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GROWTH AND BIOMASS YIELD OF DIFFERENT COTTON CULTIVARS IN RESPONSE TO MACRONUTRIENT APPLICATION

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Abstract

High or low fertilizer use efficiency has threatened the yield of cotton. In this study, four combined ratios of nitrogen (N), phosphorous (P) and potassium (K) (N1 = 150 N, 0 P_2O_5, and 0 K_2O kg ha^{-1} (control), N2 = 150 N, 45 P_2O_5, and 90 K_2O kg ha^{-1}, N3 = 150 N, 90 P_2O_5, and 135 K_2O kg ha^{-1}, and N4 = 150 N, 135 P_2O_5, and 180 K_2O kg ha^{-1}) on biomass yield and growth of two cotton cultivars (*Bacillus thuringiensis* (Bt) Siza 1 and Sikang 1) were studied in a two-year (2016-2017) field trial in a randomized complete block design with three replicates. The ratios of N2, N3, and N4 increased biomass production and growth as compared to the control group N1; the flowering period of Sikang 1 cultivar was improved by 20.6%, the boll setting period by 42.8%, and the peak boll setting period by 30.0%. Over the two growing seasons, Siza 1 cultivar increased boll opening by 6.3%, stem fresh and dry weight by 27.6% and 33.3%, leaves fresh and dry weight by 14.5% and 8.7%, and reproductive organ fresh and dry weight by 19.1% and 21.5%. In 2016 and 2017, the higher percentage of N4 reduced the fresh stem weight, fresh leaves, and dry weight of both cultivars compared to N1. The current study showed that the integrated supply of N, P and K at suitable ratios could improve cotton biomass yield and growth during boll development. However, the percentage of N3 ratios was in comparison more effective for cotton growth in the Siza 1 cultivar than in Sikang 1 cultivar.

Key words: Cotton growth, Biomass yield, and Cultivars.

Introduction

Nutrients play an indispensable role in enhancing cotton biomass growth and yield (Cianchetta & Davis, 2015; Yang *et al*., 2016). The application of nitrogen (N), phosphorous (P) and potassium (K) are considered to be an essential macronutrients that are required more frequently and in greater amounts for cotton growth and development than other nutrients, as they are the three main constituents of biomass and yield elements (Hou *et al*., 2007; Chen *et al*., 2010; Zhu *et al*., 2020). During these three elements, the effects of N has been extensively investigated for the production of Bacillus thuringiensis (Bt) cotton (Ahmad *et al*., 2019). In previous studies, it was showed the content of N, particularly in fruiting leaves and leaves under the boll (LSCB), was closely associated to boll growth and cotton development (Ma *et al*., 2009; Zhao *et al*., 2011). In China, the amount of N applied varies widely, from a low of 225 kg ha^{-1} to a very high of 450 kg ha^{-1} (Li *et al*., 2010). At low N application rate, cotton plants do not receive sufficient N, which is important for biomass production, plants development and boll growth. Conversely, when N application rates are very high, N use efficiency decreases and the release of nitrous oxide gas (N_2O; from the application of nitrogen fertilizers) increase, leading to environmental problems. Cotton growth has high nitrogen requirements and many developing countries have neglected the application of phosphorus and potassium, resulting in depletion of nitrogen in many soils, which leads to reduced cotton growth and yield (Zhang *et al*., 2010). In China, there is a rising trend toward greater application of nitrogen. This trend in China is due to many factors. First, N is more readily available to cotton producers than P and K. Second, the boosting effect of N application is easier to observe than that of P and K (Pettigrew *et al*., 2005). In recent years, China has paid more and more attention to the environmental issues produced by the incorrect use of N fertilizers and the lack of P and K (Ahmad *et al*., 2022). Therefore, cotton growers should promote the use of balanced fertilization of N, P and K (Song *et al*., 2019).

During cotton growth and yield, the deficiency of these three elements in the soil is considered as the main limiting factor for biomass yield formation due to the continued capacity of leaf photosynthetic characteristics (Ahmad *et al*., 2021). Application of these three macro-nutrients can promote cotton growth, enhance biomass production and increase the distribution proportion of reproductive organs (Ibrahim *et al*., 2018; Ibrahim *et al*., 2018; Ahmad *et al*., 2019). Studies have shown that biomass distribution in cotton organs during the crop cycle is significant determinant of final yield. During the initial stage of cotton growth, substantial foliar tissue differentiation allows access to light, which promotes better cotton growth and provides a source for fiber quantity and quality at the end of season. Accordingly, during the final growth phase, a large amount assimilates is delivered to the reproductive organs, ensuring better growth and yield of cotton (Khan *et al*., 2019).

