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Research Article

Enhancing drought resilience in maize hybrid COH (M) 8: Unravelling the role of polyamines in field performance

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Abstract
Polyamines, namely spermidine and spermine, are essential organic compounds that hold significant importance in the growth and development of plants. Their beneficial effects in crops include enhancing stress tolerance, facilitating better nutrient uptake, and regulating various physiological processes. Considering these attributes, a study aimed to evaluate the effectiveness of polyamines as seed priming agents and foliar spray treatments on the growth and seed yield of hybrid maize COH(M) 8. The research was conducted in field conditions from 2022 to 2023. Various polyamine seed priming treatments (T_0 - control, T_1, Hydropriming, T_2 – Spermine 50 ppm, T_3 – Spermine 75 ppm, T_4 – Spermidine 50 ppm, T_5 – Spermidine 75 ppm, T_6 – spermine 50 ppm + Foliar Spray, T_7 – Spermine 75 + Foliar Spray were employed, each lasting for 12 hours. The seed priming solutions were prepared at a 1:1 volume/volume ratio with concentrations of spermine at 50 and 75 ppm, spermidine at 50 and 75 ppm, and a combination of both spermine and spermidine. Hydro priming was included as another treatment, and there was also an untreated control group. Furthermore, all treatments were supplemented with a foliar spray application at a concentration of 10 ppm on the 50th and 65th days after sowing (DAS). The outcome indicated that among the various polyamines tested, the combination of spermine seed priming at 50 ppm (T_2) and spermine foliar spray (T_6) significantly improved multiple growth parameters and yield under drought stress. Notably, this treatment maximized plant height, leaf area, chlorophyll content, cob length, seed yield per plot, and seed yield per hectare.

Keywords: Drought, Foliar spray, Maize, Polyamines, Seed priming, Seed yield

INTRODUCTION

Maize (*Zea mays* L.), belonging to the Poaceae family, holds significant global importance as the third most vital cereal crop after wheat and rice. India's year-round cultivation of maize benefits from its C_4 plant classification, ensuring efficient utilization of moisture and sunlight for impressive yields and total dry matter (Bell, 2017). Originally from Mexico, this cross-pollinated crop is adaptable to diverse agro-climatic conditions, boasting the highest genetic gain among cereals. However, climate change poses a threat, potentially reducing global maize productivity and grain quality due to high temperature-induced drought stress. Such stress adversely impacts plant growth and development through various morphological, physiological, and biochemical changes. Drought-sensitive maize plants experience reduced grain yield under prolonged stress, especially during the critical reproductive phase. Suboptimal temperatures at crucial stages also hamper growth and yield formation, exacerbating drought conditions. It is essential to understand and address the challenges posed by climate change to ensure sustainable maize production to meet the rising demand for food, oil, forage and biofuel (FAO, 2020).

Drought considered the most complex yet least understood natural hazard, is a recurring challenge in India, particularly in arid and semi-arid regions. Throughout history, widespread droughts have been the primary cause of famines in the country. The adverse effects of low rainfall lead to drought, resulting in a significant drop in agricultural production, with far-reaching consequences such as loss of life, human suffering, and damage to the economy and environment reported in IPCC (2023) (https://www.ipcc.ch/assessment-report/ar6/).

In recent decades, India has been facing frequent and severe droughts, occurring approximately once every three years. To address this issue effectively, enhancing our understanding of drought climatology and establishing an integrated drought information system that considers climate, soil, and water supply factors is crucial. By implementing such a comprehensive approach, we can better prepare for and mitigate the impacts of drought events in the future. Priming significantly impacts enhancing and regulating the plant's ability to tolerate any abiotic stress. It is crucial to improve the plant's capacity to cope with this stress by facilitating various mechanisms. These include better osmotic adjustment, maintaining ion homeostasis, activating antioxidant defense mechanisms, and overall physiological adjustments. As a result of these enhanced stress tolerance traits, plants can effectively withstand any kind of environmental stress, enabling them to sustain growth and productivity even under challenging conditions reported in IPCC, 6th Assessment Report, 2023 (https://www.ipcc.ch/assessment-report/ar6/)

