



RESEARCH ARTICLE

Seed priming with silicon dioxide and potassium nitrate alleviates the ill effects of water stress in chilli (*Capsicum annuum* L.) by suppressing reactive oxygen species (ROS) accumulation and improving antioxidant defence systems

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Abstract

Deficit soil moisture is one of the most critical limiting factors affecting crop growth and overall productivity. Because of its low membrane integrity, high stomatal conductance and increased transpiring leaf surface, the essential solanaceous crop chilli is highly vulnerable to drought. One crucial stress management technique for combating water stress is seed priming. Chilli seeds were subjected to different priming agents 2.5 % potassium nitrate (KNO₃), 3 % silicon dioxide (SiO₂) and unprimed seeds with distilled water for 24 hours. Prior to the flowering stage, the crop was subjected to moisture stress by withholding irrigation in both greenhouse and field conditions. Results of both experiments revealed that seed priming with 2.5 % KNO₃ resulted in enhanced physiological traits like relative water content, specific leaf area, total chlorophyll and biochemical traits like malonaldehyde, H₂O₂, trehalose, α-amylase activity, superoxide dismutase, total soluble sugars and total soluble protein, followed by quality traits like capsaicin, vitamin C and yield traits viz., plant height, number of flowers plant⁻¹, number of fruits plant⁻¹ and fruit yield plant⁻¹, whereas 3 % SiO₂ primed seeds recorded significantly higher values for total proline content and cell membrane stability index compared to unprimed seeds. Seeds primed with 3 % SiO₂ recorded early flowering, whereas seeds primed with 2.5 % KNO₃ recorded the first fruiting stage. The results showed that seeds with 3% SiO₂ can be recommended for seed formation in cases of water shortage and seeds primed with 2.5% KNO₃ demonstrated high antioxidant levels and the maximum capacity to absorb water.

Keywords: chilli; flowering stage; physiological traits; seed priming; water stress

Introduction

Chilli (*Capsicum annuum* L.) from the genus *Capsicum* belongs to Solanaceae family. Chilli, an important commercial crop species of India, known for its berries. The quintessential nature of chilli is primarily because of its dual use both as a vegetable and as spice (1). India is the leading producer, contributing 43 % of global production and accounting for 42 % of total spice exports to various parts of the world. Uncertainty in rainfall events in tropical countries severely impacted the production of chilli. In chilli drought conditions can prompt significant changes in plant's morphological and physiological characteristics. It affects numerous traits of plant growth, including a reduction in shoot length, crop growth rate, leaf quantity, area (2) and biomass accumulation (3).

Water stress has multiple effects on the chilli crop, ranging from seedling stage to maturity. It hinders seed germination and seedling development at the seedling level (4). Chilli is a sensitive plant when it comes to drought conditions. In chilli, instances of stress during the vegetative stages were likewise linked to a decrease in the overall fruit weight and a lowered photosynthetic rate. Drought stress during the vegetative stage interferes with organ development and plant growth, resulting in a 30-50 % reduction in yield. However, severe drought stress during the flowering phase can lead to chilli yield losses ranging from 45.9 % to 100 % (5). Stress administered during the pre-anthesis stage shortens the anthesis period; nevertheless, stress provided during the post-anthesis stage reduces fruit seed formation (6). Likewise, the occurrence of water stress significantly hampered the morphological and yield-related features, such as the number of fruits per plant, fruit diameter and length (7).

To provide plants with tolerance to water stress, seed priming has evolved as a crucial stress management technique. Controlled hydration, also known as "seed priming," allows seeds to absorb water while preventing radical protrusion (8). It decreases the time between planting and seedling emergence and shields the seeds from biotic and abiotic stresses during the early phases of establishment and emergence (9). Priming acts as a signalling system, activating specific stress response pathways in primed seeds -a process known as memory or a primed state -allowing plants to respond more quickly and effectively when confronted with environmental difficulties (10). Primed seeds possess the ability to quickly ingest and revitalize the seed metabolism, thereby augmenting the rate of germination (11). As it plays a significant role in seed germination, priming with the right concentration of pre-treatment solution is essential (12).

Numerous priming methods have been extensively employed, such as hormone priming, halo priming, osmo-priming, hydro-priming and other chemical solutions. The benefits of seed priming include improved seed performance, uniform germination, robust and healthy plant establishment, increased yield across diverse ecosystems, heightened tolerance to environmental stressors and the effective overcoming of seed dormancy (13). The pre-germination metabolic activities induced by priming increase water absorption and the mobilization of enzymes, such as amylase, cellulase and xylanase, which persist even when germination is delayed or inhibited (14).

Si, KNO₃ and SA priming treatments improved the emergence, seedling growth, yield and tolerance to salinity and drought of rice, maize, wheat and barley (15). In rice grown under drought conditions, priming with potassium nitrate and silicon dioxide enhanced germination, seedling development and the expression of defensive enzymes (15). In the F2 generation of chilli, seed treatments increased seed germination and seedling metrics (16). Studies highlighting the advantages of seed priming are widely available; however, there is a dearth of data about the relative efficacy of various seed priming methods in mitigating water stress in chilli. The goal of the current study was to evaluate the impact of seed priming on physiological and biochemical mechanisms as well as yield components in chilli under water stress conditions. Specifically, the study focused on the effects of seed priming with KNO₃ and SiO₂, which confer tolerance to water deficit stress.

Materials and Methods

At the College of Agriculture, Vellayani, Kerala Agricultural University, which is located at an altitude of 29 mean sea level and is located at 80°5' N latitude and 76°0'9" E longitude, a series of studies consisting of one pot trial and one field trial were carried out in the polyhouse and instructional farm. For the study, Vellayani Anugraha was chosen and seeds were procured from the Department of Vegetable Science, College of Agriculture, Vellayani, Thiruvananthapuram. The cultivar Vellayani Anugraha yields a high of 27 t/ha and is distinguished by its long, green, visually pleasing fruits that have moderate pungency and early maturity (17).

