BIOLOGY OF GREWIA TENAX COMPLEX (TILIACEAE) FROM PAKISTAN

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Abstract

Grewia tenax (Forssk.) Fiori was hitherto considered as a highly polymorphic taxon. A biological study of this complex from Pakistan indicates the existence of two distinct taxa viz., G.tenax(Forssk.) Fiori and G. erythraea Schweinf. G. tenax is characterized by having taller growth habit, ovate-acute leaves and deeply 4-lobed glabrous ovary; while G.erythraea is delimited by smaller individuals with obovate-obtuse leaves and entire densely hairy ovary. Two taxa also differ on the basis of chemical and palynological characters. Both the species are facultative xenogamous, sympatric in distribution and the flowering period is also overlapping in southern Pakistan. An intermediate and fairly heterogeneous population was also found, thus indicating a possibility of a limited hybridization taking place with a limited exchange of genes in southern Pakistan.

Introduction

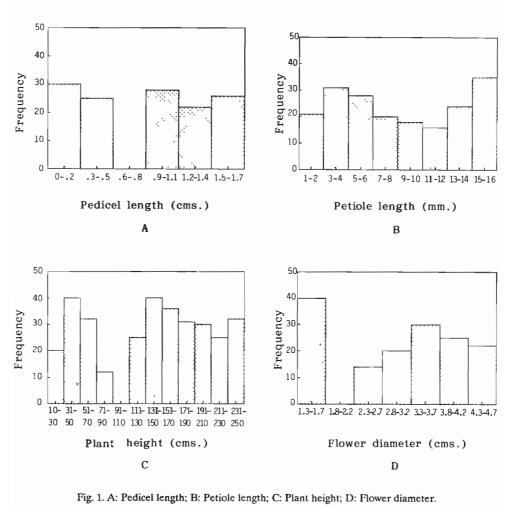
The genus Grewia L. of the family Tiliaceae, with c. 150 species is widely distributed in the tropics of the Old World (Willis, 1973). In Pakistan it is represented by 10 species (Ghafoor, 1974), of which two species viz., Grewia tenax (Forssk.) Fiori and G. villosa Willd., are quite frequent in the plains of Pakistan. Stewart (1972) reported one more species, Grewia erythraea Schwienf., from Pakistan. However, he indicated his doubts, wheather this species could be segregated from G.tenax. Several other workers viz., Fiori (1912) and Miller & Morris (1988) treated the two taxa as independent species. Ghafoor (1974) reduced Grewia erythraea to the synonymy of G.tenax considering the later species as highly polymorphic. He was of the opinion that the variation in the leaf size and shape and petiole length was continuous. On the other hand Farooqi & Khushbakht (1967) demonstrated the occurrence of two taxa on the basis of leaf shape and petiole length. They showed a clear discontinuity in the aforesaid characters. However, they did not accord any taxonomic rank to them. Rechinger & Esfandyari (1950) described a new species from Makran viz., Grewia makranica Rech.f., on the basis of leaf shape and height of the plant. Browicz (1981) while revising the family Tiliaceae for "Flora Iranica" reduced G. makranica as a subspecies of G. tenax.

In order to ascertain the actual number of taxa occurring in Pakistan, experimental studies of the complex were carried out where besides morphological features, palynological and phytochemical characteristics and breeding behavior was also examined.

Material and Methods

Population samples were collected from Karachi University campus, Manghopir hillocks, North Karachi, Malir Cantonment, Turbat, Panjgur, Kirther range (Dadu Dist.) and Jhelum (Fig 1). Herbarium specimens collected from different parts of Pakistan and available in Karachi University Herbarium (KUH) and National Herbarium (RAW), were also examined. Following parameters were studied, where about 100 plants were analyzed for each character.

Morphology: Habit, leaf length, leaf breadth, leaf shape, degree of pubescence of leaves, petiole length, pedicel length, flower size, stigma shape, degree of pubescence of fruits, shape of ovary, degree of pubescence of ovary were noted.



