

The Effect of *Viburnum opulus* L. Fruit Extract on Seed Germination and Seedling Growth of Maize (*Zea mays* L.) Under Salt Stress

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ABSTRACT

Viburnum opulus L., known for its high antioxidant activity and positive health effects, is a valuable medicinal plant. The effects of different concentrations of *Viburnum opulus* fruit extract on seed germination and seedling growth of maize (*Zea mays* L.) under salt stress were investigated. Parameters such as germination rate, speed, energy, seed viability were examined; additionally, shoot and root, as well as shoot and root length, weight, and hypocotyl length were evaluated. The findings revealed that 500 and 1000 µg/ml *Viburnum opulus* fruit extract significantly inhibited germination and growth, while 125 µg/ml had no substantial effect. However, the application of 250 µg/ml *Viburnum opulus* fruit extract notably promoted seed germination and seedling growth under salt stress. These results suggest that 250 µg/ml *Viburnum opulus* fruit extract may play a critical role in enhancing tolerance to salt stress.

Keywords: Germination, Growth, Maize, Salt stress

Tuz Stresi Altında Mısır (*Zea mays* L.) Tohumlarının Çimlenmesi ve Fide Büyümesine *Viburnum opulus* L. Meyve Ekstraktının Etkisi

ÖZ

Gilaburu (*Viburnum opulus* L.), yüksek antioksidan aktivitesi ve sağlık üzerindeki olumlu etkileriyle bilinen değerli bir tıbbi bitkidir. Bu çalışmada, *Viburnum opulus* meyve ekstraktının farklı konsantrasyonlarının tuz stresi altındaki mısır tohumlarının çimlenmesi ve fide gelişimi üzerindeki etkileri araştırılmıştır. Çimlenme oranı, hızı, enerjisi ve tohum canlılığı gibi parametreler incelenmiş; ayrıca sürgün ve kök uzunluğu, ağırlığı ve hipokotil uzunluğu değerlendirilmiştir. Bulgular, 500 ve 1000 µg/ml *Viburnum opulus* meyve ekstraktı uygulamalarının çimlenme ve büyümeyi belirgin şekilde sınırladığını, 125 µg/ml'nin ise anlamlı bir etki göstermediğini ortaya koymuştur. Ancak, 250 µg/ml *Viburnum opulus* meyve ekstrakt uygulamasının tohum çimlenmesini ve fide gelişimini tuz stresi altında anlamlı düzeyde teşvik ettiği gözlenmiştir. Bu sonuçlar, 250 µg/ml *Viburnum opulus* meyve ekstraktının tuz stresine toleransta önemli bir rol oynayabileceğini göstermektedir.

Anahtar Kelimeler: Büyüme, Çimlenme, Mısır, Tuz stresi

INTRODUCTION

Abiotic stresses, such as heavy metals, drought, extreme temperatures, salinity, and UV radiation, pose significant threats to crop productivity, leading to substantial yield losses. Among these, salt stress is particularly impactful, as it severely impedes seed germination and early seedling development. High salinity levels are especially detrimental during the germination phase, where elevated salt concentrations reduce germination rates, indices, and seedling growth parameters, including root and shoot lengths and fresh weight. This inhibition is primarily due to salt-induced interference with water uptake, which is crucial during the early stages of plant development [1]. Moreover, high salinity triggers osmotic stress and ion toxicity, disrupting essential physiological processes and

ultimately diminishing crop yields [2]. Notably, the impact of salt stress varies widely across different plant species. Wild plants, often demonstrate higher resilience to salinity compared to cultivated crops [3]. Understanding these species-specific responses is crucial for developing targeted strategies to enhance salt tolerance in crops, especially given that salinity affects approximately 20% of the world's cultivated land and 33% of irrigated agricultural land [4]. This knowledge is vital for advancing agricultural resilience in the face of growing environmental challenges.