Seed priming treatments

In order to prevent microbial infection, seeds were surface sterilized for 5-10 min using 0.1 % Mercuric chloride before being treated with 2.5 % KNO₃ and 3 % SiO₂ concentrations. Three rinses with distilled water followed this. After being primed with 2.5 % KNO₃, 3 % SiO₂ and distilled water for 24 hours, the seeds were left to dry in the shade to maintain their initial moisture content before being exposed to germination.

Evaluation of the efficiency of seed priming methods for water stress tolerance in chilli under pot conditions

The primed seeds, as well as the control counterpart, were evaluated for water stress tolerance using morpho-physiological and yield traits under a dry-down experiment initiated at the time of flowering. The design employed was a two-factor, completely randomized design (CRD), with six replications, three pots per replication and two plants in each pot for each treatment. Thirty-five DAS, the seeds were transplanted into earthenware pots (25 cm diameter, 30 cm height; 10 kg soil capacity, filled with air-dried soil, sand and farmyard manure in a 2:1:1 (v/v) ratio. The soil was loamy, having a pH of 5.8. The crop was raised according to the Package of Practices recommendations (17). Water stress was created by stopping irrigation before flowering until the relative water content (RWC) of the leaves reached 70%. At this point, water stress was imposed for three more days, followed by morpho-physiological observations. At the harvesting stage, yield and yield-attributing traits were recorded.

Evaluation of the efficiency of seed priming methods for water stress tolerance in chilli in field conditions.

The primed and unprimed seeds, 35 DAS in portrays, were transplanted into the field adhering to in well-drained alfisols (17). The experiment was designed as a two-factor RBD (randomized block design) and replicated three times. Each treatment, in each replication, was given a plot size of 20 sq m. Similar to the pot experiment, water stress was induced by withholding irrigation until the relative water content (RWC) of the leaves reached 70 %. At this point, the water stress was continued for seven more days, after which observations on physiological characteristics were made. At the harvesting stage, yield and yield components were measured.

Physiological and biochemical traits measured during experimentation I & II

Physiological and biochemical observations were conducted when the plants began to experience stress at the flowering stage. Traits viz., RWC (relative water content), CSI (Cell membrane stability index), SLA (Specific leaf area), MDA (Malondialdehyde), H₂O₂ (Hydrogen peroxide), proline, trehalose, α-amylase activity, TCC (total chlorophyll content), SOD (superoxide dismutase activity), TSS (total soluble sugar) and TSP (total soluble protein) were recorded during experimentation periods.

Relative water content, cell membrane stability index (CSI) and specific leaf area

To determine RWC, the leaf's fresh weight (FW), turgid weight (TW) and dry weight (DW) were taken into account and represented in percentages using the formula below as Equation 1(18).

$$\text{Relative water content (RWC) (\%)} = \frac{\text{FW-DW}}{\text{TW-DW}} \times 100 \quad (\text{Eqn. 1})$$

To eliminate adhered electrolytes, samples were triple-washed in deionized water. The leaf discs were sliced, put in 10 mL of deionized water, sealed in a 20 mL vial and let to sit at room temperature in the dark for a whole day. A conductivity meter (C1/T1) was used to measure the conductivity. These vials were then autoclaved, killing the leaf tissue for 15 min and releasing the electrolytes. The second reading of conductivity was taken after cooling (19). For all treatments, these two measurements have been performed individually (C₂ and T₂).

The cell membrane stability index was calculated by using the following formula and expressed as a percentage.

$$\text{CMS (\%)} = [1 - (T_1/T_2) / 1 - (C_1/C_2)] \times 100 \quad (\text{Eqn. 2})$$

T and C represent stress and control samples, respectively.

SLA was computed from a set of fully expanded leaves following the graphical method using the following formula and expressed in cm² g⁻¹.

$$\text{SLA} = (\text{LA}/\text{DW}) \quad (\text{Eqn. 3})$$

DW: dry weight of leaf and LA: leaf area

Biochemical parameters

Malondialdehyde (MDA) was quantified and the results were expressed in terms of fresh weight nmol g⁻¹ by using the thiobarbituric acid (TBA) method (20). Similarly, the hydrogen peroxide was measured and expressed as μmoles g⁻¹ FW (Fresh weight) (21). Proline content and SOD activity was determined (22,23). One unit of SOD enzyme activity was defined as the amount of enzyme that produced 50 % inhibition of the reduction of NBT (24).

Trehalose, α-amylase activity, total soluble sugar, total chlorophyll content and total soluble

To ascertain the trehalose content, methodology using anthrone reagent was adopted (25). Trehalose was expressed as micromoles per gram fresh weight (FW). For α-amylase activity was measured and expressed in % (26).

$$\% \text{ amylose} = x / 2.5 \times 100 \text{ mg amylose} \quad (\text{Eqn. 4})$$

Total soluble sugars and total soluble protein were estimated (27, 28). Total chlorophyll content was ascertained using the DMSO technique and given by following the formula (29).

$$\text{Total chlorophyll content (mg g}^{-1}\text{)} = (20.2 \times \text{A}_{645} + 8.02 \times \text{A}_{663}) \times V / (1000 \times W) \quad (\text{Eqn. 5})$$

Quality traits - Capsaicin content and vitamin C content

To determine the effect of water stress on fruit quality, quality metrics were noted from the harvested fruit samples. The bluish complex that is formed when capsaicin and Folin-Dennis reagent mix is measured calorimetrically (30). Vitamin C is assessed by dichlorophenol indophenol titration method (31).

Yield and yield attributing traits

In pot conditions, all replications were used to record observations, whereas, in field conditions, five plants were tagged in each replication, avoiding the border rows. At the vegetative stage and flowering stage, the plant height was

measured from the plant's base (soil level) to the tip of the top leaf of the shoot with the help of a 1 m scale and the average height was computed from each treatment. Followed by the number of flowers per plant during the flowering stage, the number of fruits per plant overall of all pickings and fruit yield per plant expressed in grams.

