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Effect of Light, Temperature, Salinity, and Halopriming on Seed Germination and Seedling Growth of *Hibiscus sabdariffa* under Salinity Stress

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Abstract: Salt stress is a serious and current global problem for crops. Due to climate change, the soil today has higher salinity levels than in past decades. Identifying temperature, light, and salinity that allow plants to germinate and grow is an ambitious challenge for the future. *Hibiscus sabdariffa* (*H. sabdariffa*) is a plant that undergoes abiotic stress during all stages of growth. The aim of this work was to identify the best conditions in terms of light, temperature, and salinity during the germination and growth phases of *H. sabdariffa*. To improve the germination of *H. sabdariffa* seed, the effects of abiotic stress were investigated in three experiments. In the first experiment, the factors included light at two levels (light and dark cycles) and temperature at eight levels (5, 10, 15, 20, 25, 30, 35, and 40 °C). In the second experiment, the effect of salinity was examined at seven levels (0, 30, 60, 90, 120, 150, and 180 mM NaCl). In the third experiment, the factors consisted of seed halopriming at two levels (0 and 180 mM NaCl for 24 h) and salinity at seven levels (0, 30, 60, 90, 120, 150, and 180 mM NaCl). The highest germination rate (GR), seedling dry weight, and uniformity of germination were obtained at 30 °C in dark conditions, as reported by one-way Anova analysis. Germination was restricted by temperatures lower and higher than 5 and 30 °C, respectively. By increasing the salinity, all the germination characteristics were decreased, but these effects were less pronounced by halopriming. The most suitable planting date was in the spring, when the temperature was in the range of 25–35 °C. During the germination stage, Hibiscus tea is sensitive to low salinity soils. Halopriming can be performed for enhancing GR and emergence percentage.

Keywords: germination; temperature; salt tolerance threshold; light; regression model

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1. Introduction

As a member of the Malvaceae family, Hibiscus tea (*Hibiscus sabdariffa*) originates from Africa (Angola) [1]. It is currently cultivated on a large scale in North Africa, Mexico, India, Thailand, and China [1]. In Iran, the sweet and sour petals of Hibiscus tea flower without caffeine are used to treat loss of appetite, colds, flu, and circulatory system problems, as well as allergic eczema and other skin irritations [2]. Due to its increasing use for the mentioned medicinal values in the country, we need more information about its germination ecology and production. Seed germination and seedling establishment are the critical stages of any plant in harsh habitats [3–8]. Indeed, the desert can be a difficult environment for germination. However, many plants have developed evolved strategies involving wind, rain, or animals for seed dispersal and germination [3,6]. Furthermore, the germination process can be associated with the size of the seed, the depth of sowing, and the possible treatment of the outermost layer of the seed, as observed for wheat [4]. Furthermore, the germination of maize and other crops used for food can be favored by inter-cultivation with other plant

species [5,7]. This is a strategy that improves plants' response to salt stress [8]. Therefore, successful germination is a crucial step in seedling development and establishment [9]. A seed starts to germinate in favorable conditions in response to environmental stimuli such as light, temperature, and soil components (especially salinity) [6,10]. Several environmental factors act as germination determinants, among which temperature and salinity are the governing factors for maximum germination and germination speed [11]. Appropriate temperature and light are two environmental factors of importance in the regulation of seed germination [12].Temperature significantly affects germination ability and rate [13,14]. Temperature affects the percentage and speed of germination via water imbibition and the biochemical reactions in the germination process [15]. The amount and speed of water absorption by seeds in the germination stage is affected by temperature [16]. An optimum temperature is required by seeds and organelles within plant cells to uptake sufficient water and have normal activities [17–24]. Water absorption by seeds increases at a certain temperature and may decrease at lower or higher temperatures [25]. Thus, the temperature is the major factor in determining the success or failure of plant distribution and seedling establishment [26]. The minimum, optimum, and maximum temperatures required for germination generally depend on species' adaptation to environmental conditions [27]. These temperatures are known as cardinal temperatures, within the range of which seeds of varied species can germinate differently [28]. The germination percentage usually increases linearly with temperature up to an optimal temperature, after which the germination percentage decreases sharply [29,30]. Germination rate (GR) and germination percentage (GP) are the two previously described determinants of the growth responses of some species to temperatures. Regression analysis is a useful statistical tool for investigating the relationship between these variables. Linear regression is mainly utilized to describe the relationship between the temperature and the GR [31,32]. Several authors have evaluated the germination response of black cumin (*Bunium persicum*) to temperature, and they reported that its GR was enhanced with increasing temperatures [17,32]. Light is another important factor governing germination speed [6,33]. Bhatt and colleagues stated that *Deverra triradiata* seeds had a sensitivity to light during germination, thus indicating the importance of sowing depth determination [26]. Another essential environmental factor in the germination stage is salinity in dryland areas. Due to the expansion of saline lands in arid and semi-arid areas, exploitation of salt-tolerant crop species and varieties would be a good strategy for using saline soils [34]. In saline soil ecosystems, plant tolerance to salinity is important during germination and early seedling establishment [35].