Exogenous applications have demonstrated significant potential in enhancing seed germination under salt stress by improving various physiological responses that enable plants to cope with saline conditions. Studies indicate that substances such as salicylic acid (SA), melatonin (MT), and gibberellic acid (GA) can

effectively stimulate germination even in high-salinity environments [5-7]. For instance, SA has been found to mitigate the inhibitory effects of salinity on seed germination by enhancing antioxidant activity and modulating hormonal balances that support stress tolerance [8]. Similarly, MT application has been shown to increase germination rates in salt-stressed seeds by regulating genes associated with stress response pathways, thereby facilitating improved osmotic adjustment and protection against oxidative damage [9]. Additionally, GA has been reported to counteract salt-induced reductions in germination by maintaining cellular ion balance and promoting the breakdown of germination inhibitors [10]. These findings underscore the utility of exogenous treatments in promoting seed germination and establishing robust seedlings in salt-affected soils, providing a valuable strategy for sustaining crop production in challenging environments [11], [12].

Viburnum opulus (VO) is a valuable decorative, medicinal, and edible plant belonging to the Adoxaceae family, naturally growing in Europe, Russia, and some regions of North Africa and North Asia [13, 14]. It is commonly known as guelder rose, European cranberrybush, rosol elder, European guelder, water elder, crampbark, snowball tree, and in Turkey, it is referred to as gilaburu [15, 16]. *V. opulus* holds a significant place in traditional medicine, where its fruit juice is utilized for the treatment of coughs, colds, tuberculosis, rheumatic pain, ulcers, liver diseases, diabetes, and hypertension [17-18]. Numerous studies have revealed various biological activities of *V. opulus*. Animal studies have supported VO's positive effects on the urinary system [19, 20] anti-inflammatory [21], and vasorelaxant [22] activities. In vitro studies indicate that the *V. opulus* shows antimicrobial [23,] antidiabetic [24] anti-obesity [25] anti-inflammatory [26] and anti-cancer [27] properties.

The beneficial effects of *V. opulus* are attributed to its bioactive compounds, including phenolic compounds, vitamin C, carotenoids, iridoids, and essential oils [13], [28, 29]. This rich composition positions *V. opulus* as a promising raw material for the production of functional foods. Additionally, it has been determined that *V. opulus* fruit concentrate can serve as an alternative antioxidant source to delay oxidative changes in the food industry and holds potential as a natural preservative in various products [30].

The aim of this study was to elucidate whether *V. opulus* fruit extracts (VOFE) have a role on the germination of maize seeds under salt stress conditions. The aim of this study is to investigate the bioactive compounds of *V. opulus* and their potential effects on plant development under abiotic stress conditions, particularly salt stress. Additionally, the study seeks to evaluate the traditional medicinal significance of *V. opulus*, its diverse biological activities (such as antimicrobial, anti-inflammatory, and anticancer properties), and its potential applications in functional food production. This research aims to explore the role

of *V. opulus* bioactive compounds in enhancing resilience to abiotic stresses in agricultural production while highlighting its value as a natural resource for health and food industries.

MATERIAL and METHODS

Preparation of *V. opulus* Fruit Extraction

Fresh *V. opulus* fruits were thoroughly washed with tap water and allowed to drain overnight. Following traditional extraction methods practiced by the local population, a crude extract was prepared. Specifically, 40 g of fresh fruit sample was ground with 2000 mL of tap water using a hand grinder until a homogeneous pellet formed. The mixture was then filtered through 0.22 μm filter paper to obtain a clear extract. The resulting extract was stored at +4°C in darkness for subsequent pretreatment applications on maize seedlings. The obtained extract was stored in a refrigerator at +4°C for analyses.

Plant Material, Germination Conditions and Experimental Design

Zea mays L. (cultivar, ADA 523) seeds were provided from Sakarya Maize Research Institute. The seeds were initially washed with 70% ethanol and then surface-sterilized with a 5% sodium hypochlorite solution [31] for 3 minutes. Residual hypochlorite was removed by rinsing the seeds five times with sterile distilled water, after which the seeds were placed in sterilized petri dishes. Germination was conducted in a plant growth chamber under controlled conditions of 60–65% relative humidity, a light intensity of 400 $\mu\text{mol m}^{-2} \text{s}^{-1}$, a temperature of 25°C \pm 2, and a 16-hour light/8-hour dark photoperiod. Firstly, based on the measurement of germination percentage by applying four different salt conditions (0, 150, 200 and 300 mM NaCl) to maize seeds, the salt concentration of 300 mM NaCl, which causes severe stress on seed germination, was determined.