Data analysis

For both field experiments and pot culture studies, the data were statistically analyzed using the Analysis of Variance Technique (ANOVA). The LSD (least significant difference) approach was used to compare the means. Standard Error (SE) and Critical Difference were calculated using GRAPES software (32).

Results

Effect of seed priming methods on morpho-physiological, biochemical and yield traits under well-watered and water-stressed plant under pot study

RWC was found to vary significantly at the moisture treatment level (LoS < 0.001), priming method level (LoS < 0.001) and interaction level (Seed priming × Moisture stress) (LoS < 0.05). Seed priming with KNO₃ at 2.5 % resulted in a 20.8 % higher RWC (60.16 %) compared with unprimed water-stressed plants, whereas priming agents displayed no significant impact on CMSI. Specific leaf area was found to vary significantly at both factors at P < 0.01. KNO₃ at 2.5 % under moisture stress elevated SLA by 17.44 % compared to unprimed stressed seeds. Total chlorophyll content (TCC) and Superoxide dismutase (SOD) were found to be significantly elevated (LoS < 0.01), whereas Malondialdehyde (MDA) and H₂O₂ were reduced (LoS < 0.05). Under stress conditions, KNO₃ at 2.5 % increased TCC by 13.02 % and SOD by 15.56 % and reduced levels of MDA by 13.88 % and H₂O₂ by 3 % compared with unprimed seeds (Fig. 1).

Trehalose content (0.07 to 0.17 μmol g⁻¹ FW), proline (0.59 to 3.12 μmol g⁻¹) and total soluble sugars (TSS) exhibited no significant variation, whereas α-amylase content varied significantly (LoS < 0.001) from 12.77 μmol maltose formed min⁻¹ mL⁻¹ under the unprimed stressed condition to 16.76 μmol maltose formed min⁻¹ mL⁻¹ when treated with 2.5 % KNO₃ (13.01 % increase), whereas SiO₂ resulted in 5.47 % increased activity. Total soluble protein (TSP) at interaction had a significant impact (LoS < 0.01). Among the quality parameters, capsaicin content varied significantly with moisture level and seed priming methods at P < 0.001 and there was an interaction (LP 0.05), whereas vitamin C content was not affected. 2.5 % KNO₃ increased capsaicin content by 54.63 % and by 44.68 % when treated with 3 % SiO₂ (Fig. 2).

Plant height varied significantly (LoS < 0.05) at the interaction level between moisture level and priming method. Under water-stress conditions, seed priming with 2.5 % KNO₃ retained plant height by 25 % and with 3 % SiO₂ by 17.58 % compared to unprimed stressed seeds. Seed priming methods (LoS < 0.05) and its interaction with moisture levels (LoS < 0.05) significantly impacted the number of flowers per plant, resulting a 17.58 % reduction under stress and unprimed conditions, whereas priming restricted this loss only to 4.7 % when treated with 2.5 % KNO₃ and 2.8 % with 3 % SiO₂. The number of fruits and fruit yield did not vary with moisture level

