



International Journal of Agricultural and
Environmental Research
FREE AND OPEN ACCESS
Available online at www.ijaaer.com
ISSN 2414-8245 (Online)
ISSN 2518-6116 (Print)



ECOLOGICAL AND TAXONOMIC SIGNIFICANCE OF ROOT ANATOMY IN SOME SPECIES AND CULTIVARS OF GENUS *CANNA* L.

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Abstract

*Survival potential of different plant species in varying habitats depend upon their morpho-physiological, biochemical and anatomical characteristics in that specific environment. Diversity in tissue architecture of any plant species is a promising feature in response to various environmental conditions. The present study was aimed to assess structural modifications in root of the seventeen species/cultivars of *Canna* L. (family Cannaceae) in relation to their wide distribution and cultivation in different habitats. Seventeen species/cultivars of *Canna* L. were collected from varying habitats of Faisalabad and its adjoining districts (Punjab, Pakistan). Free hand sectioning and light microscopy technique was used to prepare the double stained permanent slides for the study of different structural modifications of root in relation to variety of environmental conditions. Overall, on the basis of root anatomical studies, the wide distribution of various *Canna* species/cultivars can be attributed to their thicker root epidermis, large cortical cells, greater metaxylem area, smaller vessels and highest aerenchyma area and large pith area.*

Key words: *Canna*, ecology, root anatomy, taxonomy

INTRODUCTION

The knowledge of anatomy is an incredibly significant achievement for not only resolving plant systematic complexities, but also for the structure and function under different environmental conditions (Dickison, 2000; Nikolova and Vassilev, 2011). Anatomical characteristics are being used for systematic purposes for over a hundred years. Anatomical characters of different organs of flowering plants could be taxonomically helpful for the identification of herbarium specimens, fragmentary objects and useful for the establishment of interrelationship between taxa at and above species rank (Metcalf, 1971; Dwari and Mondal, 2011). Initially it is important to make distinction for taxonomic characters, which enable a species or genus to distinguish from other plants and species.

In plant systematics, comparative observations in plant morphology, structure and anatomy is always consider as the backbone. These studies help to clarify phylogeny, diversity and evolution in plants. In the last of 20th century plant systematics and structural studies were greatly influenced by new methods and techniques (Endress et al., 2000). In addition, plants show different anatomical responses to different environments. These anatomical responses depend on the adaptability of the plant in that environment (Stevovic et al., 2011). Plant expresses these adaptations through diverse mechanisms. Roots act as sensors of the plant, which detect soil water potential changes and are important to create resistance in plant by structural adaptive strategies for the survival in adverse environments (Shao et al., 2008).

Cannaceae is a monogeneric family with 19 species of tropical and subtropical American origin

(Tanaka, 2001); however, represented by one cultivated species (*Canna indica* L) in Pakistan (Ghazanfar, 1982). The family Cannaceae has specific anatomical characters. In leaf, mesophyll cells are solitary-prismatic having calcium oxalate crystals. Midrib region has minor leaf veins which are without phloem transfer cells. Stomata are present on leaf epidermis which may be paracytic or tetracytic and guard cells are not like typical grass type. Young stems are cylindrical having secretory cavities. In root, xylem may be with or without vessels (Watson and Dalwitz, 1992).

In this study we examined the variations in root anatomy of 17 cultivars of *Canna* collected from different areas of the Punjab province and investigated the significance of root anatomy in relation to taxonomy and ecology.

MATERIALS AND METHODS

Some cultivars of *Canna* L. (family Cannaceae) were collected from the Faisalabad and its adjoining districts including Chiniot, Jhang and Shaikhupura, (Punjab, Pakistan). Seventeen species/cultivars of *Canna* were collected for anatomical studies, namely *Canna x generalis* 'Adams Orange' (AO), *Canna x generalis* 'Apricot Dream' (AD), *Canna x generalis* 'Bangkok yellow' (BY), *Canna x generalis* 'Firebird' (F), *Canna x generalis* 'Pashion' (Ph), *Canna x generalis* 'Green Wyoming' (GW), *Canna indica*, *Canna x generalis* 'Orange Beauty' (OB), *Canna x generalis* 'Pacific Beauty' (PB), *Canna x generalis* 'President' (Pd), *Canna x generalis* 'Pretoria' (Pt), *Canna glauca* 'Ra', *Canna x generalis* 'Red Dazzler' (RD), *Canna x generalis* 'Sunny Delight' (S), *Canna x generalis* 'Wyoming' (W), *Canna x generalis* 'Yellow King Humbert' (YKH) and *Canna indica* 'Yellow' (Y) (Plate 1). These species/cultivars were assessed for structural modifications in relation to their wide distribution and cultivation in different habitats of the Punjab.

