



## PHOSPHORUS USE EFFICIENCY AND CRITICAL P CONTENT OF STEVIA GROWN IN ACID AND NON-CALCAREOUS SOILS OF BANGLADESH

Md. Maniruzzaman, <sup>1</sup>Tanzin Chowdhury, <sup>2</sup>Md. Arifur Rahman and <sup>3</sup>Md. Akhter Hossain Chowdhury

Soil Resources Development Institute, Farmgate, Dhaka, Bangladesh; <sup>1</sup>Department of Agricultural Chemistry, Sher-e-Bangla Agricultural University, Sher-e-Bangla Nagar, Dhaka-1207, Bangladesh and <sup>2&3</sup>Department of Agricultural Chemistry, Faculty of Agriculture, Bangladesh Agricultural University, Mymensingh-2202, Bangladesh

\*Corresponding author: Md. Akhter Hossain Chowdhury; E-mail: akhterbau11@gmail.com

### ARTICLE INFO

### ABSTRACT

**Received**  
10 August, 2017

**Accepted**  
22 August, 2017

**Online**  
30 August, 2017

**Key words**  
Stevia  
Leaf biomass yield  
PUE  
P requirement  
Critical P content

Knowledge of phosphorus (P) uptake and its use efficiency by crop plants is essential for adequate management of the plant nutrients to sustain food production with a minimal environmental impact. To study the effects of P on the growth, leaf biomass production, P content and uptake and to estimate P use efficiency (PUE), minimum P requirement and critical leaf P content of stevia, a pot experiment was conducted in the net house of the Department of Agricultural Chemistry, Bangladesh Agricultural University following completely randomized design (CRD) with three replications in acid and non-calcareous soils of Bangladesh. The applied treatments was six viz. 0 (P<sub>0</sub>), 25 (P<sub>25</sub>), 50 (P<sub>50</sub>), 75 (P<sub>75</sub>), 100 (P<sub>100</sub>) and 150 (P<sub>150</sub>) kg P ha<sup>-1</sup>. Plant samples were collected at 15 days interval to obtain different parameters. Collective results indicated that significantly highest values of different parameters were obtained with P @ 100 kg ha<sup>-1</sup> and the lowest from P control. Phosphorus application increased leaf dry yield at harvest by 55 to 510% in acid soil and 70 to 488% in non-calcareous soil over control. The rapid growth of the plant was recorded at the later stages (30 to 60 days after planting). Phosphorus content and uptake was directly proportional with the increased levels of P except the treatment P<sub>150</sub> in both soils. Maximum PUE and fertilizer P use efficiency (FPUE) was observed at P<sub>100</sub> treatment. Critical P content was estimated to be ca 0.19 and 0.30% in the leaves of stevia plants grown in acid and non-calcareous soils, respectively. For maximum leaf biomass production of stevia grown in acid and non-calcareous soils, the minimum requirement of P was also estimated to be ca 109 and 104 kg ha<sup>-1</sup>, respectively. The information of this finding would contribute to optimize the soil P use and improve fertilizer management for stevia cultivation.

**To cite this article:** Maniruzzaman M, T Chowdhury, MAH Chowdhury and MA Rahman, 2017. Phosphorus use efficiency and critical P content of stevia grown in acid and non-calcareous soils of Bangladesh. Res. Agric. Livest., Fish., 4 (2): 55-68.

## INTRODUCTION

Phosphorous (P) is one of the least available, least mobile, mineral nutrient to the plants in many cropping environments, based on its contribution to the biomass as a macronutrient (Goldstein et al., 1988). Many soils have large reserves of total P, often hundred-time more than the P available to the crops (Al-Abbas and Barber, 1964). Phosphorus is needed in metabolic processes such as energy transfer, signal transduction, macro-molecular biosynthesis, photosynthesis, respiration, etc. Relative to nitrogen and potassium, the recovery of P fertilizers by crop plants is usually very low due to soils' high capacity to fix P to soil constituents of little bioavailability (Manske et al., 2001; Lynch, 2007; Balemi and Negisho, 2012).

Stevia (*Stevia rebaudiana* Bertoni.) is a herbaceous perennial small bush contains the secret of stevioside, which make it the sweetest herb in the world (Soejarto et al., 1983). Dry leaves are the economic part in stevia plant. The leaves are having commercial importance due to presence of di-terpene sweet glycosides which are 300-400 times sweeter than sugar without any side effects. Hence, stevia is a potential natural source of no calorie sweeteners, alternative to the synthetic sweetening agents like saccharine, aspartame that are available in the market to the diet conscious consumers and diabetic patients. Stevia crop cultivation made significant impact in the countries like Japan, China, Korea, Mexico, USA, Thailand, Malaysia, Indonesia, Australia, Canada and Russia (Brandel and Rosa, 1992). Studies conducted in India so far could suggest only few management approaches of stevia for improving its productivity. Since the production potential of stevia in India is 2-3 t ha<sup>-1</sup> of dry leaves as against 1-2 t ha<sup>-1</sup> in China, it has definite advantage over China (Chalapathi et al., 1997b). Bangladesh being an agro-based country could easily introduce this plant as an industrial crop like sugarcane, sugar beet, tea or coffee and can commercially be cultivated in its relatively high land, char land, home stead area etc. as it grows well in open space having regular sun light.

Plants that are efficient in absorption and utilization of the absorbed nutrients greatly enhance the efficiency of applied fertilizers. Recently, suitable soil for stevia cultivation (Zaman et al., 2015), N and S requirement and critical N and S content of stevia grown in two contrasting soils of Bangladesh has been reported by Zaman et al., (2016a and 2016b). It is expected that a higher and balanced nutrient supply will result in higher foliage yield. So there is a need to set up certain protocols for cultivation of stevia in various soil conditions so that farmers can be benefited by selling, and industries also can get healthy leaves throughout the year to isolate the active components and can formulate economical market products.

Nevertheless, to the best of our knowledge, no systematic and detailed study has yet been conducted on P fertilizer requirement for stevia in acid and non-calcareous soils of Bangladesh. Therefore, optimum P requirement need to be determined for achieving maximum leaf biomass yield of stevia in the country. Critical values are quite useful and are frequently referred to when interpreting a plant analysis result. The critical P concentration of stevia is yet to be estimated. Though the level of P in the soil is one of the critical factors determining the growth and yield of the plants, no report has yet been published on the requirement of P and critical P level for the growth and leaf biomass yield of stevia in Bangladesh. Keeping in view the significant role of P in crop production systems, the present piece of research work was undertaken to investigate the effects of different levels of P on the growth, leaf biomass yield, P content and its uptake, minimum P requirement, PUE and critical leaf P concentration of stevia in two contrasting soils under the agro climatic conditions of Bangladesh Agricultural University, Mymensingh.

## MATERIALS AND METHODS

Before starting the experiment, initial soil pH, organic carbon, cation exchange capacity (CEC), available P were determined separately. Two soils viz. acid and non-calcareous of contrasting physical and chemical properties were used (Zaman et al., 2015). Approximately 40 kg soils from each location (Madhupur for acid soil and BAU farm for non-calcareous soil) were collected from 0-15cm depth of selected fellow land for the experiment. The samples were made free from plant residues and other extraneous materials, air dried, ground and sieved through a 2mm sieve. 500g sieved soil from each source was preserved in a polythene bag and the physical and chemical properties were determined following standard procedure (Page et al., 1982).