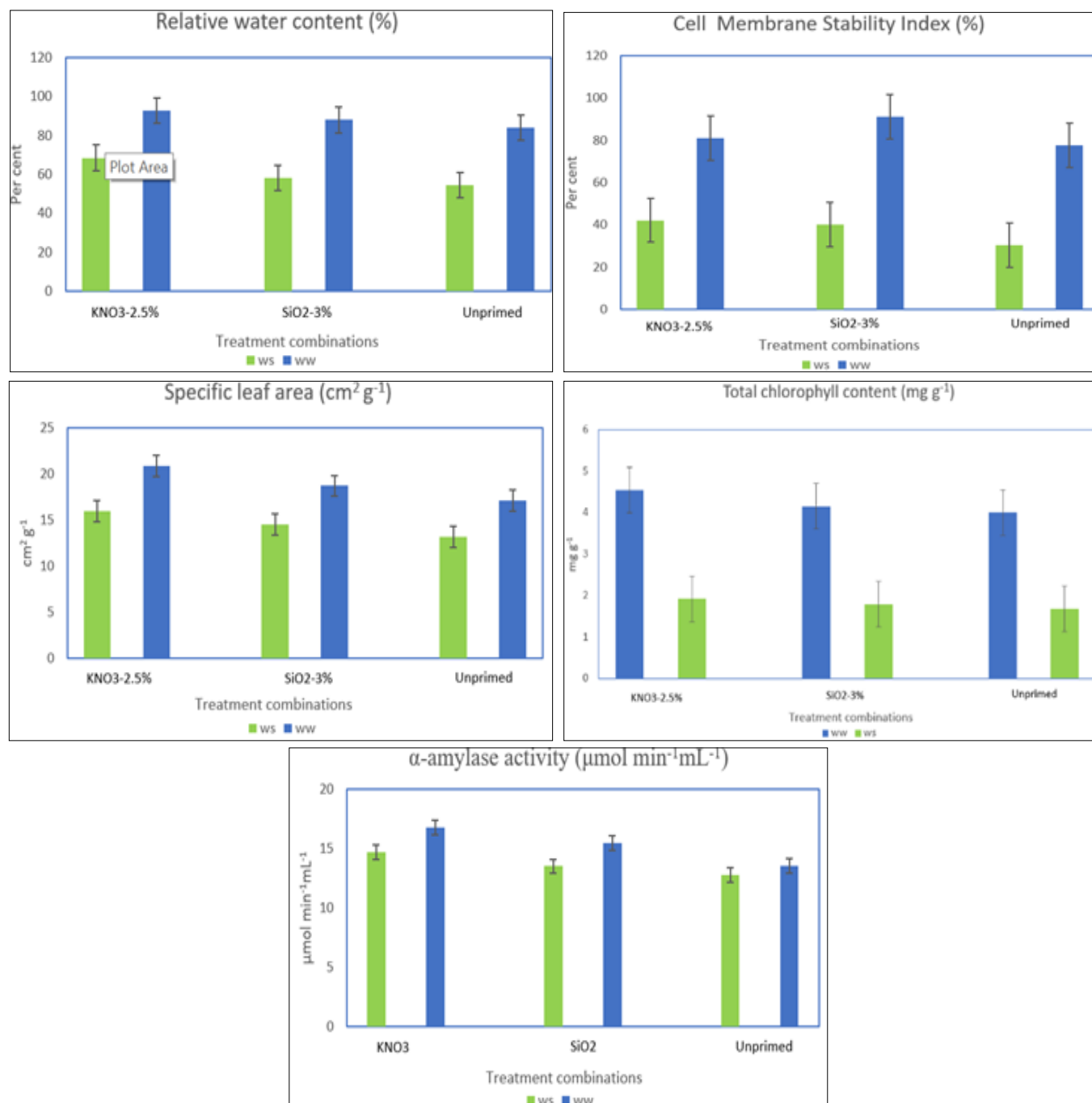


Fig. 1. Impact of seed priming agents Viz., KNO₃ and SiO₂ on Relative water content, Cell membrane stability index, Specific leaf area, Total chlorophyll content, SOD activity and α-amylase activity under moisture stress (WS) and well-watered (WW) conditions under pot study

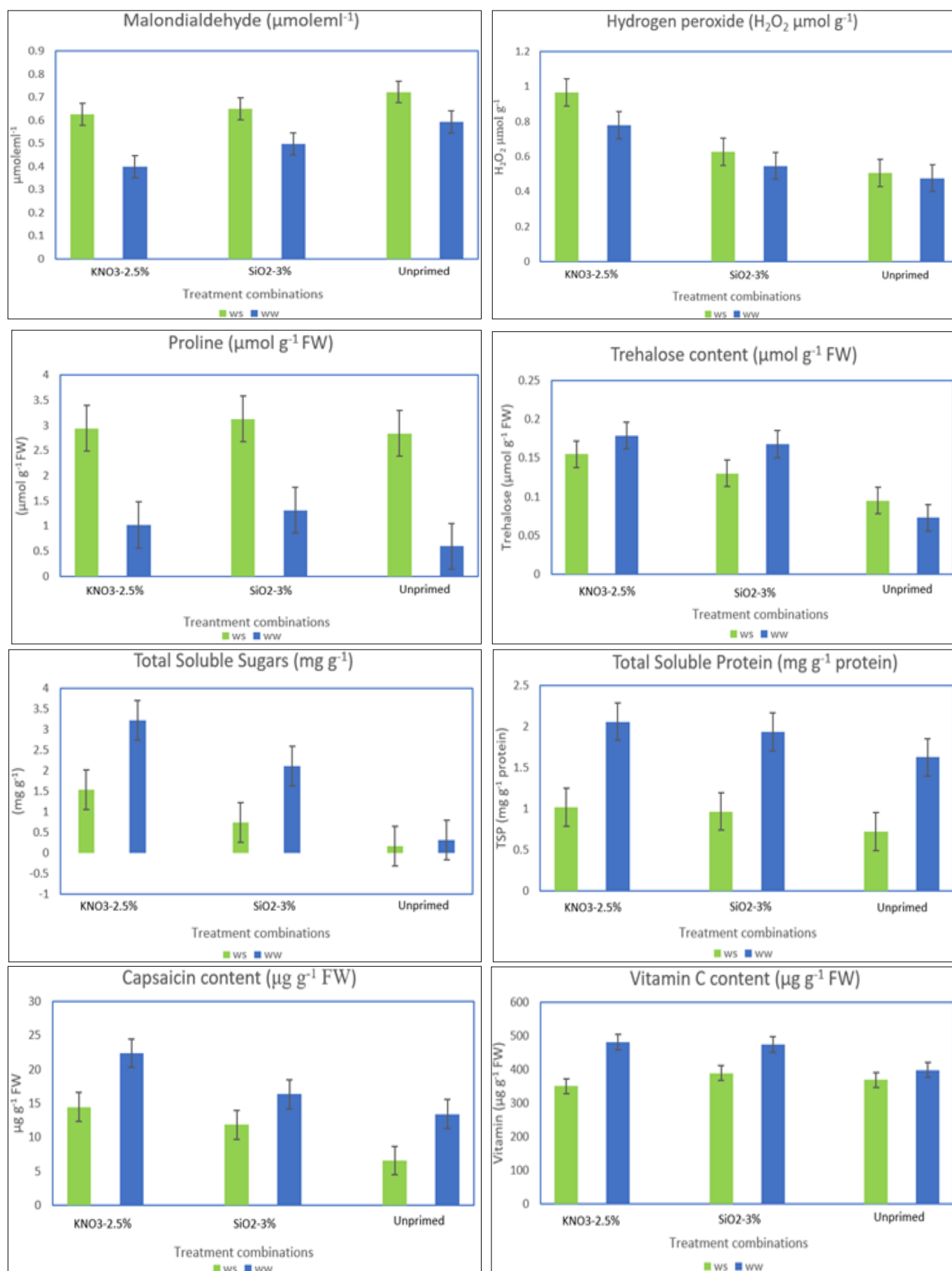


Fig. 2. Impact of seed priming agents *Viz.*, KNO₃ and SiO₂ on Malondialdehyde, Hydrogen peroxide, Proline, Trehalose, Total soluble sugar, Total soluble protein, Capsaicin content and Vitamin C under moisture stress (WS) and well-watered (WW) conditions under pot study.

and seed priming methods, but they had a significant impact together ($LoS < 0.05$). under water stress conditions, treating of seeds with 2.5 % KNO_3 increased number of fruits by 21.8 % and fruit yield by 17.8 %, followed by treating of seeds with 3 % SiO_2 increased number of fruits by 19.32 % and fruit yield by 14.4 % (Fig. 3).