Even halophyte plants are sensitive to salinity at the germination stage [36]. Their GPs decrease with increasing salinity, thus resulting in a delay in seed establishment. Therefore, it is significant to determine the salinity tolerance threshold for plants during the germination stage. Salinity tolerance of crops is estimated by comparing relative yields at different soil salinity levels. The graph response of a crop plant to salinity is linear and consists of two parts: (i) a line that specifies the range of the salinity tolerance threshold with a slope of zero and (ii) a line concentration dependent, whose slope depicts the rate of decrease in yield per unit increase in salinity. According to the above definition, the relative yield of a crop at a certain salinity level is obtained via the following equation [37]:

$$Y_r = 100 - b\,(EC_e - a)$$

where Y_r is relative germination; b is the line slope in percent; EC_e is the average salinity of the testing environment in dSm^{-1}; and a is the salt tolerance threshold in dSm^{-1}.

Assessments of light and thermal needs and tolerance to salt stress for seedling establishment in early growth stages is of great importance for introducing Hibiscus tea to an area. By providing some special pre-sowing treatments, seeds can be invigorated. There are many invigoration techniques like pre-sowing treatment (priming) and coating technologies. Results of several studies have shown that seed pretreatment with $CaCl_2$, KCl, and NaCl is an effective method to enhance speed and uniformity of emergence to achieve high early vigor under stress conditions [38,39]. Seed priming by stimulation of antioxidants

that provides seed tolerance to environmental stress conditions enhances early vigor and emergence rate [40]. Other researchers have found that microwave radiation on seeds resulted in an increase in their germination indices and also stimulated the concentration of the β-1,3-glucanase enzyme [41,42]. Jakubowski Tomasz used irradiated 2.45 GHz microwaves for 5–15 s for the germination capability of the germinated seeds of garden cress (*Lepidium sativum*) [43,44]. Abu-Elsaoud reported the positive effect of microwave electromagnetic radio frequency on the improvement of wheat seed germination [45]. Moreover, Mridha and colleagues reported that seed pretreatment with the magnetic field considerably increased the amount of indole-3-acetic acid and had favorable effects on germination and early growth [42]. Other authors reported the improvement of germination capacity of sunflowers by magnetic field seed pretreatment [46,47]. Seed priming has been used to improve germination, reduce seedling emergence time, and ameliorate stand establishment and yield [48]. This study aimed to explore the effects of light, temperature, and salinity on the germination and growth stages of Hibiscus tea for determining sowing date(s), improving threshold tolerance to salinity, and enhancing cultivation extension in slightly saline lands.

2. Materials and Methods

2.1. Seed Collection and Field Site Description

The mature seeds of Hibiscus tea (*Hibiscus sabdariffa*) were collected in Zabol City in Sistan and Balochistan Province with the coordinates of 31°0′0′′ N and 61°32′0′′ E. The seeds were stored in plastic bags at 5 °C until starting each experiment.

2.2. Experiment 1: Temperature and Light Requirements for Germination

The first experiment was a factorial experiment conducted in a randomized complete block design with 4 replications. To investigate the influences of temperature and light on germination, the seeds were placed in a growth chamber at 5, 10, 15, 20, 25, 30, 35, and 40 °C for 9-day light and dark conditions. For complete darkness, petri dishes were wrapped in two layers of aluminum foils. Twenty-five seeds were placed on wet, one-layered filter paper and put in 9-cm plastic Petri dishes under controlled light conditions at the laboratory. The filter paper was initially moistened with 5 mL of distilled water or the test solution. Seeds were considered germinated when the radicals were at least 2 mm long. The germinated seeds were counted daily. Based on the GR responses to temperature, cardinal temperature was developed through intersecting lines (ISL) by using the following model [49,50],

$$f = \text{if } (T < T_{opt}, \text{ region } 1(T), \text{ region } 2(T))$$
$$\text{Region } 1(T) = b\,(T - T_b)$$
$$\text{Region } 2(T) = c\,(T_{max} - T)$$

where T represents temperature and T_b, T_{opt}, and T_{max} stand for base, optimum, and maximum temperatures, respectively.

T_b and T_{max} were derived from the intersection of each regression line with the horizontal (X) axis, and the optimum temperature was calculated from the intersection of the two linear regression lines of GR at sub-optimal and supra-optimal temperatures.

2.3. Experiment 2: Germination Tests under Saline Conditions

The second experiment was also a factorial experiment conducted in a randomized complete block design, with 4 replications. The treatments included 7 levels of salinity (0, 30, 60, 90, 120, 150, and 180 mM NaCl). Before testing, the seeds were washed with sterile distilled water. They were incubated at 30 °C under light conditions for 9 days. A preliminary experiment showed that the temperature of 30 °C was optimal for seed germination, both in dark conditions. The germinated seeds were counted daily until 9 days. Seeds were regarded as germinated when the radicals were at least 2 mm long [51]. The fresh weight of the seedling was carefully weighed with a scale, with an accuracy of 0.0001 g, after drying the surface with a paper towel. The seedlings were dried in the oven

at 70 °C for 24 h, and, then the weight was determined by weighing with an accuracy of 0.0001 g. Seedling lengths were measured by a ruler with an accuracy of one millimeter.

