Original Research

Efficiency of Foliar Fertilizers and Growth Regulators on Cowpea Productivity and Control of Cowpea Weevil, *Callosobruchus maculatus* (Coleoptera: Bruchidae)

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Abstract

Cowpea, *Vigna unguiculata*, is an important food and forage legume worldwide. The cowpea weevil is a major cowpea pest that causes damage both in the field and during storage. This study investigated the effects of some foliar treatments, including three foliar fertilizers (Ca, K, B) and two growth regulators (mepiquat chloride (MC) and forchlorfenuron (CPPU) on the cowpea yield and its components. Furthermore, evaluating the susceptibility and resistance of cowpea seeds resulting from the previous treatments against the *C. maculatus* insect. The results showed that all foliar sprays had significant effects on yield and its components in both seasons compared with the control. The highest yield was achieved by CPPU treatment, followed by K treatment, and the lowest yield was achieved by MC treatment. The results of the free-choice and no-choice tests revealed that cowpea seeds treated with MC treatment were the most susceptible to *C. maculatus* attack. At the same time, cowpea seeds treated with K and CPPU treatments showed their resistance to infestation by the same insect. Foliar application of K and CPPU treatments could be used in integrated pest management programs to reduce the risk of pesticides to human health and increase crop production.

Keywords: foliar spraying, forchlorfenuron, legume seeds, micronutrients, mepiquat chloride, *Vigna unguiculata*, pulse beetle

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Introduction

Cowpea (Vigna unguiculata L. Walp.) belongs to the family Fabaceae. It has important nutritional and economic importance, as it contains many essential elements, including iron, sodium, potassium, and zinc, as well as fiber and folic acid [1]. Dried cowpea seeds are rich in protein, similar to meat, it is a popular seed among Egyptian customers, and the total cultivated area of cowpea plants in Egypt is approximately 1853 ha with a mean production of 7180 tons of dry seeds [2]. This confirms the need for research to improve cowpea productivity in Egypt. Foliar fertilization is an important technique in sustainable agricultural practices used to increase crop productivity. Potassium (K) is an essential element for plant growth and productivity. It is necessary for enzyme activity, osmotic regulation, and stomatal movement, among other functions [3]. This macronutrient is also important for improving crop production under biotic and abiotic stress factors. Calcium (Ca) is an essential macronutrient involved in normal plant growth, metabolism, and physiological functions [4]. Foliar spraying has advantages in correcting (Ca) deficiency [5]. Foliar fertilization is used to correct nutritional deficiencies, mainly micronutrients such as boron (B), owing to its low availability in soil [6]. Boron deficiency in legumes causes significant losses by restricting reproductive processes [6] and root growth. Plant growth regulators such as Mepiquat chloride (MC) works as a growth retardant that inhibits the production of gibberellic acid, which is involved in cell elongation. Mepiquat chloride (1,1-dimethylpiperidinum chloride) is globally used for canopy development. It changes the sequence of assimilate division, reserve remobilization, and uptake of nutrients. Furthermore, it hampers gibberellic acid biosynthesis, which finally decreases cell division and elongation [7, 8]. N-(2-Chloro-4pyridyl)- N' -phenyl-urea (CPPU) is a member of the synthetic cytokinin group. Low concentrations promote grape berry set and development, increase berry size, and improve storage quality as a plant growth regulator that promotes chlorophyll biosynthesis, cell division, and cell expansion [9]. It also increases the plant set and accelerates plant enlargement [10]. Cowpea is vulnerable to pest infestations during pod and dry seed stages in the field and storage. The pulse beetle, Callosobruchus maculatus (F.) (Coleoptera: Bruchidae), is the most destructive insect to cowpea seeds, resulting in field and storage losses of at least 60% [11]. Callosobruchus maculatus is economically significant because the serious damage caused by the insect reduces the dry weight of seeds, impacts their nutritional value, and reduces their market value. Bioactive compounds can used to increase the growth and yield of plant under insect infestation, in this concern Ahmed et al. [12] concluded that Alstonia scholaris is a natural source of bioactive compounds which can be used for control of Macrophomina phaseolina. Also, the growth and

phytoextraction capability of *Catharanthus roseus* (L.) was improved by inoculation of *P. fluorescens* RB4 and *B. subtilis* 189 [13].

Additionally, application of *B. megaterium* and Put increased level of proline and IAA in plants [14].

Farmers usually apply hazardous pesticides to preserve cowpea from infestation during storage. However, heavy and indiscriminate use of synthetic pesticides creates serious health problems for humans and the environment [15]. In addition, it develops the resistance characteristic and converts secondary pests to major pests. Seeds' color, size, hardness, texture, and biochemical compounds are usually involved in the resistance pathways in legume seeds. Biochemical resistance mechanisms in cowpea seeds involve the chemicals in the seed coat and cotyledon that cause non-preference for oviposition and antibiosis effects on the development of larvae, thus affecting the hatching and prolonged the development of insects [16]. This study was aimed to investigate the effect of using foliar feeding treatments, including fertilizers and growth regulators for increasing the productivity of cowpea crops. Furthermore, to assess their role in giving seeds the characteristic of resistance against C. maculatus insects to reduce the hazards of pesticides. Foliar application of these treatments will reduce the risk of using pesticides on the environment and human health.