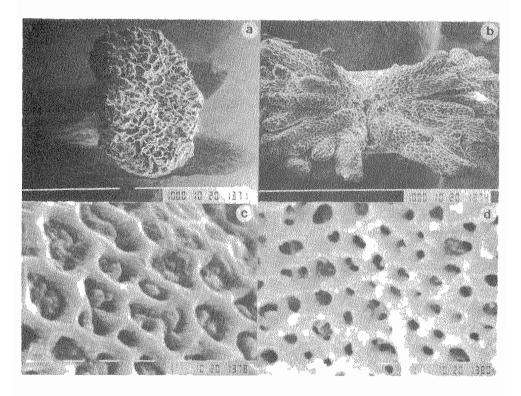


Fig. 2. Scanning Electron micrograph of G. tenax Complex A: stigma surface of G. tenax B: G. erythraea C: Pollen surface of G. tenax D: G. erythraea.

Phenolic Constituents: Phenolic constituents of the leaves of about 60 plants from different populations were analyzed according to the standard procedure of Bate-Smith (1962) and Harborne (1973). The phenolic constituents were identified with the help of authentic markers, Rf values and colour under UV light with and without ammonia fumigation.

Pollen Morphology: Pollen were acetolyzed following the procedure of Erdtman (1952). For SEM, acetolyzed as well as fresh pollen were coated with gold by conventional method. The pollen were scanned on SEM model JSM T- 200.

Pollen Fertility: For testing the pollen fertility, pollen were left overnight in acetocarmin and cotton blue. The red or blue stained pollen were scored as fertile and unstained as sterile.

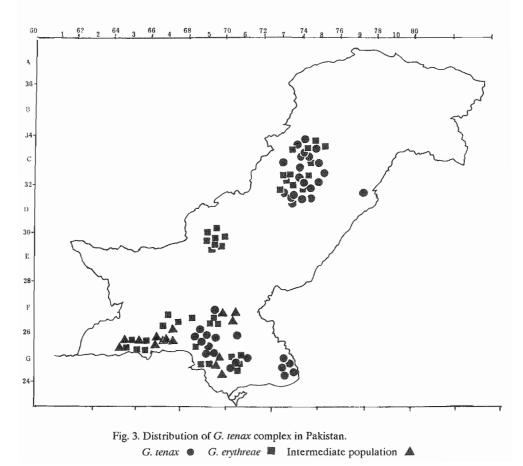
Breeding Behaviour. To check the breeding behaviour, 200 flowers were bagged, of which 50 buds were emasculated and bagged to check apomixis.

Results and Discussion

Frequency distribution of plant height, pedicel length and flower diameter were bimodal and showed the presence of two distinct populations (Fig 1). Study of corre-

lation of various characters indicates that *G. tenax* complex consists of two morphologically distinct populations which can be differentiated from each other on the basis of habit, plant height, pedicel length and flower diameter, which is also correlated with the leaf shape, indumentum type, shape of the stigma and ovary. On the basis of these characters, 2 populations were recognized. The first population includes semierect habit (not more than 90 cm tall) with stunted and profuse branches, shorter petioles (1-6 mm) leaves smaller 2-3 x 1-2.5 cm, obovate, completely glabrous, flowers of c, 1.8 cm diameter, with 1-5 mm long pedicel. Ovary entire densely hairy with two lobed stigma and hairy fruits. The second population includes erect plants (> 110 cm tall) with slender branches, longer petioles (6-12 mm long), larger leaves 3.5 - 7.8 x 3-7 cm, generally ovate to acute, stellately hairy, 6-7 mm long pedicel, larger flowers (2.2-4.5 cm in diameter), 4-lobed completely glabrous ovary and 8 - 12 lobed stigmas (Fig 2), glabrous fruits. However, few intermediates were observed from southern Pakistan which are designated here as a third population (Fig.3 & 4).