Cotton is considered one of the most important crop varieties for fiber production in China and plays an important role in production worldwide (Dong *et al*., 2006).

The population growth has led to an increase in demand for food and clothing and threatens the productivity of cotton due to climate change (Feng *et al*., 2017). Insufficient or excessive application leads to serious environmental problems, including greenhouse gas emission (Zheng *et al*., 2004), eutrophication (Le *et al*., 2010), and soil acidification (Zhang *et al.*, 2013). Therefore, the efficient use of fertilizers by reducing nutrient loss is an important environmental issue. China's cotton growing areas are mainly split into three dominant production regions, including the middle and lower reaches of the Yangtze River, the Huang huai Sea cotton areas and the northwest inland cotton areas (Feng *et al*., 2017). However, in recent years, the cultivation of cereals and vegetables in these three larger areas has significantly reduced the size of the main cotton growing areas. As a result, the competition between cotton crops and cereals has become one of the most important aspect limiting the growth of cotton productivity (Li *et al*., 2010). Appropriate nutrient supply is an effective process to improve the success of cotton growers and to address the competition between cotton and cereal crops (Geng *et al*., 2020).

The utilization efficiency of these three macro-nutrients is determined not only by the concentration of individual nutrients applied to N, P and K but also through the combination of these macro-nutrients. Suitable combined application of these three elements can improve the combined utilization efficiency of these three macro-nutrients. The interaction of single applications of N, P, and K or N and P, N and K to cotton has been well documented in many previous studies. However, to date, only a few studies have adequately documented the integrated effects of different rates of macro-nutrients works must be developed. (Shabbir *et al*., 2016; Vieira *et al*., 2018). To enhance the efficiency of macro-nutrients, use and further promote cotton growth and biomass production, a superior strategy based on a better understanding of how the combinations of these three macro-nutrients work together. The effect of macro-nutrients ratios on the biomass yield of cotton at different growth stages and in two cotton cultivars was investigated in a two-year field study.

Materials and Methods

Experimental sites and materials: A two years (2016-2017) field experiment was conducted at the Experimental Farm of Yangzhou University, Jiangsu, province, China (32.30^0 N, 1198 25^0 E) on cotton. The field experimental soil had loamy clay and the soil surface (0-20 cm) contained of 1.27 mg kg^{-1} total N, 107.2 mg kg^{-1} mineral N (sum of NO_3^- and $NH4^+$-N), 15.1 mg kg^{-1} Bray^{-1} P, and 77.9 mg kg^{-1} soil test K with pH 6.9 (1:1 in water). Soil concentrations of total nitrogen were measured according to the method of Kjeldhal (Labconco, 1998) available phosphorus (P) was determined according to the Micro-Vanadate-Molybdate method (Olsen, 1954) and available potassium (K) was determined by flame photometry according to the neutral ammonium acetate extraction method (Ahmad *et al*., 2019).

Experimental design: Two varieties of Siza 1 (Bt transgenic) and its parental variety Sikang 1 (represented as V1 and V2) were planted in a field. Three replications of four macro-nutrients ratios 1:0:0, 1:0.3:0.6, 1:0.6:0.9, and 1:0.9:1.2 (represented as N1, N2, N3, and N4) were carried out in a randomized complete block design (RCBD) in split plot arrangement. Macro-nutrients were used as a source of urea, single super phosphate, and potassium chloride at various ratios of N1 = 150 N, 0 P_2O_5, and 0 K_2O kg ha^{-1} (control), N2 = 150 N, 45 P_2O_5, and 90 K_2O kg ha^{-1}, N3 = 150 N, 90 P_2O_5, and 135 K_2O kg ha^{-1}, and N4 = 150 N, 135 P_2O_5, and 180 K_2O kg ha^{-1}at seed sowing and first flowering stage.

The seeds of cotton were sown in field on 20th April during 2016 and 2017 in 24 plots each plot of 28m^2 (8.0m x 3.5m) at a line spacing of 100cm, row spacing of 33.3 cm and planting density of 30,000 plants ha-^{1}. Application of N was practiced twice at the equal rate of 150 kg ha^{-1} after the period of seed sowing and at the beginning of flowering (Ahmad *et al*., 2021). Other field practices were carried out according to local recommendations.

Sampling and processing: In the field, 10 uniform plants from each plot were marked to determine the various growth stages of cotton including peak flowering, boll-setting, peak-boll setting, and boll-opening stages.

