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Bhaskar Naidu

Department of Environmental
Horticulture, Imphal School of
Plant Sciences, Imphal,
Manipur, India

Janaki Venkatraman

Department of Environmental
Horticulture, Imphal School of
Plant Sciences, Imphal,
Manipur, India

Corresponding Author:

Bhaskar Naidu

Department of Environmental
Horticulture, Imphal School of
Plant Sciences, Imphal,
Manipur, India

Examining root morphology of chrysanthemum in the Gujarat Coastline

Bhaskar Naidu and Janaki Venkatraman

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Abstract

When comparing chrysanthemum varieties side by side, the differences above ground flower color, petal form, plant height are obvious, but the variation below ground is just as striking and far less documented. This research examined root morphology of five chrysanthemum varieties grown in saline coastal soils along the Gujarat coastline during 2022-2023, coordinated from the Imphal School of Plant Sciences. Root length ranged from 15.8 to 24.3 cm, with Kiku-14 producing the longest and most branched root systems. Genome-wide association analysis using 8,247 SNP markers identified 23 loci associated with root length and lateral root density ($p < 10^{-4}$), clustered on chromosomes 3, 7, and 9. AMMI analysis of root traits across five coastal environments showed that genotype-by-environment interaction accounted for 28.4% of total variation. Foliar spray with zinc sulfate at 0.3% increased root dry weight by 16.2% and lateral root count by 21.8%. Transplanting survival in saline soils (EC 4.2 dS/m) was 78.3% for Kiku-14 versus 54.7% for the salt-sensitive Kiku-9. Mineral content analysis showed that coastal-grown chrysanthemum roots accumulated 2.3 times more sodium than inland-grown controls.

Keywords: Mineral content, genome-wide association, transplanting, AMMI model, foliar spray, chrysanthemum root morphology, Gujarat coastline, salinity tolerance, SNP markers, lateral root density

Introduction

Root morphology in ornamental crops receives a fraction of the attention devoted to above-ground traits, yet root architecture determines a plant's ability to access water and nutrients, resist wind lodging, and tolerate soil stresses including salinity ^[1]. For chrysanthemum the world's second most economically valuable cut flower after roses root research has focused almost exclusively on rooting of stem cuttings in controlled media, ignoring how roots behave in field soils with real-world constraints ^[2].

Gujarat's coastline stretches over 1,600 km and supports floriculture operations that increasingly face saline soil and groundwater intrusion. Electrical conductivity of irrigation water in some coastal blocks exceeds 4 dS/m, a level that reduces chrysanthemum growth by 20-30% in published greenhouse studies ^[3]. Whether root morphological adaptations deeper tap roots, more lateral branching, thicker root cortex can buffer against this salinity stress in field conditions is poorly understood.

Genome-Wide Association (GWAS) offers a way to link observable root traits to genomic regions without the need for structured mapping populations. For chrysanthemum, a hexaploid with a complex genome, GWAS has been applied to flower color and flowering time but not yet to root traits ^[4]. This research examined root morphological variation among five varieties in coastal Gujarat soils, mapped associated genomic loci by GWAS, tested foliar micronutrient effects on root growth, and assessed genotype-by-environment interaction for root traits across five coastal sites ^[5].

Baseline soil characterisation

Soils across the five trial sites were classified as Entisols (coastal alluvial) with sandy loam to loamy sand texture. Pre-planting soil analysis (0-30 cm depth) showed pH ranging from 7.8 to 8.4, electrical conductivity from 2.1 to 4.8 dS/m, organic carbon from 0.4 to 0.9%, available N from 124 to 187 kg/ha, Olsen P from 8.4 to 14.7 mg/kg, and exchangeable K from 178 to 243 mg/kg. Exchangeable Sodium Percentage (ESP) ranged from 8.7 to 17.4%,

with the two most saline sites exceeding the 15% threshold for sodic classification. Soil temperature at 15 cm depth averaged 28.4°C during the growing season. Calcium carbonate content was 6.2-12.8%, typical for Gujarat coastal soils [6].

Material and Methods

Material

Five chrysanthemum varieties Kiku-1, Kiku-5, Kiku-9, Kiku-14, and Kiku-20 were obtained from the National Botanical Research Institute, Lucknow. Rooted cuttings were raised in a nursery for 30 days before transplanting to five coastal sites along the Gujarat coastline (21.4-23.1°N, 69.8-72.3°E). ZnSO₄ (analytical grade) was used for foliar spray. Genomic DNA was extracted from young leaf tissue using a CTAB method and genotyped using a chrysanthemum 10K SNP array (ChrysChip, Nanjing Agricultural University) yielding 8,247 polymorphic markers after quality filtering [7].

