Gulf Research Reports

Volume 7 | Issue 1

January 1981

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DOI: 10.18785/grr.0701.05

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Eleuterius, L. N. and J. D. Caldwell. 1981. Effect of Mineral Deficiency on the Growth of the Salt Marsh Rush Juncus roemerianus. Gulf Research Reports 7 (1): 35-39.

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EFFECT OF MINERAL DEFICIENCY ON THE GROWTH OF THE SALT MARSH RUSH JUNCUS ROEMERIANUS

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ABSTRACT Plants of the salt marsh rush Juncus roemerianus were grown in deficient and complete nutrient solutions, and the growth responses were compared qualitatively and quantitatively. Plants grown in solutions deficient in K, S, P, and Mg were similar in appearance, which exemplified severe growth retardation. Plants grown in solutions deficient in Ca, N, and Fe also were very similar in total growth response, but had much longer leaves, more shoots, and greater biomass than those grown in the K, S, P, and Mg deficient solutions. Plants grown in complete nutrient solutions, with various sources of Fe, were very robust. Growth was better with FeCl₃ than with FeEDTA. The most robust growth occurred in plants grown in the nutrient medium deficient in micronutrients (B, Mn, Zn, Cu, Mo), indicating that J. roemerianus is very sensitive to specific or certain combined micronutrients or concentrations generally recommended for terrestrial, especially agricultural, plants. The qualitative and quantitative symptoms, such as color, length, and abundance of leaves and roots as a response to deficiency of specific elements, were similar to those deficiency symptoms previously described for numerous terrestrial plants. However, a deficiency of certain nutrients such as S and Mg has a much greater effect on J. roemerianus than is generally shown for terrestrial plants.

INTRODUCTION

Interest in mineral deficiency of agricultural plants has a long history and correspondingly voluminous literature (Chapman 1973a). Information is lacking on the effects of mineral deficiency on angiosperms inhabiting tidal marshes. This inadequacy probably exists because of the common assumption that most estuarine sediments contain an abundance of nutrients for plant growth. However, Adams (1963) found that plants of Spartina alterniflora become chlorotic in iron-deficient soils but marsh plant species found at higher elevations, such as Distichlis spicata and Juncus roemerianus, are unaffected. He also pointed out that S. alterniflora reponds to experimental foliar applications of ferrous sulphate or the addition of iron to a saline nutrient solution and is often restricted to low elevation marshes where the mean soluble iron content is in excess of 4 ppm. Adams (1963) also suggested that iron deficiency may limit the growth of S. alterniflora at high marsh elevations, because the greatest tendency toward chlorosis is in the higher, better-drained, rather than the lower, wetter sites.

Tyler (1967) obtained a significant growth response from *Juncus balticus* in salt marsh plots treated with ammonium on the Baltic coast, indicating that the rush species grew naturally in depauperate soils in which the nitrogen concentration was not optimum. Furthermore, the substrata of salt marshes are highly variable in structure, ranging from organic peat to sand, which consequently reflects considerable variation in nutrient composition (Grim 1968; Richards 1969; Buckman and Brady 1969; Chabreck 1972; Cotnoir 1974; Boyd 1970; Brupbacher et al. 1973; Eleuterius 1972, 1974). Additionally, certain plants

have relatively high or specific mineral requirements suggesting that soil nutrient deficiencies (low concentrations) may limit their growth (Brownell 1965, Rorison 1968, Wallace et al. 1966, Broyer et al. 1954). Zonation of flowering plants in coastal marshes has been shown to be influenced by mineral nutrition (Pigott 1969). Eleuterius (1974) observed red streaks on the leaves of numerous small plants of *Juncus roemerianus* which may have indicated a deficiency of phosphorous in some sandy, marsh soils. Sandy soils of salt flats, in contrast, did not appear to be deficient because dwarf plants with very dark-green leaves also were observed there.