Free hand sectioning technique was used to prepare the double stained permanent slides. One cm piece from the base of the thickest root of adventitious root was selected. The material was first preserved in FAA (formalin acetic alcohol) solution for fixation, which contained v/v formalin 5%, acetic acid 10%, ethanol 50%, and distilled water 35%. The material was then subsequently transferred in acetic alcohol solution (v/v acetic acid 25% and ethanol 75%) for long-term preservation. Ocular micrometer was used to record the

data for dermal, parenchymatous, mechanical and conducting tissues and photographs were taken by Carl-Ziess camera-equipped microscope.

The data was subjected to analysis of variance (ANOVA) in completely randomized design (CRD) for the comparison of mean including three replications (Steel and Torrie, 1997). The data was also subjected to multivariate analysis to assess the similarities and differences among different species and cultivars of *Canna*.

RESULTS

In root anatomical studies, the two regions were investigated which include epidermal region and vascular region. In the epidermal region, we checked the epidermis thickness, cortical thickness and cortical cell area and aerenchyma area while in vascular region, vascular bundle area, metaxylem area and pith area were observed.

There was high variation in the epidermal thickness in different *Canna* cultivars. C x generalis O showed maximum epidermal thickness while minimum epidermal thickness was observed in C x generalis S, C. indica Y, C x generalis W, C x generalis AO, C x generalis BY and C x generalis RD having almost same epidermal thickness (Table 1, Plate 1).

Cortical thickness was more or less equal in all *Cannas pp*. However, highest cortical thickness was noted in C x generalis Pt and C x generalis AD and minimum was calculated in C x generalis AO while cortical cell area showed huge variation in results in which C x generalis YKH and C. indica Y had the highest cortical cell area and C x generalis AD had minimum of this character (Table 1, Plate 2).

In the studies of *Canna* cultivars, aerenchyma was not present in all cultivars. Aerenchymatous tissues were observed in nine cultivars out of seventeen cultivars and having large difference among all nine cultivars in which the highest aerenchyma area was observed in C x generalis Pt while minimum of it was observed in C x generalis F.

In the vascular region, the highest metaxylem area and vascular bundle area was observed in C x generalis RD and lowest was in C x generalis AO. While in pith area characteristic C x generalis F showed maximum of this character and C x generalis AD showed minimum of this character (Table 1, Plate 3).

Table 1: Root anatomical characteristics of some cultivars of family Cannaceae from the Punjab, Pakistan (Mean ± SE)

