Effect of Organic Fertilization and Amino Acids on Growth, Chemical Composition and Capsaicin Content of Hot Pepper (Capsicum annum L var. Minimum) Plant

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Authors’ contributions

This work was carried out in collaboration among all authors. Author KAH designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors EAE and AAD managed the analyses of the study. Author AAD managed the literature searches. All authors read and approved the final manuscript.

ABSTRACT

This investigation was conducted in a private farm, Samannoud district, El-Gharbia Governorate, Egypt, during two successive seasons of 2017 and 2018 to study the effect of organic fertilizers and foliar spray of amino acid on vegetative growth, fruit yield, chemical composition and capsaicin content of hot pepper (Capsicum annum L var. Minimum) plant. The experiment included 13 treatments obtained from cattle manure at rates of 60 and 30 N-units per fed. combined with amino acid at 0, 100 and 200 ppm as well as chicken fertilizer at rates of 60 and 30 N-units per fed combined with amino acid at 0, 100 and 200 ppm, besides the treatment of NPK at recommended dose as a control. A complete randomized block design with three replicates was adopted. The obtained results showed that, the highest values from vegetative growth parameters, fruit yield, chemicals composition and capsaicin content were obtained from plants treated with cattle manure.
full dose + 200 ppm amino acids, followed by plants treated with chicken manure full dose + 200 ppm amino acids compared with all treatments except plant height, the highest value was obtained from plants treated with NPK (control) in the two seasons. Amino acid had a great effect when added with organic fertilization, whether cattle manure or chicken fertilizer at a concentration of 100 or 200 ppm. Based on the previous results, it could be recommend that spraying amino acids (glycine and glutamine) at the rate of 200 ppm along with organic fertilizers at the rate of 60 nitrogen units per feddan get the highest fruits yield, Vitamin C and capsaicin of hot pepper.

Keywords: Hot pepper plant; Capsicum annum L.; medicinal plants; organic manure; amino acids; antioxidants.

1. INTRODUCTION

Hot pepper (Capsicum annum L.) is an important vegetable crop and it is considered as one of the valuable medicinal plants in the pharmaceutical industry. It is consumed as spices or fresh. Hot pepper is being a good source of antioxidant compounds which are important for human health, such as ascorbic and capsaicinoids, which works as an antioxidant, helping to attack cancer cells in the body, expelling carcinogens outside the body and expelling toxins and vitamin A, which helps strengthen the immune system to attack infections, viruses and diseases that attack the body, as well as minerals as (potassium, phosphorus and calcium). Besides it is an excellent source of natural colors [1].

The use of organic fertilizers has become a mandatory requirement for organic regulations worldwide for crop organic production, it is not easy to provide enough nutrients for crops using only organic manure, they are low-priced alternatives that will provide nutrients to organic crops and cattle or chicken manure fertilizers are among the most important alternatives being studied. Organic fertilizers should be used to reduce the amount of toxic compounds (such as nitrates) produced by conventional fertilizers in producing crops consumed dry or fresh like hot pepper plant, hence, improving the quality of crops produced as well as human health [2].

Nowadays, amino acid glycine plays an important role in nutritional management of many plants, particularly horticultural crops. Glycine has a simple industrial synthesis process and therefore attracted interests to be in corporate with nutrient elements to produce chelates in order to facilitate nutrient uptake and translocation in plants [3].

Glutamine also is a vital amino acid in plant physiology with key roles in many metabolic processes, including nitrogen assimilation pathways and involved in many metabolic and biochemical reactions in plants [4,5].

[6] found that the application of organic fertilizers resulted in significant increases in total yield, total number of fruits per plot, fruit length and diameter of Capsicum annum. Also, [7] on green pepper (Capsicum annum L.) indicated that application of 10 t/ha of poultry manure gave the highest number of fruits, fruits weight, tallest plants and No. of branches/ plant of green pepper followed by 5 t/ha. [8] stated that the application of chicken dung showed the highest growth values, quality, yield performance and chlorophyll content of Capsicum annum L. [9] on Capsicum annum L. var. Kulai (Red Chilli Kulai) plants found that supplementing plants with chicken dung showed high rate in growth development, high yield and better quality of chili fruit.