Eight kg processed soil was taken in each earthen pot of 23 cm in height with 30 cm diameter at top and 18 cm at bottom leaving 3 cm from the top. Forty five days old stevia seedlings (*Stevia rebaudiana* Bertoni) were collected from brac biotechnology laboratory, Joydebpur, Gazipur and used for the experiment. One stevia seedling was planted in each pot during 1<sup>st</sup> week of March, 2012. N, K, S, Zn and B were applied as basal doses @ 250, , 200, 30, 3 and 1 kg ha<sup>-1</sup> from prilled urea, MoP, gypsum, zinc sulphate and boric acid, respectively (Zaman, 2015). Six levels of P viz. 0, 25, 50, 75, 100, and 150 kg ha<sup>-1</sup> were applied from TSP. Nitrogen was applied in equal three installments, 1/3<sup>rd</sup> during pot preparation, 1/3<sup>rd</sup> at 15 days after planting (DAP) and 1/3<sup>rd</sup> at 30 DAP. The experiment was laid out in completely randomized design with three replications. Intercultural operations like irrigation, soil loosening, weeding, insect pest control, removal of flowers etc. were done as and when necessary.

Data were collected at 15, 30, 45 and 60 DAP. The crop was destructively harvested at 60 DAP. After harvesting the crop, leaf samples were separated, cleaned, dried for 72 hours, weighed, ground and stored. Plant heights, number of branches and leaves, leaf area, leaf fresh and dry weight were studied. Phosphorus content of stevia leaf was determined colorimetrically using SnCl<sub>2</sub> as a reducing agent (Page et al., 1982). Uptake was calculated from P content and leaf dry yield. PUE (grain yield per unit P added) was also calculated (Moll et al., 1982).

Phosphorous uptake, utilization and use efficiency were calculated following the below mentioned formulae:

$$\text{Phosphorus Use Efficiency (PUE)} = \frac{\text{Yield (kg)}}{\text{P in soil (kg)}}$$

$$\text{Phosphorus Uptake Efficiency (PUPE)} = \frac{\text{P in plant (kg)}}{\text{P in soil (kg)}}$$

$$\text{Phosphorus Utilization Efficiency (PUTE)} = \frac{\text{Yield (kg)}}{\text{P in plant (kg)}}$$

The mathematical calculation of fertilizer P uptake, utilization and use efficiency was described as:

$$\text{Fertilizer P use Efficiency (FPUE)} = \frac{(\text{Fertilized pot yield} - \text{control pot yield})}{\text{P applied}}$$

$$\text{Fertilizer P Uptake Efficiency (FPUPE)} = \frac{(\text{P in fertilized plant} - \text{P in control plant})}{\text{P applied}}$$

$$\text{Fertilizer P Utilization Efficiency (FPUTE)} = \frac{(\text{Yield of fertilized pot} - \text{yield of control pot})}{(\text{P in fertilized pot} - \text{P in control pot})}$$

Phosphorus requirement and critical P content of stevia was also estimated following Chowdhury, 2000. The results obtained were subjected to statistical analysis using standard method of analysis (Steel et al., 1997). The differences among the treatment means were compared by using Duncan Multiple Range test (Gomez and Gomez, 1984).

## RESULTS AND DISCUSSION

Effects of different levels of P on various parameters of stevia are discussed under the following heads-

### Plant Height

Different levels of P influenced the plant height of stevia (Figure 1). At harvest, plant height was significantly and rapidly increased with the increased levels of P up to 100 kg ha<sup>-1</sup> and then slowly decreased with further increase in P levels (150 kg ha<sup>-1</sup>). P application at all levels increased plant height by 65.0 to 94.0 cm in acid soil and 59.3 to 93.0 cm in non-calcareous soil at 60 DAP. Plant height was significantly increased with the advancement of the growth period irrespective of P levels. However, the tallest plants of 94.0 cm in acid soil and 93.0 cm in non-calcareous soil were obtained from P<sub>100</sub> which was identical with P<sub>75</sub> and P<sub>150</sub> but statistically different from P<sub>25</sub> and P<sub>50</sub> and the shortest plant was obtained from the control treatment irrespective of the soils studied.

Height increase was 45% higher in acid soil and 56% higher in non-calcareous soil over control. The increase in P levels increased the plant height progressively up to P<sub>100</sub> at 60 days of growth. Absolute control (P<sub>0</sub>) recorded significantly lowest plant height of stevia.

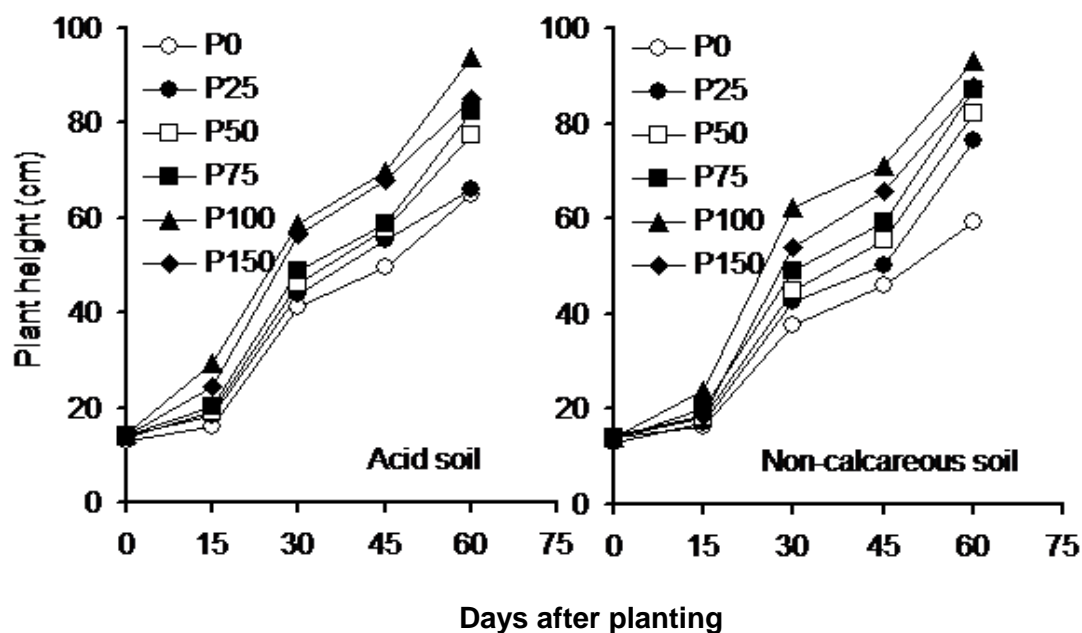


Figure 1. Effects of different levels of P on the plant height of stevia at various DAP

The results are in accordance with the findings of Chalapathi et al., (1999) who also reported increased plant height of stevia with higher nutrient levels in sandy loam soils at Bangalore. Our result is in agreement with the findings of Alam et al., (2003), who reported that the plant height of wheat significantly increased with increasing phosphorus application to the soil. On the other hand, a finding was also found by Rasul (2016), who observed highest plant height of wheat applying P<sub>2</sub>O<sub>5</sub> @ 250 kg ha<sup>-1</sup>.

