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RESEARCH ARTICLE

Seed priming - induced drought tolerance in Castor: Unravelling the physiological and molecular mechanisms

Kanimozhi Elangovan[1], R. Vijayan[2*], V. Manonmani[1], T. Eevera[1], S.R. Venkatachalam[3] & M. Tilak[4]

[1]Department of Seed Science and Technology, Coimbatore - 641 003, Tamil Nadu, India

[2]Department of Forest Biology and Tree Improvement, Forest College and Research Institute, Mettupalayam -641 301, Tamil Nadu, India

[3]Tapioca and Castor Research Station, Yethapur, Salem - 641 003, Tamil Nadu, India

[4]Department of Agroforestry, Forest College and Research Institute, Mettupalayam, Coimbatore -641 301, Tamil Nadu, India

*Email: vijayanr78@gmail.com

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Abstract

Castor is an essential nonedible oilseed crop with significant applications in the cosmetic and chemical industries. It is cultivated primarily in rainfed regions, where drought is a common abiotic stress factor. Seed germination, the critical initial stage, is particularly affected by drought. Seed priming with radical scavenging chemicals such as salicylic acid, melatonin, hydrogen peroxide and ascorbic acid has been explored to increase germination under water deficit conditions. This study aimed to evaluate the effects of different concentrations of these chemicals on the physiological and antioxidant enzyme activities of castor under drought stress. Two water regimes were applied in the study. Among the treatments, seed priming with 80 ppm salicylic acid significantly improved key parameters, including the germination % (71 %), speed of germination (4.1 %), vigour index (3141), chlorophyll content (28.3 SPAD units) and relative water content (63 %). It also increased the antioxidant enzyme activities such as CAT, POD, SOD and APX, along with the overexpression of the drought-responsive gene *RCECP63* under water deficit conditions. These findings highlight that seed priming, particularly with salicylic acid, offers a promising strategy to enhance castor resilience under prolonged drought stress, providing a practical approach for improving crop performance in drought-prone regions.

Keywords

castor; rainfed; water stress; seed priming; *RcECP63*

Introduction

Castor (*Ricinus communis)* is considered one of the most important oilseed crops in the Euphorbiaceae family. Seeds contain 45-52 % oil, which is nonedible. The oil has only one distinct fatty acid, ricinoleic acid (85-95 %) as well as stearic and palmitic acid (1 %), oleic acid (2-6 %), linoleic acid (1-5 %) and dihydroxystearic acid (1 %) (1). It has a variety of uses in the chemical industry, including lubricants for high-speed engines and aero planes, soaps, transparent paper, printing-ink, varnish, linoleum and plasticizers (2). Owing to its diverse applications, its global demand continues to grow by 3-5 % each year. However, it is mostly grown in rainfed areas where abiotic stress is a major issue that reduces the productivity of crops. A recent study found that variability in rainfall has increased, leading to longer dry spells and more intense droughts in rainfed areas (3). This has directly impacted the availability of water for crops, reducing yields and threatening the livelihoods of farmers who depend on rainfed agriculture. Among abiotic stresses, drought is a critical stress that significantly affects crop yield globally.

Drought stress induces the overproduction of reactive oxygen species such as superoxide anion radicals, singlet oxygen, hydrogen peroxide and hydroxyl radicals, causing oxidative damage (4). Under severe stress conditions, the antioxidant capacity of plants might not be adequate to reduce the damaging effects of oxidative stress. Thus, synthesizing signalling molecules in plants becomes crucial for understanding how they respond to adverse environments. Also, previous studies have demonstrated that application of external stress signalling molecules, such as salicylic acid (SA), hydrogen peroxide, melatonin and ascorbic acid (ASA) is highly promising for increasing stress tolerance in most of the plants(5-7). For instance, Melatonin has been recognized as a promising molecule in recent research for enhancing drought tolerance in plants. It has shown significant potential in improving stress resilience in oilseed crops such as sunflower, safflower, soybean and rapeseed as well as other crops like cotton (8-12). Similarly, it was indicated that pretreatment soybean (*Glycine max* L.) plants with low doses of H_2O_2 helped reduce water loss and H_2O_2 levels, while enhancing drought tolerance by activating the plant's antioxidant defense system (13). In another study, exogenous application of H_2O_2 at low concentrations (1 mM) enhanced drought tolerance in soybean by improving traits like chlorophyll content, stomatal conductance, water content and shoot length (14). Similar results were found in application of salicylic acid under drought stress in soybean and castor (15, 16). Additionally, ascorbic acid (ASA) application was reported that increase proline accumulation and reduce lipid peroxidation thereby enhancing the osmotic adjustment mechanisms under abiotic stress in several plant species (17). From the above context, by considering these radical scavenging chemicals role in response to drought, we identified a research gap in their application for enhancing drought tolerance in castor.