Regarding the advantageous effects of foliar spraying of amino acids, many investigators have revealed the valuable effects of amino acids in improving growth and yield for many crops. In this respect, [10] obtained significant increases in plant height, No. of branches/ plant, shoot dry matter, fruit length, fruit diameter, fruit dry matter and ascorbic acid content of hot peppers when plants supplied with amino acid at 0.45 g and 0.27 g/plant. Omer et al., (2013) reported that foliar spray with amino acids improved the growth and chemical composition of chamomile plant. [11] recorded significant increases in plant height, No. of branches and dry matter of shoots of pepper plants treated with amino acids at 800 mg/l, compared with control. [12] on sweet pepper, found that shoot fresh and dry weights increased with application of amino acid at 3- 6 g/l, compared with control.

Therefore, there is a need to generate information and evaluate the effect of organic fertilizers from cattle and chicken manure sources and bio-stimulation effects of glycine and
glutamine and compared with chemical fertilizer (N, P and K) on hot pepper (Capsicum annum L var. Minimum) plant and the possibility of using organic fertilizers with amino acids as an alternative to chemical fertilizers to produce a high quality yield of hot pepper.

2. MATERIALS AND METHODS

This study was performed at a private farm, Samannoud district, El-Gharbieh Governorate, Egypt during two successive seasons of 2017 and 2018, to study the effect of organic fertilization and amino acids on hot pepper (Capsicum annum L var. Minimum) plant. Seeds (self-pollinated) were obtained from Research Department, Horticulture Research Institute. In the two seasons, the seeds were sown in the nursery in the first week of January and transplanted to experimental soil in the first week of March, in rows 30 cm apart with intra-row spacing of 60 cm with plot area of 3 x 3.5 m². The physical and chemical properties of the studied soil (clay loam) were done at Water and Environment Research Institute laboratories (ARC) according to the methods of [13] are recorded in Table1. Soil and water analysis.

The organic manure sources were cattle manure (obtained from a nearby smallholder farmer), chicken manure (obtained from a nearby broiler house), both cattle and chicken manures were applied before composting them and evenly broadcasting and then thoroughly incorporated it into the experimental plots using a hand hoe to an approximate depth of 10 cm. The cattle and chicken manures were applied 21 days before transplanting, during the soil preparation to allow sufficient time for reaction with the soil. The quantity of applied manures was estimated as nitrogen units, two rates were prepared for each manure 60 and 30 units of nitrogen as full and half doses, respectively.

Two concentrations of glycine and glutamine amino acids (100 and 200 ppm) were applied as foliar sprays, three times on the plants, during growth period. The first spray was done four weeks after transplanting, and the remaining sprays were done at three-week intervals.

The chemical fertilizers applied (as control) were added as follows, N- fertilizer (ammonium sulfate ,20.5 % N) at 60 kg N/fed., P-fertilizer (calcium super phosphate ,15.5 % P₂O₅) at 30 kg P₂O₅/fed and K-fertilizer (potassium sulfate) at 48 kg K₂O / fed. The doses of both N and K were applied in two equal split doses, after month from transplanting and the remaining portion was applied four weeks after the first dose, while calcium superphosphate fertilizer was added during of soil preparation.

Thirteen treatments were suggested according to a combination of the used materials to study the effect of organic fertilization and amino acids on hot pepper (Capsicum annum L var. Minimum) plant as follows:

1. NPK at recommended dose as a control.
2. Cattle manure at full dose
3. Cattle manure full dose+ amino acids at 100 ppm
4. Cattle manure full dose+ amino acids at 200 ppm
5. Cattle manure half dose
6. Cattle manure half dose+ amino acids at 100 ppm
7. Cattle manure half dose+ amino acids at 200 ppm
8. Chicken manure full dose
9. Chicken manure full dose+ amino acids at 100 ppm
10. Chicken manure full dose+ amino acids at 200 ppm
11. Chicken manure half dose
12. Chicken manure half dose+ amino acids at 100 ppm
13. Chicken manure half dose+ amino acids at 200 ppm

2.1 Statistical Analysis

The collected data were statistically analyzed using computer package program of MSTAT-C. Data were analyzed using an analysis of variance (ANOVA) for a randomized complete block design with three replicates. Mean separation was done using least significant difference (LSD) at 5% level of significance as described by [14].

