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Salt spray as a micro-environmental factor affecting the growth of *Commelina maritima* L. at Lekki Beach, Nigeria

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***Commelina maritima* L. is a dominant perennial halophyte that is restricted to sandy beaches where it plays major ecological roles in Southern Nigeria. This study examines the response of this plant to saltwater sprays, a factor that affects the growth of coastal plants. Plants were sprayed with seawater twice per week (2/week), four times per week (4/week) or six times per week (6/week) while control was sprayed without seawater (de-mineralized water) six times per week (0/wk). Survival, growth and biomass allocation of the plants were determined. Salt spray did not affect plant survival but significantly ($p \leq 0.05$) decreased number of leaves, shoot length, stem girth, leaf area and root growth. Relative growth rate and number of branches were not significantly ($p \geq 0.05$) affected by salt spray. Sea spray significantly ($p \leq 0.05$) reduced fresh and dry mass of plant parts, total biomass and leaf total chlorophyll when compared with the control. Root : shoot ratio increased significantly ($p \leq 0.05$) under seawater treatment as the shoot growth was more negatively affected than root growth. Relatively more biomass was allocated to the root than shoot in seawater-treated plants. Salt spray increased shoot ash content and negatively affected plant organic content. *C. maritima* can be classified as a salt spray-sensitive plant. Salt spray is a micro-environmental factor affecting its survival and growth, thus influencing its distribution in strandline.**

Key words: Strandline, survival, growth, biomass allocation, Commelinaceae, distribution.

INTRODUCTION

Strandlines are created along the high water mark on a range of habitats including coastal vegetated shingle, coastal sand dunes and coastal salt marshes (Lee and Ignaciuk, 1985). Several natural and anthropogenic factors affect the vegetation of strandlines (Owen et al., 2001; Peter et al., 2003; Pichler and Oberhuber, 2007). Studies on coastal plant communities in many parts of the world have found that air-borne saltwater spray is an important natural selective abiotic factor (Boyce, 1954; Cartica and Quinn, 1980; Sykes and Wilson, 1988; Hesp,

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1991). Salt-sensitive species can be eliminated entirely from areas with high salt spray so that plants adapted to salt spray grow close to the ocean and are replaced by less salt-resistant plants farther inland (Cheplick and White, 2002). It has been found that natural salt spray accumulation on plants in the field can cause water stress, leaf necrosis, growth reduction and death depending on the frequency and severity of storms and winds that carry aerial salt drift inland (Griffiths and Orians, 2004; Griffiths, 2006).

Strandline community composition is structured by the effects of salt spray, and plants typical of coastal beaches are salt spray tolerant, which gives them a competitive advantage over others (Tominaga and Ueki, 1991; Greaver and Sternberg, 2007; De Vos et al., 2010). To date, the possible influence of natural processes on coastal vegetation has largely focused on soil salinity while air-borne salinity has been largely ignored.

Commelina maritima L. commonly called white mouth dayflower belongs to the family Commelinaceae. It is a fleshy, creeping, almost glabrous herb with blue flowers, in coastal sand bar vegetation. It is perennial and monocotyledonous with succulent stem and swollen nodes often mucilaginous (Hutchinson et al., 1986). This plant provides essential habitat, spawning sites and food for many diverse species of terrestrial and marine wildlife. The plant is able to withstand periodic coastal disturbances and it is particularly important on exposed shores, where its population can act as precursors to sand dunes. It helps in storm protection and controls shoreline erosion. It helps in shoreline stabilization by enhancing organic and moisture content; pioneer plants can then become established and sand dune formation is initiated (Williamson, 2005). *C. maritima* is widely distributed in West Tropical Africa where it is found a short distance from sea on the sandy beaches (Hutchinson et al., 1968). This plant species is dominant, wide spread and restricted to strandline habitat in Nigeria. We hypothesize that salt spray is a micro-environmental factor affecting its growth. This is because strand vegetation is affected by environmental factors such as soil salinity, sand burial, drought, waterlogging, nutrient deficiency, trampling and other anthropogenic activities. Each factor is usually considered as a micro-environmental factor among the various environmental conditions that affect strand vegetation (Barbour, 1978). It was reported that the death of *Impereta cylindrica* and *Miscanthus sinensis* transplanted to beach was as a result of salt spray, inhibiting them from becoming established on the front dunes. They concluded that salt spray is among the non-negligible factors controlling the distribution of plants in sand dune vegetation (Ogura and Yura, 2007). Plant's ability to survive in the strandline is largely influenced by salt spray. Since *C. maritima* thrives exclusively in the environment, salt spray is a natural abiotic factor that influences its growth in the strandline (Barbour, 1978). We experimentally manipulated salt spray in laboratory studies and examined its influence on the survival, growth and biomass allocation of the plant species. The results are used to discuss its tolerance and some adaptations for survival.

