Population Studies of Limonium stocksii (Plumbaginaceae) from a Salt Desert Near the Arabian Sea Coast

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Population Studies of *Limonium stocksii* (Plumbaginaceae) from a Salt Desert Near the Arabian Sea Coast

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Abstract: A population of *Limonium stocksii* was studied for one year at the upper end of Manora Creek along the Arabian Sea coast, near Karachi, Pakistan. The community was divided into three zones. Soil pH varied from 8.1 to 8.5. Soil conductivity was highest (191 dS m$^{-1}$) in September and lowest (55 dS m$^{-1}$) in June. Soil water content was relatively low and varied from 1.6% in February to 9.5% in June. *Limonium stocksii* maintained a persistent seed bank, which peaked at 5,887 seeds m$^{-2}$ in May. Very slow growth was observed; however, plants in the intermediate zone showed better growth than the other two zones. Stem and leaf tissue water content were affected by salinity fluctuations, but root tissue water remained unaffected. Sodium (Na$^{+}$) and chloride (Cl$^{-}$) accumulated in stems and leaves more than other ions. Leaves also accumulated a very high amount of magnesium (Mg$^{2+}$) compared to roots and stems.

Keywords: Arabian Sea, halophyte, ion accumulation, *Limonium*, population, salt desert, tissue

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INTRODUCTION

The Arabian Sea coast near Karachi, Pakistan, is extremely arid as a result of low rainfall and high temperature. The region is represented by approximately 108 halophytic species, mostly herbaceous perennials from 36 plant families (Khan and Gul 2002). The Plumbaginaceae is represented by Limonium stocksii (Boiss) O. Kuntze, a salt-secreting, low-branched perennial. Populations of L. stocksii occupy salt flats and rocky grounds near the seashore along the Karachi coast. Laboratory experiments on seed germination (Zia and Khan 2004) and growth of L. stocksii showed it to be a moderately salt-tolerant halophyte that could be classified as a miohalophyte (Zia 2004).

Coastal vegetation is broadly classified as salt marsh, coastal dunes, brackish water, and rocky salt planes (Day et al. 1989; Mitsch and Gosselink 2000; Migahid and Elhaak 2001). Demographic studies of many halophytic populations show that halophytes exhibit both interspecific and intraspecific variation in resistance to salinity stress based on their location (Hester mendelsohn, and McKee 2001). Generally, salinity, water, and temperature are critical environmental factors that determine the vegetation pattern in a region (Sen, Rajpurohit, and Wissing 1982; Boer 1996). However, populations are predominantly affected by specific overriding physical factors such as sand burial (Middleton 2003), inundation frequency (Hutchinson 1982; Leuschner, Landwher, and Mehlig 1998; Gul and Khan 1999; Bockelmann et al. 2002), and pulsed changes in substrate salinity (Whigham, Jordan, and Miklas 1989; La Peyre and Rowe 2003; Lillebø et al. 2003).

Halophytes utilize a number of different mechanisms at the cellular level to achieve osmotic adjustment, including 1) inorganic ion accumulation, 2) synthesis or accumulation of organic compounds, and 3) water loss (Ungar 1991).

Leaves of many halophytes play an important role in combating salt stress. Leaf evolutionary strategies to respond to salt stress include trichomes, sunken stomatas, salt glands, development of succulence, reduced leaf surface area, deposition of a thick cuticle, or a combination of the aforementioned traits (Khan and Gul 2002).

Soil seed banks are important components of vegetation dynamics affecting both ecosystem resistance and resilience (Pugnaire and Lazaro 2000), and their relationship to plant populations is crucial to understanding the development of plant desert communities (Kemp 1989). In contrast, Gul and Weber (2001) questioned the role of soil seed banks in determining population dynamics of saline deserts, because recruitment from seed banks is a rare phenomenon. In arid environments, both perennial and annual species accumulate seeds in the soil, but the distinction between persistent and transient seed banks is weak. This is because the extent of germination depends on rainfall (Baskin and Baskin 1998) and on the soil moisture threshold for germination that varies among species (Parke and Venables 1996). Pugnaire and Lazaro (2000) suggested that rainfall variability strongly affects species composition. For halophytes, the ability to form seed banks has survival value under extreme conditions that
mature plants would not be able to endure (Gul 1998). Seed bank studies from Karachi, Pakistan, show that 1) perennial shrubs and grasses dominating the vegetation maintain a persistent seed bank (Gulzar and Khan 1994; Aziz and Khan 1996) and 2) seed banks exhibit considerable spatial and temporal variation in size (Khan and Gul 1999). Khan (unpublished data) indicated that seeds of most perennial species studied lost their viability within a few months after dispersal as a result of high temperature and salinity stress when in the imbibed state. The significance of seed reserves in the soil in determining the establishment of a plant population in hypersaline environments is not clearly understood.