Polyamines such as spermidine and spermine are organic compounds that play a vital role in plant growth and development. In crops, polyamines have been found to enhance stress tolerance, improve nutrient uptake, and regulate various physiological processes. They contribute to increased cell division and elongation, leading to better plant growth. Additionally, polyamines act as antioxidants, protecting crops from oxidative damage and promoting overall resilience to environmental stresses (Mishra *et al.,* 2016). The present study aimed to enhance the growth and productivity of the Hybrid maize COH(M) 8 using seed priming with polyamines under field conditions.

MATERIALS AND METHODS

The present study utilized freshly harvested hybrid maize seeds COH (M) 8 sourced from the Department of Millets, Tamil Nadu Agricultural University, Coimbatore, as the primary materials. Field experiments were conducted at the Department of Forage Crops, Tamil Nadu Agricultural University, Coimbatore. To simulate drought stress, irrigation was withheld 65 days after sowing (DAS), while control plants received continuous and full irrigation throughout the experimental period. The study incorporated polyamines, specifically spermine and spermidine, as part of the experimental approach.

Field experiment

The field experiment was conducted in the rainout shelter at the Department of Forage Crops, TNAU, Coimbatore, from 2022 to 2023. The study involved conducting experiments with seeds that were primed with polyamines, and foliar application was also performed to observe and analyze morphological, reproductive, and seed yield attributes. For the priming process, a solution was prepared using double distilled water, and the seeds were soaked in this solution for 12 hours at a volume/volume ratio of 1:1, while maintaining a temperature of 10-15°C. After the priming period (12 hours), the seeds were removed from the solution, washed with water, and then dried back to their original moisture content.

The treatments included: T_0 - Control, T_1 - Hydro priming, T_2 - Seed priming with spermine 50 ppm, T_3 - Seed priming spermine 75 ppm, T_4 - Seed priming with spermidine 50 ppm, T_5 - Seed priming with spermidine 75 ppm, T_6- Seed priming with spermine 50 ppm and foliar spray (FS) with spermine 10 ppm and T_7 - Seed priming with spermidine 50 ppm and foliar spray with spermidine 10 ppm with three replication in a randomized block design (RBD). Foliar spray was administered at 50th and 65th days after sowing (DAS) while irrigation was withheld. The plot size measured 3.0 x 3.0 m^2 with a crop spacing of 45 cm x 65 cm. Various observations were recorded at different stages of crop growth.

Observations recorded

Field emergence (%) was assessed by counting the number of germinated hills in each plot fifteen days after sowing. Plant height (cm) was determined by measuring ten randomly selected plants from each treatment in every replication. The measurements were taken from the base to the tip of the leaf, and the mean value was then expressed in centimeters (cm). The leaf area (cm^2) of all green leaves from three specifically tagged plants was measured using Stickler's linear measurement method (Stickler and Pauli, 1961). This technique calculated the leaf area per plant and expressed in square centimeters (cm²).

Leaf area (cm^2) = L x B x 0.747 Eq.1

L = length of leaf; B = breadth of leaf;

0.747 = correction factor

The stem girth (cm) was determined by using a cotton thread to measure its widest point. The average girth was then expressed in centimetres.

The duration in days to complete tasselling for the entire population within the plot to reach 100 percent tasselling was recorded and expressed as the number of days. The duration in days for the entire population within the plot to reach 100 percent silking was recorded and expressed as the number of days.

The relative growth rate (RGR) measures the increase in plant dry weight during a specific time interval relative to the initial weight. It is calculated as the dry matter increment per unit biomass per unit time or as the grams of dry weight increase per gram of initial dry weight (Grime and Hunt, 1975) (Williams, 1946)

RGR = $\text{Log e } W_2 - \text{Log e } W_1 / t_2 - t_1$ Eq. 2

Where, W_1 and W_2 are whole plant dry weight at t_1 and t_2, respectively t_1 and t_2 are a time interval in days

Leaf chlorophyll (mg/g) content was quantified using the method proposed by Yoshida *et al*. (1971). The optical density of the extract was measured at 645 nm, 663 nm, and 652 nm using a Spectrophotometer.