On 25th july the data on peak flowering, 10th august on boll setting, 25th august on peak boll setting and 20th september on boll opening were measured by counting the number of flowers on ten chosen cotton plants in each plot.

Biomass yield: To determine biomass yield, five entire cotton plants were randomly picked from the center line, manually divided into stems, leaves and reproductive organs (squares, flowers, bolls) and put in an envelope. The fresh stems, leaves and reproductive were weighed using an electronic scale (Geng *et al*., 2015). For further determination, the subsamples were placed under the assistance of an electric fan and heated rapidly at 105°C for 30 min and followed by heating at 60°C until a constant weight (Luo *et al*., 2020). When was completely dried, the weight of the dried stems, leaves and genitalia was measured (Cordeiro *et al*., 2021).

Statistical analysis

The two years (2016-2017) field experimental trial was performed under the same environmental conditions arranged in a (two varieties and four fertilizer levels) factorial completely randomized design with three replications. During this study, two years of average data are presented. The mean of each variable was calculated for each treatment on (July 25th, August 10th, August 25th, and September 20th) for all plants in a replicated manner. The data of each changeable were subjected to analysis of variance (ANOVA) applying the statistical package of MSTAT-C (Freed *et al*., 1991). The treatment mean differences of every parameter were isolated by the least significant difference (LSD) test if the F tests were significant at the ($p\leq0.05$). The figures were created by using a SigmaPlot 10.0 (SPSS Inc., Point Richmond, CA, USA).

Results

Peak flowering, boll setting, peak boll setting, and boll opening: The peak flowering of cotton was significantly influenced by different ratios of N, P, and K and cultivar during two consecutive growing seasons (Table 1). Compared to N1, N2 increased bloom by 41.2% and 58.3%, N3 increased by 64.2% and 68.7%, and N4 increased by 69.6% and 73.6% respectively, in both years (Fig. 1). Compared to V1, V2 showed an increase of 16.6% and 20.6% in peak flowering in the two growing seasons, respectively.

In addition, different ratios of N, P, and K and cultivars had significant effects on cotton boll setting. Application of N2, N3 and N4 increased boll set by 3.8%, 10.2%, and 6.8% in 2016 compared to N1. Similarly, N2, N3, and N4 increased by 12.9%, 5.2%, and 19.4% in 2017 compared to N1 (Fig. 2). Compared to V1, V2 performed better, increasing the number of boll set by 42.8% and 46.3% in both years, respectively.

Accordingly, peak boll setting was also influenced by different proportions of N, P, and K and cultivar. During 2016 and 2017, peak boll setting increased by 6.5% and 7.6% for N2, 9.3% and 14.7% for N3, 18.3% and 16.7% for N4 compared to N1 (Fig 3). Compared with V1, the peak styles of V2 increased by 28.0% and 30.0% in two years, respectively.

Similarly, different N, P and K ratios and cultivars increased the boll opening. Compared to N1, N2, N3, and N4 increased boll opening by 8.0%, 14.9%, and 18.5% in 2016 and 8.5%, 14.3% and 19.9% in 2017 (Fig. 4). Compared to V1, V2 reduced boll opening by 5.6 and 6.3% in both years.

Table 1. Significance test for source of variation (mean square), and its effects on peak flowering, boll setting, peak boll setting, and boll opening during two growing seasons.

Source	Peak flowering	Boll setting	Peak boll setting	Boll opening
C (cultivar)	3.2ns	156.8**	237.1**	40.4 ns
R (ratio of N)	14.6*	3.8ns	14.0ns	89.3**
Y (year)	1.8ns	10.1ns	6.6 ns	52.1*
C×R	0.1 ns	0.6ns	2.9 ns	7.2ns
C×Y	0.0 ns	0.0ns	0.0 ns	0.2ns
R×Y	0.2ns	0.0ns	0.3 ns	0.11 ns
C×R×Y	0.1 ns	0.2 ns	0.2 ns	1.6 ns
Error	4.2	5.9	9.0	12.1
Year				
2016	2.2 a	5.1 a	12.7 a	30.6 b
2017	2.6 a	6.1 a	13.5 a	32.7 a
N levels				
N1	1.0 b	4.9 a	11.9 b	28.3 c
N2	2.0 ab	5.7 a	12.8 ab	30.8 bc
N3	3.0 a	5.3 a	13.4 ab	33.1 ab
N4	3.5 a	6.2 a	14.5 a	34.6 a
Cultivars				
CFSH30	2.1 a	3.7 b	10.9 b	32.6 a
Siyong3180	2.7 a	7.3 a	15.4 a	30.8 a

