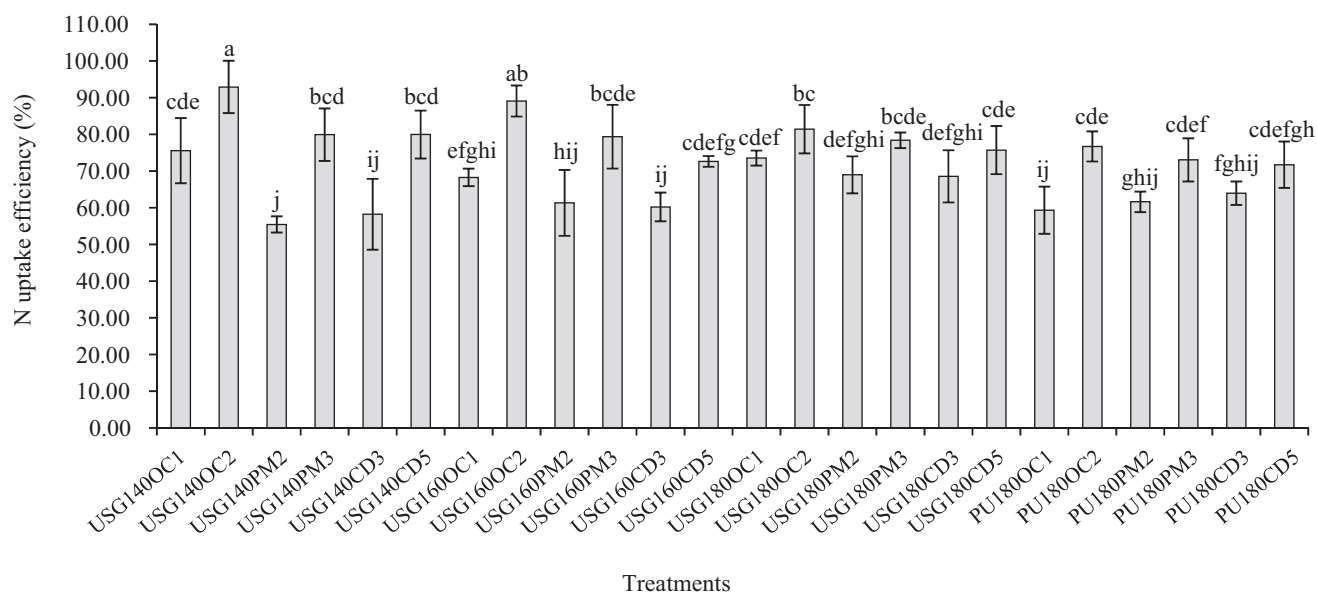
### Nitrogen use efficiency

It was speculated that N use efficiency would decrease with increasing levels of N fertilizer. The highest N use efficiency (92.89%) was obtained with USG-N<sub>140</sub>×OC<sub>2</sub> followed by USG-N<sub>160</sub>×OC<sub>2</sub> (89.06%) and the lowest N use efficiency (55.45%) was found with PU-N<sub>140</sub>×PM<sub>2</sub> (Fig. 2). USG-N<sub>160</sub>×OC<sub>2</sub> and USG-N<sub>160</sub>×PM<sub>3</sub> also showed higher N use efficiency levels. However, it was higher in the USG-treated plots compared to PU. This might have been due to the slow release but over a long time of N from the organic manure such as the mustard oilcake, cow dung and poultry litter. USG could ensure higher N use efficiency than PU with a continuous N supply and a greater recovery percentage of fertilizer-N. Similar findings were observed by Riley and Vågen (2003), where the N use efficiency of broccoli decreased with increasing amounts of fertilizer. Khalil et al. (2011) reported that USG in deeper placement (5.0–7.5 cm) resulted in greater fertilizer-N recovery (70.5–78.0%) in the crop compared to the use of prills (56.6%). An apparent fertilizer-N recovery by the aboveground part of the plant decreased linearly from between 46 to 93% with 125 kg N/ha to between 20 to 44% with 625 kg N/ha (Zebarth et al., 1995). These results were corroborated by the findings of the present study. The higher use efficiency might reflect the impact of USG with deep placement where comparatively lower numbers of nitrifying bacteria participate to convert a large granule of urea due to its smaller surface area compared to PU, thus taking a longer time to convert it to an available form that can be taken up by plants. This phenomenon occurring following deep placement of USG where a zone of concentrated urea solution was created meant that the denitrifying bacteria could not work actively and consequently the N produced there remained in the root zone for plant uptake (Mukherjee, 1986).