#### Branch Number

The number of branch plant<sup>-1</sup> of stevia was significantly influenced by different levels of P (Figure 2). At harvest, number of branches plant<sup>-1</sup> was significantly increased with the increased levels of P up to 100 kg ha<sup>-1</sup> and then slowly decreased with further increase in P levels (150 kg ha<sup>-1</sup>). Phosphorus application at all levels increased branch number by 75 to 187% in acid soil and 45 to 163% in non-calcareous soil at 60 DAP. The number of branch plant<sup>-1</sup> was significantly increased with the advancement of the growth period irrespective of P levels and soils used. However, highest number of branches plant<sup>-1</sup> in both soils was obtained from P<sub>100</sub> which was statistically identical with all P levels except control and P<sub>25</sub> and the lowest value was obtained from the control treatment in both soils. Crop performance to a great extent is governed by the number of branches plant<sup>-1</sup>. It is, therefore, imperative that if the number of branches plant<sup>-1</sup> is higher, the numbers of leaves are expected to be higher; ultimately the leaf yield will be higher. This finding is also similar with the results of Jain and Singh (2003) who reported that number of branches per plant in pea increased with the application of P. Similarly, Atif et al., (2014) found highest number of branches of pea applying P @ 100 kg ha<sup>-1</sup>. This growth parameter might have possibly contributed positively to the higher leaf yield with higher P application.

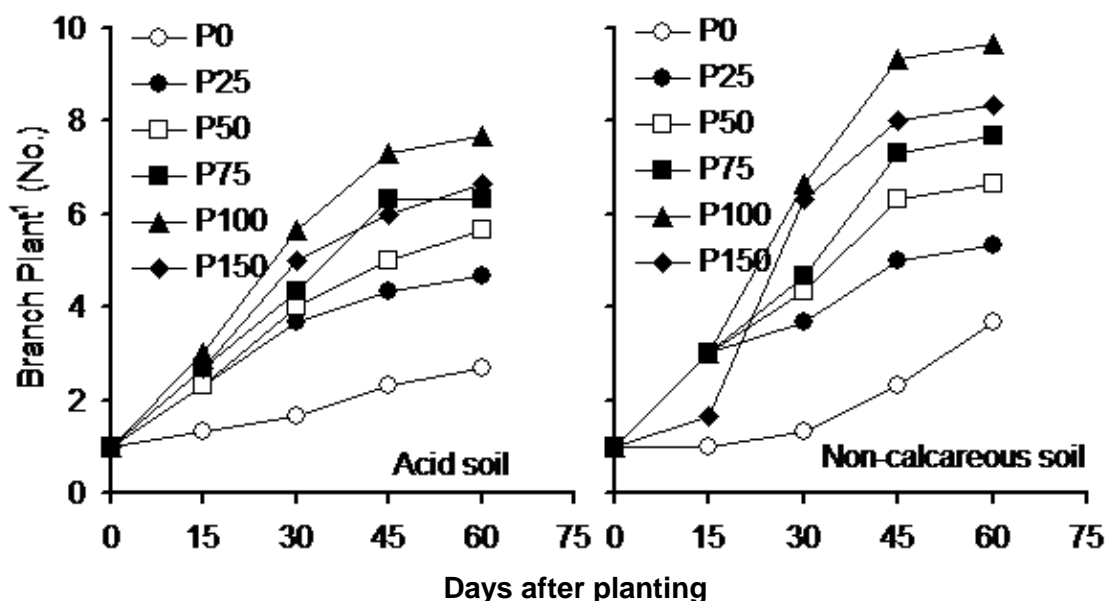


Figure 2. Effects of different levels of P on the branch number of stevia at various DAP

#### Leaf Number

The number of leaves of stevia in both acid and non-calcareous soils at all growth stages except 0 DAP (Figure 3) is also significantly influenced by different levels of P. Number of leaves plant<sup>-1</sup> was increased with the increased levels of P up to 100 kg ha<sup>-1</sup> and then declined by 65% in acid soil and 63% in non-calcareous soil with further addition (P<sub>150</sub>). Leaf number increase was very slow at the early growth stages (0-30 DAP) while it was rapid between 30 and 60 DAP irrespective of P levels except control. The most dominant increase was observed between 45 and 60 DAP particularly when P was applied @ 100 kg ha<sup>-1</sup>. P application at all levels increased the number of leaves by 26 to 248 in acid soil and 38 to 270 in non-calcareous soil. Plants fertilized with P<sub>150</sub> and P<sub>75</sub> produced identical number of leaves in non-calcareous soil. The minimum number of leaves plant<sup>-1</sup> was obtained from the plants fertilized with P control irrespective of soils and growth period. Green leaves are the site of photosynthetic activity taking place in the plants. The number of leaves plant<sup>-1</sup> would also substantiate the fact that increased number of leaves plant<sup>-1</sup> would contribute to the final yield of the plant particularly the crops like stevia in which only leaves are used as commercial product.

Kawatani et al., (1980) had also reported increased number of branches and leaves plant<sup>-1</sup> of stevia with higher inorganic nutrition in Japan. This finding is also similar with the results of Islam et al., (2013) who reported that number of leaves plant<sup>-1</sup> in tomato was increased due to the application of different doses of inorganic fertilizers. Aladakatti et al., (2012) found that plant height of stevia at harvest was significantly influenced by higher levels of nitrogen, phosphorus and potassium which in turn were responsible for higher number of branches plant<sup>-1</sup> and number of leaves plant<sup>-1</sup> resulting into higher leaf yield. Increased number of leaves plant<sup>-1</sup> of stevia with increased levels of N, P and K fertilizers was also reported by Buana and Goenadi (1985) in Brazil.

#### Leaf Area

The total leaf area plant<sup>-1</sup> at harvest as influenced by different levels of P is presented in Table 1. Leaf area plant<sup>-1</sup> responded significantly due to the application of different levels of P. The results revealed that leaf area progressively increased with increasing levels of P application up to P<sub>100</sub> in both soils and then declined with further addition (P<sub>150</sub>). The highest total leaf area plant<sup>-1</sup> (2207cm<sup>2</sup> in acid soil and 2866cm<sup>2</sup> in non-calcareous soil) observed at 60 DAP was measured from the plant receiving 100 kg P ha<sup>-1</sup> which was significantly higher than other levels of P. Second highest values (1322cm<sup>2</sup> in acid soil and 1752cm<sup>2</sup> in non-calcareous soil) were obtained from P<sub>150</sub>.