2.4. Experiment 3: Germination Tests of Primed Seeds under Saline Conditions

The third experiment was similarly a factorial experiment carried out in a randomized complete block design, with 4 replications. The treatments included priming at 2 levels (control seeds and primed seeds with 18 mM NaCl for 24 h) and salinity at 7 levels (0, 30, 60, 90, 120, 150, and 180 mM NaCl). After priming, the seeds were washed 3 times with distilled water and re-dried to their original weights with forced air under shade. They were then incubated at 30 °C under dark conditions for 9 days. In this experiment, we used the new seeds and not the non-germinated seeds from the second experiment. The germinated seeds were counted daily until 9 days. The mean germination time (MGT) was calculated to assess the germination rate (Ellis and Roberts, 1981). MGT and GR, and the coefficient of uniformity of germination (CUG) were calculated via the equation developed by [13,36] as follow:

$$\begin{gathered} CUG = GP/MGT \\ MGT = \sum DiNi/ni \\ GR = 1/MGT \end{gathered} \tag{1}$$

where N is the number of seeds that grow on day D, and D is the number of days from the date of germination. GP is the germination percentage; N is the number of seeds newly germinated at the time of t; and n is the total number of seeds germinated. For measuring the seedling dry weight (SDW), the samples were placed in an oven at 70 °C for 24 h and were then weighed with a scale, with an accuracy of 0.0001 gr (ISTA, 1985)20.

2.5. Statistical Analysis

Prior to analysis, the data population normality was verified by Kolmogorov–Smirnov with SPSS software. Next, a one-way ANOVA test was applied using SAS software.9.3 after ensuring the homogeneity of variances. Means were separated by least significant difference (LSD, $p \leq 0.05$) when treatment F values were significant at probability levels of 0.05 and 0.01.

3. Results

Germination was significantly affected by temperature. The seeds germinated more rapidly at 30 °C than at any other temperature treatments during the first 72 h and then reached a germination peak (Figure 1). Seed germination was slower at 10 °C compared to the other temperatures. The GP of the seeds reached 95% at 30 °C after 72 h, while it only reached 50% at 10 °C after 120 h. At temperatures lower and higher than 30 °C, seed germination was slower, and the slope declined (Figure 1).

Light had a significant effect on the GP. GPs were 25.71 and 87.67% in the light and dark conditions, respectively (Table 1). GR was significantly influenced by temperature. It was increased by enhancing temperatures from 5 to 30 °C and then suddenly dropped at higher temperatures. It was low at temperatures lower and higher than 30 °C. GRs were 0.17, 0.66, and 0.40 seeds per day at 10, 30, and 40 °C, respectively (Figure 2). There were positive and negative slopes in the ranges of 5–30 °C and 30–40 °C, respectively (Figure 2). The intersection of the linear regression fit between the mentioned ranges of temperatures, with the horizontal axis showing that the base and maximum temperatures were 5 and 53.75 °C, respectively (Figure 2). The intersection below and above the regression lines displayed an optimal temperature of 30 °C (Figure 2).

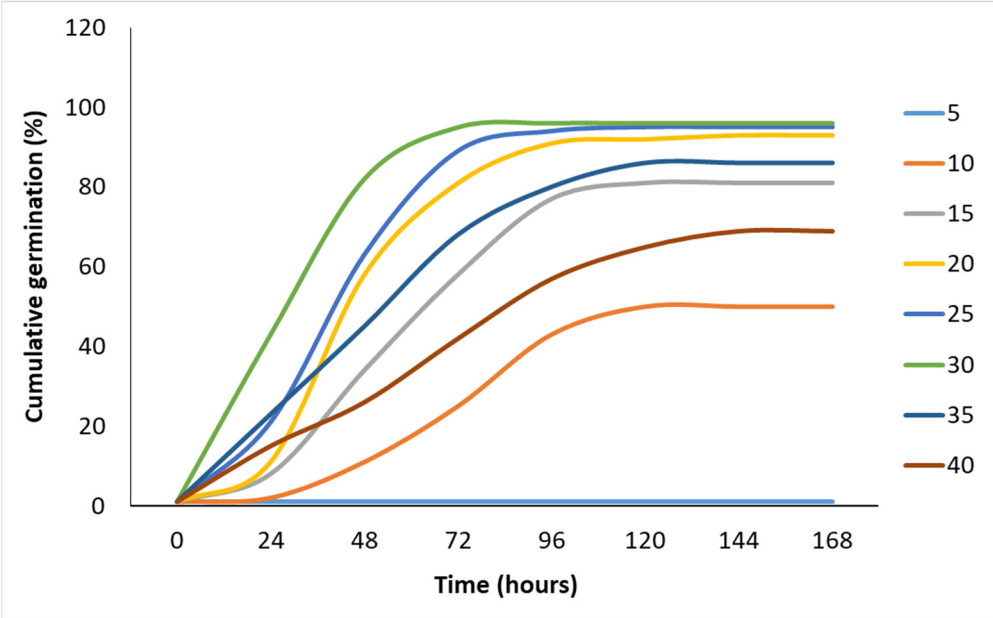

Figure 1. Hibiscus tea germination responses to temperatures of 5 to 40 °C.

Table 1. Effects of light and different temperatures on GR, SDW, and CUG of Hibiscus tea seeds.

