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RESPONSE OF TOCOPHEROL AND ZINC ON CHEMICAL-BIOCHEMICAL PARAMETERS AND YIELD OF GREEN GRAM (*Vignaradiat* L.)

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ABSTRACT

The present study was conducted at farm of Agril. Botany, College of Agriculture, Nagpur, during *kharif* 2019-2020 to study the probable response of tocopherol and zinc on chemical, biochemical and yield parameters in green gram. The experiment was laid out in Randomized Block Design (RBD) with three replications, consisting of twelve treatments of tocopherol (100 ppm, 200 ppm, 300 ppm, 400 ppm and 500 ppm) and 0.5 % zinc sulphate spray individually and in their combinations. Spraying was done at 25 and 35 DAS. Application of tocopherol and zinc enhanced chemical, biochemical parameters viz., chlorophyll content, NPK content in leaves and protein content in grains, yield parameter like seed yield plant^{-1}. Data revealed that treatment 100 ppm tocopherol and 0.5 % zinc sulphate was considered as a most effective concentration in enhancing all chemical and biochemical parameters and yield of green gram.

(Key words: Green gram, tocopherol, zinc sulphate, foliar feeding, chemical parameters, biochemical parameters and yield)

INTRODUCTION

Green gram is an important pulse crop ranked as the second most drought resistant crop after soybean. It has more protein content and better digestibility than any other pulse crop (Tabasum *et al*., 2010). Green gram grains are having 51% carbohydrates, 26% protein, 10% moisture and 3% vitamin. The residues of green gram are also used as feed for animals and enhance the soil fertility as well (Asaduzzaman, 2008).Green gram is rich source of nutrients including manganese, potassium, copper, zinc and vitamin B complex.It plays an important role not only in human diet, but also in improving the soil fertility by fixing atmospheric nitrogen into available form with the help of *Rhizobia* species present in the nodule of its roots (Ashraf *et al*., 2003).

Tocopherols are lipophilic antioxidants and together with triacontanols belong to vitamin "E" family. á-tocopherol is most biologically active form of vitamin-E. Vitamin-E exists in eight different forms, four tocopherols and four tocotrienols. The four forms of tocopherols consist of a polar chromanol ring and lipophilic prenyl chain with differences in position and number of methyl groups (Lushchack and Semchuk, 2012). There is a hydroxyl group that can donate hydrogen atom to reduce free radicals and a hydrophobic chain which allows for penetration into biological membrane. Tocopherols occur in á (alpha), â (beta), ã (gamma) and ä (delta) forms. á -Tocopherol as a fat-soluble antioxidant interrupts the propagation of reactive oxygen species.

Zinc is one of the micronutrients, plants need to grow efficiently. It is an essential component of enzymes involved in metabolic reaction. Zinc plays a special role in synthesizing proteins, RNA and DNA (Kobrace *et al.,* 2011).Its deficiency symptoms vary among different plant species. Zinc sulphate is most commonly used source of zinc in granular fertilizer due to its high solubility in water and it's relatively low cost of production. Zinc sulphate is an inorganic compound with molecular formula $ZnSO_4$. It is colourless and available in liquid form. Zinc affects several biochemical processes in the plant, such as cytochrome and nucleotide synthesis, auxin metabolism, chlorophyll production, enzyme activation and membrane integrity. Yashona *et al.* (2018) stated that zinc servers as an essential component of enzymes and acts as a functional, structural and /or regulatory cofactor of a large number of enzymes. With the above back ground, it was thought worthwhile to assess the effect of tocopherol and zinc on green gram.

The present research work has been designed to study the effect of tocopherol and zinc sulphate on chemical, biochemical parameters and yield of green gram.

