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Plant Extracts Are a Promising Solution to Alleviate Salinity Stress at the Chili Pepper Germination Stage

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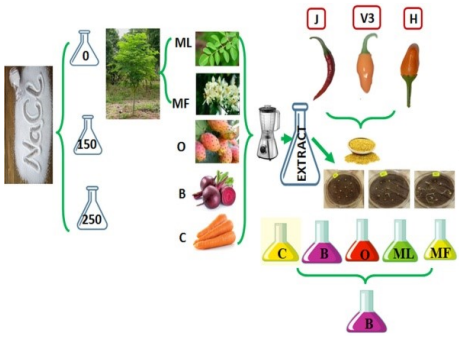
Abstract



Salinity is one of the most pervasive abiotic stressors limiting global agricultural productivity, particularly by impairing seed germination in sensitive species, such as chili peppers (Capsicum spp.). This study rigorously evaluated the potential of nature-derived biostimulants to alleviate salinity-induced germination inhibition in three agronomically important Yemeni chili pepper genotypes: Haimi (H), Hajjah (J), and Jawfi3 (V3). Aqueous extracts from red beetroot, prickly pear fruit, carrot roots, and moringa leaves and flowers were subjected to an optimized 18-hour seed priming protocol. The seeds were germinated at three salinity levels (0, 150, and 250 mM NaCl) in a randomized complete block design. The results revealed a striking mitigation of salinity stress at 150 mM NaCl by all plant extracts, with substantial protective effects observed at the extreme 250 mM NaCl concentration. Notably, beetroot extract emerged as the most potent biostimulant, consistently delivering the highest Extract Stimulation Index (ESI%) across all germination parameters including germination percentage (GrP%), mean germination time (MGT), germination speed (GSC), and radicle length. Its performance was robust across all genotypes and salinity levels, thereby highlighting its broad-spectrum efficacy. These findings provide the first empirical evidence that cost-effective natural extracts, particularly from red beetroot and prickly pear fruit, can match or surpass synthetic priming agents in enhancing seed germination under saline conditions.

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1. INTRODUCTION

Chili pepper(Capsicum spp.) is one of the most important vegetable crops belonging to the Solanaceae family, commonly known as "Chili," a name derived from its origin in Mexico[1]. Cultivated worldwide in both warm and temperate regions, chili peppers have significant economic and nutritional value. It is widely used as a spice, consumed fresh or dried, and plays a crucial role in the food industry, such as in the production of hot sauces [2, 3]. Additionally, chili peppers are rich in essential nutrients, particularly vitamin C and capsaicin, which have been extensively studied for their potential health benefits [4, 5, 6, 7] Moreover, chili pepper exhibits antimicrobial properties[8], making it a staple ingredient in

various global cuisines[2, 3].

According to the 2020 Annual Agricultural Statistics, chili pepper is a vital crop in Yemen, ranking seventh in terms of cultivated area and production among vegetables, accounting for approximately 4% of the country's total vegetable production[9]. It surpasses crops, such as carrots and zucchini, highlighting its relative importance in the agricultural sector. Furthermore, Food and Agriculture Organization (FAO) data indicate a linear increase in chili pepper production over the past five decades. In 2022, Yemen produced approximately 18,223.37 tons from a cultivated area of 3,240 hectares, representing 2.3% of the global production of 788,032.04 tons from 689,336 hectares [10].

Despite its significance, chili pepper cultivation in arid and semi-arid regions faces substantial challenges, primarily owing to soil salinity. This issue arises from the accumulation of salts due to improper irrigation practices, the use of saline water, and excessive fertilization, and climate change further exacerbates soil salinity, posing a threat to agricultural productivity[11, 12, 13].

Over 800 million hectares of agricultural land worldwide are severely affected by salinity [14], making it one of the most critical challenges to global food security. Salinity is a major abiotic stress that significantly constrains crop production, particularly in arid and semi-arid regions [15]. Approximately 23% of the world's cultivated land is classified as saline, whereas 37% is sodic [16].

Yemen features a diverse range of climates, including semi-humid, semi-arid, and arid tropical regions [17]. Within the country, approximately 37,100 ha of non-desert agricultural land are affected by salinity levels. Additionally, 12 million hectares are impacted by erosion, and 3.8 million hectares exhibit varying degrees of salinity, with 3–5% of this land at risk of desertification [18, 19]. Notably, highland regions remain unaffected by salinity [20].

Research on salt stress tolerance in various crops, including pepper[21, 22] and fenugreek [23], often focuses on specific salt concentrations and exposure durations. Responses to salt stress also vary according to plant species, genotype, climatic conditions, and water and soil characteristics [24, 25]. Chili peppers can be classified as either sensitive to salinity [26, 27, 28] or moderately salt-tolerant [29, 30]. Among the plants tested by Loganayaki et al.. *et al.* [31], chili was found to be the most sensitive compared to tomato, okra, and cucumber.

Salinity, a form of abiotic stress, significantly impairs germination, growth, and productivity of chili pepper plants [21, 29, 32, 33]. For instance, studies have shown that chili pepper and tomato seeds fail to germinate at a salinity level of 200 mM NaCl [31]. Elevated salinity levels not only delay germination but also reduce germination rates, further exacerbating the challenges of cultivating these crops under saline conditions [21, 34, 35].

The germination stage is particularly sensitive to salt stress [12, 36]. Elevated salinity levels hinder seed germination by decreasing water absorption under osmotic stress and by causing ionic stress. Increased salt concentrations in the germination medium adversely affect seed embryo vitality due to disrupted ion transport[37]. An inverse correlation exists between salinity and germination in various vegetable plants, including cucumber [38], sweet pepper[29, 39, 40], and tomato[41].

Ongoing research efforts are focused on developing agricultural technologies to mitigate the adverse effects of salinity on crop production, including breeding salt-tolerant plants [42, 43], employing grafting techniques on vegetables [44, 45] or fruits [46], utilizing growth regulators and plant biostimulants [32, 47, 48], and managing soil salinity through excessive irrigation [49].

Plant biostimulants are non-pesticidal and non-nutritional organic compounds that enhance plant growth and development while improving stress resistance [50]. They can be applied through foliar spraying, soil application, or seed treatment[51, 52]. According to du Jardin [53]**,** biostimulants are classified into seven categories: humic and fulvic acids, seaweed and plant extracts, microbial inoculants, protein hydrolysates and amino acids, chitosan and other biopolymers, inorganic compounds, and beneficial elements.

Plant extracts, derived from natural sources, are widely used as cost-effective biostimulants to enhance plant growth[54]. This prompted numerous studies to explore the use of plant extracts to mitigate their effects. For instance, extracts from moringa leaves [55] and carrot roots[56] have shown promise in reducing salinity stress during seed germination.

In a previous study conducted by Arraf and Al-Madhagi[54], the authors demonstrated the effectiveness of using extracts from prickly pear, red beet, and moringa flowers to improve the quality of pepper seedlings. However, the potential of these extracts to mitigate the adverse effects of salinity stress has not yet been investigated, leaving a significant gap in literature. This presents a critical opportunity to explore the role of bioactive compounds derived from these plants in addressing salinity-related challenges and enhancing crop resilience under saline conditions.

The central hypothesis of this study was that plant-based extracts, particularly those derived from moringa flowers, prickly pear fruits, red beetroots, and carrot roots, may possess significant potential to alleviate salinity stress. For instance, prickly pear, which thrives in arid and saline environments, is rich in proline and other bioactive compounds [57], making it a strong candidate for reducing salinity stress. Similarly, red beetroot, known for its high salt tolerance (up to 4 dS/m) [58], offers a promising solution for mitigating salinity stress in sensitive chili pepper genotypes.

The primary objective of this study was to evaluate the efficacy of aqueous plant extracts obtained from moringa

leaves and flowers, prickly pear fruits, red beetroots, and carrot roots in alleviating the detrimental effects of salinity on chili pepper seed germination.

2. MATERIALS AND METHODS

2.1. Study Location and Chili Pepper Genotypes

The experiment for this study was conducted in the laboratory of the Horticulture and Its Technologies Department, Faculty of Agriculture, Food, and Environment at Sana'a University during the academic year 2021–2022.

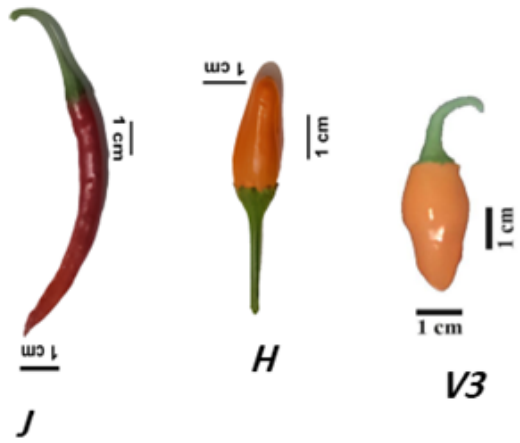

Figure 1. Local chili pepper genotypes were used in this experiment. **Genotypes**: *H= Haimi, J =Hajjah,* and *V3 = Jawfi3*.

A Randomized complete block design (RCBD) was employed with three replicates, each containing 10 seeds. The study utilized seeds from three local chili pepper genotypes: *Haimi* (*H*), *Jawfi3* (*V3*), and *Hajjah* (*J*), as illustrated in Figure(1).

2.2. Preparation of Plant Extracts

Plant extract samples were obtained from local markets, and moringa leaves and flowers were collected from a mature tree grown in the Bani Asim area of Sana'a.

The samples were sterilized by rinsing with running water and multiple washes with distilled water. Prickly pear fruits were cut and immediately juiced. Moringa leaves and flowers were manually extracted using an electric blender, followed by grinding with a ceramic mortar. Similarly, beetroot and carrot roots were grated and blended using an electric mixer. Table(1) provides the details of the extract characteristics.