Secondly, four different VOFE treatments (125, 250, 500, 1000 $\mu\text{g/ml}$) were applied to maize seeds to test the hypothesis that VOFE applications would increase the germination limited by salt stress conditions and contribute strongly to salt stress tolerance. The seeds were pretreated with VOFE solutions for 24 hours and then subjected to 300 mM NaCl for 10 days. For each petri dish, 10 mL of the salt solution was added, and germination parameters were recorded daily for 10 days. Germination percentage, germination energy, germination rate index, mean germination time, root length, hypocotyl length, root weight, and shoot weight were evaluated. This study aimed to assess the potential effects of VOFE on mitigating the adverse effects of salt stress on maize seed germination and seedling growth.

Collection of *Viburnum opulus* Fruits

The mature fruits of *V. opulus* (VO) were collected on August 29, 2023, from the vicinity of Şavşat Karagöl National Park, Artvin, along roadsides and near streams (600 m, 41°18'15"N, 42°28'10"E). Specimen identification was verified through herbarium materials (Aksu 459). These collected fruits are preserved in the personal collection at the Medical Plants Application and Research Center of Artvin Çoruh University.

Germination Assessment

Germination Percentage (%GP)

The number of seeds germinated was recorded daily for up to 10 days. From these germination counts, several germination attributes were calculated to characterize the salt tolerance, including germination percentage (%). Seeds with 2 mm long rootlets emerging 3 days after sowing were counted as germinated, and the seeds were counted as germinated when cotyledons appeared at the end of the 10th day.

$$FGP\% = [TNG/TNP] \times 100$$

where FGP % refers to the final germination percentage, TNG refers to the total number of germinated seeds, and TNP refers to the total number of seeds [32].

Germination Rate Index (GRI)

The germination rate index (GRI) represents the daily germination percentage throughout the germination period. Higher GRI values indicate both a greater percentage of seeds germinating and a faster overall germination process.

$$GI = \sum(Gt/Tt)$$

where Gt is the number of seeds germinated on day t, and Tt is the number of days [33].

Germination Energy (GE)

Germination energy was assessed on the fourth day by counting the number of typical seedlings [34].

Mean Germination Time (MGT)

The lower the MGT, the faster a population of seeds has germinated.

$$MGT = \sum(Ti \times Ni) / \sum Ni$$

where MGT refers to mean germination time, Ni refers to the number of germinated seeds on germination days, and Ti refers to the number of days during the germination interval (between 0 and 10 days) [35].

Seed Vigour (SV)

$$SV = [SDM(g) \times GP/100]$$

where SI = seed vigour, SDM is the dry seedling mass (g), and GP = germination (%) [36].

Seedlings Growth Assessment

To assess seedling growth, ten seedlings were randomly selected from each petri dish at the end of the germination period. After selection, shoot length (SL), root length (RL) and hypocotyl length (HL) were measured in cm. We also assessed shoot fresh weight (SFW) and root fresh weight (RFW). SFW and RFW were measured in grams (gr), while the SL, RL, and HL were shown in centimeters (cm).

Statistical Analysis

Each experiment was conducted with a minimum of three replicates. Using one-way analysis of variances (ANOVA), data were analyzed via SPSS software version 27.0 in relation to the Duncan's multiple range test, which was used for comparisons among the treatment means.

RESULTS and DISCUSSION

Effect of Different Salt Stress Conditions on Germination Percentage (%) and Morphological Changes

Firstly, the effective salt stress concentration was selected based on the germination percentage. It was determined that 150 mM NaCl and 200 mM NaCl treatments did not cause a statistically significant difference in germination percentage compared to the control group (non-stressed group). However, 300 mM NaCl treatments decreased the germination percentage in maize seedlings by 25% (Fig 1). According to results, 300 mM NaCl was selected as the salt stress concentration that severely affected seed germination.