However, there are different methods used for the application of these chemicals in plants, such as seed treatment, foliar application and application through soil. Among these methods, seed priming is a potential and cost-effective technique for improving plant drought tolerance, especially in the early stages of plant growth (18). Previous studies have demonstrated that seed priming can improve plant resilience to abiotic stresses, including drought. For example, seed priming with various chemicals has been shown to enhance seed germination, seedling vigor and stress tolerance in crops like *Jatropha curcas*, soybean and sunflower (19-21). In castor, limited research has been conducted on seed priming, but studies on similar oilseed crops suggest that priming can improve drought tolerance by promoting antioxidant activity, water retention and overall plant vigor under stress conditions. These findings provide a foundation for exploring the potential of seed priming in castor cultivation. Therefore, on the basis of the role of these radical-scavenging chemicals and seed priming treatments, we hypothesized that these chemicals can make plants tolerant to drought during the early growth stage such as germination stage. Thus, this study aimed to assess the impact of various seed priming treatments with radical-scavenging chemicals under low moisture conditions, in order to identify the most effective treatment for enhancing drought tolerance.

Materials and Methods

Seeds of the Castor hybrid YRCH 1 used for this study were obtained from Tapioca and Castor Research Institute, Tamil Nadu Agricultural University, Salem, Tamil Nadu, India.

Castor seeds were subjected to different seed priming treatments at 30 °C room temperature for 16 h, such as hydropriming (T_2), salicylic acid at 40 ppm (T_3), salicylic acid at 80 ppm (T_4), melatonin at 0.1 mM (T_5), melatonin at 0.5 mM (T_6), H_2O_2 at 1 mM (T_7), H_2O_2 at 10 mM (T_8), ascorbic acid at 200 mM (T_9) and ascorbic acid at 100 mM (T_{10}), along with the control (T_1). After the priming treatment, the soaked seeds were placed on blotting paper to absorb surface moisture and dried until they reached their original moisture. After drying, the seeds were subjected to a germination test.

The best performing treatments under laboratory conditions were performed under field conditions. A split-plot design was used to sow primed seeds alongside a control group under 2 different irrigation regimes in a alkaline soil (pH 8.0). One is irrigation after sowing, life irrigation and 5 days interval (W_1). Another one is irrigation after sowing, without life irrigation, with a 10 days interval (W_2).

Physiological analysis

Physiological parameters, viz., germination %, speed of germination, root length, shoot length, dry matter production and vigour indices I and II, were evaluated according to the ISTA seed testing procedure (22). The relative water content of the leaves was measured (23).

Biochemical analysis

To assess the activity of antioxidants, e.g., catalase, peroxidase, superoxide dismutase activity, ascorbate peroxide, chlorophyll index and relative water content, leaf samples were collected and estimated. Catalase (24), Peroxidase (25) and SOD (26) activity was analysed. A chlorophyll meter from Minolta (model 502 of Minolta Japan) was utilized to measure the chlorophyll index of the castor leaves.

RT-PCR analysis

RT-PCR analysis was performed for best performed one treatment and control under 2 irrigation regimes to know expression of drought responsive gene *RcECP63* (27). This gene was selected for analysis because it plays a critical role in drought tolerance mechanisms. Specifically, *RcECP63* is associated with the regulation of proline metabolism, hydroxyl radical scavenging and improved antioxidant defense, which are key processes in mitigating oxidative stress and cellular damage during water deficit conditions.

Statistical analysis

The significance of the data from various studies was assessed using the F-test (28). The critical difference was calculated at a 5 % level of probability. Values with different letters indicate significant differences ($p < 0.05$) between groups, while groups with the same letter are not significantly different. Graphs were generated using Microsoft Excel (2010) and GraphPad Prism version 5.8.