CUG		SDW (g)		GR (Seeds per Day)		GP %		
dark	light	dark	light	dark	light	dark	light	Temperature
0 ^{e}	0 ^{j}	0 ^{j}	0 ^{e}	0 ^{e}	0 ^{e}	0	0	5
3.65 ^{d}	2.17 ^{f}	0 ^{f}	0 ^{c}	0.17 ^{d}	0.16 ^{d}	50	36	10
7.38 ^{c}	5.02 ^{e}	0.014 ^{e}	0.012 ^{d}	0.36 ^{bc}	0.28 ^{c}	81	72	15
9.57 ^{bc}	8.19 ^{c}	0.016 ^{d}	0.015 ^{bc}	0.41 ^{b}	0.36 ^{ab}	93	85	20
10.88 ^{b}	9.05 ^{b}	0.018 ^{ab}	0.017 ^{ab}	0.46 ^{ab}	0.39 ^{ab}	95	89	25
14.16 ^{a}	11.15 ^{a}	0.020 ^{a}	0.018 ^{a}	0.66 ^{a}	0.51 ^{a}	98	90	30
8.52 ^{c}	8.28 ^{c}	0.019 ^{ab}	0.017 ^{ab}	0.45 ^{b}	0.35 ^{ab}	89	86	35
6.05 ^{d}	6.23 ^{d}	0.017 ^{c}	0.016 ^{b}	0.40 ^{bc}	0.35 ^{b}	69	70	40

Means followed by the same letter within a column are not significantly different ($p \leq 0.05$), according to the least significant difference (LSD) test, germination percentage (GP), germination rate (Gr), seedling dry weight (SDW) and coefficient of uniformity of germination (CUG).

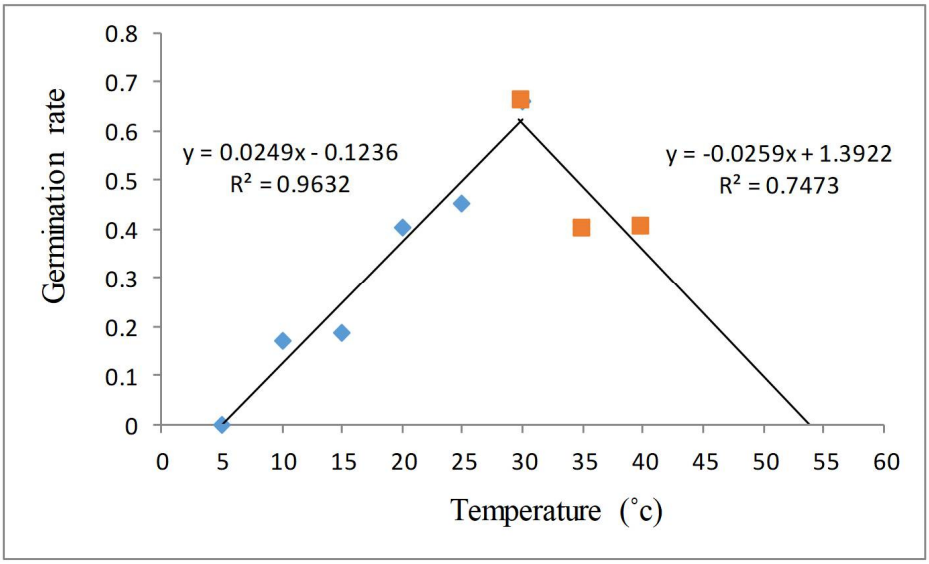

Figure 2. Germination rate response of *Hibiscus sabdariffa* seeds to temperature.

By increasing temperatures, the GRs reached 0.51 and 0.66 seeds per day at 30 °C in light and dark conditions, respectively. Temperatures above 30 °C reduced GR and thus, by increasing temperature from 30 to 40 °C, the GRs were decreased to 0.35 and 0.40 seeds per day in light and dark conditions, respectively (Table 1). By increasing temperature from 15 to 30 °C, SDWs reached from 0.012 to 0.018 gr and 0.018 to 0.016 gr in light and dark conditions, respectively (Table 1). CUG was significantly affected by light. A comparison of the means revealed that CUG reached from 2.17 and 3.65 at 10 °C to 11.15 and 14.16 at 30 °C, while decreasing to 6.23 and 6.05 at 40 °C in light and dark conditions, respectively (Table 1).

3.1. Effects of Salinity on Seed Germination

GP was significantly affected by salinity. The results demonstrated that GP decreased with increasing salinity (Table 2). The highest GP (96%) was observed in distilled water, while it reached 15 and 0% at NaCl concentrations of 150 and 180 mM NaCl (Table 2). GR was significantly reduced by salinity. A comparison of the means showed no significant differences between the control group and treatment with 30 mM NaCl (Table 2). Seedling length (SL) was significantly affected by salinity. A comparison of the means revealed that SL was decreased to 7.26 cm by increasing the concentration to 30 mM NaCl (Table 2). Seedling fresh weight (SFW), SDW, and CUG were significantly influenced by salinity. Mean germination time (MGT) was significantly influenced by salinity, and it was decreased significantly with increasing salinity concentration. CUG was lowered to 20.04 by enhancing the concentration from the control level to 30 mM NaCl (Table 2).

Table 2. Comparisons of GP and GR means and morphological traits of Hibiscus tea seeds at different salt (NaCl) levels.