2.3. Seed Sowing and Treatment Application

Seed sterilization was performed using a solution containing 10% sodium hypochlorite, 90% distilled water, and a drop of Tween 20 for 5 min. After sterilization, the seeds were rinsed with running water and then multiple times with distilled water before being dried for planting. The filter paper was immersed in Petri dishes containing 3 mL NaCl at three concentrations (0, 150, and 250 mM), with distilled water as the control.

Seeds of the local chili pepper genotypes (*V3*, *H*, and *J*) (Figure 1) were soaked overnight (18 h) in the different plant extract concentrations listed in Table(1) for over the night (18 hours). Seeds in the control group were soaked in distilled water. The seeds were then planted in Petri dishes containing saline concentrations (0, 150, and 250 mM).

2.4. Experimental Measurements

Germination data were recorded daily for 21 days, as outlined in Table(2).

2.5. Seedling Morphometric Analysis

Hypocotyl and radicle elongations (cm) in normal seedlings were quantified using standardized digital morphometry. Each specimen was imaged against a calibrated black background setup featuring precision reference scales (ruler and coin) using high-resolution photography on a mobile device. Raw images underwent rigorous computational processing: (1) resolution-optimized preprocessing in *IrfanView* software, followed by (2) metrically calibrated morphometric analysis using *Fiji/ImageJ* software with traceable scale standardization (Figure(2)).

2.6. Statistical Analysis

The experimental data were subjected to comprehensive statistical analysis using GenStat 12 software (VSN International, UK). A multifactorial ANOVA was conducted to assess the main and interaction effects of the factors studied. Significant differences between treatment means were further determined using Duncan's Multiple Range Test (DMRT) at a significance level of $P \leq 0.05$.

Additionally, the Salinity Stress Mitigation Efficiency (%SEI) of the plant extracts was calculated for significant interactions. This was determined by computing the salinity sensitivity index under extract-treated conditions, using the applying modified equation of Horuz. et al.[62]:

$$SEI\% = \left(\frac{P0 - PE}{PE} x\ 100 \right) \quad (1)$$

Where;
P0 = parameter with extract in salt level,
PE = parameter without extract in same salt level.

Table 1. Extract characteristics

Extract	Plant part	Research Code	Stock preparation W(g)/ V(ml)	Concentration								
				Higher			Medium			lower		
				pH	EC	TSS	pH	EC	TSS	pH	EC	TSS
Control	-	NO	-	7	0	0	7	0	0	7	0	0
Prickly pear	Fruit	O	300/0	6.7	1.5	8.2	6.8	0.8	5.4	6.8	0.4	3.6
Beetroot	Root	B	100/150	7	2.7	4.2	7	1.4	3.4	6.9	0.7	3.2
Carrot	Root	C	100/ 150	6.8	1.6	3.6	6.8	0.9	3.4	6.8	0.4	3.2
Moringa	Leaf	ML	100/30	6.3	2	3.6	6.2	1.1	3.5	6.2	0.8	3.4
Moringa	Flower	MF	82.5/30	6.2	1.4	4	6.1	0.8	3.8	6.2	0.5	3.6

The study utilized water extracts exclusively, with beetroot, prickly pear fruit, and carrot extracts tested at low, medium, and high concentrations of 10%, 25%, and 50%, respectively, while Moringa leaves and flowers were evaluated at 10%, 20%, and 30%. pH levels were measured with a pH meter, electrical conductivity (EC, mS/cm) was determined using an EC meter, and total soluble solids (TSS, °Brix) were assessed via a hand refractometer.

Table 2. The various metrics used to calculate the process of seed germination in the experiment.

Measurements	Unit	Equation	Ref
Germination Percentage (GrP)	%	$GrP = \left(\frac{\sum_{i=1}^{k} n_1}{N}\right) \times 100$ (1)	[59]
Mean Germination Time (MGT)	Day	$MGT = \frac{\sum_{i=1}^{k} n_i t_i}{\sum_{i=1}^{k} n_i}$ (2)	[60]
Mean Germination Rate (MGR)	day^{-1}	$MGR = \frac{1}{MGT}$ (3)	[61]
Germination speed coefficient	%	$GSP = \left(\frac{\sum_{i=1}^{k} G_i}{\sum_{i=1}^{k} G_i X_i}\right) \times 100$ (4)	[61]
Coefficient of Velocity of Germination	%	$CVG = \left(\frac{SDG}{MGT}\right) \times 100$ (5)	[61]

In the germination equations: N, the total number of seeds in each experimental unit; n_i, the number of seeds germinated in the i^{th} time; k, the last day of germination evaluation; t_i, the period from the commencement of the experiment to the i^{th} observation; G_i, the number of seeds germinated in the i^{th} time; and X_i, the number of days from sowing; SDG denotes the germination standard deviation

3. RESULTS

3.1. Germination percentage (GrP):

The results generally indicated that the independent factors genotype, salinity levels, and plant extracts had a significant effect on the total germination percentage (%) at a probability level of ($P < 0.001$), except for the effect of extract concentrations, which was not statistically significant (Table 3).

As shown in Table 3, the *Haimi* genotype exhibited the highest germination percentage, which was significantly different from those of the *Hajjah* and *Jawfi3* genotypes. Among the plant extracts, beetroot extract was the most effective in terms of germination percentage, showing no significant difference from the moringa flower and prickly pear fruit extracts. Most two-way interactions were statistically significant, except for those involving extract concentrations. In contrast, the three-way interactions were not significant except for the interaction between genotypes, salinity, and extracts (G×N×E), which demonstrated a significant effect at a probability level of less than 5% ($P < 0.05$). The four-way interaction (between all factors) was not significant (Table 3).

The discovery of a significant three-way interaction (G×N×E; $P < 0.05$) between genotype, salinity, and plant extracts represents a major advance in our understanding of the stress mitigation mechanisms of GrP% (Table 4). Under non-saline conditions, plant extracts had no significant effect on germination, as they did not exhibit statistically significant values, either positive or negative, in terms of the extract efficiency coefficient (SEI%). However, the extracts clearly mitigated the impact of salinity stress on the germination percentage (Table 4).

For the *Hajjah* (*J*) genotype, germinating at a salinity level of 150 mM, all plant extracts significantly increased

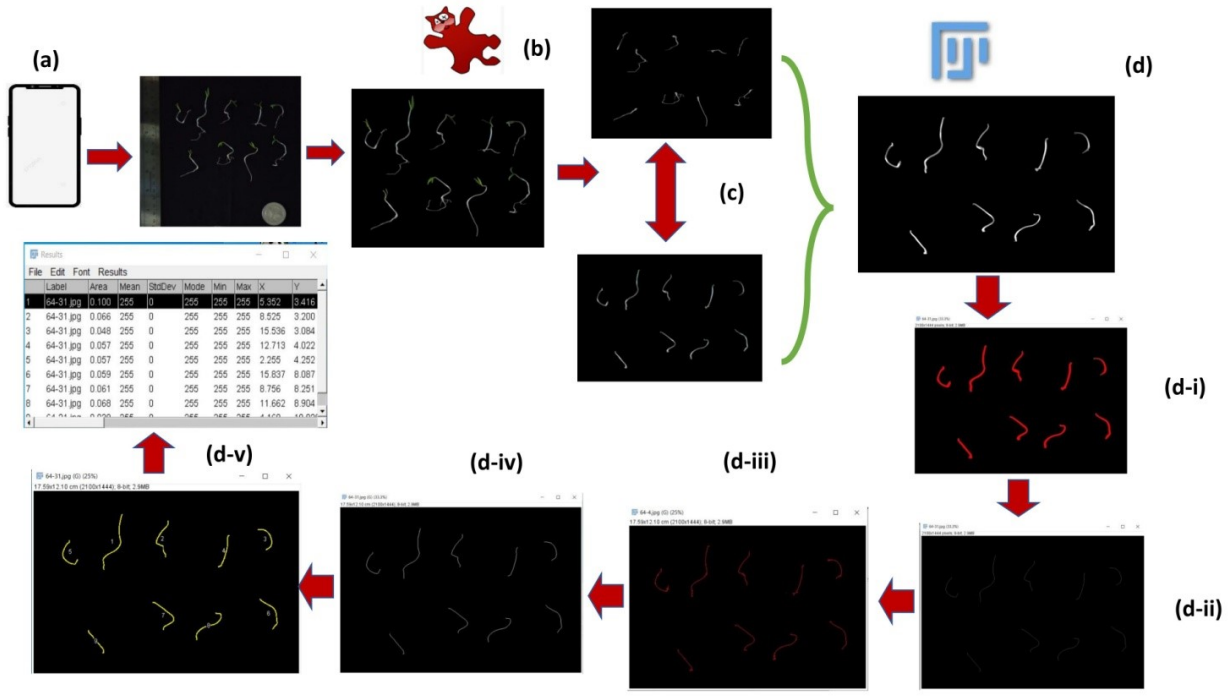

Figure 2. Digital morphometric analysis protocol for radicle and hypocotyl measurements.
(a) Image acquisition: Seedlings were photographed using a smartphone camera against a black background containing a ruler and a coin (scale reference). (b) Image processing: Raw images were optimized by resizing to the original dimensions using *Irfan View* software. (c) Each image was digitally divided into the hypocotyl (stem) and radicle (root) components. (d) *Fiji* software processing: (d-i) threshold adjustment to isolate plant structures and (d-ii) binary conversion with skeletonization. (d-iii) Secondary threshold refinement. (d-iv) Automated measurement of hypocotyl and radicle lengths. (d-v) Final output.

the germination percentage, ranging from the lowest value (30%) in the control group (without extract) to the highest value (82.22 $\pm$4.01%) when using the beetroot extract (B).