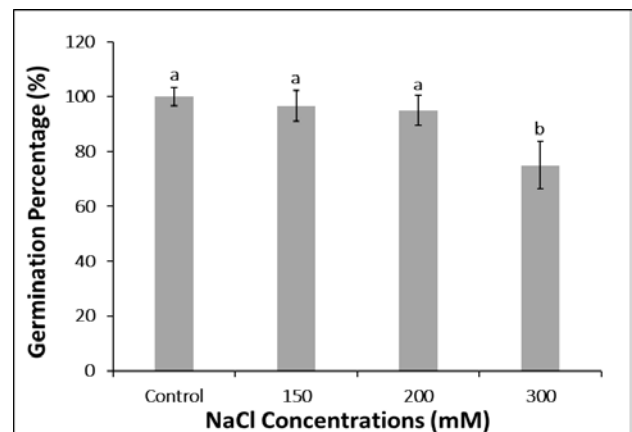


Figure 1. Effects of Different Salt Concentrations on GP (%) in Maize. Bars marked with the same letters indicate no significant difference ($p \leq 0.05$), VOFE: *Viburnum opulus* fruit extract, GP: Germination percentage (%).

To visually demonstrate the effects of salt stress, representative images of maize seedlings subjected to different salt stress concentrations were provided (Figure 2). In the control group (Figure 2a), healthy root and shoot development was observed. Similarly, treatments with 150 mM NaCl (Figure 2b) and 200 mM NaCl (Figure 2c) did not show significant reductions in root and shoot development compared to the control. However, seedlings exposed to 300 mM NaCl (Figure 2d) exhibited a marked reduction in root and shoot growth, clearly demonstrating the detrimental effects of salt stress.

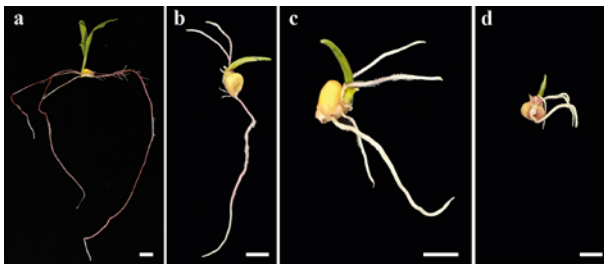


Figure 2. Morphological Alterations of Maize Seedlings After 10 Days of Germination under Different Salt Stress Concentrations. (a) Control group (no salt stress applied); (b) 150 mM NaCl treatment; (c) 200 mM NaCl treatment; (d) 300 mM NaCl treatment. Scale bars represent 1 cm.

Effect of VOFE on Germination Percentage (%) and Germination Rate Index under Salt Stress

To minimize the aging process and protect seedlings from environmental stressors, seeds must meticulously regulate the germination process. Germination and seedling development are essential stages in a plant's life cycle, greatly influenced by temperature and moisture conditions. In this context, the germination percentage (GP) plays a critical role in determining seed viability, plant establishment, and final crop yield, particularly under adverse environmental conditions [37, 38]. Many studies have highlighted the beneficial effects of exogenous antioxidant applications, such as MT and SA, 2,4-epibrassinolide (EBR) etc. on improving seed germination in some crops under salt stress conditions. For instance, MT and EBR have been shown to significantly enhance GE and GRI and improve seedling growth by activating antioxidant defense mechanisms [39]. Likewise, as shown Table 1, compared to the salt stressed group alone, 125 and 250 $\mu\text{g/ml}$ VOFE increased the GP by 11.2% and 19.6%, respectively, whereas 500 and 1000 $\mu\text{g/ml}$ VOFE decreased in the germination percentage by 8.8% and 6.6% (Table 1).

The germination rate index (GRI), a key indicator of seedling vigor and speed of development, can be significantly improved under salt stress conditions through the application of growth regulators or protective treatments, helping to mitigate the inhibitory effects of salinity and promote more uniform and rapid germination [40]. Studies have shown that improving

the GRI under salt stress can be achieved through various methods. For example, exogenous GA3 treatment significantly increased the germination rate index, seed vigor, and other parameters under salt stress conditions, thereby reducing the inhibitory effects of salinity on seed germination [11]. Similarly, seed priming techniques have proven effective in enhancing germination indices in salt-stressed wheat seeds, showing improved germination rates even at higher NaCl concentrations [40]. As shown Table 1, 125, 250, 500 and 1000 $\mu\text{g/ml}$ VOFE increased the GRI by 24.3%, 31.27%, 12.19% and 6.32%, respectively, in comparison with the salt stressed group alone. The highest the GRI was showed in 250 $\mu\text{g/ml}$ VOFE. These results suggest that 250 $\mu\text{g/ml}$ VOFE could be more effective in promoting seed germination under salt stress due to its high antioxidant properties, providing a promising approach to enhancing maize resilience to salinity and improving crop productivity [41].