C: Cultivar; R: Rates of nitrogen; Y: Year * and ** Significant at 5 and 1% probability level, respectively

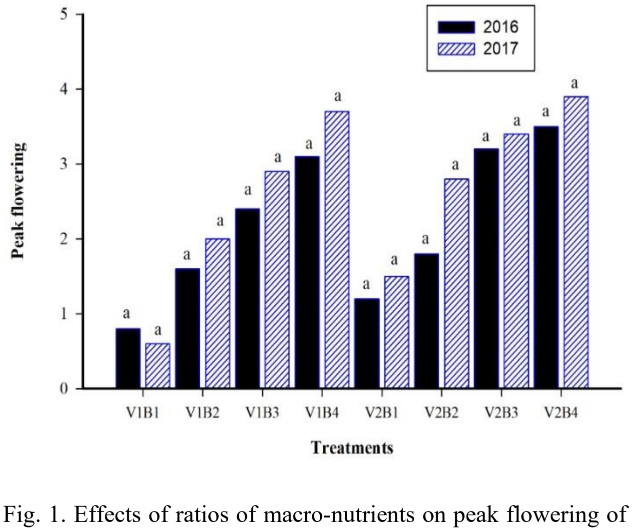

Fig. 1. Effects of ratios of macro-nutrients on peak flowering of Siza 1 and Sikang 1 cotton plants in 2016 and 2017 years. Various letters on the bars are statistically dissimilar at the 0.05 probability level by an ANOVA-protected test. Bars with same letters showed no significant differences in each figure.

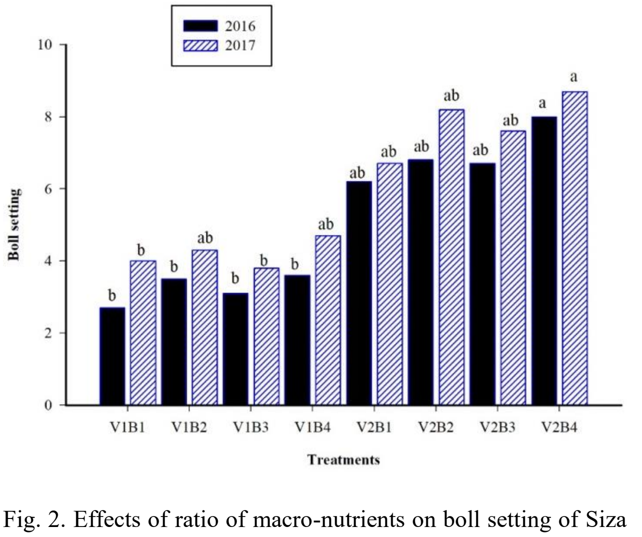

Fig. 2. Effects of ratio of macro-nutrients on boll setting of Siza 1 and Sikang 1 cotton plants in both years. Various letters on the bars are statistically dissimilar at the 0.05 probability level by an ANOVA-protected test. Bars with same letters represented no significant differences in each figure.

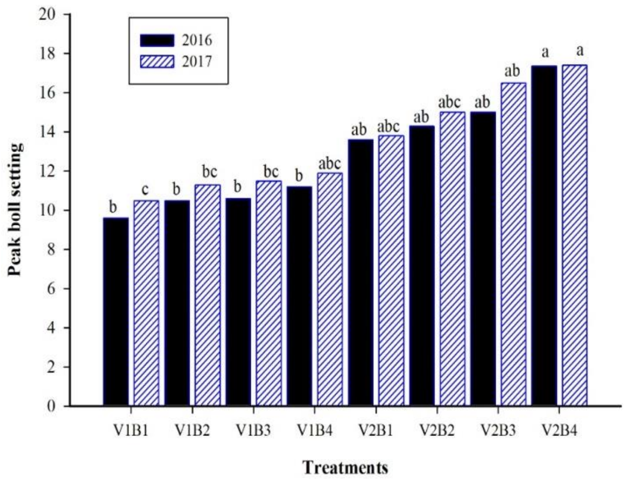

Fig. 3. Effects of ratio of macro-nutrients on peak boll setting of Siza 1 and Sikang 1 cotton plants during two years. Various letters on the bars are statistically dissimilar at the 0.05 probability level by an ANOVA-protected test. Bars with same letters showed no significant differences in each figure.