Materials and Methods

The cowpea field experiment was performed at El-Baramon Experimental Farm, El Dakahllia Horticulture Research Institute, Egypt, during two successive seasons, 2018/2019 and 2019/2020. The study area is located at latitude 31°2'15.238 and 31.394"N longitude 31°23'11"E and at an elevation of about 11 m above mean sea level.

Experimental Soil Analysis

The physical and chemical properties of the soil of the experimental area (0.0 to 50 cm depth) are given in (Table 1).

Experimental Design

The randomized complete block design was the statistical design of this experiment, with six treatments and three replicates. The plot area was about 12.6 m² (6 ridges, each 3 m length, and 0.7 m width). The Qaha-1 cultivar was sown with 15 cm spacing between plants within the same raw on one side of ridges at a rate of 3-5 seeds at hills by hand at a depth of 2-3 cm and then covered with wet and dry soil. All agriculture practices were carried out as recommended by Hort. Res Inst. Egypt.

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|-------------------------|-----------------------|-------|-------|
| Soil ch | naracters | 2018 | 2019 |
| | Coarse sand | 2.55 | 2.58 |
| | Fine sand | 12.95 | 12.97 |
| Mechanical analysis (%) | Silt | 17.68 | 17.71 |
| , , , | Clay | 66.82 | 66.74 |
| | Texture class | Clay | Clay |
| EC ds | /m (1:5) | 1.47 | 1.52 |
| I | ъН | 7.79 | 7.84 |
| 0. | M.% | 2.52 | 2.49 |
| Са | ACO3 | 1.84 | 1.86 |
| | Fe | 12.06 | 12.11 |
| Micronutrients (ppm) | Mn | 7.03 | 7.06 |
| | Zn | 3.04 | 3.01 |
| | Cu | 0.96 | 0.94 |
| | N | 32.50 | 33.01 |
| Available (ppm) | Р | 6.02 | 6.08 |
| (Tr) | K | 340 | 348 |

Table 1. Physical and chemical analysis of the investigated soil.

Treatment

This experiment had six treatments, including five foliar applications as well as control as follows:

1. Control (sprayed with tap water).

2. Boron tri-ethanol amine (10% B) applied at 1 mL/L.

3. Calcium acetate (5% Ca) applied at 5 mL/L.

4. Potassium acetate (30% K_2O) applied at 5 mL/L.

5. Mepiquat chloride (MC) (25%) applied at 1 mL/L.

6. Forchlorfenuron (1-(2-Chloro-4-pyridyl)-3-phenylurea) (CPPU) (1%) applied at 0.5 mL/L.

Cowpea plants were sprayed three times (20 days after sowing and again every 20 days) with the previous concentrations of each treatment.

Yield and Its Components

Ten plants from each plot at the maturing stage were randomly chosen and marked, and the following characteristics were recorded:

Pod length (cm), pod diameter (cm), pod weight (g), number of pods /plant, number of seeds/pod, and total pods yield (ton/feddan).

Chemical Composition of Pods

Pod samples were prepared to determine the following chemical analysis:

The solution of pods' juice can be titrated with a 2,6-dichlorophenol indophenol blue dye and the volume of the titration used to calculate the ascorbic acid content according to the AOAC method [17].

Total Carbohydrates (%)

Carbohydrates were extracted and determined according to the method described by Yemm and Willis [18]. In this method, the carbohydrates content can be determined by hydrolysis into simple sugars using diluted hydrochloric acid. Glucose is dehydrated to hydroxymethyl furfural which forms a green color solution with anthrone. The color can be measured spectrophotometrically at 630 nm using glucose standard.

Total Soluble Solids (TSS %)

Total soluble solids were directly recorded by using Digital-Bench- Refractometer (HANNA Instruments Inc, Washington, USA) after calibration it with distilled water according to the protocol of AOAC [17].

Protein (%)

The samples are digested in H_2SO_4 to convert the protein to $(NH4)_2SO4$. Ammonia is liberated by alkaline steam distillation and quantified titrimetrically with acidic standard. The crude protein of each sample was determined using the Kjeldahl Method and calculated according to the method of Thiex et al. [19].

Potassium (%)

The potassium content determined in acid digested samples using a flame photometer, according to the method mentioned by Piper [20].

Boron (%)

Boron concentration in digested ground seed tissues was colorimetrically measured at 412 nm using boron standard solution according to the azomethine-H method [21].

Calcium (%)

Calcium was measured in wet digestion of dried and ground samples using a PerkinElmer (1100 B) Atomic Absorption Spectrometer and determined according to Chapman and Pratt [22].

Effect of Treated Cowpea on C. maculatus

Adults of cowpea beetle, C. maculatus, obtained from The Department of Plant Protection Research

Institute, Egypt. Insects were kept in glass jars (one-liter capacity) provided with 500 g of cowpea seeds from the tested fertilizers. These seeds were preheated in an oven at 60°C for one hour to eliminate any infestation by other pests. After one week, these adults were removed from the jars, and the seeds were incubated for two weeks in an incubator set at $30\pm2^{\circ}$ C and $75\pm5\%$ RH. The newly emerged adults (0-2 days old) were used for following bioassay tests.

