



## Article

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## COMPARATIVE PRODUCTIVITY AND SEED NUTRITION OF COTTON BY PLANT GROWTH REGULATION UNDER DEFICIENT AND ADEQUATE BORON CONDITIONS

*Produtividade Comparativa e Nutrição de Sementes de Algodão com o Uso de Regulador de Crescimento Vegetal sob Condições Adequadas e na Deficiência de Boro*

**ABSTRACT** - Plant growth regulators (PGRs) potentially improve the productivity and nutritional quality of crops through modulation of morphology, physiology and nutrient dynamics within plants. However, the effect of PGRs may differ under environments with deficient and adequate nutrients. Here the role of mepiquat chloride (MC) on the yield and seed nutritional quality of cotton was explored under boron (B) deficiency and adequate conditions in a two-year field study. Mepiquat chloride was foliar-applied at different dosages (0 and 70 mg L<sup>-1</sup>) and growth stages (squaring and flowering) and B was applied into soil (0, 1, 1.5, 2 and 2.5 kg ha<sup>-1</sup>) to establish B deficiency and adequate levels. Seed cotton yield and seed nutritional quality were substantially higher under adequate B conditions, compared to B deficiency. Nonetheless, MC applications improved the seed cotton yield, seed oil yield, seed protein yield and concentration of seed mineral nutrients (N, P, K, B, Zn and Fe except for Mn) under both B deficiency and adequate conditions. Nonetheless, the improvement in all traits caused by MC application was greater under adequate B conditions (2-2.5 kg ha<sup>-1</sup> B), compared to B deficiency conditions. Furthermore, the application of MC at squaring proved more beneficial in improving the seed cotton yield and nutritional quality, compared to the flowering stage. In conclusion, MC application improves seed cotton yield and cotton seed nutritional quality under both deficient and adequate B conditions, through an improved nutrient accumulation in seed tissues; however, the efficacy of MC is affected by B deficiency.

**Keywords:** Boron deficiency; Mepiquat chloride; Seed cotton yield; Cotton seed nutritional quality; Nutrient Dynamics

**RESUMO** - Os reguladores de crescimento vegetal (RCV) melhoram potencialmente a produtividade e a qualidade nutricional das culturas através da modulação da morfologia, fisiologia e dinâmica dos nutrientes nas plantas. No entanto, o efeito dos RCV pode diferir sob ambientes com deficiência ou conteúdos adequados de nutrientes. O papel do cloreto de mepiquat (CM) na produtividade e na qualidade nutricional da semente de algodão foi explorado sob deficiência ou condições adequadas de boro em um estudo de campo de dois anos. O cloreto de mepiquat foi aplicado nas folhas em diferentes doses (0 e 70 mg L<sup>-1</sup>) e estádios de crescimento (formação de botões florais e abertura das flores), e o B foi aplicado no solo (0, 1, 1,5, 2 e 2,5 kg ha<sup>-1</sup>) para estabelecer deficiência e níveis adequados desse nutriente. O rendimento do algodão e a qualidade nutricional das sementes foram substancialmente maiores em condições de B adequadas, comparadas à deficiência

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*de B. No entanto, as aplicações de CM melhoraram o rendimento do algodão, o rendimento de óleo de sementes, o rendimento de proteína de sementes e a concentração de nutrientes minerais de sementes (N, P, K, B, Zn e Fe, exceto Mn) sob deficiência e condições adequadas de B. Entretanto, a melhoria em todas as características causada pela aplicação de CM foi maior em condições de B adequadas (2-2,5 kg ha<sup>-1</sup> de B), quando comparadas às condições de deficiência. Além disso, a aplicação de CM durante a formação dos botões mostrou-se mais benéfica na melhoria do rendimento do algodão e da qualidade nutricional, em comparação com o estágio de abertura das flores. Em conclusão, a aplicação de CM melhora o rendimento do algodão e a qualidade nutricional das suas sementes em condições de deficiência e condições adequadas de B, por meio de melhor acumulação de nutrientes nos tecidos das sementes; contudo, a eficácia do CM é afetada pela deficiência de B.*

**Palavras-chave:** deficiência de boro, cloreto de mepiquat, rendimento do algodão, qualidade nutricional de sementes de algodão, dinâmica dos nutrientes.

## INTRODUCTION

Cotton (*Gossypium hirsutum* L.) is cultivated worldwide for fibers and cottonseeds, due to a diverse seed composition, i.e. oil, fatty acids, protein and mineral nutrition (Bellaloui et al., 2015). Its oil is used for human consumption and meal for animal feed. The production of cottonseed oil has reached the third position after rapeseed and soybean (USDA-ERS, 2013). Cottonseeds are rich in protein (17-27%), oil (12-30%), saturated fatty acids (~29%) and unsaturated fatty acids (~70%) (Dowd et al., 2010; Pettigrew and Dowd, 2011). Various environmental and management practices influence the cottonseed composition despite genetic control (Pettigrew and Dowd, 2011; Bellaloui et al., 2015). Thus, a better management of crops and environmental factors is essential to maintain and ensure high productivity and nutritional quality of cottonseeds.