2.2 Plant Sampling and Measurements

A random sample of five plants was taken from each experimental treatment at the final harvesting stage and the data were recorded on vegetative growth parameters (plant height, number of branches, fresh and dry weights g/plant) and fruit yield during harvest period (number of fruits/plant, weights of fresh and dry fruits yield g/ plant and weight of dry fruits yield ton /fed) during the two seasons.
Table 1. Some physical and chemical properties of experimental soil in the two seasons

| Season | Clay % | Silt % | Sand % | Organic Matter % | pH | Available nutrients (ppm) |
|--------|--------|--------|--------|------------------|----|--------------------------|
|        |        |        |        |                  |    | N  | P  | K  | Zn | Fe | Mn |
| 2017   | 46.5   | 29.2   | 24.3   | 1.26             | 7.50| 2.12 | 28.10 | 391 | 1.35 | 8.01 | 12.2 |
| 2018   | 46.7   | 29.1   | 24.2   | 1.77             | 7.40| 2.81 | 30.15 | 389 | 1.28 | 8.11 | 12.3 |

Table 2. Some physical and chemical properties of different organic manure used in the two seasons

| Season | pH  | EC dS/m | C:N ratio | Macro-elements (%) | Micro-elements (ppm) |
|--------|-----|---------|-----------|--------------------|----------------------|
|        |     |         |           | N     | P     | K     | Fe  | Mn  | Zn  | Cu  |
|        |     |         |           | Cattle manure     |Chicken manure        |
| 2017   | 7.1 | 4.2     | 20.1      | 1.16  | 0.54  | 0.86  | 556 | 116 | 159 | 36  |
| 2018   | 7.1 | 4.2     | 20.2      | 1.15  | 0.57  | 0.88  | 552 | 117 | 162 | 34  |
| 2017   | 8.1 | 6.2     | 14.60     | 0.22  | 0.72  | 1.14  | 418 | 184 | 225 | 43  |
| 2018   | 8.1 | 6.2     | 15.30     | 0.29  | 0.71  | 1.16  | 416 | 186 | 231 | 41  |

Fig. 1. HPLC chromatograms of capsaicin standard

Fig. 2. Calibration curve of peak area versus concentration for capsaicin

However, vitamin C and carotenoids contents in the pepper fruit (as mg /100 g fresh weight) was estimated according to [15] and dried fruits samples was used to determine the concentration of carbohydrates (%DW) according to [16]. The fruit mineral concentration of N was estimated by the micro-Kjeldahl according to [17] and P, K were determined according to [18].

2.3 Capsaicin Determination in Fruits

2.3.1 The content of capsaicin in fruits (µg/g) was estimated by high-performance liquid chromatography (HPLC) as follows:

2.3.1.1 Capsaicin extraction

Each powder sample for all treatments was put in a Petri dish and kept in the oven at 60°C for 1 hour to dry. One gram of each powder sample was transferred into the volumetric flask (25 ml) filled with methanol and kept in the water bath at 50°C for 4 hour, then it was left overnight at room temperature. The final volume was adjusted to 25 ml by methanol. The samples were centrifuged and passed through a Millipore (Billerica, MA) nylon filter (pore size, 0.45 mm). The samples were packed in the vials and kept in the refrigerator until examination. The samples and standard capsaicin solution (50 µL) were directly injected into the HPLC apparatus [19].

2.3.1.2 HPLC chromatographic analysis

The conditions used: Hewlett Packard apparatus HPLC system (Agilent, USA) constructed of a quaternary HP 1260 pump and HP 1260 multiple wave length detector, analytical HPLC column C18 (5 µm, 4.6x250 mm), Germany. The mobile
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phase consisted of 0.1% phosphoric acid = acetonitrile (3:2V=V). The temperature of column was maintained at 30°C during the separation, the flow rate was 1 ml/minute and the chromatography run was 15 minutes. The eluting compounds were mentioned at wavelength of 281 nm.

