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SUPPRESSION OF HERBS BY *INULA GRANTIOIDES* BOISS. IN THE SIND DESERT, PAKISTAN.

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Abstract

In the Sind desert, rocky soils are occupied by *Inula grantioides* dominated communities with low species diversity and high dominance. Pattern analysis disclosed that *I. grantioides* was negatively correlated with many co-occurring species. Analysis of interspecific association also revealed that a number of herbaceous species were negatively associated with *I. grantioides*. In the year following the experimental removal or clipping of *I. grantioides* the density and total dry matter production of herbs increased which suggested suppressive effects due to *I. grantioides*.

Experimental evidence is presented for the release from *I. grantioides* shoot of volatile water soluble substances which strongly inhibit the germination and seedling growth of a number of test species. The decaying shoots and roots of *Inula* were also inhibitory to germination and seedling growth of *Cenchrus pennisetiformis* and *Pennisetum americanum*. Artificial rain drip collected from *Inula* shoots arrested seedling growth of the test species. Wheat coleoptile bioassay of ether fraction of aqueous extract indicated a number of inhibitors of phenolic nature in both shoot and root extracts of *Inula*. The possible role of chemical inhibition of certain herbs in stand of *I. grantioides* is discussed.

Introduction

The phenomenon of allelopathy i.e., one higher plant exerting a detrimental influence on another through the production of germination and growth inhibiting substances has been widely reported (Martin & Rodemacher, 1960; Muller & Muller, 1964; Naqvi & Muller, 1975; Friedman *et al.*, 1975; Bokhari, 1978). The ecological significance of phytotoxic substances in oldfield succession and in other natural plant communities has attracted the attention of many workers (Muller, 1968; Grodzinsky, 1971; Whittaker & Feeny, 1971; Muller & Chou, 1972). Prominent among the chemicals (allelochemics) involved in such interactions are phenols, terpenes, glucosides, alkaloids, amino acids and sugars (Harborne, 1977; Whittaker & Feeny, 1971). The exact nature (both qualitative and quantitative) of phytotoxins depends upon the species and the environmental characteristics of the site. In many cases toxic substances are produced that inhibit other species under experimental conditions. Little information is, however, available which would adequately explain the role of one or more species in affecting the distribution and performance of others in the field.

Inula grantioides Boiss., an aromatic perennial herb, is a pre-dominant species on rocky soils in the lower Sind desert. In such xeric communities where *I. grantioides* is

dominant, the herb density and diversity is poor suggesting that *Inula* suppresses the growth and abundance of other species. This led us to examine the distribution pattern of various species of *I. grantioides* dominated communities, their interspecific association and the role of *Inula* in affecting the distribution and performance of various herbs in field as well as laboratory conditions.

Material and Methods

a) *Analysis of* Inula *dominated communities:*

The vegetation of 5 *Inula grantioides* dominated communities was sampled by 1 m^2 quardrats, 30 in each, in November, 1978. The following analytic attributes were calculated; relative frequency, relative density and relative cover. The three relative measures were combined by summation into a single importance value index (I.V.I.) following Curtis & McIntosh (1951). The species diversity of the stands was ascertained by the popular Shannon-Wiener information function $\bar{H} = \Sigma\ pi\ \log_2 pi$ (Margalef, 1957). Equitability was expressed as the ratio of observed to maximal diversity, $e = \bar{H}/\bar{H}\ max.$ (Pielou, 1969). The dominance concentration was measured using Simpeson's index (Simpson, 1949), $c = \Sigma pi^2$ where pi is the relative importance value of species i. Furthermore, dominance-diversity curves (Whittaker, 1965) were plotted for each stand to portray the relative abundance patterns.

b) *Population structure:*

The technique of 'pattern analysis' developed by Greig-Smith (1952, 1961) and Kershaw (1957, 1961) provide a means for detection of pattern and the determination of the scale and intensity of aggregation. The method allows the elucidation of interactions between species population (Brereton, 1971; Pamadasa *et al*, 1974).

The data on small scale spatial pattern were collected from two sites (arbitrarily named A and B) in the vicinity of Karachi University Campus where the soils were somewhat shallow and rocky and *I. grantioides* was the leading dominant species. Species chosen for the analysis of population structure were: *I grantioides, Blepharis sindica, Salvia santolinifolia, Hibiscus micranthus, Cencharus pennisetiformis* and *Dactyloctenium scindicum.*

Each site was systematically sampled by a grid of contiguous quardrats. Each grid was 2 x 8 m in size, consisting of 64 square grid units of side 0.5 m. Density data were collected from each grid unit for the six species listed above.