Boron is an essential micronutrient that directly or indirectly influences plant growth and development, and physiological and biochemical processes. It is mainly involved in the metabolism of nucleic acid, sugar and starch, protein, indole acetic acid and phenol, cell wall biosynthesis, and it regulates membrane integrity and functioning (Goldbach et al., 2001; Barker and Pilbeam, 2007). Boron deficiency is spreading throughout the world, with wide areas in some regions of the world and smaller areas in others (Shorrocks, 1997; Niaz et al., 2002). Several factors have been held responsible for B deficiency in soil, including drought, high soil pH, calcareousness, low organic matter in soil, B leaching and fixation with clay minerals, and microbial activity (Shorrocks, 1997; Mengel and Kirkby, 2001; Barker and Pilbeam, 2007).

Boron deficiency is associated with the abnormal growth of apical regions, normally due to the disturbance in cell wall growth and cell division. However, the most prominent effects of B deficiency are perceived on the plant reproductive growth. Its deficiency affects pollen development and pollen tube growth, resulting in male sterility and poor seed set. Moreover, seeds and fruits abscise prematurely; this consequently leads to reduced crop yield (Barker and Pilbeam, 2015). Although cotton is conceived as well adapted to a wide range of soils and growing conditions, it is quite sensitive to B deficiency. In cotton, its deficiency causes stunted growth, reduced carbon assimilation and translocation, poor fruit retention, reduced yield, and poor quality (Zhao and Oosterhuis, 2002; Dordas, 2006).

Boron is essential for growth and development, and its deficiency negatively affects the yield and seed nutritional quality of cotton (Ahmed et al., 2011; Ahmed et al., 2013). Boron affects cottonseed composition and nutritional quality through the modulation in uptake and translocation of other nutrients as well (Ahmed et al., 2011; Bellaloui et al., 2015). Many studies have revealed boron interactions with other nutrients, such as positive interactions with N, P, K, Cu, Zn and Fe, whereas there were negative interactions with Ca, Mg (Patel and Golakiya, 1986; Lopez-Lefebvre et al., 2002; Dursun et al., 2010), and Mn (Sotiropoulos et al., 2002). Although B nutrition improves the yield and quality of crops, B use efficiency is low due to many plant and soil factors. Therefore, finding means to improve the nutrient uptake by plants and translocation to seeds to sustain productivity and seed nutritional quality is an essential goal and challenge of the present era.

Plant growth regulators may be employed to improve crop performance in terms of yield and seed nutritional quality through the modulation of plant growth and physiological processes such as the photosynthetic efficiency and nutrient dynamics within the plant body (Khan et al., 2005; Anjum et al., 2016). Some studies have provided the ground basis that exogenously applied PGRs improve crop productivity and the nutritional quality of crop plants through improved photosynthesis and nutrient uptake and through accumulation within the plant body (Nagel and Lambers, 2002; Agegnehu and Taye, 2004; Niu et al., 2016).

Mepiquat chloride (1,1-dimethylpiperidinium chloride), a plant growth regulator, inhibits the biosynthesis of gibberellic acid concentrations within plant cells, which results in reduced cell size and plant growth (Rademacher, 2000; Almeida and Rosolem, 2012). It modulates plant architecture by reducing leaf area and internodes length, improves light penetration, exalts boll set at lower sympodial branches and first position bolls, and results in improved yield (Nuti et al., 2006; Mao et al., 2014). Furthermore, it exhilarates leaf Chl, CO<sub>2</sub> exchange rate, photosynthesis and alters source-sink relationship (Zhao and Oosterhuis, 2000; Gwathmey and Clement, 2010; Rosolem et al., 2013). It is known that MC affects cottonseed composition and seed nutrient contents in cotton. It enhances cottonseed protein and oil contents, and affects the fatty acid profile (Sawan et al., 2007). Mepiquat chloride enhances cotton root growth by increasing the number of lateral roots, and improves the nutrient uptake and partitioning within the plant body (Zhang et al., 1990; Duan et al., 2004). Sawan et al. (2009) and Yang et al. (2014) noticed an increase in uptake and translocation of N, P, and K to cottonseed in MC-treated plants, thus improving yield and cottonseed quality.

Many studies have been conducted on MC involving cotton; however, the comparative effect of MC on cotton productivity and cottonseed nutritional quality under deficient and adequate B conditions has seldom been studied. This study was conducted to investigate the comparative effect of MC on seed cotton yield, oil and protein yield, and cottonseed nutritional quality under B deficiency and adequate B content conditions.

## MATERIALS AND METHODS

### Experimental details

This 2-year study was conducted at the Agronomic Research Area, of the University of Agriculture, Faisalabad (31.25° N, 73.06° E and 183 m above the sea level), Pakistan during the summer season of 2014 and 2015. The net plot size was 6 m × 3 m. Before sowing the crop, soil samples were collected with the help of an auger at a 0-30 cm depth at different locations of the experimental field during both years. Composite samples were air dried, ground and passed through a 2 mm sieve to remove clods and materials other than soil. Soil was sandy loam, having saturation percentage (28%, 28%) organic matter (1.20, 0.97%), pH (8.1, 8.1), electrical conductivity (1.80, 1.72 dS m<sup>-1</sup>), total soluble salts (18.30, 17.80 mmol L<sup>-1</sup>), exchangeable sodium (8.90, 8.80 mmol L<sup>-1</sup>), calcium carbonate (5.21, 5.12%), total nitrogen (0.051, 0.049%), available phosphorous (7.10, 6.90 ppm), exchangeable potassium (260, 246 ppm), boron (0.47, 0.50 ppm), zinc (1.69, 1.48 ppm) and iron (5.21, 5.10 ppm) during both years (2014 and 2015), respectively. The soil of the experimental site belongs to the Lyallpur soil series (fine silty, mixed, hyperthermic Ustalfic aridisol), Haplargid in the USDA classification and Haplic Yermosols in the FAO classification. Meteorological data during both experimental seasons are shown in Table 1.