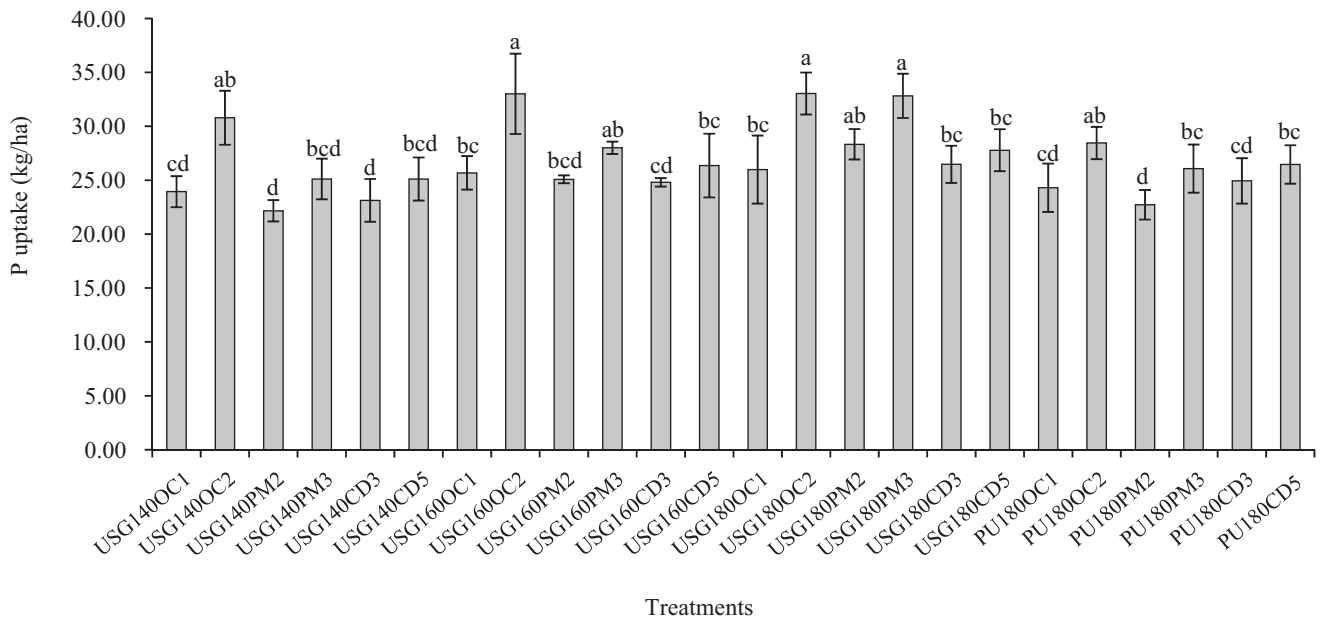
### Phosphorus and potassium uptake

The phosphorus uptake was significantly increased with increasing levels of nitrogenous fertilizer and the highest P uptake (33.04 kg/ha) was recorded with USG-N<sub>180</sub>×OC<sub>2</sub> followed by USG-N<sub>160</sub>×OC<sub>2</sub> (33.01 kg/ha) which was not significantly different from USG-N<sub>180</sub>×PM<sub>3</sub>, PU-N<sub>180</sub>×OC<sub>2</sub> and USG-N<sub>140</sub>×OC<sub>2</sub>. The lowest P uptake (22.16 kg/ha) was with USG-N<sub>140</sub>×PM<sub>2</sub>. However, the P uptake was higher in the USG-treated plants with organic manure than for PU (Fig. 3).

