



# Improving Salt Stress Tolerance in *Rosmarinus officinalis* through Intercropping with Halophytic Plants: A Biochemical Emphasis on Evaluating Morpho-Physiological Parameters

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## Abstract

The challenge of salt stress in plant growth is a major concern in agriculture. Rosemary (*Rosmarinus officinalis*) holds particular interest as a domesticated crop, but it faces challenges related to salinity that hinder its optimal development. In this study, we assessed the effect of salt stress on rosemary growth by associating it with certain halophytic plants, namely *Plantago coronopus* and *Spergularia salina*. We examined various morphological and physiological parameters to understand the impact of salinity on these plant associations. The results showed that increasing NaCl concentration in the soil led to a significant reduction in rosemary growth, particularly in root length, stem length, root weight, and stem weight. However, the combination of rosemary with certain halophytic plants appeared to mitigate the adverse effects of salt stress on rosemary growth. The physiological parameters studied included the relative water content of leaves, stems, and roots, as well as sugar content in the leaves. The increase in NaCl concentration resulted in a significant decrease in relative water content in different parts of rosemary, while the sugar content in the leaves increased. However, the association of rosemary with *Plantago coronopus* did not lead to higher sugar secretion compared to the control. Nevertheless, by associating rosemary with certain halophytic plants, we could enhance its resilience to salt stress, offering promising prospects for more salt-resistant and sustainable agriculture.

**Keywords:** Rosemary, *Plantago*, *Spergularia*, NaCl.

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## 1. Introduction

Salinization stands out as a major hurdle in agriculture [1]. Soil salinization is now affecting the world's most productive regions for sustainable agriculture, posing a major obstacle to plant growth and development due to high salinity [2]. Salt represents one of the main threats that significantly limits plant productivity at both local and global scales, with an increase in soil salinity being perceived as evident for plant populations [3]. According to FAO, salinization affects approximately 20 to 30 million hectares out of the 260 million hectares of irrigated land worldwide. This information is incorporated into the Global Map of Salt-Affected Soils (GSASmap), a product that gathers contributions from over

118 countries, encompassing 257,419 sites containing measured soil data [4]. Projections indicate that more than half of the world's agricultural land will experience salinization by 2050 [5]. This highlights the urgent need to address the challenge of soil salinization and its negative consequences on global food production. It is vital to implement effective strategies with implications both at the local and global levels [6]. Two distinct groups of plants have been identified based on their ability to endure salt: "halophytes," which are salt-tolerant, and "glycophytes," which are salt-sensitive. This categorization is determined by their respective capabilities to thrive in saline environments [7-8].

As a result, there has been an increasing focus on enhancing salt tolerance in glycophytic crop species in recent years [9-10]. Halophytic plants, which thrive in saline soils, are defined as plants that complete their life cycle in conditions of high salt concentration [7]. These fascinating plants have received special attention due to their ability to tolerate high-salt waters and soils through specific mechanisms [11]. This growing interest in halophytic plants and their salt tolerance mechanisms has led to a comparative study to explore their potential in improving the salt stress tolerance of glycophytic crop species. *Rosmarinus officinalis* L. is an evergreen plant in the form of small shrubs, with aromatic leaves and delicate blue flowers [12]. It belongs to the Lamiaceae family, one of the largest families among flowering plants, comprising 252 genera and 7200 to 6900 species worldwide [13]. The genus *Plantago* (Plantaginaceae) comprises different species that exhibit varying salt sensitivity [14]. *Plantago coronopus* L. is found in various habitats, such as nitrified and degraded pastures, trampled regions, roadside ditches, and coastal areas [15]. Additionally, these plants can thrive in environments with diverse salinity conditions [14]. *Spergularia salina* a halophilic plant species from primary salt-influenced environments in central Europe, is currently extending its range to roadside areas that receive de-icing salt treatments in the winter [16]. The objective of this study is to evaluate the salt stress tolerance of *Rosmarinus officinalis*, a glycophytic plant, when associated with two halophytic plant species, *Plantago coronopus* and *Spergularia salina*. We aim to determine whether this association enhances the tolerance of rosemary by monitoring relevant morpho-physiological and biochemical parameters.

## 2. Materials and methods

### 2.1. Plant Material

As part of our experimentation, we selected three plant species, including one glycophytic and two halophytic species. One-year-old rooted cuttings of *Rosmarinus officinalis*, our glycophytic plant, were obtained from a reputable nursery. As for the halophytic plant species, *Plantago coronopus* and *Spergularia salina*, they were collected from Oued Malah, located near the Brikcha territorial commune, which belongs to the Ouazzane region in northwestern Morocco (Figure 1). Oued Malah is a stream in Morocco, situated in the North Africa region. It is located at the geographical coordinates of 34.82593° north latitude and 5.58803° west longitude, with an elevation of 180 meters (591 feet). This location provides a suitable environment to study these halophytic plants under specific saline conditions, thus contributing to enriching our understanding of their adaptation mechanisms to salt stress.

