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SEED CHARACTERISTICS, GERMINATION AND PHENOTYPIC PLASTICITY OF *TEPHROSIA UNIFLORA* POPULATIONS IN SOUTHERN SINDH

A. Zarina , I.A. Siddiqui, S. S. Shaukat and R. Shaukat

Department of Botany, Federal Urdu University of Arts, Science and Technology
Department of Botany, University of Karachi, Karachi-75270, Pakistan

ABSTRACT

Three populations of *Tephrosia uniflora* Pers. were studied in various habitats of the southern Sindh, namely: (i) rocky soil (ii) calcareous sandy loam soil and (iii) saline sandy soil. The populations differed considerably with regard to number of flowers and fruits per plant. The mean seed weight of *Tephrosia uniflora* for the saline soil population was 0.0214g ± 0.00076, for calcareous soil 0.0276g ± 0.00081 and for the rocky soil population it was .0184g± 0.0074.

Seed weight variation within pods showed that seeds at distal end of 5-seeded pods of *Tephrosia uniflora* had lower weight in saline habitat. In calcareous sandy loam soil, seeds at distal end of 4,5 and 7-seeded pods had lower weight. In rocky soil distal end seeds of 4,6,7,8 and 9-seeded pods had lower weights than the rest of the seeds though such differences were non-significant..

Tephrosia uniflora exhibited external seed dormancy owing to hard seed coat which could be broken by physical scarification or puncturing the seed coat. Large seeds of *Tephrosia uniflora* invariably produced larger seedlings than those produced by smaller seeds.

The crude reproductive effort of *Tephrosia uniflora* was high and it was highest for the population associated with saline soil.

Key words: Seed characteristics, germination, *Tephrosia*

INTRODUCTION

Seed is considered as a stored part of the population. Different studies have been conducted to evaluate response of seeds to environmental complex. Seed is the commonest form of dormancy organ. Fresh seeds of knot-grass (*Polygonum aviculare*) show innate dormancy which prevents germination before the winter but this is broken by stratification and seedling emergence in the spring. Seeds which have broken their innate dormancy but remain in enforced dormancy till the end of May and then acquire renewed (induced) dormancy that prevents germination until another period of winter (Roberts and Feast, 1970).

The distribution of biomass among plant organs is not fixed. It is influenced by the environment, habit of the plant, life span of the plant, competitive interactions, etc. (Rodin and Bazilevich,1967; Gadgil and Solbrig, 1972; Abrahamson and Gadgil, 1973; Abrahamson, 1980). Reproductive allocation did vary significantly with plant density (Snell and Burch 1975; Ogden, 1968).

Seed weight is an ecologically important characteristics in plant because it influences dispersal ability (Harper *et al.*, 1970; Baker, 1972; Werner and Platt 1976, Zhang and Maun, 1990), growth (Manasse, 1990) and seedling establishment (Winn, 1985; Gross and Werner, 1982).

Relatively few studies, however, have examined whether seed weight effect on seedling growth and survival are comparable in different environment (Gross, 1984; Winn 1985; Wulff, 1986a). Length of fruit is also important. It has relation with seed number (Holsinger and Ellstrand,1984). Seed mass is positively correlated with seedling size in many herbaceous plant species (Weis, 1982; Gross, 1984). Several recent studies have also shown interspecific differences in seed mass can influence seedling competitive ability, growth and establisment (Stanton, 1984; Gross, 1984; Winn, 1985, Wulff, 1986b). However, it is not clear how long the effects of seed mass had diminished in *Lupinus texensis* at the time when genetic differences in seedling performance were first selected (Schaal, 1980).

Tephrosia uniiflora Pers. (family Fabaceae) a perennial herb, is widely distributed in southern Sindh. It has small seeds with hard seed coat that exhibit imposed dormancy. It can grow in different soil types *i.e.,* saline, rocky and calcareous soils. Germination commences after rainfall in July-August and seed ripening begins in October. Seed dispersal occurs during October to November.