**Standard curve of capsaicin:** A stock solution of capsaicin (8-methyl-N-vanillyl-6-nonanamide) was made by dissolving standard capsaicin in methanol at the following concentrations (50, 100, 300, 550, 800, 1100 and 1500 µg/mL).

### 2.3.1.3 Capsaicin identification

Capsaicin peak identification in the extract was done by matching the retention time with that of standard capsaicin, which was further confirmed by co-injecting standard capsaicin together with the extract for quantitative analysis of capsaicin in chosen samples of *Capsicum* fruits in this study.

### 3. RESULTS AND DISCUSSION

#### 3.1 Effect on Vegetative Growth

Data on the comparative effect of inorganic (NPK at recommended dose as a control) and organic fertilizers with or without amino acids treatments (glycine and glutamine at 100 or 200 ppm) on growth parameters of hot pepper (*Capsicum annuum* L var. Minimum) plants are given in Table 3.

Concerning the effect of organic manures, the data obtained during 2017 and 2018, seasons, revealed that all growth parameters of hot pepper (*Capsicum annuum* L var. Minimum) plant treated with the high dose of either cattle manure or chicken manure alone were higher than that of the half dose. In both seasons, the application of NPK fertilizer gave the tallest plants (44.66 and 45.88 cm, respectively), followed by cattle manure at full dose plus foliar spray treatment of amino acids at 200 ppm which increased plant height to 44.30 cm in the first season and 45.75 cm in the second one. On the other hand, the application cattle manure at half dose alone gave the shortest in the first season (37.77 cm) and in the second one (39.42 cm).

In the first season, the application of cattle manure at full dose combined with amino acid treatment at 200 ppm, followed by chicken manure at full dose combined with amino acid treatment at 200 ppm increased the formation of branches/plant, than the control and other treatments. In the second season, treating hot pepper plants with cattle manure at full dose combined with amino acid treatments at 100 and 200 ppm, followed by chicken manure at full dose plus amino acid treatment at 200 ppm, were the most effective treatments in this respect.

Concerning the response of fresh and dry weights of herb to the application of organic manures (cattle or chicken) and amino acids treatments, the obtained data showed that, in both seasons, the application of organic manures (cattle or chicken) at full dose combined with amino acid treatment at 200 ppm produced the heaviest fresh as well as dry weight of herbs. Generally, it can be mentioned that the best results obtained for the growth parameters came from cattle manure treatment combined with amino acids at 200 ppm, and this promoting effect on plant growth could be due to increase in microbial biomass in soils after receiving cattle manure or may be due to production of plant growth regulators by microorganisms, which is in line with the work of [20]. In this regard, [21] stated that the high rates of manure increased soil contents of K, Mg, and P and increased the cation exchange capacity of soils and these factors contribute toward higher crop yields on manure treated plots. Also, [22] stated that cattle manure amendments could increase the pH of soils and this effect on soil pH was immediate and persisted during an 8wk incubation and it was attributed to buffering from bicarbonates and organic acids in cattle manure and the mineral N as well as available P, K, Ca and Mg increased immediately after manure application and there was no change in the cation exchange capacity (CEC) of manure amended soils. The effect of animal manure on soil pH may persist over several years. While, [23] found that cattle manure application promoted the growth of bacteria, but not fungi, when compared with the control soil and enriched K, which were positively correlated with soil productivity. Furthermore, the foliar application of amino acids (glycine plus glutamine) in plants treated with cattle manure or chicken manure showed a marked stimulating effect on hot pepper growth. The foliar spray of amino acids at 200 ppm in plants treated with cattle manure at full dose significantly increased vegetative growth parameters (No. of branches, fresh and dry weights g/plant) compared to the NPK fertilizer or organic manure without amino acids at 200 ppm, followed by chicken manure at full dose combined with amino acid treatment at 200 ppm.
acid. The use of amino acids to improve plant growth and productivity was studied on several horticultural crops, because of their role in increasing protein content, chlorophyll and enzymes, which in turn increase the performance of plants and raise the efficiency of photosynthesis, giving the best growth of plants. In this regard, [24] found that the use of amino acids led to a pronounced increase in pepper plant height, number of branches and dry weight of shoots compared to non-treated plants. Amino acids act as prerequisite matter and activator of hormones and effective causes of growth, and they also increased the amount of chlorophyll which increases the rate of photosynthesis of sweet pepper plants [25,26] sprayed plants of bell pepper (Capsicum annum L.) cultivars “Sven Rz F-1” and “Red Knight.” with different concentrations of amino acids and obtained significant increases in the number of leaves, leaf area along with better physicochemical quality. [27] sprayed Romain lettuce (Lactuca sativa subvar Sahara) with glycine or glutamine at 250-1000 ppm, and found significant increase in shoot fresh and dry weights with 500 ppm and the different amino acid treatments had no significant effect on plant height.