In the same genotype (J) at the highest salinity level (250mM), the plant extracts increased the germination percentage, with the lowest germination rate being 16.67% in the control group and the highest reaching 40.0 $\pm$ 6.01% when using the moringa flower extract. The ESI values for all extracts at this salinity level were positive, recording 86.62%, 19.98%, 139.95%, 59.99%, and 113.32% for beetroot (B), carrot (C), moringa flowers (MF), moringa leaves (ML), and prickly pear fruit (O), respectively (Table 4).

For the *Haimi* (H) genotype under a salinity level of 150 mM, all plant extracts showed a significant effect on germination percentage, with results ranging from the lowest value of 76.67 $\pm$ 8.82% in the control group (without extract) to the highest germination rate of 96.67 $\pm$ 1.67% when using the moringa flower extract (MF).

The plant extracts demonstrated their ability to mitigate salinity stress, as indicated by positive ESI values of 21.73% for beetroot (B), 13.04% for carrot (C), 26.09% for moringa flowers (MF), 8.7% for moringa leaves (ML), and 14.49% for prickly pear fruit (O).

When the salinity level was increased to 250 mM in the *Haimi* (*H*) genotype, the plant extracts significantly improved the germination percentage compared to the control group. The extraction efficiency coefficients were 90.18%, 110.21%, 60.15%, 66.85%, and 73.51% for beetroot (B), carrot (C), moringa flowers (MF), moringa leaves (ML), and prickly pear fruits (O), respectively.

In the *Jawfi3* (*V3*) genotype, the effects of the plant extracts were limited, except for beetroot (B), carrot (C), and moringa flower (MF) extracts, which had positive efficiency coefficients. The remaining extracts yielded negative values at salinities below 150 mM. Under the 250 mM salinity stress, the plant extracts did not show a significant effect, except for the moringa leaf extract, which achieved the highest reduction rate of 44.4% compared to the control group.

3.2. Mean germination time (MGT):

The results demonstrated that all independent factors (genotypes, salinity levels, extracts, and extract concentrations) significantly affected the mean germination time (in days). The effects of genotype and salinity level were significant at a probability level of less than $P < 0.001$, whereas the effects of extract and extract concentration were significant at a probability level of less than $P < 0.01$ (Table 3).

Regarding the two-way interactions, the interactions G$\times$N (genotypes $\times$ salinity) and E$\times$C (extract $\times$ concen-

Table 3. Probability values (*F*-values) for the effects of the single factors and their interaction, including the means of the single factors [Genotypes (G), NaCl Levels (N), Plant Extracts (E) and Extract Concentrations (C)]

Source of variation	GrP	MGT	MGR	GSC	CVG	HL	RL
Genotypes (G)	***	***	***	***	***	ns	***
J	59.20 ± 2.35^{b}	14.17 ± 0.19^{a}	0.073 ± 0.001^{b}	7.29 ± 0.11^{b}	22.75 ± 0.59^{b}	1.90 ±0.06	1.83 ±0.06^{c}
H	79.88 ± 1.72^{a}	11.52 ± 0.22^{b}	0.092 ± 0.002^{a}	9.19 ± 0.18^{a}	27.56 ± 0.72^{a}	2.04 ±0.05	2.37 ±0.07^{a}
V3	59.07 ± 1.67^{b}	14.21 ± 0.16^{a}	0.072 ± 0.001^{b}	7.18 ± 0.08^{b}	22.66 ± 0.51^{b}	1.99 ±0.06	2.05 ±0.08^{b}
NaCl (N)	***	***	***	***	***	***	***
0	85.56 ± 1.26^{a}	11.77 ± 0.21^{c}	0.09 ± 0.002^{a}	8.97 ± 0.17^{a}	27.71 ± 0.66^{a}	2.81 ±0.04^{a}	2.91 ±0.07^{a}
150	69.26 ± 1.69^{b}	13.01 ± 0.18^{b}	0.08 ± 0.001^{b}	7.93 ± 0.11^{b}	24.07 ± 0.57^{b}	1.80 ±0.03^{b}	1.92 ±0.05^{b}
250	43.33 ± 1.73^{c}	15.12 ± 0.17^{a}	0.07 ± 0.001^{c}	6.75 ± 0.08^{c}	21.19 ± 0.57^{c}	1.41 ±0.05^{c}	1.43 ±0.06^{c}
Extract (E)	***	**	**	**	**	**	***
B	72.22 ± 2.74^{a}	13.2 ±0.28^{b}	0.079 ± 0.002^{a}	7.91 ± 0.20^{a}	24.27 ± 0.75^{ab}	2.13 ±0.08^{a}	2.24 ±0.10^{a}
C	66.67 ±2.85 ^{b}	13.96 ± 0.27^{a}	0.07 ± 0.002^{b}	7.40 ± 0.15^{b}	22.23 ± 0.86^{b}	2.06 ±0.08^{ab}	2.32 ±0.12^{a}
MF	69.51 ± 2.74^{ab}	13.39 ±0.34^{b}	0.08 ± 0.002^{a}	7.92 ± 0.24^{a}	24.16 ± 1.01^{ab}	1.89 ±0.10^{c}	1.98 ±0.11^{b}
ML	60.86 ±3.02^{c}	13.37 ± 0.33^{b}	0.08 ± 0.002^{a}	7.92 ± 0.24^{a}	25.82 ± 0.98^{a}	1.97 ±0.07^{bc}	1.82 ±0.09^{b}
O	67.78 ±2.68^{ab}	13.12 ±0.29^{b}	0.08 ±0.002^{a}	7.96 ± 0.19^{a}	25.91 ± 0.89^{a}	2.02 ±0.08^{ab}	1.99 ±0.10^{b}
NO	59.26 ± 3.39^{c}	12.77 ±0.31^{c}	0.08 ± 0.002^{a}	8.20 ± 0.20^{a}	23.55 ±0.89^{b}	1.94 ±0.06^{bc}	2.15 ±0.10^{ab}
Concentration (C)	ns	**	**	**	ns	0.052	ns
C1	65.37 ±0.07^{a}	13.03 ±0.22^{b}	0.081 ± 0.002^{a}	8.06 ± 0.15^{a}	24.69 ± 0.28^{a}	2.04 ±0.07	2.15 ±0.08
C2	67.53 ± 2.05^{a}	13.56 ± 0.22^{a}	0.077 ±0.001^{b}	7.73 ±0.15^{b}	23.98 ± 0.65^{a}	1.96 ±0.07	2.11 ±0.09
C3	65.25 ± 2.13^{a}	13.31 ± 0.22^{ab}	0.079 ± 0.001^{ab}	7.87 ±0.15^{ab}	24.29 ±0.59^{a}	2.07 ±0.07	2.10 ±0.09
G × N	***	***	***	***	***	ns	***
G × E	**	ns	ns	ns	ns	***	***
N × E	*	**	**	**	**	***	***
G × C	ns	ns	ns	ns	**	ns	ns
N × C	ns	ns	ns	ns	ns	**	ns
E × C	ns	*	**	**	ns	ns	ns
G ×N ×E	*	ns	ns	ns	**	***	***
G × N × C	ns	ns	ns	ns	ns	ns	ns
G ×E × C	ns	ns	*	*	ns	ns	**
N × E × C	ns	*	*	*	ns	ns	ns
G × N × E × C	ns	ns	ns	ns	ns	*	ns

GrP (%) Total germination percentage, MGT (day) Germination time, MGR (seed/day) Relative germination rate, GSC (%) Germination speed coefficient, CVG (%) Germination variation coefficient, HL (cm) Hypocotyl Length**,** RL (cm) Radicle Length**. Significant. Codes**: '***' 0.001 '**' 0.01 '*' 0.05 and ns = not significant **. Genotypes**: *Haimi* (*H*), *Hajjah* (*J*), and *Jawfi3* (*V3*). **Concentrations:** C1, C2, and C3 represent low, medium, and high concentrations, respectively**.**

trations) showed significant effects. Among the three-way interactions, the interaction between salinity, extracts, and extract concentrations (E×N×C) demonstrated a significant effect at a probability level of less than $P <$ 0.05. However, the three-way interaction involving genotypes did not show any significant effect, indicating that these genotypes responded similarly to plant extracts during the germination stage. However, the four-way in-

Table 4: Means of the three-way interaction between genotypes (G), salinity (N), and plant extracts (E) (G × N × E), and plant extracts (E) (G × N× E), and extract efficiency coefficients (%) (ESI) for the total germination percentage (GrP%)