No-Choice Test (Susceptibility)

A no-choice test was conducted to examine the beetle oviposition preference among the tested treatments by following Messina and Renwick [20]. Twenty-five seeds of each treatment were transferred to small glass jars (11.5 cm in height and 86 cm in diameter), then 25 adult insects of *C. maculatus* (0-2 days old) were placed in each jar. Three replicates of each treatment were used, and each jar was covered with muslin cloth and kept under laboratory conditions. By the end of this test, the total number of deposited eggs and emerged adults for each treatment was recorded. The reduction percentage was calculated using the following equation:

% Reduction = (mean No of emerged adults in control -mean No of emerged adults in treatment)/ (mean No of emerged adults in control) ×100

Free-Choice Test (Preferences)

The food preference chambers technique by Messina and Renwick [23] was used to conduct multiple-choice bioassays. The wood chamber was cylindrical (6 cm height, 30 cm diameter) with a raised arena (2 cm height, 10 cm diameter) in the center of the chamber. It was divided into 6 equal sections by wood partitions. The sections were filled with 25 g of treated grain. One hundred beetles (0-2 days old) *C. maculatus* adults were transferred into the center of the arena (race chamber), confined by a wood ring (2.5 cm height, 5 cm diameter). A circular black-painted plate was used to cover the top of the chamber. The chamber was kept at $30\pm1^{\circ}$ C and $70\pm5\%$ RH for 48 h, and the number of eggs laid was recorded and kept under the same conditions until the newly emerged adult was released. The number of emerged adults recorded, and the percentage of reduction were calculated using the equation mentioned above.

Statistical Analysis

All data were statistically analyzed by analysis of variance (ANOVA), and Tukey's test was used for post hoc comparison using DSAASTAT 2.8 (Onofri, Italy).

Results

Field Experiment

The objective of this experiment was to investigate the effects of three foliar fertilizers (K, Ca, B) and two growth regulators (MC, CPPU) on cowpea productivity. The data in Table 2 show that the tested foliar treatments significantly increased the yield and its components, i.e., pod length (cm), pod diameter (cm), pod weight (g), number of pods/plant, number of seeds/pod and total yield ton/fed compared to the control during the two seasons. As for pod length, CPPU treatment had the greatest effect on this parameter with (14.34 and 15.34 cm) followed by K treatment (13.45, 13.97cm) compared to the control (10.27, 10.76 cm) during the first and

Table 2. Effect of the tested foliar treatments on the pod characters of cowpea plants during the seasons of 2018/2019 and 2019/2020.

| Treatments | Pod leng | gth (cm) | Pod dian | neter (cm) | Pod we | eight (g) | No. of p | ods/plant | No. of s | eeds/pod | | l yield /fed |
|------------|-------------------|-----------------|-----------------|------------|----------|-----------|-----------------|-----------|-----------------|----------|--------------|-----------------|
| | 1^{st} | 2 nd | 1 st | 2^{nd} | 1^{st} | 2^{nd} | 1 st | 2^{nd} | 1 st | 2^{nd} | $1^{\rm st}$ | 2^{nd} |
| Ca | 12.04 | 14.0 | 0.48b | 0.87 | 4.64 | 6.64 | 18.33 | 19.33 | 10.33 | 13.33 | 1.83 | 2.16 |
| | ±0.06d | ±0.06d | ±0.02c | ±0.02bc | ±0.06d | ±0.06d | ±2.6ab | ±2.6ab | ±0.33ab | ±0.33ab | ±0.02d | ±0.02b |
| K | 13.45 | 15.45 | 0.55± | 0.94 | 6.14 | 8.14 | 19.33 | 20.33 | 11.33 | 14.33 | 2.34 | 2.56 |
| | ±0.07b | ±0.07b | 0.02ab | ±0.02ab | ±0.06b | ±0.06b | ±0.2ab | ±2.02ab | ±0.33ab | ±0.33ab | ±0.02b | ±0.04a |
| В | 12.73 | 14.73 | 0.52± | 0.92 | 5.45 | 7.45 | 22.33 | 23.33 | 10.67 | 13.66 | 2.12 | 2.34 |
| | ±0.07c | ±0.07c | 0.02ab | ±0.02ab | ±0.07c | ±0.07c | ±1.4ab | ±1.45ab | ±0.33ab | ±0.33ab | ±0.05c | ±0.05b |
| МС | 11.12 | 12.26 | 0.35± | 0.75 | 4.14 | 5.53 | 15.67 | 16.66 | 9.67 | 11.33 | 1.65 | 1.96 |
| | ±0.06e | ±0.06f | 0.01d | ±0.01d | ±0.05e | ±0.06f | ±0.2ab | ±2.02ab | ±0.33bc | ±0.33c | ±0.02e | ±0.04c |
| CPPU | 14.34 | 16.33 | 0.62 | 1.02 | 6.75 | 8.75 | 24.00 | 25.00 | 12.00 | 15.00 | 2.66 | 2.74 |
| | ±0.04a | ±0.04a | ±0.02a | ±0.02a | ±0.06a | ±0.06a | ±1.7a | ±1.73a | ±0.57a | ±0.57a | ±0.04a | ±0.04a |
| Control | 10.27 | 13.12 | 0.40 | 0.80 | 3.53 | 6.14 | 13.33 | 14.33 | 8.33 | 12.66 | 1.35 | 1.67 |
| | ±0.06f | ±0.05e | ±0.02cd | ±0.02cd | ±0.07f | ±0.05e | ±1.4b | ±1.45b | ±0.33c | ±0.33bc | ±0.02f | ±0.05d |