Results and Discussion

Standardization of promising seed priming treatments under laboratory conditions

The results of this study revealed that, compared with nonprimed seeds (control), primed seeds presented increased physiological and biochemical parameters under laboratory conditions. With respect to the speed of germination, 40 ppm (6.2) and 80 ppm (7) salicylic acid, 0.1 mM (6.4) and 0.5 mM (6.7) melatonin and 1 mM (6.4) and 10 mM (6.5) H_2O_2 had similar results (Table 1).

The present study revealed a significant increase at P-value<0.05 in the % of germination when the seeds were treated with salicylic acid (80 ppm) and hydrogen peroxide (H_2O_2-10 mM), with a 21 % improvement compared with the control. The germination rate increased from 49 % in the control to 70 % in both treatment groups. A similar increase in root length was noted, with seeds primed with salicylic acid (80 ppm), melatonin (0.5 mM) and H_2O_2 (10 mM) showing increased root lengths of 13.2 cm, 13.5 cm and 13.8 cm respectively, compared with the 9.7 cm in the control group. Shoot length also improved across the treatments, with lengths comparable to those of the control (25.6 cm) but slightly greater than those of the T_4 (29.2 cm), T_6 (28.6 cm), T_7 (28.2 cm) and T_8 (29.5 cm) treatments (Table 1).

These findings align with earlier research on castor and soybean, where similar improvements in germination traits were observed (29). The beneficial effects of salicylic acid (SA) on seed germination may be attributed to its role in reducing abscisic acid (ABA) levels, which in turn prevents the decline of essential growth hormones such as cytokinin and indole-3-acetic acid (IAA) under water stress, as reported in wheat (30). This hormonal regulation likely plays a significant role in enhancing germination and early seedling growth, as observed in the present study.

The improvements in root and shoot growth could be due to the stimulatory effects of SA, melatonin and H_2O_2 on key physiological processes, which help plants cope better with stress conditions. Overall, these treatments appear to promote better seedling establishment and growth, potentially offering a useful strategy for improving crop resilience in stressful environments.

Compared with those in the control (1.3 mg), dry matter production in seeds primed with hydrogen peroxide (H_2O_2, 10 mM) or salicylic acid (SA, 80 ppm) was notably greater, with values reaching 2.3 mg and 2.2 mg respectively (Table 1). Additionally, the vigour index significantly increased by 65 % in seeds treated with salicylic acid (80 ppm), reaching 2955.8, followed closely by H_2O_2 (10 mM) at 2926.6, compared with 1789.5 in the control group (Fig. 1). Similar findings have been reported in maize exposed to chilling stress, where salicylic acid treatments resulted in improved plant performance (31). The exogenous application of salicylic acid in barley plants under drought stress was also found to increase net CO_2 assimilation and stomatal conductance, leading to increased dry matter production (32). These findings suggest that SA enhances the physiological processes critical for plant growth, especially under stress conditions.

Table 1. Determination of the best seed priming treatments under laboratory conditions.

Treatments	Speed of germination	Germination %	Root length (cm)	Shoot length (cm)	Dry matter production (mg/10 seedlings)	Vigour index I	Vigour index II
T_1-Control	4.8^{d}	49^{g}	9.7^{f}	25.6^{e}	1.3^{g}	64^{f}	1789^{f}
T_2-Hydro priming	6.1b^{c}	57^{f}	10.2^{ef}	27.4^{cd}	1.5^{f}	85de	2164^{e}
T_3-Salicylic acid 40 ppm	6.2^{abc}	65^{cde}	11.0^{def}	28.4^{abc}	1.8^{d}	114^{c}	2563^{bcd}
T_4-Salicylic acid 80 ppm	7.0^{a}	70^{ab}	13.2^{ab}	29.2^{ab}	2.2^{ab}	150^{a}	2955^{a}
T_5-Melatonin 0.1 mM	6.4^{abc}	66^{bc}	12.4^{bcd}	27.8^{bcd}	1.8^{de}	120^{c}	2694^{abc}
T_6-Melatonin 0.5 mM	6.7^{ab}	69^{ab}	13.5^{ab}	28.6^{abc}	2.0^{bc}	138^{b}	2834^{ab}
T_7-H_20_2 1 mM	6.4^{abc}	65^{cd}	12.6^{abc}	28.2^{abcd}	1.8^{d}	116^{c}	2660^{abcd}
T_8-H_2O_2 10 mM	6.5^{abc}	70^{a}	13.8^{a}	29.5^{a}	2.3^{a}	159^{a}	2926^{a}
T_9-Ascorbic acid 50 ppm	5.8^{c}	61^{e}	11.2^{cde}	26.8^{de}	1.6^{ef}	97^{d}	2346de
T_{10}-Ascorbic acid 100 ppm	5.9^{c}	63de	11.5^{cde}	27.9^{bcd}	1.9^{cd}	118^{c}	2383^{cde}
Mean	**6.18**	**64**	**11.91**	**27.94**	**1.82**	**116**	**2531**
SEd	**0.396**	**3.023**	**0.677**	**0.739**	**0.085**	**4.712**	**163.630**
CD (P=0.5)	**0.809**	**6.350**	**1.389**	**1.554**	**0.174**	**9.668**	**343.791**