G	NaCl (mM)	NO	B		C		MF		ML		O	
			M ±ES	ESI%	M ±ES	SEI%	M ±ES	ESI%	M ±ES	SEI%	M ±ES	ESI%
	0	93.33 $\pm 3.33^{a-c}$	90.00 $\pm 4.72^{a-c}$	-3.57	86.7 $\pm 3.33^{a-e}$	-7.1	85.56 $\pm 2.94^{a-e}$	-8.33	81.11 $\pm 6.11^{a-h}$	-13.09	95.56 $\pm 1.76^{ab}$	2.39
J	150	30.0 $\pm 0.00^{s-v}$	82.22 $\pm 4.01^{a-g}$	174.07	48.9 $\pm 5.39^{m-r}$	63	68.89 $\pm 2.61^{d-l}$	129.63	64.44 $\pm 3.38^{f-n}$	114.8	68.89 $\pm$ 5.64^{d-l}	129.63
	250	16.67 $\pm 8.82^{v}$	31.11 $\pm 6.76^{r-v}$	86.62	20.0 $\pm 4.08^{uv}$	19.98	40.00 $\pm 6.01^{o-t}$	139.95	26.67 $\pm 4.08^{t-v}$	59.99	35.56 $\pm 5.80^{p-u}$	113.32
	0	100.0 $\pm 0.00^{a}$	97.78 $\pm 1.47^{a}$	-2.22	95.56 $\pm 1.76^{ab}$	-4.44	98.89 $\pm 1.11^{a}$	-1.11	91.11 $\pm 3.51^{a-c}$	-8.89	96.67 $\pm 1.67^{a}$	-3.33
H	150	76.67 $\pm 8.82^{b-i}$	93.33 $\pm 2.89^{a-c}$	21.73	86.67 $\pm 2.89^{a-e}$	13.04	96.67 $\pm 1.67^{a}$	26.09	83.33 $\pm 4.41^{a-f}$	8.69	87.78 $\pm 3.24^{a-d}$	14.49
	250	33.3 $\pm 12.02^{q-v}$	63.33 $\pm 3.33^{g-n}$	90.18	70.0 $\pm$ 5.27^{d-k}	110.21	53.33 $\pm 6.67^{k-p}$	60.15	55.56 $\pm 4.12^{j-o}$	66.85	57.78 $\pm 3.64^{i-o}$	73.51
	0	76.7 $\pm 3.33^{b-i}$	81.11 $\pm 3.51^{a-h}$	5.75	74.4 $\pm 5.03^{c-j}$	-3	67.78 $\pm 7.41^{e-m}$	-11.63	65.56 $\pm 5.56^{f-n}$	-14.52	62.22 $\pm 5.72^{h-n}$	-18.88
V3	150	56.7 $\pm 12.02^{j-o}$	63.33 $\pm 5.00^{g-n}$	11.69	67.8 $\pm 5.72^{e-m}$	19.58	63.33 $\pm 5.77^{g-n}$	11.69	52.22 $\pm 8.63^{k-p}$	-7.9	55.56 $\pm 4.75^{j-o}$	-2.01
	250	50.0 $\pm$ 15.28^{l-q}	47.78 $\pm 2.21^{n-s}$	-4.44	50.0 $\pm 4.41^{l-q}$	0	51.11 $\pm 8.24^{k-q}$	2.22	27.78 $\pm 8.46^{t-v}$	-44.44	50.00 $\pm 6.24^{l-q}$	0

Means ± standard error. Means sharing the same Latin letters are not significantly different according to Duncan's Multiple Range Test (MRDT) at a significance level of less than 0.05. **Genotypes**:*Haimi* (***H***), *Jawfi3* (***V3***), and *Hajjah* (***J***). **Plant Extracts**: **NO:** Control, **B**:Beetroot, **C**: Carrot, MF:Moringa flower,**ML**:Moringa leaf, and **O**: Prickly pear fruit. **%SEI: Extract Efficiency Coefficient (%) in mitigating the effects of salinity stress. Negative values indicate the extent** of reduction, and positive values indicate the extent of improvement compared to the control treatment (NO) at the same salinity level and for the same genotype

Table 5: The three-way interaction means between salinity, plant extracts, and their concentrations (N × E × C), and the efficiency values of the extracts (ESI) % for the Mean Germination Time (MGT) trait (in days).

NaCl (mM)	Con	NO	B		C		MF		ML		O	
			M ±ES	ESI%	M ±ES	ESI%	M ±ES	ESI%	M ±ES	ESI%	M ±ES	ESI%
	C1	11.32	12.53 $\pm$ 0.84 $^{h-n}$	10.69	12.15 $\pm 0.59^{h-o}$	7.33	10.14 $\pm 0.76^{p}$	-10.42	11.23 $\pm 0.84^{m-p}$	-0.8	10.73 $\pm 0.85^{n-p}$	-5.21
0	C2	±	11.83 $\pm 1.08^{i-p}$	4.51	13.31 $\pm 0.93^{e-k}$	17.58	11.46 $\pm 0.99^{e-l}$	1.24	12.48 $\pm 1.05^{h-o}$	10.25	12.14 $\pm 0.99^{h-o}$	7.24
	C3	0.98^{l-p}	11.71 $\pm$ 0.87 $^{j-p}$	3.45	12.88 $\pm 0.59^{f-m}$	13.78	11.5 $\pm 0.79^{k-p}$	1.59	11.23 $\pm 1.00^{m-p}$	-0.8	12.52 $\pm 0.84^{h-n}$	10.6
	C1	13.36	13.72 $\pm 0.4^{e-i}$	2.69	13.33 $\pm 0.76^{e-k}$	-0.22	12.88 $\pm 0.74^{f-m}$	-3.59	12.49 $\pm 0.51^{h-n}$	-7.19	10.62 $\pm 0.45^{op}$	-20.51
150	C2	±	12.69 $\pm 0.81^{g-m}$	-5.01	13.23 $\pm 0.56^{e-l}$	-0.97	13.15 $\pm 0.61^{e-l}$	-1.57	13.89 $\pm$ 0.77^{d-h}	3.97	12.89 $\pm 0.49^{f-m}$	-3.52
	C3	1.02^{e-k}	12.67 $\pm 1.04^{g-m}$	-5.16	13.74 $\pm 0.75^{e-i}$	2.84	13.36 $\pm 0.45^{e-k}$	0	11.91 $\pm$ 0.93^{i-p}	-10.85	13.29 $\pm 0.52^{e-k}$	-0.52
	C1	13.52	13.26 $\pm 0.43^{e-k}$	-1.92	15.04 $\pm 0.94^{a-e}$	11.24	16.08 $\pm 0.86^{a-c}$	18.93	16.66 $\pm 0.58^{a}$	23.22	14.62 $\pm 0.69^{b-f}$	8.14
250	C2	±	14.47 $\pm 0.57^{c-g}$	7.03	15.78 $\pm 0.64^{a-c}$	16.72	16.33 $\pm 0.83^{ab}$	20.78	15.98 $\pm 0.36^{a-c}$	18.2	16.02 $\pm 0.49^{a-c}$	18.49
	C3	0.63^{e-j}	15.89 $\pm 0.48^{a-c}$	17.53	15.87 $\pm 0.79^{a-c}$	17.38	15.61 $\pm 0.74^{a-d}$	15.46	14.46 $\pm$ 0.99^{c-g}	6.95	14.85 $\pm 0.56^{a-e}$	9.84

Means ± standard error. This means sharing the same Latin letters is not significantly different according to Duncan's Multiple Range Test (MRDT) at a significance level of less than 0.05. **Concentration**: C1, C2, and C3 represent low, medium, and high concentrations respectively. **Plant Extracts: NO:** Control, **B**: Beetroot, **C**: Carrot, **MF**: Moringa flower, **ML**: Moringa leaf, and **O**: Prickly pear fruit. **%SEI: Extract** Efficiency Coefficient (%) in mitigating the effects of salinity stress. Negative values indicate the extent of reduction, and positive values indicate the extent of improvement compared to the control treatment (NO) at the same salinity level and for the same genotype.

teraction (between all factors) was not significant (Table 3).

Table 5 illustrates the effect of the three-way interaction between salinity, extracts, and extract concentrations (N×E×C) on mean germination time (days).

The MGT increased significantly with higher salinity levels, from 11.32 ± 0.98 days under non-saline conditions to 13.52 ± 0.63 days at a salinity level of 250 mM.

However, under non-saline conditions (control treatment), the plant extracts did not significantly affect the mean germination time (MGT). Nevertheless, the use of 10 % moringa flower extract resulted in a non-significant reduction of approximately 10.42%.

At a salinity level of 150 mM, all extracts reduced germination time. According to the ESI values, this reduction ranged from 0.00% when using moringa flower extract at a 50% concentration to 20.51% when using prickly pear fruit extract at a 10% concentration, compared to the control treatment. However, only the reduction achieved with 10% prickly pear fruit extract was statistically significant.

When plant extracts were used at a salinity level of 250 mM, all extracts at the three concentrations significantly prolonged germination time, except for the 10% beetroot extract, which did not significantly reduce germination time by 1.92%. The highest germination time was observed, with no significant difference compared to concentration C3 (Table 3). At the same time, among the genotypes, the prolongation of germination time under 250 mM salinity was 23.22%, achieved using 10% moringa leaf extract (ML). The results indicated that the *Hajjah* (*J*) and *Jawfi3*(*V3*) genotypes exhibited the longest germination times and did not differ significantly from each other. By contrast, the *Haimi* (*H*) genotype differed significantly and achieved the shortest germination time (Table 3).

3.3. Germination speed coefficient (GSC):

The effects of genotypes and salinity levels were significant at a statistical level of less than $P < 0.001$, whereas the effects of extracts and their concentrations were significant at a level of less than $P<0.01$ on the germination speed coefficient (%) (Table 3).

As shown in Table 3, genotype textitHaimi (textitH) achieved the highest germination rate of 9.19%. All plant extracts, except carrot extract, resulted in higher germination speeds. The concentration C1 recorded the highest relative germination and did not differ significantly from C3 (Table 3).

Regarding two-way interactions, the analysis indicated that the interactions between genotypes and salinity (G×N), between extracts and salinity (N×E), and between extracts and their concentrations (E×C) had significant effects at a probability level of less than $P <$

Table 6: The three-way interaction means between salinity, plant extracts, and their concentrations (N × E × C), and the efficiency values of the extracts (ESI) % for the Germination Speed Coefficient (GSC) trait (%).