MATERIALS AND METHODS

Stem cuttings of *C. maritima* collected from Lekki Beach in Lagos, Nigeria were used to raise uniform plants in perforated plastic pots filled with 2:1 ratio of washed river sand to top soil (Khan et al., 2000a, b). Seawater was collected off the shore, the source of the air-borne salts and stored in a plastic keg at room temperature. Salinity of the seawater was 30.00 ppt and later measurements with conductivity meter did not change with short time storage. Plants were randomly assigned to either salt-spray treatments or control. On Mondays and Thursdays, plants were sprayed with seawater twice per week (one seawater spray on each of the two days), four times per week (two seawater sprays on each of the two days) or six times per week (three seawater sprays on each of the two days) while control were sprayed without seawater six times per week (three de-ionized water prays on each of the two days) following the methods of Cheplick and Demetri (1999), Cheplick and White (2002) and De Vos et al. (2010). Individual plant was sprayed to run-off with all the aerial parts equally exposed. The accumulated salt onto the shoot for 1, 2 and 3 sprays was estimated following the method described by Cheplick and Demetri (1999), which equaled on average 4, 8 and 12 mg NaCl dm^{-2} leaf area day^{-1}. These fall within the levels found in the natural habitat of beach plants (Barbour et al., 1985; Griffiths, 2006). A plant mist bottle held about 20 cm from the shoot was used for spraying. Pots were arranged on the Greenhouse bench of the Plant Science and Biotechnology Department, Adekunle Ajasin University, Akungba Akoko, Ondo State Nigeria (Lat. 7° 28^1N, Long. 5° 44^1 E). Seawater was applied as a fine mist from a spray bottle held 20 cm from the side of each shoot. Plants in all treatments were repositioned after each saltwater treatment. Salt that might possibly deposit on soil during misting was flushed out weekly with water. There were 6 single-plant replicates per treatment in a completely randomized experimental design. Prior to beginning the salt spray treatments, five plants were harvested to determine the initial growth parameters and successive measurements were taken at an interval of two weeks.

Percentage survival and growth parameters were recorded. Shoot length was measured from the soil level to the terminal bud and leaf area was obtained with a leaf area meter. Stem girth was measured at about 10 cm from stem base using model 0-200 mm digital caliper. Number of leaves and lateral branches of individual plants were counted manually. The experiment was terminated at 12 weeks after initiation of treatment. Plants were harvest, major roots were counted and their lengths measured. The harvested plants were separated into leaves, stems and roots and their fresh mass determined. Dry mass was obtained after oven-drying to constant mass at 80°C. Root dry mass was divided by that of the shoot to obtain root : shoot ratio while total biomass of all plant parts was weighed. Relative growth rate was calculated using the formula, RGR (relative growth rate) = (ln mass2-ln mass1)/time.

Total chlorophyll was extracted in 80% (v/v) aqueous acetone and absorption was read in a spectrophotometer at 645 and 663 nm (Arnon, 1949). Each sample of the dried plant materials was ground to fine powder using Philips model blender. Percentage ash content was determined by weighing the plant sample (powder) in a Pyrex beaker and placed in muffle furnace set at 500°C. Ash and organic mass components were also determined. The soil physico-chemical properties were analyzed in the Central Laboratory of The National Institute for Oil Palm Research (NIFOR), Nigeria, following the standard methods of Association of Official Analytical Chemists

Table 1. The physico-chemical properties of soil used for planting.