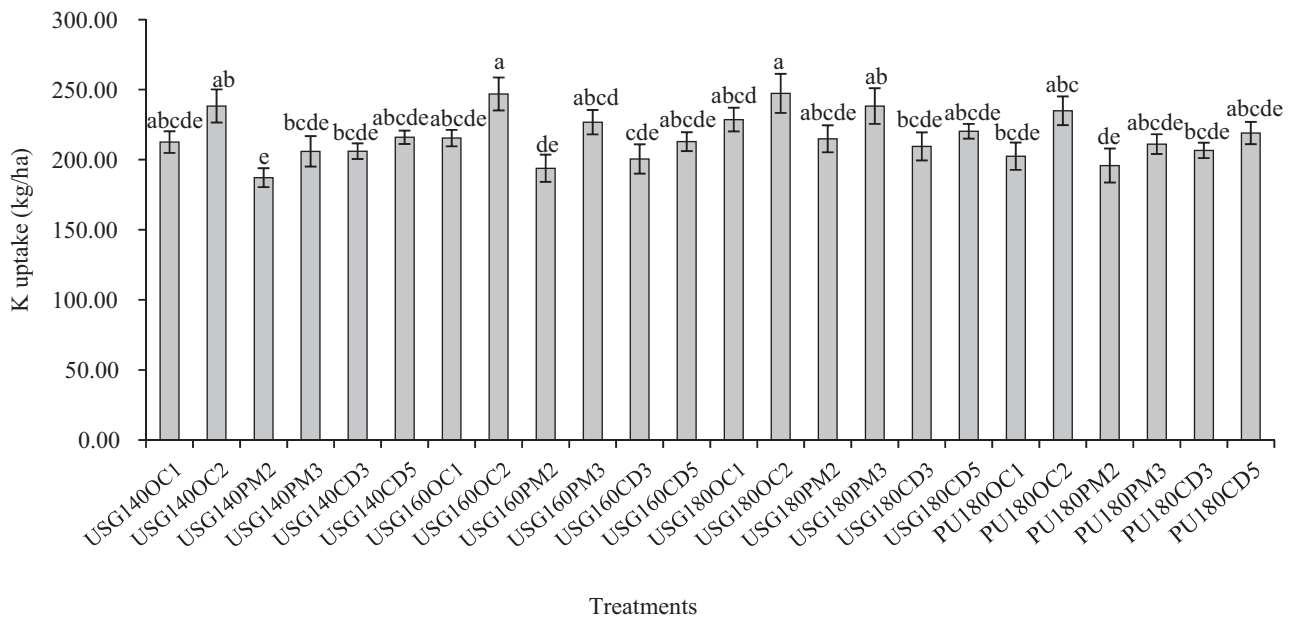
Similarly, the K uptake increased with increasing levels of nitrogenous fertilizer and the highest K uptake (247.32 kg/ha) was recorded with USG-N<sub>180</sub>×OC<sub>2</sub> followed by USG-N<sub>160</sub>×OC<sub>2</sub> (246.96 kg/ha) and both USG-N<sub>140</sub>×OC<sub>2</sub> and USG-N<sub>180</sub>×PM<sub>3</sub> (238.32 kg/ha) that were not significantly different. The lowest K uptake (187.20 kg/ha) was recorded with the treatment USG-N<sub>140</sub>×PM<sub>2</sub> (Fig. 4). This was due to the continuous supply and greater recovery of P and K fertilizer with USG with organic manure than for PU. This result was supported by Yoldas et al. (2008) who reported that application of nitrogen increased the N, P, K and Fe concentrations in broccoli heads. Similar results were obtained by Abdelrazzag (2002) and Magnusson (2002) with several vegetable crops. The possible reason for the higher P and K uptakes was the conversion of N to nitrate in the soil and the subsequent nitrate absorption by roots resulting in a negative charge in root cells with charge equilibrium being driven by cation absorption and consequently, P and K absorption by the plant increased (Moniruzzaman et al., 2007).



**Fig. 2** Integrated effect of inorganic and organic sources of N on N use efficiency by the broccoli plants, where values are mean and error bars represent  $\pm$ SD; treatments are detailed in the experimental design section.



**Fig. 3** Integrated effect of inorganic and organic sources of N on P uptake by the broccoli plants, where values are mean and error bars represent  $\pm$ SD; treatments are detailed in the experimental design section.



**Fig. 4** Integrated effect of inorganic and organic sources of N on K uptake by the broccoli plants, where values are mean and error bars represent  $\pm$ SD; treatments are detailed in the experimental design section.

### Sulfur uptake

The sulfur uptake increased significantly with increasing levels of nitrogenous fertilizer and the highest S uptake (3.45 kg/ha) was recorded in USG-N<sub>180</sub>×OC<sub>2</sub> followed by USG-N<sub>160</sub>×OC<sub>2</sub> (3.24 kg/ha) with these results not being significantly different. Here, the higher uptake was recorded in the USG-treated plots rather than for PU (Fig. 5) and there was greater S uptake from the higher amount of organic manure. This might have been due to the continuous supply and the greater amount of S recovery with USG in combination with organic manure than for PU. The higher N supply from USG synergistically induced a higher uptake of S by the crop.

### Head yield

The head yield was significantly affected by the treatment combinations and the highest head yield (14.75 t/ha) was recorded with USG-N<sub>160</sub>×OC<sub>2</sub> followed by USG-N<sub>180</sub>×OC<sub>2</sub> (14.48 t/ha), USG-N<sub>160</sub>×PM<sub>3</sub> (13.84 t/ha) and PU-N<sub>180</sub>×PM<sub>3</sub> (13.72 t/ha) which were not significantly different but were significantly higher than those of all other treatment combinations (Table 5). The lowest head yield (11.87 t/ha) was obtained from USG-N<sub>140</sub>×CD<sub>3</sub>. Here, the treatment combinations USG-N<sub>160</sub>×OC<sub>2</sub>, USG-N<sub>180</sub>×OC<sub>2</sub>, USG-N<sub>160</sub>×PM<sub>3</sub> and PU-N<sub>180</sub>×PM<sub>3</sub> produced more or less similar performance in terms of yield. This might have been due to the higher availability and continuous and uniform supply of N that caused optimum growth and induced a maximum marketable head yield than was achieved from the highest level of USG. However, mustard oilcake supplied more available N to the crop where maximum vegetative growth occurred. The low yield from crops harvested from the treatments were due to the insufficient supply of N to the plants, leading to limited carbon assimilation and resulting in a reduction in plant productivity (Lawlor,