Means in a column with the same letters are not significantly different at 5%; Ca, calcium; K, potassium; B, boron; MC, mepiquat chloride; CPPU, 1-(2-Chloro-4-pyridyl)-3-phenylurea.

second seasons, respectively. Regarding pod diameter and pod weight, CPPU still has the highest effect, followed by K treatment. The mean number of pods/ plant was the highest in the case of CPPU treatment (24.0 and 26.0 pods/plant), followed by B treatment (22.33 and 23.67 pods/plant) compared to the control (13.33 and 14.67 pods/plant). Additionally, the highest cowpea yield was recorded by CPPU treatment, followed by K, and the lowest production was achieved by MC treatment.

Chemical Content of the Pods

Table 3 shows that all the tested foliar treatments used in this experiment had a statistically significant improvement in the quality of chemical pod contents (carbohydrates, crude protein, vitamin C, and total soluble solids) during the two seasons 2018/2019 and 2019/2020. The highest values of total carbohydrates during the two successive seasons were obtained by using CPPU treatment (52.99%, and 54.85%) followed by K treatment (49.15%, and 53.18%), respectively, compared to the control values (46.51, 47.62%) in the two seasons respectively. In addition, for total soluble solids content, the highest contents were obtained by CPPU treatment (7.14%, 7.85%) followed by k treatment (7.01%, 7.76%) compared to the control (6.53%, 7.07%) during the two seasons respectively. As for crude protein, CPPU treatment (25.33%, 27.09%) recorded the highest values, followed by Ca treatment (24.5%, 25.52%), compared to the control (21.22%, 22.25%). In the case of vitamin C content, CPPU treatment had the highest effect, followed by Ca treatment with values (1.95, 2.16 mg) and (1.56, 1.72 mg), respectively, compared with control (0.43, 0.47%) during the two seasons.

Chemical Content of the Seed

The seed contents of Ca, K, B, carbohydrates, and protein are presented in Table 4 during the two seasons 2018/2019 and 2019/2020. All foliar treatments

significantly enhanced the chemical seed compositions of the cowpea plants during the two seasons when compared to the control. The measurements in Table 4 show that among all treatments, the highest substantial values were recorded for the CPPU treatment. The highest Ca content (%) was recorded for CPPU treatment during the two seasons, followed by MC treatment (1.06%, 1.18%), (0.097%, 1.08%), respectively. CPPU treatment recorded the highest value for K (%) (2.74%, 3.04%), followed by K treatment (2.6%, 2.9%), respectively. In the case of B (%), CPPU treatment recorded the greatest value, followed by Ca treatment (16.75%, 17.92%) and (16.05%, 16.68%), respectively. CPPU treatment gave the highest percentage contents of carbohydrates (61.29%, 65.6%), and protein (27.13%, 29.3%) followed by K treatment (59.14%, 61.32%) for protein and (26.15%, 27.2%) for carbohydrates during the two investigated seasons. The control treatment recorded the lowest value of all chemical seed characters.

Laboratory Experiment

Susceptibility of Treated Cowpea Seeds to C. maculatus Infestation

The data given in Table 5 show the preference among cowpea seed treatments for C. maculatus attack using the free-choice test. The results showed that cowpea seeds treated with K and CPPU were not preferred for oviposition with 220.0 ± 30.5 and 222.0 ± 10.53 eggs, respectively, compared to the control with 283.3±12.01 eggs. While MC treatment was preferred for oviposition with 276.6±12.01 of deposited eggs. The highest reduction percentage in emerged adults was calculated in the case of K and CPPU treatments, with 45% and 40.7%, respectively, while MC treatment had the least effect in the reduction percentage of emerged adults, with 3.98%. The preference for C. maculatus among the treated cowpea seeds in terms of deposited egg numbers and progeny numbers can be arranged as follows MC>B>Ca>CPPU>K in the free-choice test.

Table 3. Effect of the tested foliar treatments on the chemical pod characters of cowpea plant during the seasons of 2018/2019 and 2019/2020.

