



# Article The Effects of Suaeda salsa/Zea mays L. Intercropping on Plant Growth and Soil Chemical Characteristics in Saline Soil

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Abstract: Halophytes possess the capacity to uptake high levels of salt through physiological processes and their root architecture. Here, we investigated whether halophyte/non-halophyte intercropping in saline soil benefits plant growth and contains root-dialogue between interspecific species. Field and pot experiments were conducted to determine the plant biomasses and salt and nutrient distributions in three suaeda (*Suaeda salsa*)/maize (*Zea mays* L.) intercropping systems, set up by non-barrier, nylon-barrier, and plastic-barrier between plant roots. The suaeda/maize intercropping obviously transferred more Na<sup>+</sup> to the suaeda root zone and decreased salt and Na<sup>+</sup> contents. However, the biomass of the non-barrier-treated maize was significantly lower than that of the nylon and plastic barrier-treated maize. There was lower available N content in the soil of the non-barrier treated groups compared with the plastic barrier-treated groups. In addition, the pH was lower, and the available nutrient content was higher in the nylon barrier, which suggested that rhizospheric processes might occur between the two species. Therefore, we concluded that the suaeda/maize intercropping would be beneficial to the salt removal, but it caused an adverse effect for maize growth due to interspecific competition, and also revealed potential rhizospheric effects through the role of roots. This study provides an effective way for the improvement of saline land.

Keywords: maize; intercropping; Suaeda salsa; saline soil

# 1. Introduction

Salt stress is an abiotic factor that limits crop productivity and agricultural development worldwide. How to improve soil quality and the viability of non-halophytes (crops) in saline land is always an important issue. Previous studies showed that halophyte is a plant that can complete its entire life cycle on a high-salt soil owing to a series of adaptive strategies in its coordinated evolution with the environment [1-3]. Halophytes develop special mechanisms to resist and alleviate salinity stress over evolutionary time: (i) cells produce some osmotic substances to reduce water potential, and would favor the uptake of water from the outside; and (ii) halophytes can absorb Na<sup>+</sup> and compartmentalize it into vacuoles by an Na<sup>+</sup>/H<sup>+</sup> antiporter [4–6]. They can survive in media containing more than 200 mM NaCl and direct root damage in the surficial soil [7–9]. Wang et al. (2021) found that the decrement in salt content has occurred after halophytes were initially cultivated in virgin saline soil [10]. Etesami and Beattie (2018) reported that halophyte salinity-tolerant microorganisms improve non-halophyte adaptability to saline soils [11]. Shultana et al. (2020) also determined that the salt-tolerant PGPR (Bacillus tequilensis and Bacillus aryab*hattai* strains) produces a positive role on photosynthesis and stomatal conductance to increase the rice yield [12]. This gives us an idea that the efficient use of halophytes helps decrease the salt concentrations of saline soils, which might optimize the soil conditions for non-halophytes by intercropping system.