MATERIALS AND METHODS

1. Studies on phenotypic plasticity of populations

The populations of *Tephrosia uniflora* were studied in three different habitats: a rocky site located at Pipri, a site with calcareous sandy loam soil and a saline sandy habitat, the latter two being located at Karachi University campus, Karachi. Data was obtained on number of fruit / plant, number of flower / plant and number of seed / plant. Twenty plants were examined at each site. Fruit length and the number of seeds / fruit were examined for hundred fruits from each population. Seeds of *Tephrosia uniflora* Pers. were collected during October 2002 and stored in glass bottles at room temperature (25º C). Two hundred seeds from each population were individually weighed to examine the seed size distribution. In addition, within fruit seed weight pattern was also investigated.

2. Studies on seed and fruit characteristics

The fruit weight variation of 100 randomly collected pods of *Tephrosia uniflora* from their respective habitats was investigated The length of each fruit was also measured.

3. Dormancy and germination

Seeds from only one population, namely calcareous soil were examined. Ten freshly collected seeds were treated with 2 % sodium hypochlorite for 5 minutes and placed on Whatman No.1 filter paper in sterile 9 cm diameter Petri plates and the filter paper moistened with 5 ml distilled water. Petri plates were placed at room temperature. Each of the following treatments were replicated four times.

Over-night soaking

10 seeds were soaked overnight and subsequently placed on Whatman No.1 filter paper in Petri plates at room temperature.

Chemical scarification

Ten seeds were scarified by hydrochloric acid. Seeds were treated with 4N HCl for two minutes and five minutes. The seeds were then washed and placed in Petri plates on filter paper. The plates were placed on laboratory bench at room temperature.

Mechanical scarification

Seeds were scarified with the help of sand paper No.1.5. The seeds were then placed on Whatman No. 1 filter paper in sterile Petri plates kept at room temperature.

Punctured seeds

Surface sterlized seeds (2 percent sodium hypochlorite) were punctured with the help of needle and subsequently soaked over night The seeds were either placed on Whatman No.1 filter paper in sterile Petri plates or sown in pots containing loamy sand. Germination counts were made daily.

(d) Relation between seed weight and seedling length

Seeds belonging to different size classes were germinated in Petri plates and pots. Seedling lengths were measured at 12,13,14 and 15th days after sowing.

(e) Crude reproductive effort

Firstly we took fresh and dry vegetative parts *i.e*. root, stem and leaves of *Tephrosia uniflora* were weigh secondly, we took fresh and dry weight of reproductive parts (pedicels, flower, fruits and seeds) in order to determine crude reproductive effort (C.R.E.):

$$\text{C. R. E.} = \frac{\text{Dry weight of reproductive parts}}{\text{Total dry biomass}} \times 100$$

Statistical analysis

Statistical analysis was performed in accordance with Zar (1996). For seed charaecteristics, descriptive statistics were computed. Population data were subjected to one-way analysis of variance

RESULTS

(a). Phenotypic plasticity of the three populations

The results are presented in Table 1. The three populations differed considerably with regard to number of flowers, number of fruits and the number of seeds per plant..The number of fruits / plant, number of flowers / plant and number of seeds / plant were highest for rocky soil, followed by saline soil and calcareous soil.

Table 1. shows that phenotypic plasticity of *Tephrosia uniflora* in three different habitats

Soil classes	# of fruits / plant	# of flowers / plant	# of seeds / plant
Saline soil	15.4 ± 15.12	22.6 ± 20.2	112
Calcareous soil	14.8 ± 13.95	20.0 ± 21.38	91
Rocky soil	24.8 ± 24.58	28.2 ± 28.10	129

(b). Studies on seed and fruit characteristics
(i). Overall seed weight distribution

Six hundred and twentythree seeds of *Tephrosia uniflora* were studied in saline, calcareous and rocky soil. Seed weight ranged from .0140-.042g. Mode occurred at left side of the distribution. The overall mean was 0.0224g ± 0.0007.

ii) Fruit weight distribution

Hundred pods of *Tephrosia uniflora* were studied in saline, calcarious, and rocky soil. Overall fruit weight ranged from .0829-.3496 g. Mode occurred at central seed size class = .1897-.2430. Mean was .1745, standard error .00487, cofficient of variation 27.91 %, coefficient of skewness = .523 and coefficient of kurtosis = 3.567.