| Tracting and a | Total carbo | hydrates % | Prote | ein % | Vitamin C | (mg/100 g) | Total solub | ole solids % |
|----------------|-----------------|-----------------|-----------------|-----------------|------------------|-----------------|-------------|-----------------|
| Treatments | 1 st | 2 nd | 1 st | 2 nd | 1 st | 2 nd | 1^{st} | 2 nd |
| Ca | 52.11±0.60a | 50.20±0.61bc | 24.54±0.07b | 27.52±0.07b | $1.56 \pm 0.04b$ | 1.72±0.04b | 6.78±0.04cd | 8.77±0.04cd |
| K | 49.15±0.70bc | 51.18±0.70bc | 22.84±0.03d | 25.76±0.03d | 0.98±0.05cd | 1.09±0.05d | 7.00±0.03ab | 9.01±0.03ab |
| В | 50.54±0.60ab | 52.66±0.34ab | 23.74±0.05c | 26.17±0.05c | 1.21±0.06c | 1.31±0.06c | 6.88±0.03bc | 8.88±0.03bc |
| MC | 47.45±0.55c | 49.88 ±0.70c | 21.82±0.06e | 24.13±0.06e | 0.75±0.06d | 0.83±0.06e | 6.66±0.03de | 8.65±0.03de |
| CPPU | 52.99±0.34a | 54.85 ±0.61a | 25.33±0.06a | 28.09±0.06a | 1.95±0.05a | 2.16±0.05a | 7.14 ±0.02a | 9.14±0.03a |
| Control | 46.51±0.72c | 47.62±0.70d | 21.22±0.06f | 23.25±0.06f | 0.43±0.06e | 0.47±0.06f | 6.53 ±0.03e | 8.53±0.03e |

Means in a column with the same letters are not significantly different at 5%; Ca, calcium; K, potassium; B, boron; MC, mepiquat chloride; CPPU, 1-(2-Chloro-4-pyridyl)-3-phenylurea.

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| Two devices to | Ca | Ca % | K % | % | B (mg/kg) | g/kg) | Total carbohydrates % | hydrates % | Protein % | in % |
|------------------|--------------------|----------------------|----------------------|-------------------|---------------------|-----------------|-----------------------|---|-------------------|-------------|
| HEAUNCHUS | 1 st | 2^{nd} | 1 st | 2 nd | 1 st | 2 nd | 1 st | 2 nd | 1 st | 2^{nd} |
| Ca | 0.81±0.02c | 0.90±0.02d | 2.51±0.02b | 2.76±0.02c | 16.05±0.07b | 16.68±0.07b | 57.32±0.54bc | 59.84±0.54bc | 25.19±0.04c | 25.86±0.04c |
| K | 0.73±0.02c | 0.81±0.02e | 2.60±0.02b | 2.90±0.02b | 14.56±0.07d | 15.15±0.07d | 59.14 ±0.7ab | 59.14 ±0.7ab 61.32±0.70ab | 26.15±0.04b | 27.20±0.04b |
| B | 0.92±0.01b | 1.00±0.01c | 2.34±0.02c | 2.54±0.02d | 15.34±0.06c | 16.24±0.06c | 54.67±0.45cd | 54.67±0.45cd 56.79±0.45cd | 24.09±0.37d | 25.53±0.37c |
| MC | 0.97±0.01b | 1.08±0.01b | 2.22±0.02d | 2.46±0.02e | 13.91±0.05e | 14.76±0.05e | 52.46±0.56de | 54.83±0.56de | 23.54±0.04d | 24.94±0.04d |
| СРРU | 1.06±0.02a | 1.18±0.02a | 2.74±0.02a | 3.04±0.02a | 16.75±0.07a | 17.92±0.07a | 61.29 ±1.10a | 63.60 ± 1.10a | 27.13±0.06a | 29.03±0.06a |
| Control | 0.64±0.01d | 0.71±0.01f | 2.14±0.01d | 2.33±0.01f | 13.35±0.07f | 14.03±0.07f | 49.49 ±0.79e | 51.81 ±0.80e | 22.61±0.04e | 23.75±0.04e |
| Means in a colur | nn with the same l | etters are not signi | ficantly different a | t 5%; Ca, calcium | ı; K, potassium; B, | boron; MC, mepi | iquat chloride; CPI | Means in a column with the same letters are not significantly different at 5%; Ca, calcium; K, potassium; B, boron; MC, mepiquat chloride; CPPU, 1-(2-Chloro-4-pyridyl)-3-phenylurea. | -pyridyl)-3-pheny | lurea. |

The data summarized in Table 6 show the susceptibility of five cowpea treatments to *C. maculatus* infestation using the no-choice test. Based on the obtained results, the mean number of deposited eggs and emerged adults was the highest in B treatment with 239.6±15.1 and 118.3±16. 41, respectively. While K (141.6±13.01 eggs and 70±5.7 emerged adults) and CPPU (148.6±14.3 eggs and 73.6±8.92 emerged adults) treatments were the best treatment in reducing the mean number of deposited eggs and emerged adults, respectively. It was concluded from both the free-choice and no-choice tests that cowpea seeds treated with B and MC were more susceptible and preferred to *C. maculatus* infestation, whereas cowpea seeds treated with K and CPPU were less susceptible to infestation by the same insect.