(Values with different letters indicate significant differences ($p < 0.05$) between groups. Groups sharing the same letter are not significantly different)

Fig.1. Effect of different seed priming treatments under laboratory conditions.

In the present study, the increase in dry matter production and vigour index can likely be attributed to the role of SA and H_2O_2 in mitigating stress effects, improving seedling vigour and promoting metabolic activities that increase plant growth. The increased dry mass reflects better resource allocation and utilization, likely due to improved photosynthesis and water-use efficiency, as seen in previous studies (33). These treatments, therefore, hold potential for improving crop performance under stress conditions, offering a valuable tool for sustainable agricultural practices.

Effects of seed priming on physiological parameters under well-watered and water stress conditions

Compared with unprimed seeds, seeds primed with 80 ppm salicylic acid presented the highest % of germination under both well-watered (W_1) (75 %) and low-moisture (W_2) conditions (71 %). Similarly, primed seeds took less time to germinate under both irrigation regimes, as indicated by the increased speed of germination. Seed priming with salicylic acid at 80 ppm was more responsive than priming with melatonin or hydrogen peroxide. The vigour index was highest in seeds primed with 80 ppm salicylic acid, followed by those primed with melatonin and hydrogen peroxide, compared with the control (un primed seeds) (Table 2). These results were consistent with previous studies on cantaloupe seeds primed with salicylic acid (34). In the present study, an improvement in shoot length was observed, which is agree with previous studies on the effects of salicylic acid *on Phaseolus vulgaris* (35).

In addition to SA, melatonin improved germination traits such as germination %, the speed of germination and the vigour index. Similar findings were recorded in waxy maize following the application of melatonin via seed priming (36). In rapeseed, seed priming via melatonin maintains the photosynthetic rate, stomatal conductance and transpiration. Additionally, the accumulation of reactive oxygen species (ROS) can be prevented by increasing the production of different antioxidants for scavenging (37). Similarly, the exogenous application of melatonin increased root growth by increasing the endogenous levels of free IAA in the roots of young *Brassica juncea* seedlings (38).

Effect of the seed priming on relative water content (RWC) of shoots under well watered and water stress conditions

The RWC in shoots was found to be in the range of 50–70 % in the plants raised from both primed and unprimed seeds. As a result of low moisture (W_2), the RWC of primed and unprimed plants decreased. The decrease in RWC was greater in unprimed seeds than in primed seeds under W_2. The RWC of the seedlings primed with salicylic acid (60 %) was the highest, followed by those of the melatonin (57 %) and hydrogen peroxide (54 %) treated seedlings. The leaf relative water content was significantly different under well-watered and water deficit conditions in seeds primed with SA compared with the control (Table 2). Similar results were obtained in maize seeds. The accumulation of osmolytes may increase, which decreases the osmotic potential, leading to the maintenance of the RWC or SA priming may regulate the behavior of stomata, which leads to the maintenance of the water status of plants under water deficit conditions (39).

Effect of seed priming on various antioxidant enzyme activities under well-watered and water stress conditions

Catalase activity was greater in the SA treatment (1.85 µg of reduced H_2O_2 g^{-1} min^{-1}) against unprimed seeds (1.78 µg of reduced H_2O_2 g^{-1} min^{-1}) under W_1. Similarly, in W_2, the catalase activity of seeds primed with salicylic acid (2.89 µg of reduced H_2O_2 g^{-1} min^{-1}) was greater than that of unprimed seeds (2.31 µg of reduced H_2O_2 g^{-1} min^{-1}).