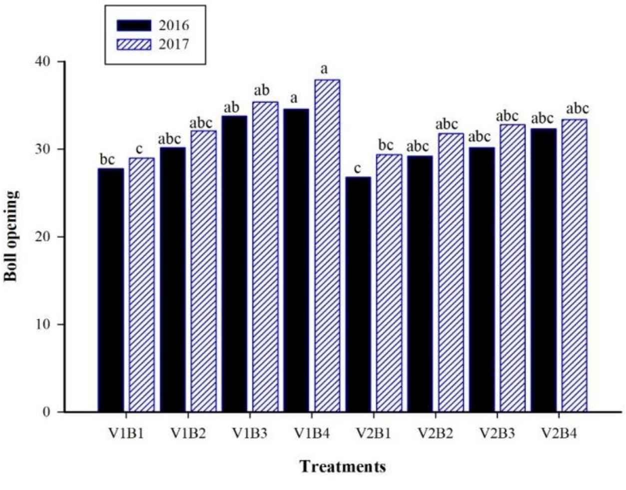

Fig. 4. Effects of ratio of macro-nutrients on boll opening of Siza 1 and Sikang 1 cotton plants in two years. Various letters on the bars are statistically dissimilar at the 0.05 probability level by an ANOVA-protected test. Bars with same letters represented no significant differences in each figure.

Table 2. Significance test for source of variation (mean square), and its effects on stem, leaves, and reproductive fresh and dry weight during two growing seasons.

Source	Stem fresh weight (g)	Stem dry weight (g)	Leaves fresh weight (g)	Leaves dry weight (g)	Reproductive fresh weight (g)	Reproductive dry weight (g)
C (cultivar)	360.6 ns	0.0 ns	11.5 ns	5.0 ns	391.1 ns	138.3 ns
R (ratio of N)	304.9 ns	41.2 ns	354.5 ns	50.1 ns	314.9 ns	184.8*
Y (year)	26739.4**	2022.2**	15793.4**	1246.9**	44884.1**	989.9**
C×R	453.5 ns	26.2 ns	268.5 ns	25.3 ns	245.0 ns	39.6 ns
C×Y	47.0 ns	154.2 ns	242.1 ns	4.9 ns	3.5 ns	0.8 ns
R×Y	130.9 ns	11.0 ns	9.1 ns	3.2 ns	16.7 ns	46.0 ns
C×R×Y	60.2 ns	7.0 ns	4.9 ns	3.2 ns	25.7 ns	31.3 ns
Error	686.8	73.5	481.4	50.4 ns	1430.1 ns	53.6 ns
Year						
2016	17.9 b	9.1 b	23.3 b	9.2 b	13.0 b	10.3 b
2017	65.1 a	22.1 a	59. 6 a	19.4 a	74.2 a	19.4 a
N levels						
N1	39.0 a	14.7 a	41.7 a	13.9 a	42.6 a	11.4 b
N2	42.0 a	16.2 a	44.0 a	15.3 a	45.7 a	16.7 ab
N3	48.4 a	17.8 a	46.3 a	16.5 a	49.2 a	19.5 a
N4	36.8 a	13.5 a	33.6 a	11.6 a	37.0 a	11.7 b
Cultivars						
CFSH30	44.3 a	15.5 a	41.9 a	14.7 a	46.5 a	16.5 a
Siyong3180	38.8 a	15.6 a	40.9 a	14.0 a	40.7 a	13.1 a

C: Cultivar; R: Rates of nitrogen; Y: Year * and ** Significant at 5 and 1% probability level, respectively

Biomass yield: The biomass of cotton was significantly affected by different ratios of N, P, and K and cultivar (Table 2). As compared to N1, stem fresh weight enhanced by 8.5% in 2016 for N3 and by 11.3% and 22.9% in 2017 for N2 and N3, respectively (Fig. 5). In addition, an increased in N, P, and K ratios led to a decrease in the fresh weight of the stem. V2 decreased stem fresh weight by 10.5% and 27.6% during both growing seasons compared to V1.

In addition, different ratios of N, P, and K and cultivar increased stem dry weight. In N2, N3, and N4 increased stem dry weight by 3.3%, 10.3%, and 17.1% in 2016 year compared to N1 (Fig. 5). Similarly, In N2 and N3 there was an increase of 11.5% and 17.1% in 2017 as compared to N1. In N4, stem dry weight decreased by 18.9% in 2017. Compared to V1, V2 decreased 33.3% of stem dry weight in the first year and showed a 10.5% and 27.6% reduction in fresh weight of stems in the two growing seasons, respectively.

In addition, leaves fresh weight was also influenced by different N, P, and k and cultivar. Compared to N1, the fresh weight of leaves increased by 10.8% and 2.6% for N2 and by 11.5% and 9.1% for N3, respectively (Fig. 6). However, N4 reduced leaf fresh weight by 20.7% and 25.4% in both years compared to N1. Compared to V1, V2 decreased 14.4% of leaf fresh weight in 2016 and increased by 2.1% in 2017.