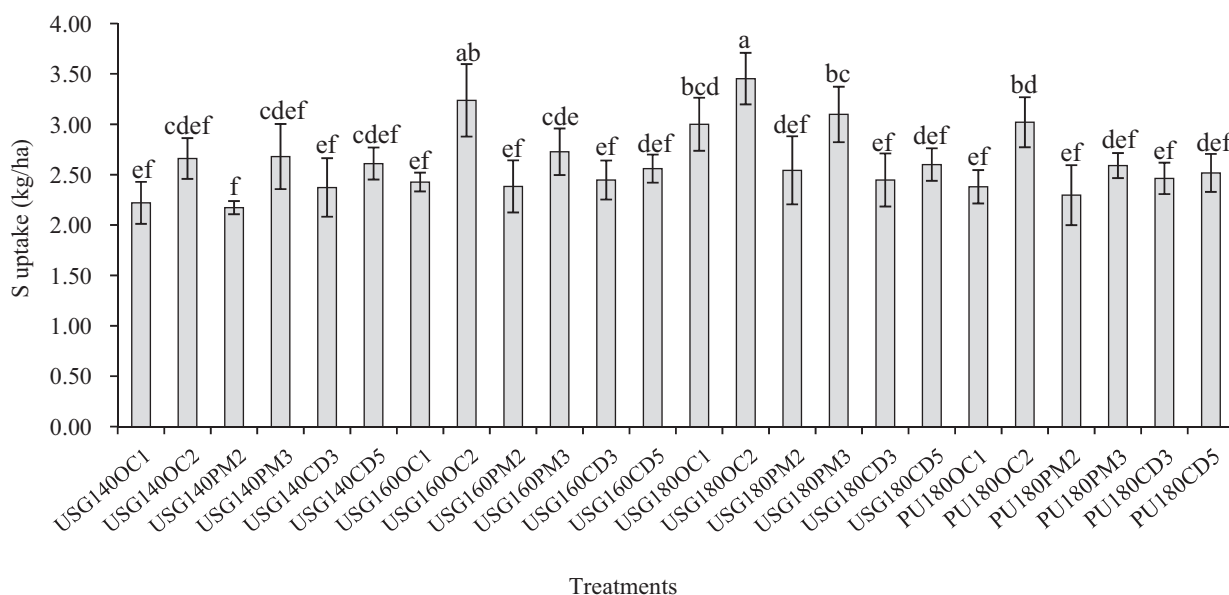
2002). This result was in agreement with Kandil and Gad (2009) who concluded that using organic manure plus inorganic solution fertilizers had a significant promotive effect on plant growth, head yield, chemical constituents and the mineral composition of broccoli. Abou El-Magdet al., (2006) found the highest total yield of broccoli for the variety ‘Premium crop’ and indicated that broccoli plants with 100% cattle manure produced the highest vegetative growth and the highest total yield, with better quality achieved by adding poultry manure. Similar results were reported by Rickard (2008) for broccoli.

### Compactness coefficient

The CC is an important quality indicator of the broccoli head. It was significantly influenced by the treatment combinations (Table 5). The highest CC (21.92) was recorded in USG-N<sub>160</sub>×OC<sub>2</sub> followed by USG-N<sub>180</sub>×OC<sub>2</sub> (21.63) and the lowest CC (19.00) was recorded with USG-N<sub>140</sub>×PM<sub>2</sub>. These results indicated that USG-organic manure might increase head compactness as well as head quality due to the higher availability and continuous, uniform supply of N resulting in higher uptake and use efficiency of N which had maximum assimilation for higher compactness and quality of broccoli head. This result was supported by the findings of Wojciechowska et al. (2005) in broccoli.

### Vitamin C and β-carotene content

The vitamin C content of broccoli head was significantly influenced by the treatment combinations (Table 5) and the highest vitamin content (87.28 mg/100g FW) was found in USG-N<sub>160</sub>×PM<sub>3</sub> followed by USG-N<sub>180</sub>×PM<sub>2</sub> (85.20 mg/100g FW). The lowest vitamin C (56.06 mg/100g FW) was noted with PU-N<sub>180</sub>×OC<sub>2</sub>. Karitonas (2001) reported that an increased level of N supply slightly reduced vitamin C from



**Fig. 5** Integrated effect of inorganic and organic sources of N on S uptake by broccoli plants, where values are mean and error bars represent  $\pm$ SD; treatments are detailed in the experimental design section.