CUG	SDW (g)	SFW (g)	SL (cm)	GR (n/d)	MGT (d)	GP (%)	NaCl (mM)
21.82 ^{a}	0.034 ^{a}	2.73 ^{a}	9.14 ^{a}	0.91 ^{a}	1.098 ^{e}	96 ^{a}	0
20.04 ^{b}	0.030 ^{a}	0.343 ^{b}	7.26 ^{b}	0.87 ^{ab}	1.149 ^{c}	92 ^{b}	30
13.08 ^{c}	0.021 ^{b}	0.238 ^{bc}	4.63 ^{c}	0.80 ^{b}	1.25 ^{bc}	75 ^{c}	60
10.62 ^{d}	0.020 ^{bc}	0.226 ^{bc}	4.20 ^{d}	0.72 ^{c}	1.39 ^{b}	59 ^{d}	90
3.82 ^{e}	0.016 ^{cd}	0.19 ^{cd}	3.29 ^{e}	0.61 ^{d}	1.64 ^{ab}	35 ^{e}	120
1.67 ^{f}	0.014 ^{d}	0.16 ^{d}	1.91 ^{f}	0.45 ^{e}	2.3 ^{a}	20 ^{f}	150
0 ^{g}	0 ^{e}	0 ^{e}	0 ^{g}	0 ^{f}	0 ^{f}	0 ^{g}	180

Means followed by the same letter within a column are not significantly different ($p \leq 0.05$), according to the least significant difference (LSD) test, germination percentage (GP), germination rate (GR), seedling length (SL), seedling dry weight (SDW), and the coefficient of uniformity of germination (CUG).

Although Gp, GR, CUG, and all the morphological traits of seedlings decreased with increasing salinity, the declining CUG slope was still at a higher level than that of the other traits (Table 3).

Table 3. Reductions of GP, MTG, GR, SL, SFW, SDW, and CUG of Hibiscus tea seeds with increasing salinity.

Morphological Traits of Germination	Equation	R^2
GP	$Y = -1.024x^2 - 8.67x + 109$	0.99
MGT	$Y = 0.0582x^2 - 0.2012x + 1.279$	0.98
GR (day)	$Y = -0.00003x^2 + 0.0016x + 0.8733$	0.98
SL (cm)	$Y = -0.0305x^2 + 0.102x + 0.81$	0.98
SFW(g)	$Y = -0.0037x^2 - 0.097x + 0.55$	0.91
SDW (gr)	$Y = -0.0002x^2 - 0.003x + 0.038$	0.933
CUG	$Y = -0.189x^2 - 5.49 + 28.34$	0.97

Germination percentage (GP), mean germination time (MGT), germination rate (Gr), seedling length (SL), seedling fresh weight (SFW), seedling dry weight (SDW), and the coefficient of uniformity of germination (CUG).

Based on the regression equation, the relationships of the different salinity levels with GP and GR of the seeds were marked by 99 and 98% of the points, respectively (Figures 3 and 4).

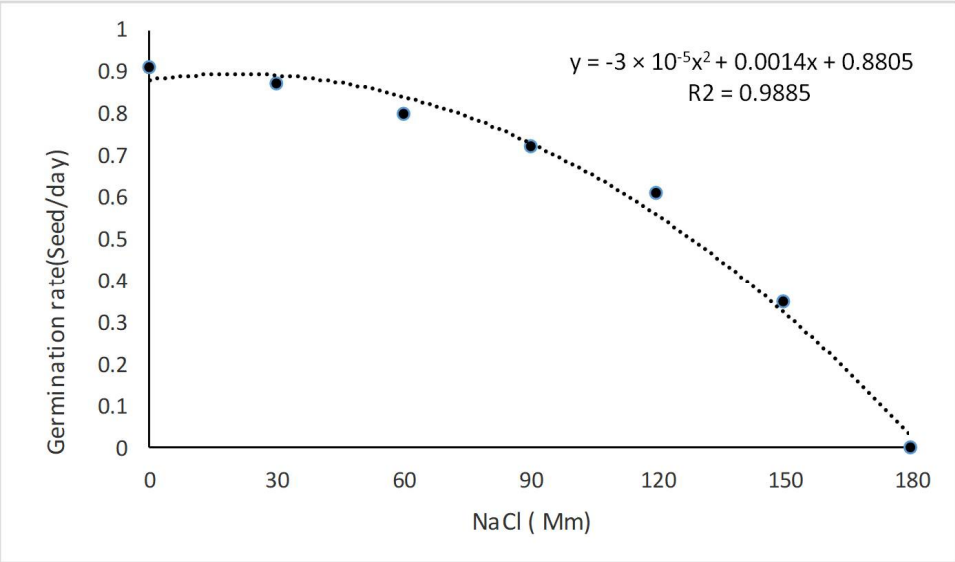

Figure 3. Effects of salinity stress on the GR of Hibiscus tea seeds.