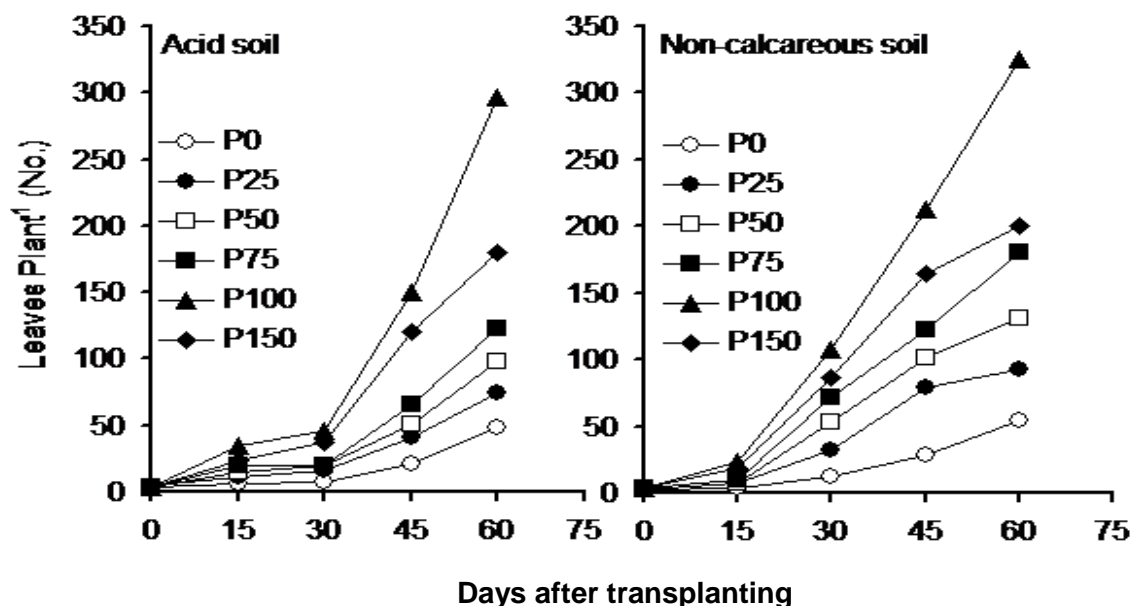


Figure 3. Effects of different levels of P on the leaf number of stevia at various DAP

Identical leaf area was obtained from the plants fertilized with P<sub>50</sub> and P<sub>75</sub> in non-calcareous soil. The lowest leaf area was found from the control treatment which was identical with P<sub>25</sub> irrespective of soils used. P application at all levels increased leaf area by 55 to 510% and 70 to 488% in acid and non-calcareous soils, respectively at harvest. Leaf area is an important growth indices determining the capacity of plant to trap solar energy for photosynthesis and has marked influence on the growth and yield of plant. Like other yield attributes, leaf area also followed increasing trends with the progress of plant growth with maximum value at 60 DAP irrespective of treatments. Higher leaf area of stevia with higher P levels could be attributed to more number of branches and leaves plant<sup>-1</sup> due to higher plant height. Khanom (2007) reported highest leaf area of stevia plant grown in non-calcareous soil applying chemical fertilizers.

#### Leaf dry weight

The data pertaining to the dry weight of stevia leaves plant<sup>-1</sup> at harvest as influenced by different levels of P fertilizer have been presented in Table 1. Results revealed that leaf dry weight significantly and progressively increased with the increased levels of P application up to 100 kg ha<sup>-1</sup> in both soils and then declined with further addition (P<sub>150</sub>). The highest leaf dry weight plant<sup>-1</sup> (9.02g in acid soil and 9.70g in non-calcareous soil) at harvest was measured from the plant receiving 100 kg P ha<sup>-1</sup> which was significantly higher than other levels of P. Second highest values (5.42g in acid soil and 6.10g in non-calcareous soil) were obtained from P<sub>150</sub> in both soils. Identical dry weight was also obtained from the plants fertilized with P<sub>75</sub> and P<sub>150</sub> in non-calcareous soil. The lowest values were obtained from the control treatment (1.48g in acid soil and 1.65g in non-calcareous soil). Phosphorus application at all levels increased leaf dry yield at harvest by 55 to 510% and 70 to 488% in acid and non-calcareous soils, respectively over control.

Dry matter accumulation by the crop is another important growth parameter to be considered for determining the economic yield while assessing the effects of different treatments. Phosphorus fertilizers showed significant influence on the dry weight of stevia leaves. These results are in conformity with the findings of Pramanik and Singh (2003) who reported that the application of P<sub>2</sub>O<sub>5</sub> at 60 kg ha<sup>-1</sup> significantly increased yield attributes and yield over control in chickpea. The lowest growth, yield and yield attributing characters of grass pea were recorded under control (P<sub>0</sub>) treatment. Murayama et al., (1980) in Japan experimentally proved that no fertilization resulted in lowest leaf yield of stevia. Thus, the findings of our study were in agreement with that of Ojeniyi et al., (2007) who reported that application of N, P, K and animal manure increased the dry weight of tomato as compared to control. Sood and Kumar (1994) also reported that

green and dry foliage yield increased with increasing levels of N and P, which also confirmed the results obtained in the present study. In case of simultaneous increase biomass yield, it was reported with the application of N, P and K at 60, 30 and 45 kg/ha, respectively, produced higher dry leaf yield with the simultaneous higher nutrient uptake by stevia plant (Chalapathi et al., 1997 and 1999).

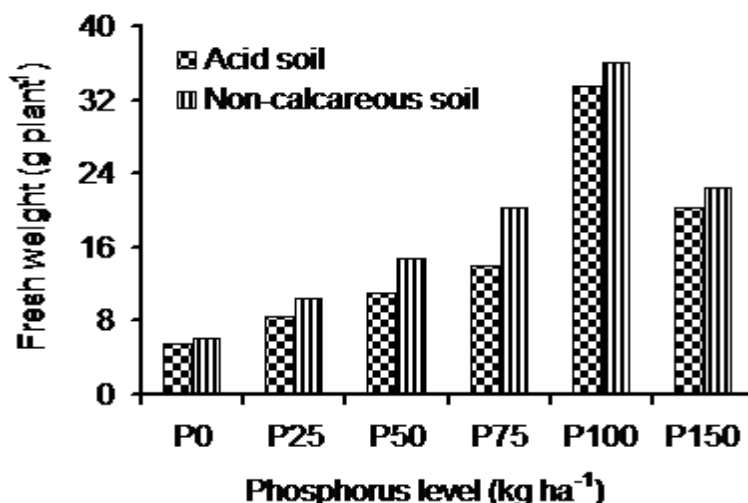
**Table 1.** Effects of different levels of P on leaf area, dry weight and yield increase of stevia leaves over control at harvest

P level	Leaf area plant <sup>-1</sup> (cm <sup>2</sup> )		Leaf dry weight (g plant <sup>-1</sup> )		Yield increase over control (%)	
	Acid soil	Non-calcareous soil	Acid soil	Non-calcareous soil	Acid soil	Non-calcareous soil
P <sub>0</sub>	243e	268e	1.48e	1.65e	-	-
P <sub>25</sub>	494de	657de	2.30d	2.81d	55	70
P <sub>50</sub>	708cd	1032d	2.99cd	4.05c	102	145
P <sub>75</sub>	886c	1532c	3.74c	5.49b	153	233
P <sub>100</sub>	2207a	2866a	9.02a	9.70a	510	488
P <sub>150</sub>	1322b	1752b	5.42b	6.10b	266	270
CV (%)	5	5	4.92	4.29	-	-
LSD <sub>0.05</sub>	177	237	0.51	0.56	-	-
SE±	161	209	0.61	0.64	-	-

CV = Coefficient of variance, LSD = Least significant difference, SE± = Standard error of means

#### Leaf fresh weight

The data pertaining to the fresh weight of stevia leaves plant<sup>-1</sup> at harvest as influenced by different levels of P fertilizer have been presented in Figure 4. Results revealed that leaf fresh weight significantly and progressively increased with the increased levels of P application up to P<sub>100</sub> in both soils and then declined with further addition (P<sub>150</sub>).