### 3.2 Effect on Fruit Yield and its Components

It is clear from Table 4. that the yield of hot pepper plant and its components (number fruit, fresh and dry weights of fruits per plant and fruits yield / feddan) were affected by all treatments of organic fertilizers either with or without amino acids application when compared to NPK (control). The fruit number per plant did not show any significant differences in respect to fertilizer treatments applied. It was found that, cattle manure followed by chicken manure gave the best mean values of fruit number per plant when compared to NPK fertilizer treatment. Treating plants with organic manures had no significant effect on increasing the number of fruits/plant, fruit fresh weight/plant and fruit dry weight/plant as compared with NPK fertilization (except the fruits yield / feddan, in both seasons).

The data indicated that, the application of cattle manure plus amino acids followed by chicken manure plus amino acids resulted in a significant increase in fresh and dry weights of fruits per plant and fruits yield / feddan when compared to NPK fertilizer. Furthermore, an increase in the fruit yield was obtained in plants fertilized with organic manure with the application of amino acid at 200 ppm, follow by the treatment of organic manure plus amino acids at 100 ppm, with no significant difference between them.

It is clear that organic fertilizers have increased soil porosity and supported root growth and increased soil organic matter thus, increased efficiency in hot pepper production. Whereas, amino acids i.e., glycine plus glutamine amino acids have several important roles in plant metabolism, and have stimulation effects on plant growth and quality. The use of amino acids could improve plant productivity in several horticultural crops, as it increases photosynthesis, the contents of protein, chlorophyll and enzymes, which in turn increase plant growth, resulting in best the fruit yield. In this regard, [26] on bell pepper (Capsicum annum L.) cultivars “Sven Rz F-1” and “Red Knight.” sprayed plants with different concentrations of amino acids and obtained significant increases in the fruit yield per plant, number of marketable fruits per plant, average fruit diameter, fruit wall thickness, and average single fruit weight and storage life along with better physicochemical quality. Our results are also in line with those of [25] who pointed out that the use of organic manure plus amino acids increased the growth and yield of sweet pepper significantly compared to the use of mineral fertilization at the recommended dose.

### 3.3 Effect on Chemical Composition of Fruits

Data presented in Table 5 indicated that the fruit content of total carbohydrates, carotenoids and vitamin C of hot pepper plants were responded positively to the applications of organic manure with or without amino acids treatments comparing with the NPK at recommended dose without significant differences. While, the results showed that there was a significant difference in the content of total carbohydrates, carotenoids and vitamin C of hot pepper with the application of different organic manure plus amino acids at rate of 100 or 200 ppm treatments when compared to NPK or half dose of organic manure with or without amino acids. The results of this study are in agreement with those obtained by [28] on sweet pepper and [29] on hot pepper, who reported that the foliar application of amino acids gave higher content of vitamin C and β-carotene contents than the control [27] sprayed Romain lettuce (Lactuca sativa subvar Sahara) with glycine or glutamine at 250 ppm, and found significant in leaf content of carotenoids compared with the control plants.
Table 3. Effect of organic fertilization and amino acids on plant height (cm), No. of branches / plant, fresh and dry weights/plant (g) of hot pepper (*Capsicum annum* L var. Minimum) plant during 2017 and 2018, seasons