	Epi. Thickness (μm)	Cortical thickness (μm)	Cortical cell area (μm^2)	Aerenchyma cell area (μm^2)	Vascular bundle area (μm^2)	Metaxylem area (μm^2)	Pith area (μm^2)
C x generalis. F	16.34±4.71	302.29±9.43	227.264±63.03	1502.188±796.57	58599.177±6080.47	1477.217±191.71	4352.982±996.46
C x generalis O	19.0633±2.72	305.013±5.45	340.896±26.22	4640.886±2310.67	45452.822±5340.64	1110.098±176.77	3723.635±793.65
C x generalis Pt	12.255±2.36	345.863±11.87	467.639±45.63	7858.896±3894.86	53844.113±3876.40	1494.698±163.76	1346.103±303.29
C x generalis PB	9.53166±1.36	280.503±5.45	305.932±53.17	0	53844.113±3876.4	2167.749±242.86	2377.532±489.49
C x generalis S	13.61667±1.36	305.013±5.45	349.637±69.93	2232.178±1127.87	35593.056±3149.84	1686.999±130.53	839.129±0
C. glauca	4.085±0	296.843±9.81	257.857±34.13	1798.555±960.53	25523.508±2665.51	1101.357±109.17	716.7560±122.37
C x generalis Ph	5.44667±1.36	315.9067±7.21	467.639±90.94	2656.878±1708.4	35593.056±3149.84	1206.248±52.44	629.347±209.78
C. indica	9.531667±1.36	296.843±2.72	397.713±41.69	0	47340.863±5634.21	987.725±132.27	1888.0403±0
C. indica Y	4.085±0	337.693±7.21	590.013±100.14	4921.883±2439.36	56291.572±6133.32	2045.377±157.34	2342.568±227.26
C x generalis GW	12.255±1.36	302.29±8.17	397.712±41.69	0	25943.073±5336.52	978.984±69.93	2185.232±608.86
C x generalis W	4.085±0	332.2467±7.21	323.414±38.1	0	30348.4998±2907.67	1232.4708±149.1	1346.1028±303.29
C x generalis AO	4.085±0	220.59±4.72	249.1164±39.33	0	10419.185±1697.09	262.228±30.28	996.466±157.34
C x generalis AD	9.531667±1.36	343.14±4.72	166.0776±23.13	0	19649.6047±1328.62	948.3906±64.38	384.6008±87.41
C x generalis BY	4.085±0	231.4833±9.43	305.9324±38.1	4806.5043±2387.96	16992.363±0	1193.1366±187.42	0
C x generalis Pd	9.531667±1.36	343.14±2.72	437.0463±34.96	0	47340.863±5634.21	1573.3669±181.67	2604.796±424.27
C x generalis RD	4.085±0	266.8866±4.72	502.6033±73.52	3145.942±1608.99	63284.3142±16705.63	2185.2318±378.59	2115.3044±227.26
C x generalis YKH	8.17±2.36	313.1833±2.72	598.7535±73.52	0	46012.2417±2801.14	1608.3306±279.71	0



Canna x generalis
'Adam's Orange'



Canna x generalis
'Apricot Dream'



Canna x generalis
'Bangkok Yellow'



Canna x generalis
'Firebird'



Canna x generalis
'Phasion'



Canna x generalis
'Green Wyoming'



Canna indica



Canna x generalis
'Orange Beauty'



Canna x generalis
'Pacific Beauty'



Canna x generalis
'President'



Canna x generalis
'Pretoria'



Canna glauca 'Ra'



Canna x generalis
'Red Dazzler'



Canna x generalis
'Sunny Delight'



Canna x generalis
'Wyoming'

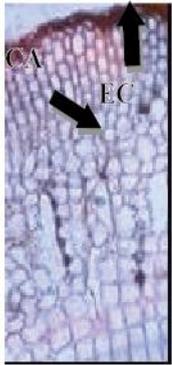


Canna x generalis
'Yellow King Humbert'

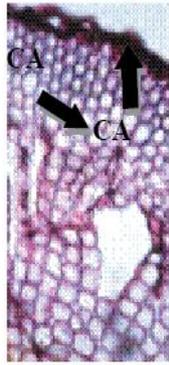


Canna indica 'Yellow'

Plate 1: Seventeen Canna cultivars/species



Canna x generalis
'Firebird'



Canna x generalis
'Orange Beauty'



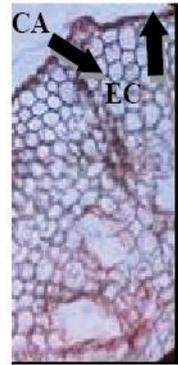
Canna x generalis
'Pretoria'



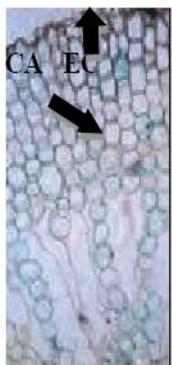
Canna x generalis
'Pacific Beauty'



Canna x generalis
'Sunny Delight'



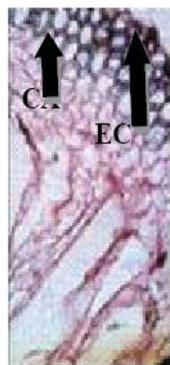
Canna glauca
'Ra'



Canna x generalis
'Phasion'



Canna indica



Canna indica
'Yellow'



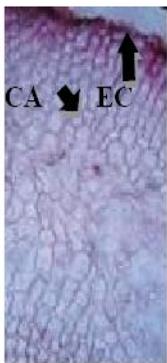
Canna x generalis
'Green Wyoming'