The cob length and breadth (cm) were measured in randomly selected ten plants using a measuring scale and the mean value was calculated and expressed in centimeters. The cob weight (g) was measured using an electronic balance. The result was averaged and the mean value was expressed in grams. The weight of a hundred seeds (g) from four different replications was measured and recorded in grams, with precision up to three decimal places. Subsequently, the average weight was calculated. Seeds from each plot (kg) were threshed individually, dried to attain 13% moisture content, and weighed, with the results expressed in kilograms. The computed seed yield per hectare (kg/ha) was calculated based on the plot yield and presented in kilograms per hectare.

Statistical analysis

The analysis of variance was carried out and a comparison was made by Duncan's Multiple Range Test (DMRT). The mean difference is significant at the P-values < 0.05. Statistical analysis was performed using the SPSS 16.0 software (SPSS Inc., Chicago, USA).

RESULTS AND DISCUSSION

Seed priming and foliar spray with polyamines have gained significant attention in agricultural research as potential strategies to enhance the performance of maize crops. Polyamines, such as spermine and spermidine, play essential roles in various physiological processes, including stress tolerance, cell division, and growth regulation (Afzal *et al*., 2022)

Priming and foliar spraying with spermine @ 50 ppm (T_6) for maize crop recorded the maximum increase of field emergence (91%), which was 5.0% more over nonprimed seeds (T_0) (86%) under drought stress (Fig 1). The increase in germination percentage observed in this study can be attributed to the combined effect of polyamine seed treatments on the production of hydrolytic enzymes and growth hormones such as IAA and gibberellins. This finding aligns with previous research indicating that priming improves germination by enhancing water uptake, improving water relations, and promoting hydrolytic enzyme production in primed brassica seeds (Lechowska *et al.,* 2019). Furthermore, the results of this study are consistent with earlier reports by Shi *et al.* (2013) for Bermuda grass (*Cynodon dactylon*) seeds treated with polyamines, where the survival rate and germination were significantly higher compared to untreated seeds without priming. The findings from the Dursun and Ekinci (2010) studies collectively highlight the positive influence of polyamine seed treatments on germination performance, indicating their potential as a beneficial practice to enhance seedling establishment and overall crop productivity.

The maximum plant height at 30 and 45 DAS (75.2 cm and 205.7 cm) was observed in seed priming and foliar

Table 1. Effect of seed priming and foliar spray treatment with spermine and spermidine on plant height (cm) of maize under drought condition

Treatments	Plant height (cm)		
	30 DAS	45 DAS	65 DAS
T_0 - Control	65.31 ± 0.36 ^{i}	185.6 ± 0.65 ^{i}	199.8 ± 1.01 ^{i}
T_1 - Hydropriming	68.35 ± 0.25 ^{gh}	189.3 ± 0.67 ^{gh}	223.6 ± 1.05 ^{gh}
T_2 - spermine 50 ppm	73.21 ± 0.63 ^{b}	202.3 ± 0.48 ^{b}	241.5 ± 1.22 ^{b}
T_3 - spermine 75 ppm	73.01 ± 0.25 ^{c}	197.2 ± 0.55 ^{c}	240.9 ± 1.31 ^{c}
T_4 - spermidine 50 ppm,	71.61 ± 0.33 ^{e}	192.4 ± 0.48 ^{e}	233.5 ± 0.87 ^{e}
T_5 - spermidine 75 ppm	72.15 ± 0.42 ^{cd}	193.5 ± 0.64 ^{cd}	237.6 ± 1.19 ^{cd}
T_6 - spermine 50 ppm + Folair spray	75.2 ± 0.31 ^{a}	205.7 ± 0.52 ^{a}	245.3 ± 1.36 ^{a}
T_7 - spermine 75 ppm + Foliar spray	70.28 ± 0.22 ^{f}	190.8 ± 0.47 ^{f}	228.5 ± 1.23 ^{f}