### Experimental design and treatments

The experiment was laid out in randomized complete block design with a factorial arrangement having three replications. The experiment consisted of single foliar application of MC at different doses (0 and 70 mg L<sup>-1</sup>) at different growth stages (squaring and flowering stage) and soil application of B at different doses (0, 1, 1.5, 2 and 2.5 kg ha<sup>-1</sup>). The control treatment received sprayed water. B was applied at the time of sowing, using boric acid (17% B). MC [98% SP from Henan Haoyuhang Economic and Trade Co., Ltd] was foliar applied using a Knapsack hand sprayer at a pressure of 207 kPa. The spraying volume used for MC application at the squaring stage was 300 L ha<sup>-1</sup> while at the flowering stage was 350 L ha<sup>-1</sup>.

**Table 1** - Meteorological conditions during the cotton growing seasons of 2014 and 2015

Month	Total rainfall (mm)		Relative humidity (%)		Temperature (°C)						Sunshine (h)	
					Monthly maximum		Monthly minimum		Daily mean			
	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015
May	41.2	17.0	33.2	27.5	36.6	38.7	23.7	24.9	30.1	31.8	10.4	10.4
June	7.1	11.6	33.5	39.0	40.9	38.0	28.1	25.6	34.5	31.8	9.4	9.4
July	57.5	128.0	53.9	61.1	37.0	34.9	28.0	27.0	32.5	31.0	9.0	5.1
August	4.8	48.4	52.7	60.4	37.1	35.9	27.3	26.7	32.2	31.3	9.1	7.0
September	140.2	75.2	61.2	51.6	33.9	35.4	24.5	24.4	29.2	29.9	7.7	8.2
October	3.6	14.5	54.6	52.9	31.3	32.2	19.1	19.1	25.2	25.4	7.8	8.3
November	10.0	8.8	61.7	61.5	26.3	27.1	11.5	12.1	18.9	19.6	7.6	6.6

Source: Agro-meteorology Cell, Department of Crop Physiology, University of Agriculture, Faisalabad, Pakistan.

### Crop husbandry

The cotton cultivar MNH-886 (Containing Bt gene, developed by Central Cotton Research Institute, Multan, Pakistan) was sown on May 22<sup>nd</sup> 2014 and May 25<sup>th</sup> 2015. Seeds were delinted with commercial sulphuric acid (1:10 ratio of seed and acid) and treated with fungicide (dynasty CST 125 FS @ 3 g kg<sup>-1</sup> seed) before sowing. The crop was sown on beds using a dibbler and keeping a plant to plant distance of 25 cm, and a row to row distance of 75 cm. A seed rate of 15 kg ha<sup>-1</sup> was used and 2-3 seeds were sown per hill. Thinning was done 25 days after sowing (DAS) to maintain the recommended plant population. Fertilizers were 200 kg ha<sup>-1</sup> N, 120 kg ha<sup>-1</sup> P and 110 kg ha<sup>-1</sup> K, applied using urea (46% N), diammonium phosphate (18% N: 46% P<sub>2</sub>O<sub>5</sub>) and sulfate of potash (50% K<sub>2</sub>O), respectively, as sources of fertilizers during both cotton growing seasons. All P, K and 1/3 N were applied at sowing, while the remaining N was applied at squaring and boll formation stages in equal splits. Irrigations were performed according to crop and weather conditions. The first irrigation was applied 4 DAS and the second irrigation was applied 7 days after the first irrigation. Subsequent irrigations were applied with an interval of 2 weeks. During the first year, 8 irrigations were applied besides the one applied at sowing, whereas during the second year, 5 irrigations were applied in addition to the one applied at sowing, due to the occurrence of high rainfall (Table 1). Local plant protection measures were followed to keep weeds, diseases and insect pests below the economic threshold level.

### Seed cotton yield

The crop was harvested in two manual pickings. The seed cotton yield obtained from both pickings was summed to calculate the whole seed cotton yield from each plot. The seed cotton yield was expressed as kg ha<sup>-1</sup>. The seed cotton obtained from each picking was mixed and ginned using a roller-type, hand-fed laboratory gin to obtain cottonseeds for further analysis.

### Seed nutritional quality

Cottonseed ash content was determined according to AOAC (1990) by burning dry ground seed samples in porcelain crucibles in a muffle furnace at 550 °C for 4 hours. Seed oil was extracted with petroleum ether using a Soxhlet extractor and percent oil content was determined by using the procedure described by AOAC (1990). Cottonseed oil yield was calculated by using the formula: cottonseed oil yield (kg ha<sup>-1</sup>) = cottonseed yield (kg ha<sup>-1</sup>) × seed oil contents (%). N content in seeds was assayed by Kjeldhal method (Estefan et al., 2013) and the seed protein content was calculated by using the method by AOAC (1990). Cottonseed protein yield was calculated as: cottonseed protein yield (kg ha<sup>-1</sup>) = cottonseed yield (kg ha<sup>-1</sup>) × seed protein contents (%).