(iii). Fruit length distribution

Two hundred pods of *Tephrosia uniflora* were studied in on an overall basis. Fruit length ranged from 10-81 mm. Mode occurred at the central size class 38.6-52.8 mm. Mean was 43.15, standard error =1.431, coefficient of variation = 33.17 %, coefficient of skewness 0.00725 and coefficient of kurtosis = 2.921.

(iv). Seed number and seed weight variation in the three populations

Tephrosia uniflora fruits studied in saline, calcareous and rocky soil had 4,5,6,7,8,9 or10 seeds per pod. Seeds located at distal end generally, but not always, had lesser weight than the average. The average seed weight for 4,5,6,7,8,9 and 10-seeded pods differed. Grand mean of 7-seeded pods was higher (data not presented). The mean seed weight of *Tephrosia uniflora* for the saline soil population was 0.0214g ± 0.00084, for calcareous soil 0.0276g ± 0.00081 and for the rocky soil population it was .0184g± 0.00062 (Table 2).

Table 2. Mean seed weights of the three populations studied

Habitat	Mean	SD	CV%
Calcareous soil	0.0276	0.0081	29.34
Saline soil	0.0214	0.0076	35.51
Rocky soil	0.0184	0.0062	33.69

(v). Relationship between seed number and fruit length and weight

The overall relationship between seed number (SN) and fruit length (FL) is described by the following equation:

$$SN = 2.1904 + 0.1077\ FL \qquad r = 0.9104$$

The relationship between seed number (SN) and fruit weight is expressed by the following linear regression equation:

$$SN = 3.1913 + 20.847\ FW \qquad r = 0.5995$$

The seed number and fruit length relationships for the saline, calcareous and rocky soil populations are given below:

i)saline soil:

$$SN = 1.5636 + 0.1197\ FL \qquad r = 0.9803$$

ii) calcareous soil:

$$SN = 2.3042 + 0.0980\ FL \qquad r = 0.8146$$

iii) rocky soil:

$$SN = 2.1319 + 0.1091\ FL \qquad r = 0.9267$$

(c). Demographic studies

Germination behaviour: The results of seed germination study are presented in Table 3.

Table 3. Effect of various treatments on germination of *T. uniflora* seeds

General procedure	Over night soaking	Chemical scarification	Mechanical scarification	Punctured seeds
0	0	0	20	100

Table 3 shows that the seeds of *Tephrosia uniflora* by general procedure, over- night soaking and chemical scarification failed to germinate. Scarification using sand paper gave 20% germination while puncturing the seed coat resulted in 100% germination.

Table 4. Seed size classes of the three populations and their seedling lengths (cm) at various time (days) after seed sowing.

		Number of Days			
Habitat	Seed class g	12	13	14	15
Rocky Soil	.01 -.02	1.8 ± 1.6	3.0 ± 3.3	3.9	4.0 ± 2.2
Saline Soil	0.01- 0.02	1.2 ± 0.8	2.6 ± 3.0	3.2 ± 2.7	4.1 ± 3.9
	0.02- 0.03	2.2 ± 1.7	2.6 .2 ± 1.7	3.3 ± 2.2	4.9 ± 2.8
	0.03-0.04	3.1 ± 1.5	4.2 ± 1.0	5.7 ± 4.3.	6.2 ± 3.5
Calcareous Soil	0.01-0.02	1.5 ± 1.2	2.2 ± 2.1	3.2 ± 1.9	3.4 ± 2.1
	0.02 -0.03	2.5 ± 1.9	2.7 ± 1.2	4.5 ± 3.5	5.0 ± 4.2

(d) Relationship between seed weight and seedling length.

The relationship between seed size and the seedling length can be examined in Table 4. It is apparent from that in each of the population there is a direct relationship between seed size and seedling length. In all cases seedling length increased with the increase in seed size.

(e) Crude reproductive effort.

The crude reproductive effort was determined for the three populations and is given in Table 5.

Table 5. Crude reproductive efforts (C.R.E.) of *Tephrosia uniflora* in three different habitats.