The *L. stocksii* population in this study extended from the edge of Manora Estuary to the farthest end of Manora Creek. The area near the edge received regular inundation from seawater, and inundation progressively decreased toward the landward side. However, it may get inundated occasionally during the year with high monsoons. The present study was designed to monitor the variation in demography and growth parameters along the inundation gradient.

**MATERIALS AND METHODS**

The study was carried out from July 2000 to June 2001 in a salt flat near Hawks Bay, Karachi, Pakistan (24° 52′–647′ N and 66° 53′–321′ E). The main source of moisture for the population is seawater seepage from the backwaters of Manora Creek. The mean ambient summer and winter temperatures are approximately 36°C and 25°C, respectively (Gul and Khan 1999). Rains are received during the monsoon season from June to September; however, no rainfall was observed during the study year. The study site was dominated by *Limonium stocksii* along with few individuals of *Aeluropus lagopoides* (Linn.) Trin. Ex Thw., *Arthrocnemum macrostachyum* (Moric.) C. Koch, *Suaeda fruticosa* Forssk. ex J. F. Gmelin, *Tamarix* spp., and *Urochondra setulosa* (Trin.) C. E. Hubbard.

**Plant Sampling**

Three transects of 100 m were laid across the population from the landward to seaward sides. These transects were further divided into three zones perpendicular to the transects. Nine plants from each zone were randomly harvested each month to measure the morphological growth parameters (root and stem length, number of branches, fresh and dry weight).

**Ion Analysis**

For ion analyses, 0.5 g of ground dry plant material was boiled in 25 mL of deionized water for 2 h at 100°C using a dry heat bath. This hot-water extract
was cooled and filtered using Whatman no. 2 filter paper. One mL of this hot-water extract was diluted with de-ionized water for ion analysis. Inorganic cations potassium ($\text{K}^+$), sodium ($\text{Na}^+$), calcium ($\text{Ca}^{2+}$), and magnesium ($\text{Mg}^{2+}$) from the plant organs were analyzed using a Perkin-Elmer® 238

**Table 1.** Monthly variation in moisture content (%) of the soil in different zones of the *Limonium stocksii* community during a 1-year study from July 2000 to June 2001

<table>
<thead>
<tr>
<th>Months</th>
<th>Landward</th>
<th>Seaward</th>
<th>Intermediate</th>
</tr>
</thead>
<tbody>
<tr>
<td>July</td>
<td>1.72 ± 0.26</td>
<td>3.37 ± 0.30</td>
<td>3.33 ± 0.53</td>
</tr>
<tr>
<td>August</td>
<td>5.44 ± 0.53</td>
<td>6.12 ± 0.46</td>
<td>5.56 ± 0.51</td>
</tr>
<tr>
<td>September</td>
<td>2.63 ± 0.46</td>
<td>4.03 ± 0.46</td>
<td>2.81 ± 0.24</td>
</tr>
<tr>
<td>October</td>
<td>3.98 ± 0.35</td>
<td>3.51 ± 0.54</td>
<td>6.56 ± 0.78</td>
</tr>
<tr>
<td>November</td>
<td>2.70 ± 0.15</td>
<td>2.93 ± 0.45</td>
<td>2.06 ± 0.25</td>
</tr>
<tr>
<td>December</td>
<td>7.76 ± 0.64</td>
<td>4.24 ± 0.65</td>
<td>4.67 ± 0.53</td>
</tr>
<tr>
<td>January</td>
<td>1.80 ± 0.25</td>
<td>3.79 ± 0.39</td>
<td>4.44 ± 0.39</td>
</tr>
<tr>
<td>February</td>
<td>1.67 ± 0.20</td>
<td>2.10 ± 0.26</td>
<td>2.00 ± 0.43</td>
</tr>
<tr>
<td>March</td>
<td>2.28 ± 0.28</td>
<td>3.13 ± 0.36</td>
<td>3.50 ± 0.41</td>
</tr>
<tr>
<td>April</td>
<td>2.69 ± 0.24</td>
<td>2.72 ± 0.31</td>
<td>3.69 ± 0.35</td>
</tr>
<tr>
<td>May</td>
<td>3.00 ± 0.35</td>
<td>2.91 ± 0.25</td>
<td>4.88 ± 0.51</td>
</tr>
<tr>
<td>June</td>
<td>6.78 ± 0.42</td>
<td>8.30 ± 0.99</td>
<td>9.49 ± 0.86</td>
</tr>
</tbody>
</table>
atomic absorption spectrophotometer. Chloride ion content analyzed using a Corning pH/ion meter 135 with an Accumet chloride-specific probe.