### Seed mineral nutrients

Cottonseed samples from each replication were assayed to determine the concentration of macro and micronutrients. The delinted seed samples were sun-dried and in an electric oven at 70 °C for 24 h, and they were ground to powder form (Cyclotec 1093 Sample Mill, Sweden) to pass through a 30 mesh screen. The samples were digested using sulfuric acid and catalyst mixture and N was determined by Kjeldhal distillation method as described by Estefan et al. (2013). P was analyzed by the ammonium-vanadomolybdate colorimetric method at 410 nm, using the same extract (Estefan et al., 2013). In order to determine K and micronutrients, viz. Zn, Fe and Mn, seed samples were soaked over-nightly in di-acid mixture (1:2 of nitric acid and perchloric acid) and digested on a block digester. K was determined by a flame photometer using the method by Chapman and Pratt (1961); while, Zn, Fe and Mn were assayed using an atomic absorption spectrometer, according to the method described by Estefan et al. (2013). Boron was determined by dry ashing the ground seed material in a muffle furnace at 550 °C for 6 h (Chapman and Pratt, 1961). Then, the ash was taken in 0.36N H<sub>2</sub>SO<sub>4</sub> and the B concentration was determined by a spectrophotometer at 420 nm wavelength, using the azomethine-H colorimetric method (Bingham, 1982).

### Statistical analysis

The analysis of the data collected was done by using Microsoft Excel and the statistical software program SPSS19.0. The Fisher's analysis of variance technique was used to analyze data (Steel et al., 1997) and treatment means were compared by using the honest significance difference (HSD) Tukey's test at 5% probability.

## RESULTS AND DISCUSSION

### Seed cotton yield

Seed cotton yield was substantially low under B deficiency, compared to B adequate conditions. Nevertheless, MC application improved significantly the seed cotton yield under both deficient and adequate B conditions, compared to the control treatment, during both years. Maximum increase in seed cotton yield (34-40%) was caused by MC application at squaring along with 2.5 kg ha<sup>-1</sup> B, compared to the control treatment. Furthermore, MC application at squaring and 2 kg ha<sup>-1</sup> B as well as MC application at flowering and 2.5 kg ha<sup>-1</sup> B application produced similar results (Table 2).

### Cottonseed nutritional quality

Poor cottonseed nutritional quality was perceived under B deficiency, compared to B adequate conditions, during both years. Maximum increase in cottonseed ash (7%), oil (4-6%) and protein contents (5%) was caused by 2.5 kg ha<sup>-1</sup> B, compared to the control treatment. However, the effect of 1-2.5 kg ha<sup>-1</sup> B was at par with each other except for ash contents during the first year, in which the effect of 1.5-2.5 kg ha<sup>-1</sup> B was similar. Furthermore, MC application improved considerably the seed nutritional quality under both B conditions, compared to the control treatment. Although the interaction of MC and B was non-significant, a maximum increase in all studied traits was caused by MC application under adequate B conditions. Mepiquat chloride application at squaring exalted ash (5-8%), oil (4-5%) and protein (4%) contents during both years, compared to the control treatment, and it was followed by MC application at flowering stage. However, during the second year, the effect of MC application at both growth stages differed for seed ash content (Table 2).

### Cottonseed oil and protein yield

Under B deficiency conditions, cottonseed oil and protein yield were substantially low, compared to adequate B conditions. However, applying MC enhanced oil and protein yield under

**Table 2** - Comparative effect of MC on seed cotton yield and seed nutrition of cotton under deficient and adequate B conditions