Discussion

Foliar fertilization is the most efficient way to increase yield production and improve crop health [24]. The results obtained in this study showed that all foliar nutrients, including Ca, K, B, CPPU, and MC, play an important role in cowpea productivity. Our findings showed that the use of CPPU as a foliar growth regulator was superior to other foliar treatments in improving the quality and productivity of the cowpea crop, followed by K treatment as a foliar fertilizer. Based on the chemical analysis of the pods and seeds, our findings showed that CPPU followed by K treatment resulted in the highest carbohydrate and protein values compared to other treatments. This increase in protein and carbohydrate content could be explained by increasing productivity in the case of CPPU and K treatments. Potassium (K) is an essential element in crop growth and is classified as a macronutrient because the plant needs it in large quantities. Many important enzymes, including those involved in protein synthesis, sugar transport, N and C metabolism, and photosynthesis, are activated by K [3]. Regarding the mechanism of K as a growth promoter, there is evidence that K regulates ATPase in the plasma membrane to provide acid stimulation, which then induces the loosening of the cell wall and activation of hydrolysis enzymes [3]. Our findings are in agreement with those of Babita [25], who reported that CPPU is one of the most significant growth regulators with a highly active synthetic cytokinin that promotes protein and chlorophyll biosynthesis and increases cell division, and cell expansion. These effects reflect positively the quality and quantity of yield [26].

Furthermore, Zeng et al. [9] proposed that CPPU encouraged the mobilization of carbohydrates from the leaves to macadamia fruit. Our results on the effect of K were assured by Gowthami et al. [27], who found that foliar application of K had a significant effect on the soybean seed yield of soybean at 2506.67 kg/ha compared to the control at 2330.0 kg/ha. As for B as a foliar fertilizer on cowpea crops, the results showed that this treatment occupied an average position in

| Treatment | Mean no. (±SE) of eggs | Mean no. (±SE) of emerged adults | % Reduction of emerged adults |
|-----------|------------------------|----------------------------------|-------------------------------|
| Са | 230.0±11.16 bc | 130.0±5.32 b | 35.32 |
| K | 220.0±11.55 c | 110.0±10.0 b | 45.27 |
| В | 257.3±9.33 abc | 135.0±12.27 b | 32.83 |
| CPPU | 229.0±10.53 bc | 126.0±8.02 b | 37.3 |
| MC | 276.6±12.01 ab | 193.3±8.22 a | 3.98 |
| Control | 283.33±12.01 a | 201.0±2.08 a | - |

Table 5. Preferability of treated cowpea seeds to Callosobruchus maculatus infestation under free choice conditions.

Means in a column with the same letters are not significantly different at 5%; Ca, calcium; K, potassium; B, boron; MC, mepiquat chloride; CPPU, 1-(2-Chloro-4-pyridyl)-3-phenylurea.

Table 6. Susceptibility of treated cowpea seeds to Callosobruchus maculatus infection under no-choice conditions.

| Treatment | Mean no. (±SE) of eggs | Mean no. (±SE) of emerged adults | % Reduction of emerged adults |
|-----------|------------------------|----------------------------------|-------------------------------|
| Са | 161.6 ±9.6 b | 81.6±4.27 b | 2.04 |
| K | 141.6±13.01 c | 70.0±5.77 c | 16 |
| В | 239.6±15.1 a | 118.3±6.41 a | -41.6 |
| CPPU | 156.6±14.3 b | 81.6±8.92 b | 2.04 |
| MC | 196.6±6.0 ab | 90.0±5.77 ab | - 8.0 |
| Control | 163.3±14.27 b | 83.3±6.81 b | - |

Means in a column with the same letters are not significantly different at 5%; Ca, calcium; K, potassium; B, boron; MC, mepiquat chloride; CPPU, 1-(2-Chloro-4-pyridyl)-3-phenylurea.

increasing the production of cowpea yield after CPPU and K treatments. Boron (B) is a micronutrient that plants need in low amounts compared to others. B plays an important role in cell wall biosynthesis and plasma membrane structure and integrity. Boron-deficient plants have been reported to possess reduced pollen viability [28]. Soil B application gives a grain yield (4.7 t/ha), but foliar B application decreased grain yield by 10.9% compared to the control in rice plants [29].

The calcium foliar fertilizer ranked fourth among the treatments for increasing the productivity of cowpea. These results might be related to the effect of calcium on pollen germination and pollen tube growth, as well as increasing fertility. Our results showed that MC treatment had the lowest rank among the treatments in cowpea yield production. It is known that MC, a plant growth regulator, has been used worldwide to reduce excessive vegetative growth, speed up crop maturation, and prevent yield losses in cotton production [30]. The effect of MC on cowpea yield may be related to its role as a plant growth retardant. Similar observations were made by Naik et al. [30], who reported that spraying of MC at 500 ppm on cluster beans caused the lowest pod yield per plot.

The results of our study proved that the cowpea seeds treated with K and CPPU during cultivation were resistant and not preferred to attack by *C. maculatus*

compared to the other treatments. While the cowpea seeds treated with B and MC were more susceptible and preferred to C. maculatus infestation. Referring to the chemical analysis of seeds, we observed that the highest carbohydrate, protein, and K contents were in the seeds treated with CPPU and K. While the lowest contents were recorded in seeds treated with MC and B. Our results regarding susceptibility and resistance of cowpea seeds against C. maculatus are less identical to some previous works. Some earlier studies mentioned that the high resistance of seeds against stored product insects is correlated with the high carbohydrate and low protein content of seeds. Other previous studies have suggested that the seed coat of cowpea cultivars may be the main factor affecting oviposition and larval development of C. maculatus. Biochemical resistance mechanisms in cowpea seeds involve the chemical characteristics in the seed coat and cotyledon exerting non-preference for oviposition and antibiosis effects on the development of larvae, thus influencing the larval hatching and prolonging the individual development [31]. Additionally, Saad et al. [32] showed that Romanian, American, and American wheat cultivars were the most damaged by Sitophilus oryzae, which may be attributed to the high moisture content and chemical composition of wheat varieties. In summary, the specific reason why some seeds were kept without being attacked by pests is still under debate and may be related to the variation in the physical and chemical contents of seeds. In a recent study, the application of potassium 30 kg/ha in combination with 100 ppm GA3 (T7) recorded higher performance in growth parameters, yield, and yield attributes in Cowpea plants [33].