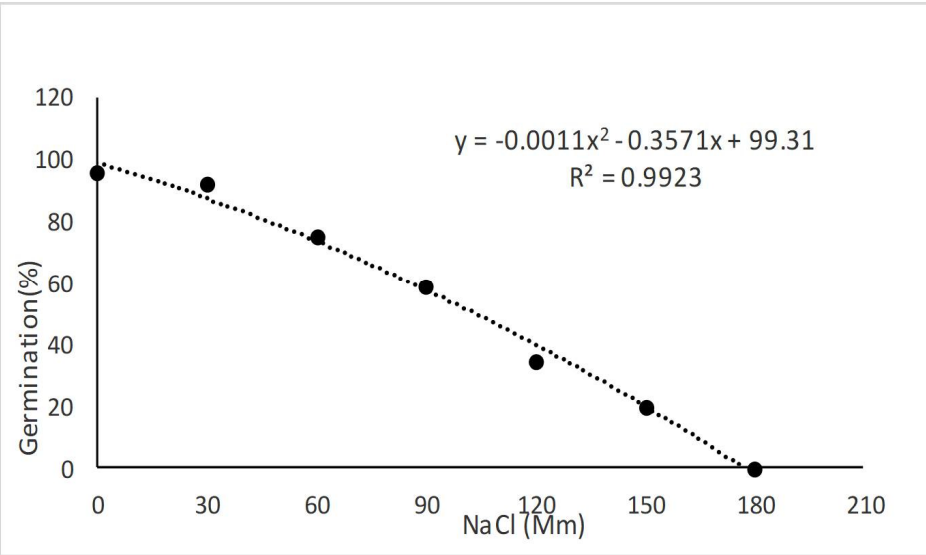

Figure 4. Effects of salinity stress on the GP of Hibiscus tea seeds.

3.2. Hibiscus Sabdariffa Tolerance Threshold and Germination Tolerance to Salinity before and after Priming

The salt tolerance threshold is a critical parameter for establishing plant salt tolerance, which is generally known as the relative yield response to increasing salinity. In this study, the results showed a negative trend between salinity and germination traits (Table 3). The germination processes of *H. sabdariffa* tolerated a salinity of up to 30 mM of NaCl (Table 2, Figure 5). Higher concentrations led to a collapse of the germination processes. Therefore, the 30 mM salinity of NaCl was considered the salinity tolerance threshold of *H. sabdariffa*. The salinity tolerance threshold of the seedlings was also influenced by the priming of the seed. Priming with NaCl (18 mM for 24 h) increased the tolerance threshold for salinity in the germination phase. The germination tolerance threshold increased from 30.23 ds/mM (in non-primed seeds) to 30.97 mM NaCl ds/m^2 in primed seeds (Figures 5 and 6). The

results showed that priming with NaCl (18 mM for 24 h) increased the salinity tolerance threshold in the *H. subdariffa* germination phase. Furthermore, the slope of germination decreasing with increasing salinity was affected by priming, so that with increasing salinity, the slope of germination reduction after the tolerance threshold at the salinity point was lower in the primed seeds than in the non-primed seeds (Figures 5 and 6). Priming the stem strengthened the salinity tolerance threshold and the salinity tolerance level after this point in the germination phase.

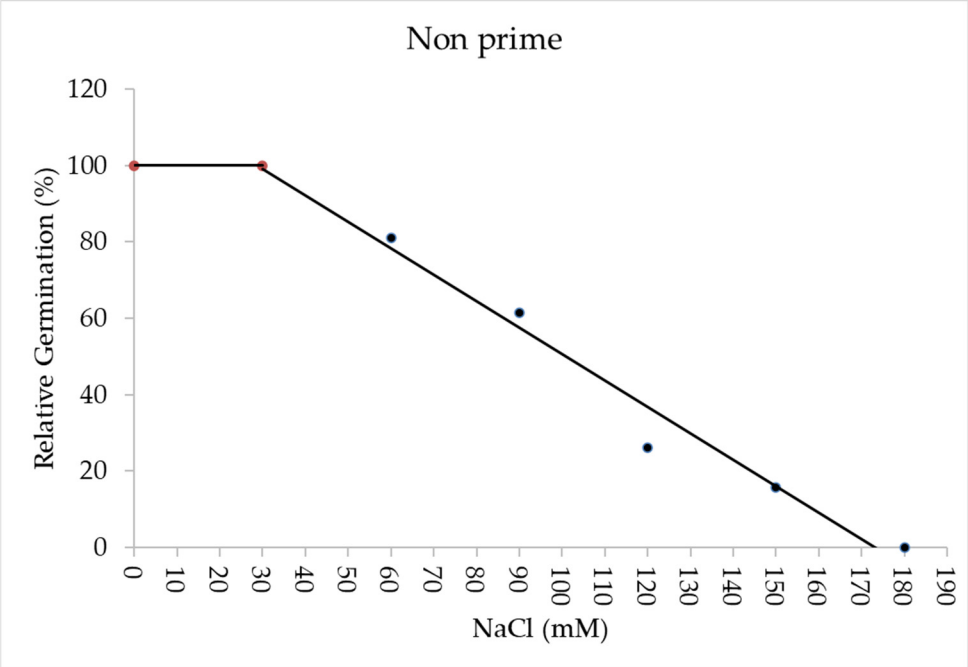

Figure 5. Hibiscus tea relative germination response to increasing salinity in non-primed seeds.