There was a slight difference in peroxidase activity between primed (0.37 unit mg^{-1} protein min^{-1}) and unprimed seeds (0.31 unit mg^{-1} protein min^{-1}) under W_1. Compared with un primed seeds (0.46 unit mg-1 protein min^{-1}), seeds primed with salicylic acid (0.76 unit mg^{-1} protein min^{-1}) presented greater peroxidase enzyme activity, followed by those primed with melatonin (0.70 unit mg-1 protein min^{-1}) and H_2O_2 (0.61 unit mg^{-1} protein min^{-1}).

Table 2. Effects of different seed priming treatments on the germination %, speed of germination, vigour index I and relative water content under two different irrigation regimes.

	Germination %					**Speed of germination**					**Vigour index I**					**Relative water content (%)**				
	T_1	**T_2**	**T_3**	**T_4**	**Mean**	**T_1**	**T_2**	**T_3**	**T_4**	**Mean**	**T_1**	**T_2**	**T_3**	**T_4**	**Mean**	**T_1**	**T_2**	**T_3**	**T_4**	**Mean**
W_1	58	75	65	61	259	3.6	4.7	4.2	3.9	16.4	2221	3654	2618	2449	2221	66	70	67	66	269
W_2	48	71	61	58	238	2.7	4.1	3.8	3.4	14	1634	3141	2694	2286	1634	51	63	57	54	225
Mean	106	146	126	119		6.3	8.8	8	7.3		3855	6795	5312	4735		117	133	124	120	
	W	**T**	**W at T**	**T at W**		**W**	**T**	**W at T**	**T at W**		**T**	**W**	**W at T**	**T at W**	**T**	**T**	**W**	**W at T**	**T at W**	
SEd	**0.56**	**0.93**	**1.27**	**1.31**		**0.066**	**0.083**	**0.122**	**0.118**		**7.32**	**38.93**	**48.23**	**55.05**	**7.32**	**0.33**	**0.47**	**0.67**	**0.67**	
CD ($P<0.05$)	**1.79**	**1.95**	**2.94**	**2.77**		**0.212**	**0.176**	**0.297**	**0.249**		**23.28**	**81.78**	**102.53**	**115.65**	**23.28**	**1.05**	**1.01**	**1.59**	**1.42**	

Under the first irrigation schedule (W_1), there was no that much significant difference in SOD activity. However, under water stress condition (W_2), the SOD activity significantly differed from that of the treatment and control (7.46 units g^{-1} protein min^{-1}). Among the treatments, seeds primed with salicylic acid presented greater SOD activity (9.36 units g^{-1} protein min^{-1}). This is followed by melatonin and hydrogen peroxide.

In addition, ascorbate peroxidase (APX) activity did not significantly differ between primed seeds and unprimed seeds. However, SA at 80 ppm (11.5 units g^{-1} protein min^{-1}) resulted in greater activity than unprimed seeds (10.2 units g^{-1} protein min^{-1}). Similar results were found in W_2, where SA at 80 ppm (17.8 units g^{-1} protein min^{-1}) resulted in greater APX activity followed by melatonin (16.6 units g^{-1} protein min^{-1}) (Fig. 2). This increase in the levels of antioxidants such as CAT, POD, SOD and APX under water deficit stress caused by the application of SA was observed in previous studies on *Brassica napus* and *Pisum sativum* (40, 41).

Under drought conditions, reactive oxygen species (ROS) levels tend to increase, leading to oxidative damage within plants. Salicylic acid (SA) plays a crucial role in managing ROS production in mitochondria by activating the antioxidant defense mechanisms of plants. This helps reduce oxidative stress and prevents cellular damage. Water stress often triggers excessive ROS accumulation, which can harm cellular structures and diminish photosynthetic efficiency, ultimately affecting plant growth.