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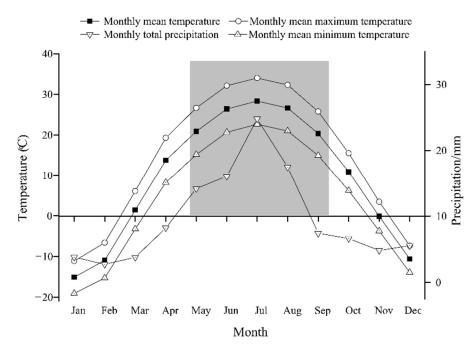
Generally, intercropping or crop rotations has greatly induced plant growth, and the interspecific interactions give some crops nutrient competitive advantages and significantly superior yield levels [13–16]. The intercropping of water spinach and corn not only significantly weakens the toxic action of nitrate in the former plant, and it also reduces the nitrate accumulation in the soil [17]. These intercropping patterns usually involve rhizospheric dialogues, in which root exudates play important roles in delivering soil nutrients [18,19]. Sufficient transformational levels of nitrogen (N), phosphorus (P), and potassium (K) are necessary to drive vegetative growth that promotes further nutrient acquisition in a plantroot system [20,21]. Non-halophytes might increase the soil nutrient availability by root exudates, which is good for halophyte requirements in intercropping systems. Liang and Shi (2021) also discovered an intercropping system involving cotton and halophytes, which significantly influenced salt removal, and accelerated crop productivity in saline soil [22]. Under the salt stress, halophytes are subject to the base ions (Na<sup>+</sup> and Cl<sup>-</sup>), leading to osmotic stress. Thus, roots may produce excretions in response to the environment, which effectively are associated with soil salt, microbes, and nutrients [19,23]. Root excretions are necessary for altering and adapting to salt conditions to enhance the availability of soil nutrients and microbial activities [24,25], which affects root metabolism and development of non-halophytes. However, it is difficult to extrapolate the promotive effects on nonhalophytes in intercropping systems that contain distinct species. In intercropping systems, whether or not halophytes can reduce the salt content of saline soil through salt absorption to reduce the salt stress of non-halophytes, is an issue that should be further investigated.

*Suaeda salsa*, an annual halophyte, could complete body development in saline soil, and is able to assimilate Na<sup>+</sup>, which is considered an ideal halophyte for improving saline soil [23,26]. Maize always has an important influence on nutrients through rhizospheric activities in intercropping systems. How do the soil properties and the growth of two species vary in the suaeda and maize intercropping? We hypothesized that the suaeda and maize intercropping would decrease the soil salt content on the maize side, and increase nutrient movements through root interactions. In this study, three treatments (non-barrier, nylon-barrier, and plastic-barrier) between intercropping plant roots were adopted as per previous studies [14,27]. The objectives were as follows: (a) to analyze the effect of the suaeda/maize intercropping on biomasses; (b) to investigate the variations in pH and salt between the two root systems; and (c) to assess the interspecific competition or cooperation for nutrients (available N and P) in the suaeda and maize intercropping system.

## 2. Materials and Methods

## 2.1. Field Experiment

The study was conducted at the Salt Farm Botanical Garden in Karamay (45°28′6.38″ N, 84°59′41.61″ E), Xinjiang Province, China. The study area is characterized by cold, windy winters and hot, dry summers. The annual mean temperature and precipitation are 8.1 °C and 108.9 mm (Figure 1), respectively, and the light and heat are sufficient in this region, with 2734 h of sunshine, 200 frost-free d, and effective and accumulated temperatures of up to 3968.1 °C. The soil chemical and physical properties in this experiment were measured, as indicated in Table 1.



**Figure 1.** Monthly total precipitation, mean temperature, and maximum and minimum temperatures in the study area from NOAA's National Centers for Environmental Information (NCEI). The shadow represents the main growing season of halophyte from May to Sep.

Table 1. The field soil's chemical and physical properties.

Soil Depth (cm)	pН	EC (dS m <sup>-1</sup> )	Total Salt (g kg <sup>-1</sup> )	Available N (mg kg <sup>-1</sup> )	Olsen-P (mg kg <sup>-1</sup> )	Available K (mg kg <sup>-1</sup> )
0–10	7.247	1.260	1.308	7.556	5.282	2.858
10-20	7.403	1.027	2.567	13.208	5.369	3.099
20-30	7.383	0.616	1.833	7.032	4.876	2.336
30-50	7.373	0.691	2.025	6.308	4.110	2.448
50-100	7.490	0.582	1.570	4.964	4.683	2.691

