Annals of Arid Zone 49(2): 101-106, 2010

Variation in Yield, Free Proline Content and Cation Accumulation of Lemongrass (*Cymbopogon flexousus*) Varieties Grown in Sodic Soils

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Abstract: The variations in yield, free proline content and cation accumulation in five varieties of lemongrass (Pragati, Praman, CKP-25, Cauvery and Nima) in sodic soils of ESP (exchangeable sodium percentage) 18.0 and 36.0 were studied in a pot experiment. Irrespective of the soil sodicity level, all the varieties of lemongrass differed significantly in their herb and oil yield, free proline content and cation accumulation. The herb and oil yield of lemongrass significantly decreased with an increase in soil ESP from 18.0 to 36.0, however, the extent of decrease in yield was lower for the varieties Praman and Nima and higher for the varieties Pragati and CKP-25. The total citral content in the essential oil was highest for the variety Nima and lowest in CKP-25. The free proline content was significantly higher in shoot tissues of lemongrass grown in the sodic soil of ESP 18.0 than in the sodic soils of ESP 36.0. The concentration of Na in shoot and root tissues of lemongrass increased and that of K and Ca decreased with an increase in the soil ESP. The concentrations of Na, K, and Ca were relatively higher in the shoot than in the root tissues of lemongrass. The relatively lower accumulation of Na and the capacity to maintain higher K/Na and Ca/Na ratios in shoot tissues of varieties Praman and Nima at high soil ESP showed their higher tolerance of soil sodicity as compared to varieties Pragati, CKP-25 and Cauvery.

Key words: Lemongrass, herb and oil yield, soil exchangeable sodium percentage, cation accumulation, free proline content.

Soil salinity and sodicity are major factors limiting the crop productivity in the arid and semi-arid regions. Excess of exchangeable sodium in sodic soils inhibits the growth and yield of crops through its influence on physical and hydrological properties of soils and/or due to nutritional disorders and ion toxicity (Naidu and Rengasamy, 1993). In sodic soils high Na concentration reduces the amount of K, Ca and Mg in tissues of many plant species, even to a degree that symptoms of K and Ca deficiency appear (Greenway and Munns, 1980).

The reclamation of sodic soils for crop production is considered economically unviable. An alternate approach is the utilization of sodic soils for growing tolerant crops. Lemongrass (*Cymbopogon flexuosus* steud Wats) is a high value aromatic crop. The essential oil obtained from the hydro-distillation of fresh herbage is widely used in perfumery, cosmetic and pharmaceutical industries. Lemongrass can be grown on a wide variety of soils ranging from rich loam to poor laterite. Singh and Anwar (1985) reported that the lemongrass could be grown in saline soil of the ECe 10.0 dS m^{-1} without significant reduction in the yield. Adequate information on the effect of soil sodicity on the yield, free proline content and cation accumulation in different varieties of lemongrass, however, is lacking. In this study we report variations in yield, free proline content and cation accumulation of five varieties of lemongrass grown under two levels of soil sodicity (ESP 18.0 and 36.0).

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Materials and Methods

A surface (0-15 cm) soil (Typic Natrustalfs) was collected from the Banthra Research Farm of the National Botanical Research Institute, Lucknow. The soil was silty clay loam, with pH (1:2.5 soil water suspension) 10.0, electrolyte conductivity (EC) 1.0 dS m^{-1}, ESP (exchangeable sodium percentage) 60.0, CEC 14 c mol P^+/kg, organic carbon 0.15% and available N (alkaline $KMnO_4$ extractable), P (0.5 M $NaHCO_3$ extractable) and K (1 N neutral ammonium acetate extractable) 60.0, 2.0 and 50.0 mg kg^{-1} soil, respectively. Two levels of soil sodicity viz., soil ESP 18.0 and 36.0, corresponding pH (1:2.5 soil water suspension) 8.9 and 9.5 and EC 0.26 and 0.43 dS m^{-1} were simulated by applying laboratory grade gypsum ($CaSO_4.2H_2O$) to the above soil @ 4.7 and 3.2

Table 1. Herbage yield (g plant^{-1}) of different varieties of lemongrass grown in sodic soils