The objectives of this study regarding the mineral nutrition of *Juncus roemerianus* were (1) to determine the individual qualitative and quantitative effects that seven deficient minerals and an array of micronutrients would have on growth of the rush, (2) to compare these responses to those obtained in complete nutrient solutions, (3) to assess and rank the resulting condition of the test plants, and (4) to evaluate the nutritional requirements of the tidal marsh plant.

MATERIALS AND METHODS

Standard nutrient media (Machlis and Torrey 1956) were used to determine the effects of mineral deficiency on *J. roemerianus*. Seven nutrient solutions were prepared, each deficient (—) in one of the following elements: nitrogen (N), phosphorus (P), potassium (K), sulfur (S), calcium (Ca), iron (Fe), and magnesium (Mg). One solution deficient in an array of micronutrients (B, Mn, Zn, Cu. Mo) also was prepared as a test medium. Two complete nutrient solutions, differing in the source of iron, also were prepared as controls. One complete nutrient solution contained FeCL₃, and another the chelated form FeEDTA. Distilled water also was used as a control. Plants grown from seed collected

from a single parental plant to reduce genetic variability were used in triplicate for each culture solution. The experiment was carried out during the spring in a greenhouse. Temperature varied from about 18°C to 30°C during the experiment.

Seedlings of similar size were selected, based on the number of leaves and leaf length, from a large number of seedlings grown as individual units. Roots were thoroughly washed and freed of all peat and other organic material, and then placed in clean (washed) river sand for 5 weeks. A dilute (1%) solution of 10-30-20 soluble fertilizer (Robert Peters Co. Inc.) was used in a single application as a nutrient source. Prior experience showed that this procedure resulted in a proliferation of new roots, which replaced those lost and injured in the initial washing.

New roots formed within a few weeks and the remaining old ones subsequently were easy to clean. Seedlings were carefully rinsed six times in distilled water. The number and length of leaves were determined. For statistical comparison, a "total" leaf length was obtained by adding the length of all leaves on each plant. A single plant then was placed in each battery jar filled with nutrient media and supported with specially prepared, waxed, plywood covers (Machlis and Torrey 1956). Observations were made weekly for survival and deficiency symptoms, and the general condition of the plants was recorded. Distilled water was added to the nutrient solutions as needed. At the end of 68 days, number of leaves, total leaf length, maximum leaf length (plant height), dry mass of leaves, maximum root length, and dry mass of roots were determined. Multidimensional plotting, a method of cluster analysis (Andrews 1972, Nance et al. 1975), was used for statistical analysis of the quantitative data representing the seedling at the beginning and at the end of the experiment. Statistical significance between treatments was tested by a 1-way analysis of variance (ANOVA) test and a least significant difference (LSD) test.

RESULTS AND DISCUSSION

Seedlings were shown to have no significant difference at the beginning of the experiment by a 1-way ANOVA statistical test. Multidimensional plots also showed similarity between the seedlings for number of leaves, maximum leaf length (plant height), and total leaf length (Figure 1). Qualitative characteristics of the plants in each treatment at the end of the experiment are shown in Table 1. The ANOVA test, which compared all test groups for each variable simultaneously, showed a statistically significant difference between the 11 groups, except for the number of leaves produced per plant (Table 2). This means, because of the nature of the ANOVA test, that at least one test group was statistically different from at least one of the other groups. However, the ANOVA test does not indicate which groups were significantly different. When the resulting data were subjected to a LSD test all measured variables were shown to have a significant difference between certain test groups.

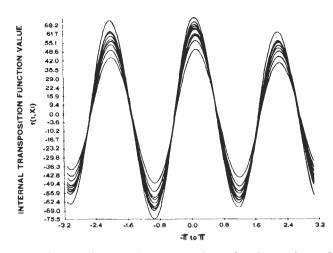


Figure 1. Multidimensional computer plots using the number and length of leaves for seedlings of *Juncus roemerianus* immediately prior to placing in complete and deficient nutrient solutions. Similarity and closeness of plotted lines indicate that plants are similar in size.