Phenolic constituents: The results of phenolic constituents are also in accordance with the morphological findings (Table 1). In the first group, luteolin 7-3'-0 diglucoside,



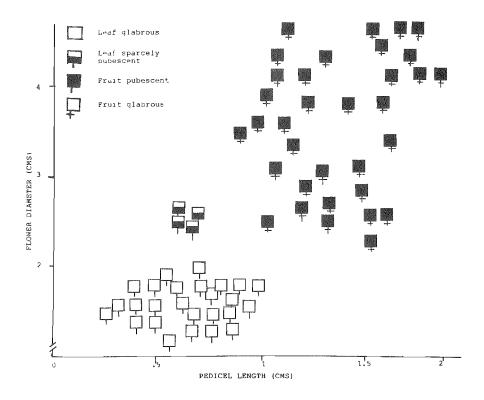


Fig. 4. Scatter diagram of Grewia tenax complex.

luteolin 7-9 diglucornide, Rutin, Chrysoreol 7-0- glucornide and 4 unidentified spots were observed. In group two, Rutin, Chrysoreol- 7-0-glucornide and unidentified spots were common with group 1, while luteolin 7-3'-0- diglucoside, luteolin 7-glucornide, unidentified spots (Rf 8-10 in BAW and 25 in 15% HOAC; Rf 20 in BAW and 92 in 15% HAOC) were absent, while Apigenin 7-0 glucoside and spot 1 was absent. Table 2 indicates that the third population is fairly heterogeneous in its chemical constituents, where ferulic acid, p-coumaric acid, pelargonidin, myricetin, quercetin 4-glucoside were present from all localities, while peonidine was absent from the population of Makran region. Quercetin 3-arabinose and quercetin 3-glactoside were absent in specimen from Sibi and Dadu, respectively. Chrysoereol was absent in specimen from Makran region. Unknown spot 1 with Rf 82 in Forestal was absent in specimen from Sibi and spot 111 with Rf 92 was absent in specimen from Karachi and Dadu.

Pollen Morphology: No appreciable differences were observed in the pollen size. In both the cases the pollen are tricolporate, however, in case of the first group the tectum is coarsely reticulate as compared to the group two. (Fig 2)

Pollen fertility: In case of first and second group the pollen fertility was very high as compared to third population, which was as low to 36% (Table 3).

Table 1. Phenolic constituents of Grewia tenax and G. erythraea.

Flavonoles/Flavonol Glycoside	G. erythraea	G. tenax
Luteolin 7-3'-0 diglucornide	+	_
Luteolin 7- glucornide	+	-
Unidentified	+	+
Unidentified	+	-
Rutin (Quercetin 3-rutinose)	+	+
Chrysoeriol 7-0-glucornide	+	+
Apeginine	-	+
Phenolic Acid		
Unidentified	+	-
Unidentified	+	-
Unidentified	-	+

Pollen Ovule ratio: Table 4 indicates that both the groups have high number of pollen/anther, 235 and 250 respectively and in the third population as low as 115.

Breeding System: In both the population all the bagged flowers developed into fruits. All the emasculated flowers did not produce fruits, thus indicating that basically both the populations are amphimictic and inbreeders (Table 5). However pollen-ovule ratio clearly indicates that both the populations are facultative xenogamous (Cruden, 1977).

Insect Visitation: Inspite of the inbreeding a constant visit of the pollinators was observed.

Browicz (1981) has recognized two subspecies in this complex where the former taxon was regarded as G. tenax subsp. makranica (Rech.f.) Browicz, and the latter taxon with larger leaves and erect habit as the type subspecies. Comparing the 2 groups with the type of G. tenax the group 2 conforms fully with G. tenax (Forssk)

Table 2. Phenolic constituents of Intermediate Population of *Grewia tenax* complex from different localities of Pakistan.