### 2.2. Experimental Setup and Treatments applied

In this study, we conducted an experiment with cuttings of *Rosmarinus officinalis* grown both individually and in association with two halophytic plant species, *Plantago coronopus* and *Spergularia salina*. The plants were cultivated in a mixture of Maamora soil (Kenitra) combined with peat and natural compost. To examine salt stress tolerance, we used different salt treatments using sodium chloride (NaCl) at specific concentrations of 3g/l (51 mmol/l), 6g/l (102 mmol/l), and 9g/l (153 mmol/l). Each group of plants was

exposed to one of these salt treatments by adding the corresponding NaCl solution to the irrigation water. We also maintained a control group without salt treatment, where plants were irrigated only with water. This experimental setup allowed us to compare the responses of plants subjected to different salt concentrations with the control group. The plants were watered with 100 ml of water in the morning, and the watering interval varied from 2 to 3 days as needed. For each of the following salt concentrations, 3g/l, 6g/l, 9g/l NaCl, 8 manipulations were performed. While for the control, we used 4 manipulations. This experimental design was applied to all three experiments: *Rosmarinus officinalis* alone, *Rosmarinus officinalis* with *Plantago coronopus*, and *Rosmarinus officinalis* with *Spergularia salina*.

### 2.3. Measurement of soil electrical conductivity

Soil electrical conductivity was assessed by conducting measurements on a soil-water extract (1:5, w/v) using a conductivity meter [17].

### 2.4. Morphological Parameters Examined

Various morphological parameters were examined, including root length, stem length, root volume, root weight, and stem weight. Root length was measured from the main root tip to the point of branching or the end of lateral roots, while stem length referred to the height of the above-ground portion of the plant from the soil level to the topmost point of the stem. Root volume represented the total space occupied by the plant's root system in the soil. Additionally, root weight was determined as the mass of the plant's root system, and stem weight referred to the mass of the above-ground portion of the plant, encompassing the main stem and branches.

### 2.5. Physiological Parameters Examined

Two physiological parameters were examined in this study: Relative Water Content of Leaves, Stem, Root and Sugar Content in Leaves. Relative Water Content is calculated according to the formula of Martínez et al., and Sugar Content are determined using the phenol method of Dubois et al. [18-19].

### 2.6. Statistical Analysis

The data obtained from the experiment underwent descriptive statistical analysis, analysis of variance (ANOVA), correlation testing, and Principal Component Analysis (PCA) using the IBM SPSS Statistics program version 23.

## 3. Results and Discussion

### 3.1. Soil electrical conductivity

Figure 3 shows a comparison of soil electrical conductivity among different salt concentrations for three plant groups: *Rosmarinus officinalis* alone, *Rosmarinus officinalis* with *Plantago coronopus*, and *Rosmarinus officinalis* with *Spergularia salina*. As expected, the results show that soil electrical conductivity increases significantly with the increase in NaCl concentration in the three experiments.

These experiments include *Rosmarinus officinalis* alone, *Rosmarinus officinalis* with *Plantago coronopus*, and *Rosmarinus officinalis* with *Spergularia salina*. This

indicates that the presence of NaCl in the soil leads to a significant increase in electrical conductivity, indicating a higher level of salt content in the soil.

### 3.2. Morphological parameters

In the experiment with *Rosmarinus officinalis* alone, increasing salt concentrations lead to a significant reduction in root length, stem length, and root weight, while stem weight and root volume remain unchanged. A similar trend is observed in the experiment with *Rosmarinus officinalis* and *Plantago coronopus*, where higher salt concentrations result in significant reductions in root length, stem length, root weight, and stem weight. However, root volume is not significantly affected. In the experiment with *Rosmarinus officinalis* and *Spergularia salina*, an increase in salt concentrations also leads to significant decreases in root length, stem length, root weight, and stem weight, without significantly affecting root volume. The plant's morphology, measured through root length, stem length, root weight, and stem weight, provides essential information about its ability to explore the soil, undergo development, and allocate its biomass to support its aerial structures and reproductive efforts. Root volume also reflects the plant's capacity to spread its roots and access a larger area for nutrient and water uptake. These parameters play a crucial role in the overall understanding of the plant's growth and development. The negative effects of sodium chloride (salt) on plant growth are generally attributed to osmotic effects and/or ionic imbalances resulting from nutritional deficiencies or excessive ions. This inability of plants to adequately hydrate their tissues under saline conditions causes water stress, and it has been observed that salt tolerance is often associated with drought tolerance [20-21]. Previous studies have indicated that salt stress has a detrimental effect on plant growth, resulting in a slight reduction in overall development [22]. Specifically concerning Rosemary, research has shown that concentrations of 2, 5, and 8 g/l of NaCl led to a decrease in morphological growth parameters [23]. Additionally, further studies have highlighted that Rosemary plants can preserve their turgor through osmotic adjustment in response to salt stress [24]. On the other hand, the study results indicate that salinity led to a reduction in the growth of both the aerial and root parts of Rosemary, which is a normal phenomenon. Indeed, prior research conducted by Hamrouni et al., and Thouraya et al., has demonstrated that plants respond to salt stress by initially reducing their root system to protect the aboveground portion and sustain photosynthesis. Subsequently, as salinity levels rise, there is a decrease in the aboveground portion as well [25-26].