Physico-chemical parameter	Value
pH	5.48
% C	3.67
N (ppm)	20.42
P (ppm)	3.56
K (meg/100 g)	3.56
Ca (meg/100 g)	2.32
Mg (meg/100 g)	2.60
CEC (meg/100 g)	8.2
Sand (%)	80.68
Silt (%)	12.06
Clay (%)	8.36

(AOAC, 1985). Data were subjected to single factor analysis of variance and means were separated with Duncan's MRT using SPSS version 17.0 (SPSS Inc., Chicago, IL, USA) at $P \leq 0.05$.

RESULTS

Table 1 shows the physico-chemical properties of the soil used for planting. The ratio 2:1 of washed river sand to top soil is typical of sandy beach soil in strandline. The sandy soil supported the plant growth while the top soil was for nutrient supply. The soil was low in nutrients in comparison with nutrient composition of agricultural soils. Damage caused by salt spray include leaf browning, chlorosis and necrosis. Salt spray led to leaf firing and leaf injury which resulted in early leaf senescence and defoliation. Leaf death and defoliation in plants sprayed with seawater resulted in leaf number reduction. Leaf number decreased with increase in level of seawater (Figure 1). One-way ANOVA however revealed that reduction in leaf number became significant ($p \leq 0.05$) only at 4/week and 6/week when compared with the control treatment.

Table 2 shows the influence of salt spray on the growth parameters of *C. maritime*. Generally, plant growth was inhibited by salt spray but there was no plant mortality. Leaf area decreased significantly ($p \leq 0.05$) in plants sprayed with seawater relative to the control. At the highest level of salt application, leaf area was reduced by as high as 53.45%. In plants exposed to salt spray, root growth was negatively affected. This is evidenced by a significantly ($p \leq 0.05$) reduced root length and root number under seawater treatment as compared to the control. There was however no statistical difference ($p \geq 0.05$) when different levels of salt treatments were compared. Plants sprayed with seawater exhibited a significantly reduced ($p \leq 0.05$) stem girth in comparison with those exposed to de-ionized water. Relative growth rate likewise decreased with increase in salt spray but there was no significant difference ($p \geq 0.05$) between control and seawater-treated plants. Total biomass and chlorophyll content of plants subjected to salt treatment were reduced significantly ($p \leq 0.05$) as compared to the control. Root : shoot ratio increased significantly ($p \leq 0.05$) under seawater treatments relative to de-mineralized water treatment. This revealed that shoot growth was more negatively affected by salt spray than root growth. Shoot length of *C. maritima* treated with seawater decreased progressively with increasing salt level. This became significant ($p \leq 0.05$) from eight weeks after treatment. At the end of the investigation, shoot length was reduced by 37.14% at 6/week relative to the 0/week treatment (Figure 2). However, plants sprayed with seawater had reduced number of branches but there was but there was no significant difference ($p \geq 0.05$) between the control and those treated with salt spray (Table 2).

Fresh and dry mass of plant parts declined progressively with increasing salt spray levels. One-way ANOVA showed that fresh and dry mass were significantly lower ($p \leq 0.05$) under salt sprays than in the control treatment (Table 3). Figure 3 shows the biomass allocation pattern of *C. maritime* under different spray treatments. Relatively more organic biomass was allocated to the root than the shoot in plants subjected to salt spray. This was due to salt accumulation on the shoot following foliar spray with seawater. This led to an increase in the shoot ash content thus having a negative effect on percentage organic content. Organic biomass allocation was not affected by salt treatment in roots since they were not in direct contact with salt. When compared with the control, shoot ash mass was significantly increased ($p \leq 0.05$) by salt sprays. However, there was no significant difference ($p \geq 0.05$) when root ash mass was compared between control and seawater treatments (Figure 4). Organic mass was significantly ($p \leq 0.05$) reduced both in the shoot and root due to the influence of salt deposition on shoot surfaces (Figure 5).