**Seed Bank Analysis**

Nine soil cores were randomly collected monthly from each zone using a soil corer (1.5 cm × 6 cm) for seed bank analysis. Seeds from these samples were manually extracted with the help of a binocular microscope and identified with the help of precollected seed samples. Nine surface soil samples were also collected monthly for analysis of conductivity and moisture content. These data were analyzed using two-way and three-way ANOVA with SPSS software (SPSS®, 2002). Bonferroni comparisons were used to determine significant differences between groups in a data set, demonstrating a statistically significant ANOVA.

**RESULTS**

Soil pH varied from 8.1 to 8.5 along the transects, with the lowest pH in the intermediate zone. Soil conductivity varied from 55 dS m⁻¹ to 191 dS m⁻¹ in different zones throughout the year (Figure 1). Soil conductivity was highest in the intermediate zone and lowest in the landward zone.
Low soil-moisture content was recorded as there was no rainfall during the study. Generally, soil moisture increased from the landward to the seaward zone (Table 1). Lowest soil moisture (1.67%) was recorded in February in the landward zone, and highest soil moisture (9.49%) was recorded in June in the intermediate zone (Table 1). Soil moisture was maintained at higher levels in the intermediate zone from July to November and in the seaward zone from December to June (Table 1).

Seed bank size of *L. stocksii* was generally small. Seed density decreased from the landward zone to the seaward zone and was highest (5,887 seeds m$^{-2}$) in May and lowest (68 seeds m$^{-2}$) in June (Figure 2). Mean annual average value for seed density was highest in the landward zone, whereas it

![Figure 3. Mean (± SE) root and stem length of *Limonium stocksii* in different zones along a transect. Bars with different letters denote significant differences (P < 0.05, Bonferroni).](image-url)
was almost equal in the intermediate and seaward zones (Figure 2). A few seeds of *Aeluropus lagopoides*, *Cyperus* spp., and *Suaeda fruticosa* were also encountered in the landward zone.

Growth parameters such as root and stem length (Figure 3), number of branches (Figure 4), and fresh and dry biomass (Figures 5 and 6) varied little in different zones. A two-way ANOVA indicated significant main effects of month ($F = 34.152, P < 0.0001$), zone ($F = 3.798, P < 0.05$), and their interaction ($F = 1.88, P < 0.05$) in affecting the root length. Root length was highest in September in the intermediate zone, whereas stem length showed no variation throughout the year (Figure 3). A two-way ANOVA of stem length indicated a significant individual effect of month ($F = 49.778, P < 0.0001$), whereas zone ($F = 2.279^{ns}$) and its interaction with month ($F = 1.109^{ns}$) were nonsignificant. The number of branches was highest in November and June (Figure 4). A two-way ANOVA indicated a significant main effect of month ($F = 7.026, P < 0.0001$), zone ($F = 2.55, P < 0.05$), and their interaction ($F = 1.652, P < 0.05$) in affecting the number of branches.

A three-way ANOVA indicated significant main effects of month, zone, plant part, and their interactions in affecting fresh weight, dry weight, and tissue water content of *L. stocksii* (Table 2). In general, leaf and stem biomass was much higher than root biomass. Biomass values varied in the order intermediate > landward > seaward zones. *Limonium stocksii* plants

![Figure 4. Number (+ SE) of branches of *Limonium stocksii* in different zones of the community from July 2000 to June 2001. Bars with different letters denote significant differences (P < 0.05, Bonferroni).](image)
collected in June had the highest fresh and dry weight, whereas plants with the lowest fresh and dry weights were collected in December (Figures 5 and 6). Fresh and dry weights were highest for leaf tissue in the landward zone and for stem tissue in the intermediate zone (Figures 5 and 6). Tissue water content in leaf tissue was higher in comparison to stem and root tissue (Figure 7), particularly from January to March, and the lowest values were recorded in November (Figure 7). Stem tissue exhibited highest water content in July and decreased in August. Root tissue did not vary much throughout the year (Figure 7).

Figure 5. Mean (± SE) fresh weight of roots, stems, and leaves of *Limonium stocksii* in different zones of the community from July 2000 to June 2001. Bars with different letters denote significant differences (P < 0.05, Bonferroni).
A two-way ANOVA indicated significant individual effects of zone, plant part, and their interactions in affecting the ion content of *L. stocksii* (Table 3). In general, all ions accumulated more in the leaves except for K\(^+\), which had similar values among plant parts. This was followed by stems with slightly lower values, except for Mg\(^{2+}\), which was substantially lower in comparison to the leaves. Roots had comparatively much lower ion values than other plant parts except for chloride (Cl\(^-\)) and K\(^+\) (Figure 8). Potassium varied little in different plant parts and had low values in comparison to Na\(^+\) and Cl\(^-\) (Figure 8).