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MATERIALS AND METHODS

A field experiment was conducted at Botany farm, College of Agriculture, Nagpur to known the response of tocopherol and zinc on its chemical, biochemical parameters and yield of green gram. The experiment was carried out in Randomized Block Design with three replications. The experiment consisted of twelve treatments viz., T_1 (Control), T_2 (100 ppm tocopherol), T_3 (200 ppm tocopherol), T_4 (300 ppm tocopherol), T_5 (400 ppm tocopherol), T_6 (500 ppm tocopherol), T_7 (0.5% $ZnSO_4$), T_8 (100 ppm tocopherol +0.5% $ZnSO_4$), T_9 (200 ppm tocopherol +0.5% $ZnSO_4$), T_{10} (300 ppm tocopherol +0.5% $ZnSO_4$), T_{11} (400 ppm tocopherol +0.5% $ZnSO_4$) and T_{12} (500 ppm tocopherol +0.5% $ZnSO_4$). The seeds were sown with a spacing of 30 cm × 10 cm. Two foliar sprays were given at 25 and 35 DAS. Observations were taken on chemical, biochemical parameters such as leaf chlorophyll content, N, P, K content in leaves and protein content in seed and yield parameter like seed yield plant^{-1}. Leaf chlorophyll content was determined by acetone method as given by Bruinsma (1982), Determination of nitrogen and protein was carried out by micro-kjeldhal method as given by Somichi *et al*. (1972). Phosphorus and potassium estimated by vanadomolybdate yellow colour method, (using colorimeter and flame photometer respectively) given by Jackson (1967). The data were analyzed statistically using analysis of variance at 5 % level of significance (Panse and Sukhamate, 1954).

RESULTS AND DISCUSSION

Chlorophyll content

Chlorophyll is a pigment directly associated with the photosynthetic activity, which is responsible for the conversion of sunlight into chemical energy in the form of ATP and NADPH (Taiz and Zeiger, 2009), favoring the fixation of atmospheric carbon dioxide and providing thereby the production of plant biomass during the process of Malvin Calvin cycle (Salisbury and Ross, 2012).

At 40 DAS significantly highest chlorophyll content in leaves was observed in treatment T_8 (100 ppm tocopherol+0.5% $ZnSO_4$) followed by treatmentsT_9 (200 ppm tocopherol+0.5% $ZnSO_4$), T_{10} (300 ppm tocopherol+0.5% $ZnSO_4$), T_{11} (400 ppm tocopherol+0.5% $ZnSO_4$), T_{12} (500 ppm tocopherol+0.5% $ZnSO_4$), T_2 (100 ppm tocopherol), and T_3 (200 ppm tocopherol) when compared with treatment T_1 (control) and other treatments. At 55 DAS significantly maximum chlorophyll content was recorded in treatment T_8 (100 ppm tocopherol+0.5% $ZnSO_4$) followed by treatments T_9 (200 ppm tocopherol+0.5% $ZnSO_4$), T_{10} (300 ppm tocopherol+0.5% $ZnSO_4$), T_{11} (400 ppm tocopherol+0.5% $ZnSO_4$), T_{12} (500 ppm tocopherol+0.5% $ZnSO_4$), T_2 (100 ppm tocopherol),T_3 (200 ppm tocopherol) and T_4 (300 ppm tocopherol) when compared with treatment T_1 (control) and other treatments.

Chlorophyll is a photosynthetic pigment. The main function of anti-oxidants such as vitamin E was protective of cell membranes and their binding transporter proteins, maintained their structure and function against the toxic and destructive effects reactive oxygen species during stress, in turn, more absorption and translocation of minerals (Dicknson *et al*., 1991).The effects of zinc application on chlorophyll and leaf area mainly attributed to more availability of zinc both during seedling and subsequent stages of plant growth which has been known to increase photosynthates and 'N' fixation (Hegazy *et al*., 1990). The zinc have important role in chlorophyll formation which enhanced chlorophyll content in leaf of the plants (Sharma *et al.,* 2010).