Effect of VOFE on Germination Energy Index under Salt Stress

The ability to optimize germination energy (GE) is key to ensuring plant establishment in adverse environmental conditions, as seeds with higher GE show better resilience and growth potential under stress, such as salinity. As shown Table 1, compared to the salt stressed group alone, 125 $\mu\text{g/ml}$ VOFE $\mu\text{g/ml}$ did not cause a statistically significant ($p \leq 0.05$) difference in the GE. However, 250 $\mu\text{g/ml}$ VOFE increased the GE by 15.28%, but 500 and 1000 $\mu\text{g/ml}$ VOFE decreased by 18.5% and 16.08% in comparison with the salt stressed group alone (Table 1). Some studies has shown that the positive effects of exogenous treatments on improving seed GE and resilience some plants under salt stress conditions [42, 43]. For instance, exogenous proline improved GE in rice under salt stress [43]. These results provide valuable insights into how about 250 $\mu\text{g/ml}$ VOFE can improve GE and seedling development under salt stress conditions?

Effect of VOFE on Mean Germination Time under Salt Stress

Mean germination time (MGT) serves as an indicator of both the spread of time and the speed of germination across the seed population [44]. Therefore, a high germination rate generally corresponds with a lower MGT, which means the seeds germinate faster and more efficiently [44]. As shown Table 1, it was detected that 125, 500 and 1000 $\mu\text{g/ml}$ VOFE $\mu\text{g/ml}$ did not cause a significant difference in MGT compared to the salt stressed group alone. However, 250 $\mu\text{g/ml}$ VOFE decreased the MGT by 7.24%. The lowest the MGT was observed with 250 $\mu\text{g/ml}$ VOFE (Table 1). The relationship between GP and MGT is significant as MGT is used to assess the speed of germination. A study showed that maize seeds with a higher GP typically have a lower MGT, indicating that they

complete germination more quickly [45]. The results for GP and rate provide strong evidence that, with the increased GP and rate due to 250 µg/ml VOFE, the germination time decreased under salt stressed group alone. The results provide strong evidence that the 250 µg/ml VOFE not only increased germination percentage rate but also effectively reduced mean germination time in comparison with the salt stressed group alone.

Effect of VOFE on Seed Vigour under Salt Stress

Seed vigour, a complex agronomic trait that encompasses seed longevity, germination speed, seedling growth, and early stress tolerance, determines the duration and success of the establishment period. In this process, the germination rate-defined as the percentage of seeds that successfully sprout within a specific timeframe-serves as a key indicator of seed viability and vigour under various environmental conditions [46]. The results showed that 125 and 250 µg/ml VOFE increased the SV by 25.04% and 32.8%. However, 500 and 1000 µg/ml VOFE decreased the SV by 19.54% and 29.14%, respectively, in comparison with salt stressed group alone (Table 1). The SV is strongly influenced by germination percentage, as a higher germination percentage typically reflects a more vigorous seed lot, indicating that seeds are not only viable but also capable of rapid and uniform development [47]. Compared to the salt stressed group alone, the increase in germination percentage observed with the application of 250 µg/ml VOFE aligns with the enhanced vigour index, further reinforcing its potential role in mitigating the adverse effects of salinity.