DISCUSSION

The ratio 2:1 washed river sand to top soil has been reported to have properties of beach soil where strand plants naturally grow (Khan et al., 2000a, b). Although, the soil was low in nutrients, strand plants have low nutrient requirements for growth (Lee and Ignaciuk, 1985). Since no negative effect was observed in control plants, the observed effect on the plant was not due to nutrient unavailability but salt spray. The changes in plant morphology due to salt sprays are consistent with the previous studies demonstrating damage to plants as a result of salt spray accumulation (Griffiths, 2006). Necrosis on *Myrica pensylvanica* leaves decreased as distance from the dune crest increased (Griffiths and Orians, 2003). Sea spray likewise resulted in leaf necrotic

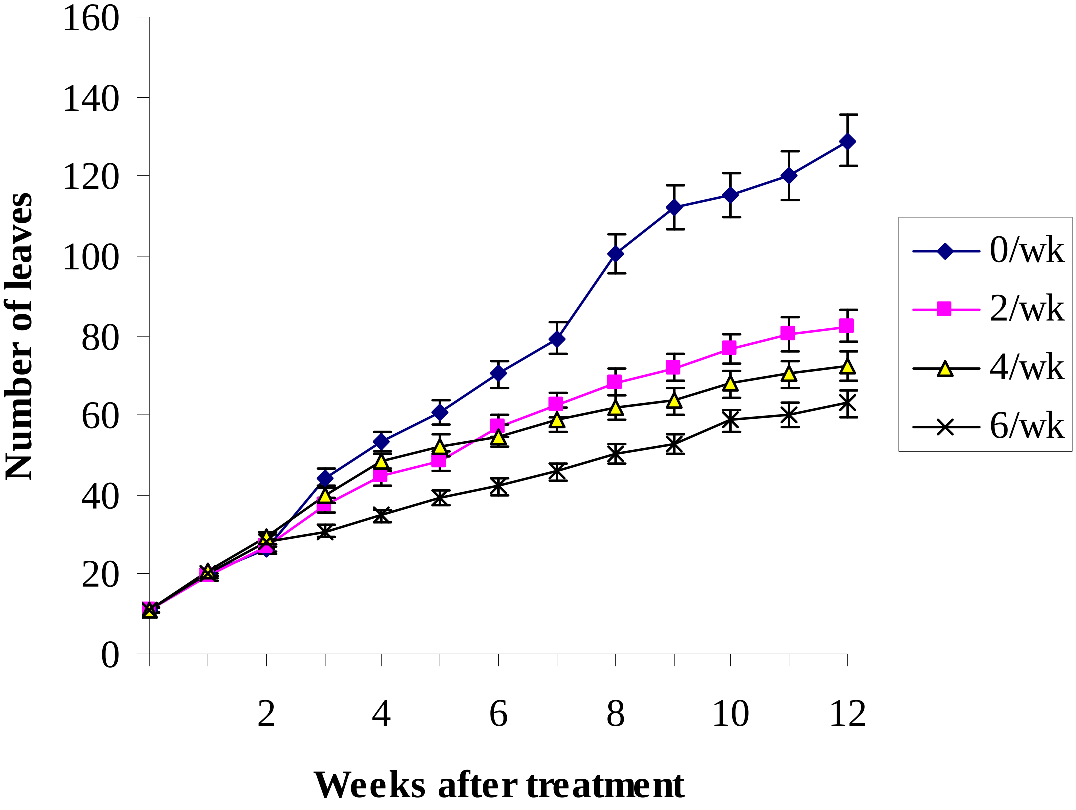

Figure 1. Effect of salt sprays on the leaf number of *C. maritima*, taken at 2-week interval for 12 weeks. Each value is a mean ± standard error of 6 determinations. 0/week = sprayed without seawater (demineralized water) six times per week, 2/week = sprayed with seawater twice per week, 4/week = sprayed with seawater four times per week, 6/week = sprayed with seawater six times per week.

damage in seedlings of *Pinus rigida* (Griffiths and Orians, 2004), *Solidago puberula*, *Solidago rugosa*, *Gaylussacia baccata* and *Quercus ilicifolia* (Griffiths and Orians, 2003) and in some coastal plant species (Griffiths et al., 2006). Kim et al. (2004) also observed browning symptoms followed by defoliation after the foliar spray of 3% NaCl in apple, pear, peach and grape trees. These could result in a decrease in net photosynthesis that might be expressed in reduced growth (Griffiths and Orians, 2004).