### 3.3. Physiological Parameters

Table 2 presents the effects of salt stress on various physiological parameters in three different experiments involving *Rosmarinus officinalis* with different halophytic plant species. The study results indicate that the increase in NaCl concentration led to a significant decrease in the relative water content in the leaves, stems, and roots of *Rosmarinus officinalis*. However, the sugar content in the leaves increased with higher NaCl concentrations. This trend was also observed in the experiments with *Plantago coronopus* and *Spergularia salina*. During salt stress, the relative water content in plants can decrease. Salt stress leads to an increase in the salt concentration in the soil, creating an osmotic

imbalance between the plant roots and the soil. In response to this imbalance, water tends to leave the plant cells and move towards the soil with a higher concentration of dissolved salts [27-28]. This water loss results in dehydration of plant cells, leading to a decrease in water content within the tissues. A reduction in relative water content can have adverse effects on plant physiology, particularly disrupting growth and development processes. Faced with dehydration caused by salt stress, some plants develop adaptive mechanisms, such as the accumulation of higher levels of soluble sugars. This adaptive response to osmotic stress, caused by the presence of salts in the soil, allows soluble sugars to act as osmoprotectants, maintaining cellular water balance and preventing water loss from the cells [29]. Conversely, by increasing their soluble sugar content, plants can reduce the impact of water deficit caused by salt stress and ensure a more stable physiological state. Halophytic plants have developed effective adaptations for their survival, supported by molecular evidence from recent studies [30]. In our study, we measured the soluble sugar content in three groups: *Rosmarinus officinalis* alone, *Rosmarinus officinalis* with *Plantago coronopus*, and *Rosmarinus officinalis* with *Spergularia salina*. The results showed significant differences in the concentration of soluble sugars between these groups, revealing their response to salt stress. The results of our study demonstrate significant variations in sugar secretion depending on the NaCl concentration. For the *Rosmarinus officinalis* alone group, we observed a progressive increase in sugar secretion with increasing NaCl concentration. At a concentration of 3g/l NaCl, the *Rosmarinus officinalis* with *Plantago coronopus* group showed a significantly lower sugar content ( $0.66 \pm 0.20$  mg/ml FW) compared to the *Rosmarinus officinalis* alone group ( $1.42 \pm 0.16$  mg/ml FW) and the *Rosmarinus officinalis* with *Spergularia salina* group ( $1.03 \pm 0.05$  mg/ml FW). Similarly, at a concentration of 6g/l NaCl, the *Rosmarinus officinalis* with *Plantago coronopus* group also exhibited a significantly lower sugar content ( $0.54 \pm 0.09$  mg/ml FW) compared to the *Rosmarinus officinalis* alone group ( $1.75 \pm 0.31$  mg/ml FW). However, there was no significant difference between the *Rosmarinus officinalis* with *Spergularia salina* group ( $1.08 \pm 0.17$  mg/ml FW) and the other two groups at this concentration. Finally, at a concentration of 9g/l NaCl, the *Rosmarinus officinalis* with *Plantago coronopus* group showed a significantly lower sugar content ( $0.33 \pm 0.04$  mg/ml FW) compared to the *Rosmarinus officinalis* alone group ( $2.46 \pm 0.30$  mg/ml FW). However, there was no significant difference between the *Rosmarinus officinalis* with *Spergularia salina* group ( $2.35 \pm 0.29$  mg/ml FW) and the *Rosmarinus officinalis* alone group at this concentration. These observations suggest that the association of rosemary with *Plantago coronopus* may lead to a different regulation of sugar secretion in response to salt stress, offering new perspectives for understanding the importance of halophytic plants in enhancing rosemary's tolerance to salt stress.

### 3.4. Effects of NaCl treatment, plant association and their interaction

Analysis of variance was carried out to assess the impact of different sources of variation on various morpho-physiological parameters of the study. Results reveal significant differences between association combinations

between plants for all parameters except stem length, with notable effects on root length, leaf weight, leaf number, sugar, and the relative water content of roots, stems, and leaves. Likewise, the applied NaCl treatment had a significant impact on all measured parameters. In addition, the interaction between the type of plant association and the treatments also influenced several parameters, in particular root length, stem length, root weight, leaf weight, number of leaves, root volume, sugar content, and relative root water content. The residual errors remain relatively low, indicating good precision in the measurements carried out. These results highlight the crucial importance of interactions between associations between plants and NaCl treatments, as well as variations between individuals in the response of the morpho-physiological parameters studied.