Treatment	2014				2015			
	M <sub>0</sub>	M <sub>1</sub>	M <sub>2</sub>	Mean	M <sub>0</sub>	M <sub>1</sub>	M <sub>2</sub>	Mean
Seed cotton yield (kg ha <sup>-1</sup> )								
Control treatment	2219 f	2440 def	2384 ef	2348 C	2125 f	2319 ef	2250 ef	2231 C
1 kg ha <sup>-1</sup> B	2366 ef	2479 c-f	2471 c-f	2439 BC	2204 ef	2410 cde	2315 ef	2310 C
1.5 kg ha <sup>-1</sup> B	2423 def	2647 cd	2542 cde	2537 B	2273 ef	2565 bcd	2415 cde	2418 B
2 kg ha <sup>-1</sup> B	2501 cde	2925 ab	2727 bc	2718 A	2329 def	2696 ab	2598 bc	2541 A
2.5 kg ha <sup>-1</sup> B	2487 cde	3099 a	2917 ab	2834 A	2309 ef	2841 a	2738 ab	2629 A
Mean	2399 C	2718 A	2608 B		2248 C	2566 A	2463 B	
Ash content (%)								
Control treatment	3.97	4.31	4.39	4.22 C	4.16	4.55	4.40	4.37 B
1 kg ha <sup>-1</sup> B	4.05	4.42	4.43	4.30 BC	4.36	4.59	4.47	4.47 AB
1.5 kg ha <sup>-1</sup> B	4.21	4.52	4.42	4.38 ABC	4.55	4.69	4.55	4.60 AB
2 kg ha <sup>-1</sup> B	4.30	4.56	4.48	4.45 AB	4.55	4.80	4.55	4.63 A
2.5 kg ha <sup>-1</sup> B	4.36	4.70	4.52	4.53 A	4.58	4.78	4.60	4.66 A
Mean	4.18 B	4.50 A	4.45 A		4.44 B	4.68 A	4.52 B	
Oil content (%)								
Control treatment	19.11	20.15	19.84	19.70 B	18.13	19.24	18.87	18.75 B
1 kg ha <sup>-1</sup> B	19.67	20.19	19.97	19.94 AB	18.82	19.88	18.87	19.19 AB
1.5 kg ha <sup>-1</sup> B	19.97	20.59	20.02	20.19 AB	19.14	19.89	19.56	19.53 AB
2 kg ha <sup>-1</sup> B	20.28	20.93	20.41	20.54 AB	19.35	20.14	19.56	19.68 AB
2.5 kg ha <sup>-1</sup> B	20.23	20.93	20.53	20.56 A	19.38	20.44	19.79	19.87 A
Mean	19.85 B	20.56 A	20.15 AB		18.96 B	19.92 A	19.33 AB	
Oil yield (kg ha <sup>-1</sup> )								
Control treatment	262 f	304 cde	291 def	286 C	240 g	278 def	265 efg	261 C
1 kg ha <sup>-1</sup> B	282 ef	303 cde	298 c-f	294 BC	255 fg	295 cde	269 efg	273 C
1.5 kg ha <sup>-1</sup> B	292 def	328 bcd	305 cde	308 B	266 efg	312 bc	289 cde	289 B
2 kg ha <sup>-1</sup> B	304 cde	365 ab	333 bc	334 A	276 ef	330 ab	308 bcd	305 A
2.5 kg ha <sup>-1</sup> B	302 cde	386 a	356 ab	348 A	273 efg	352 a	329 ab	318 A
Mean	288 C	337 A	317 B		262 C	313 A	292 B	
Protein content (%)								
Control treatment	19.63	20.52	20.11	20.08 B	19.42	20.37	19.72	19.84 B
1 kg ha <sup>-1</sup> B	20.19	20.86	20.61	20.56 AB	19.65	20.48	20.05	20.06 AB
1.5 kg ha <sup>-1</sup> B	20.38	21.21	20.95	20.85 AB	19.86	20.73	20.37	20.32 AB
2 kg ha <sup>-1</sup> B	20.71	21.46	21.10	21.09 AB	20.42	20.98	20.79	20.73 AB
2.5 kg ha <sup>-1</sup> B	20.71	21.58	21.27	21.18 A	20.47	21.29	20.79	20.85 A
Mean	20.32 B	21.12 A	20.81 AB		19.97 B	20.77 A	20.34 AB	
Protein yield (kg ha <sup>-1</sup> )								
Control treatment	270 g	310 def	295 fg	291 C	258 g	294 def	277 efg	276 C
1 kg ha <sup>-1</sup> B	289 fg	313 def	308 ef	303 BC	267 fg	304 cde	286 efg	285 C
1.5 kg ha <sup>-1</sup> B	298 fg	337 cde	319 def	318 B	276 efg	325 bcd	301 cde	301 B
2 kg ha <sup>-1</sup> B	311 def	374 ab	344 bcd	343 A	291 ef	344 ab	327 bc	321 A
2.5 kg ha <sup>-1</sup> B	309 def	398 a	369 abc	359 A	288 efg	367 a	346 ab	333 A
Mean	295 C	346 A	327 B		276 C	327 A	307 B	

Interactions and main effects sharing the same letter for a parameter during an experimental year are not significantly different according to Tukey's honest significance difference (HSD) test at  $p = 0.05$ ; M<sub>0</sub>: control treatment, M<sub>1</sub>: Mepiquat chloride application at squaring, M<sub>2</sub>: Mepiquat chloride application at flowering.

both deficient and adequate B conditions, compared to the MC-free control treatment; however, the improvement by MC application was much lower under B deficiency conditions than under adequate B conditions, during both years. Maximum increase in cottonseed oil yield (47%) as well as protein yield (42-47%) was caused by MC application at squaring, along with 2.5 kg ha<sup>-1</sup> B, during both years. However, the effect of applying MC at squaring, along with 2 kg ha<sup>-1</sup> B and MC application at flowering in combination with 2.5 kg ha<sup>-1</sup> B was similar (Table 2).

### Cottonseed macronutrients concentration

A considerable reduction in the concentration of macronutrients in cottonseed tissues was caused by B deficiency, compared to adequate B conditions. Under adequate B conditions, a maximum increase in the concentration of N (5%), P (13-19%) and K (10-13%) in cottonseed tissues was noticed at 2.5 kg ha<sup>-1</sup> B application during both years, compared to the control treatment; however, at 2 kg ha<sup>-1</sup> B the results were similar. Furthermore, the application of MC improved the concentration of macronutrients in cottonseed tissues under both deficient and adequate B conditions. Nonetheless, the interaction of MC and B was non-significant. Mepiquat chloride application at squaring caused a great increase in the concentration of N (4%), P (8-11%) and K (9-14%) in cottonseed tissues during both years, compared to the control treatment; however, the increase in the N contents of seeds by MC application at flowering was statistically similar with a slightly lower effect (Table 3).