Purushottam *et al.* (2018) observed 3-8 % increment in leaf chlorophyll of pigeonpea with foliar application of 0.5% zinc sulphate. Foliar spray of 200 ppm ascorbic acid and $ZnSO_4$. @ 0.5 % significantly increased leaf chlorophyll content of chickpea (Raut *et al*., 2020). Blesseena *et al.* (2020) tested α- tocopherol and zinc as a foliar application on chickpea and recorded significant increase in chlorophyll content of leaf by the application of 100 ppm tocopherol + 0.5 % $ZnSO_4$.

Leaf nitrogen content

Nitrogen is most imperative element for proper growth and development of plants which significantly increases and enhances the yield and its quality by playing a vital role in biochemical and physiological functions of plant. Nitrogen (N) is responsible for the development of leaf area. Plant cells require adequate supply of N for normal cell division and growth of the plant. Tender shoots, tips of shoots, buds, leaves contains higher nitrogen (Jain, 2010).

At 40 DAS significantly maximum leaf nitrogen content was obtained with treatment T_8 (100 ppm tocopherol +0.5% $ZnSO_4$) followed by treatments T_9 (200 ppm tocopherol+0.5% $ZnSO_4$), T_{10} (300 ppm tocopherol+0.5% $ZnSO_4$), T_{11} (400 ppm tocopherol+0.5% $ZnSO_4$), T_{12} (500 ppm tocopherol+0.5% $ZnSO_4$), and T_2 (100 ppm tocopherol) over control and rest of the treatments under study. Treatments T_3 (200 ppm tocopherol), T_4 (300 ppm tocopherol), T_5 (400 ppm tocopherol), and T_6 (500 ppm tocopherol) also observed significantly higher nitrogen content when interrelated with treatment T_1 (control) and other treatments. At 55 DAS nitrogen was maximum and significantly more in treatment T_8 (100 ppm tocopherol+0.5% $ZnSO_4$) followed by treatments T_9 (200 ppm tocopherol+0.5% $ZnSO_4$), and T_{10} (300 ppm tocopherol+0.5% $ZnSO_4$) when compared with treatment T_1 (control) and remaining treatments under study. Treatments T_{11} (400 ppm tocopherol +0.5% $ZnSO_4$), T_{12} (500 ppm tocopherol+0.5% $ZnSO_4$), T_2 (100 ppm tocopherol) and T_3 (200 ppm tocopherol) also increased leaf nitrogen over control and rest of the treatments under observation. However, treatments T_4 (300 ppm tocopherol), T_5 (400 ppm tocopherol), T_6 (500 ppm tocopherol), and T_7 (0.5% $ZnSO_4$) were found also significantly superior in a descending manner when compared with treatment T_1 (control).

At the vegetative period, physiological and metabolic activities are at higher rate and this might be the reason for increase in uptake of nitrogen content from soil at early stage of crop growth. Results obtained by Poonkodi (2003) also stated that decrease in nitrogen content in leaves might be due to translocation and utilization of nutrient for flower and pod formation.

Raut *et al.* (2020) investigated the effect of foliar application of ascorbic acid and zinc on chickpea plant. Investigation showed that application of 200 ppm ascorbic acid + 0.5% $ZnSO_4$ significantly gained higher nitrogen content in leaf. Blesseena *et al.* (2020) noticed that the foliar application of 100 ppm tocopherol + 0.5 % $ZnSO_4$ recorded highest nitrogen content in leaf of chickpea. Nagwa *et al.* (2013) observed that, the application of bioregulator and vit E at 100 ppm resulted highest N content in bulb tissues of onion.

Leaf phosphorus content

Phosphorus (P) is part of the nuclei acid structure of plants which is responsible for the regulation of protein synthesis. Without phosphorus, photosynthesis could not occur. Phosphorus plays a key role in complex energy transformations that are necessary to all life, as a main ingredient in ATP (adenosine triphosphate). It is essential for the formation of seed.