Table 2. Effect of seed priming and foliar spray treatment with spermine and spermidine on vegetative and phenological parameter of maize under drought condition

Treatments	Leaf area (m^2)	Stem girth (cm)	Days to complete tasseling	Days to complete silking
T_0 - Control	566.3 ± 1.37 ^{i}	4.5 ± 0.03 ^{i}	53 ± 0.28 ^{ns}	52 ± 0.15 ^{ns}
T_1 - Hydropriming	578.6 ± 1.33 ^{gh}	4.9 ± 0.01 ^{gh}	53 ± 0.23 ^{ns}	52 ± 0.22 ^{ns}
T_2- spermine 50 ppm	608.3 ± 1.21 ^{b}	7.4 ± 0.04 ^{b}	51 ± 0.13 ^{ns}	50 ± 0.23 ^{ns}
T_3 - spermine 75 ppm	594.6 ± 0.45 ^{c}	6.4 ± 0.06 ^{c}	53 ± 0.17 ^{ns}	52 ± 0.27 ^{ns}
T_4 - spermidine 50 ppm,	585.4 ± 0.44 ^{e}	5.5 ± 0.07 ^{e}	53 ± 0.22 ^{ns}	52 ± 0.22 ^{ns}
T_5 - spermidine 75 ppm	590.5 ± 0.42 ^{cd}	5.9 ± 0.04 ^{cd}	53 ± 0.18 ^{ns}	51 ± 0.18 ^{ns}
T_6 - spermine 50 ppm + Foliar spray	608.8 ± 1.31 ^{a}	7.7 ± 0.05 ^{a}	51 ± 1.19 ^{ns}	50 ± 1.15 ^{ns}
T_7 - spermine 75 ppm +Foliar spray	580.4 ± 0.89 ^{f}	5.1 ± 0.05 ^{f}	52 ± 0.25 ^{ns}	51 ± 0.16 ^{ns}

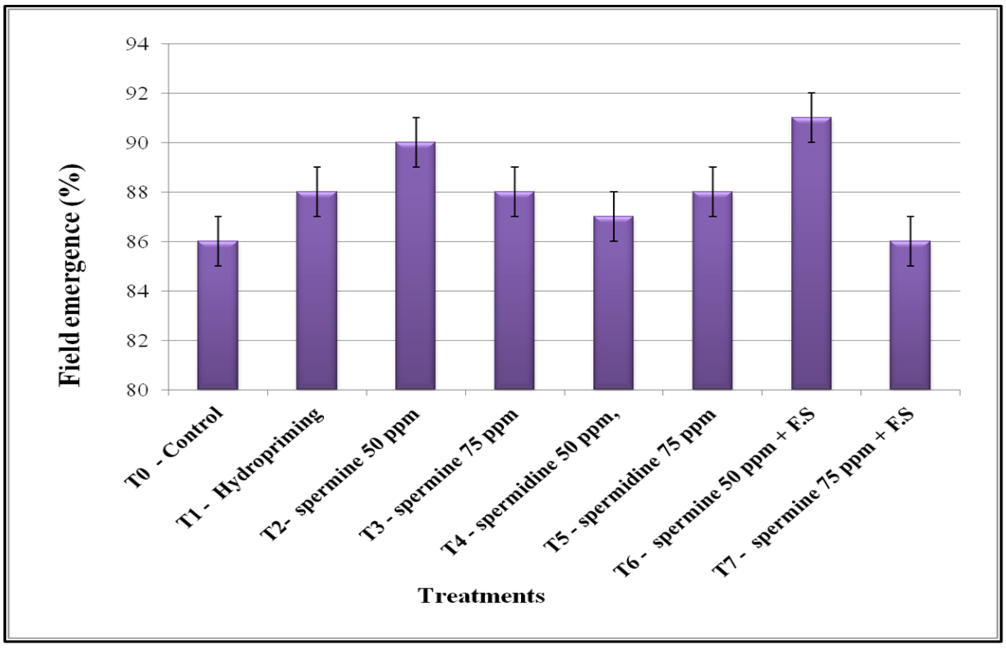

Fig. 1. *Effect of seed priming and foliar spray treatment with spermine and spermidine on field emergence % of maize under drought condition*