Varieties	First harvest			Second harvest			Third harvest			Fourth harvest			Total		
	ESP 18.0	ESP 36.0	Mean	ESP 18.0	ESP 36.0	Mean	ESP 18.0	ESP 36.0	Mean	ESP 18.0	ESP 36.0	Mean	ESP 18.0	ESP 36.0	Mean
Pragati	42.5	20.0	31.2^{b}	58.9	40.1	49.5^{c}	67.8	31.3	49.5^{a}	57.2	34.8	46.0^{b}	226.4	126.2	176.3b
Praman	61.9	40.7	51.3^{a}	80.5	48.5	64.5a	62.1	37.7	49.9^{a}	64.2	41.9	53.0^{a}	268.7	168.8	218.7a
CKP-25	42.0	26.6	34.3^{b}	48.1	23.9	36.0^{d}	65.2	36.1	50.6^{a}	66.1	35.3	50.7^{a}	221.4	121.9	171.6^{b}
Cauvery	36.0	20.9	28.4^{c}	54.2	32.8	43.5^{c}	58.2	35.8	47.0^{a}	45.3	25.8	35.5^{c}	194.5	115.3	154.9^{d}
Nima	35.7	21.4	28.5^{c}	70.9	41.9	56.4^{b}	52.6	40.7	46.6^{a}	47.7	23.5	35.6^{c}	206.9	127.5	167.2^{c}
Mean	43.6^{a}	25.9^{b}	34.7	62.5^{a}	37.4^{b}	49.9	61.8^{a}	36.3^{b}	48.7	56.1^{a}	32.3^{b}	44.2	223.6^{a}	131.9b	177.7
LSD	S : 2.39			S : 3.93			S : 2.34			S : 3.36			S : 5.99		
(P=0.05)	V : 3.79			V : 6.29			V : NS			V : 5.31			V : 9.47		
	S x V : NS			S x V : NS			S x V : 5.25			S x V : NS			S x V : 13.40		

Notes: Within treatment means followed by the same letter in the same column or rows are not significantly different at P=0.05. NS: Not significant.

g kg^{-1} soil, separately and leaching with ponding water for 15 days. Nine kg of air-dried soil (<2 mm) of two different ESP levels was used to fill the polythene lined earthen pots separately. The sodicity was maintained during the course of experiment by not draining the pots and irrigating plants with de-ionized water. A basal dose of 60 mg N, 14 mg P and 25 mg K kg^{-1} soil was applied by adding urea, KH_2PO_4 and KCl to each pot. The pots were maintained in a wire-net house with natural light and temperature and protected from precipitation by a transparent glass roof.

One rooted slip each of lemongrass varieties viz. Pragati, Praman, CKP-25, Cauvery and Nima was transplanted separately in each pot on 10th August, 2004. The soil water content in pots was maintained near field capacity throughout the experiment. The plants were harvested four times (11th April 2005, 5th August 2005, 20th November 2005 and 10th June 2006) during the 22 months of growth trial. At each harvest, the fresh herbage was weighed and sampled for the extraction of essential oil and chemical analyses. At the end of experiment, the plants were uprooted and roots were separated and sampled for the chemical

Table 2. Essential oil yield (ml plant^{-1}) of different varieties of lemongrass grown in sodic soils

Varieties	First harvest			Second harvest			Third harvest			Fourth harvest			Total		
	18.0	36.0	Mean	18.0	36.0	Mean	18.0	36.0	Mean	18.0	36.0	Mean	18.0	36.0	Mean
Pragati	0.30	0.14	0.22^{c}	0.32 (81.0)*	0.22 (81.6)	0.27^{b}	0.27	0.13	0.20^{c}	0.29 (81.0)	0.17 (83.7)	0.23^{b}	1.18	0.66	0.92^{c}
Praman	0.34	0.22	0.28^{b}	0.40 (75.6)	0.29 (78.6)	0.34^{a}	0.22	0.15	0.18^{c}	0.26 (74.6)	0.17 (77.5)	0.21^{b}	1.22	0.83	1.02^{a}
CKP-25	0.38	0.24	0.31^{a}	0.38 (60.8)	0.24 (60.3)	0.31^{b}	0.35	0.22	0.28^{a}	0.43 (74.3)	0.23 (77.9)	0.33^{a}	1.54	0.93	1.23^{a}
Cauvery	0.29	0.17	0.23^{c}	0.33 (78.3)	0.23 (79.4)	0.28^{b}	0.23	0.16	0.19^{c}	0.25 (81.2)	0.14 (80.8)	0.19^{b}	1.10	0.70	0.90^{c}
Nima	0.29	0.17	0.23^{c}	0.46 (86.4)	0.36 (85.3)	0.41^{a}	0.26	0.22	0.24^{b}	0.26 (88.4)	0.13 (89.6)	0.19^{b}	1.27	0.88	1.07^{b}
Mean	0.32^{a}	0.19^{b}	0.25	0.38^{a}	0.27^{b}	0.32	0.27^{a}	0.18^{b}	0.22	0.30^{a}	0.17^{b}	0.23	1.26^{a}	0.80^{b}	1.03
LSD	S : 0.016			S : 0.52			S : 0.011			S : 0.018			S : 0.041		
(P=0.05)	V : 0.26			V : 0.085			V : 0.017			V : 0.028			V : 0.065		
	S x V: NS**			S x V: NS			S x V : 0.025			S x V : 0.40			S x V : 0.092		