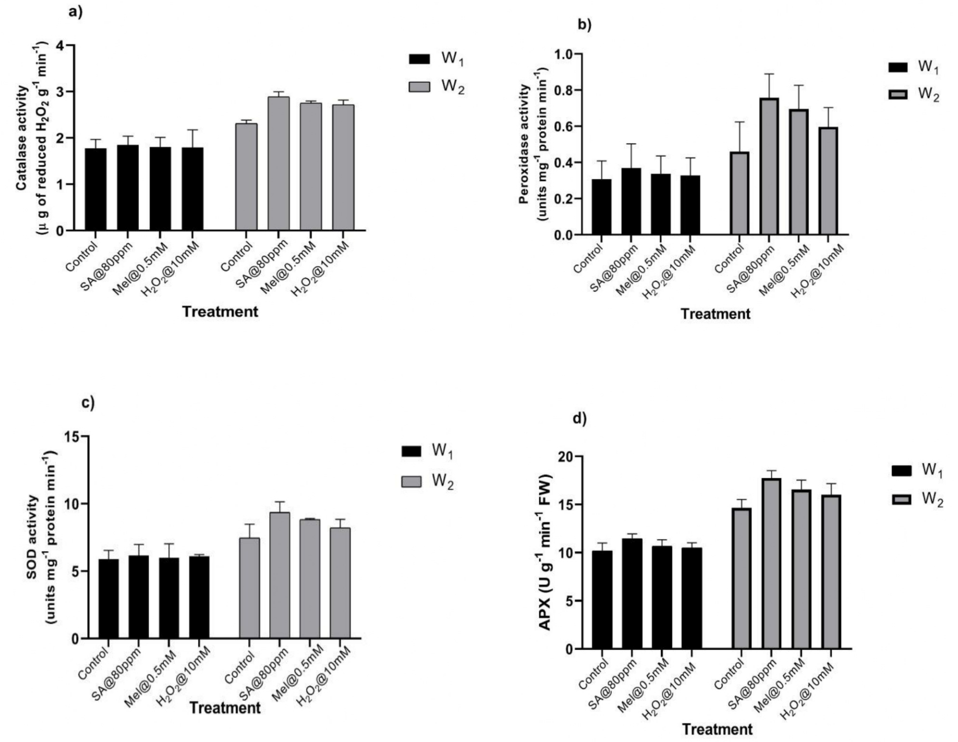

Fig. 2. Effect of different seed priming treatments and water schedules on biochemical traits under field condition a) Catalase activity b) Peroxide activity c) SOD activity d) Ascorbate peroxide (APX) activity.

In the present study, the increased enzyme activities observed in plants treated with SA likely increased their ability to detoxify ROS, thereby mitigating oxidative damage. By reducing oxidative stress, these primed plants are better equipped to maintain cellular integrity, support metabolic functions and sustain improved growth and development under water-limited conditions.

This regulation of ROS by SA highlights its protective role in drought stress, allowing plants to maintain photosynthesis and other key physiological processes. The activation of antioxidant systems enables plants to counteract the negative effects of ROS accumulation, supporting resilience and improving overall plant performance. These findings suggest that the use of SA, along with other priming agents, can be a valuable strategy for enhancing plant tolerance to environmental stress, particularly drought.

Effect of seed priming on chlorophyll content under well-watered and water stress conditions

The chlorophyll content was greater in seeds primed with 80 ppm salicylic acid (22.5) than in unprimed seeds (18) under W_1. In W_2, similar results were observed. Compared with the control (17.45), seed priming with 80 ppm salicylic acid resulted in significantly increased chlorophyll content (20.55), followed by melatonin (19.45) and hydrogen peroxide (19.1). With respect to the water schedule, irrigation schedule one (W_1) had the greatest effect on the chlorophyll content in all the treatments compared with water stress condition (W_2).

Under irrigation schedule 1 (W_1), the chlorophyll content significantly differed between primed and unprimed seeds. However, seed priming with SA at 80 ppm (25.2) resulted in a greater chlorophyll content than the control (20.95). Similarly, in W_1 the chlorophyll content of seeds primed with SA at 80 ppm (20.55) was greater than that of unprimed seeds (17.475) (Fig. 3).

Seed priming with salicylic acid improved the chlorophyll index even under drought stress. Similar results were reported in tomato and soybean following the application of salicylic acid through seed priming (42, 43). Similarly, it was reported that foliar application of SA increased the chlorophyll content in tomato plants under water deficit conditions. This might be due to an improvement in the photosynthetic rate, resulting in an increase in plant growth (44). According to a report, plants with relatively high chlorophyll contents tend to have relatively high tolerance to drought (45).