| Treatments                                           | First season 2017 |                                                      | Second season 2018 |                                                      |
|------------------------------------------------------|-------------------|------------------------------------------------------|-------------------|------------------------------------------------------|
|                                                      | Plant height      | No. of branches / plant                             | Fresh weight / plant | Dry weight / plant                                   | Plant height | No. of branches / plant | Fresh weight / plant | Dry weight / plant |
| NPK at recommended dose as a control                | 44.66             | 13.00                                                | 358.48             | 68.23                                                | 45.88        | 13.53                                                | 368.37             | 71.23                                                |
| Cattle manure at full dose                           | 41.62             | 13.15                                                | 358.90             | 68.89                                                | 42.52        | 13.92                                                | 368.86             | 72.81                                                |
| Cattle manure full dose + amino acids at 100 ppm     | 42.97             | 13.45                                                | 359.46             | 70.23                                                | 44.53        | 14.12                                                | 370.89             | 73.02                                                |
| Cattle manure full dose + amino acids at 200 ppm     | 44.30             | 14.48                                                | 379.87             | 74.03                                                | 45.75        | 14.93                                                | 390.57             | 77.85                                                |
| Cattle manure half dose                              | 37.77             | 12.00                                                | 328.41             | 61.08                                                | 39.42        | 12.34                                                | 349.74             | 63.60                                                |
| Cattle manure half dose + amino acids at 100 ppm.    | 39.82             | 12.72                                                | 333.08             | 63.13                                                | 41.45        | 12.62                                                | 350.42             | 65.00                                                |
| Cattle manure half dose + amino acids at 200 ppm     | 41.85             | 13.00                                                | 335.45             | 64.31                                                | 43.82        | 13.10                                                | 354.45             | 67.67                                                |
| Chicken manure full dose                             | 41.94             | 12.81                                                | 357.08             | 67.62                                                | 43.17        | 13.42                                                | 376.74             | 70.94                                                |
| Chicken manure full dose + amino acids at 100 ppm    | 42.30             | 13.54                                                | 357.44             | 68.05                                                | 43.50        | 13.87                                                | 377.77             | 71.15                                                |
| Chicken manure full dose + amino acids at 200 ppm    | 43.86             | 14.00                                                | 368.82             | 72.50                                                | 45.18        | 14.08                                                | 379.82             | 73.18                                                |
| Chicken manure half dose                             | 39.07             | 11.84                                                | 320.03             | 59.58                                                | 41.38        | 11.86                                                | 339.03             | 59.23                                                |
| Chicken manure half dose + amino acids at 100 ppm    | 41.09             | 12.41                                                | 330.38             | 61.59                                                | 43.41        | 12.09                                                | 346.05             | 63.78                                                |
| Chicken manure half dose + amino acids at 200 ppm    | 43.10             | 12.76                                                | 334.73             | 62.94                                                | 44.12        | 12.53                                                | 350.06             | 65.79                                                |
| LSD at 5 %                                           | 1.07              | 0.99                                                 | 20.38              | 3.89                                                 | 0.98         | 0.81                                                 | 19.77              | 3.56                                                 |
Table 4. Effect of organic fertilization and amino acids on number of fruits/plant, fruit fresh weight/plant (g), fruit dry weight/plant (g) and fruits yield/fed (Kg) of hot pepper (*Capsicum annuum* L var. Minimum) plant during 2017 and 2018, seasons