In coastal areas, salt spray often influences the distributions of species (Cheplick and Demetri, 1999; Griffiths and Orians, 2003; Ogura and Yura, 2007). Complete survival of plants under salt sprays conforms to the findings of Gagne and Houle (2002) on *Leymus mollis* and De Vos et al. (2010) on *Crambe maritima*. It is however in contrast with that of Gagne and Houle (2002) who reported a significantly reduced seedling survival in *Honckenya peploides*.

Growth reduction in *C. maritima* in this investigation confirms the report that salt spray usually leads to growth reduction (Tominaga and Ueki, 1991; Griffiths, 2006). Salt spray caused growth reduction in *Leymus mollis* (Gagne and Houle, 2002) and *M. pensylvanica* (Griffiths and Orians, 2003). Reduction in root elongation has been reported on many coastal plant species (Griffiths and Orians, 2004; Griffiths et al., 2006). The reduction in plant shoot length as obtained in this study affirms the report that salt spray inhibits the shoot elongation in coastal plant species (Griffiths et al., 2006). This could be one mechanism through which the characteristic dwarf stature of strand vegetation is maintained (Griffiths and Orians, 2003).

Reduction in number of branches agrees with the previous work of Cheplick and Demetri (1999) who recorded reduced number of tillers in *Triplasis purpurea* sprayed with salt relative to control plants. Reduction in leaf number in plants sprayed with seawater likewise supports the earlier report on *Scaevola sericea* seedlings by Goldstein et al. (1996) and De Vos et al. (2010) on *Crambe maritima*. Reduced leaf number in this study was due to leaf firing leading to leaf death and defoliation. Leaf area reduction is in conformity with that of Ogura and Yura (2007) on *Miscanthus sinensis* and *Imperata cylindrica* and in *Pinus rigida* (Griffiths and Orians, 2004). Leaf length was also reduced in *Crambe maritima* seedlings 13 weeks after spraying with salt water. Reduced leaf size was as a result of reduction in leaf area expansion and hence reduction of light interception. Reduction in leaf area also provided reduced area for water loss through transpiration and changes in

Table 2. Effect of salt sprays on leaf total chlorophyll and growth parameters of *C. maritima*.

No. of spray (s) per week	Survival (%)	Stem Girth (cm)	Leaf area (cm^2)	Number of branches	Total biomass (gplant^{-1})	Root: Shoot	RGR (gg^{-1}d^{-1})	Total chlorophyll (mgg^{-1})	Root number	Root length (cm)
0	100	0.40^{a}	5.04^{a}	22.72^{a}	20.67^{a}	1.34	0.08^{a}	3.24^{a}	67.43^{a}	44.71^{a}
2	100	0.31^{b}	2.41^{b}	19.55^{a}	13.45^{b}	1.95	0.07^{a}	1.43^{b}	49.87^{b}	32.04^{b}
4	100	0.31^{b}	2.36^{b}	16.89^{a}	11.50^{b}	2.15	0.07^{a}	1.71^{b}	47.65^{b}	34.11^{b}
6	100	0.28^{b}	2.54^{b}	16.87^{ab}	8.47^{bc}	1.87	0.07^{a}	1.68^{b}	48.55^{b}	34.67^{b}

RGR = Relative growth rate. Each value is a mean of 6 replicates taken at 12 weeks after initiation of treatments. Means with the same letter (in superscript) in the same column are not significantly different at $P \geq 0.05$.