**Figure 6.** Mean (± SE) dry weights of different *Limonium stocksii* organs from different zones along transects from July 2000 to June 2001. Bars with different letters denote significant differences (P < 0.05, Bonferroni).
DISCUSSION

The main source of water for *L. stocksii* population near Hawks Bay, Karachi, was seepage from the sea, as very little rainfall was recorded for the previous years (1998–2000). Soil pH was basic in nature, varying from 8.1 to 8.5. Soil conductivity varied from 55 dS m$^{-1}$ to 100 dS m$^{-1}$ throughout the year, which was manyfold higher than the optimal salinity level for growth under greenhouse conditions (Zia 2004). Differences in salinity between laboratory conditions are seldom comparable to those encountered in the field because of the variation in salt concentration at and around root surface and the differences in soil salinity with changes in soil moisture (Barrilleaux and Grace 2000). In addition, salinity measurements are limited only to the salt concentrations and do not indicate variations either in the composition or combinations of salts. Variation in soil salinity increased from the landward to the seaward zone throughout the year. Coastal substrate salinity varies considerably, from 0.1% to 3% (Barbour, DeJong, and Pavlik, 1985) and depends on the time of year and proximity to the sea (Weber and D’Antonio 1999). The fluctuating substrate salinity is reported to affect growth of many halophytes (Whigham, Jordan, and Miklas 1989; La Peyre and Rowe 2003; Lillebø et al. 2003).

Seed bank studies from Karachi, Pakistan, demonstrated that dominant perennial shrubs and grasses maintain a persistent seed bank (Gulzar and Khan 1994; Aziz and Khan 1996). Most salt desert and salt marsh halophytes maintain a persistent seed bank, which serves as a long-term seed-storage mechanism during unpredictable cold, dry, or hypersaline periods (Ungar 1995). *Limonium stocksii* appears to have a persistent seed bank; however, seed bank size was very small with a maximum of 5,887 seeds m$^{-2}$. Seeds, when tested for viability, germinated showing no sign of dormancy.

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Table 2. Three-way ANOVA of characteristics by month (M), zone (Z), and plant part (P) and their interactions affecting the fresh and dry weight and tissue water content in *Limonium stocksii*

<table>
<thead>
<tr>
<th>Dependent variable</th>
<th>Fresh weight</th>
<th>Dry weight</th>
<th>Tissue water</th>
</tr>
</thead>
<tbody>
<tr>
<td>Month (M)</td>
<td>11.8***</td>
<td>10.5***</td>
<td>6.3***</td>
</tr>
<tr>
<td>Zone (Z)</td>
<td>14.6***</td>
<td>10.6***</td>
<td>0.7ns</td>
</tr>
<tr>
<td>Plant part (P)</td>
<td>107.1***</td>
<td>62.1***</td>
<td>261.1***</td>
</tr>
<tr>
<td>M × Z</td>
<td>3.4***</td>
<td>2.8***</td>
<td>2.7***</td>
</tr>
<tr>
<td>M × P</td>
<td>2.0**</td>
<td>1.7*</td>
<td>3.9***</td>
</tr>
<tr>
<td>Z × P</td>
<td>5.6***</td>
<td>3.8***</td>
<td>1.6ns</td>
</tr>
<tr>
<td>M × Z × P</td>
<td>0.5ns</td>
<td>0.3ns</td>
<td>1.1ns</td>
</tr>
</tbody>
</table>

Notes: Numbers indicate F-values at * = P < 0.01, ** = P < 0.001, *** = P < 0.0001, ns = nonsignificant.
Seeds of *L. stocksii* were ready to germinate (Zia and Khan 2004) and remained nondormant even after long periods of storage at ambient temperature (Zia 2004). Lack of rainfall during the study period was the reason for absence of any seedling data; however, a large number of seedlings was observed in the same community after monsoon rains the following year. Mature seeds remained attached on the mother plant for 2–3 months before dispersal. After this time, seeds still enclosed in their bracts were collected by ants in large numbers. This predator activity may well be the reason for the patchy seed bank data and the pattern of seedling emergence after rainfall.