TABLE 1.

Mineral deficiency symptoms. Plant groups are ranked by treatments according to qualitative characteristics.*

Micronutrients Leaves dark green. Minimal dead leaf tissue. Roots
white or light tan, about 25 cm long, with pliable root hairs. Root
hairs slightly shorter (2-8 cm) than those in complete FeEDTA.

Complete (FeCL₃) Leaves dark green. Very little (about 1%) dead leaf tissue. Roots white to light tan, 30 to 40 cm long, with obviously more mass than roots of plants in complete FeEDTA. Root hairs long (4-10 cm), pliable.

Complete (FeEDTA) Leaves dark green. Very little (about 5%) dead leaf tissue. One plant dead. White to light tan roots, about 25 cm long, with long (4-10 cm), pliable root hairs.

- Ca Leaves dark green, some dead young leaves. Roots short (10-15 cm), reddish-brown. Short stumpy root hairs about 0.5 cm long.
- N Leaves pale or light green, about 15-20% dead leaf tissue.
 Roots white to light tan, very long (about 60 cm). Root hairs about 2 cm in length.
- Fe Leaves brownish-yellow with four pale or light-green leaves, about 40% dead tissue and 50% dying, 10% living. Roots, white to tan, coarse, about 15 cm long. Short root hairs about 0.5 cm long.
- K Leaves dark green, 25% dead leaf tissue (primarily young leaves). Roots long (30-35 cm), reddish-brown. Root hairs long (2-8 cm).
- S Leaves dark to light green. About 15% leaf tissue dead. One plant almost dead (very little growth). Roots reddish-brown to tan, about 25 cm long. Root hairs coarse, less than 1 cm long.
- P Leaves dark green, some leaves deep purplish-red or dark green with streaks of red. About 20% leaf tissue dead. One plant dead. Roots reddish-brown to tan. Few roots formed but extremely long, about 60 cm. Root hairs sparse, long (2-10 cm).
- Mg Leaves yellow-green. Leaf tissue, 10% dead, 70-75% dying. Roots short (about 15 cm), light tan. Root hairs short, about 0.5 cm.

Distilled H₂O Leaves light-green to yellow, brown or mottled with yellow or light to dark brown. About 20% leaf tissue dead, the remainder dying. Roots reddish-brown, short (10 cm), coarse. Root hairs short (0.5 cm) except on extreme tips of roots.

^{*}Three plants were grown in nutrient media for 63 days.

TABLE 2.

Mineral nutrition characteristics of Juncus roemerianus. Plant groups are ranked by treatments according to quantitative characteristics. Values represent mean and standard error of the mean of three plants grown in nutrient media for 68 days.

Nutrient Solution	Number of Leaves	Total Leaf Length (cm)	Leaf Length Maximum (cm)	Dry Mass Leaves (g)	Root Length Maximum (cm)	Dry Mass Roots (g)
- Micronutrients	22 ± 3.2	732.4 ± 78.1	78.1 ± 2.8	2.790 ± 0.08	28.3 ± 3.7	0.421 ± 0.02
Complete FeCL ₃	15 ± 2.5	415.2 ± 31.2	65.9 ± 5.9	1.593 ± 0.06	39.5 ± 5.0	0.313 ± 0.01
Complete FeEDTA*	12 ± 5.9	348.0 ± 198.7	49.0 ± 13.2	1.383 ± 0.72	27.7 ± 10.2	0.286 ± 0.10
- Ca	12 ± 3.1	255.2 ± 44.0	48.0 ± 5.7	1.103 ± 0.04	15.9 ± 0.3	0.164 ± 0.03
– N	13 ± 1.7	257.9 ± 10.7	44.9 ± 4.2	0.763 ± 0.01	70.2 ± 4.1	0.457 ± 0.08
– Fe	14 ± 1.2	212.6 ± 41.9	27.5 ± 4.7	0.413 ± 0.09	14.9 ± 4.3	0.081 ± 0.01
– K	8 ± 0.3	241.4 ± 12.5	56.1 ± 5.8	0.869 ± 0.11	37.1 ± 2.8	0.108 ± 0.01
− S †	9 ± 2.4	200.5 ± 62.6	36.8 ± 4.2	0.734 ± 0.29	25.0 ± 5.4	0.165 ± 0.11
− P *	9 ± 2.6	194.8 ± 69.1	38.0 ± 5.2	0.614 ± 0.27	56.1 ± 22.0	0.331 ± 0.17
– Mg	9 ± 1.2	186.7 ± 25.2	36.9 ± 1.9	0.435 ± 0.09	18.0 ± 2.0	0.075 ± 0.01
Distilled H ₂ O	11 ± 2.4	90.8 ± 16.0	19.9 ± 0.5	0.240 ± 0.04	11.3 ± 2.4	0.082 ± 0.02
F(10,22)	1.97	5.34††	8.13††	7.84††	5.18††	3.44††
LSD _{0.05}	7.7	205.0	16.2	0.716	22.3	0.211