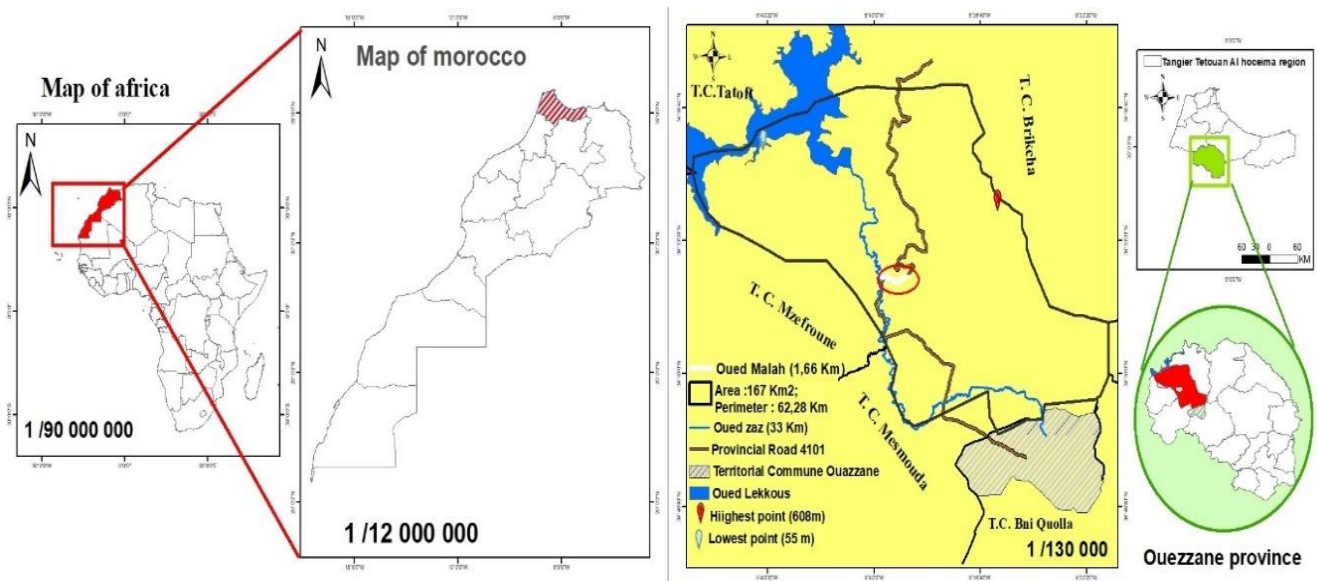
### 3.5. Correlation analysis

Correlation coefficients exceeding 0.5 were taken into consideration in this analysis in order to identify significant relationships between the different traits studied. Root length showed a positive correlation with stem length ( $r = 0.625^{***}$ ), root weight ( $r = 0.618^{***}$ ), and leaf weight ( $r = 0.552^{***}$ ). This correlation suggests coordination between the root system and aboveground biomass, potentially suggesting a structural adaptation aimed at optimizing growth. Stem weight has a positive correlation with number of leaves ( $r = 0.658^{***}$ ) and leaf weight ( $r = 0.607^{***}$ ). Additionally, root relative water content displays significant positive correlations with stem length ( $r = 0.833^{***}$ ), leaf weight ( $r = 0.722^{***}$ ), root length ( $r = 0.661^{***}$ ), the weight of the roots ( $r = 0.651^{***}$ ), and the number of leaves ( $r = 0.609^{***}$ ). These associations highlight the importance of efficient water uptake by roots to support overall plant growth. Relative stem water content is positively associated with relative root water content ( $r = 0.738^{***}$ ), leaf weight ( $r = 0.630^{***}$ ), number of leaves ( $r = 0.556^{**}$ ), root length

( $0.553^{**}$ ), and stem length ( $r = 0.503^{**}$ ), indicating coordination between the different parts of the plant to maintain water homeostasis. Furthermore, relative leaf water content showed significant positive correlations with leaf weight ( $0.892^{***}$ ), relative root water content ( $r = 0.839^{***}$ ), relative root water content ( $r = 0.738^{***}$ ), the weight of the stem ( $r = 0.691^{**}$ ), the relative water content of the stem ( $0.662^{**}$ ), the length of the stems ( $0.573^{**}$ ), and the length of the roots ( $0.517^{**}$ ). These associations suggest integrated water regulation to support leaf physiological function. These results highlight the complex interconnection of physiological mechanisms underlying the growth and adaptation of plants to their environment.

### 3.6. Principal component analysis

In the absence of saline stress (0 g/L NaCl), the first component, F1, exhibits correlations with Root length, Stem weight, Weight of leaves, Number of Leaves, Root volume, Sugar content, Relative Water Content (RWC) in stem and leaves. Conversely, the second component, F2, is primarily associated with Stem length and Root weight. When *Rosmarinus officinalis* is cultivated alone, it is characterized by a high RWC in the stem and number of leaves, as well as lower root weight, while the remaining traits display average values. In the presence of the association *R. officinalis* + *S. salina*, *R. officinalis* demonstrates increased Stem length but reduced RWC in the stem, root volume, and sugar content, with the other traits exhibiting moderate values. In contrast, in the association *R. officinalis* + *P. coronopus*, *R. officinalis* displays elevated values of sugar and root volume but lower values for leaf and stem weight, leaf RWC, root length, and stem root length.



**Figure 1:** Geographical origin of halophytic species: *Plantago coronopus* and *Spergularia salina* collected from Oued Malah.