At 40 DAS, treatment T_8 (100 ppm tocopherol + 0.5% $ZnSO_4$) gave significantly maximum phosphorus content followed by treatments T_9 (200 ppm tocopherol+0.5% $ZnSO_4$), T_{10} (300 ppm tocopherol+0.5% $ZnSO_4$), T_{11} (400 ppm tocopherol+0.5% $ZnSO_4$), T_{12} (500 ppm tocopherol+0.5% $ZnSO_4$), T_2 (100 ppm tocopherol), T_3 (200 ppm tocopherol), T_4 (300 ppm tocopherol), T_5 (400 ppm tocopherol) and T_6 (500 ppm tocopherol) over control.The best results related to leaf phosphorus content obtained at 55 DAS in treatment T_8 (100 ppm tocopherol+0.5% $ZnSO_4$) followed by treatments T_9 (200 ppm tocopherol+0.5% $ZnSO_4$), T_{10} (300 ppm tocopherol+0.5% $ZnSO_4$), T_{11} (400 ppm tocopherol+0.5% $ZnSO_4$), T_{12} (500 ppm tocopherol+0.5% $ZnSO_4$), T_2 (100 ppm tocopherol) when compared with treatment T_1 (control) and other treatment under study. Whereas, treatments T_3 (200 ppm tocopherol), T_4 (300 ppm tocopherol), T_5 (400 ppm tocopherol), T_6 (500 ppm tocopherol), T_7 (0.5% $ZnSO_4$) were found significantly superior in descending manner when compared with treatment T_1 (control).

It is evident from the data that phosphorus content in leaves was increased gradually up to 40 DAS and decreased at 55 DAS. It might be because of translocation of leaf phosphorus and its utilization for developing of food storage organ.

Raut *et al.* (2020) demonstrated that among the various treatments, foliar application of 200 ppm ascorbic acid + 0.5% $ZnSO_4$ was found to be significantly superior over control in phosphorus content in leaves of chickpea. Blesseena *et al.* (2020) found out that combined spray of 100 ppm tocopherol + 0.5% $ZnSO_4$ at 25 and 40 DAS recorded significantly more phosphorus content in leaves of chickpea.

Leaf potassium content

Potassium is an essential macronutrient for the process of respiration, photosynthesis and many physiological processes in plant. It plays important role in stomatal movements and acts as an activator of many enzymes involved in protein synthesis. It is important for crop yield as well as for the quality of edible parts of crops. Plant responses to low K involve changes in the concentrations of many metabolites as well as alteration in the transcriptional levels of many genes.

Leaf potassium at 40 DAS was significantly enhanced by the treatments T_8 (100 ppm tocopherol+0.5% $ZnSO_4$) and T_9 (200 ppm tocopherol+0.5% $ZnSO_4$). Next to these treatments the treatments T_{10} (300 ppm tocopherol +0.5% $ZnSO_4$), T_{11} (400 ppm tocopherol+0.5% $ZnSO_4$), T_{12} (500 ppm tocopherol+0.5% $ZnSO_4$), T_2 (100 ppm tocopherol), and T_3 (200 ppm tocopherol) also increased potassium content significantly over treatment T_1 (control). Similarly treatments T_4 (300 ppm tocopherol), T_5 (400 ppm tocopherol), T_6 (500 ppm tocopherol) and T_7 (0.5% $ZnSO_4$) were also found significantly superior in descending manner when compared with treatment T_1 (control).At 55 DAS the highest value was recorded in treatment T_8 (100 ppm tocopherol +0.5% $ZnSO_4$) followed by treatments T_9 (200 ppm tocopherol+0.5% $ZnSO_4$), T_{10} (300 ppm tocopherol+0.5% $ZnSO_4$), T_{11} (400 ppm tocopherol+0.5% $ZnSO_4$), T_{12} (500 ppm tocopherol+0.5% $ZnSO_4$) and T_2 (100 ppm tocopherol), when compared with treatment T_1 (control) and rest of the treatment under study. Similarly treatments T_3 (200 ppm tocopherol), T_4 (300 ppm tocopherol), T_5 (400 ppm tocopherol), T_6 (500 ppm tocopherol) and T_7 (0.5% $ZnSO_4$) were found significantly superior in a descending order when compared with treatment T_1 (control).