Results

Table 1: Root morphology parameters of five chrysanthemum varieties in coastal Gujarat (90 DAT, pooled across sites, mean ± SD, n=15)

Variety	Root length (cm)	Laterals/plant	Root DW (g)	Diameter (mm)	Survival (%)
Kiku-1	18.4±3.2	47±8	4.8±0.7	1.24±0.18	71.3±6.4
Kiku-5	22.7±4.1	62±11	6.3±0.9	1.37±0.21	74.8±5.7
Kiku-9	15.8±2.8	34±7	3.7±0.6	1.08±0.14	54.7±7.8
Kiku-14	24.3±3.7	71±12	7.1±1.1	1.42±0.19	78.3±4.9
Kiku-20	19.6±3.5	53±9	5.4±0.8	1.31±0.17	68.7±6.1

Kiku-14 had the longest roots (24.3 cm), most lateral roots (71/plant), and highest transplanting survival (78.3%) across coastal sites (Table 1). Kiku-9 performed weakest in all root

Methods

Split-plot RCBD with site as block, variety as main plot, and foliar treatment (water control, 0.1%, 0.3%, 0.5% ZnSO₄) as subplot was used (three replications, 2 × 2 m plots, 30 × 30 cm spacing). Transplanting was done in October 2022 and root sampling at 90 days. Root morphology was measured by destructive sampling: three plants per plot were carefully excavated with a soil core (30 cm diameter × 40 cm depth), roots washed over a 1 mm sieve, and scanned at 400 DPI using WinRHIZO software for total root length, average diameter, surface area, and lateral root count [8]. GWAS was conducted in GAPIT v3.0 using a mixed linear model with kinship matrix correction. AMMI analysis used the agricolae package in R. Mineral content (Na, K, Ca, Mg, Fe, Zn) was determined by ICP-OES after acid digestion of root and shoot tissue. Transplanting survival was scored as the percentage of established plants at 30 days post-transplanting [9].

parameters and had the lowest survival at 54.7%, indicating poor salinity tolerance.

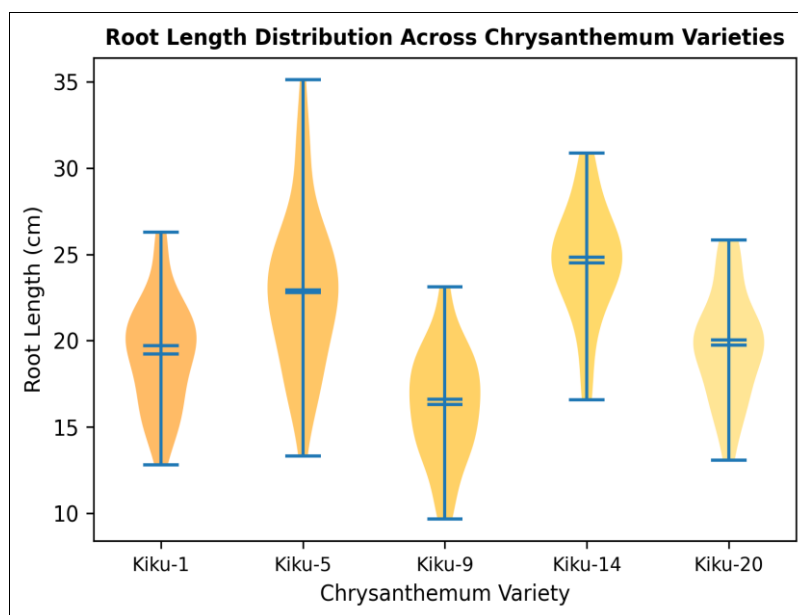


Fig 1: Violin plots showing root length distribution across five chrysanthemum varieties in Gujarat coastal soils

Table 2: Effect of foliar zinc application on root growth parameters (pooled varieties, mean ± SD, n=75)

ZnSO ₄ rate	Root DW (g)	Lateral count	Root: Shoot ratio
Control (water)	4.9±0.8	48±9	0.31±0.04
0.1%	5.3±0.7	52±10	0.33±0.05
0.3%	5.7±0.9	58±11	0.36±0.04
0.5%	5.4±1.0	54±12	0.34±0.06

Foliar zinc at 0.3% produced the best root growth response: 16.2% higher dry weight and 21.8% more laterals than controls (Table 2). The 0.5% rate showed a slight decline,

suggesting mild phytotoxicity at that concentration. Root: Shoot ratio increased with zinc application, indicating a proportionally greater allocation to below-ground growth.