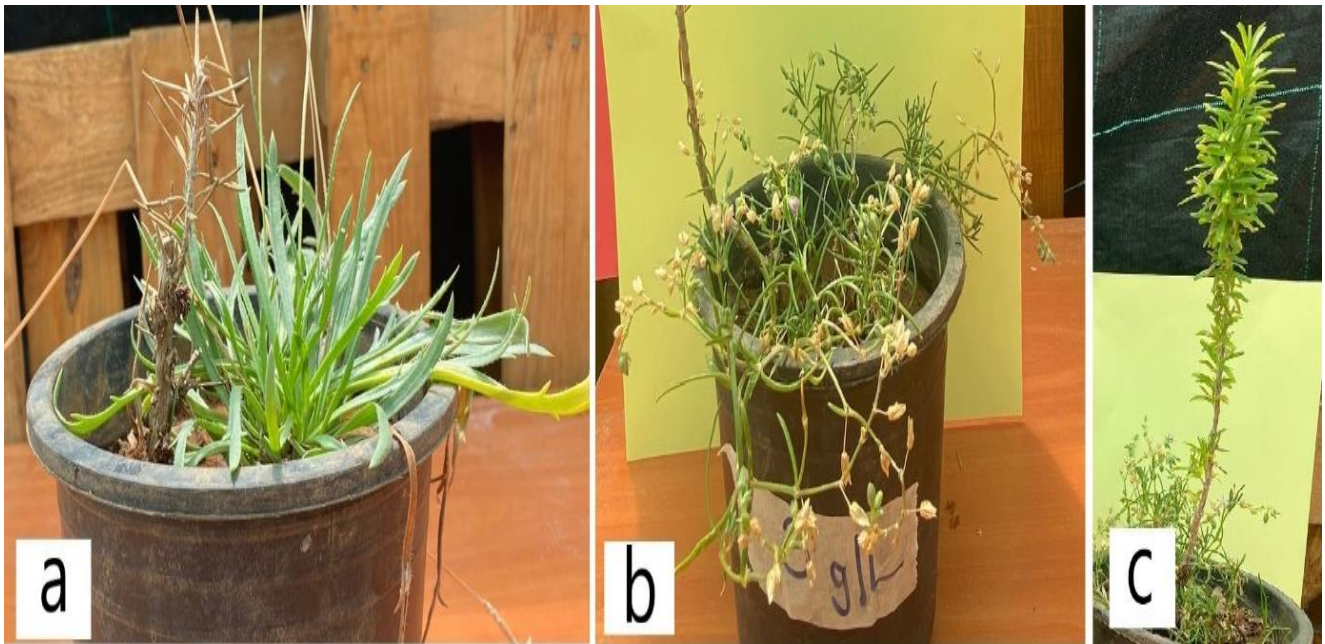


Figure 2: (a) *Plantago coronopus* (b) *Spergularia salina* (c) *Rosmarinus officinalis*.