The results of this study recommend the use of CPPU and K treatments as foliar sprays on cowpea plants to improve their quality, productivity, and resistance to *C. maculatus* infestation. In particular, the infestation by this insect usually starts in the field and transfers to storage places. This will reduce the risk of using pesticides on the environment and human health. Although, further toxicological studies are needed to investigate the safety of these treatments on human health.

Conclusion

The current study demonstrated that using K and CPPU as foliar nutrients in cowpea crop led to a significant increase in quality and production yield. In addition, increasing the cowpea resistance against C. maculatus infestation. Foliar spraying of K and CPPU treatments could be used in integrated pest management programs to reduce the risk of pesticides.

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Conflict of Interests

The authors declare no conflict of interest.

References

- MAHDI S.F. The effect of foliar spraying with the amino acid arginine on some vegetative and yield for cowpea plant *Vigna unguiculatal* L. exposed to salt stress. Journal of Survey in Fisheries Sciences. **10** (38), 2796, **2023**.
- FAOSTAT. Statistical Databases. Food and Agriculture Organization of the United Nations. https://www.fao.org/ faostat/en/. Accessed 20 July 2021.
- WHITE P.J. Potassium in Crop Physiology. In Achieving Sustainable Crop Nutrition; Rengel, Z., Ed.; Burleigh Dodds Science Publishing: Cambridge, UK, 2020; pp. 213-236. ISBN 978-0-429-27584-5.
- 4. YOU X., TENG J., CHEN Y., LONG Y., YU G., SHEN L., LIN H. New insights into membrane fouling by alginate: Impacts of ionic strength in presence of calcium ions. Chemosphere **246**, **2020**.
- CORREIA S., QUEIRÓS F., FERREIRA H., MORAIS M.C., AFONSO S., SILVA A.P. GONÇALVES B. Foliar

application of calcium and growth regulators modulate sweet cherry (*Prunus avium* L.) tree performance. Plants **9**, 410, **2020**.

- CHATTERJEE M., TABI Z., GALLI M., MALCOMBER S., BUCK A., MUSZYNSKI M., GALLAVOTTI A. The boron efflux transporter ROTTEN EAR is required for maize inflorescence development and fertility. Plant Cell, 26, 2962, 2014.
- RAUT S.A., JAYANT M.H, LAL E.P. Effect of mepiquat chloride on cotton var Suraj shoot and root growth behavior. International Journal of Communication Systems. 7 (3), 946, 2019.
- ABBAS H., WAHID M.A., SATTAR A., TUNG S.A., SALEEM M.F., IRSHAD S. Foliar application of mepiquat chloride and nitrogen improves yield and fiber quality traits of cotton (*Gossypium hirsutum* L.). PLoS ONE 17 (6), e0268907, 2022.
- SENTHILKUMAR S., VIJAYAKUMAR R.M., SOORIANATHASUNDARAM, K., DURGA DEVI D. Role of CPPU and Application Stages with Other PGR's on Bunch and Berry Characters in New Grape cv. Italia under Tamil Nadu Conditions Int. J. Curr. Microbiol. App. Sci. 7 (4), 353, 2018.
- ZENG H., YANG W., LU C., LIN W., ZOU M., ZHANG H., HUANG X. Effect of CPPU on carbohydrate and endogenous hormone levels in young macadamia fruit. PLoS One, 11 (7), 2016.
- 11. OGUNKANMI L.A., OGUNDIPE O.T., AMUSA O.D., BOLARINWA B., AKINDELE O.O. Comparative evaluation of cold and container storage preservation efficiencies on cowpea grains against *Callosobruchus maculatus* Fab. Agricultura, **3**, 20, **2018**.
- AHMAD A., KHAN T.A., MUBEEN S., SHAHZADI I., AKRAM W., SAEED T., BASHIR Z., WANG R., ALAM M., AHMED S., HU D., LI G., WU T. Metabolic and Proteomic Perspectives of Augmentation of Nutritional Contents and Plant Defense in Vigna unguiculata. Biomolecules. 10 (2), 224, 2020.
- KHAN W.U., AHMAD S.R., YASIN N.A., ALI A., AHMAD A. Effect of *Pseudomonas fluorescens* RB4 and *Bacillus subtilis* 189 on the phytoremediation potential of *Catharanthus roseus* (L.) in Cu and Pb-contaminated soils, International Journal of Phytoremediation, **19** (6), 514, **2017**.
- TARIQ M., SHAH A.A., YASIN N.A., AHMAD A., RIZWAN M. Enhanced performance of *Bacillus megaterium* OSR-3 in combination with putrescine ameliorated hydrocarbon stress in *Nicotiana tabacum*, International Journal of Phytoremediation, 23 (2), 119, 2021.
- ISMAIL T., SALAMA M.A., EL-EBIARY M. Entomotoxic effects of synthesized aluminum oxide nanoparticles against Sitophilus oryzae and their toxicological effects on albino rats. Toxicol Ind Health, 37 (10), 594, 2021.
- ARONG G.A, USUA E.J. Comparative development of cowpea bruchid (*Callosobruchus maculates* F.) (Coleoptera: bruchidae) on pods and seeds of some cowpea cultivars (*Vigna unguiculata*). Global J Pure Appl Sci. 12, 35, 2006.
- AOAC. Official Methods of Analysis of the Association of Official Analytical Chemists: J AOAC Int 22th, AOAC, Washington DC, 2023.
- YEMM E.W., WILLIS A. The estimation of carbohydrates in plant extracts by anthrone. Biochem J. 57 (3), 508, 1954.