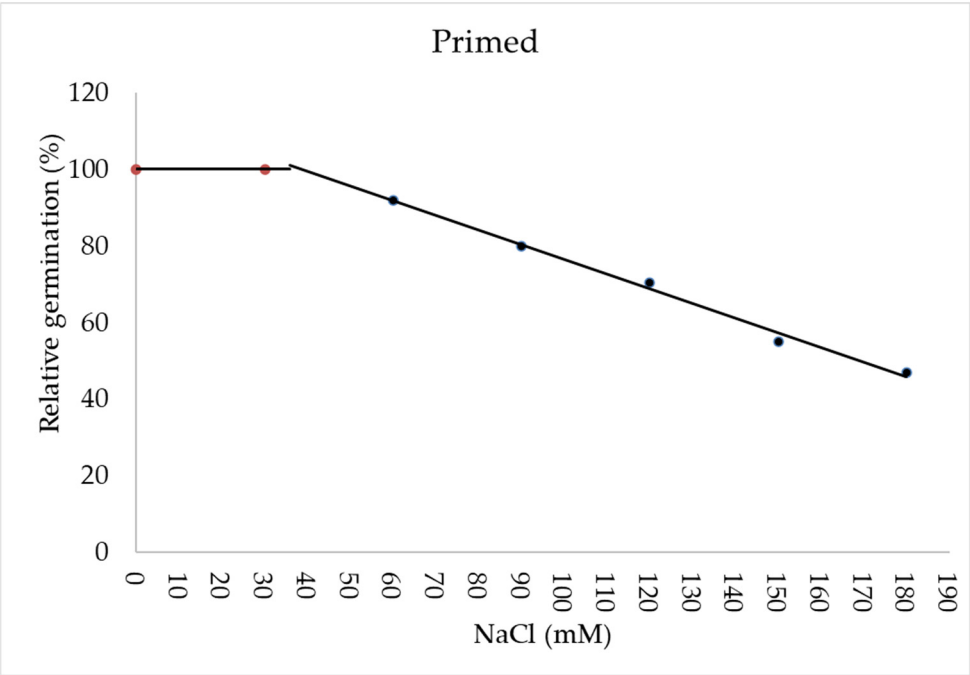

Figure 6. Hibiscus tea relative germination response to increasing salinity in primed seeds.

The results showed that seed priming, in addition to the index of salinity tolerance threshold during germination, increased the salinity tolerance threshold to the salinity of seedlings, especially SL and SDW (Table 4).

Table 4. Effects of priming on the reduction of GR and enhancement of morphological traits per unit of salinity (EC dSm^{-1}) above the threshold value.

	GP (%)	GR (Seeds per Day)	SL (cm)	SDW (g)	CUG
Non-primed seeds	30.23 [b]	30.29 [ab]	31.38 [b]	30.70 [ab]	30.44 [ab]
Primed seeds	30.97 [a]	30.52 [a]	31.50 [a]	30.97 [a]	30.58 [a]

Means followed by the same letter within a column are not significantly different ($p \leq 0.05$), according to the least significant difference (LSD) test, germination percentage (GP), germination rate (Gr), seedling length (SL), seedling dry weight (SDW), and coefficient of uniformity of germination (CUG).

4. Discussion

This study aimed to recognize the effects of cardinal temperature, light, salt tolerance, and halopriming on *H. sabdariffa* during the germination and seedling stages for its extensive cultivation. Seed germination is a critical phase that is affected by environmental and genetic factors. Among all the environmental factors of seed germination, cardinal temperature (minimum, optimum, and maximum temperatures), light, and salinity are considered as the major factors [28,52]. Cardinal temperatures (minimum or base temperature T_b, optimum temperature, T_{opt} and maximum temperature, T_{max}) describe the temperature range over which the seeds of a particular species can germinate [53–56]. The cardinal temperatures for germination have been determined via the use of germination rate for different plants [55,56]. Our results confirmed that *H. sabdariffa* seed germination was also affected by temperature, as previously observed for other plants [11,39,50]. Our results suggested that the appropriate temperature for *H. sabdariffa* seed germination was 30 °C. This data is essential for planning *H. sabdariffa* cultivation strategies. In fact, errors in the timing of sowing in the field can affect the yield of the final crop [57]. Furthermore, sudden climatic changes can affect the germination of *H. sabdariffa*, making its cultivation even more precarious [58]. In general, considering all the weather-climatic parameters, spring cultivation could be recommended in cold regions. Identification of T_b, T_{opt}, and T_{max} for seed germination is essential for determining the best planting date [59]. Seed germination response to temperature is a basis for predicting germination timing and introducing suitable regions for cultivation in a range of climates [51,60]. To describe the relationship between temperature and GR, researchers have mostly utilized linear regression [50,61]. Using the ISL regression model, it is possible to estimate the cardinal temperature for plants [62]. Our results showed that the T_b, T_{opt}, and T_{max} for *H. sabdariffa* germination were 5, 30, and 40 °C, respectively. These temperatures for *H. sabdariffa* seed germination are similar to those recorded by other authors (4.4, 29.30, and 50 °C, respectively) [63]. Similar findings were reported for *Haloxylon aphyllum*, *Bouteloua curtipendula*, and *Calotropis procera* [39,64].