NaCl (mM)	Con	NO	B		C		MF		ML		O	
			M ±ES	ESI%	M ±ES	ESI%	M ±ES	ESI%	M ±ES	ESI%	M± ES	ESI%
0	C1	9.31 ± 0.72 $^{b-d}$	8.28s ±0.56^{d-m}	-11.06	8.43 ±0.51^{c-l}	-9.45	10.41 ±0.92^{a}	11.82	9.34 ±0.73^{a-d}	0.32	9.83 ± 0.81^{ab}	5.59
	C2		9.05 ±0.82^{b-g}	-2.79	7.86 ±0.61^{h-p}	-15.57	9.3 ±0.87^{b-e}	-0.11	8.58 ±0.86^{c-k}	-7.84	8.72 ± 0.75^{b-j}	-6.34
	C3		8.99 ±0.77^{b-h}	-3.44	7.90 ±0.36^{g-p}	-15.15	9.10 ±0.75^{b-f}	-2.26	9.53 ±0.89^{a-c}	2.36	8.28 ±0.56^{d-m}	-11.06
150	C1	7.83 ± 0.57 $^{h-p}$	7.33 ±0.2^{l-u}	-6.39	7.70 ±0.44^{i-q}	-1.66	7.99 ±0.51^{f-p}	2.04	8.14 ±0.43^{e-o}	3.96	9.54 ±0.39^{a-c}	21.84
	C2		8.19 ±0.61^{d-n}	4.6	7.68 ±0.36^{j-q}	-1.92	7.74 ±0.36^{i-p}	-1.15	7.37 ±0.39^{l-u}	-5.87	7.85 ±0.3^{h-p}	0.26
	C3		8.38 ±0.76^{c-l}	7.02	7.44 ±0.38^{k-u}	-4.98	7.55 ±0.26^{j-s}	-3.58	8.87 ±0.77^{b-i}	13.28	7.63 ±0.33^{j-r}	-2.55
250	C1	7.5 ± 0.33 $^{k-t}$	7.61 ±0.28 $^{j-r}$	1.47	6.84 ±0.45^{p-v}	-8.8	6.39 ±0.41^{s-v}	-14.8	6.05 ±0.21^{v}	-19.33	6.97 ±0.33^{o-v}	-7.07
	C2		7.01 ±0.3^{1-v}	-6.53	6.42 ±0.27^{s-v}	-14.4	6.27 ±0.36^{uv}	-16.4	6.28 ±0.15^{uv}	-16.27	6.28 ±0.19^{uv}	-16.27
	C3		6.34 ±0.18^{t-v}	-15.47	6.46 ±0.39^{r-v}	-13.87	6.53 ±0.32^{q-v}	-12.93	7.15 ±0.48^{m-v}	-4.67	6.81 ±0.26^{p-v}	-9.2

Means±standard error. This means sharing the same Latin letters is not significantly different according to Duncan's Multiple Range Test (MRDT) at a significance level of less than 0.05. **Concentration**: C1, C2, and C3 represent low, medium, and high concentrations respectfiency Coefficient(%)in mitigating the effects of salinity stress. Negative values indicate the extent of reduction, and positive values indicate the extent of improvement compared to the control treatment (NO) at the same salinity level and for the same genotype.

0.001. In contrast, the other two-way interactions (N×C, G×C, and G×E) did not show any significant statistical effects.

Table 6 illustrates the significant effects of the three-way interaction between salinity, extracts, and extract concentrations (N×E×C) on the germination speed coefficient. The results showed a significant decrease in germination rate with increasing salinity levels, declining from 9.31 ± 0.72% under non-saline conditions to 7.5 ± 0.33% at a salinity level of 250 mM.

In the control treatment, most extracts did not show any significant effect on the germination speed coefficient, except for the 10% moringa flower extract, which increased germination speed by 11.82%.

At a salinity level of 150 mM, seeds treated with 10% prickly pear fruit extract achieved the highest significant increase in germination speed, reaching 21.84%, compared to seeds grown under the same salinity level without extracts. As for the other extracts, although some achieved slight increases, such The 30

At a salinity level of 250 mM, all extracts reduced the germination speed coefficient. The decrease was significant when using MF, ML, and O extracts, whereas it was not significant for the other extracts. However, the 10% beetroot extract (B) showed a slight increase of 1.47%, which was not statistically significant.

3.4. Mean germination rate (MGR):

The results demonstrated that all independent factors studied (genotypes, salinity levels, extracts, and extract concentrations) had a significant effect on the relative germination rate (seed/day) at a probability level of less than $P < 0.001$ (Tables 3). As shown in Table 3, the *Haimi*(*H*) genotype achieved the highest relative germination, differing significantly from the *Hajjah*(*J*) and *Jawfi3*(*V3*) genotypes. Similarly, the control group, compared to salinity levels and all plant extracts, resulted in higher relative germination, except for carrot extract.

The concentration C1 recorded the highest relative germination and did not differ significantly from that of C3. Regarding the two-way interactions, the interactions between genotypes and salinity (G×N), salinity and extracts (N×E), and extracts and their concentrations (E×C) showed significant effects at a probability level of less than $P < 0.001$. In contrast, the other two-way interactions did not show significant effects.

For the three-way interactions, no significant effects were observed with genotypes, except for the interaction between extracts, extract concentrations, and genotypes (E×C×G), which showed a significant effect at a probability level of $P < 0.05$. Similarly, the interaction between salinity, extract, and extract concentration (E ×C×N) had a significant effect at the same probability level. Conversely, the four-way interaction between genotypes, salinity, extracts, and extract concentrations

Table 7: The three-way interaction means between salinity, plant extracts, and their concentrations (N × E × C), and the efficiency values of the extracts (ESI) % for the Mean Germination Rate (MGR) (seed/day) trait

NaCl (mM)	Con	NO	B		C		MF		ML		O	
			M ±ES	ESI%	M ±ES	ESI%	M ±ES	ESI%	M ±ES	ESI%	M ±ES	ESI%
0	C1	0.093	0.083 ±0.006^{d-m}	-15.8	0.084 ±0.005^{c-l}	-9.68	0.104 ±0.009^{a}	11.83	0.093 ±0.007^{a-d}	0	0.098 ±0.008^{ab}	5.38
	C2	±	0.09 ±0.008^{b-g}	-3.23	0.079 ±0.006^{h-p}	-15.1	0.093 ±0.009^{b-e}	0	0.086 ±0.009^{c-k}	-7.53	0.087 ±0.008^{b-f}	-6.45
	C3	0.007^{b-d}	0.09 ±0.008^{b-h}	-3.23	0.079 ±0.004^{s-p}	-15.1	0.091 ±0.008^{b-f}	-2.15	0.095 ±0.009^{a-c}	2.15	0.083 ±0.006^{d-m}	-10.75
150	C1	0.078	0.073 ±0.002^{l-u}	-6.41	0.077 ±0.004^{i-q}	-1.28	0.08 ±0.005^{f-p}	2.56	0.081 ±0.004^{e-o}	3.85	0.095 ±0.004^{a-c}	21.79
	C2	±	0.082 ±0.006^{d-n}	5.13	0.077 ±0.004^{j-q}	-1.28	0.077 ±0.004^{i-p}	-1.28	0.074 ±0.004^{l-u}	-5.13	0.079 ±0.003^{h-p}	1.28
	C3	0.006^{h-p}	0.084 ±0.008^{c-l}	7.69	0.074 ±0.004^{k-u}	-5.13	0.076 ±0.003^{j-s}	-2.56	0.089 ±0.008^{b-i}	14.1	0.076 ±0.003^{j-r}	-2.56
250	C1	0.075	0.076 ±0.003^{j-r}	1.33	0.068 ±0.004^{p-v}	-9.33	0.064 ±0.004^{s-v}	-14.7	0.061 ±0.002^{v}	-18.67	0.07 ±0.003^{o-v}	-6.67
	C2	±	0.07 ±0.003^{n-v}	-6.67	0.064 ±0.003^{s-v}	-14.7	0.063 ±0.004^{uv}	-16	0.063 ±0.001^{uv}	-16	0.063 ±0.002^{uv}	-16
	C3	0.003^{k-t}	0.063 ±0.002^{l-v}	-16	0.065 ±0.004^{r-v}	-13.3	0.065 ±0.003^{q-v}	-13.3	0.071 ±0.005^{m-v}	-5.33	0.068 ±0.003^{p-v}	-9.33

Means ± standard error. This means sharing the same Latin letters is not significantly different according to Duncan's Multiple Range Test (MRDT) at a significance level of less than 0.05.**Concentration**: C1, C2, and C3 represent low, medium, and high concentrations respectively. **Plant Extracts: NO:** Control, B: Beetroot, C: Carrot, MF: Moringa flower, **ML**: Moringa leaf, and O: Prickly pear fruit. **%SEI: Extract Efficiency Coefficient** (%) in mitigating the effects of salinity stress. Negative values indicate the extent of reduction, and positive values indicate the extent of improvement compared to the control treatment (NO) at the same salinity level and for the same genotype.

(G×N×E×C) did not have any significant effect (Table 3).

Table 7 illustrates the effect of the three-way interaction between extract concentration, salinity, and type of extract (C×N×E) on the relative germination rate (seed/day). The results showed a significant decrease in the relative germination rate with increasing salinity levels, declining from 0.093 ± 0.007 seed/day under non-saline conditions to 0.075 ± 0.003 seed/day at a salinity level of 250 mM.

Under non-saline conditions, the 10% moringa flower extract (MF) showed a significant increase in the relative germination rate by 11.83%, while the 10% prickly pear fruit extract (O) resulted in a non-significant increase of 5.38%. Similarly, the 30% moringa leaf extract (ML) showed a non-significant increase compared to the control treatment. However, the other extracts did not significantly affect the relative germination rate. Conversely, the application of carrot extract at concentrations of 25% and 50% resulted in a significant decrease in the relative germination rate by 15.05% compared to the control.

At a salinity level of 150 mM, 10% prickly pear fruit extract and 30% moringa leaf extract (ML) significantly increased the relative germination rate, with values of approximately 21.79% and 14.10%, respectively. In contrast, the other extracts did not exhibit any significant effects.