**Table 5** Integrated effect of different forms and levels of inorganic and organic sources of N on head yield and quality of broccoli

Treatment	Head yield (t/ha)	Compactness coefficient (CC)	Vitamin-C (mg/100g FW)	$\beta$ -carotene (mg/100g FW)	Chlophyll-a (mg/100g FW)	Chlophyll-b (mg/100g FW)
USG-N <sub>140</sub> ×OC <sub>1</sub>	12.16 <sup>hi</sup>	19.43 <sup>de</sup>	74.45 <sup>cdefg</sup>	0.263 <sup>fg</sup>	0.663 <sup>i</sup>	0.885 <sup>gh</sup>
USG-N <sub>140</sub> ×OC <sub>2</sub>	13.19 <sup>bcd</sup>	20.46 <sup>bcd</sup>	72.07 <sup>cdefg</sup>	0.380 <sup>ab</sup>	0.724 <sup>def</sup>	1.015 <sup>cde</sup>
USG-N <sub>140</sub> ×PM <sub>2</sub>	11.91 <sup>i</sup>	19.00 <sup>e</sup>	82.60 <sup>abcd</sup>	0.330 <sup>cd</sup>	0.705 <sup>fg</sup>	0.840 <sup>hi</sup>
USG-N <sub>140</sub> ×PM <sub>3</sub>	12.69 <sup>defgh</sup>	19.46 <sup>de</sup>	79.96 <sup>abcde</sup>	0.327 <sup>cd</sup>	0.715 <sup>f</sup>	1.081 <sup>bc</sup>
USG-N <sub>140</sub> ×CD <sub>3</sub>	11.87 <sup>i</sup>	19.06 <sup>e</sup>	80.62 <sup>abcde</sup>	0.250 <sup>g</sup>	0.609 <sup>i</sup>	0.776 <sup>i</sup>
USG-N <sub>140</sub> ×CD <sub>5</sub>	12.95 <sup>bcd</sup>	19.79 <sup>cde</sup>	74.46 <sup>cdefg</sup>	0.380 <sup>ab</sup>	0.710 <sup>f</sup>	1.070 <sup>bcd</sup>
USG-N <sub>160</sub> ×OC <sub>1</sub>	13.42 <sup>bcd</sup>	20.03 <sup>bcd</sup>	79.81 <sup>abcde</sup>	0.260 <sup>fg</sup>	0.688 <sup>ghi</sup>	0.844 <sup>hi</sup>
USG-N <sub>160</sub> ×OC <sub>2</sub>	14.75 <sup>a</sup>	21.92 <sup>a</sup>	75.38 <sup>bcd</sup>	0.387 <sup>ab</sup>	0.747 <sup>abc</sup>	1.143 <sup>a</sup>
USG-N <sub>160</sub> ×PM <sub>2</sub>	12.81 <sup>cdefgh</sup>	19.90 <sup>cde</sup>	84.53 <sup>abc</sup>	0.343 <sup>c</sup>	0.720 <sup>ef</sup>	0.957 <sup>efg</sup>
USG-N <sub>160</sub> ×PM <sub>3</sub>	13.84 <sup>a</sup>	20.78 <sup>bcd</sup>	87.28 <sup>a</sup>	0.393 <sup>a</sup>	0.733 <sup>cde</sup>	1.036 <sup>cde</sup>
USG-N <sub>160</sub> ×CD <sub>3</sub>	13.02 <sup>bcd</sup>	19.29 <sup>de</sup>	78.71 <sup>abcde</sup>	0.277 <sup>ef</sup>	0.669 <sup>i</sup>	0.980 <sup>def</sup>
USG-N <sub>160</sub> ×CD <sub>5</sub>	13.29 <sup>bcd</sup>	19.93 <sup>bcd</sup>	71.18 <sup>efg</sup>	0.383 <sup>ab</sup>	0.738 <sup>bcd</sup>	1.068 <sup>bcd</sup>
USG-N <sub>180</sub> ×OC <sub>1</sub>	13.40 <sup>bcd</sup>	20.86 <sup>bcd</sup>	77.40 <sup>abcde</sup>	0.283 <sup>e</sup>	0.715 <sup>f</sup>	0.866 <sup>h</sup>
USG-N <sub>180</sub> ×OC <sub>2</sub>	14.48 <sup>a</sup>	21.63 <sup>bc</sup>	78.13 <sup>abcde</sup>	0.317 <sup>d</sup>	0.761 <sup>a</sup>	1.168 <sup>a</sup>
USG-N <sub>180</sub> ×PM <sub>2</sub>	12.67 <sup>efgh</sup>	19.91 <sup>cde</sup>	85.20 <sup>ab</sup>	0.370 <sup>b</sup>	0.634 <sup>k</sup>	0.906 <sup>fgh</sup>
USG-N <sub>180</sub> ×PM <sub>3</sub>	13.58 <sup>bc</sup>	20.92 <sup>bcd</sup>	81.77 <sup>abcde</sup>	0.387 <sup>ab</sup>	0.755 <sup>ab</sup>	1.140 <sup>ab</sup>
USG-N <sub>180</sub> ×CD <sub>3</sub>	12.47 <sup>fghi</sup>	20.25 <sup>bcd</sup>	81.35 <sup>abcde</sup>	0.267 <sup>efg</sup>	0.713 <sup>f</sup>	1.093 <sup>bc</sup>
USG-N <sub>180</sub> ×CD <sub>5</sub>	13.31 <sup>bcd</sup>	20.20 <sup>bcd</sup>	66.23 <sup>g</sup>	0.387 <sup>ab</sup>	0.691 <sup>gh</sup>	1.132 <sup>ab</sup>
PU-N <sub>180</sub> ×OC <sub>1</sub>	12.91 <sup>bcd</sup>	20.97 <sup>bcd</sup>	81.06 <sup>abcde</sup>	0.283 <sup>e</sup>	0.663 <sup>i</sup>	0.897 <sup>fgh</sup>
PU-N <sub>180</sub> ×OC <sub>2</sub>	13.67 <sup>b</sup>	21.14 <sup>bcd</sup>	56.06 <sup>h</sup>	0.383 <sup>ab</sup>	0.751 <sup>abc</sup>	1.136 <sup>ab</sup>
PU-N <sub>180</sub> ×PM <sub>2</sub>	13.09 <sup>bcd</sup>	19.75 <sup>cde</sup>	82.93 <sup>abc</sup>	0.263 <sup>fg</sup>	0.661 <sup>i</sup>	0.861 <sup>hi</sup>
PU-N <sub>180</sub> ×PM <sub>3</sub>	13.72 <sup>a</sup>	20.52 <sup>bcd</sup>	68.25 <sup>fg</sup>	0.393 <sup>a</sup>	0.677 <sup>hij</sup>	1.030 <sup>cde</sup>
PU-N <sub>180</sub> ×CD <sub>3</sub>	12.16 <sup>ghi</sup>	20.88 <sup>bcd</sup>	75.55 <sup>bcd</sup>	0.260 <sup>fg</sup>	0.672 <sup>ij</sup>	1.016 <sup>cde</sup>
PU-N <sub>180</sub> ×CD <sub>5</sub>	13.25 <sup>bcd</sup>	21.29 <sup>bcd</sup>	74.46 <sup>cdefg</sup>	0.383 <sup>ab</sup>	0.718 <sup>ef</sup>	1.038 <sup>cde</sup>
CV (%)	3.09	4.91	6.96	4.79	3.03	5.18