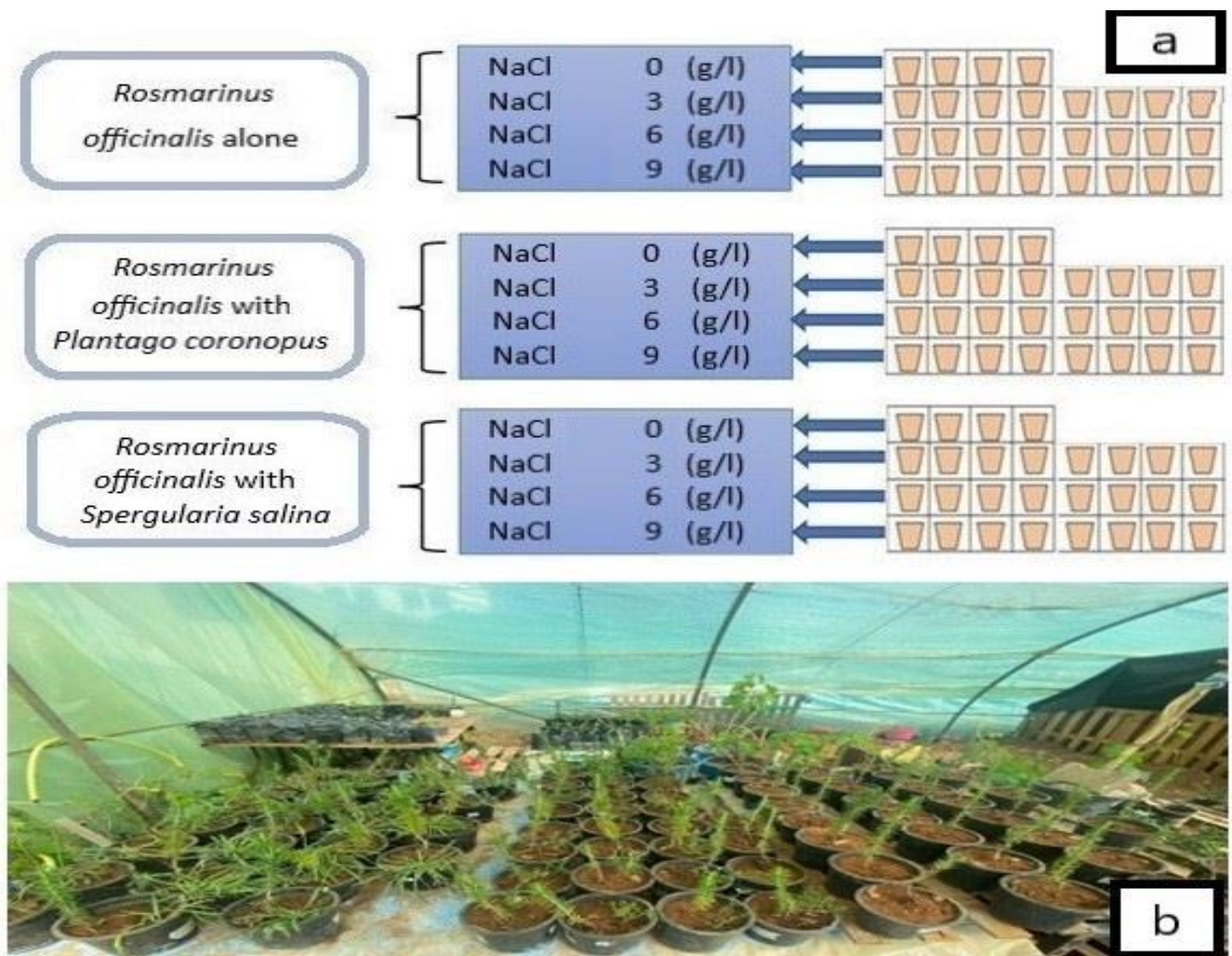
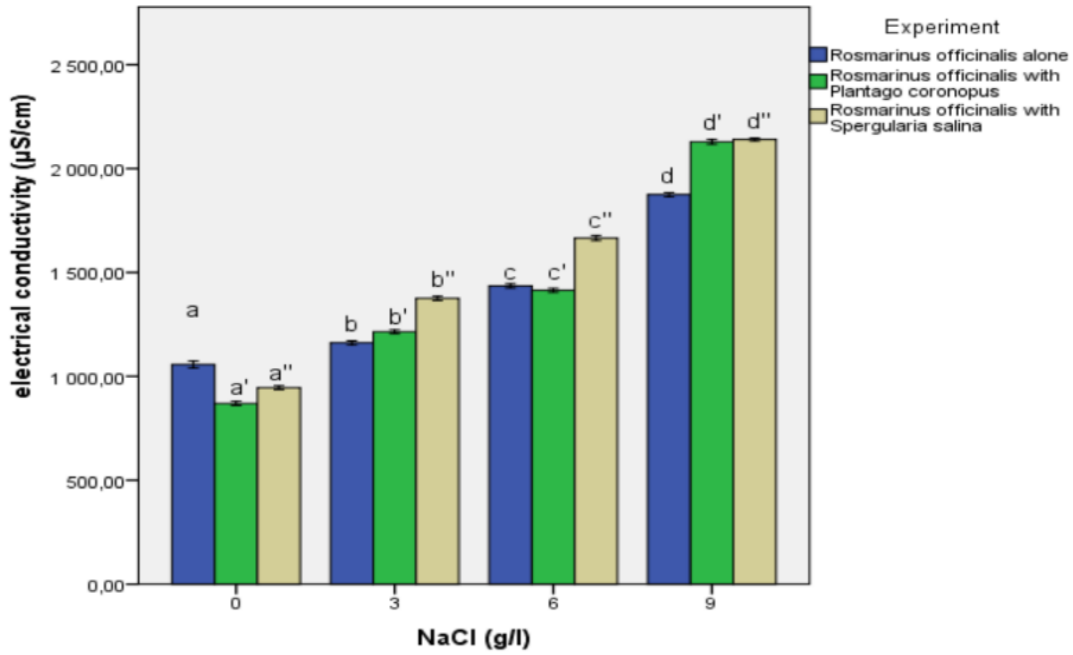
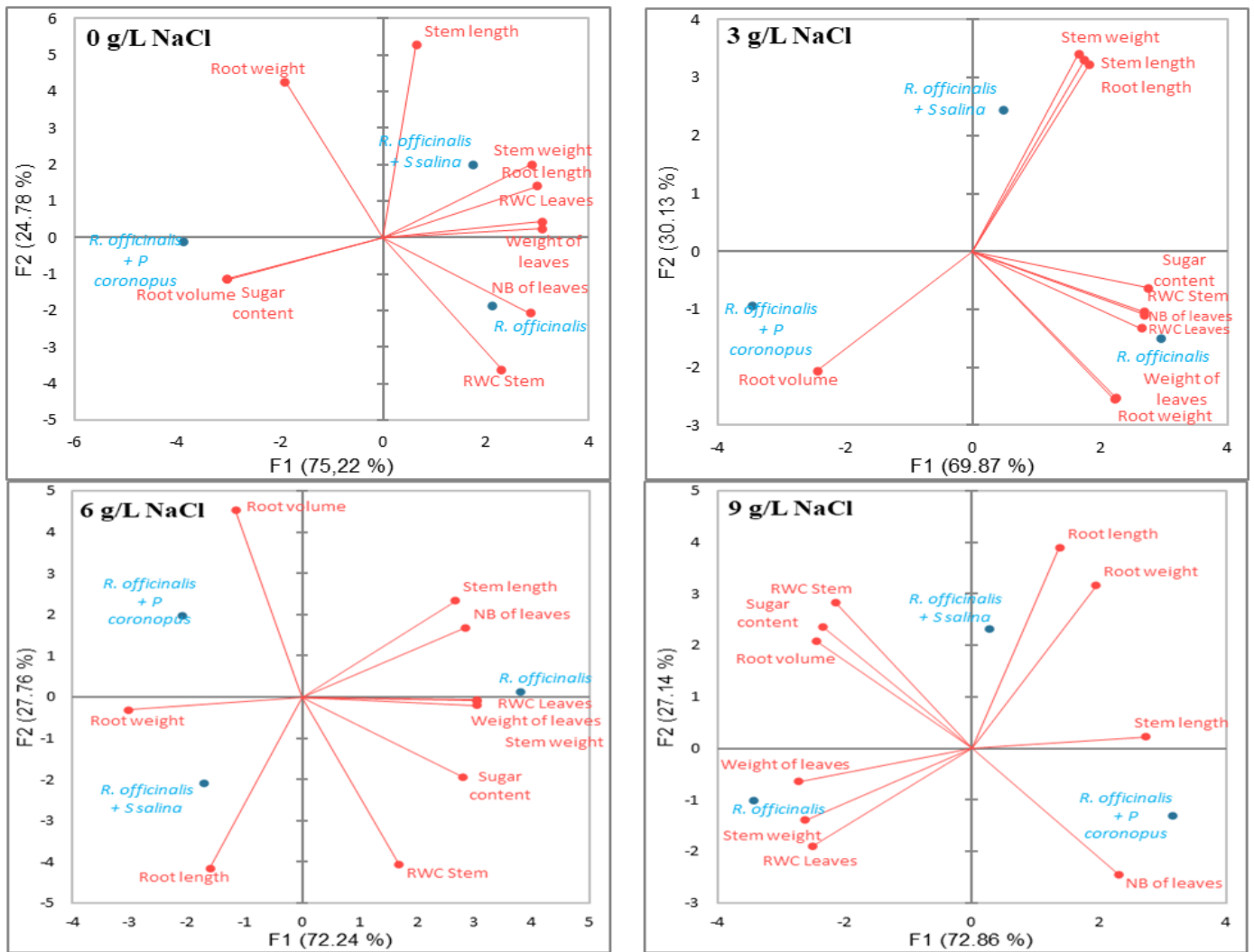


Figure 3: (a) Representative diagram of the manipulation (b) Experimental Arrangement for Salinity Treatment in the Greenhouse at the Faculty of Sciences, Ibn Tofail University in Kenitra.