Canna x generalis
'Wyoming'



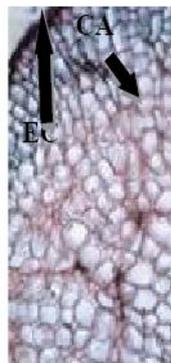
Canna x generalis
'Adam's Orange'



Canna x generalis
'Apricot Dream'



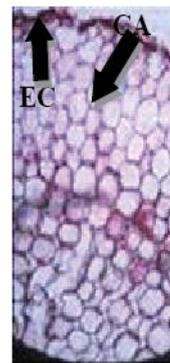
Canna x generalis
'Bangkok Yellow'



Canna x generalis
'President'



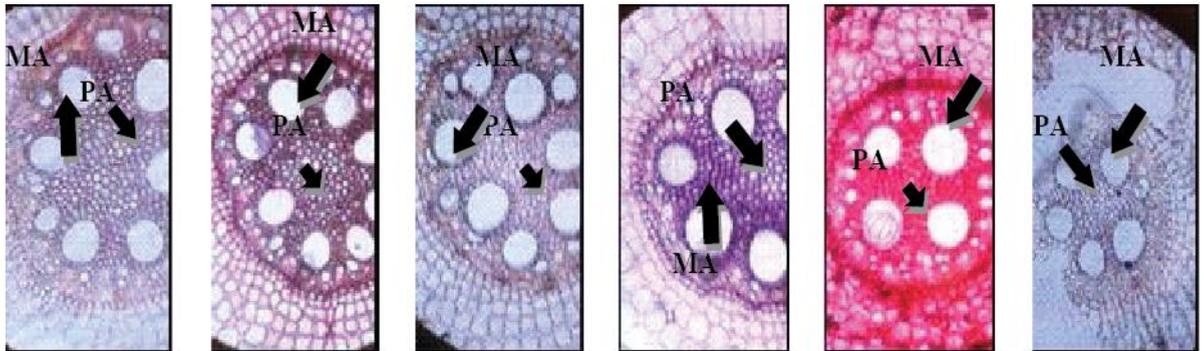
Canna x generalis
'Red Dazzler'



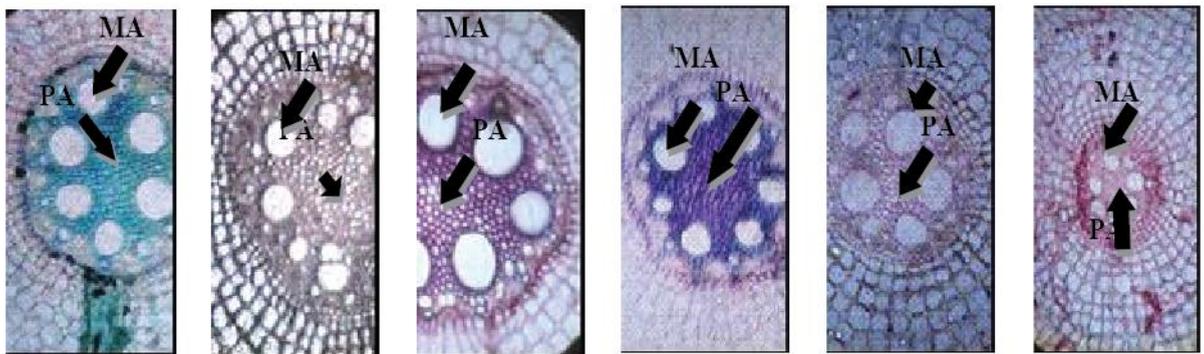
Canna x generalis
'Yellow King Humbert'