Notes: Within treatment means followed by the same letter in the same column or rows are not significantly different at P=0.05; *Figures in parentheses indicate total citral content in the essential oil; **NS: Not significant.

*Table 3. Cation accumulation in shoot tissues of lemongrass varieties in sodic soil (ESP 18.0 and 36.0)**

Varieties	Na				K				Ca			
	First harvest		Fourth harvest		First harvest		Fourth harvest		First harvest		Fourth harvest	
	18.0	36.0	18.0	36.0	18.0	36.0	18.0	36.0	18.0	36.0	18.0	36.0
Pragati	0.28 ±0.03	0.37 ± 0.04	0.22 ± 0.02	0.27 ±00.30	1.33 ± 0.13	1.02 ± 0.10	1.20 ± 0.11	1.05 ± 0.10	1.05 ± 0.10	0.94 ± 0.09	0.96 ± 0.10	0.48 ± 0.05
Praman	0.14 ± 0.01	0.15 ±0.015	0.12 ± 0.01	0.16 ± 0.01	1.19 ± 0.12	1.06 ±0.10	0.73 ± 0.08	0.73 ± 0.07	0.94 ± 0.09	0.84 ± 0.08	0.84 ± 0.07	0.72 ± 0.06
CKP-25	0.30 ± 0.03	0.43 ± 0.04	0.31 ± 0.03	0.63 ± 0.07	0.95 ± 0.09	0.85 ± 0.08	0.81 ± 0.08	0.59 ± 0.05	1.05 ± 0.10	0.63 ±0.05	0.72 ± 0.07	0.48 ± 0.05
Cauvery	0.25 ± 0.03	0.37 ± 0.04	0.58 ± 0.06	0.75 ± 0.08	0.94 ± 0.08	0.68 ± 0.06	0.81 ± 0.08	0.69 ± 0.07	1.26 ± 0.13	1.05 ± 0.10	0.72 ± 0.07	0.60 ± 0.07
Nima	0.10 ± 0.01	0.15 ± 0.02	0.10 ± 0.01	0.12 ± 0.01	1.34 ± 0.14	1.34 ± 0.15	0.93 ± 0.09	0.62 ± 0.06	1.05 ±0.12	0.94 ± 0.10	0.72 ± 0.08	0.6 ± 0.06

* Values are means of three replicates (± standard deviation).

analysis. The essential oil content in the fresh herbage was determined by hydro-distillation on Clevengers (1928) apparatus. The oil yield was computed by multiplying the herb yield by oil content.

The analysis of essential oil for quality was performed on GLC mode HP with FID and a capillary column DB-Wax (30 m x 0.32 mm x 0.25 μm) using a temperature program from 100°C to 140°C at 5°/m with initial and final hold of 2 minutes, then up to 220°C at 6°/m and H_2 carrier at 10 psi inlet pressure. The injector and detector temperature of 220°C and 240°C, respectively, was applied in the analysis. Components were identified by retention time as compared with those produced by standard samples. The free proline content in shoot tissues at first and second harvest was estimated following Bates *et al.* (1973). Plant samples were thoroughly washed with distilled water, oven-dried at 65°C, ground in Wiley mill to pass through 0.5 mm screen and digested in mixture of HNO_3:$HClO_4$ (10:4, v/v). Calcium in the digested tissues was determined by versanate method and sodium and potassium by flame photometer (Jackson, 1973).

All the treatments were replicated three times in a completely randomized design. Differences in the herbage and oil yield were statistically analyzed for significance by analysis of variance and treatment means were compared using the least significant differences (LSD) test at 0.05 probability.