Evaluation of various seed priming methods for water stress tolerance in chilli at the flowering stage under field condition

Morpho and physiological traits

Seed priming exhibited a profound impact on physiological traits of plants under stress viz., RWC and CMSI. Seeds treated with KNO_3 at 2.5 % significantly elevated levels of RWC under stress condition by 19.55 % (68.16 %, $LoS < 0.01$) and in case of CMSI, treating the seeds with 3 % SiO_2 increased CMSI by 20.69 % (48.33 %, $LoS < 0.01$) compared with unprimed seeds under stress. SLA was found significantly varying under interaction with moisture levels and priming agent ($LoS < 0.05$). Though 50 % reduction in TCC was noticed under stress, priming resulted an increase in TCC by 18.65 % with 2.5 % KNO_3 and by 19.07 % with 3 % SiO_2 (Table 1).

MDA and H_2O_2 content

Water stress and seed priming agents significantly influenced MDA levels ($LoS < 0.01$). At 3 % SiO_2 , significantly reduced the levels of MDA by 14.86 % and 2.5 % KNO_3 by 1.3 %. Similarly, H_2O_2 content under stress conditions reported 43.75 % when seeds are treated with 3 % SiO_2 ($0.54 \mu mol g^{-1}$), followed by 2.5 % KNO_3 ($0.62 \mu mol g^{-1}$) with 35 % reduction (Table 1).

Proline, α -amylase activity, SOD activity and total soluble protein assay.

In water stress conditions, proline accumulation varied from $0.59 \mu mol g^{-1}$ under unprimed seeds to $3.19 \mu mol g^{-1}$ ($LoS < 0.01$ at the interaction level) when treated with 3 % SiO_2 , showing an increase of 81.50 % and 10.92 % when treated with 2.5 % KNO_3 .

In stress conditions, the rate of α -amylase production increased from 10.26 to $13.93 \mu mol$ maltose formed $min^{-1} mL^{-1}$, showing an increase of 23.94 %, when treated with 2.5 % KNO_3 followed by 3 % SiO_2 ($12.37 \mu mol$ maltose formed $min^{-1} mL^{-1}$) with 17.05 % increased activity. In moisture stress conditions, treating with 2.5 % KNO_3 exhibited higher SOD activity by 15.96 % ($520.03 units mg^{-1}$ of protein), whereas SiO_2 exhibited an increase of 15.25 % ($515.65 units mg^{-1}$ of protein) compared to unprimed seeds. Seed priming also elevated the levels of total soluble protein significantly ($LoS < 0.01$). Under stressed conditions seed priming with KNO_3 at 2.5 % increased total soluble protein by 25.71 %, followed by SiO_2 with 7.6 % over the unprimed stressed condition (Table 2).

Trehalose, total soluble sugars, capsaicin content and vitamin C content.

Moisture stress had a significance influence on trehalose content ($LoS < 0.05$). Under moisture stress condition trehalose content increased from 0.07 to $0.13 \mu mol g^{-1}$ FW (46.15 %), when treated seeds with 2.5 % KNO_3 , followed by $0.12 \mu mol g^{-1}$ FW (41.66 %), when treated with 3 % SiO_2 . Total soluble sugars varied from 0.15 to $3.16 mg g^{-1}$ FW under unprimed stressed and control conditions, under stressed conditions treating