Light is another environmental factor that is effective in germination [28]. The sensitivity of seeds to light in the germination stage is very variable according to the species of seed [6]. Plant physiology divides seeds into photoblastic (light-dependent germination) and aphotoblastic (light-independent germination) [65]. The response of seeds to light is regulated by specific receptors, the photoreceptors, capable of being activated with red light and triggering a series of metabolic reactions associated with germination and governed by hormones such as gibberellins [66]. In other species, germination is inhibited by continuous white light [53]. In the present research, continuous light and dark conditions resulted in a decrease and increase in the germination of *H. sabdariffa* seeds by up to 90 and 98%, respectively, at 30 °C. The trend of the germination percentage from 5 at 30 °C ranged from 0 to 90% in the dark and from 0 to 98% in light. The highest percentage and germination rate were reported at 30 °C in both light and dark. The darkness and the far-red light favor germination because they represent the conditions of the deep soil [66].

In fact, when *H. sabdariffa* seeds are sown close to the soil surface, shallow depth, elevated soil temperature, and soil moisture loss caused by direct sunlight in the summer inhibit germination in arid areas. Conversely, to achieve maximum germination, *H. sabdariffa* seeds should be sown deeper. Taghvaei and Ghaedi reported similar results about black Haloxylon (*Haloxylon aphyllum* L.) [11]. In this study, the mean SDW increased in dark conditions. Khan and Rizivi [67] and Flores [68] reported similar findings about *Atriplex graffithii*, black Haloxylon and *Agave salmiana*, and *Mammillaria compressa* responses of seedling characteristics, respectively.

To evaluate the tolerance of different species to salinity, it is appropriate to investigate a plant's early growth phase, since germination and seedling traits are the most appropriate aspects on which a plant's final yield strongly depends [35]. Our results demonstrated that the GP, GR, SDW, and CUG parameters of *H. sabdariffa* decreased with an increasing degree of salt stress. Yuan and colleagues reported similar results about the responses of *H. sabdariffa* seedling traits to salinity [69]. A similar response in terms of percentage of germination in the presence of salt stress was observed in *Pisum sativum* L., *Calotropis procera* L., and *Chenopodium glaucum* L. crops [70,71]. Reginato and colleagues reported sensitivities of hypocotyls and radicals of *Prosopis strombulifera* L. to salinity [72]. Finally, Yildirim and Karlidag reported similar results about the effects of salinity stress on the dry weights of both the *Physalis ixocarpa* and *Physalis peruviana* species [73].

It is well known that salinity has a negative impact on seed germination and seedling traits [74]. However, this negative correlation varies depending on salt concentration. The development of the organs of the seedling is strongly reduced by the increase in salinity, since it induces a high osmotic potential, due to the accumulation of salt and the specific toxicity of the Na^+ and Cl^- ions in the germination phase [74]. The excess of Na^+ and Cl^- ions alter the transport of other ions fundamental for the metabolism of plant cells such as the ions K^+, Ca^{2+}, and Mg^{2+}. Thus, identifying a parameter that indicates the salt tolerance threshold of plants is essential. Usually, the salt tolerance threshold corresponds to how the yield of the plant varies with increasing salt concentration. Hence, the plant's tolerance to salinity can be expressed by plotting the relative yield as a continuous function of salinity [75]. The relationship between plant tolerance and salinity is represented by two straight lines, which, when intersecting, allow the identification of the threshold, after which the morphological traits of germination begin to decline [76]. Our results revealed a decrease of more than 30.23 mM NaCl in *H. sabdariffa* germination, with a steep slope in salinity conditions. Seed halopriming is known to often alleviate salinity stress during germination and seedling growth [77–79]. Seed halopriming is a practice that enhances antioxidant stimulation by improving the tolerance of seeds to environmental conditions, improving early vigor and seedling emergence rate [80]. In this research, halopriming enhanced the salinity tolerance threshold besides augmenting other morphological traits of the seedlings. Hence, relative germination increased from 30.23 to 30.97 mM NaCl (Table 4). Our results were well consistent with the results of Masondo and colleagues, who showed that seed priming increased salt tolerance of *Ceratotheca triloba* at the germination stage [81]. Halopriming can elevate germination in a saline environment by enhancing the rate of metabolism [82]. The type of priming agents ($CaCl_2$, KCl, and NaCl) affects fresh and dry shoot biomass in crop plants [38]. It is concluded that seed priming with NaCl reduces the adverse effects of salinity stress on the early growth characteristics of *H. sabdariffa*.

5. Conclusions

In this study, the best conditions of temperature, light, and salinity of the germination processes of Hibiscus tea have been identified. Therefore, Hibiscus tea showed the typical characteristics of a plant of temperate latitudes. Based on the data collected, it can be assumed that Hibiscus tea can also be cultivated in other areas of the planet. For example, in cold ecosystems, such as the province of Fars in Iran, Hibiscus tea can be grown after mid-spring (mid-May to mid-June). Its germination phase is sensitive to salinity. Therefore, Hibiscus tea cannot be easily sown in saline soil conditions. However, seed halopriming

could increase the salinity tolerance of Hibiscus tea, as observed in this study, and facilitate sowing in relatively saline soils.

In conclusion, the Fars Province and Mediterranean areas of Iran may have the potential for spring cultivation of Hibiscus tea in a double-cropping system, even in slightly saline soils, since these regions normally lack frost, with soil temperatures above 20 °C from May through June. This study indicates that halopriming may increase salinity tolerance in the germination stage and make it possible to establish these plants in saline lands.

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