Plate 2: Root anatomy (EC and CA) of different *Canna* cultivars/species

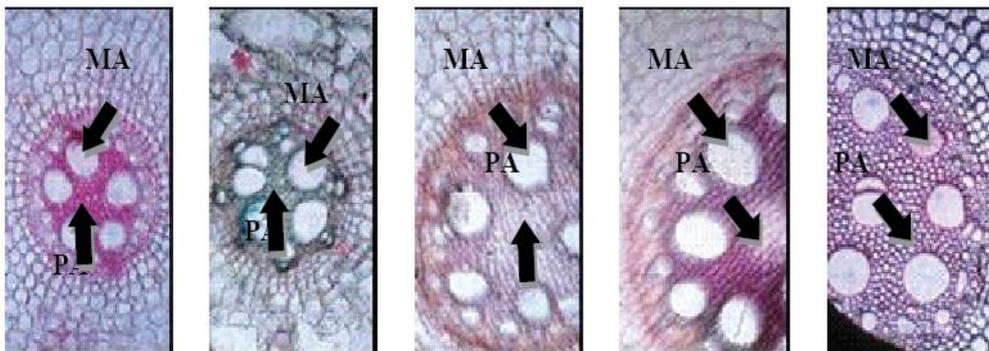
EC=Epidermal Cell, CA=Cortical Cell Area



Canna x generalis 'Firebird' *Canna x generalis* 'Orange Beauty' *Canna x generalis* 'Pretoria' *Canna x generalis* 'Pacific Beauty' *Canna x generalis* 'Sunny Delight' *Canna glauca* 'Ra'



Canna x generalis 'Phasion' *Canna indica* *Canna indica* 'Yellow' *Canna x generalis* 'Green Wyoming' *Canna x generalis* 'Wyoming' *Canna x generalis* 'Adam's Orange'



Canna x generalis 'Apricot Dream' *Canna x generalis* 'Bangkok Yellow' *Canna x generalis* 'President' *Canna x generalis* 'Red Dazzler' *Canna x generalis* 'Yellow King Humbert'

Plate 3: Root anatomy (VA) of different *Canna* cultivars/species

VA=Vascular Area, MA=Metaxylem area, PA=Pith area

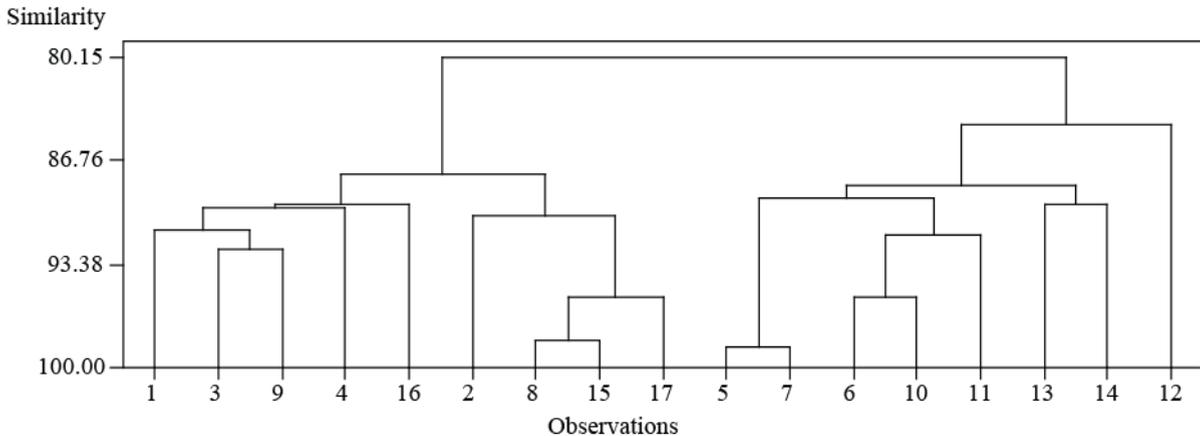


Fig 1: Dendrogram of root anatomical parameters of some *Canna* cultivars from Punjab, Pakistan
 1. *C x generalis*. F 2. *C x generalis* O 3. *C x generalis* Pt 4. *C x generalis* PB 5. *C x generalis* S 6. *C. glauca*
 7. *C x generalis* Ph 8. *C. indica* 9. *C. indica* Y 10. *C x generalis* GW 11. *C x generalis* W 12. *C x generalis* AO
 13. *C x generalis* AD 14. *C x generalis* BY 15. *C x generalis* Pd 16. *C x generalis* RD 17. *C x generalis* YKH