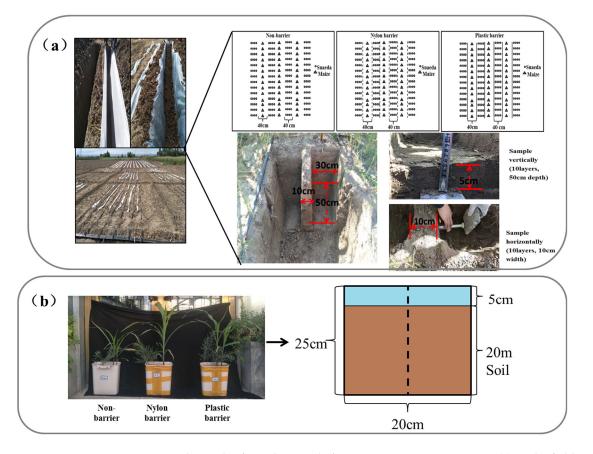
Field experimental methods were used to analyze the effects of halophyte (suaeda) and non-halophyte (maize) intercropping systems on plant growth and soil chemical characteristics. Additionally, the root systems of the two species were separated by placing different materials between them, as follows: (1) the control, not separated (non-barrier treatment): the root systems of the different plants coexisted, allowing direct competitive and promotive effects; (2) roots separated by a 30  $\mu$ m nylon mesh (nylon-barrier treatment): the transmission of soil nutrients and microbial products could proceed belowground; and (3) roots separated by a plastic film (plastic-barrier treatment): there was no interaction between the roots of the two species. These conditions would expose the regulatory mechanisms behind the interactions at the root–soil interface of halophytes and non-halophytes.

The experiment had a split-plot design, with nine plots, and three replicates. The area of each individual plot was 6 m × 5.6 m. To satisfy nutrient requirements, the site was treated with urea of 590 kg ha<sup>-1</sup> (N  $\geq$  46%), potassium sulfate of 163 kg·ha<sup>-1</sup> (K<sub>2</sub>O  $\geq$  50%) and superphosphate of 150 kg ha<sup>-1</sup> (P<sub>2</sub>O<sub>5</sub>  $\geq$  14%). The nylon and plastic barriers were placed in the soil at 1 m depths, and the inter-row distance was 40 cm.

The suaeda seeds were gathered in October 2018 from a botanical garden in Karamay, Xinjiang Province, China. The maize seeds (Cultivars: Silage Tieyan 53) were gather from Hejiayuan Agricultural Science and Technology Co. LTD, Beijing, China. Suaeda was sown in May 2019, and maize was sown 15 d later. Each intercropped plot included four rows of suaeda and three rows of maize. All rows of plant were irrigated with each drip line, and the interval of drip line and the inter-species distance were 40 cm. The harvesting and gathering of root and soil samples were completed in August 2019. During the growth period, the plots were manually irrigated and weeded regularly with fresh water of 8 m<sup>3</sup>, where the discharge rate of emitters was 2 L h<sup>-1</sup>.

The shoot biomasses of the two species were determined by harvesting from the intercropping systems after 60 d of growth. To clearly determine the root spatial distributions and correlations with soil properties, the soil was stratified in the vertical direction to acquire the roots. At each plot, two  $30 \times 30 \times 50$  cm soil samples were excavated for suaeda and for maize in the adjacent row. The samples were stratified every 5 cm and then divided into 10 parts. The roots were obtained from the soil samples and washed free of soil. All the samples were over dried by treating at 105 °C for 30 min and then dried at 65 °C for 48 h.

The potential deliveries of soil substances were investigated between the two species by horizontal soil stratification. The junction between the two root systems was determined to occur at a 10 cm depth in the middle of the plants. The 5 cm topsoil layer was initially removed and then the soil was divided into ten 1 cm thick layers (Figure 2).



**Figure 2.** Soil samples from the suaeda/maize intercropping systems: (**a**) In the field experiment, soil samples were collected horizontally at 5 cm depths and vertically at 1 cm depths. (**b**) The pot experiment was conducted in concert with the field experiment.

The soil pH was measured using a pH meter. The soil conductivity (EC) and slat content were measured using a conductivity meter and residue drying quality method (10 g soil in 50 mL water), and diluted extracts were analyzed for Na<sup>+</sup> (Flame Photometer, 735 ICP-OES). The available N was measured using the alkali hydrolysis diffusion method (5 g soil in 50 mL solution). The available P (Olsen-P) was measured using the molybdenum antimony anti-colorimetric method (2.5 g soil in 50 mL solution) [28].