**Figure 4.** Effects of different levels of P on the fresh weight of stevia leaves at 60 DAP

The highest fresh weight plant<sup>-1</sup> (33.50g in acid soil and 36.02g in non-calcareous soil) at harvest was measured from the plant receiving 100 kg P ha<sup>-1</sup> which was significantly higher than other levels of P. Second highest values (20.23g in acid soil and 22.45g in non-calcareous soil) were obtained from P<sub>150</sub> in both soils. Identical fresh weight was also obtained from the plants fertilized with P<sub>75</sub> and P<sub>150</sub> in non-calcareous soil. The lowest values were obtained from the control treatment (5.49g in acid soil and 6.17g in non-calcareous soil).

Phosphorus application at all levels increased fresh weight at harvest by 2.96 to 28.01g plant<sup>-1</sup> in acid soil and 4.26 to 29.85g plant<sup>-1</sup> in non-calcareous soil.

#### Leaf P concentration and uptake

The data on the P concentration and uptake by stevia leaves as influenced by different levels of P have been presented in Table 2. Both the concentration and uptake was significantly influenced by the application of P fertilizers. Phosphorus concentration of the leaf was increased with the increased levels of P irrespective of soils used. The highest concentration (0.21% in acid soil and 0.33% in non-calcareous soil) was obtained when P was applied @ 100 kg ha<sup>-1</sup> in both soils which was statistically identical with the P contents of the leaves of stevia plant fertilized with P<sub>75</sub> and P<sub>150</sub> but significantly different from other treatments. The lowest P content was obtained from the plants receiving no P fertilizer in both soils. Phosphorus uptake was also significantly affected by its additions. The uptake of P did not follow the same trend like P concentration of stevia leaves. P uptake varied from 1.09 to 17.76 mg pot<sup>-1</sup> in acid soil and 1.71 to 32.12 mg pot<sup>-1</sup> in non-calcareous soil. The uptake of P as expected increased with increasing P levels up to 100 kg ha<sup>-1</sup> and then decreased with further additions (P<sub>150</sub>). The lowest P uptake was observed in the control treatment of both soils. The nutrient content of a plant varies not only among its various plant parts but changes with age and stage of development. Phosphorus contents and its uptake by stevia leaf varied significantly in both soils with their additions. The increase in concentration and nutrient uptake was proportional with the rate of application up to treatment P<sub>100</sub>. Higher nutrient uptake may be related to higher biomass yield due to the highest dry leaf yield harvested from that treatment. Because nutrient uptake was calculated from their concentrations and corresponding dry leaf yield.

**Table 2.** Effects of different levels of P on its content and uptake by stevia leaf at harvest

P level	Phosphorus			
	Acid soil		Non-calcareous soil	
	Content (%)	Uptake (mg pot <sup>-1</sup> )	Content (%)	Uptake (mg pot <sup>-1</sup> )
P <sub>0</sub>	0.07b	1.09e	0.10b	1.71e
P <sub>25</sub>	0.11b	2.45e	0.16b	4.98e
P <sub>50</sub>	0.14a	4.58d	0.22b	9.36d
P <sub>75</sub>	0.18a	7.45c	0.28b	15.55c
P <sub>100</sub>	0.21a	17.76a	0.33b	32.12a
P <sub>150</sub>	0.19a	10.54b	0.29b	17.88b
CV (%)	3.7	8.3	3.9	7.9
LSD <sub>0.05</sub>	0.06	0.78	0.06	1.12
SE±	0.01	1.42	0.02	2.55

CV = Coefficient of variance, LSD = Least significant difference, SE± = Standard error of means

These results are in agreement with the results found by Sushanta et al., (2014), who found that the total P uptake by wheat increased with increasing P fertilizer application.

#### Critical P concentration of stevia leaf

We followed the “Critical nutrition concentration” concept advanced by Ulrich (1952) for plant to determine critical P concentration in stevia leaf. Critical values as used by Ulrich and Hills (1973) are determined from the relationship of nutrient concentration and relative yield at the time of sampling. The critical P concentration in stevia leaf was estimated from the relative amount of leaf biomass to achieve 80% of the maximum production of stevia leaf following the procedure of Kouno et al., (1999). For both the soils, relative leaf biomass yield was plotted on the ordinate (Y axis) against the respective P concentration of stevia leaf on the abscissa (X axis) in Figure 5. The Phosphorus concentration corresponding to the arbitrary point at 80% to achieve the maximum leaf biomass production was estimated by the fitted curve to be ca 0.19 and 0.30% in the leaves of stevia plants grown in acid and non-calcareous soils, respectively (Figure 5).



Sharif et al., (1988) reported the critical concentration of P in maize plant as 1.4 mg P/g dry matter at 95% of relative yield. They found the maximum 100% relative yield at P tissue level of 1.8 mg P/g dry matter. Another study of Fageria et al., (2007) reported that the average P content of 0.16% in rice leaves is adequate for rice growth and yield.

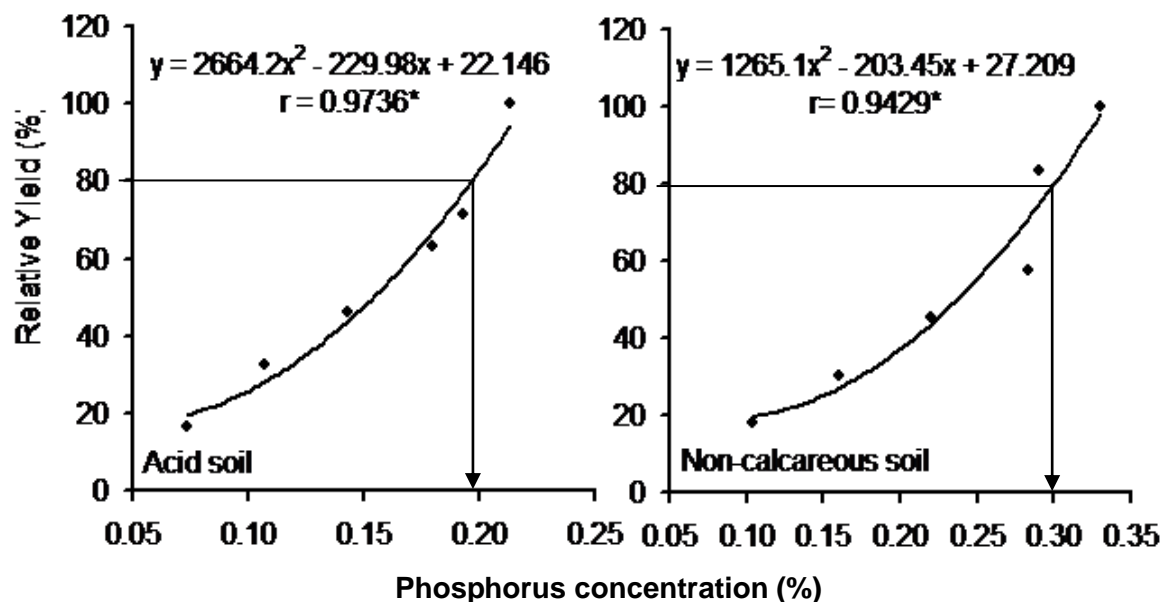


Figure 5. Correlation between leaf P concentration and relative leaf biomass yield of stevia grown in acid and non-calcareous soils. Values are the means of all treatments. \*\*Correlated significantly at  $P < 0.01$