CV = coefficient of variation;

Means in a column superscripted by different lowercase letters are significantly different ( $p < 0.05$ ).

Treatments are detailed in the experimental design section.

83 mg/100g FW to 73 mg/100g FW in broccoli flowers. Similar results were reported by Chao-Jiong et al. (2010) who observed that the concentrations of ascorbic acid in the broccoli floret and stem decreased with increasing N application and a significantly lower amount of ascorbic acid was detected following N fertilization at the rate of 300–400 kg/ha N which was agreed with the present study. Similarly, the  $\beta$ -carotene content of broccoli heads was significantly influenced by the treatments (Table 4) and the highest  $\beta$ -carotene content (0.393 mg/100g FW) was obtained with both USG-N<sub>160</sub>×PM<sub>3</sub> and PU-N<sub>180</sub>×PM<sub>3</sub> followed by USG-N<sub>160</sub>×OC<sub>2</sub>, USG-N<sub>180</sub>×PM<sub>3</sub> and USG-N<sub>180</sub>×CD<sub>5</sub> (0.387 mg/100g FW). The lowest  $\beta$ -carotene content (0.250 mg/100g FW) was recorded in USG-N<sub>140</sub>×CD<sub>3</sub>. This finding was similar to the results reported by Decoteau (2000) for broccoli.

#### Chlorophyll-a and chlorophyll-b contents

The chlorophyll-a content of broccoli heads was significantly influenced by the treatment combinations and the highest chlorophyll-a content (0.761 mg/100g FW) was obtained with USG-N<sub>180</sub>×OC<sub>2</sub> followed by USG-N<sub>180</sub>×PM<sub>3</sub> (0.755 mg/100g FW), as shown in Table 5. The lowest chlorophyll-a content (0.661 mg/100g FW) was recorded in PU-N<sub>180</sub>×PM<sub>2</sub>. Similarly, the chlorophyll-b content of broccoli heads was significantly influenced by the treatment combinations and the highest chlorophyll-b content (1.168 mg/100g FW) was

found in USG-N<sub>180</sub>×OC<sub>2</sub> followed by USG-N<sub>160</sub>×OC<sub>2</sub> (1.143 mg/100g FW). The lowest content (0.776 mg/100g FW) was produced by the treatment USG-N<sub>140</sub>×CD<sub>3</sub>. These findings were similar to those of Ouda et al. (2008) who reported that the chlorophyll content was higher when a combination of organic and inorganic fertilizers was added compared with their individual addition.