spray with spermine @ 50 ppm (T_6), whereas control (T_0) registered the minimum plant height (65.31 cm and 185.6 cm). Seed priming combined with foliar spray of spermine @ 50 ppm (T_6) recorded higher plant height at 65 DAS (245.6 cm), with an increase of 22.7% over control (Table 1). In terms of vegetative growth, polyamine treatments influence the plant's leaf area and stem girth. In the case of leaf area, seed priming and foliar spraying with spermine @ 50 ppm (T_6) recorded a higher value (608.8 m^2), and control (T_0) registered the lowest one (Table 2).

In a study by Tiburcio *et al*. (2014), spermine application in crops viz., rice, tomato, wheat and maize led to a notable increase in plant biomass, specifically enhancing plant height, root length, and the dry biomass of both roots and shoots. Moreover, under drought conditions, the spermine treatment positively influenced various aspects, viz., photosynthesis, chlorophyll content, stress tolerances, growth parameters and yield characters of plant physiology under abiotic stress condition. Floral induction, fruit set, leaf senescence, DNA synthesis, and osmolyte balance were significantly upregulated in spermine-treated plants compared to the control group. The findings align with a study by Zheng *et al*. (2016), which demonstrated that spermine priming improved rice seed growth. The positive effects of spermine on plant growth appear to be attributed to its role in enhancing the production of phytohormones, particularly gibberellins (GAs) and indole-3-acetic acid (IAA). These phytohormones play vital roles in cell division and elongation, consequently promoting overall plant growth.

The phenological parameters, such as days to complete tasseling and days to complete silking were not influenced by the exogenous application of polyamine. The significant difference was observed in polyamine treatments' relative growth rate and total chlorophyll content. The result revealed that seed priming and foliar spraying of spermine @ 50 ppm (T_6) recorded maximum value of relative growth rate (3.84 mg g^{-1} day^{-1}) and total chlorophyll (0.483 mg/g). Control (T_0) recorded the lowest value of relative growth rate (1.13 mg g^{-1}

Table 3. Effect of seed priming and foliar spray treatment with spermine and spermidine on yield paramters of maize under drought condition

Treatments	Cob length (cm)	Cob breadth (cm)	Cob weight (g)	100 seed weight (g)
T_0 - Control	16.4 ± 0.016 ^{i}	13.2 ± 0.13 ^{i}	60.17 ± 0.63 ^{h}	23.54 ± 0.15 ^{h}
T_1 - Hydropriming	17.5 ± 0.002 ^{h}	14.3 ± 0.24 ^{h}	61.36 ± 0.06 ^{g}	23.82 ± 0.36 ^{g}
T_2 - spermine 50 ppm	21.5 ± 0.019 ^{b}	15.2 ± 0.21 ^{b}	73.52 ± 1.08 ^{b}	27.13 ± 0.03 ^{b}
T_3 - spermine 75 ppm	19.8 ± 0.018 ^{c}	14.9 ± 0.28 ^{c}	72.45 ± 0.39 ^{c}	26.82 ± 0. 23 ^{c}
T_4 - spermidine 50 ppm,	18.7 ± 0.057 ^{f}	14.0 ± 0.19 ^{f}	64.82 ± 0.70 ^{e}	25.47 ± 0.47 ^{e}
T_5 - spermidine 75 ppm	19.3 ± 0.018 ^{de}	15.0± 0.24 ^{de}	69.25 ± 0.55 ^{d}	26.42 ± 0.23 ^{bd}
T_6 - spermine 50 ppm + Foliar spray	22.2 ± 0.057 ^{a}	15.4 ± 0.16 ^{a}	74.52 ± 0.78 ^{a}	29.34 ± 0.01 ^{a}
T_7 - spermine 75 ppm + Foliar spray	17.9 ± 0.025 ^{g}	15.3 ± 0.28 ^{g}	63.11 ± 0.55 ^{f}	24.02 ± 0.02 ^{f}