^{*}Contained 1 dead plant.

†Contained 1 plant which was almost dead.

††Significant at the 0.05 level.

We believe the LSD test indicates a better analysis than the ANOVA test for our purposes because it compares each group of values against every other group individually for each variable (Table 2). Furthermore, all variables, which included all groups, were subjected to cluster analysis or multidimensional plotting (MDPLT). This analysis compares and evaluates collectively and simultaneously the entire array of data. MDPLT evaluates all variables measured for each group, resulting in an averaged or total value for each test group. The group values are then ranked. Ranking all quantitative responses by MDPLT resulted in six distinctly different groups as shown in Figure 2. The closer the curves, the more closely related or similar were the test plant groups based on total response (all variables).

The most unexpected response was the robust growth of plants in nutrient medium without micronutrients (B, Mn, Zn, Cu, Mo). Table 2 shows that the longest leaves and greatest dry mass were produced on plants without micronutrients. Plants grown in both complete nutrient media were also robust, but the best growth was achieved with FeCl₃ rather than FeEDTA. Iron appeared to be necessary for strong root development in *J. roemerianus*. Plants in Fe, K, and Mg deficient media had very poorly developed root systems (Table 2).

Although chelation (FeEDTA) may ensure availability of Fe to terrestrial plants (Steiner and Van Winden 1970), the requirement of *Juncus roemerianus* is apparently more readily satisfied by the FeCl₃ source. Apparently most of the Fe of FeCl₃ became chemically bound in soils and in

nutrient media, where it precipitated out of solution. Chemically bound Fe was not available for plant growth (Brown 1961, Hewitt 1963). Conversely, *J. roemerianus* may have a low Fe requirement and a high concentration of Fe in the nutrient medium or soil may have a toxic effect.

Plants grown in solutions deficient in Ca, N, and Fe had similar responses. These plants had shorter leaves, about one half of the number of shoots (reflected as a threefold

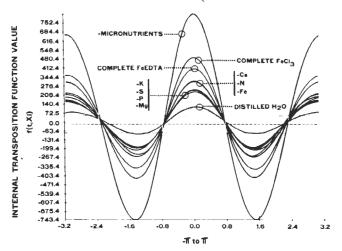


Figure 2. Multidimensional plots of plants of Juncus roemerianus grown in complete and deficient solution cultures for 68 days. The dotted, horizontal lines indicate the relative size of young plants at the beginning of the experiment. Curves shown in Figure 1 would center on the middle and fall within the upper and lower dotted lines. These dotted lines are provided to show the relative amounts of plant growth. Each curve represents the combined and relative condition of three plants in a particular series (triplicate) of deficient or complete nutrient media.

reduction in total leaf length), and one half of the biomass compared with plants in media without micronutrients. Plants grown in media deficient in K, S, P, and Mg were similar in appearance and all had a retarded growth habit.