At a salinity level of 250mM, a significant decrease in the relative germination rate was observed for all plant extracts, except for the 10% beetroot extract, which increased the relative germination rate by 1.33%. However, these values were not significant, except for the reductions caused by the 10% and 20% moringa leaf extract (ML), which decreased by 18.67% and 16.0%, respectively. Additionally, the 20% moringa flower extract (MF) and 25% prickly pear fruit extract (O) showed a significant reduction of 16% compared to the control.

3.5. Coefficient of velocity of germination (CVG):

The results indicated that the independent factors (genotypes, salinity levels, and extracts) significantly affected the coefficient of variation of germination (CVG) at a probability level of $P<0.001$, except for the effect of extract concentration, which was not statistically significant.

As shown in Table 3, the *Haimi*(*H*) genotype exhibited the highest coefficient of variation in germination. Similarly, the control group and the extracts of moringa leaves and prickly pear fruits resulted in the highest coefficient of variation in germination, with no significant differences among the concentrations.

Most two-way interactions showed significant effects, except for those involving extract concentrations and the interaction between genotypes and extracts. On the other hand, the three-way interactions did

However, the extracts clearly mitigated the impact of salinity stress on the coefficient of variation of germination. For the *Hajjah* (*J*) genotype, the extracts did not have a significant effect on the coefficient of variation of germination for seeds grown at a salinity level of 150 mM NaCl, despite positive increases in the extract efficiency coefficient (SEI) for extracts B, ML, and O, which reached 12.34%, 5.47%, and 18.79%, respectively (Table 8).

In the same genotype, *Hajjah* (*J*) at the highest salinity level (250mM), the plant extracts reduced the coefficient of variation of germination compared to the control group, which recorded the highest coefficient of variation of germination at 28.32 ± 1.76%. The Plant extracts showed significant negative values for the extract efficiency coefficient ESI) under this salinity level, with values of -18.47%, -33.79%, -40.61%, -31.29%, and -26.55% for beetroot (B), carrot (C), moringa flowers (MF), moringa leaves (ML), and prickly pear fruit (O), respectively.

For the *Haimi* (*H*) genotype, which showed a linear decrease in the coefficient of variation of germination with increasing salinity levels, all plant extracts demonstrated their ability mitigated salinity stress and had a significant effect on the coefficient of variation of germination at a salinity level of 150 mM. Positive values for the extract efficiency coefficient (ESI) were recorded, reaching 49.31% for beetroot (B), 72.52% for carrot (C), 90.69% for moringa flowers (MF), 89.75% for moringa leaves (ML), and 104.21% for prickly pear fruit (O).

When the salinity level increased to 250 mM in the *Haimi* (*H*) genotype, the plant extracts showed a clear positive effect on increasing the coefficient of variation of germination compared to the control group. The extract efficiency coefficients were 29.71%, 9.39%, and 8.03% for beetroot (B), carrot (C), and moringa flowers (MF), respectively, in comparison to the control. However, these increases were not significant, except for the moringa leaf extract (ML), which achieved a significant increase of 44.91% compared to the control (without extract under 250 mM), and the prickly pear fruit extract (O), which achieved a significant increase of 49.57% compared to the control.

For the *Jawfi3* (V3) genotype under a salinity level of 150 mM, the plant extracts did not show a significant effect on the coefficient of variation of germination, although some extracts recorded positive values for the extract efficiency coefficient (ESI), reaching 3.02% for carrot (C), 0.78% for moringa flowers (MF), and 8.55% for prickly pear fruit (O). When the salinity level increased to 250 mM for the same genotype, the plant extracts also showed a non-significant effect, with negative values for the extract efficiency coefficient (ESI) for all extracts, except for the moringa leaf extract (ML), which recorded a positive value of 9.75%.

Table 8: Means of the three-way interaction between genotypes (G), salinity (N), and plant extracts (E), and the efficiency values of the extracts (ESI) % for the coefficient of variation of germination (CVG) trait (seed/day).

G	NaCl (mM)	NO	B		C		MF		ML		O	
			M ±ES	ESI%	M ±ES	SEI%	M ±ES	ESI%	M ±ES	SEI%	M ±ES	SEI%
J	0	$0.5^{i-q} \pm 22.24$	$22.62 \pm 1.88^{h-q}$	1.71	$22.06 \pm 1.4^{i-q}$	-0.81	$23.28 \pm 2.3^{h-q}$	4.68	$28.65 \pm 2.06^{b-f}$	28.8	$27.83 \pm 2.71^{c-k}$	25.13
	150	$2.6^{i-q} \pm 21.4$	$24.04 \pm 1.39^{f-p}$	12.3	$20.77 \pm 1.42^{k-q}$	-2.94	$21.28 \pm 1.99^{j-q}$	-0.56	$22.57 \pm 1.33^{h-q}$	5.47	$25.42 \pm 2.65^{e-m}$	18.79
	250	$1.76^{b-k} \pm 28.32$	$23.09 \pm 4.72^{h-q}$	-18.5	$18.75 \pm 5.05^{m-q}$	-33.8	$16.82 \pm 1.87^{o-q}$	-40.6	$19.46 \pm 3.71^{m-q}$	-31.3	$20.80 \pm 2.29^{k-q}$	-26.6
H	0	$1.31^{a} \pm 38.73$	$33.95 \pm 1.73^{a-d}$	-12.3	$31.47 \pm 1.86^{b-f}$	-18.8	$35.04 \pm 3.74^{a-c}$	-9.53	35.22 ± 3.41^{ab}	-9.06	$31.32 \pm 2.61^{b-g}$	-19.1
	150	$1.25^{g} \pm 15.90$	$23.74 \pm 2.3^{g-p}$	49.3	$27.43 \pm 2.93^{d-l}$	72.5	$30.32 \pm 2.42^{b-h}$	90.7	$30.17 \pm 2.48^{b-h}$	89.8	$32.47 \pm 3.22^{a-e}$	104.2
	250	$1.14^{n-q} \pm 17.57$	$22.79 \pm 0.74^{h-q}$	29.7	$19.22 \pm 1.72^{m-q}$	9.39	$18.98 \pm 1.37^{m-q}$	8.03	$25.46 \pm 1.6^{e-m}$	44.9	$26.28 \pm 1.48^{e-m}$	49.57
V3	0	$1.19^{i-q} \pm 21.73$	$24.48 \pm 1.25^{f-o}$	12.7	$19.85 \pm 1.62^{l-q}$	-8.65	$29.40 \pm 3.05^{b-i}$	35.3	$26.52 \pm 2.6^{e-m}$	22	$24.38 \pm 2.79^{f-p}$	12.2
	150	$1.66^{h-q} \pm 23.16$	$23.02 \pm 1.67^{h-q}$	-0.6	$23.86 \pm 2.21^{f-p}$	3.02	$23.34 \pm 2.07^{h-q}$	0.78	$19.18 \pm 2.02^{m-q}$	-17.2	$25.14 \pm 1.82^{e-n}$	8.55
	250	$1.14^{h-q} \pm 22.88$	$20.68 \pm 1.37^{k-q}$	-9.62	16.63 ± 1.88^{pq}	-27.3	$18.99 \pm 2.25^{m-q}$	-17	$25.11 \pm 3.03^{e-n}$	9.75	$19.52 \pm 1.92^{m-q}$	-14.7

Means ± standard error. This means sharing the same Latin letters is not significantly different according to Duncan's Multiple Range Test (MRDT) at a significance level of less than 0.05. **Genotypes**: *Haimi* (*H*), *Jawfi3* (*V3*), and *Hajjah* (*J*). cance level of less than 0.05. **Genotypes**: ***Haimi* (*H*), *Jawfi3* (*V3*) and *Hajjah* (*J*). Plant Extracts: NO: Control, B: Beetroot, C: Carrot, MF: Moringa flower, ML: Moringa leaf, and O: Prickly pear fruit. %SEI: Extract:NO:** Control, **B**: Beetroot, **C**: Carrot, **MF**: Moringa flower, **ML**: Moringa leaf, and **O**: Prickly pear fruit. **%SEI: Extract Efficiency Coefficient** (%) in mitigating the effects of salinity stress. Negative values indicate the extent of reduction, and positive values indicate the extent of improvement compared to the control treatment (NO) at the same salinity level and for the same genotype.

3.6. Radicle Length (RL):

The results demonstrated that individual factors (genotypes, salinity levels, and plant extracts) independently exerted significant effects on mean radicle length, except for extract concentration (Table 3). Significant variations were observed among genotypes, with the *Haimi* genotype (H) exhibiting the maximum radicle length (2.37 ±0.07 cm), while genotype *Hajah* (*J*) showed the lowest length (1.83±0.06 cm). A significant linear reduction in radicle length was observed with increasing salinity. Among the tested extracts,red beet*root* (B,) and carrot root (C) extracts produced significantly greater radicle elongation than the other extracts, while showing no statistical difference from the control treatment. Other extracts demonstrated inferior performance relative to extracts B and C, but remained statistically comparable to the control. The extract concentration had no significant effect.

Analysis of variance revealed significant two-way interactions (G×N, G×E, N×E; $p<0.001$) for all factors except those involving extract concentrations. Higher-order interactions (three-way and four-way) were non-significant, with the notable exception of the genotype × salinity × extract (G×N×E) interaction, which showed highly significant effects ($p<0.001$) on radicle elongation (Table 3),

Table 9 details the G×N×E interaction effects. Under non-saline conditions, genotype *Hajah* (*J*) showed significant radicle elongation when treated with prickly pear(O), *carrot* (C), or *red beetroot.* (B) extracts compared to controls, with maximum stimulation (2.90 ± 0.21 cm) from the carrot extract. No stress mitigation was observed with 150 mM NaCl. However, at 250 mM NaCl, both B and C extracts significantly enhanced radicle growth relative to the untreated (control) group, demonstrating salt stress amelioration. The extraction efficiency index (%ESI) quantified this mitigation, showing 32.99% and 22.68% improvements for extracts B and C, respectively (Table 9).