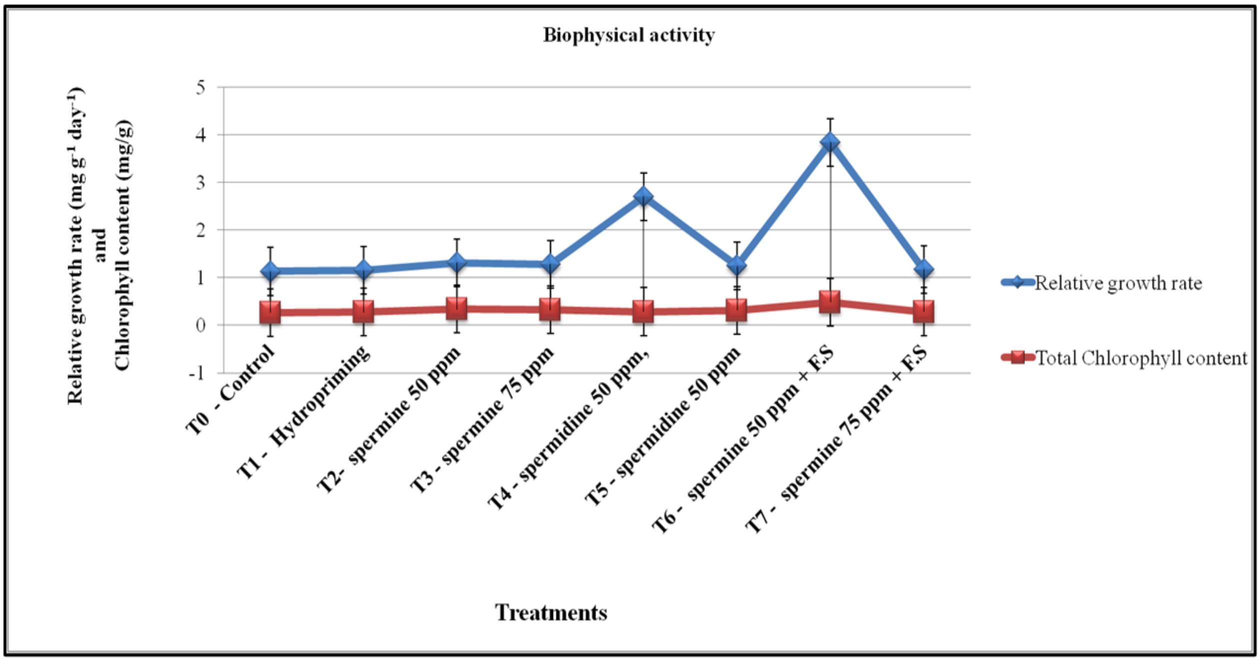

Fig. 2. *Effect of seed priming and foliar spray treatment with spermine and spermidine on biophysical acitivity of maize under drought condition*

day^{-1}) and total chlorophyll (0.266 mg/g) (Fig 2).

Tiburcio *et al*. (2014) observed that both spermine and spermidine treatments enhanced chloroplast metabolism, increasing chlorophyll content in salt-stressed rice plants. This finding aligns with the results presented by Ebeed *et al*. (2017), where spermine treatments in sorghum and maize under drought stress also resulted in elevated chlorophyll levels. Furthermore, the synthesis of indole-3-acetic acid (IAA) was found to play a crucial role in this process, as it was shown to increase the production of photosynthetic pigments and metabolites. The studies mentioned above are consistent with the research conducted by Du and Tuong (2002) and the present research study, which demonstrated that polyamines, including spermine and spermidine, positively affect chlorophyll content. Hence, in the present study, these polyamines significantly increased the activity of enzymes necessary for chlorophyll biosynthesis, even under moisture-deficit conditions. Overall, the evidence suggests that spermine and spermidine play a beneficial role in enhancing chloroplast metabolism and increasing chlorophyll content, vital for improved photosynthesis and metabolic processes in plants, particularly under drought-stress conditions.