**Figure 4:** Comparison of Soil Electrical Conductivity Among Different Salt Concentrations for Three Plant Groups: Rosmarinus, Rosmarinus + Plantago, and Rosmarinus + Salina.



**Figure 5:** Distribution of variables and types of association between *R. officinalis*, *S. salina*, and *P. coronopus* in principal components F1 and F2 as affected by NaCl concentration.

**Table 1:** Effects of Salt Stress on Morphological Parameters (Root Length, Stem Length, Root Volume, Root Weight, Stem Weight) in the Three Experiments.

Experiment	NaCl (g/l)	Root length (cm)	Stem length (cm)	Root weight (g)	Stem weight (g)	Root volume (cm <sup>3</sup> )
<i>Rosmarinus officinalis</i> alone	0	31,93 ± 3,93 <sup>a</sup>	31,6 ± 1,76 <sup>a</sup>	21,51 ± 2,89 <sup>a</sup>	3,65 ± 1,23 <sup>a</sup>	13 ± 2,64 <sup>a</sup>
	3	24,26 ± 1,32 <sup>b</sup>	29,83 ± 2,25 <sup>a</sup>	21,50 ± 2,17 <sup>a</sup>	2,36 ± 0,47 <sup>a</sup>	11 ± 1 <sup>a</sup>
	6	20,5 ± 1,93 <sup>b</sup>	26,83 ± 0,76 <sup>a</sup>	10,99 ± 1,36 <sup>b</sup>	2,79 ± 0,16 <sup>a</sup>	10,33 ± 1,52 <sup>a</sup>
	9	20,16 ± 1,60 <sup>b</sup>	16,66 ± 2,75 <sup>b</sup>	9,08 ± 1,76 <sup>b</sup>	2,62 ± 0,55 <sup>a</sup>	10 ± 1 <sup>a</sup>
<i>Rosmarinus officinalis</i> with <i>Plantago coronopus</i>	0	24,66 ± 1,15 <sup>a</sup>	32,53 ± 1,5 <sup>a</sup>	24,25 ± 1,32 <sup>a</sup>	2,96 ± 0,66 <sup>a</sup>	15 ± 1 <sup>a</sup>
	3	21,33 ± 2,51 <sup>a</sup>	26,4 ± 0,79 <sup>b</sup>	17,31 ± 2,02 <sup>b</sup>	2,04 ± 0,19 <sup>a</sup>	14,33 ± 2,08 <sup>a</sup>
	6	21 ± 1,73 <sup>a</sup>	25,5 ± 1,8 <sup>b</sup>	16,14 ± 3,64 <sup>b</sup>	1,87 ± 0,66 <sup>a</sup>	10,66 ± 1,15 <sup>b</sup>
	9	21 ± 2 <sup>a</sup>	22 ± 1,73 <sup>bc</sup>	12,74 ± 1,11 <sup>b</sup>	1,67 ± 0,26 <sup>a</sup>	9,33 ± 1,15 <sup>b</sup>
<i>Rosmarinus officinalis</i> with <i>Spergularia salina</i>	0	34 ± 2,5 <sup>a</sup>	35,1 ± 4,65 <sup>a</sup>	24 ± 2 <sup>a</sup>	3,99 ± 0,65 <sup>a</sup>	12,66 ± 2,51 <sup>a</sup>
	3	26,95 ± 1,92 <sup>b</sup>	32,63 ± 2,76 <sup>a</sup>	17,36 ± 2,92 <sup>a</sup>	2,69 ± 0,99 <sup>a</sup>	10,66 ± 1,15 <sup>a</sup>
	6	25,66 ± 3,05 <sup>b</sup>	24,5 ± 0,43 <sup>b</sup>	16,18 ± 1,05 <sup>b</sup>	1,97 ± 0,19 <sup>b</sup>	10,33 ± 1,52 <sup>a</sup>
	9	24,33 ± 0,76 <sup>b</sup>	20,5 ± 1,5 <sup>c</sup>	15,41 ± 2,99 <sup>b</sup>	1,94 ± 0,38 <sup>b</sup>	10 ± 0,5 <sup>a</sup>

Values with the same letter for each species in the column are not significantly different ( $P \geq 0,05$ ).

**Table 2:** Effects of Salt Stress on Physiological Parameters (Relative Water Content of Leaves, Stem, Root and Sugar Content in Leaves) in the Three Experiments.