#### 2.2. Pot Experiment

A pot experiment was conducted in 2019 to verify the field results. Saline soil was collected from the experimental station at Changji, Xinjiang Province, China (44°09′59′′ N, 87°04′56′′ E). Then, the soil was air-dried and passed through a 2 mm sieve. The soil properties were as follows: pH, 7.75; EC, 1.32 dS m<sup>-1</sup>; total salt, 0.50%; available N (NO<sub>3</sub><sup>-</sup> and NH<sub>4</sub><sup>+</sup>), 33.68 mg kg<sup>-1</sup>; Olsen-P, 4.62 mg kg<sup>-1</sup>; and available K, 0.25 g kg<sup>-1</sup>.

To identify variations in plant growth in the suaeda and maize intercropping, the pot experiment was established using three inter-species root treatments. The intercropping system's equipment was shown in Figure 2b. The pot tube had an inner length of 20 cm, width of 10 cm, and height of 25 cm. The cultivars of maize and suaeda seeds were consistent with those in the field experiment.

To ensure an adequate nutrient supply for plant growth, the soil was fertilized with 0.20 g kg<sup>-1</sup> of N and 0.15 g kg<sup>-1</sup> of P<sub>2</sub>O<sub>5</sub> [29]. The pot was filled with 8.0 kg of airdried soil, which was divided using a nylon or plastic barrier in the middle position. We always established three treatments (non-barrier, nylon-barrier, and plastic-barrier) and four replicates for each treatment. Then, two liters of water was added, and the soil was allowed to stand for 5 d. In July 2019, on one side per pot, 20 suaeda seeds were sown and grown to 4 cm before thinning. After 15 d, on the other side of each pot, four maize seeds were sown. The weighing method was used to supply water to field capacity (20%, w/w) to reduce the influence of water on plant growth.

The aboveground parts of suaeda and maize were harvested at approximate 45 d after sowing. All the samples were treated at 105 °C for 30 min and then dried at 65 °C for 48 h, and the rhizosphere soil of suaeda and maize were collected by shaking off the soil adhering to the root surface (0–3 mm) [30]. Then, the determination methods of soil chemical characteristics were consistent with that in the field experiment.

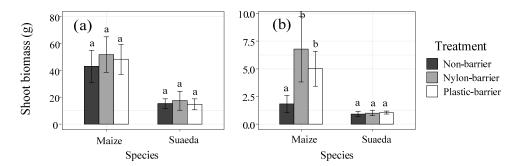
#### 2.3. Statistical Analyses

An analysis of variance was used to determine the differences in the shoot, soil salt, and nutrient contents among the three treatments with SPSS statistical software (version 19.0, IBM SPSS Inc, Chicago, IL, USA) and R software (version 4.0.3-win, R Foundation for Statistical Computing, Vienna, Austria). Significant differences among means were separated using least-significant difference (LSD) at the p < 0.05 probability level. Using the smooth fit method, we also detected spatial variations in the soil pH, salt content, and nutrients of the two species.

## 3. Results

#### 3.1. Plant Biomasses

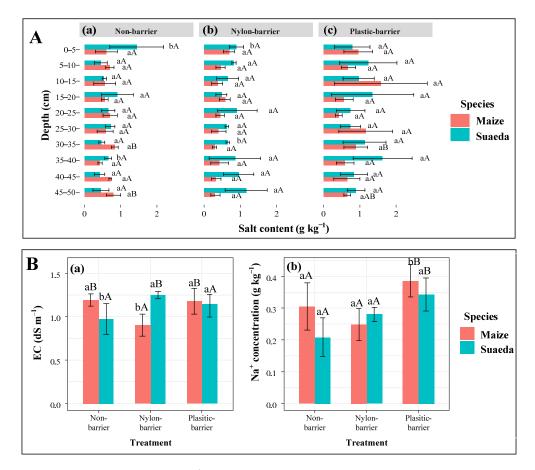
The suaeda/maize shoot biomasses and N absorptions were determined in the three intercropping systems (Figure 3). The maize biomass in the non-barrier treatment was 11.2% and 17.1% lower than that in the plastic-barrier and nylon-barrier treatment, respectively (Figure 3a). The plant development observed in the pot experiment was different to that observed in the field experiment. There was not a significant difference in the suaeda biomasses, but the maize biomass in the non-barrier treatment was significantly lower 3.2 g per plant than in the plastic-barrier treatment (Figure 3b).