Table 1. Effects of Salt Stress and Different VOFE on GP, GRI, GE and MGT and SV in Maize

Treatment	GP (%)	GRI	GE	MGT	SV
300 mM NaCl	75±8.6 ^c	16.35±0.3 ^c	71.5±3 ^b	6.25±0.17 ^a	317.7±2.2 ^c
<i>Under salt stress conditions</i>					
125 µg/ml VOFE	84.4±3.5 ^b	21.6±0.6 ^b	78.3±5.5 ^b	6.25±0.14 ^a	423.9±9.8 ^f
250 µg/ml VOFE	93.3±4.2 ^a	23.8±1.2 ^a	84.4±6.3 ^a	5.8±0.11 ^b	473.1±8.4 ^f
500 µg/ml VOFE	68.3±5.5 ^c	18.6±1.5 ^c	58.3±8.6 ^c	6.2±0.11 ^a	255.6±2.3 ^f
1000 µg/ml VOFE	70±7.5 ^c	17.4±1.7 ^d	60±4.7 ^c	6.24±0.15 ^a	225.1±3.2 ^f

Bars marked with the same letters indicate no significant difference ($p \leq 0.05$), VOFE: *Viburnum opulus* fruit extract, GP: Germination percentage (%), GRI: germination rate index, GE: germination energy, MGT: mean germination time and SV: seed vigor

Effect of VOFE on Seedling Growth Parameters on Salt Stress

As shown Table 2, the effect of various concentrations of VOFE on shoot fresh weight (SFW), shoot length (SL), root fresh weight (RFW), root weight (RW), hypocotyl length (HL) were evaluated and also as

shown Figure 2, the morphological alterations were observation under salt stress.

The shoot fresh weight (SFW) was decreased with 125, 500, and 1000 µg/ml VOFE by 20.37%, 5.28% and 27.16% respectively, in comparison with salt stressed group alone. However, the 250 µg/ml VOFE concentration was observed to increase SFW by 5.69% comparison with salt stressed group alone (Table 2). Compared to salt stressed group alone, 125, 250, 500 µg/ml VOFE increased the shoot length (SL) by 81.3%, 87.5%, 62.5%, respectively. However, SL did not show any statistically significant ($p \leq 0.05$) change with the 1000 µg/ml VOFE under salt stressed group alone. The highest SL was observed in 250 µg/ml VOFE.

Likewise, the root fresh weight (RFW) was decreased with 125, 500 and 1000 µg/ml VOFE by 11.94%, 34.82%, and 43.78%, respectively, but 250 µg/ml VOFE increased RFW by 15.55% in comparison with salt stressed group alone (Table 2). Additionally, in root length (RL), 125, 500 µg/mL and 1000 µg/mL VOFE did not result in a statistically significant ($p \leq 0.05$) difference compared to the salt stressed group alone. However, the 250 µg/mL VOFE increased RL by 37.4% under salt stressed group alone (Table 2).

The hypocotyl length (HL) of plants tends to reduce significant or not? due to the inhibitory effects of salinity on cell elongation and overall growth compared to salt stressed alone. For instance, studies have shown that exposure to high salt stress leads to a reduction in the length of the hypocotyl, indicating that inhibition of normal hypocotyl growth [48]. As shown in Table 1, HL did not show any statistically significant ($p \leq 0.05$) change with the 125 and 500 µg/ml VOFE under salt stressed group alone. However, it was observed that the 250 µg/ml VOFE increased the HL by 18.46%, in comparison with salt stress treatment alone. Additionally, no hypocotyl development was detected with 1000 µg/ml VOFE (Table 2). Many studies investigated that several exogenous substances, have a potential antioxidant properties, have been shown to mitigate the adverse effects of salt stress. For example, in a study on *Leptochloa fusca*, it was observed that increased salinity up to 300 mM NaCl led to improved root and shoot length under salt [49].

Table 2. Effects of Salt Stress and Different VOFE on SFW, SL, RFW, RL and HL in Maize.

Treatment	SFW (gr)	SL (cm)	RFW (gr)	RL (cm)	HL (cm)
300 mM NaCl	2.65±0.02 ^c	0.3±0.03 ^d	2.01±0.03 ^b	3.6±1.2 ^b	1.06±0.12 ^b
<i>Under salt stress conditions</i>					
125 µg/ml VOFE	2.68±0.03 ^b	1.06±0.01 ^b	1.77±0.04 ^c	3.6±1.1 ^b	1.06±0.07 ^b
250 µg/ml VOFE	2.81±0.04 ^a	2.4±0.02 ^a	2.38±0.02 ^a	5.75±0.67 ^a	1.3±0.08 ^a
500 µg/ml VOFE	2.51±0.02 ^d	0.8±0.04 ^c	1.31±0.06 ^d	3.4±1.3 ^b	1.1±0.07 ^b
1000 µg/ml VOFE	1.93±0.05 ^e	0.36±0.03 ^d	1.13±0.07 ^e	2.52±0.8 ^b	-