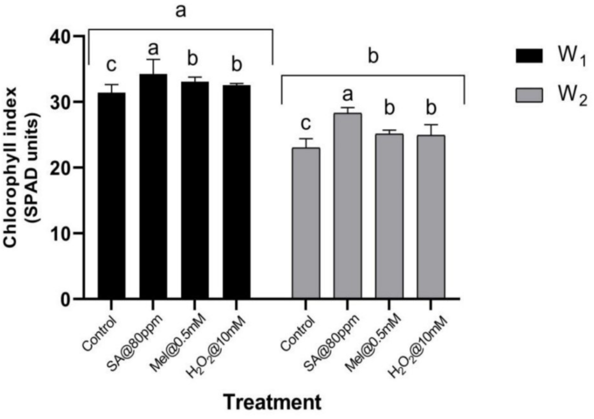

Fig. 3. Effect of different seed priming treatments and water schedules on chlorophyll index under field condition.

Effect of seed priming on *RcECP63* gene expression under well-watered and water stress conditions

The *RcECP63* gene expression was significantly upregulated under drought conditions in seeds primed with SA (Salicylic Acid) at 80 ppm, compared to the control (unprimed seeds). Gene expression was notably higher in the W_2 water schedule compared to the W_1 schedule. In both water schedules, seeds primed with SA at 80 ppm exhibited higher expression of the gene compared the control (Fig. 4). Likewise, salicylic acid treated *Mitragyna speciosa* plants showed that variety of genes were upregulated, many of which are involved in stress responses and signalling pathways (46). In wheat, application of SA enhanced the gene expression such as GST1, GST2 and GR, which play a key roles in improving stress tolerance during drought stress (47). When mitochondrial function is altered by drought, SA influences mitochondrial retrograde signalling, in which signals are sent to the nucleus that trigger the expression of specific genes related to drought tolerance (48).

This drought-responsive gene, *RcECP63*, was overexpressed under drought conditions in seeds primed with SA at 80 ppm. The *RcECP63* gene is linked to proline accumulation, hydroxyl radical scavenging capacity and total antioxidant activity in castor under drought stress (49). In the present study, the observed overexpression of this gene suggested that SA treatment may increase the ability of plants to mitigate the effects of drought by promoting these protective mechanisms.

RcECP63 plays a crucial role in enhancing proline metabolism, which helps to maintain osmotic balance and protect cellular structures during water-deficit conditions. The upregulation of this gene also enhance the antioxidant defense system by increasing the activity of enzymes such as catalase (CAT), peroxidase (POD) and superoxide dismutase (SOD) which helps to neutralize reactive oxygen species (ROS) generated during drought stress.

These physiological changes such as improved antioxidant enzyme activity and reduced oxidative damage were evident in the primed plants, correlating with higher *RcECP63* expression levels. By mitigating oxidative stress and enhancing the plant's ability to maintain cellular homeostasis, *RcECP63* likely contributes to the observed improvements in seedling establishment and overall drought tolerance in castor. This gene's role in coordinating both proline accumulation and antioxidant activity provides a molecular basis for the enhanced physiological responses seen in the study.

The findings of this study suggest that seed priming, particularly with salicylic acid, has significant potential to enhance drought tolerance in castor plants during critical early growth stages. This has important practical