Phenolic constituents	Localities				
	Karachi	Turbat	Panjgur	Dadu	Sibi
Phenolic Acid					
Ferulic Acid	+	+	+	+	+
P-Coumaric acid	+	+	+	+	+
Leucoanthocyanidines					
Pelargonodi	+	+	+	+	+
Peonidin	+	-	-	+	+
Flavonols					
Myricetin	+	+	+	+	+
Quercetin 4-glucoside	+	+	+	+	+
Quercetin 3-arabinoside	+	+	+	-	+
Quercetin 3-glactoside	+	-	-	-	+
Flavone					
Apigenine	+	+	+	+	+
Chrysoeriol	+	-	-	+	+
Unknown in Forestal					
Spot 1 with Rf (X 100) 82	+	+	+	+	
Spot 11 with Rf (X100) 86	+	+	+	+	-
Spot 111 with Rf (X100)92		+	+	+	+

Table 3. Pollen fertility in Grewia tenax complex.

Taxa	Pollen fertility %			
	Min.	Mean	Max.	
G. tenax	67.0	81.5	90.0	
G. erythraea	66.4	76.2	85.5	
Intermediates	30.0	36.5	45.0	

Table 4. Pollen Ovule ratio in Grewia tenax complex.

Taxa	Avg. No. of pollen/anther	Total No. of pollen/flower		Pollen/ ovule	Breeding system
G. tenax	226	13560	81695	Facultative	xenogamy*
G. erythraea	164	9240	8	1155	-do-
Intermediate population	118	4720	8	590	-do-**

^{*} classification followed is that of Cruden (1977).

Table 5. Breeding System in Grewia tenax Complex.

TAXA	Sample Size	Selfing Percentage	
G. tenax	30	80.3	
G. erythraea	30	70.8	
Intermediate Population	30	40.1	

^{**} Almost intermediate between facultative autogamy and facultative xenogamy.

Fiori, while group 1 is similar to *G. erythraea* Schweinf., which was doubtfully recorded from our area or even sometimes treated as conspecific with *G. tenax* by previous workers. A key and complete synonymy of both the species is as follows.

Key To The Species

+ Plants erect, leaves stellately pubescent, fls 2.2-4.5 cm in diameter, ovary 4 lobed, glabrous, stigma 8-12 lobed ------ G. tenax - Plants semi-erect, leaves glabrous, fls 1.2-1.8 cm in diameter, ovary entire, hairy, stigma 2 lobed. ------ G. erythraea

Grewia tenax (Forssk.) Fiori in Agric. Colon 5: Supp. 23, 1912; Stewart R.R.,(1972) An Annotated Catalogue of the Vascular Plants of West Pakistan and Kashmir. In: Flora of Pakistan (Eds.) E. Nasir and S.I.Ali, 473-474.

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Chadara tenax Forssk. Fl. Aegypt-Arab., 105. 1753.

Type: Egypt Forsskal 388 (C) IDC (!)

Grewia erythraea Schwienf.,in verl. Zool. Bot. Ges. Wien 18: 671 1868; Stewart l.c.pp.; Ghafoor l.c.pp.

Syntypes: Ehrenberg (Djara Prope Gumfuda), Schwienfurth, 24488-2491 (M. Uaratab Prope Suakin).

G. makranica Rech.f., & Esfand, Anz. Math.- Nat. Kl. Oesterr. Akad. Wiss. 87300 (1950).

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Type: Iran - Makran: Chah Bahar, 8-4. 1949, Sharif 771 (E!,W)

HYBRIDIZATION: Inspite of the fact that both the species are inbreeders, the presence of a third intermediate population (Fig 4) indicates that both the taxa are involved in hybridization on a limited scale, particularly in southern region of Pakistan. This is also substantiated by the fact that both the species are facultative xenogamous (Table 4), sympatric in distribution and probably the flowering period is also overlapping in southern Pakistan. However, in northern Pakistan the flowering period of both the taxa is different. G. tenax flowers from June to August, while the flowering period of G. erythraea is from March to June. Since the flowering period coincides in southern parts, it is possible that a limited number of cross fertilization is taking place with a limited exchange of genes.

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