Based on the findings, the SPAD value, nutrient uptake and N use efficiency as well as the head quality (compactness co-efficient, vitamin C,  $\beta$ -carotene and chlorophyll content) were all superior in the USG-organic manure integration compared to PU-organic manure. The SPAD value, nutrient (NPKS) uptake and head quality (compactness co-efficient,  $\beta$ -carotene and chlorophyll content) all increased with incremental rates of inorganic and organic fertilizer; however, N use efficiency and the vitamin C content slightly decreased. The highest amounts of N, P, K and S uptake (208.33 kg/ha, 33.04 kg/ha, 247.32 kg/ha and 3.45 kg/ha) were produced with USG-N<sub>180</sub>×OC<sub>2</sub> followed by USG-N<sub>160</sub>×OC<sub>2</sub>, but the maximum N use efficiency (92.89%) was obtained with USG-N<sub>140</sub>×OC<sub>2</sub>. USG-organic manure and the highest head CC (21.92) was from USG-N<sub>160</sub>×OC<sub>2</sub> followed by USG-N<sub>180</sub>×OC<sub>2</sub>. The highest vitamin C content (87.28 mg/100g FW) was recorded with USG-N<sub>160</sub>×PM<sub>3</sub> and the maximum  $\beta$ -carotene content (0.393 mg/100g FW) was recorded from both the USG-N<sub>160</sub>×PM<sub>3</sub> and PU-N<sub>180</sub>×PM<sub>3</sub> treatments.

## Conflict of Interest

The authors declare that there are no conflicts of interest.

## Acknowledgements

The authors acknowledge the Bangladesh Agricultural Research Council (BARC) for providing a fellowship award as financial support for the successful completion of this work.