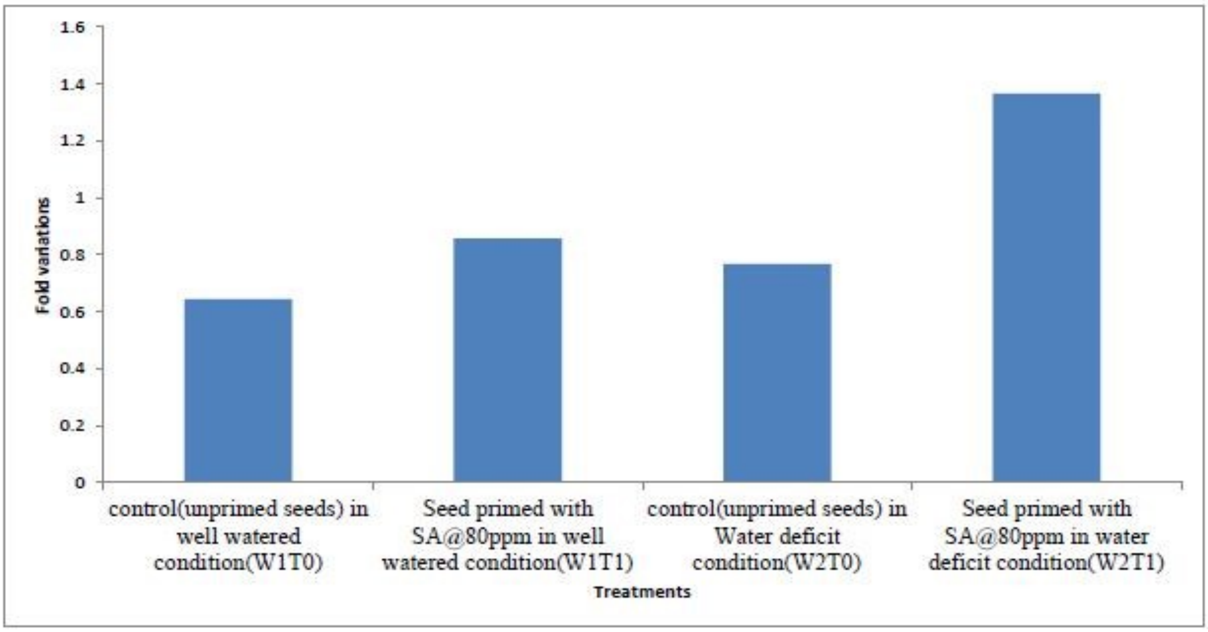

Fig. 4. Effect of best performing seed priming treatment and control (unprimed seeds) under well-watered and water deficit conditions on drought responsive RcECP 63 gene expression.

implications for castor cultivation, especially in regions prone to water scarcity or irregular rainfall. Implementing seed priming on a large scale is feasible due to its relatively low cost and ease of application. The process requires only small quantities of the priming agents and can be performed using simple soaking techniques that are already familiar to many farmers (Fig. 5).

Moreover, since the priming treatment occurs before sowing, it does not require additional labor or field equipment during the growing season, making it an attractive option for farmers. However, scaling up this technique would require further validation across different environmental conditions and castor varieties to ensure consistent benefits. Additionally, demonstrating its long-term effects on crop yield and quality up to the harvest stage would be essential for promoting widespread adoption.

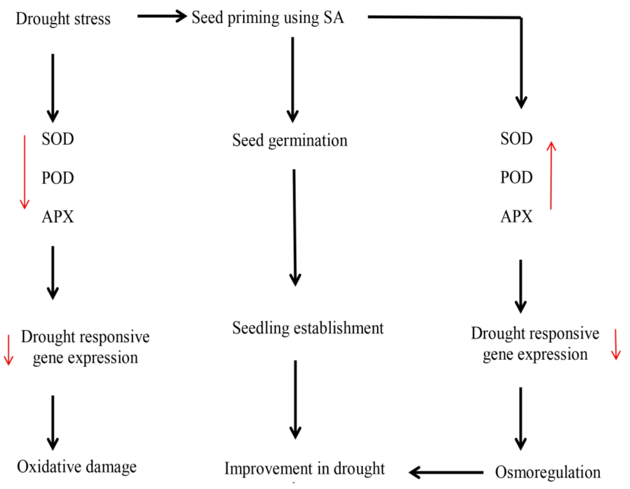

Fig 5. Overall effect of drought and seed priming using SA on seed germination. Upwards arrow indicates the positive correlation and downwards arrow indicates the negative correlation.

Conclusion

This study highlights the effectiveness of seed priming as a strategy to increase water stress tolerance in castor plants. Priming with salicylic acid (SA) significantly improved key growth parameters, including the germination, root and shoot length, dry matter production and seedling vigour. In particular, SA is instrumental in regulating reactive oxygen species (ROS) levels and activating the plant's antioxidant defense system, helping to reduce oxidative stress and protect cellular integrity under drought conditions.

These findings demonstrate that seed priming not only mitigates the adverse effects of water stress but also promotes better growth and development by enhancing stress resistance mechanisms. The use of SA as a priming agent offers a promising approach to improve castor crop performance, contributing to more sustainable and resilient agricultural practices that are better equipped to handle the growing threat of climate change-induced drought. However, further research is needed to determine how long the salicylic acid-induced expression of the *RcECP63* gene lasts.

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Authors' contributions

KE carried out the entire research work and performed statistical analysis, VR participated in the design of the study and manuscript editing, MV helped to facilitate the instruments for the study, ET helped in PCR analysis and manuscript writing, VSR participated in design and coordination of the study, TM helped in manuscript writing. All authors read and approved the final manuscript.

Compliance with ethical standards

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Declaration of generative AI and AI-assisted technologies in the writing process

During the preparation of this work the author(s) had not used any AI tools.

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