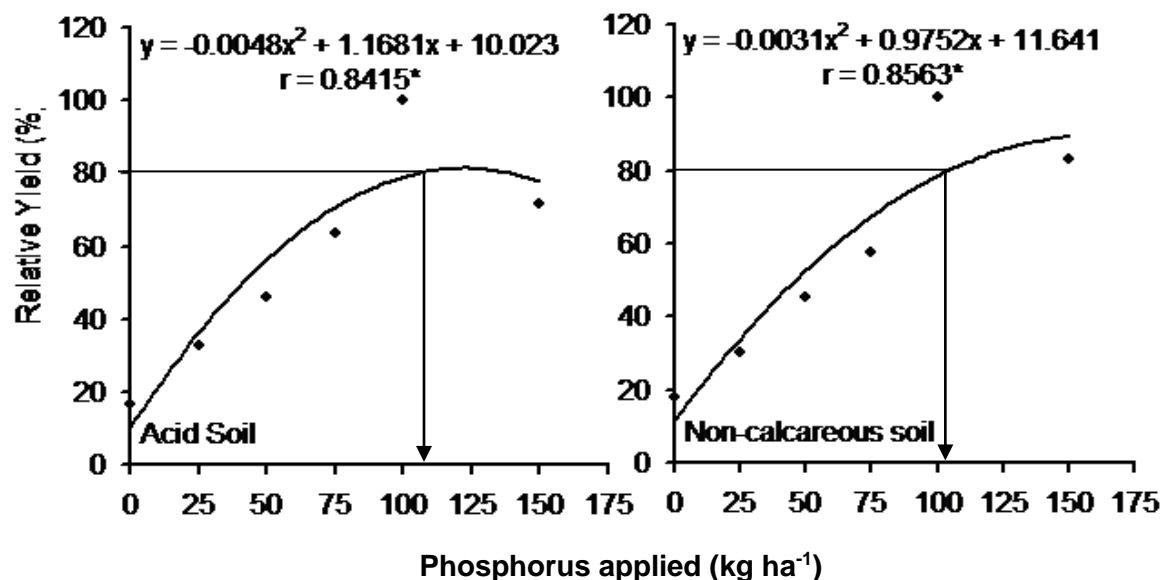


Figure 6. Correlation between applied P and relative leaf biomass yield of stevia grown in acid and non-calcareous soils. Values are the means of all treatments. \*\*Correlated significantly at  $P < 0.01$

### Phosphorus requirement of stevia plant

To determine the requirement of P in soil to obtain 80% of maximum leaf biomass yield, the applied P was plotted on the X axis against the relative leaf biomass yield on the Y axis. From the fitted curve, the corresponding estimated minimum amount of P for leaf biomass production in the plant grown in acid soil and non-calcareous soil to be ca 109 and 104 kg ha<sup>-1</sup>, respectively (Figure 6). A crop's requirement for a specific nutrient is commonly defined as "the minimum content of that nutrient associated with the maximum yield" or "the minimum rate of intake of the nutrient associated with the maximum growth rate" (Loneragan, 1968).

Phosphorus requirement vary greatly depending upon fertilizer applied and whether or not the investigation was performed in the field or green house, choice of crop etc. High phosphorus plant levels may cause imbalances and deficiencies of other elements, such as Zn, Cu, Fe, etc. Soils with inherent pH values between 6 and 7.5 are ideal for P availability, while pH values below 5.5 and between 7.5 and 8.5 limits P availability to plants due to fixation by aluminum, iron, or calcium. Phosphorus does not readily leach out of the root zone; potential for P loss is mainly associated with erosion and runoff. Phosphorus deficiency symptoms often occur as young plants are exposed to cool/wet growing conditions, resulting in a phase where vegetative growth exceeds the roots' ability to supply P. Recently, Zaman et al., (2016b) estimated minimum amount of S for leaf biomass production in stevia grown in acid and non-calcareous soils to be ca 40 and 45 kg ha<sup>-1</sup>, respectively.

### Phosphorous use, uptake and utilization efficiency of stevia

Our current understanding of PUE greatly varies depending upon crop species. Moll et al., (1982) using N, defined nutrient use efficiency as grain yield per unit of nutrient supplied (from soil plus fertilizer).

### Phosphorus use efficiency (PUE)

PUE is a measure of the economic yield produced per unit phosphorus in the soil. PUE was significantly influenced by the application of different levels of P. The results revealed that maximum PUE of 74.5% in acid soil and 69.6% in non-calcareous soil were observed at P<sub>100</sub> treatment. Similarly, the minimum PUE of 44.1% in acid soil and 33.9% in non-calcareous soil were obtained with P<sub>50</sub> treatment and control (P<sub>0</sub>), respectively (Table 3). Similarly, at higher P application rates plants used smaller proportion of fertilizer P due to the greater fixation that resulted in low PUE (Sultani et al., 2004; Alam et al., 2005; Rahim et al., 2010). Marschner (1995) reported that P retention in roots is improved sufficient levels, which may increase PUE at low P level.

**Table 3.** Phosphorus use, uptake and utilization efficiency of stevia leaf

P level	Acid soil			Non-calcareous Soil		
	PUE	PUPE	PUTE	PUE	PUPE	PUTE
P <sub>0</sub>	57.35ab	0.041c	1476a	33.94b	0.04c	997a
P <sub>25</sub>	45.75b	0.048c	976ab	45.05b	0.07bc	631b
P <sub>50</sub>	44.09b	0.062bc	708b	46.78b	0.10bc	469bc
P <sub>75</sub>	47.79b	0.085b	566b	48.88ab	0.14b	364c
P <sub>100</sub>	74.54a	0.156a	480b	69.65a	0.23a	310c
P <sub>150</sub>	41.90b	0.080b	527b	35.35b	0.10bc	351bc
CV (%)	2.66	5.04	5.22	2.98	5.90	5.06
SE±	3.25	0.01	97.10	3.27	0.02	62.11

PUE = Phosphorus use efficiency, PUPE = Phosphorus uptake efficiency; PUTE = Phosphorus utilization efficiency. Means with dissimilar letter(s) are significantly different from other treatments according to Scheffé-test ( $p < 0.05$ )

### Phosphorous uptake efficiency (PUPE)

PUPE is the ratio of kg P in plants and kg P in soil. PUPE increased with the increased levels of P up to 100 kg ha<sup>-1</sup> and then declined. The highest PUPE (0.156 in acid soil and 0.23 in non-calcareous soil) was observed when the plants were amended with 100 kg P ha<sup>-1</sup>. The lowest PUPE (0.041 in acid soil and 0.04 in non-calcareous soil) was recorded from the control treatment (Table 3). These results are in line with the previous work of Pongsakul and Gensen (1991).