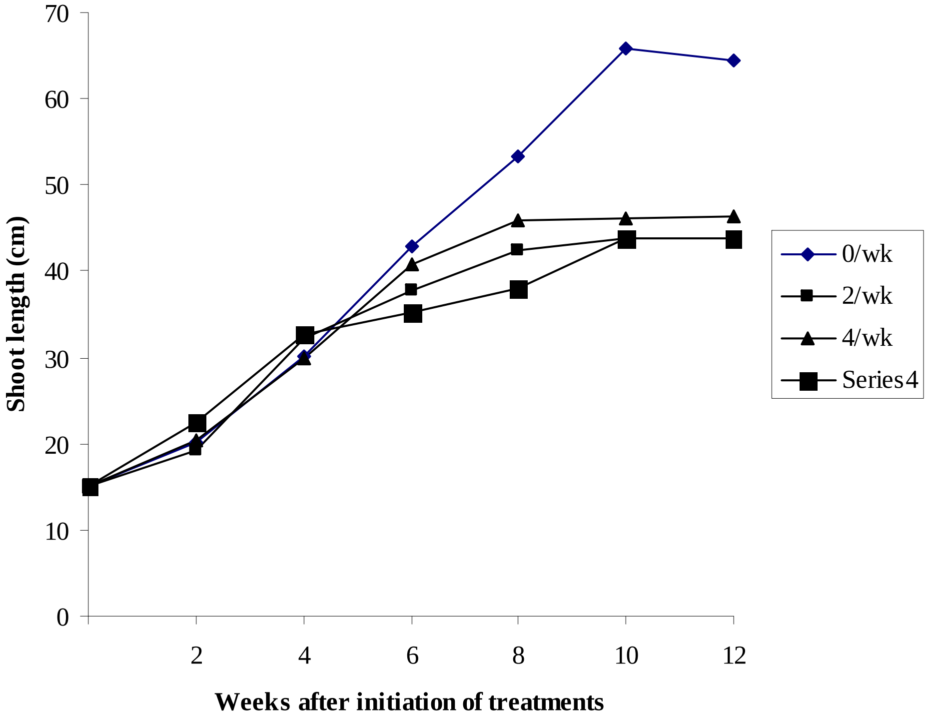

Figure 2. Effect of salt sprays on the shoot length (cm) of *C. maritima*, taken at 2-week interval for 12 weeks. Each value is a mean of 6 replicates. 0/week = sprayed without seawater (de-mineralized water) six times per week, 2/week = sprayed with seawater twice per week, 4/week = sprayed with seawater four times per week, 6/week = sprayed with seawater six times per week.

Table 3. Effect of salt sprays on the fresh and dry mass of plant parts in *C. maritima*.

Parameter	**Plant part**	**No of spray(s)per week**			
		0	**2**	**4**	**6**
Fresh mass (g)	Leaf	5.10^{a}	2.56^{b}	2.11^{b}	1.25^{bc}
	Stem	10.03^{a}	5.94^{b}	4.70b	5.16^{b}
	Root	14.87^{a}	11.14^{a}	9.76^{a}	7.38^{c}
Dry mass (g)	Leaf	3.77	2.04	1.67	1.01
	Stem	5.07	2.53	1.97	1.96
	Root	11.88	8.91	7.81	5.56

Each value is a mean of 6 replicates, taken at 12 weeks after initiation of treatments. Means with the same letter (in superscript) in the same row are not significantly different at P ≥ 0.05. 0/week = sprayed without seawater (de-mineralized water) six times per week, 2/week = sprayed with seawater twice per week, 4/week = sprayed with seawater four times per week, 6/week = sprayed with seawater six times per week.