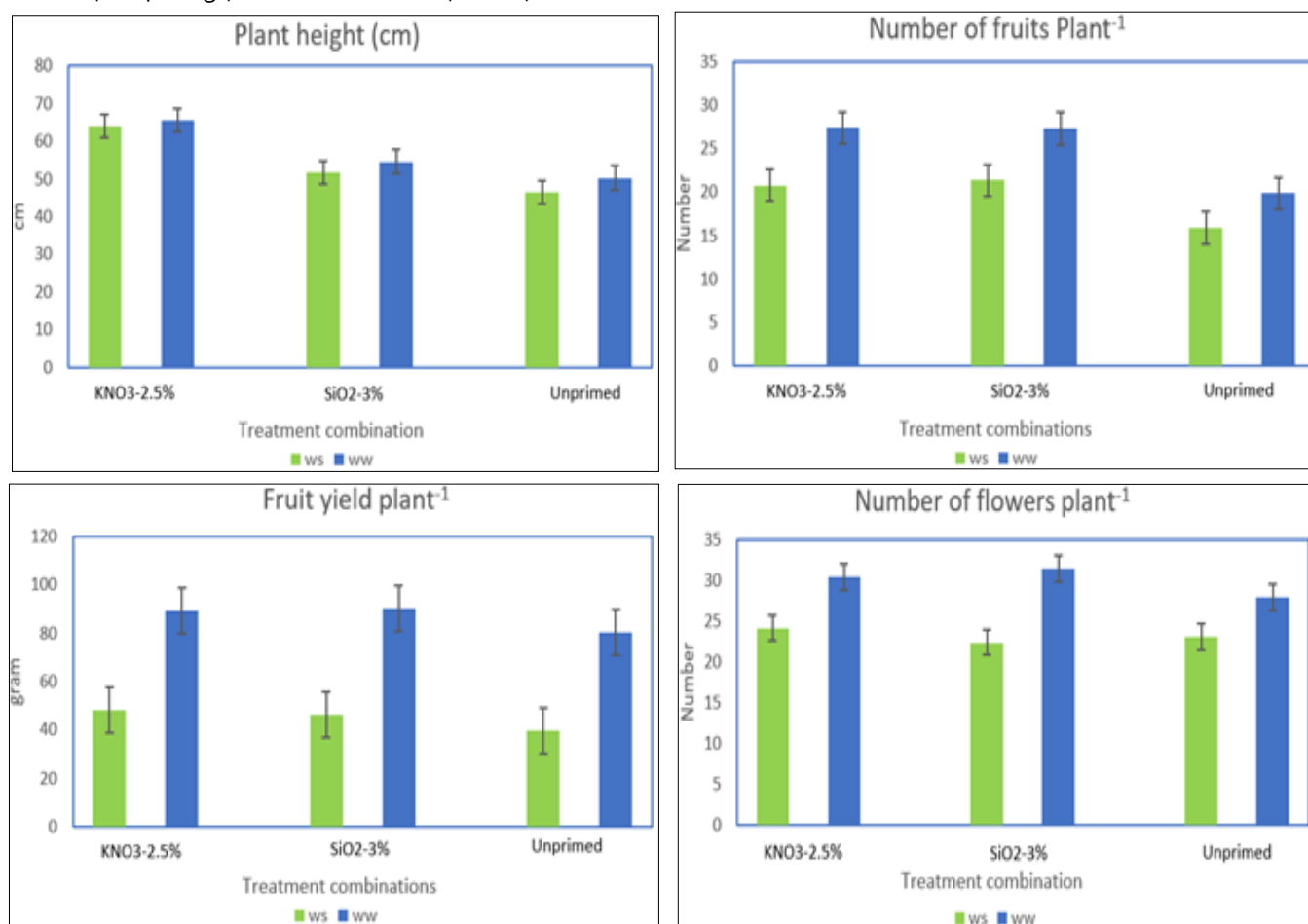


Fig. 3. Impact of seed priming agents viz., KNO_3 and SiO_2 on Plant height, number of fruits per plant, Fruit yield per plant and number of flowers per plant under moisture stress (WS) and well-watered (WW) conditions under pot study.

Table 1. Effect of seed priming agents on biochemical traits of chilli under well-watered and moisture-stressed field study

Stress levels	Priming agents	RWC (%)	CMSI (%)	MDA ($\mu\text{mole ml}^{-1}$)	H ₂ O ₂ ($\mu\text{mol g}^{-1}$ FW)	Proline ($\mu\text{mol g}^{-1}$)	TCC (mg g^{-1})	SOD (units mg^{-1} of protein)
Control	2.5 % KNO ₃	79.3 $\pm$ 6.14	76.66 $\pm$ 2.0	0.49 $\pm$ 0.01	0.08 $\pm$ 0.02	1.01 $\pm$ 0.08	3.86 $\pm$ 0.21	405.60 $\pm$ 3.24
	3 % SiO ₂	70.1 $\pm$ 2.56	91.16 $\pm$ 0.76	0.53 $\pm$ 0.04	0.53 $\pm$ 0.02	1.31 $\pm$ 0.01	3.6 $\pm$ 0.21	385.33 $\pm$ 3.91
	Unprimed	68.83 $\pm$ 3.01	76.0 $\pm$ 2.0	0.64 $\pm$ 0.04	0.56 $\pm$ 0.04	0.59 $\pm$ 0.01	3.45 $\pm$ 0.24	337.13 $\pm$ 2.62
	2.5 % KNO ₃	68.16 $\pm$ 2.02	46.66 $\pm$ 4.72	0.73 $\pm$ 0.03	0.71 $\pm$ 0.03	3.02 $\pm$ 0.08	1.93 $\pm$ 0.02	520.03 $\pm$ 12.21
Moisture stress	3 % SiO ₂	59.83 $\pm$ 3.01	48.33 $\pm$ 1.15	0.63 $\pm$ 0.05	0.59 $\pm$ 0.07	3.19 $\pm$ 0.15	1.94 $\pm$ 0.16	515.65 $\pm$ 12.44
	Unprimed	54.83 $\pm$ 3.88	38.33 $\pm$ 6.42	0.74 $\pm$ 0.03	0.97 $\pm$ 0.01	2.69 $\pm$ 0.14	1.57 $\pm$ 0.232	437.00 $\pm$ 8.19
Source of Variation								
Factor A (Moisture regimes)		***	***	***	***	***	***	***
Factor B (Seed priming agents)		**	**	**	***	***	*	***
A×B		*	*	*	***	**	**	**
CD _A		3.76	4.013	0.043	0.044	0.137	0.227	12.908
CD _B		4.612	4.937	0.053	0.054	0.168	0.279	15.809
CD _{AB}		NS	6.982	0.075	0.076	NS	NS	NS
SE(m)		2.07	2.216	0.024	0.024	0.075	0.125	7.095
CV (%)		5.367	6.106	6.551	5.999	6.008	7.896	2.603

***, ** and * indicate LoS at 0.001, 0.01 and 0.05 respectively. Different alphabets indicate the significant differences across the mean values calculated using the LSD method of means comparison test. RWC (relative water content), CMSI (Cell membrane stability index), MDA (Malondialdehyde), H₂O₂ (Hydrogen peroxide), SLA (Specific leaf area), TCC (total chlorophyll content) and SOD (superoxide dismutase activity).

Table 2. Effect of seed priming agents on quality traits of chilli under well-watered and moisture-stressed field conditions