### Phosphorous utilization efficiency (PUTE)

PUTE is the kg of dry matter yield divided by kg of P in plant, measure the efficiency with which P in plant is utilized for producing economic yield. The PUTE rate decreased with the increased level of P application. The PUTE was maximum (1476 in acid soil and 997 in non-calcareous soil) when no P was applied and it was minimum (480 and 310 in acid and non-calcareous soils, respectively) when the soil was amended with P @ 100 kg/ha (Table 3).

### Fertilizer P use, uptake and utilization efficiency of Stevia

#### Fertilizer P use efficiency (FPUE)

The results revealed that maximum FPUE of 31.9% in acid soil and 37.7% in non-calcareous soil was observed at P<sub>100</sub> treatment. Similarly, the minimum FPUE of 15.2% in acid soil and 23.9% in non-calcareous soil was obtained with P<sub>25</sub> treatment (Table 3). Thus the finding of our study is in good agreement with the finding of Yamoah et al., (2002) who reported that fertilizer P use efficiency improved significantly, when integrated (organic and inorganic) source of P was used.

**Table 4.** Fertilizer P use, uptake and utilization efficiency of stevia

P level	Acid soil			Non-calcareous Soil		
	FPUE	FPUPE	FPUTE	FPUE	FPUPE	FPUTE
P <sub>0</sub>	-	-	-	-	-	-
P <sub>25</sub>	15.24b	0.011b	1368a	27.25b	0.027b	1014a
P <sub>50</sub>	15.96b	0.015b	1107b	24.30b	0.032b	787b
P <sub>75</sub>	16.51b	0.018b	947b	23.85b	0.039b	642b
P <sub>100</sub>	31.92a	0.035a	938b	37.71a	0.064a	608b
P <sub>150</sub>	12.35c	0.013b	951b	14.04c	0.023b	636b
CV (%)	3.91	2.13	5.12	3.24	2.75	4.58
SE±	1.856	58.324	0.002	2.126	52.371	0.004

FPUPE = Fertilizer P uptake efficiency; FPUE = Fertilizer P use efficiency; FPUTE = Fertilizer P utilization efficiency, Means with dissimilar letter(s) are significantly different from other treatments according to Scheffe-test ( $p < 0.05$ )

#### Fertilizer P uptake efficiency (FPUPE)

FPUPE was significantly influenced by the application of different levels of P. The FPUPE reached its maximum (0.035 in acid soil and 0.064 in non-calcareous soil) value when the soils were amended with 100 kg P ha<sup>-1</sup> and it was minimum (0.011 in acid soil and 0.023 in non-calcareous soil) value those were fertilized by no P and 150 kg P ha<sup>-1</sup>, respectively (Table 4).

#### Fertilizer P utilization efficiency (FPUTE)

FPUTE decreased gradually with increased levels of P application up to treatment P<sub>100</sub>. The highest FPUTE (1368 in acid soil and 1014 in non-calcareous soil) was recorded in soil fertilized with 25 kg P ha<sup>-1</sup> and the lowest (938 and 608 in acid and non-calcareous soils, respectively) value was found in soil amended with 100 kg P ha<sup>-1</sup> (Table 4).

## CONCLUSION

The results clearly showed that all the parameters examined in this study were significantly affected by different doses of phosphorus. The highest values of most parameters were obtained from 100 kg P ha<sup>-1</sup> and the lowest values from control. However the decreasing trends in these parameters with further increase beyond 100 kg P ha<sup>-1</sup> indicated that higher doses of P could be detrimental to stevia and it should be avoided. Application of P @ 100 kg ha<sup>-1</sup> gives highest leaf dry yield at harvest (510% in acid soil and 488% in non-calcareous soil). Same responses were found in case of P content and uptake by stevia in both soils. The

highest PUE and FPUE were also recorded at P<sub>100</sub> treatment. The overall results suggest that farmers can be advised to apply P @ 109 kg ha<sup>-1</sup> in acid soil and 104 kg ha<sup>-1</sup> in non-calcareous soil to ensure optimum yield and maximum PUE. However, such studies should be conducted with different combination of soils and applied P before widespread recommendations.

## ACKNOWLEDGEMENTS

We express our sincere thanks to the concerned authority of Bangladesh Agricultural Research Council (BARC), Farmgate, Dhaka for financial support.

## CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this paper.

## REFERENCES

1. Aladakatti YR, YB Palled, MB Chetti, SI Halikatti, SC Alagundagi, PL Patil, VC Patil and AD Janawade, 2012. Effect of nitrogen, phosphorus and potassium levels on growth and yield of stevia (*Stevia rebaudiana* Bertoni). Karnataka Journal of Agricultural Science, 25: 25-29.
2. Alam SM, S Azam, S Ali and M Iqbal, 2003. Wheat yield and P fertilizer efficiency as influenced by rate and integrated use of chemical and organic fertilizers. Pakistan Journal of Soil Science, 22(2): 72-76.
3. Alam SM, SA Shah, and MM Iqbal, 2005. Evaluation of method and time of fertilizer application for yield and optimum P-efficiency in wheat. Songklanakarin Journal of Science and Technology, 27: 457-463.
4. Al-Abbas AH and SA Barber, 1964. A soil test for phosphorous based upon fractionation of soil phosphorous: I Correlation of soil phosphorous fraction with plant available phosphorous. Soil Science Society of America Proceedings, 28: 218-221.
5. Atif MJ, SA Shaukat, ASZ Shah, YA Choudry and SK Shaukat, 2014. Effect of Different Levels of Phosphorus on Growth and Productivity of Pea (*Pisum Sativum* L.) Cultivars Grown as Off-Season under Rawalakot Azad Jammu and Kashmir Conditions. Journal of Recent Advances in Agriculture, 2: 252-257.
6. Balemi T and K Negisho, 2012. Management of soil phosphorus and plant adaptation mechanisms to phosphorus stress for sustainable crop production: a review. Journal of Plant Nutrition Soil Science, 12: 547-561.
7. Buana L and DH Goenadi, 1985. A study on the correlation between growth and yield of stevia. Menara Perkebunan, 53: 68-71.
8. Chalapathi MV, B Shivaraj and VR Parama, 1997a. Nutrient uptake and yield of Stevia (*Stevia rebaudiana* Bertoni) as influenced by methods of planting and fertilizer levels. Crop Research, 14: 205-208.
9. Chalapathi MV, S Timmegowda, VR Prama and TG Prasad, 1997b. Natural non calorie sweetener stevia (*Stevia rebaudiana* Bertoni): A future crop of India. Crop Research, 14: 347-350.
10. Chalapathi MV, S Thimmegowda, DN Kumar, GGE Rao and J Chandraprakash, 1999. Influence of fertilizer levels on growth, yield and nutrient uptake of ratoon crop of stevia. Crop Research, 21: 947-949.
11. Chowdhury MAH, 2000. Dynamics of microbial biomass sulphur in soil and its role in sulphur availability to plants. PhD Thesis, Laboratory of Plant Environmental Science, Graduate School of Biosphere Sciences, Hiroshima University, Japan.