From given data, it was observed that potassium content was increased at 40 DAS but thereafter, at 55 DAS it was decreased gradually. The decrease in potassium content at advance stage might be due to diversion of potassium towards developing part i.e. pods of the green gram crop.

Raut *et al.* (2020) carried out an experiment to understand the efficacy of foliar sprays of ascorbic acid and zinc on chickpea. Study found that application of 200 ppm ascorbic acid + 0.5 % $ZnSO_4$ significantly increased potassium content in leaf.Blesseena *et al.* (2020) tested the effect of foliar application of á – tocopherol and zinc on chickpea. However, foliar application of 100 ppm tocopherol + 0.5 % $ZnSO_4$ gave highest potassium content of leaf. K content was increased in bulb tissues of onion during first season with foliar application of bio regulator and vit E at 100 ppm in the study conducted by Nagwa *et al.* (2013).

Protein content in seed

Grain legumes are known to be an important source of protein. The variability in seed protein content among different varieties of legumes is due to the genetic difference and to the interaction with differences physiological conditions of the plants. Protein content of the seed is one of the considerable factors for seed quality determination also.

The maximum seed protein was recorded in treatment T_8 (100 ppm tocopherol+0.5% $ZnSO_4$) followed by treatments T_9 (200 ppm tocopherol+0.5% $ZnSO_4$), T_{10} (300 ppm tocopherol+0.5% $ZnSO_4$), T_{11} (400 ppm tocopherol +0.5% $ZnSO_4$), T_{12} (500 ppm tocopherol+0.5% $ZnSO_4$), T_2 (100 ppm tocopherol), T_3 (200 ppm tocopherol), and T_4 (300 ppm tocopherol) when compared with T_1 (control) and rest of the treatments.

Foliar application of á – tocopherol and zinc increases the uptake and availability of nutrients and its further assimilation for biosynthesis of protein. This might be the reasons for increased protein content in seed in the present investigation.

Raut *et al.* (2020) studied the effect of foliar spray of 200 ppm ascorbic acid + 0.5 % $ZnSO_4$ in chickpea and reported that both á – tocopherol and zinc increased total protein content of leaf.The foliar application of 100 ppm tocopherol + 0.5 % $ZnSO_4$ in chickpea at 25 and 40 DAS recorded in highest protein content in the study conducted by Blesseena *et al*. (2020). The foliar application of 0.5% zinc sulphate in cluster bean at 25 and 45 DAS resulted in highest protein content (28.9%) in the study conducted by Selvaraj and Prasanna in 2012. Habbasha *et al.* (2013) conducted a field experiment for 2 years and observed that application of 0.2 % $ZnSO_4$ at seed filling stage resulted significant effect on seed protein content (21.05%) in chickpea during both years.

Yield parameter

Seed yield plant^{-1}

Seed yield is a quantitative trait which is final result of physiological activities of plant. Application of á – tocopherol and zinc also significantly enhanced chlorophyll, nitrogen, phosphorus and potassium content in leaves which might have helped in increase in yield.

Significantly, maximum seed yield plant^{-1} was recorded in treatment T_8 (100 ppm tocopherol+0.5% $ZnSO_4$) followed by treatment T_9 (200 ppm tocopherol+0.5% $ZnSO_4$). Next to these treatments, treatments T_{10} (200 ppm tocopherol +0.5% $ZnSO_4$) T_{11} (200 ppm tocopherol+ 0.5% $ZnSO_4$) and T_{12} (200 ppm tocopherol+0.5% $ZnSO_4$) also significantly enhanced seed yield plant^{-1} as compared to control and rest of the treatments under study. While, treatments T_2 (100 ppm tocopherol), T_3 (200 ppm tocopherol), T_4 (300 ppm tocopherol), T_5 (400 ppm tocopherol), T_6 (500 ppm tocopherol) and T_7 (0.5% $ZnSO_4$) were also recorded maximum seed yield plant^{-1} when compared with treatment T_1 (control).