**Figure 3.** The shoot biomasses of maize and suaeda in the different treatments: (**a**) Field experiments; (**b**) Pot experiments. Lower case letters on the column indicate the significant differences among three treatments.

### 3.2. Soil EC, Salt, and Na<sup>+</sup> Contents

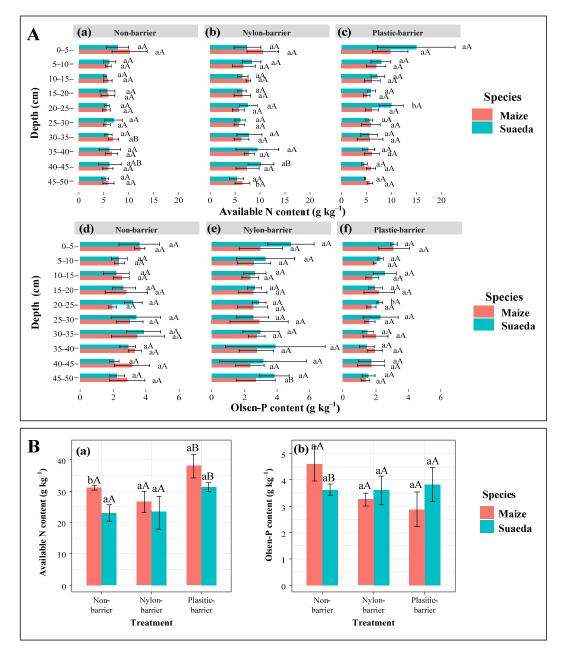
The salt contents in the different suaeda-associated soil layers in the non-barrier treatment were lower than those in the nylon- and plastic-barrier treatments, as measured in the 5–15 cm and 20–50 cm soil layers (Figure 4A(a–c)). Compared with the plastic-barrier treatment, the nylon-barrier treatment resulted in a higher soil salt concentration on the suaeda side than on the maize side (Figure 4A(b)), and the EC and Na<sup>+</sup> values were significantly higher (exceeded 0.345 dS m<sup>-1</sup>, 0.032 g kg<sup>-1</sup>) in the pot experiments (Figure 4B(a)). The Na<sup>+</sup> concentration displayed lower in the non-barrier treatment than that in the plastic-barrier (Figure 4B(b)).



**Figure 4.** The soil salt, EC, and Na<sup>+</sup> in the different treatments: (A(a-c)) Salt content in non-, nylon-, and plastic-barrier treatment of field experiment; (B(a,b)) EC, Na<sup>+</sup> content of pot experiment. Capital letters on the column indicate the significant difference differences among three treatments. Lower case letters on the column indicate significant differences between two species.