MULTIVARIANT ANALYSIS

Dendrogram of root anatomical characteristics of seventeen cultivars of *Canna* showed two distinct groups in which nine cultivars were clustered in one and eight were clustered in other group. First cluster with nine cultivars showed two sub groups and second cluster with eight cultivars showed three subgroups. In first cluster, one subgroup contains three cultivars (*C. indica*, *C x generalis* Pd and *C x generalis* YKH) showing close relation to *C x generalis* O while second subgroup contains five cultivars (*C x generalis*. F, *C x generalis* Pt, *C. indica* Y, *C x generalis* PB and *C x generalis* RD) in which *C x generalis* Pt and *C. indica* Y more closely related to each other following the *C x generalis*. F, *C x generalis* PB and *C x generalis* RD cultivars. In second cluster, *C x generalis* S and *C x generalis* Ph in one subgroup showed more close relation. In second subgroup *C. glauca* and *C x generalis* GW showed close relationship with each other following the *C x generalis* W cultivar while third subgroup have three cultivars (*C x generalis* AO, *C x generalis* AD and *C x generalis* BY (Fig. 1).

DISCUSSION

Roots as the plant sensors are important for the plant adaptability in a variety of environments (Shao et al., 2008). Root epidermis being outermost layer

of the plant is very important structure, which connect the internal plant environment with external environment. It plays a critical role to control water movement inside the plant (Saleem et al., 2010). Thicker root epidermis of *C x generalis* O may provide it an ecological benefit for the adaptability in different environmental types.

Cortical region is the fundamental part of root and structurally and functionally represents the important constituent that might serve as a storage area for water and nutrients (Hameed et al., 2009). Large cortical cells of *C x generalis* YKH and *C. indica* Y may provide ecological success to these *Canna* cultivars for the adaptation to survive under environmental stresses and indicate their wide distribution (Zwieniecki and Newton, 1995), while small cortical cells indicate the plant response to low water potential or drought conditions. It is also described by Pena-Valdivia et al. (2010) in common Beans (*Phaseolus vulgaris* L.).

C x generalis RD cultivar had greater metaxylem area and it is well documented that larger vessels have low resistance to water conduction and dramatically increase the water conduction efficiency (Nicotra et al., 2002) but on the other hand these larger vessels are more prone to embolism/cavitation under water stress conditions. Whereas *C x generalis* AO. had smaller vessels which support its wide distribution as small vessels indicate the increased hydraulic safety (Durante et al., 2011).

Aerenchyma is the spongy tissue having large air spaces (Evans, 2003), which provide low internal resistant pathway for storage and gas exchange within plant (Visser et al., 1997, 2003). Aerenchyma were present in nine cultivars, while highest aerenchyma area observed in roots of *C x generalis* Pt cultivar, it support the distribution of these cultivars in anaerobic environments as it is well documented that maximum penetration and survival of roots in anaerobic sediments depends on aerenchyma tissue in roots which provide adequate oxygen supply to the surrounding cells (Armstrong et al., 1991) Maximum pith area was observed in *C x generalis* F cultivar and it is reported that additional water storage occurs in pith cells which is helpful in severe climatic conditions (Akram et al., 2002). So, that it could be helpful for Canna cultivar to survive under a variety of environmental conditions.

CONCLUSION

In the end, it can be said that different Canna cultivars showed different anatomical characteristics in relation to their ecology. Thick root epidermis in *C x generalis* O, Large cortical cells of *C x generalis* YKH and *C. indica* Y, greater metaxylem area in *C x generalis* RD cultivar, smaller vessels of *C x generalis* AO and highest aerenchyma area observed in roots of *C x generalis* Pt cultivar and maximum pith area in *C x generalis* F revealed their wide distribution in a variety of environment.

On the basis of multivariate analysis, it can be predicted that anatomical characters can support for taxonomic studies to some extent. As in present investigation, seventeen canna species/cultivars were divided in main two clusters in accordance to anatomical similarities, in which seven (*C x generalis* O, *C x generalis* YKH, *C. indica* Y, *C x generalis* RD, *C x generalis* Pt, *C x generalis* F) were concluded as more adaptive to environmental heterogeneity than the others on the basis of anatomical features. Six canna species/cultivars of these are included in first cluster in multivariate analysis. This showed their resemblance in accordance with anatomical features and their adaptability to environmental heterogeneity and therefore, the validity of anatomical data cannot be ignored in the taxonomic studies.

ACKNOWLEDGEMENT

The authors greatly acknowledge the department of Botany, University of Agriculture, Pakistan for

providing lab space and other necessary equipment for research work. This is the part of MPhil work of Ms. Nighat Noreen.

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