Plants grown in distilled water were severely retarded compared to other plants. J. roemerianus appears to have a great capacity for survival under extremely depauperate conditions because the plants placed in distilled water survived and increased slightly in size, obviously utilizing stored nutrients. Gallagher (1975) stated that J. roemerianus was capable of storing reserves of nitrogen in leaves (luxury supply or reserve) without an observable increase in plant size. It was possible that the distilled water contained trace amounts of nutrients but certainly not at concentrations to support growth or survival.

The results of the present study indicate that the presence of Mg, P, S, and K, which are found in great quantities in highly organic tidal marsh soils (Pomeroy et al. 1969, Tyler 1971, Brupbacher et al. 1973), are extremely important for growth of J. roemerianus. The presence of Fe, N, and Ca are also very important for growth, but are probably required in much lower quantities than other nutrients. The growth of J. rocmerianus was suppressed in solutions with micronutrients. Apparently, one or more of the micronutrients was inhibitory to J. roemerianus. Variable soil fertility accounts, at least in part, for length of leaves (stand height), rhizome and root development, inflorescence size, and other phenotypic characteristics which are known to vary between populations of J. roemerianus found in Gulf and Atlantic coastal marshes (Eleuterius 1975). The qualitative and quantitative diagnostic symptoms of the rush J. roemerianus are very similar for each treatment to those mineral deficiency symptoms generally described for many agricultural and terrestrial plants (Epstein 1972, Devlin 1966, Chapman 1973b), with the exception of an apparent sensitivity to certain heavy metals. These similarities in physiological responses to an array of deficient elements links the intertidal and estuarine J. roemerianus to the adjacent upland or terrestrial flora. However, the effects are different in J. roemerianus in comparison to those generally seen in terrestrial plants especially in regard to the degree of severity. For example, the absence of Ca apparently

had less of an effect on the growth of *J. roemerianus* than that described for terrestrial plants (Chapman 1973b, McMurtrey 1932). The growth of *J. roemerianus* was severely retarded by a deficiency of S and Mg, while such a severe effect on growth has not been generally described for terrestrial plants (Carolus 1935, Embleton 1973). The S and Mg requirements of *J. roemerianus* were much greater than those of most terrestrial plants.

The greater S and Mg requirements of J. roemerianus compared to those for terrestrial plants are obviously related to the higher concentrations of those elements in salt water and soils of tidal marshes. Furthermore, concentrations of P and K are much higher in the soils of tidal marshes than in those of most terrestrial areas. In contrast, there is little available Ca. Nor Fe in estuarine waters or tidal marsh soils. The micronutrients B, Mn, Zn, Cu, and Mo occur in very low concentrations in the relatively pristine aquatic habitats of estuaries and tidal marshes in comparison to the concentration of the respective nutrient medium used in this study. The abundance of water probably accounts for the dilution of naturally occurring micronutrient concentrations in tidal marshes. The present study indicates that J. roemerianus is well adapted to the peculiar concentration and array of mineral nutrients in the soils and saline waters of pristine tidal marshes.

The heavy metals B, Mn, Zn, Cu, and Mo are compounds of domestic and industrial effluent which enter into estuaries from factories, municipalities, and through runoff from highways, marinas, farms, lawns, etc. Therefore, the sensitivity of the salt marsh rush J. roemerianus to heavy metal concentrations recommended for the culture of agricultural plants has profound and far-reaching importance to the future of estuaries, and indicates that J. roemerianus is physiologically distinct. The latter is not surprising because of the long evolutionary history of halophytes, and it points out the need for further physiological studies on J. roemerianus and other tidal marsh plants.

ACKNOWLEDGMENTS

We thank other members of the Botany Section of the Gulf Coast Research Laboratory, for assistance with various aspects of this study. Mrs. Helen Gill and Mrs. Lucia O'Toole typed the manuscript.

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