Tocopherol found to be universal constituents of all higher plant (Bafeel and Ibrahim, 2008). It is a powerful biological antioxidant that assists the transport of electrons in photosystem – II protein complex. Zinc takes part in the metabolism of plant as an activator of several enzymes which in turn can directly or indirectly affect the synthesis of carbohydrates and protein. The increase in seed yield due to zinc application could possibly be due to the enhanced synthesis of carbohydrate and protein and their transport to the site of seed formation (Mali *et al.,* 2003).

Raut *et al*. (2019) investigated the effect of foliar spray of ascorbic acid and zinc on yield of chickpea. The foliar application of 200 ppm ascorbic acid + 0.5% $ZnSO_4$ at 25 and 40 DAS significantly enhanced seed yield of chickpea. Habbasha *et al.* (2013) observed that the application of 0.2 % $ZnSO_4$ at seed filling stage in chickpea showed significantly higher number of seeds plant^{-1} (83.92) in chickpea as compared to no foliar application.

Table 1. Effect of tocopherol and zinc on chemical, biochemical parameters and yield of green gram

Treatments	Chlorophyll content (mg g^{-1})		Nitrogen content in leaves in leaves (%)		Phosphorus content in leaves (%)		Potassium content (%)		Protein content in seed (%)	Seed yield $plant^{-1}$ (g)
	40 DAS	55 DAS	40 DAS	55 DAS	40 DAS	55 DAS	40 DAS	55 DAS		
T_1 (Control)	1.10	1.19	1.65	1.03	0.41	0.37	0.71	0.51	0.71	0.51
T_2 (100 ppm tocopherol)	1.25	1.43	2.66	2.30	0.76	0.72	1.30	1.29	1.30	1.29
T_3 (200 ppm tocopherol)	1.31	1.44	2.57	2.24	0.73	0.65	1.28	1.26	1.28	1.26
T_4 (300 ppm tocopherol)	1.27	1.43	2.45	2.16	0.72	0.66	1.24	1.11	1.24	1.11
T_5 (400 ppm tocopherol)	1.26	1.32	2.36	2.06	0.67	0.63	1.20	1.10	1.20	1.10
T_6 (500 ppm tocopherol)	1.21	1.30	2.24	1.96	0.65	0.61	1.18	1.10	1.18	1.10
T_7 (0.5% $ZnSO_4$)	1.15	1.24	1.97	1.62	0.51	0.47	1.09	0.83	1.09	0.83
T_8 (100 ppm tocopherol+0.5% $ZnSO_4$)	1.58	1.65	3.12	2.52	0.84	0.79	1.60	1.55	1.60	1.55
T_9 (200 ppm tocopherol+0.5% $ZnSO_4$)	1.55	1.60	3.07	2.45	0.83	0.77	1.51	1.37	1.51	1.37
T_{10} (300 ppm tocopherol+0.5% $ZnSO_4$)	1.52	1.57	3.00	2.38	0.81	0.76	1.43	1.35	1.43	1.35
T_{11} (400 ppm tocopherol+0.5% $ZnSO_4$)	1.48	1.54	2.96	2.37	0.80	0.76	1.39	1.35	1.39	1.35
T_{12} (500 ppm tocopherol+0.5% $ZnSO_4$)	1.46	1.50	2.88	2.35	0.79	0.75	1.37	1.34	1.37	1.34
SE (m)±	0.0915	0.0802	0.169	0.046	0.074	0.029	0.050	0.032	0.050	0.032
CD at 5%	0.2683	0.2353	0.495	0.135	0.217	0.084	0.147	0.094	0.147	0.094

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