Stress levels	Priming agents	Trehalose ($\mu\text{mol g}^{-1}$ FW)	α -amylase ($\mu\text{mol min}^{-1}$ mL ⁻¹)	TSS (mg g^{-1} FW)	TSP (mg g^{-1} protein)	Capsaicin ($\mu\text{g g}^{-1}$ FW)	Vitamin C ($\mu\text{g g}^{-1}$ FW)
Control	2.5 % KNO ₃	0.17 $\pm$ 0.006	14.77 $\pm$ 1.75	3.16 $\pm$ 0.12	2.0 $\pm$ 0.07	232.0 $\pm$ 2.73	4.75 $\pm$ 8.53
	3 % SiO ₂	0.13 $\pm$ 0.006	13.51 $\pm$ 1.17	1.99 $\pm$ 0.08	1.84 $\pm$ 0.08	154.7 $\pm$ 2.16	4.78 $\pm$ 8.06
	Unprimed	0.05 $\pm$ 0.005	10.36 $\pm$ 1.56	0.41 $\pm$ 0.04	1.64 $\pm$ 0.04	128.2 $\pm$ 1.66	3.96 $\pm$ 4.36
	2.5 % KNO ₃	0.13 $\pm$ 0.007	13.93 $\pm$ 1.7	1.55 $\pm$ 0.02	1.05 $\pm$ 0.16	115.9 $\pm$ 1.38	3.51 $\pm$ 9.36
Moisture stress	3 % SiO ₂	0.12 $\pm$ 0.007	12.37 $\pm$ 1.27	0.83 $\pm$ 0.05	0.97 $\pm$ 0.08	90.5d $\pm$ 1.73	3.78 $\pm$ 17.48
	Unprimed	0.07 $\pm$ 0.008	10.26 $\pm$ 0.61	0.15 $\pm$ 0	0.78 $\pm$ 0.09	65.5 $\pm$ 0.63	3.62 $\pm$ 1.00
Source of Variation							
Factor A (Moisture regimes)		*	**	***	***	***	***
Factor B (Seed priming agents)		***	*	***	*	***	***
A×B		***	*	***	**	*	***
CD _A		0.007	1.132	0.078	0.113	1.893	10.657
CD _B		0.009	1.386	0.095	0.139	1.923	13.052
CD _{AB}		0.013	NS	0.134	NS	3.279	18.45
SE(m)		0.004	0.702	0.043	0.062	1.041	5.858
CV (%)		5.971	9.102	5.47	7.791	13.743	2.492

***, ** and * indicate LoS at 0.001, 0.01 and 0.05 respectively. Different alphabets indicate the significant difference across the mean values calculated by using LSD method of means comparison test. TSS (total soluble sugar) and TSP (total soluble protein)

seeds with KNO₃ at 2.5 % increased total soluble sugars (1.55 mg g⁻¹ FW) by 90.3 %, followed by SiO₂ at 3 % (0.83 mg g⁻¹ FW) by 81.92 %. Capsaicin content increased by 43.48 %, under moisture stress when treated with KNO₃ at 2.5 % (115.9 $\mu\text{g g}^{-1}$ FW), whereas treating with SiO₂ at 2.5 % (90.5 $\mu\text{g g}^{-1}$ FW) exhibited increase in capsaicin content by 27.62 %. In case of vitamin C content, it varied from 3.51 to 4.78 $\mu\text{g g}^{-1}$. In water stress condition, treating of seeds with KNO₃ 2.5 % (3.51 $\mu\text{g g}^{-1}$) decreased the vitamin C by 3.0 %, whereas SiO₂ at 3 % (3.78 $\mu\text{g g}^{-1}$) increased capsaicin content by 4.2 % (Table 2).

Yield and yield attributing traits

Plant height, number of flowers per plant, number of fruits per plant and fruit yield per plant were significantly impaired under water stress ($LoS < 0.01$). Under moisture stress, plant height (42.3 cm) exhibited a reduction of 8.6 % under unprimed conditions, whereas priming with 2.5 % KNO₃ recorded 23.09 % higher plant height than unprimed, followed by SiO₂ at 3 % (49 cm) with 14.02 %. The data generated for the number of flowers per plant was found to be significant ($LoS < 0.001$) at all three levels. Under water stress conditions, a reduction of 14.86 % was noticed in the number of flowers per plant. KNO₃ at 2.5 % increased flowers per plant (22.8) by 18.59 %, whereas SiO₂ at 3 % reduced flowers (20.5) by 9.4 % (Table 3).

The number of fruits per plant varied from 16.26 under unprimed conditions to 22.43 fruits when treated with KNO₃ at 2.5% under water stress conditions. Seed priming had a beneficial effect in increasing the yield. A 14.55 % increase was noticed when treated with KNO₃ (19.03), followed by SiO₂ at 2.5 % (17.63 %) by 7.77 % compared with unprimed seeds. Similarly, fruit yield per plant varied from 34.86 g to 84.93g. A 54.8 % reduction in fruit yield was noticed from unprimed seeds exposed to moisture stress. Under moisture stress conditions, KNO₃ at 2.5 % (45.3g) recorded a 23.04 % increase in fruit yield, followed by SiO₂ at 3 % (44.26 g) by 21.23 % over unprimed seeds (Table 3).

Discussion

Aberrations constantly influence plant processes in the surrounding environment and under water stress, their status was significantly reduced (33). It has been observed that priming treatments with SiO₂ and KNO₃ enhance emergence, seedling development, yield and tolerance to salinity and drought in rice, maize, wheat and barley (34). Seed priming a straightforward pre-sowing technique, ensuring uniform establishment and compatible growth across various plant species. In the current study, treating of seeds of KNO₃ and SiO₂

Table 3. Effect of seed priming agents on yield traits of chilli under well-watered and moisture-stressed plants field study