**Table 3** - Comparative effect of MC on macronutrient concentrations in seed tissues of cotton under deficient and adequate B conditions

Treatment	2014				2015			
	M <sub>0</sub>	M <sub>1</sub>	M <sub>2</sub>	Mean	M <sub>0</sub>	M <sub>1</sub>	M <sub>2</sub>	Mean
Seed N content (mg g <sup>-1</sup> DW)								
Control treatment	31.41	32.83	32.17	32.14 B	31.08	32.59	31.55	31.74 B
1 kg ha <sup>-1</sup> B	32.31	33.38	32.98	32.89 AB	31.44	32.77	32.08	32.10 AB
1.5 kg ha <sup>-1</sup> B	32.62	33.93	33.52	33.36 AB	31.78	33.17	32.59	32.51 AB
2 kg ha <sup>-1</sup> B	33.13	34.35	33.76	33.75 A	32.67	33.57	33.27	33.17 AB
2.5 kg ha <sup>-1</sup> B	33.13	34.52	34.04	33.90 A	32.75	34.06	33.27	33.36 A
Mean	32.52 B	33.80 A	33.29 AB		31.94 B	33.23 A	32.55 AB	
Seed P content (mg g <sup>-1</sup> DW)								
Control treatment	3.45	3.88	3.64	3.66 C	3.27	3.52	3.37	3.39 C
1 kg ha <sup>-1</sup> B	3.72	4.00	4.00	3.90 B	3.36	3.72	3.46	3.52 BC
1.5 kg ha <sup>-1</sup> B	3.82	4.07	3.99	3.96 AB	3.53	3.80	3.63	3.65 B
2 kg ha <sup>-1</sup> B	3.97	4.22	4.09	4.09 A	3.63	3.98	3.98	3.86 A
2.5 kg ha <sup>-1</sup> B	3.97	4.29	4.12	4.13 A	3.72	4.33	4.00	4.02 A
Mean	3.79 C	4.09 A	3.97 B		3.50 C	3.87 A	3.69 B	
Seed K content (mg g <sup>-1</sup> DW)								
Control treatment	18.65	20.88	19.83	19.79 C	17.67	19.24	18.63	18.51 C
1 kg ha <sup>-1</sup> B	19.27	20.82	20.78	20.29 BC	18.36	19.74	19.30	19.13 BC
1.5 kg ha <sup>-1</sup> B	19.76	22.40	21.61	21.25 ABC	18.76	20.05	19.72	19.51 ABC
2 kg ha <sup>-1</sup> B	19.84	23.24	21.60	21.56 AB	19.26	21.19	20.09	20.18 AB
2.5 kg ha <sup>-1</sup> B	20.40	24.17	22.48	22.35 A	19.57	21.77	19.95	20.43 A
Mean	19.58 C	22.30 A	21.26 B		18.72 C	20.40 A	19.54 B	

Interactions and main effects sharing the same letter for a parameter during an experimental year are not significantly different according to Tukey's honest significance difference (HSD) test at  $p = 0.05$ ; M<sub>0</sub>: control treatment, M<sub>1</sub>: Mepiquat chloride application at squaring, M<sub>2</sub>: Mepiquat chloride application at flowering.

### Cottonseed micronutrients concentration

B deficiency lead to a decrease in micronutrient concentration in cottonseed tissues, compared to adequate B conditions. However, a significant improvement was caused by MC application under all B treatments. Application of MC at squaring, along with 2.5 kg ha<sup>-1</sup> B, exaggerated the B concentration (49-59%) in cottonseed tissues to the uppermost level compared to the control treatment, during both years. However, during the first year, the concentration of B in seeds was similar by the influence of MC application at the squaring stage at 2 kg ha<sup>-1</sup> of B application, as well as MC application at the flowering stage at 2.5 kg ha<sup>-1</sup> of B. During the second year, B concentration in seed tissues was similar by the effect of MC application at flowering and 2.5 kg ha<sup>-1</sup> of B application.

Similar to B, the concentration of Zn and Fe was greater under adequate B conditions, compared to B deficiency, except for Mn, which was reduced with increased doses of B. The maximum concentration of Zn (9-12%) and Fe (9-14%) occurred at 2.5 kg ha<sup>-1</sup> of B. However, the application of 1.5-2.5 kg ha<sup>-1</sup> of B produced statistically similar results. The interaction of MC and B was non-significant, but a great increase in the concentration of Zn and Fe by MC application occurred under adequate B conditions. The application of MC at squaring exaggerated the concentration of Zn (8%) and Fe (6-9%) in cottonseed tissues during both years, compared to the control treatment; however, the effect of MC application at flowering was similar during both years. Nevertheless, the concentration of Mn in cottonseed decreased with the increase in soil applied with B dosages, with maximum decrease occurring at 2.5 kg ha<sup>-1</sup> of B. Similarly, MC application at the squaring stage exhibited maximum decrease in the Mn concentration of seeds (6-9%) during both years, compared to the control treatment; however, during the second year, the effect of MC application at flowering was similar (Table 4).