Bars marked with the same letters indicate no significant difference ($p \leq 0.05$), VOFE: *Viburnum opulus* fruit extract, SFW: shoot fresh weight, SL: shoot length, RFW: root fresh weight, RL: root length and HL: hypocotyl length

Similarly, jasmonic acid was found to significantly increase root and shoot growth parameters in *Hibiscus sabdariffa* seedlings [50]. These findings suggest that specific antioxidant substance treatments can enhance growth and stress resilience in plants exposed to high salinity conditions. These results support that, compared to salt stressed group alone, 250 VOFE $\mu\text{g/ml}$, due to its antioxidant potential, can significantly promote plant growth and development, thereby enhancing salt stress tolerance in plants.

As shown Figure 2, the effects of different concentrations of *V. opulus* fruit extract (VOFE) on the germination and morphology of maize seeds under 300 mM NaCl salt stress. In the absence of VOFE application (Figure 2a), the adverse effects of salt stress are evident, with root and shoot growth severely inhibited, highlighting the significant inhibitory impact of 300 mM NaCl on germination and seedling development. When 125 $\mu\text{g/mL}$ VOFE was applied (Figure 3b), slight improvements in root and shoot elongation were observed, indicating a partial alleviation of salt stress. The most notable improvement was observed at 250 $\mu\text{g/mL}$ VOFE (Figure 3c), where the seedlings exhibited maximum root and shoot lengths and enhanced vigor, suggesting that this concentration was the most effective in mitigating the effects of salt stress. However, at 500 $\mu\text{g/mL}$ VOFE (Figure 3d), the growth improvement was limited and less pronounced compared to 250 $\mu\text{g/mL}$. At the highest concentration of 1000 $\mu\text{g/mL}$ VOFE (Figure 3e), seedling growth was severely inhibited, producing results similar to or worse than those observed under salt stress alone (Figure 3a). This suggests potential toxicity or reduced efficacy at excessive concentrations. Overall, 250 $\mu\text{g/mL}$ VOFE emerges as the optimal concentration for supporting maize seed germination and seedling development under salt stress, demonstrating its potential as a biostimulant for alleviating plant stress.

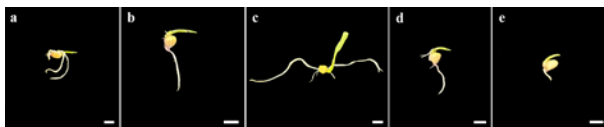


Figure 3. Morphological Effects of Different Concentrations of *Viburnum opulus* Fruit Extract (VOFE) on Maize Seed Germination and Seedling Growth under 300 mM NaCl. (a) 300 mM NaCl; (b) 125 $\mu\text{g/mL}$ VOFE; (c) 250 $\mu\text{g/mL}$ VOFE; (d) 500 $\mu\text{g/mL}$ VOFE; (e) 1000 $\mu\text{g/mL}$ VOFE. Scale bars represent 1 cm.

CONCLUSION

Maize seeds were treated with varying concentrations of *Viburnum opulus* fruit extract (VOFE) to evaluate its effectiveness in enhancing germination and growth under salt stress conditions. The results showed that especially 500 and 1000 $\mu\text{g/ml}$ VOFE had a detrimental effect on seed germination and seedling development,

including root and shoot length/weight, as well as hypocotyl length under 300 mM salt stress. However, the exogenous treatment of 250 $\mu\text{g/ml}$ VOFE significantly improved germination and promoted plant growth, indicating increased tolerance to high salinity. These findings strongly suggest that the antioxidant properties of *Viburnum opulus*, 250 $\mu\text{g/ml}$ VOFE plays a crucial role in mitigating the harmful effects of salt stress, providing strong evidence for its potential use in enhancing crop resilience in saline environments. Consequently, the exogenous application of VOFE during the germination stage is an effective strategy to improve seed performance under salt stress. It is therefore recommended to prime crop seeds with VOFE before planting in saline conditions.

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