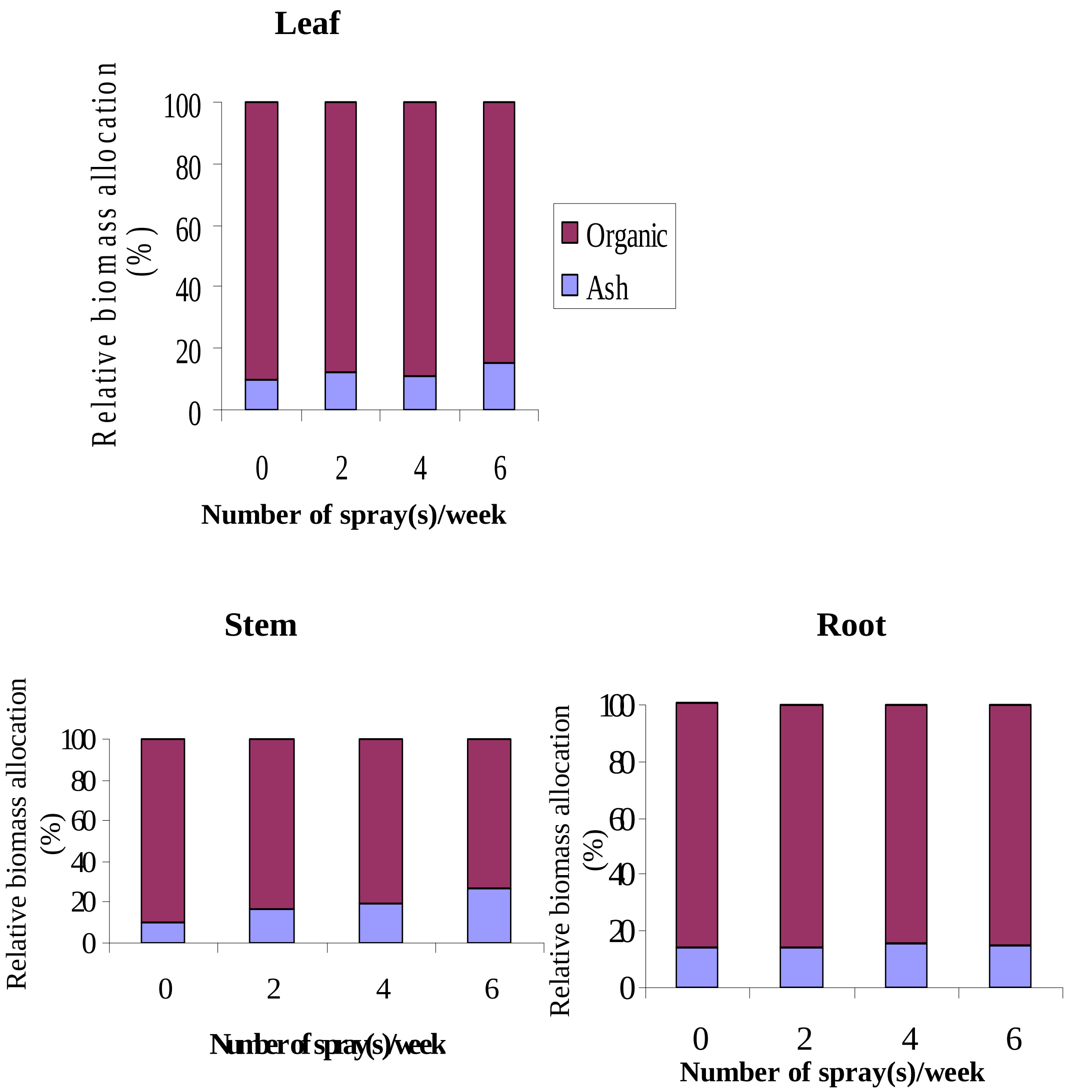

Figure 3. Effect of salt sprays on the relative biomass allocation (%) of *C. maritima.* Each bar represents mean of six determinations, taken at 12 weeks after initiation of treatments.