## 3.3. Soil Available N and Olsen-P Contents

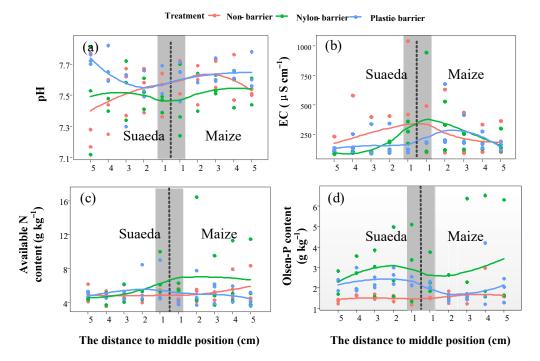
Differences in the available N and Olsen-P contents in the three intercropping systems existed to a certain extent (Figure 5). The soil available N content in the non-barrier treatment was also  $0.0104 \text{ g kg}^{-1}$  lower than in the plastic-barrier treatment (Figure 5A(b,c)). The similar results were found in pot experiment as showed in Figure 5B(a). The Olsen-P content in the plastic-barrier treatment was lower than in the non-barrier and nylon-barrier treatments though there was not significant (Figure 5A(d-f)).



**Figure 5.** The available N and Olsen-P contents in the different treatments: (A(a-c)) Available N contents in non-, nylon-, and plastic-barrier treatment of field experiment; (A(d-f)) Olsen-P contents in non-, nylon-, and plastic-barrier treatment of field experiment; (B(a,b)) Available N, Olsen-P concentration of pot experiment. Capital letters on the column indicate the significant differences among three treatments. Lower case letters on the column indicate significant differences between two species.

#### 3.4. Horizontal Variations in Soil Properties

An increasing trend in soil pH was found in the horizontal direction from suaeda to maize under non-barrier treatment conditions (Figure 6a), but the pH value was lower, and the EC was higher at the interface near the nylon barrier (Figure 6b). An increasing trend in available N developed from suaeda to maize in the nylon-barrier treatments (Figure 6c). As a whole, the Olsen-P in the nylon-barrier treatment was significantly greater than in the non-barrier and plastic-barrier treatments (Figure 6d). Higher available N, Olsen-P levels were found at the interface near the nylon barrier, which also confirmed the possibility of synergy in the non-barrier treatment through the physiological root-dialogue process (Figure 6c,d).



**Figure 6.** Variations in soil properties in the different treatments: (**a**) pH; (**b**) EC; and (**c**) available N content; (**d**) Olsen-P content of the soil from the middle positions. Dotted black lines and gray shadowing represent the middle positions and the interfaces of the two species, respectively, in the different treatments.

## 4. Discussion

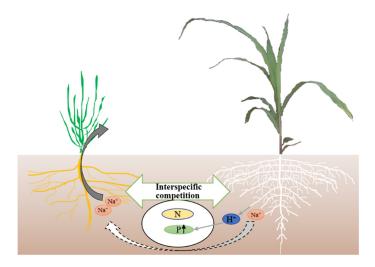
## 4.1. The Effects of Intercropping Systems on Biomass

The promotion of non-halophyte growth was not found in the suaeda/maize intercropping systems, which was inconsistent with the results of previous studies [31,32]. In this study, the non-barrier treated maize biomass was lower than that during the plastic-barrier treatment (Figure 2). Root interaction of two species may have restrained each other's growth in the intercropping systems. Roots of the halophyte have their own adaptive strategy-related responses to saline soil, and physiological responses to the nylon barrier may also exist. A greater salt content near suaeda roots influences maize belowground growth and suppresses the shoot biomass [33]. In addition, inhibitory effects during the intercropping are greatly affected by the planting distance and density, as well as the plant species [34].

#### 4.2. The Soil Salt's Spatial Variations in The Intercropping Systems

The study was conducted to determine whether salt migrated from non-halophytes to halophytes in the intercropping systems. The salt level variation in the horizontal direction was not significant in the non-barrier and nylon-barrier treatments, but the salt content was higher at the 20 cm soil depth on the suaeda side compared to that on the maize side