Accordingly, leaf dry weight of cotton was affected by different proportions of N, P, and K and cultivar. In N2 and N3, the dry leaves increased by 18.8%, and 19.6% in the first year, and increased by 4.0% and 13.5% in the second year compared to N1 (Fig. 6). In N4, stem weight decreased by 7.5% in 2016 and by 25.4% in 2017. V2 reduced stem weight by 2.6% and 8.7% in both years compared to V1.

Moreover, the various ratios of N, P, and K and cultivar was significantly increased the reproductive fresh weight of cotton (squares, flowers, bolls). The reproductive fresh weight increased by 34.1% for N2, 46.8% for N3, and 16.2% for N4 in the first year compared to N1 (Fig. 7). Likewise, N2 and N3 showed an increase of 1.2% and 5.4%, respectively, in the second year compared to N1. In N4, the reproductive fresh weight reduced by 26.3% in the second year as compared to N1. In V2 reproductive fresh weight reduced by 13.3% and 19.1%, respectively, during both years compared to V1.

Correspondingly, the reproductive dry weight (squares, flowers, bolls) was significantly affected by different ratios of N, P, and K and cultivar. The ratios of N2, N3, and N4 enhanced the reproductive dry weight by 56.3%, 70.1%, and 54.2% in 2016 and increased by 19.6% and 21.3% in 2017 for N2 and N3 compared to N1. However, there was a 39.5% reduction in reproductive dry weight in the second year as compared to N1. As compared to V1, V2 variety had 21.5% and 32.9% reduction in reproductive dry weight in both years, respectively.

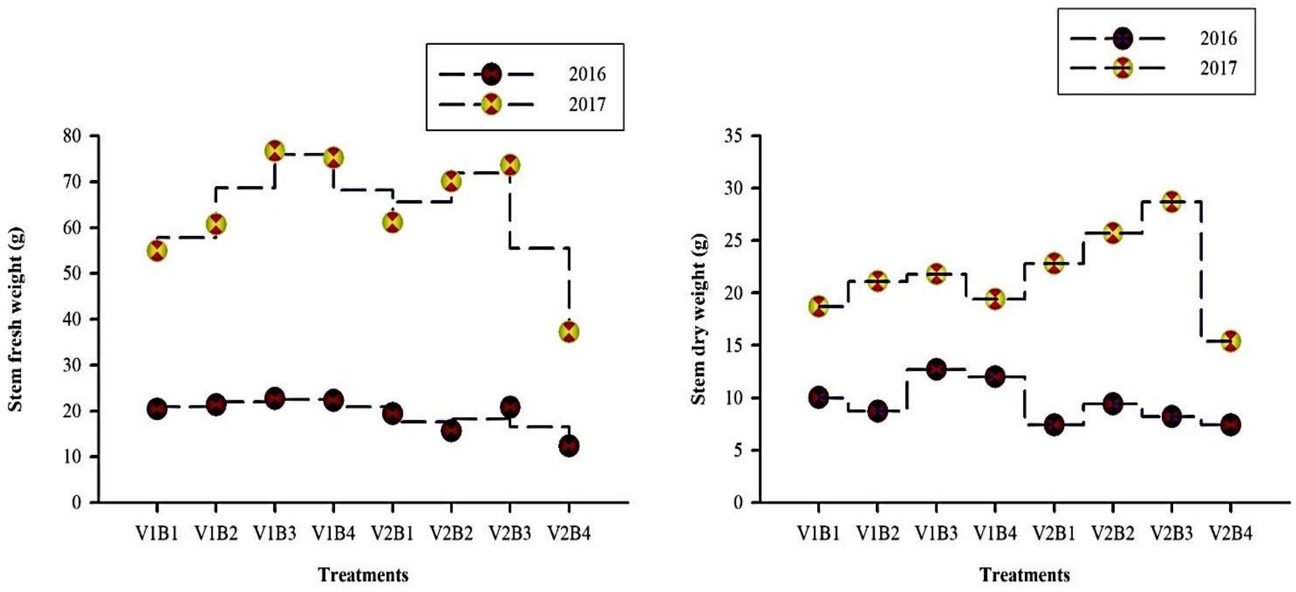

Fig. 5. Effects of ratio of macro-nutrients on stem fresh and dry weight (g) of Siza 1 and Sikang 1 cotton plants in two years (2016-2017). Various letters on the bars are statistically dissimilar at the 0.05 probability level by an ANOVA-protected test. Bars with no letters showed no significant differences in each figure.