## References

- Abdelrazzag, A. 2002. Effect of chicken manure, sheep manure and inorganic fertilizers on yield and nutrient uptake by onion. *Pak. J. Biol. Sci.* 5: 266–268. doi: 10.3923/pjbs.2002.266.268
- Abou El-Magd, M.M., El-Bassiony, A.M., Fawzy, Z.F. 2006. Effect of organic manure with or without chemical fertilizers on growth, yield and quality of some varieties of broccoli plants. *J. Appl. Sci. Res.* 2: 791–798.
- Black, C.A. 1965. *Methods of Soil Analysis, Part-I and II*. Amer. Soc. Agron. Inc. Pub. Madison, WI, USA.
- Bremner, J.M. 1965. Inorganic forms of nitrogen. In: Norman, A.G. (Ed.). *Methods of Soil Analysis. Part 2. Chemical and Microbiological Properties*, American Society of Agronomy. Madison, WI, USA, pp. 1179–1237.
- Chao-Jiong, X., Rong-Fang, G., Hui-Zhuan, Y., Jing, Y., Bo, S., Gao-Feng, Y., Qiao-Mei, W. 2010. Effect of nitrogen fertilization on ascorbic acid, glucoraphanin content and quinine reductase activity in broccoli floret and stem. *J. Food Agric. Environ.* 8: 179–184. doi.org/10.1234/4.2010.1479
- Costa, C., Frigon, D., Dutilleul, P., Dwyer, L.M., Pillar, V.D., Stewart, D.W., Smith, D.L. 2003. Sample size determination for chlorophyll meter readings on maize hybrids with a broad range of canopy types. *J. Plant Nutr.* 26: 1117–1130. doi.org/10.1081/PLN-120020079
- Craswell, E.T., Godwin, D.C. 1984. The efficiency of nitrogen fertilizers applied to cereals in different climates. In: Tinker, P.B., Auchli, A. (Eds.). *Advances in Plant Nutrition*. Praeger Publishers, New York, NY, USA, pp. 1–55.
- Decoteau, D.R. 2000. *Vegetable Crops*. Prentice Hall. Upper Saddle River, NJ, USA.
- Everaarts, A.P., de Willigen, P. 1999. The effect of the rate and method of nitrogen application on nitrogen uptake and utilization by broccoli (*Brassica oleraceavar. italica*). *Neth. J. Agri. Sci.* 47: 201–214. doi.org/10.18174/njas.v47i3.462
- Fox, R.L., Olsen, R.A., Rhoades, H.F. 1964. Evaluating the sulphur status of soils by plant and soil tests. *Soil Sci. Soc. Amer. Proc.* 28: 243–246.
- Gomez, K.A., Gomez, A. 1984. *Statistical Procedure for Agricultural Research—Hand Book*. John Wiley & Sons. New York, NY, USA.
- Jackson, M.L. 1973. *Soil Chemical Analysis*. Prentice Hall of India Private Limited. New Delhi, India.
- Kandil, H., Gad, N. 2009. Effects of inorganic and organic fertilizers on growth and production of broccoli (*Brassica oleracea* L.). *Factorişi Processe Pedogenetice din Zona Temperată 8 S. nouă* (2009): 61–69.
- Kapotis, G., Zervoudakis, G., Veltsistas, T., Salahas, G. 2003. Chlorophyll concentration in *Amaranthus vltius*: Correlation with physiological processes. *Russ. J. Plant Physiol.* 50: 395397–. doi.org/10.1023/A:1023886623645
- Karitonas, R. 2001. Effect of nitrogen supply on yield and quality of broccoli. In: Horst, W.J., Schenk, M.K., Bürkert, A., et al. (Eds.). *Kluwer Academic Publishers*. Hannover, Germany, pp. 298–299.
- Khalil, M.I., Schmidhalter, U., Gutser, R., Heuwinke, H. 2011. Comparative efficacy of urea fertilization via supergranules versus prills on nitrogen distribution, yield response and nitrogen use efficiency of spring wheat. *J. Plant Nutr.* 34: 779–797. doi.org/10.1080/01904167.2011.544349
- Lawlor, D.W. 2002. Carbon and nitrogen assimilation in relation to yield: Mechanisms are the key to understanding production systems. *J. Exp. Bot.* 53: 773–787. doi.org/10.1093/jexbot/53.370.773
- Magnusson, M. 2002. Mineral fertilizers and green mulch in Chinese cabbage (*Brassica pekinensis* Rupr): Effect on nutrient uptake, yield and internal tipburn. *Acta Agr. Scand. B-S. P.* 52: 25–35. doi.org/10.1080/090647102320260017
- Moniruzzaman, M., Rahman, S.M.L., Kibria, M., Grahman, M.A., Hossain, M.M. 2007. Effect of boron and nitrogen on yield and hollow stem of broccoli. *J. Soil. Nature.* 1: 24–29.
- Mukherjee, S.K. 1986. Chemical technology for producing fertilizer nitrogen in the year 2000. In: Swaminathan, M.S., Sinha, S.K. (Eds.). *Global Aspects of Food Production*. Tycooly Publishing Ltd. London, UK, pp. 227–237.
- Nagata, M., Yamashita, I. 1992. Simple method for simultaneous determination of chlorophyll and carotenoids in tomato fruit. *J. Japan. Soc. Food Sci. Technol.* 39: 925–928. doi.org/10.3136/nskkk1962.39.925
- Ouda, B.A., Mahadeen, A.Y. 2008. Effect of fertilizers on growth, yield, yield components, quality and certain nutrient contents in broccoli (*Brassica oleracea*). *Int. J. Agri. Biol.* 10: 627632–.
- Rickard, M. 2008. Effect of irrigation and nitrogen treatments on yield, quality, plant nitrogen uptake and soil nitrogen status and the evaluation of sap test, SPAD chlorophyll meter and Dualex to monitor nitrogen status in broccoli. M.Sc. thesis, Horticultural Science Programme, Swedish University of Agricultural Sciences, SLU. Alnarp, Sweden.
- Riley, H., Vågen, I. 2003. Critical N-concentration in broccoli and cauliflower, evaluated in field trials with varying levels and timing of fertilizer. *Acta Hortic.* 627: 241–249. doi.org/10.17660/ActaHortic.2003.627.31
- Samotus, B., Leja, M., Scigalski, A. 2013. A comparison of four methods of determination of ascorbic acid in fruits and vegetables [1982]. *Acta Agr. Silv. Ser. Agr.* 21: 105–122.
- Thomson, H.C., Kelly, W.C. 1985. *Vegetable Crops*, 5<sup>th</sup> ed. McGrawHill Co. Ltd., New Delhi, India.
- Tremblay, N., Scharpf, H.C., Weier, U., Laurence, H., Owen, J. 2001. Nitrogen management in field vegetables: A guide to efficient fertilization. <http://publications.gc.ca/collections/Collection/A42-92-2001E.pdf>, 19 December 2020.
- Varvel, G.E., Schepers, J.S., Francis, D.D. 1997. Ability for in season correction of nitrogen deficiency in corn using chlorophyll meters. *Soil Sci. Soc. Am. J.* 61: 1233–1239. doi.org/10.2136/sssaj1997.03615995006100040032x
- Wang, Q., Chen, J., Li, Y. 2004. Nondestructive and rapid estimation of leaf chlorophyll and nitrogen status of peace lily using a chlorophyll meter. *J. Plant Nutr.* 27: 557–569. doi.org/10.1081/PLN-120028878
- Wojciechowska, R., Rozek, S., Rydz, A. 2005. Broccoli yield and its quality in spring growing cycle as dependent on nitrogen fertilization. *Folia Horticulturae Ann.* 17: 141–152.
- Yamamoto, A., Nakamura, T., Adu-Gyamfi, J.J., Saigusa, M. 2002. Relationship between chlorophyll content in leaves of sorghum and pigeonpea determined by extraction method and by chlorophyll meter (SPAD-502). *J. Plant Nutr.* 25: 2295–2301. doi.org/10.1081/PLN-120014076
- Yoldas, F., Ceylan, S., Yagmur, B., Mordogan, N. 2008. Effect of nitrogen fertilizer on yield quality and nutrient content in broccoli. *J. Plant Nutr.* 31: 1333–1343. doi.org/10.1080/01904160802135118
- Zebarth, B.J., Bowen, P.A., Toivonen, P.M.A. 1995. Influence of nitrogen fertilization on broccoli yield, nitrogen accumulation and apparent fertilizer-nitrogen recovery. *Can. J. Plant Sci.* 75: 717–725. doi.org/10.4141/cjps95-122