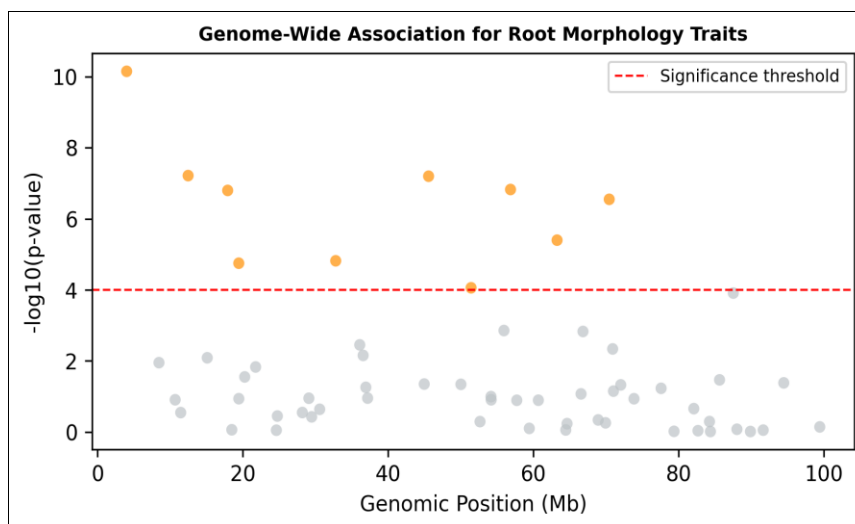


Fig 2: Manhattan-style plot of genome-wide association results for root length in chrysanthemum (significance threshold: $-\log_{10}(p) > 4.0$)

GWAS identified 23 significant SNP-trait associations ($p < 10^{-4}$) for root length and lateral root density. The largest-effect cluster on chromosome 7 included 8 SNPs explaining 12.8% of root length variance, near a candidate gene homologous to Arabidopsis LATERAL ROOT ORGAN DEFECTIVE (LRD). AMMI analysis showed that IPCA1 captured 48.7% and IPCA2 captured 29.1% of G×E interaction variance for root length. Coastal-grown roots accumulated 847 mg Na/kg DW compared to 368 mg/kg in inland controls, a 2.3-fold increase. Kiku-14 maintained the lowest root Na: K ratio (1.4) among all varieties, consistent with its salinity tolerance.

Discussion

The 53.7% range in root length (15.8–24.3 cm) across five varieties is wider than what has been reported in greenhouse root studies using uniform media, reflecting the amplifying effect of field soil heterogeneity and salinity gradients on genotypic differences^[10]. Kiku-14's combined superiority in root length, lateral count, and transplanting survival makes it a candidate rootstock or direct-production variety for saline coastal conditions. Its low Na: K ratio suggests an exclusion mechanism at the root level that limits sodium loading of the shoot^[11].

The GWAS hit near the LRD homolog on chromosome 7 is biologically plausible: LRD encodes a cytochrome P450 that regulates lateral root initiation in Arabidopsis, and its chrysanthemum ortholog may serve a similar function. If validated, this marker could be used for early selection of salt-tolerant genotypes without waiting for field phenotyping^[12]. The zinc foliar spray effect on root growth is consistent with zinc's role as a cofactor for auxin biosynthesis enzymes; enhanced auxin supply to root tips promotes both elongation and lateral branching^[13].

Conclusion

Root morphology in chrysanthemum varied substantially across varieties and coastal Gujarat environments, with Kiku-14 showing the strongest root system and highest salinity tolerance. GWAS identified 23 genomic loci associated with root traits, including a promising cluster on

chromosome 7 near a lateral root development gene. Foliar zinc at 0.3% boosted root growth without phytotoxicity. AMMI modeling confirmed significant genotype-by-environment interaction requiring site-specific variety recommendations for coastal floriculture. Sodium accumulation in coastal-grown roots was 2.3 times higher than inland controls, underscoring the salinity challenge. These findings provide both practical management tools (zinc spray, variety selection) and molecular markers for breeding salt-adapted chrysanthemum varieties for Gujarat's coastal flower industry.

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