The results revealed that seed cotton yield was low under B deficiency conditions compared to adequate B conditions (Table 2). The higher yield under adequate B conditions might be attributed to an increased boll retention and boll weight because B is involved in the reproductive

**Table 4** - Comparative effect of MC on micronutrient concentrations in seed tissues of cotton under deficient and adequate B conditions

Treatment	2014				2015			
	M <sub>0</sub>	M <sub>1</sub>	M <sub>2</sub>	Mean	M <sub>0</sub>	M <sub>1</sub>	M <sub>2</sub>	Mean
Seed B content (µg g <sup>-1</sup> DW)								
Control treatment	35.23 g	39.05 efg	37.91 fg	37.40 C	31.16 h	35.68 fg	33.15 gh	33.33D
1 kg ha <sup>-1</sup> B	40.03 d-g	42.94 c-f	40.77 def	41.25 B	34.79 fgh	37.11ef	35.93 fg	35.94 C
1.5 kg ha <sup>-1</sup> B	41.91 def	44.56 cd	42.72 c-f	43.06 B	36.14 fg	41.23 bcd	39.83 cde	39.07 B
2 kg ha <sup>-1</sup> B	43.52 cde	52.53 ab	47.93 bc	47.99 A	37.49 ef	44.02 ab	41.83 bcd	41.11 A
2.5 kg ha <sup>-1</sup> B	43.86 cde	56.10 a	50.82 ab	50.26 A	38.24 def	46.37 a	43.07 abc	42.56 A
Mean	40.91 C	47.04 A	44.03 B		35.56 C	40.88 A	38.76 B	
Seed Zn content (µg g <sup>-1</sup> DW)								
Control treatment	41.71	45.81	43.67	43.73 C	38.37	41.61	40.47	40.15 B
1 kg ha <sup>-1</sup> B	43.43	45.76	45.80	45.00 BC	39.67	42.89	41.41	41.32 AB
1.5 kg ha <sup>-1</sup> B	44.97	47.41	45.88	46.09 ABC	41.27	43.77	42.28	42.44 AB
2 kg ha <sup>-1</sup> B	45.71	48.23	47.56	47.17 AB	41.47	45.01	43.97	43.48 A
2.5 kg ha <sup>-1</sup> B	45.90	51.47	49.33	48.90 A	42.43	45.69	43.67	43.93 A
Mean	44.34 B	47.73 A	46.45 A		40.64 B	43.79 A	42.36 AB	
Seed Mn content (µg g <sup>-1</sup> DW)								
Control treatment	24.37	23.04	23.97	23.79 A	21.73	20.94	20.90	21.19 A
1 kg ha <sup>-1</sup> B	23.82	21.88	22.78	22.83 AB	21.42	20.17	20.65	20.75 AB
1.5 kg ha <sup>-1</sup> B	23.18	21.16	22.49	22.28 BC	20.43	19.41	19.59	19.81 BC
2 kg ha <sup>-1</sup> B	22.70	20.20	21.96	21.62 BC	20.09	18.33	19.75	19.39 C
2.5 kg ha <sup>-1</sup> B	22.30	20.09	20.47	20.95 C	19.43	18.16	19.19	18.92 C
Mean	23.27 A	21.27 C	22.33 B		20.62 A	19.40 B	20.02 AB	
Seed Fe content (µg g <sup>-1</sup> DW)								
Control treatment	172.57	173.56	174.78	173.64 D	166.57	175.09	170.39	170.68 C
1 kg ha <sup>-1</sup> B	175.76	180.68	181.19	179.21 CD	169.84	178.66	178.59	175.69 BC
1.5 kg ha <sup>-1</sup> B	180.87	187.79	184.19	184.28 BC	175.65	185.71	180.32	180.56 AB
2 kg ha <sup>-1</sup> B	184.37	197.32	188.43	190.04 AB	178.26	190.76	185.35	184.79 AB
2.5 kg ha <sup>-1</sup> B	187.63	207.98	196.68	197.43 A	178.03	195.96	185.71	186.57 A
Mean	180.24 B	189.47 A	185.06 AB		173.67 B	185.24 A	180.07 A	

Interactions and main effects sharing the same letter for a parameter during an experimental year are not significantly different according to Tukey's honest significance difference (HSD) test at p = 0.05; M<sub>0</sub>: control treatment, M<sub>1</sub>: Mepiquat chloride application at squaring, M<sub>2</sub>: Mepiquat chloride application at flowering.



growth, mainly in pollen development, pollen germination and pollen tube growth (Lee et al., 2009). Furthermore, B is involved in sugar synthesis, metabolism and translocation, thus affecting the source-sink relationship (Mengel and Kirkby, 2001; Barker and Pilbeam, 2007). In this study, the application of MC exhilarated the seed cotton yield under both deficient and adequate B conditions, compared to the control treatment; though the improvement under adequate B conditions was much higher than B deficiency conditions (Table 2). Mepiquat chloride alters the source-sink relationship indicating the bolls on treated plants as a larger sink for photosynthates (Gwathmey and Clement, 2010), which might be the reason for enhanced boll retention and boll weight, leading to a higher yield. Higher seed cotton yield by MC application under adequate B conditions might be due to higher B uptake and translocation under adequate B conditions than under B deficiency conditions, which led to greater assimilate partitioning and boll retention. Similar results were reported by Yang et al. (2014), that MC application improved the lint yield under both deficient and adequate K conditions, due to improved K uptake and translocation.