Figure 7. Mean (+ SE) tissue water content (g g\(^{-1}\) dry wt) of roots, stems, and leaves in *Limonium stocksii* from different zones of the community during July 2000 to June 2001. Bars with different letters denote significant differences (P < 0.05, Bonferroni).
Resources are often limited in saline environments. Plants allocate the available resources to various plant structures or functions according to several possible compromises maximizing their fitness (Harper 1977). Life history strategies partly depend on selection for optimal resource allocation to growth, reproduction, or maintenance of vegetative structures (Abrahamson 1979). Limonium stocksii allocated most of its biomass to aboveground parts. Similar results were found in a laboratory experiment on the growth of L. stocksii (Zia 2004). Clarke and Jacoby (1994) studied the biomass allocation in aboveground parts of Juncus kraussii Hochst., Sarcocornia quinqueflora (Ung.-Sternb.) A. J. Scott, and Sporobolus virginicus (L.) Kunth and concluded that soil moisture and soil salinity were the major controlling factors. Growth parameters of L. stocksii varied little throughout the year, probably due to both water deficit and a subsequent rise in soil salinity. Slow growth rate in saline habitats has survival value because there is little interspecific competition (Adu, Yeo, and Okusanya 1994). Stem length and fresh weight data correlate with soil moisture content. Tissue water content of L. stocksii was also not high in any plant part and remained unchanged throughout the year. Similar results for tissue water were also observed in a greenhouse study (Zia 2004).

Plants growing in saline environments encounter salt toxicity, water deficit, and nutrient deficiency or imbalance (Munns and Termaat 1986; Munns 1993; Adu, Yeo, and Okusanya 1994; Niu et al. 1995). Many halophytes control high Na⁺ and Cl⁻ concentrations either through ion exclusion at the root or secretion of ions from the leaves through salt glands (Flowers 2004). Other halophytes accumulate salts in their tissues for osmotic adjustment and compartmentalize ions in the vacuole (Gorham, Wyn Jones, and McDonell 1985). Ion accumulation data showed that L. stocksii, like most halophytes (Flowers 2004), accumulated high concentrations of Na⁺ in leaves and stems as an osmoticum. Accumulation of Na⁺ was twofold greater than Cl⁻. The electrical and chemical gradient analyses across salt glands in Limonim spp. showed that it is an electrogenic Cl⁻ pump with an affinity for K⁺ and Na⁺ (Long and Mason 1983). This could be the reason for low Cl⁻ content in L. stocksii, or there may be other anions such as sulfates and organic acids doing the balancing act. Donovan,

### Table 3. Two-way ANOVA for ions in Limonium stocksii due to zone (Z), plant part (P), and their interactions

<table>
<thead>
<tr>
<th>Dependent variables</th>
<th>Na⁺</th>
<th>K⁺</th>
<th>Ca⁺⁺</th>
<th>Mg⁺⁺</th>
<th>Cl⁻</th>
</tr>
</thead>
<tbody>
<tr>
<td>Zone (Z)</td>
<td>3.1*</td>
<td>49.5ns</td>
<td>14.0***</td>
<td>9.6***</td>
<td>11.3***</td>
</tr>
<tr>
<td>Plant part (P)</td>
<td>2.9*</td>
<td>65.9ns</td>
<td>191.5***</td>
<td>65.5***</td>
<td>5.5**</td>
</tr>
<tr>
<td>Z x P</td>
<td>6.5***</td>
<td>70.8ns</td>
<td>6.3***</td>
<td>6.9***</td>
<td>3.5***</td>
</tr>
</tbody>
</table>

**Notes:** Numbers indicate F-values at * = P < 0.05, ** = P < 0.001, *** = P < 0.0001, ns = nonsignificant.
Richards, and Schaber (1997) correlated higher leaf Na\(^+\) with the dominance of Na\(^+\) nutrition in halophytes. Relationship of ion accumulation and soil moisture content showed an increase in soil moisture content and a decrease in Na\(^+\) accumulation in leaves and stems.

In conclusion, the *Limonium stocksii* population studied was found to be highly salt tolerant. It maintained a viable and persistent seed bank, and after monsoon rains recruitment mainly takes place by seed germination. Growth of plants continued at a slow rate, and most of the biomass accumulated in upper plant parts. *Limonium stocksii* accumulated large amounts of Na\(^+\) followed by Cl\(^-\) and K\(^+\). Sodium appears to have a role as an osmoticum.

**Figure 8.** Mean (± SE) content of Na\(^+\), Mg\(^{2+}\), Cl\(^-\), Ca\(^{2+}\), and K\(^+\) in various organs of *Limonium stocksii* collected from (A) landward, (B) intermediate, and (C) seaward zones. Bars with different letters denote significant differences (P < 0.05, Bonferroni).
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