12. Fageria NK and MPB Filho, 2007. Dry matter and grain yield, nutrient uptake, and phosphorus use efficiency of lowland rice as influenced by phosphorus. *Communications in Soil Science and Plant Analysis*, 38(9-10): 1289-1297.
13. Gomez KA and AA Gomez, 1984. *Statistical Procedure for Agricultural Research*. 2nd Eds., Los Banos, Philippines: International Rice Research Institute, pp: 207-215.
14. Goldstein AH, DA Baertlein and RG McDaniel, 1988. Phosphate starvation inducible metabolism in *Lycopersicon esculentum*. *Plant Physiology*, 87(3): 716-720.
15. Islam MR, MAH Chowdhury, BK Saha and MM Hasan, 2013. Integrated nutrient management on soil fertility growth & yield of tomato. *Journal of Bangladesh Agricultural University*, 11: 33-40.
16. Jain LK and P Singh, 2003. Growth and nutrient uptake of chickpea as influenced by phosphorus and nitrogen. *Crop Research*, 25: 401-413
17. Kawatani T, Y Kaneki, T Tanabe and T Takahashi, 1980. On cultivation of Kaa-He-E (*Stevia rebaudiana* Bert). VI. Response of stevia to potassium fertilization rates and to the three major elements of fertilizer. *Japanese Journal of Tropic Agriculture*, 24: 105-112.
18. Khanom S, 2007. Growth, leaf yield and nutrient uptake by stevia as influenced by organic and chemical fertilizers grown on various types of soil. MS Thesis, Department of Agricultural Chemistry, Bangladesh Agricultural University, Mymensingh-2202.
19. Kouno K and S Ogata, 1988. Sulphur supplying capacity of soils and critical S values of forage crops. *Soil Science and Plant Nutrition*, 34: 327-339.
20. Loneragan JF, 1968. Nutrient requirements of plants. *Nature*, 220: 1307-1308.
21. Lynch J, 2007. Roots of the second green revolution. *Australian Journal of Botany*, 55: 493-512.
22. Marschner H, 1995. *Mineral nutrition of higher plants*, 2<sup>nd</sup> Eds., Annals of Botany Company, London: Academic Press, pp: 889.
23. Manske G, J Ortiz-Monasterio, MV Ginkel, R González, R Fischer, S Rajaram and P Vlek, 2001. Importance of uptake efficiency versus P utilization for wheat yield in acid and calcareous soils in Mexico. *European Journal of Agronomy*, 14: 261-274.
24. Murayama SM, OR Kayan, K Miyazato and A Nose, 1980. Studies on the cultivation of *Stevia rebaudiana* Bert. II. Effects of fertilizer rates, planting density and seedling clones on growth and yield. *Science Bulletin of the college of Agriculture, University of the Ryukyus, Okinawa*, 27: 1-8.
25. Ojeniyi SO, MA Awodun and SA Odedina, 2007. Effect of animal manure amended spent grain and cocoa husk on nutrient status, growth and yield of tomato. *Middle East Journal of Scientific Research*, 2: 33-36.
26. Page AL, RH Miller and DR Keeney, 1982. *Method of Soil Analysis, Part-2 Chemical and Microbiological Properties*, 2nd Eds., American Society of Agronomy, Inc. Madison, Wisconsin, USA.
27. Pongsakul P, ES Gensen, 1991. Dinitrogen fixation and soil N uptake by soybean as affected by phosphorus application. *Journal of Plant Nutrition*, 14: 809-823.
28. Pramanik K and RK Singh, 2003. Effect of levels and mode of phosphorus and biofertilizers on chickpea under dryland conditions. *Indian Journal of Agronomy*, 48: 294-296.
29. Rahim A, AM Ranjha, Rahamtullah and EA Waraich, 2010. Effect of phosphorus application and irrigation scheduling on wheat yield and phosphorus use efficiency. *Soil Environment*, 29: 15-22
30. Rasul GAM, 2016. Effect of Phosphorus Fertilizer Application on Some Yield Components of Wheat and Phosphorus Use Efficiency in Calcareous Soil. *Journal of Dynamic Agricultural Research*, 3: 46-52.
31. Sharif MZ, R Amin, FE Qayum and M Aslam, 1988. Plant tissue concentration and uptake of phosphorus by maize as affected levels of fertilization. *Pakistan Journal of Agricultural Research*, 9: 335-338.
32. Sushanta S, S Bholanath, M Sidhu, P Sajal and DR Partha, 2014. Grain yield and phosphorus uptake by wheat as influenced by long-term phosphorus fertilization. *African Journal of Agricultural Research*, 9: 607-612.
33. Soejarto DD, CM Compadre, PJ Medon, SK Kamath and AD Kinghorn, 1983. Potential sweetening agents of plant origin. II. Field search for sweet tasting stevia species. *Economic Botany*, 37: 71-79.

34. Sood BK, N Kumar, 1994. Effect of nitrogen and phosphorus on forage yield and nutrient uptake of oat-berseem mixture. *Crop Research*, 8: 239 – 244.
35. Steel RGD, JH Torrie and D Dickey, 1997. *Principles and Procedures of Statistics: A biometrical approach*, 3rd Eds., McGraw-Hill Book Co., New York, USA.
36. Sultani MI, M Shaukat, IA Mehmood and MF Joyia, 2004. Wheat growth and yield response to various green manure legumes and different P levels in pothowar region. *Pakistan Journal of Agricultural Research*, 41: 102-108
37. Ulrich A, 1952. Physiological basis for assessing the nutritional requirements of plants. *Annual Review. Plant Physiology*, 3: 207-228.
38. Ulrich A and FJ Hills, 1973. Plant analysis as an aid in fertilizing sugar crops: Part I. Sugar beets. In: *Soil testing and plant analysis*, Eds., Walsh, L.M. and J.D. Beaton Madison, Wisconsin, USA, pp: 271-288.
39. Yamoah CF, A Bationo, B Shapiro and S Koala, 2002. Trend and stability analysis of millet yields treated with fertilizer and crop residues in the Sahel. *Field Crops Research*, 75: 53-62.
40. Zaman MM, 2015. Nutrient requirement leaf yield and stevioside content of stevia (*Stevia rebaudiana* Bertoni) in some soil types of Bangladesh. PhD Thesis, Department of Agricultural Chemistry, Bangladesh Agricultural University, Mymensingh.
41. Zaman MM, MAH Chowdhury and T Chowdhury, 2015. Growth parameters and leaf biomass yield of stevia (*Stevia rebaudiana*, Bertoni) as influenced by different soil types of Bangladesh. *Journal of Bangladesh Agricultural University*, 13: 33-40.
42. Zaman MM, MAH Chowdhury, KM Mohiuddin and T Chowdhury, 2016a. Nitrogen requirement and critical N content of stevia grown in two contrasting soils of Bangladesh. *Research in Agriculture, Livestock and Fisheries*, 3: 87-97.
43. Zaman MM, MAH Chowdhury, T Chowdhury and ABMM Hasan, 2016b. Critical leaf S concentration and S requirement of stevia grown in two different soils of Bangladesh. *Fundamental and Applied Agriculture*, 1(3): 106-111.