Stress levels	Priming agents	Plant height (cm)	Number of flowers (number)	Number of fruits (number)	Fruit yield (gram plant ⁻¹)	SLA(cm ² g ⁻¹)
Control	2.5 % KNO ₃	60.0 ^d ±3.0	25.6 ^a ±1.75	22.43 ^d ±2.46	84.93 ^a ±4.3	18.47 ^c ±0.93
	3 % SiO ₂	58.3 ^a ±2.5	23.3 ^a ±1.30	20.33 ^a ±1.25	82.76 ^a ±2.18	16.62 ^c ±0.61
	Unprimed	46.3 ^f ±1.5	21.8 ^b ±1.93	20.43 ^f ±1.04	77.26 ^b ±1.84	14.79 ^d ±0.94
Moisture Stress	2.5 % KNO ₃	55 ^a ±4	22.8 ^c ±1.76	19.03 ^a ±2.34	45.3 ^c ±3.2	13.47 ^a ±0.73
	3 % SiO ₂	49 ^b ±2	20.5 ^d ±1.25	17.63 ^b ±2.08	44.26 ^d ±3.73	13.23 ^a ±0.45
	Unprimed	42.3 ^c ±2.5	18.56 ^{cd} ±1.0	16.26 ^c ±2.31	34.86 ^e ±4.15	10.96 ^b ±1.01
Source of Variation						
Factor A (Moisture Regimes)		*	*	***	***	***
Factor B (Seed Priming)		***	*	*	*	***
A×B		**	**	**	**	*
CD _A		3.029	0.715	1.391	3.366	0.912
CD _B		3.787	2.1	1.703	4.122	1.117
CD _{AB}		Ns	NS	NS	NS	NS
SE(m)		1.7	0.943	0.764	1.85	0.502
CV (%)		5.68	7.387	6.84	5.205	5.953

***, ** and * indicate LoS at 0.001, 0.01 and 0.05 respectively. Different alphabets indicate significant differences across the mean values calculated using the LSD method of means comparison test.

significantly elevated the levels of morpho-physiological, biochemical and yield traits of chilli plants exposed to moisture stress. In the current study, RWC was reduced by 20.10% under moisture stress conditions and seed treatment with KNO₃ at 2.5% alleviated the reduction in RWC levels. Treating seeds with 1% KNO₃ increased the relative water content (RWC) under water stress in mung beans. SiO₂ at 3 % increased levels of CMSI by 20.69 %, followed by 17.8 % with 2.5 % KNO₃ (35).

Seed priming dosage of 100 mM KNO₃ at 100 % FC produced the highest membrane stability index (36). At the same soil moisture level, the results were 7 % higher than the non-primed control, but statistically equal to the cantaloupe seed priming dosages of 25 and 50 mM KNO₃. In cucumbers, KNO₃ at 5 % with SiO₂ nanoparticles as a seed priming agent greatly increased the amounts of carotenoid and chlorophyll a, b and a+b. (37). A similar phenomenon was noticed in current study by increasing TCC (18.65 %) with KNO₃ and 19.07 % with SiO₂ under stress. MDA levels is indicative of oxidative stress-induced lipid peroxidation. Drought circumstances cause a considerable rise in malondialdehyde levels (MDA), a marker of lipid peroxidation (38). Treated spinach seeds with 1.8 moles g⁻¹ had lower MDA concentrations than untreated seeds when subjected to water stress (39). In maize, seed priming with SiO₂ reduced the levels of MDA, which were elevated to 122 % upon water stress (40). When soybean seeds are stressed by drought, osmotic potential variations can affect both H₂O₂ levels and CAT activity (41). Seedlings treated with KNO₃ (2.8 mole g⁻¹) showed reduced hydrogen peroxide levels than untreated seeds after undergoing various forms of water stress (39).

The roles of SiO₂ and KNO₃ as priming agents in elevating the levels of proline in rice and maize crops are emphasized (15, 42). There is an increase in proline from 2.7 to 3.5 moles proline g⁻¹ in chilli primed with SiO₂ exposed to water stress (43). A positive correlation has been observed between the accumulation of trehalose and increased resistance to drought stress (44). In addition to serving as a vital carbon source for the growth of early seedlings and preserving turgor pressure, which is necessary for the expansion of tissues during germination, sugars are crucial in controlling the metabolic processes of germination. The activity of α-amylase and trehalose were severely impaired by 32 and 23 %, respectively, under chilling stress in chickpea.

In contrast, Osmo priming significantly elevated the levels of α-amylase, trehalose and total soluble sugars by 83, 75 and 11.7 %, respectively (45). Similar outcomes were observed under the impact of SiO₂ and KNO₃ in water stress conditions in the current investigation. Greater germination properties of cantaloupe in response to KNO₃ seed priming may be linked to more significant α-amylase activity. Subsequently, increased starch breakdown (36), except for KNO₃, SiO₂ and SA priming, significantly increased the SOD activities of rice seedlings under all drought circumstances (15). The tolerance and severity of the stress have the most effects on how antioxidant enzymes in plants respond to water stress. With increasing water stress, SOD activity rises in the leaves and roots of the same species of tomato (46).

Water stress impaired the capsaicin content in chilli during the pod development stage (47). Chilli seeds primed with KNO₃ and SiO₂ increase the capsaicin content than unprimed seeds (48). Ascorbic acid uptake in chilli under water stress circumstances is significantly influenced by the duration of the stress, genetic variables and cultivar performance (3). In the current study, treating seeds with 2.5% KNO₃ increases plant height under water stress. Nitrates, which control growth and transfer photoassimilates to particular plant sections, enhance development and yield, as seen by the higher growth in plants grown from KNO₃ primed seeds (49). Water stress in *C. chinense* may cause flowering to occur later and result in a decline in flower yield (47). Fruit yield plant⁻¹ showed a significant difference under control conditions between seeds primed with 2.5 % KNO₃, 3 % SiO₂ and unprimed seeds, which were 89.3, 90.43 and 80, respectively. In tomatoes, exogenous application of SiO₂ at 401 kg ha⁻¹ maximized the crop yield under well-watered conditions (50). Fruit yield is impacted by water shortage during the vegetative stage and plants become stunted. Control plants produced substantially more fruit than drought stresses in chilli (47). Cantaloupe plants cultivated from 50 and 100 mM KNO₃-primed seeds had favourable responses in all growth, fruit yield, fruit quality, irrigation water productivity and physiological measurements (36).

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Authors' contributions

RB conceptualized the study. MPK performed the work. Data analysis was done by MPK, MChLN, RM and PPG. Technical guidance and support were given by MSN and RB. All authors reviewed and approved the manuscript.

Compliance with ethical standards

Conflict of Interest :The authors declare that they have no conflict of interest

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