The polyamines significantly influenced seed yield parameters such as cob length & breadth, cob weight, and 100 seed weight. The maximum cob length (22.2 cm), cob breadth (15.4), cob weight (74.52 g) and 100 seed weight (29.34 g) were noticed in priming and foliar spraying of spermine @ 50 ppm (T_6). While analyzing the seed yield of various treatments, seed priming and foliar spray of spermine @ 50 ppm (T_6) recorded a maximum seed yield of 5.73 kg/plot and the control (T_0) recorded the lowest seed yield 4.52 kg/plot. Similarly, seed priming and foliar spray of spermine @ 50 ppm (T_6) increased seed yield (kg/ha) 26.7% over control (Table 4)

Polyamines have positively impacted plant growth by promoting favorable photosynthesis and chlorophyll content effects. These beneficial effects include higher

Table 4. Effect of seed priming and foliar spray treatment with spermine and spermidine on yield paramters of maize under drought condition

Treatments	Seed yield per plot (kg)	Seed yield per ha (kg)
T_0 - Control	4.52 ± 0.73 ^{h}	5022.2 ± 1.22 ^{h}
T_1 - Hydropriming	4.61 ±1.02 ^{g}	5122.2 ±1.23 ^{g}
T_2 - spermine 50 ppm	5.62 ±1.25 ^{b}	6244.4 ±1.33 ^{b}
T_3 - spermine 75 ppm	5.44 ± 0.14^c	6044.4 ± 1.45 ^{c}
T_4 - spermidine 50 ppm,	4.86 ± 0.36 ^{e}	5400 ± 1.39 ^{e}
T_5 - spermidine 75 ppm	5.31 ± 0.69 ^{d}	5900 ± 1.46 ^{d}
T_6 - spermine 50 ppm + Foliar spray	5.73 ± 1.42 ^{a}	6366.6 ± 1.55 ^{a}
T_7 - spermine 75 ppm + Foliar spray	4.76 ± 0.79 ^{f}	5288.8 ± 1.25 ^{f}

photosynthetic activity and increased nutrient exchange, accumulating more dry weight in the form of primary metabolites such as sugars, proteins, and fatty acids. This accumulation of essential nutrients may contribute to an overall increase in seed yield. This finding is consistent with a study by Munir and Aftab (2009), which found that polyamine seed priming in salt stressed tomato plants resulted in higher plant yields attributed to enhanced photosynthetic activities. Moreover, recent work by Li and Wang (2020) showed that treated plants exhibited a significant increase in seed yield, aligning with current research results that demonstrated a similar impact of polyamine treatments on maize seeds, leading to a notable increase in seed yield. Additionally, Muthumanickam *et al*. (2011) documented that when polyamines and other phytohormonal seed priming treatments were applied, soybean growth and seed yield were notably increased, further supporting the positive influence of polyamines on photosynthesis and chlorophyll content, contributing to improved plant growth and ultimately leading to increased seed yield, as demonstrated in various studies involving different plant species.

Conclusion

The present study concluded that the detailed outcomes of the experiments emphasize critical parameters like field emergence, plant height, leaf chlorophyll content, and seed yield. The present findings illuminate the potential advantages of polyamines in enhancing maize crop performance under water deficit stress. Seed priming and foliar spray with spermine 50 ppm to drought-stressed maize crop resulted in enhanced growth parameters such as improved plant height, field emergence, chlorophyll content, and yield characters. Thus, seed priming with polyamines like spermine improved the physiological, morphological and biochemical parameters of maize crop under drought stress. Furthermore, the results of this present study (seed priming and foliar spray with spermine 50 ppm) advance our understanding of the utility and efficacy of polyamine-based seed priming and foliar spray as sustainable agricultural approaches. This research underscores their significance in meeting the escalating food production demands amid shifting environmental conditions.

Conflict of Interest

The authors declare that they have no conflict of interest.

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