Habitat	Dry vegetative biomass	Dry reproductive biomass	Total biomass	C.R.E.
Rocky soil	4.81 4.07	3.38 3.14	7.47	45.24
Saline soil	1.11 1.01	1.06 .98	2.20	48.18
Calcareous soil	5.81 5.40	2.47 2.21	8.29	29.79

Table 5 shows that in *Tephrosia uniflora* a high proportion of the total biomass was allocated to reproductive parts. Therefore, crude reproductive efforts of *Tephrosia uniflora* populations were high. The three populations differed in C.R.E. Saline soil and rocky soil populations exhibited considerably higher C.R.E compared to the calcareous soil population.

DISCUSSION

Environment plays an important role in determining patterns of seed production and therefore, fitness in natural populations. Seed is also closely linked to the sexual process that it becomes in effect the means of release of genetic variability to external environment. The total mass of seeds produced by a plant is determined by the product of several components *e.g.* the number of fruit / plant, the number of seeds / fruit and the biomass / seed. Each of these components of seed yield is subject to genetic and environmental influences that will directly determine the seed production. A high degree of phenotypic plasticity in *Tephrosia uniflora* plants was observed in the three different habitats. This is in agreement with Begon *et al*., (2003) who emphasized that yield component relationship can vary depending on the environment in which plants grow. The three populations differed in density. It has been observed that a density increase above certain levels reduces the production of seeds more that the production of vegetative parts of a plant. The reduced seed production is caused by a lowered seed number more than by a reduced seed size (Hakansson, 2003). Evolutionary botanists have demonstrated that populations in nature can actually diverge and become reproductively isolated, and they have attributed this to disruptive selection (Antonovics, 1968). However, it is difficult to presume complete reproductive isolation. On the other hand, variation in biomass allocation in *Plantago lanceolata,* which occupies a range of successional habitats, was found to be mostly environmental (Primack and Antonovics, 1981). It appears, therfore, that the difference between populations in the size of seeds produced were due more to phenotypic plasticity rather genetic difference between population experiencing different selection regime.

Fresh seeds of *Tephrosia uniflora* did not germinate well unless the seed coat was punctured or scarified which indicated that smooth, black seed coat was impervious to water and imposed an external dormancy. Several families of plants bear hard coated seeds (Baskin and Baskin, 1998) and different methods of chemical and physical scarification have been successfully used to break dormancy imposed by hard seed coat (Teketay, 1996). The impermeability of seed coat to water is most widespread in the Leguminosae (Tran and Cavanagh, 1984).

Yield components that determine seed numbers are often more variable than seed mass (Harper *et al*.,1970; Marshall, 1986). Therefore, effect of seed mass on seedling establishment need to be considered to evaluate the relative importance of seed mass and seed number in determining parental fitness. The distribution of seed weight of *Tephrosia uniflora* exhibited moderate coefficient of variability. Such variability in the seed weight has also been reported for many species by Harper *et al.* (1970).

The shoot growth of seedling is faster in case of larger seed. Such a difference in seedling growth may be a significant factor in interspecific competition (Black 1958). He discovered that the individuals derived from larger seeds could germinate successfully from greater depth and grow more rapidly than those from smaller seeds (Black, 1958; see also Zhang and Maun, 1990) and the individuals from large seeds may have an advantage early in the life cycle, particularly if seed weight effects seedling size and competitive ability (Howell, 1981; Howe and Richter, 1982; Gross, 1984) or resistance to abiotic stress (Weis, 1982).

The optimal temperature for germination of *Tephrosia uniflora* is nearly 30 ºC and seed germinate during monsoon season in the field. Studies on germination of *Tephrosia uniflora* showed that higher seed weight classes give rise to greater length of seedlings as compared to lower seed weight classes. Seedling in *Tephrosia uniflora* derived from larger seed weight classes produced greater seedling length and higher fresh and dry weights. The increased seedling length as well the fresh and dry weights of seedlings with the increase in seed size has also been reported for many other species (Tecklin and McCreary, 1991; Bonfil, 1998). This undoubtedly confers an advantage to large-sized seeds.

There is evidence that plant species differ in the proportion of their net annual assimilated income which is devoted to reproductive plant parts of the species. The crude reproductive effort of *Tephrosia uniflora* was sufficiently high as compared to many other perennial species although annuals in general allocate greater energy and resources to reproduction. The differences in crude reproductive effort between populations are presumably environmentally induced rather than of genetic origin.

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