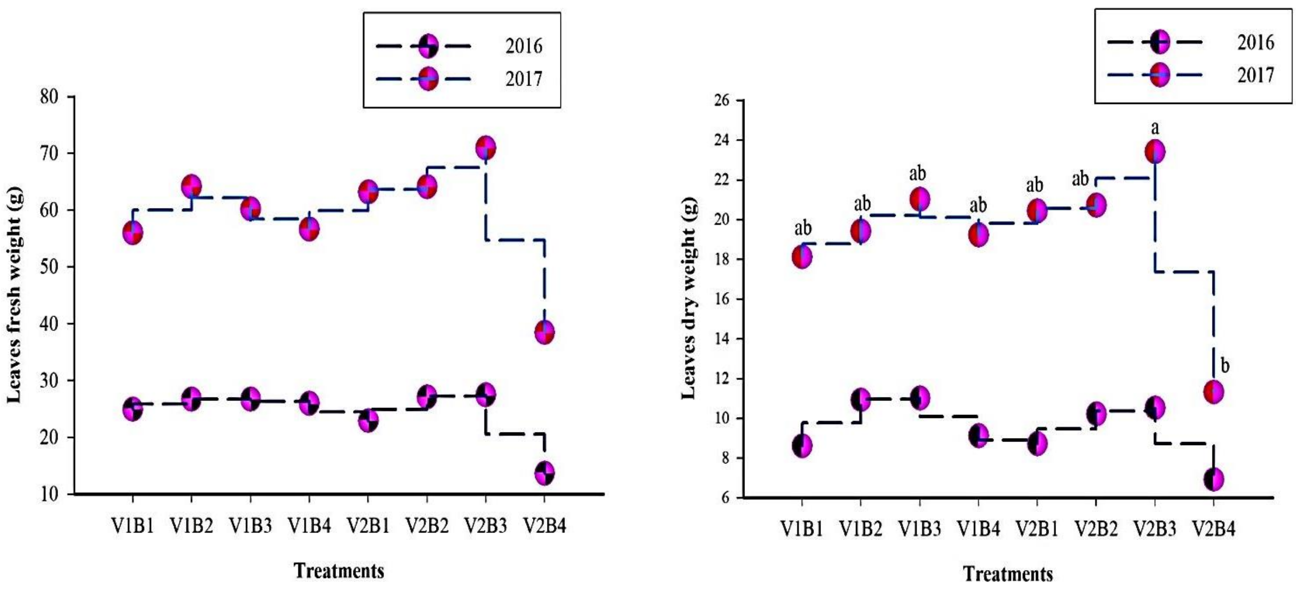

Fig. 6. Effects of ratio of macro-nutrients on leaves fresh and dry weight (g) of Siza 1 and Sikang 1 cotton plants during two consecutive years. Various letters on the bars are statistically dissimilar at the 0.05 probability level by an ANOVA-protected test. Bars with no letters showed no significant differences in each figure.