Results and Discussion

Herbage and essential oil yield

All the five varieties of lemongrass evaluated differed significantly in their herbage and essential oil yields when grown in sodic soils of ESP 18.0

*Table 4. K/Na and Ca/Na ratives in shoot tissues of lemongrass varieties in sodic soil (ESP 18.0 and 36.0)**

Varieties	K/Na ratios				Ca/Na ratios			
	First harvest		Fourth harvest		First harvest		Fourth harvest	
	18.0	36.0	18.0	36.0	18.0	36.0	18.0	36.0
Pragati	4.75 ± 0.50	2.75 ± 0.28	5.45 ± 0.56	3. 81 ± 0.39	3. 75 ± 0.40	2.54 ± 0.25	4.36 ± 0.45	1.77 ± 0.10
Praman	8.50 ± 0.90	7.06 ± 0.70	6.08 ±0.70	4.56 ± 0.45	6. 71 ± 0.70	5.60 ± 0.60	7.0 ± 0.65	4.50 ± 0.35
CKP-25	2.37 ± 0.24	1.98 ± 0.20	2.61 ± 0.30	0.93 ±0.09	2. 60 ± 0.28	1.46 ± 0.15	2.32 ± 0.23	0.76 ± 0.08
Cauvery	3.76 ± 0.40	1.83 ± 0.20	1. 39 ± 0.15	0. 92 ± 0.09	5. 04 ± 0.50	2.83 ± 0.30	1.24 ± 0.13	0.80 ± 0.09
Nima	13.4 ± 1.40	8. 93 ± 0.91	9. 30 ± 0.90	5. 16 ± 0.55	10. 5 ± 1.10	6.16 ± 0.60	7.20 ± 0.75	5.00 ± 0..55

*Values are means of three replicates (± standard deviation).

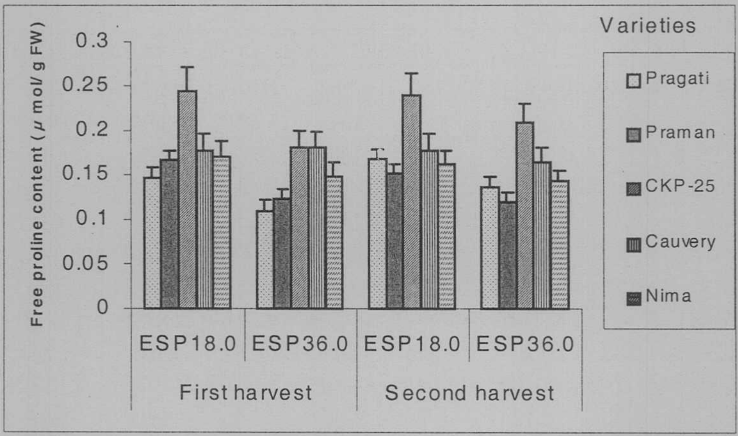

Fig. 1. Free proline content in shoot tissues of lemongrass varieties grown in sodic soils (ESP18.0 and 36.0). Vertical bar represent standard deviation of three replicates.

or 36.0 (Tables 1 and 2). The total herbage yield was higher in variety Praman; whereas, the total oil yield was higher in CKP-25, as compared to other varieties. The herbage and oil yield of lemongrass significantly decreased with the increase in soil ESP from 18.0 to 36.0. The decrease in the total fresh herbage yield was 44.2% 37.1%, 44.9%, 40.7% and 38.3% over soil ESP 18.0 by the increase in soil ESP to 36.0 in the varieties Pragati, Praman, CKP-25, Cauvery and Nima, respectively; whereas decrease in the total oil yield by the application of same treatments was 44.0%, 31.9%, 39.6%, 36.4% and 30.7% over soil ESP 18.0, respectively. The significant decrease in yield with increase in soil sodicity might be due to increase in the concentration of Na and decrease in the concentration of K and Ca in shoot tissues. Mass and Hoffman (1977) reported that the effect of salinity and sodicity on non-halophytes was associated with the reduction in growth and yield. However, the adverse effects of soil sodicity on the crop yield depend on the level of sodicity, the ability of crop to tolerate sodicity and agronomic management practices (Gupta and Abrol, 1990). In this study, the lower reduction in the yield of Praman and Nima with the increase in soil ESP showed their higher degree of tolerance to soil sodicity as compared to other varieties. Inter species or varietal differences in the tolerance to salinity and sodicity have been demonstrated by Gupta and Abrol (1990).