For *Haimi* (H) under control conditions, *Moringa oleifera* leaf (ML) and flower (MF) extracts significantly inhibited radicle growth, whereas the other extracts (B, C, and O) showed neutral effects. No extract provided significant salinity protection at either 150 mM or 250 mM NaCl.

In genotype *Jowfi 3* (V3), prickly pear (O), and *Moringa* extracts significantly suppressed radicle growth under non-stressed conditions, whereas B and C extracts maintained control-equivalent performance. All extracts conferred significant salinity tolerance at 150 mM NaCl, with the MF extract showing maximum efficacy (58.01% ESI), followed by the B extract (33.40% improvement). At 250 mM NaCl, all extracts significantly alleviated salt stress, with the MF extract demonstrating a remarkable 245.02%

Table 9: Means of the three-way interaction between genotypes (G), salinity (N), and plant extracts (E) (G × N × E), and extract efficiency coefficients (%) (ESI) for the Radicle length (RL, cm)

G	NaCl (mM)	NO	B		C		MF		ML		O	
			M ±ES	ESI%	M ±ES	SEI%	M ±ES	ESI%	M ±ES	SEI%	M ±ES	ESI%
J	0	2.11 ±0.05^{d-k}	2.29 ±0.21^{c-i}	**8.96**	2.90 ±0.21^{bc}	**37.49**	1.92 ±0.15^{f-n}	**-9.00**	2.04 ±0.21^{e-m}	**-3.27**	2.25 ±0.22^{c-j}	**6.40**
	150	1.98 ±0.13^{e-n}	2.07 ±0.16^{e-l}	**4.55**	1.96 ±0.16^{e-n}	**-1.06**	1.51 ±0.22^{j-p}	**-23.80**	1.86 ±0.15^{g-n}	**-6.27**	1.73 ±0.11^{h-n}	**-12.53**
	250	1.46 ±0.25^{k-q}	1.95 ±0.51^{e-n}	**32.99**	1.79 ±0.35^{g-n}	**22.68**	0.39 ±0.23^{r}	**-73.36**	1.23 ±0.17^{n-q}	**-15.92**	1.54 ±0.19^{j-p}	**5.05**
H	0	3.71 ±0.21^{a}	3.78 ±0.21^{a}	**1.92**	3.78 ±0.34^{a}	**1.94**	2.69 ±0.20^{b-e}	**-27.46**	2.87 ±0.37^{bc}	**-22.61**	3.56 ±0.20^{a}	**-3.94**
	150	2.13 ±0.17^{d-k}	2.24 ±0.12^{c-j}	**5.07**	2.44 ±0.34^{c-h}	**14.45**	2.04 ±0.13^{e-m}	**-4.36**	1.98 ±0.16^{e-n}	**-6.95**	1.95 ±0.14^{e-n}	**-8.35**
	250	1.96 ±0.11^{e-n}	1.34 ±0.17^{l-q}	**-31.62**	1.69 ±0.11^{h-o}	**-13.77**	1.72 ±0.22^{h-n}	**-12.34**	1.29 ±0.10^{m-q}	**-34.17**	1.49 ±0.14^{l-q}	**-23.92**
V3	0	3.71 ±0.06^{a}	3.29 ±0.17^{ab}	**-11.03**	3.57 ±0.18^{a}	**-3.83**	2.51 ±0.35^{c-s}	**-32.23**	2.49 ±0.29^{c-s}	**-32.66**	2.81 ±0.27^{b-d}	**-24.27**
	150	1.491 ±0.06^{j-q}	1.989 ±0.17^{e-n}	**33.40**	1.88 ±0.23^{g-n}	**26.36**	2.36 ±0.30^{c-h}	**58.01**	1.33 ±0.21^{l-q}	**-10.87**	1.58 ±0.25^{j-p}	**5.84**
	250	0.773 ±0.19^{qr}	1.23 ±0.20^{n-q}	**59.12**	0.90 ±0.16^{pr}	**16.95**	2.67 ±0.46^{p-f}	**245.02**	1.30 ±0.23^{m-q}	**68.31**	0.97 ±0.32^{o-r}	**25.23**

Means ± standard error. This means sharing the same Latin letters is not significantly different according to Duncan's Multiple Range Test (MRDT) at a significance level of less than 0.05.**Genotypes**: *Haimi* (*H*), *Jawfi3* (*V3*), and *Hajjah* (*J*). **Plant Extracts**: **NO:** Control, **B**: Beetroot, **C**:Carrot, **MF**: **rot, MF: Moringa flower, ML: Moringa leaf, and O: Prickly pear fruit.%SEI: Extract Efficiency Coefficient (%) in mitigating the effects of salin-**ity stress. Negative values indicate the extent of reduction, and positive values indicate the extent of improvement compared to the control treatment (NO) at the same salinity level and for the same genotype.

enhancement in radicle length relative to stressed controls.

3.7. Hypocotyl Length (HL):

Table 3 demonstrates that among the individual factors examined, only salinity levels and plant extracts exhibited statistically significant effects on mean hypocotyl length ($P<0.05$). A dose-dependent linear reduction in hypocotyl length was observed with increasing salinity, decreasing from 2.81 ± 0.04 cm in the control to 1.41 ± 0.05 cm at 250 mM NaCl concentration. Among the tested extracts, beetroot (B) extract produced the maximum hypocotyl elongation (2.13 ± 0.08 cm), showing significant enhancement compared to the control ($P<0.05$) while remaining statistically comparable to both prickly pear (O) and carrot (C) extracts. The remaining extracts yielded lower values that did not differ significantly from those of the control.

Analysis of variance revealed significant two-way interaction effects ($P<0.05$) for all factor combinations involving plant extracts, except for the extract × concentration interaction. Notably, the salinity × extract concentration interaction demonstrated significant effects ($P<0.05$), whereas the other two-way interactions showed no statistical significance. Among higher-order interactions, only the genotype × salinity × extract (G×N×E) three-way interaction significantly influenced hypocotyl length ($P<0.001$, Table 3), with no detectable four-way interaction effects.

Genotype-specific responses to the three-way interaction (G×N×E) are detailed in Table 10. For the *Hajjah* (J) genotype under non-saline conditions, treatment with prickly pear (O), carrot (C), and beetroot (B) extracts resulted in significant hypocotyl elongation relative to the controls ($P<0.05$), with the O extract producing maximal growth (3.18 ±0.13 cm; 17.93% increase). In contrast, Moringa leaf (ML) and flower (MF) extracts showed no significant inhibitory effects. At moderate salinity (150 mM NaCl), none of the extracts demonstrated significant stress mitigation. However, at high salinity (250 mM NaCl), the MF extract exhibited remarkable stress-alleviating properties, as evidenced by a 170.66% increase in hypocotyl length relative to that of the salt-stressed controls (Table 10).

The *Haimi* (H) genotype displayed distinct responses, with all extracts except ML and MF significantly promoting hypocotyl growth under non-saline conditions ($P<0.05$). However, this positive effect was completely abolished at both 150 mM and 250 mM salinity levels, with no extract showing significant protective effects against salt stress.

For the Jawfi 3 (V3) genotype under control conditions, all extracts except carrot extract significantly enhanced hypocotyl elongation ($P<0.05$), with the MF extract producing the greatest response (3.04 cm; 27.88% increase). Notably, while no extracts showed significant

Table 10: Means of the three-way interaction between genotypes (G), salinity (N), and plant extracts (E) (G × N × E), and extract efficiency coefficients (%) (ESI) for the Hypocotyl Length (HL, cm)

G	NaCl (mM)	NO	B		C		MF		ML		O	
			M ± ES	ESI%	M ± ES	SEI%	M ± ES	ESI%	M ± ES	SEI%	M ± ES	ESI%
J	0	2.7 ±0.08^{b-f}	3.08 ±0.15^{ab}	14.00	3.12 ±0.11^{ab}	15.56	2.52 ±0.13^{e-g}	-6.56	2.58 ±0.13^{d-g}	-4.59	3.18 ±0.13^{a}	17.93
	150	1.73 ±0.11^{l-o}	1.80 ±0.08^{j-o}	4.40	1.71 ±0.09^{j-o}	-0.98	1.88 ±0.13^{j-n}	8.57	1.69 ±0.06^{j-o}	-2.37	1.68 ±0.08^{k-o}	-3.01
	250	0.93 ±0.20^{rs}	1.13 ±0.21^{p-s}	21.41	1.05 ±0.17^{q-s}	12.85	2.53 ±0.29^{e-g}	170.66	1.36 ±0.16^{o-r}	45.50	1.06 ±0.20^{q-s}	13.49
H	0	2.565 ±0.09^{d-g}	3.08 ±0.16^{ab}	20.23	3.02 ±0.12^{a-d}	17.58	2.59 ±0.15^{c-g}	1.29	2.49 ±0.14^{e-h}	-3.00	3.00 ±0.11^{a-d}	16.96
	150	2.144 ±0.07^{g-k}	2.16 ±0.13^{g-j}	0.51	2.07 ±0.12^{h-l}	-3.50	1.67 ±0.05^{k-o}	21.92	1.59 ±0.04^{l-p}	-25.79	1.82 ±0.09^{j-o}	-15.35
	250	1.549 ±0.04^{m-p}	1.49 ±0.07^{m-q}	-3.49	1.49 ±0.05^{m-q}	-3.29	0.89 ±0.19^{s}	42.16	1.56 ±0.06^{m-p}	0.90	1.55 ±0.07^{m-p}	0.26
V3	0	2.378 ±0.14^{f-i}	3.01 ±0.16^{a-d}	26.75	2.69 ±0.14^{b-f}	12.99	3.04 ±0.27^{a-c}	27.88	2.86 ±0.20^{a-e}	20.14	2.59 ±0.18^{c-g}	9.00
	150	1.935 ±0.12^{j-n}	1.92 ±0.15^{j-n}	-0.72	1.75 ±0.09^{j-o}	-9.82	1.15 ±0.25^{p-s}	40.72	1.98 ±0.13^{i-m}	2.22	1.74 ±0.19^{j-o}	-9.92
	250	1.565 ±0.04^{m-p}	1.48 ±0.10^{n-q}	-5.50	1.67 ±0.08^{k-o}	6.77	0.75 ±0.31^{s}	52.20	1.66 ±0.29^{l-o}	6.13	1.59 ±0.17^{l-p}	1.79