- THIEX N., MANSON H., ANDERSON S., PERSSON J.Å.J. Determination of crude protein in animal feed, forage, grain, and oilseeds by using block digestion with a copper catalyst and steam distillation into boric acid: collaborative study. J AOAC Int. 85 (2), 309, 2002.
- PIPER C.S. Soil and plant analysis. International Public Inc., New York, 1950.
- WOLF B. Improvements in the azomethine-H method for the determination of boron. Commun Soil Sci Plant Anal. 5, 39, 1974.
- CHAPMAN H.D., PRATT P.F. Method of Analysis for Soil Plant and Water. Uni. of California, Dep. Of Agric. Sci. U.S.A. 1, 1978.
- 23. MOHAMED A.A., MAZROU Y., EL-HENAWY A.S., AWAD N.M., ABDELAAL KH., HAFEZ Y., HANTASH R.M.A. Effect of sulfur applied and foliar spray of yeast on growth and yield of sugar beet under irrigation deprivation conditions, Fresenius Environmental Bulletin, **31** (4), 4199, **2022**.
- 24. SHAFEEK M.R., ALI, A.H., MAHMOUD A.R., HELMY Y.I., OMAR N.M. Effects of Foliar Application of Amino acid and bio fertilizer on growth and yield of onion plant under newly reclaimed land conditions. Middle-East J Appl Sci. 8, 1197, 2018.
- BABITA RANA V.S. Effect of Manual Fruit Thinning and CPPU on the Fruit Yield and Changes in Physico-chemical Composition at Harvest and after Storage of Allison Kiwifruit. Int. J. Bio-Resour. Stress Manag. 6, 781, 2015.
- 26. RANA V.S., SHARMA V., SHARMA S., RANA N., KUMAR V., SHARMA U., ALMUTAIRI K.F., AVILA-QUEZADA G.D., ABD_ALLAH E.F., GUDETA K. Seaweed Extract as a Biostimulant Agent to Enhance the Fruit Growth, Yield, and Quality of Kiwifruit. *Horticulturae.* 9 (4), 432, 2023.

- GOWTHAMI P., RAMA RAO G., RAO K.N., LAL M.A. Effect of foliar application of potassium, boron and zinc on quality and seed yield in soybean. Int J Chem Stud. 6, 142, 2018.
- 28. YUAN S., STUART A.M., LABORTE A.G., EDREIRA J.I.R., DOBERMANN A., KIEN L.V.N., THÚY L.T., PAOTHONG K., TRAESANG P., TINT K.M. Southeast Asia must narrow down the yield gap to continue to be a major rice bowl. Nat. Food, 3, 217, 2022.
- SONGSRIIN J, YAMUANGMORN S, LORDKAEW S, JUMRUS S, VEERADITTAKIT J, JAMJOD S, PROM-U-THAI C. Efficacy of Soil and Foliar Boron Fertilizer on Boron Uptake and Productivity in Rice. *Agronomy*. 13 (3),692, 2023.
- NAIK T.M., SRIHARI D., DORAJEERAO A.V.D., SASIKALA K., UMAKRISHNA K., SUNEETHA D.R.S., PRADEEPKUMAR C.M. Influence of plant growth regulators on growth and development of cluster bean varieties. Plant Archives 19, 2551, 2019.
- CRUZ L.P., DE SÁ L.F.R., SANTOS L.A., GRAVINA G.A., CARVALHO A.O., FERNANDES K.V.S., FILHO F.R.F., OLIVEIRA A.E.A. Evaluation of resistance in different cowpea cultivars to Callosobruchus maculatus infestation. J Pestic Sci. 89, 117, 2016.
- 32. SAAD A.S., TAYEB E.H.M., EL-SHAZLI M.M., BAHEEG S.A. Susceptibility of certain Egyptian and imported wheat cultivars to infestation by *Sitophilus oryzae* and *Rhyzopertha dominica*. Arch Phytopathol Plant Prot. 51 (1-2), 14, 2018.
- KRISHNA M.R., MEHERA B., KUMAR P. Effect of Potassium and Gibberellic Acid on the Growth and Yield of Cowpea (*Vigna unguiculata* L.). International Journal of Plant & Soil Science. 35(5), 31, 2023.