Irrespective to soil sodicity level, all the varieties of lemongrass tested in this study differed markedly in the citral content in its essential oil (Table 2) The total citral content in the essential oil was highest in variety Nima and lowest in variety CKP-25. The total citral content was not significantly affected with the increase in soil ESP from 18.0 to 36.0. The non-significant effects of soil salinity on the citral content in the essential oil of lemongrass has also been reported by Singh and Anwar (1985).

Proline accumulation

The concentration of free proline in shoot tissues was significantly higher in CKP-25 at both the harvests as compared to other varieties of lemongrass tested in this study (Fig. 1). The free proline content in shoot tissues increased with increase in salinity and sodicity stress (Rao, 2000). But in this study, the free proline content was relatively lower in all varieties of lemongrass grown at higher soil sodicity (ESP 36.0) than in lower sodicity soils (ESP 18.0). This might be due to the substitution of free proline for sugar as a respiratory substrate in the perennial lemongrass at high salt stress. Substitution of free proline accumulated in salt stressed lentil plants for sugar as a respiratory substrate leading to a reduction in free proline content at later stages (90 and 120 days after sowing) of crop growth has been reported by Tewari and Singh (1991).

Cation accumulation

The concentrations of Na, K and Ca in shoot and root tissues of all the varieties of lemongrass differed significantly in the cation accumulation when grown in sodic soils of ESP 18.0 or 36.0 (Tables 3, 4 and 5). Interspecies and varietal differences in the cation composition of plants grown on saline or sodic soils were expected as

*Table 5. Cation accumulation in root tissues of lemongrass varieties in sodic soil (ESP 18.0 and 36.0) at the fourth harvest**

Varieties	Na		K		Ca	
	18.0	36.0	18.0	36.0	18.0	36.0
Pragati	0.05 ± 0.005	0.09 ± 0.01	0.25 ± 0.02	0.14 ± 0.015	0.36 ± 0.04	0.24 ± 0.024
Praman	0.05 ± 0.005	0.05 ± 0.005	0.07 ± 0.007	0.03 ± 0.003	0.36 ± 0.035	0.24 ± 0.024
CKP-25	0.09 ± 0.01	0.10 ± 0.012	0.12 ± 0.013	0.10 ± 0.011	0.48 ± 0.05	0.24 ± 0.025
Cauvery	0.07 ± 0.006	0.14 ± 0.015	0.16 ± 0.015	0.16 ± 0.016	0.36 ± 0.04	0.24 ± 0.025
Nima	0.05 ± 0.005	0.05 ± 0.005	0.13 ± 0.015	0.12 ± 0.011	0.48 ± 0.055	0.24 ± 0.026

*Values are means of three replicates (standard deviation).

the chemical composition of growing or developed tissue is genetically determined (Mengel and Kirkby, 1982). The concentrations of Na, K and Ca were relatively lower in the root than in the shoot tissues of lemongrass. Salt tolerance of many crops depend upon the efficiency of root system which regulates the excess Na ions to reach the shoots (Khan *et al.*, 1995). In this study a relatively lower accumulation of Na ions in root than in shoot tissues indicated that the lemongrass did not possess a Na regulatory mechanism that prevents the translocation of Na from root to shoot.

The concentration of Na in shoot and root tissues of lemongrass significantly increased and that of K and Ca decreased with the increase in soil ESP from 18.0 to 36.0 (Tables 3 and 5). This might be due to relatively higher availability of Na and lower availability of K and Ca to plants grown in moderately sodic soils (ESP 36.0) than in the slight sodic soil (ESP 18.0). Mengel and Kirkby (1982) reported that the absorption of cations by plant is more or less non-specific process that depends upon cation species in soil solution. Soil sodicity had significant impact on the ionic balance in shoot tissues of lemongrass as evident from the decrease in the K/Na and Ca/Na ratios with increase in soil ESP from 18.0 to 36.0 (Table 4). The K/Na and Ca/Na ratios in shoot tissues were relatively higher in the varieties Praman and Nima than in the other varieties of lemongrass tested in this study. In general, the plants that limit the uptake of toxic ions and maintain normal ranges of nutrient ions could be more salt tolerant than those do not restrict ion accumulation and loose nutrient balance (Shannon, 1990). In the present study a relatively lower accumulation of Na ion and the capacity to maintain higher K/Na and Ca/Na ratios in the Praman and Nima showed their greater tolerance to soil sodicity than the other varieties of lemongrass such as Pragati, CKP-25, and Cauvery.

Acknowledgements

The authors are thankful to the Director, Central Institute of Medicinal and Aromatic Plants, Lucknow, for facilities and encouragement.

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