Means ± standard error. This means sharing the same Latin letters is not significantly different according to Duncan's Multiple Range Test (MRDT) at a significance level of less than 0.05. **Genotypes**: *Haimi* (**H**), *Jawfi3* (**V3**), and *Hajjah* (**J**). **Plant Extracts**: **NO:** Control, **B**:Beetroot, **C**: Carrot, **MF**: Moringa flower, **ML**: Moringa leaf, and **O**: Prickly pear fruit. **SEI%: Extract Efficiency Coefficient** (%) In mitigating the effects of salinity stress. Negative values indicate the extent of reduction, and positive values indicate the extent of improvement compared to the control control treatment (NO) at the same salinity level and for the same genotype.

positive effects on salt stress mitigation, the MF extract consistently demonstrated significant negative effects on hypocotyl growth across all salinity levels ($P<0.05$).

4. DISCUSSION

Under natural conditions, the slow germination of chili pepper seeds presents a significant challenge, requiring an extended period and consequently delaying their readiness for cultivation.

Salinity further exacerbates this issue by not only reducing germination speed but also significantly lowering the overall germination percentage. Salinity is a major abiotic stress that significantly hampers seed germination and plant growth, ultimately reducing agricultural productivity [21, 29, 32, 33].

The current study highlights the deleterious impact of increasing salinity levels on germination parameters in the control treatments, confirming a clear linear decline in germination metrics. These findings are consistent with previous reports by Al-madhagi and Arraf, [21] and Loganayaki *et al* [31]. This reinforces the notion that elevated sodium chloride concentrations negatively influence sugar metabolism, organic acid dynamics, and phenolic compound accumulation [30]. These physiological imbalances contribute to reduced germination percentage, delayed germination onset, and diminished seedling biomass [63].

Among the various mitigation strategies explored to counteract salinity stress, the use of plant extracts has gained considerable attention because of their natural origin, environmental safety, and potential to replace or reduce synthetic inputs [32, 47, 48, 64].

However, our findings also revealed a promising intervention: the application of plant extracts significantly attenuated the adverse effects of salinity, particularly at moderate salinity levels (150 mM). These extracts effectively disrupted the linear decline in germination parameters observed in the untreated controls, suggesting their potential as biostimulant agents. The observed genotype-dependent responses further underscore the importance of the crop genetic background in modulating extract efficacy, which has crucial implications for precision agriculture and cultivar selection in stress-prone environments.

The tolerance of chili pepper genotypes and cultivars to salinity varies, as previously demonstrated by Al-Madhagi and Arraf [21], who classified Yemeni chili pepper genotypes into four groups based on their sensitivity to salinity. In this study, salt-sensitive chili pepper genotypes were selected to evaluate the potential of plant extracts in mitigating the adverse effects of salinity stress.

Plant extracts are rich in bioactive compounds that act as natural biostimulants, enhancing seed germination and promoting plant growth. A study by Arraf and

Al-madhagi[54], demonstrated that the application of beetroot extract, a mixture of Moringa flower and seed extracts, and prickly pear extract resulted in significant improvements in shoot and root growth, as well as total biomass accumulation in chili pepper seedlings. These findings underscore the potential of plant extracts as effective agents for enhancing growth performance and biomass yield. Moreover, they lay a solid foundation for further investigation into the role of these genes in alleviating abiotic stresses, particularly salinity, and their broader implications for sustainable agricultural practices.

The mechanistic insights from our study suggest distinct modes of action among the tested extracts. Beetroot extract, characterized by its high electrical conductivity (EC = 2.7 mS/cm, Table 1), may likely operate through ion competition, mitigating sodium toxicity by providing alternative cations that compete with Na? uptake. In contrast, prickly pear extract, despite its relatively low EC (1.5 mS/cm, Table 1), may exhibit strong performance, indicating a greater reliance on non-ionic mechanisms, such as antioxidant activity and osmoprotection. This divergence highlights the multifaceted nature of plant extract efficacy and the need for tailored applications based on the extract composition and crop genotype.

The effectiveness of plant extracts in mitigating the effects of salinity was evident, as they enhanced the germination percentage and germination rate while reducing the mean germination time under salinity stress conditions. These results highlight the significance of plant extracts in alleviating salinity stress.

The genotype-specific performance of extracts, particularly those grown under 150 mM NaCl conditions, is of significant horticultural interest. While not all treatments showed statistically significant improvements, several extract-genotype combinations, such as beetroot across all genotypes and carrot or moringa flower extracts in *Haimi* (*H*) (*Capsicum frutescens*) and *Jawfi3*(*V3*) (*Capsicum annuum*), produced germination outcomes comparable to non-saline controls. Therefore, it can be concluded that these extracts mitigated the effects of salinity up to a concentration of 150 mM and reduced the impact of salinity at a higher sodium chloride concentration (250 mM).

The ion competition mechanism proposed for beetroot extract is particularly compelling given our measured EC values, suggesting that the high ion content in extracts may indeed compete with Na+ uptake, although this requires direct ion flux measurements for confirmation. Previous studies have suggested that beetroot, known for its betaine content, has been shown to effectively reduces salinity stress [65]. The high electrical conductivity (EC) of the beetroot extract (2.7 mS/cm, Table 1) suggests a substantial concentration of dissolved ions. This is relevant in saline conditions, where competitive ion interactions may help mitigate sodium (Na^+) toxicity.

Similarly, carrot extract, a source of beta-carotene, has been shown to have salinity-mitigating properties [66]. Although the current study did not directly measure betaine or beta-carotene levels, the observed improvements under salinity stress may align with these proposed mechanisms.

The novel identification of prickly pear extract as an effective salinity mitigator, particularly for the *Haimi*(*H*) genotype, is a key contribution of this study. Although limited prior work has examined this extract in the context of salinity stress, its known composition, as identified in literature reports, including proline, vitamin C, and stress-responsive amino acids, supports its efficacy [67]. These compounds likely enhance membrane stability, reduce oxidative damage, and improve osmotic adjustments. Our findings broaden the scope of bioactive plant resources available for abiotic stress mitigation.

Previous research on carrot extract in drought-stressed faba beans has shown enhanced proline accumulation and decreased oxidative stress [68]. Although we did not directly assess these biomarkers, the improved germination parameters in our study suggest that carrot-derived compounds may also confer cross-tolerance to salinity stress, offering a broader stress mitigation profile than previously recognized. The rich nutritional and biochemical profiles of carrot roots include phenolics, carotenoids, vitamins A and C, and essential minerals [69, 70]. Moreover, carrot extract plays a crucial role in promoting the biosynthesis of indole-3-acetic acid (IAA)[70], further supporting their role as biostimulants

Moringa-derived extracts, particularly those from flowers, have emerged as highly effective in enhancing germination under salinity stress. Our data align with previous studies highlighting the effectiveness of aqueous moringa leaf extract (ML) in reducing salinity stress, as observed in tomatoes [71] and mint [72]. Although moringa leaf extracts (ML) have been widely studied for reduced zeatin, with concentrations reaching up to 200 micrograms per gram of fresh leaves[73], and contain higher levels of vitamin C (mg per 100 g)[74], our data indicate that moringa flower extract (MF) provided superior results. This may be attributed to the flower's function as a physiological sink for nutrients and its elevated levels of arginine, a known enhancer of stress resilience and plant growth [75].

5. CONCLUSION

Our findings demonstrate that plant extracts have significant potential as cost-effective biostimulants for enhancing salinity tolerance during germination and early seedling growth in chili. The results indicated that plant extracts completely prevented the negative effects of moderate salinity (150mM) on chili pepper seed germination and partially alleviated the impact of high salinity levels (250mM). Among the tested extracts, beetroot ex-

tract demonstrated the highest efficiency in enhancing germination percentage under saline conditions compared to the other extracts.

Further research is essential to explore the physiological and chemical mechanisms underlying the effectiveness of these extracts on the skin. While the observed effects are particularly promising at these critical early developmental stages, we acknowledge that salinity stress affects plants throughout their life cycle. Future studies should evaluate the efficacy of these extracts across the vegetative and reproductive phases to determine their full agricultural potential as alternatives to synthetic stimulants. A deeper understanding of their chemical composition will provide valuable insights into their roles in promoting plant growth and improving salt stress tolerance.

The delayed germination of chili pepper seeds is attributed to their polyphenol content, which exists in both free and sugar-bound forms. Therefore, we propose investigating the dynamic changes in phenolic compounds during germination and evaluating the role of plant extracts as biostimulants to enhance seed germination. Additionally, exploring their interaction with phenolic compounds as novel biostimulants with potential germination- and growth-enhancing properties could provide valuable insights.

Authors' contributions:

All authors contributed to the study's conception and design. E.A. designed and performed the experiments, collected the data, and prepared the manuscript draft. I.M. supervised the research, conducted formal analysis, performed software analysis, and contributed to the manuscript writing, review, and editing. H.H. provided support and contributed to the manuscript review and editing. All authors have read and approved the final version of the manuscript.

Competing Interests:

The authors declare no conflicts of interest.

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