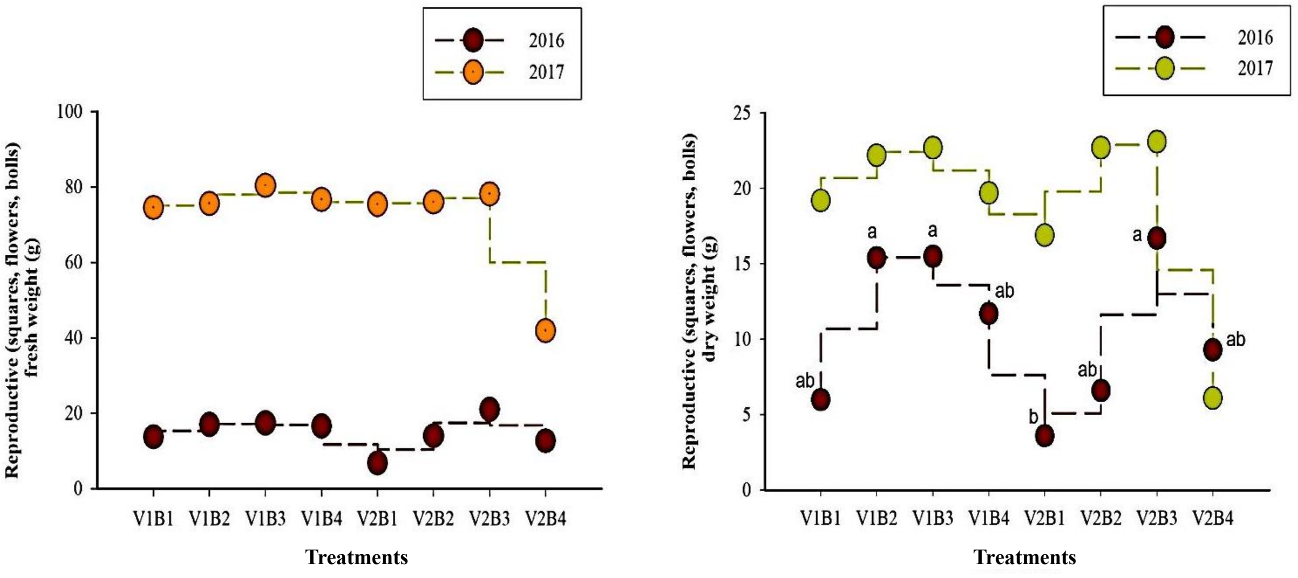

Fig. 7. Effects of ratio of macro-nutrients on reproductive (squares, flowers and bolls) fresh and dry weight (g) of Siza 1 and Sikang 1 cotton plants in two years. Various letters on the bars are statistically dissimilar at the 0.05 probability level by an ANOVA-protected test. Bars with no letters represented no significant differences in each figure.

Discussion

Cotton plants needs a regular supply of nutrients for its growth, including nitrogen (N), phosphorus (P) and potassium (K). However, they absorb these nutrients at different growth stages and in different amounts (Xiaoping *et al*., 2008). Previous studies have shown that early fertilization (peak flowering) can reduce labor costs without reducing yields (Luo *et al*., 2020). It is well known that the supply of N application has a significant impact on the growth and development of cotton (Hachiya & Sakakibara, 2017; Ahmad *et al*., 2019). However, different N concentrations in relation to the ratio and source of P and K affecting the growth of cotton have not been studied so far. Therefore, the premier purpose of the current study was to examine how the combined application of these macro-nutrients affects the growth and biomass yield of cotton. Efficient biomass production is a prerequisite for cotton fiber yield formation. In the present study, the biomass yields of N2 and N3 were significantly higher than the respective controls in terms of nitrogen to phosphorus ratio. However, our results are in unison to those of (Ullah *et al.*, 2017), who showed that plants increased the growth characteristic and stem weight of ramie crops in the integrated presence of these three macro-nutrients compared to their respective K, P and P, K treatments. The corresponding results were supported by (Iqbal *et al*., 2020) for cotton crop.

In addition, in 2017, the biomass production was higher compared to the 2016 cropping season, which could be due to the unfavorable environmental conditions in 2016 (Figs. 5, 6, 7) which negatively affected the performance of all plants. Overall, the V1 cultivar performed best in terms of biomass yield compared to the respective V2 cultivar. The increase in biomass yield of V1 cultivars may lie in the genetic background of the cultivars.

In addition, cotton plants are very small and require few nutrients until the peak of flowering (Yang *et al*., 2016). The application of macro-nutrients corresponded well during the cotton growth phase (Geng *et al*., 2015; Ahmad *et al*., 2021). Earlier investigations shown that supplying fertilizer at time of first flower emergence should be more conducive to increasing the rate of nutrient accumulation in cotton, as well as accumulating nutrients in a short period of time (Luo *et al*., 2020). This approach will help our environment by reducing greenhouse gas emissions and water pollution (Shen *et al*., 2011). In the current study, the supply of macro-nutrients ratios at the early stages significantly increased the yield at peak flowering, boll setting, peak boll setting, and boll opening, and the maximum reproductive biomass yield and growth of cotton compared to the respective controls (Luo *et al*., 2020). Our results are inconsistent with the findings of (Yang *et al*., 2016), who showed that the application of N, and K increased the cotton yield at the initial stage but slightly decreased at the maturity stage (boll opening). In this research, these differentiations may be responsible for the contribution of P with K and N. Moreover, the V1 cultivar was reduced in all growth stages of boll development compared to V2, except at boll opening in both years.

Conclusion

Application of macro-nutrients at early growth stages of cotton significantly increased biomass production and cotton growth as compared to N1. Both cotton cultivars showed potential increment but V1 cultivar performed higher biomass yield as compared to the respective V2 cultivar. All these indices sort out the best practices for receiving higher biomass yield and cotton growth but N3 application is more suitable in both growing seasons, especially for V1 cultivar. However, further examination is required to study the effects of various sources of macro-nutrients ratios on a wide range of cotton cultivars. Therefore, macro-nutrients fertilizer management is required to ensure cotton growth and yield.

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