| Treatments                                                | First season, 2017 | Second season, 2018 |
|-----------------------------------------------------------|--------------------|---------------------|
|                                                           | No. of fruits/plant | Fruit fresh weight/plant | Fruit dry weight/plant | Fruits yield/fed | No. of fruits/plant | Fruit fresh weight/plant | Fruit dry weight/plant | Fruits yield/fed |
| NPK at recommended dose as a control                     | 207.67             | 178.18              | 36.55                 | 812.21           | 210.67             | 180.22              | 37.15                 | 825.54           |
| Cattle manure at full dose                                | 210.67             | 182.33              | 38.83                 | 862.88           | 212.33             | 187.17              | 39.52                 | 878.21           |
| Cattle manure full dose + amino acids at 100 ppm          | 212.00             | 186.63              | 39.63                 | 880.66           | 214.67             | 188.47              | 40.15                 | 888.21           |
| Cattle manure full dose + amino acids at 200 ppm          | 214.33             | 188.71              | 41.88                 | 930.66           | 217.67             | 192.54              | 42.38                 | 941.77           |
| Cattle manure half dose                                   | 206.33             | 168.37              | 32.87                 | 797.10           | 208.67             | 171.54              | 33.71                 | 749.10           |
| Cattle manure half dose + amino acids at 100 ppm          | 206.67             | 168.72              | 33.22                 | 738.21           | 209.00             | 171.88              | 34.88                 | 775.10           |
| Cattle manure half dose + amino acids at 200 ppm          | 207.00             | 169.38              | 34.38                 | 763.99           | 209.67             | 172.21              | 35.05                 | 778.88           |
| Chicken manure full dose                                  | 210.33             | 181.73              | 38.57                 | 857.10           | 212.33             | 185.90              | 38.87                 | 863.77           |
| Chicken manure full dose + amino acids at 100 ppm         | 211.00             | 182.77              | 38.93                 | 865.10           | 212.67             | 187.93              | 39.43                 | 876.21           |
| Chicken manure full dose + amino acids at 200 ppm         | 212.00             | 184.13              | 40.13                 | 891.77           | 216.33             | 190.97              | 39.80                 | 884.44           |
| Chicken manure half dose                                  | 205.33             | 162.02              | 32.02                 | 711.55           | 207.33             | 168.86              | 32.36                 | 719.10           |
| Chicken manure half dose + amino acids at 100 ppm         | 205.67             | 164.87              | 32.21                 | 715.77           | 207.67             | 170.71              | 33.87                 | 752.66           |
| Chicken manure half dose + amino acids at 200 ppm         | 206.33             | 165.38              | 33.54                 | 745.33           | 209.00             | 171.21              | 34.04                 | 756.44           |
| LSD at 5 %                                                | 7.40               | 7.03                | 2.61                  | 40.05            | 7.40               | 7.24                | 2.66                  | 41.04            |
Table 5. Effect of organic fertilization and amino acids on carbohydrates, carotenoids, vitamin C, nitrogen, phosphorus and potassium of hot pepper (*Capsicum annuum* L var. Minimum) plant during 2018, season