(Figure 4A(b)). This suggests that suaeda has an important ability to gather salt to improve the saline soil. Salt-tolerance has evolved in a variety of vegetative types over long periods, during which non-halophytes might have lost their genetic variation and developed special traits related to high salt-tolerance, such as in crop plants [1,35,36]. Some glycophytes, such as maize and wheat, also tend to remove Na<sup>+</sup> from roots [37]. Suaeda appears to gather and uptake salt into the root zone [9,38], which might reduce the salt injury level and improves the saline-related root environment for maize. This would be explained by the results in the pot experiments that the Na<sup>+</sup> content displayed higher on the suaeda sides (Figure 4B(b)). This indicates that the intercropping was more beneficial to the absorption of salt and Na<sup>+</sup> by suaeda (Figure 7). In addition, it suggests that Na<sup>+</sup> is an essential factor for suaeda growth and metabolism during the life cycle. However, though the decrement in salt content was displayed, the maize growth was obviously inhibited in the non-barrier treatment, which might have been caused by interspecific competition (Figure 5). Ai et al. discovered that the intercropping disadvantage of jujube was due to niche invasion of cotton, which increased the competition for water [39].



**Figure 7.** The potential mechanism in the suaeda/maize intercropping systems. Maize's excretion decreases soil pH to activate more Olsen-P, but it facilitates the Na<sup>+</sup> shift to the suaeda root zone and interspecific competition for nutrients.

## 4.3. Soil Nutrients' Variations in The Suaeda/Maize Intercropping Systems

The variations in the available N were obvious among the three intercropping systems. The soil available N was lower in the non-barrier treatment than that in the plastic treatment, which suggests that suaeda/maize intercropping facilitated interspecific competition to a certain content. Root interactions in an intercropping system may improve resource acquisition and adaptation to nutrient constraints [40,41]. For instance, the disadvantage of maize/barley intercropping was mainly due to the increased interspecific competition for N, P, and K between the two plants [42].

However, the competition of soil nutrients between maize and suaeda might be reflected in nitrogen, but not phosphorus. The trends in the soil Olsen-P in the intercropping system were not similar to those of the available N. The soil Olsen-P levels in the non-barrier treatment were higher than in the plastic-barrier treatment (Figure 5), which indicates that P release and absorption were accelerated by root interactions in the suaeda/maize intercropping systems. The significantly higher Olsen-P levels in these systems indicates that intercropping may increase the available nutrient levels, but these results are not consistent with those of previous studies [43–45]. The higher P level on the suaeda side of the nylon barrier, compared with the maize side, may be attributed to maize excretions that acidize the salt. The prominent impact of pH on soil Olsen-P in the rhizosphere has been demonstrated previously [46]. It could be illustrated that a high nutrient availability was

displayed at the interface near the nylon barrier, and this was coincident with the low pH value (Figure 6). This suggested that suaeda/maize intercropping shifted nutrients and ions between the inter-species roots, as reported in previous researches [14,31,47,48]. In this study, though we observed a negative effect on maize growth, which was probably due to the nitrogen competition, the issues regarding the role that the root excretions played, and how the excretion–microorganism–soil properties related in this system, were unclear, which should be explored to deeply figure out the mechanism of halophyte/non-halophyte intercropping in subsequent studies.

### 5. Conclusions

From the experimental data, it can be shown that there is an obvious interspecific ecological competition between suaeda and maize. The decrements in salt and Na<sup>+</sup> content in the non-barrier treatment suggest that the intercropping plays a positive effect on salt absorption. Comparing the experimental results of nylon barrier treatment with other treatments, it can be shown that the roots of suaeda and maize interact through the exchange of soluble substances. This study on suaeda/maize intercropping may provide new insights into the amelioration of saline soil. Whether it is possible to change the planting pattern (row spacing, fertilization method, etc.) of maize/suaeda or crop selection to address the intercropping disadvantage of maize is a focus of further research.

**Author Contributions:** Conceptualization, S.W., W.M. and C.T.; methodology, S.W., Z.Z. and W.M.; software, S.W. and S.G.; validation, Z.Z. and C.T.; formal analysis, S.W.; investigation, S.W., S.G. and K.Z.; writing—original draft preparation, S.W.; writing—review and editing, S.W., W.M. and C.T.; visualization, S.W. and S.G.; supervision, Z.Z. and C.T. All authors have read and agreed to the published version of the manuscript.

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