Experiment	NaCl (g/l)	Relative Water Content (RWC) in Leaves (%)	Relative Water Content (RWC) in Stem (%)	Relative Water Content (RWC) in Root (%)	Sugar Content in Leaves mg/ml MF
<i>Rosmarinus officinalis</i> alone	0	83,19 ± 1,22 <sup>a</sup>	77,47 ± 0,74 <sup>a</sup>	85,62 ± 2,15 <sup>a</sup>	0,70 ± 0,14 <sup>a</sup>
	3	76,75 ± 1,49 <sup>b</sup>	67,19 ± 1,11 <sup>b</sup>	81,26 ± 1,9 <sup>a</sup>	1,42 ± 0,16 <sup>b</sup>
	6	72,10 ± 0,89 <sup>c</sup>	40,96 ± 1,55 <sup>c</sup>	79,59 ± 3,24 <sup>ab</sup>	1,75 ± 0,31 <sup>b</sup>
	9	49,16 ± 0,77 <sup>d</sup>	18,91 ± 1,24 <sup>d</sup>	71,37 ± 1,52 <sup>c</sup>	2,46 ± 0,30 <sup>c</sup>
<i>Rosmarinus officinalis</i> with <i>Plantago coronopus</i>	0	55,43 ± 1,16 <sup>a</sup>	42,28 ± 0,67 <sup>a</sup>	81,81 ± 2,13 <sup>a</sup>	1,09 ± 0,24 <sup>a</sup>
	3	25,14 ± 0,98 <sup>b</sup>	33,69 ± 1,51 <sup>b</sup>	73,10 ± 0,66 <sup>b</sup>	0,66 ± 0,20 <sup>ab</sup>
	6	18,17 ± 0,83 <sup>c</sup>	32,75 ± 1,22 <sup>b</sup>	73,13 ± 0,80 <sup>b</sup>	0,54 ± 0,09 <sup>b</sup>
	9	18,12 ± 1,01 <sup>c</sup>	10,96 ± 1,24 <sup>c</sup>	68,11 ± 1,07 <sup>c</sup>	0,33 ± 0,04 <sup>bc</sup>
<i>Rosmarinus officinalis</i> with <i>Spergularia salina</i>	0	84,07 ± 1,09 <sup>a</sup>	50,05 ± 0,96 <sup>a</sup>	85,35 ± 0,76 <sup>a</sup>	0,62 ± 0,09 <sup>a</sup>
	3	42,52 ± 1,54 <sup>b</sup>	45,82 ± 1,34 <sup>b</sup>	81,53 ± 0,63 <sup>b</sup>	1,03 ± 0,05 <sup>a</sup>
	6	22,64 ± 1,29 <sup>c</sup>	37,93 ± 1,30 <sup>c</sup>	73,39 ± 1,35 <sup>c</sup>	1,08 ± 0,17 <sup>a</sup>
	9	18,28 ± 1,87 <sup>d</sup>	25,21 ± 1,73 <sup>d</sup>	71,21 ± 0,78 <sup>c</sup>	2,35 ± 0,29 <sup>b</sup>

Values with the same letter for each species in the column are not significantly different ( $P \geq 0,05$ ).

**Table 3:** Mean squares for eleven parameters in plant association evaluated in different treatments.

Source of variation	df	Root length	Stem length	Root weight	Stem weight	Weight of leaves	NB of leaves	Root volume	Sugar content	Relative Water Content Root	Relative Water Content Stem	Relative Water Content Leaves
<b>Plant</b>	2	88.47***	11.85 <sup>ns</sup>	20.38*	1.36*	34.44***	52537.59***	3.43*	2.52***	82.01***	1294.73** *	5116.12***
<b>Treatment</b>	3	123.88***	259.80***	193.29***	3.79***	24.32***	87534.35***	14.51**	1.19***	283.56***	1543.85** *	3379.44***
<b>Plant * Treatment</b>	6	12.41*	18.59**	20.54*	0.330 <sup>ns</sup>	7.42***	38620.31***	6.540 <sup>ns</sup>	0.99***	13.47**	163.310 <sup>ns</sup>	384.95***
<b>Error</b>	23	4.791	4.717	5.221	0.402	0.543	2276.138	3.159	0.042	2.643	105.458	1.485

ns: not significant; \* significant at 5%; \*\* highly significant at 1%, \*\*\* very highly significant at 1 %.

**Table 4:** Correlation coefficients of Pearson among the morpho-physiological parameters studied.

	Stem length	Root weight	Stem weight	Weight of leaves	NB of leaves	Root volume	Sugar content	Relative Water Content Root	Relative Water Content Stem	Relative Water Content Leaves
<b>Root length</b>	0.625***	0.618***	0.580***	0.552***	0.597***	0.047	-0.241	0.661***	0.553**	0.517**
<b>Stem length</b>		0.735***	0.582***	0.567***	0.514**	0.105	-0.478**	0.833***	0.503**	0.573***
<b>Root weight</b>			0.432**	0.493**	0.410*	0.338*	-0.418*	0.651***	0.541**	0.429**
<b>Stem weight</b>				0.607***	0.658***	0.223	-0.041	0.657***	0.453**	0.691***
<b>Weight of leaves</b>					0.808***	-0.027	-0.107	0.722***	0.630***	0.892***
<b>NB of leaves</b>						0.005	-0.361*	0.609***	0.556**	0.707***
<b>Root volume</b>							-0.002	0.229	0.302	0.151
<b>Sugar content</b>								-0.181	-0.145	0.044
<b>Relative Water Content Root</b>									0.738***	0.839***
<b>Relative Water Content Stem</b>										0.662***

\*, \*\*, \*\*\*: correlation is significant at 5, 0.1, and 0.01%, respectively.



#### 4. Conclusions

In conclusion, our study highlights the negative impact of salt stress on the growth and development of *Rosmarinus officinalis*. In response to this environmental constraint, by associating *Rosmarinus officinalis* with certain halophytic plants, such as *Plantago coronopus* and *Spergularia salina*, during cultivation under salt stress, we could mitigate the detrimental side effects and enhance the overall resilience of the plant. These findings offer promising prospects for developing agriculture that is more resistant to saline constraints and underscore the importance of studying interactions between different plant species for better management of stressful environments. By doing so, we can envision more sustainable and efficient agricultural practices, contributing to food security and the preservation of ecosystems in a world facing increasing environmental challenges.

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