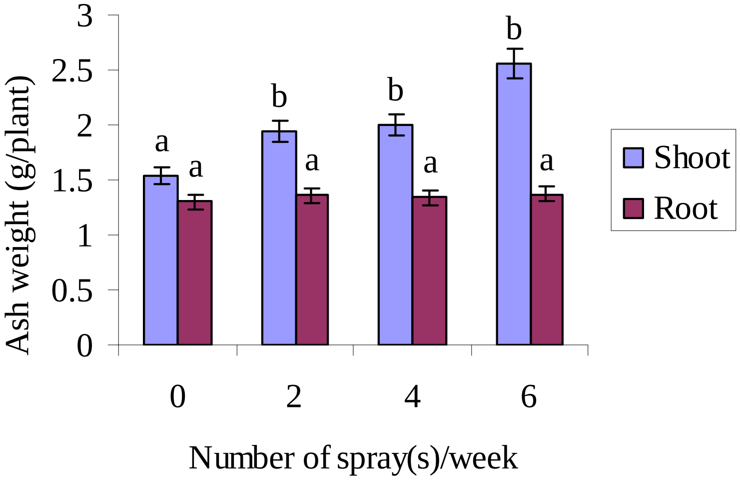

Figure 4. Effect of salt sprays on the ash mass (g plant^{-1}) of *C. maritima*. Each bar represents mean + standard error of 6 determinations. Bars with the same letter are not significantly different at P ≥ 0.05.

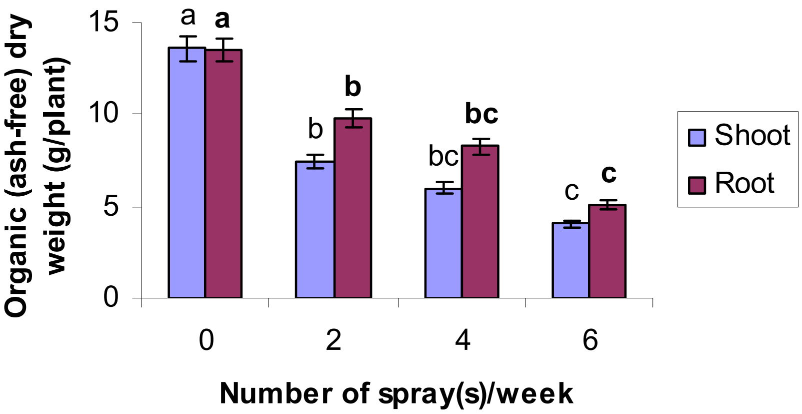

Figure 5. Effect of salt sprays on the organic mass (g plant^{-1}) of *C. maritima*. Each bar represents mean + standard error of 6 determinations, taken at 12 weeks after initiation of treatments. Bars with the same letter are not significantly different at P ≥ 0.05.

water use efficiency in plants, which have been identified to be adaptive mechanisms under water stress (Morant-Manceau et al., 2004). Reduced photosynthetic leaf area was due primarily to chloride and sodium ions from the salt spray (Cartica and Quinn, 1980). However, leaf area reduction is in contrast with that of Touchette et al. (2009) on *Spartina alterniflora*. It has been reported that salt penetrates leaves through lesions or via stomata and is consequently translocated to other plant parts (Boyce, 1954; Barbour et al., 1985) which could affect root development.

Reduction in chlorophyll content was due to leaf damage resulting in increased necrotic spots. Necrotic damage by salt spray has been reported by many authors (Sykes and Wilson, 1988; Griffiths and Orians, 2004). This usually results in a decreased net photosynthesis that might be expressed in reduced growth and long-term survivorship (Griffiths and Orians, 2004). Chlorophyll reduction can also be due to ion deficiency or ion toxicity.

Factors such as soil salinity, salt spray, sand burial, water deficit, flooding and nutrient deficiency are the natural factors that determine the distribution of strand vegetation (Lee and Ignaciuk, 1985). Strand plants vary considerably in the factors that affect their survival and growth, with each considered as a micro-environmental factor among the various environmental conditions including soil salinity, sand burial, drought, waterlogging and nutrient deficiency that affect strand vegetation (Barbour, 1978). This experiment has shown that *C. maritima* growth was negatively affected by salt spray. Therefore, salt spray is a micro-environmental factor affecting *C. maritima* growth, which determines its presence and distribution in the strandline.

Conclusions

Strand plants vary considerably in the specific factor(s) among the various abiotic factors that affect them in the strandline, hence the question: which of the factors determine the distribution of every plant species that grow naturally in the environment? This research has revealed that salt spray affects the growth of *C. maritima*, and it can be categorized as a salt spray-sensitive plant. Salt spray is therefore a micro-environmental factor (among the various abiotic factors) that contributes to the survival, growth and distribution of *C. maritima* in the strandline.

Conflict of Interests

The author(s) have not declared any conflict of interests.

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