| Treatments                                           | Second season 2018 |
|------------------------------------------------------|--------------------|
|                                                      | Total carbohydrates Content | Carotenoids mg/100g FW fruit | Vitamin C mg/100g FW fruit | N % | P % | K % |
| NPK at recommended dose as a control                | 20.83              | 1.54                         | 126.3                          | 2.30 | 0.267 | 0.324 |
| Cattle manure at full dose.                          | 21.06              | 1.59                         | 126.6                          | 2.29 | 0.266 | 0.323 |
| Cattle manure full dose + amino acids at 100 ppm     | 21.49              | 1.67                         | 130.5                          | 2.39 | 0.269 | 0.325 |
| Cattle manure full dose + amino acids at 200 ppm     | 22.41              | 1.72                         | 134.9                          | 2.40 | 0.272 | 0.330 |
| Cattle manure half dose.                             | 20.66              | 1.40                         | 124.1                          | 2.07 | 0.254 | 0.309 |
| Cattle manure half dose+ amino acids at 100 ppm      | 20.78              | 1.44                         | 125.6                          | 2.13 | 0.256 | 0.316 |
| Cattle manure half dose+ amino acids at 200 ppm      | 20.81              | 1.50                         | 126.0                          | 2.20 | 0.259 | 0.320 |
| Chicken manure full dose.                            | 21.00              | 1.56                         | 126.4                          | 2.31 | 0.266 | 0.324 |
| Chicken manure full dose + amino acids at 100 ppm    | 21.45              | 1.65                         | 129.7                          | 2.38 | 0.267 | 0.327 |
| Chicken manure full dose + amino acids at 200 ppm    | 22.04              | 1.69                         | 133.1                          | 2.39 | 0.270 | 0.333 |
| Chicken manure half dose.                            | 20.63              | 1.39                         | 123.2                          | 2.08 | 0.251 | 0.314 |
| Chicken manure half dose + amino acids at 100 ppm    | 20.75              | 1.42                         | 125.1                          | 2.10 | 0.252 | 0.318 |
| Chicken manure half dose + amino acids at 200 ppm    | 20.79              | 1.44                         | 125.7                          | 2.19 | 0.257 | 0.321 |
| LSD at 5 %                                           | 0.37               | 0.12                        | 1.6                            | 0.24 | 0.014 | 0.012 |
On the other hand, data on the contents of N, P and K % of fruits of hot pepper plants in response to the applied organic manures with or without amino acids (Table 5), revealed that application of organic manures with or without amino acids, generally, showed no significant differences in the contents of N, P and K %, as compared with the with NPK application (control) in the second season of study. Meanwhile, the highest values of N and P (% D.W) of fruits of hot pepper were recorded with the treatment of cattle manure combined with the foliar application of amino acids at rates of 200 ppm. While, the highest value of K % of fruits was recorded from treating plants with cattle manure at full dose +200 ppm amino acids and chicken manure at full dose +200 ppm amino acids, respectively. On the other side, the lowest value of capsaicin content (594.53 µg/g) was resulted from the control plants, whereas the highest one resulted from treating plants with cattle manure at full dose combined with the foliar application of 200 ppm amino acids giving capsaicin content of 1046.8 µg/g. This means, that cattle manure fertilizer and amino acids has a major role in increasing the content of capsaicin in hot pepper fruits [31] showed that there was a significant effect of poultry manure and amino acid by giving the highest value of capsaicin compared with the treatment of mineral fertilization of Capsicum annuum Plants.

3.4 Capsaicin Content in Fruits (µg/g)

Data in Table 6. and Figs. 3,4,5 and 6 showed the analysis of the capsaicin content in the chosen samples of capsicum fruits in this study. The results showed that, the highest capsaicin contents of 834.4, 1046.8 and 1007, 73 µg/g were obtained from fruits of plants treated with cattle manure at full dose, cattle manure at full dose +200 ppm amino acids and chicken manure at full dose +200 ppm amino acids, respectively.

Table 6. The capsaicin content in the chosen samples of hot pepper (Capsicum annuum L.) fruits (µg/g), in the second season

| Treatments                  | NPK (Control) | Cattle manure (C M) full dose | C. M. full dose + 200 ppm A. A. | Ch. M. (full dose) +200 ppm A.A. | mean     |
|-----------------------------|--------------|-------------------------------|-------------------------------|--------------------------------|----------|
| Capsaicin content           | 594.53       | 834.4                         | 1046.8                        | 1007, 73                       | 825.24   |

Fig. 3. HPLC chromatograms
NPK (Control)

Fig. 4. HPLC chromatograms
cattle manure (C M) full dose
Authors have declared that no competing interests exist.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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4. CONCLUSION

From the overall results, the highest values from growth parameters, fruit yield, chemicals composition and capsaicin content were obtained from plants treated with organic manure whether cattle manure full dose or chicken manure full dose + 200 ppm amino acids. Application of organic fertilizers helps to supply nutrient as required for growth and yield of hot pepper (Capsicum annum L var. Minimum) plant. Thus, it could be recommend that spraying amino acids (glycine and glutamine) at the rate of 200 ppm along with organic fertilizers at the rate of 60 nitrogen units per feddan get the highest fruits yield, Vitamin C and capsaicin content.

Fig. 5. HPLC chromatograms
cattle manure full dose + 200 ppm A. A.

Fig. 6. HPLC chromatograms
chicken manure (full dose) +200 ppm A.A.
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