The nutritional quality of cottonseed was reduced by B deficiency, which was improved by MC application under either soil B conditions, compared to the control treatment (Table 2). Higher seed ash, oil and protein contents under adequate B conditions might be due to improved C and N assimilation and metabolism (Ahmed et al., 2014). It is also evident from this study that results exhibiting the improved N concentration in seed tissues by B application indicate enhanced uptake and translocation (Table 3). Bellaloui et al. (2015) reported improved cottonseed protein and oil contents in response to B application, compared to the control treatment. Likewise, the improvement in seed protein contents by MC application is attributed to its role in protein synthesis, through the enhanced conversion of amino acids into protein (Wang and Chen, 1984). On the other hand, the increase in oil content by MC application may be attributed to improved assimilation and translocation of photosynthates (Zhao and Oosterhuis, 2000; Gwathmey and Clement, 2010). A similar increase in protein and oil contents was reported by Sawan et al. (2007).

Oil and protein yields were lower under B deficiency than under adequate B conditions. The application of MC exaggerated oil and protein yields under both B conditions, compared to the control treatment (Table 2). Higher cottonseed oil and protein yield might be due to higher yield, oil and protein contents under adequate B conditions. Results from the present study indicate that the application of MC is an important management strategy to enhance cottonseed oil and protein yield under both deficient and adequate B conditions. Moreover, higher oil and protein yield by MC application under adequate B conditions also indicates that potential benefits from MC can be obtained under adequate B supply. Similarly, Sawan et al. (2001) reported higher oil and protein yields in cottonseeds by applying MC under adequate N and Zn conditions, compared to deficient conditions.

In this study, the concentration of macronutrients was lowered by B deficiency compared to adequate B conditions. Nevertheless, the application of MC exalted cottonseed N, P and K concentrations under all B treatments (Table 3). Exalted levels of macronutrients under adequate B conditions indicate the enhanced uptake and translocation in cotton plants as detected by seed nutrient concentrations. According to Ruiz et al. (1998), B application enhances the uptake and translocation of N, directly by exerting a positive effect on the activation of enzymes involved in the N metabolism and/or indirectly by promoting and regulating the input of substrate through cellular membranes to the interior of cells. Boron has been found to have a synergistic effect with P, and that might be due to its positive effect on P assimilation (Ahmed et al., 2011). Similarly, B has a positive correlation with K, that is, a hyperpolarization function of cell membranes by B, resulting in enhanced accumulation of K in cells (Schon et al., 1990). In this study, MC application enhanced the concentration of N, P and K in seed tissues. Moreover, the higher concentration of nutrients was noticed by applying MC at squaring, which might be attributed to a greater growth reduction than by applying MC at the flowering stage (Table 4). The enhanced N, P and K concentrations in seed tissues by applying MC might be due to exalted uptake and translocation. Mepiquat chloride application enhances nutrient uptake and translocation through improved root growth, CO<sub>2</sub> assimilation and photosynthetic rate, and assimilate partitioning (Zhao and Oosterhuis, 2000; Duan et al., 2004; Gwathmey and Clement, 2010). Sawan (2013) noticed extolled N and K concentration in cotton plants in response to MC applications.

The concentration of micronutrients, viz. B, Zn and Fe in seed tissues was lower under B deficiency than adequate B conditions; however, Mn showed a negative relation with B (Table 4). The results indicate that B has a positive effect on the uptake, as well as on the translocation of micronutrients, except for Mn. The decrease in uptake and translocation of Mn may be attributed to its antagonistic effect with B (Mouhtaridou et al., 2004). Ahmed et al. (2011) reported that the application of B enhanced the concentration of B, Zn and Fe, whereas it decreased the concentration of Mn in leaves and seed tissues in cotton. Our study results show that the application of MC enhanced the concentration of micronutrients, except for Mn, in cottonseed tissues under both B conditions, compared to the control treatment; however, the MC treatments at both growth stages did not differ among each other (Table 4). The improvement in micronutrients, viz. B, Zn and Fe by MC might be due to better root growth and enhanced assimilation rate; however, the mechanism of micronutrient uptake in response to MC application is unknown. Uptake and translocation of B were increased by applying MC, which might be the reason of decreased Mn concentrations due to their antagonistic effect with each other. Furthermore, the range of deficiency and toxicity of micronutrients in plants is very low, and plants develop mechanisms to keep them in balance (Mengel and Kirkby, 2001; Miwa and Fujiwara, 2010). This seems to be the reason why there was no difference among MC treatments at the squaring and flowering stages as for micronutrients.

Seed cotton yield and cottonseed nutritional quality decreased under B deficiency conditions, compared to adequate B conditions. However, the application of MC improved yield and enhanced cottonseed nutritional quality under both deficient and adequate B conditions; however, the improvement was much greater under adequate B conditions. The results conclude that MC has the potential to improve the yield and nutritional quality of cottonseeds under both deficient and adequate B conditions, through improved nutrient dynamics within the plant body. However, the improvement in yield and nutritional quality is much higher under adequate B conditions due to the reduced efficiency of MC caused by B deficiency.

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