The pattern shown by the data was analysed by analysis of variance of successive block sizes (Ns) (Greig-Smith, 1961). In the graphs relating the mean square to block size the different scales of pattern appear as peaks at block size corresponding to the mean area of 'clump'.

The interaction between species populations was evaluated by correlation analysis as described by Kershaw (1961).

c) *Analysis of interspecific association:*

Interspecific associations between pairs of associates were detected by X^2-test (with Yate's correction) using 2 x 2 contingency tables for each pair of species (cf. Mueller-Dombois & Ellenberg, 1974). The quardrat data of section (a) of "material and methods" was utilised for the analysis of interspecific association.

d) *Density and biomass of annuals following removal of* **Inula grantioides**:

In order to assess the phytotoxic effects of *I. grantioides* in the field conditions, following treatments were applied in 1.5 m^2 plots in May, 1978 at sites A and B.

i) All *I. grantioides* plants were removed with most of their root system.
ii) Shoots of *Inula* were clipped.
iii) Untreated plots served as controls.

In all the treatments, seeds of *I. grantioides* lying on the soil surface in the plots were also removed. Density of annuals were recorded in October, 1978 and subsequently the shoots of annuals were harvested and dried to constant weight. Treatments and controls were replicated five times each.

e) *Phytotoxicity of aqueous extracts of* **Inula grantioides** *against some associated plants and* **I. grantioides** *itself:*

Vigorously growing plants of *I. grantioides* were collected from a community near Karachi University Campus, that was dominated by *I. grantioides*. The plants were dried at 60°C for 2 days and aqueous extracts were prepared by soaking 10 g dry material of either roots or shoots, in 400 ml distilled water for 24 hr. The filtrates obtained were taken as stock water extract (S) and from these 25%, 50% and 75% dilutions were prepared.

Toxicity was tested against the following: *H. micranthus* L. f., *C. pennisetiformis* Hochst. & Steud., *D. scindicum* Boiss., *C. holosericea* Fresen and *I. grantioides*. The response of *I. grantioides* to the extracts was tested for possible autotoxicity.

Twenty surface sterilized (2% sodium hypochlorite for 5 min.) seeds were placed on Whatman No. 1 filter paper in 9 cm. diameter sterile petri plates and 5 ml. of either shoot or root extract were added to each; controls received deionized distilled water. The seeds of *H. micranthus* were chemically scarified with 4N-HCl for 1 min. before placing them in petri plates and the extract used against *D. scindicum* contained 200 ppm KNO_3. Seeds of *H. micranthus* did not germinate without scarification and those of *D. scindicum* had very low percentage germination in the absence of KNO_3.

Treatments and controls were replicated thrice and the plates were kept in a growth chamber maintained at 30 ± 1 C° and light intensity at the top of petri plates of 4000 Lux for 14 h day length.

f) *Effect of volatile substance (s) of* **Inula grantioides** *on germination and seedling growth of* **Cenchrus pennisetiformis** *Hochst. & Steud. and* **Pennisetum americanum** *(L.) Schumann at different temperatures:*

Fifty seeds of *C. pennisetiformis* or *P. americanum* were placed on Whatman No. 1. filter paper in 15 cm. diameter glass dish in the centre of which was placed a small 6 cm. diameter dish containing 5 gm. fresh shoot material of *I. grantioides*. The filter paper was moistened with 8 ml. distilled water and the larger dish was covered with another indentical dish and the adjoint rim sealed with masking tape. The inner volume of the twin dishes was 883 ml. Controls consisted of similar set up without shoot material of *I. grantioides*. The dishes were kept in incubators maintained at 25 ± 1, 30 ± and 35 ± 1 C^{o} and germination percentage was noted at 24, 48 and 72 h of imbibition. Root and shoot lengths were measured at 72 h of growth. Treatment and controls were replicated thrice.

g) *Phytotoxicity of decaying* **Inula grantioides:**

To ascertain the phytotoxic capacity of decaying *I. grantioides*, the plant material (either shoot or root) was allowed to wither for a month in laboratory. The shoot and root were crushed separately and mixed thoroughly with sandy loam (76.1% sand, 15.3% silt and 8.6% clay) at the rate of 5, 10 or 20g root or shoot material per 400 g soil. These were kept in 8 cm. diameter plastic pots. sprinkled with some water and kept for one week to allow microbial activity. Control pots contained sandy loam only Subsequently, 10 seeds of *C. pennisetiformis* or *P. americanum* were sown in each pot. Each treatment and control was replicated four times. The pots were kept in a growth chamber, maintained at 30 ± 1 C^{o} and light intensity at the top of pots was 4000 lux for 14 h day length. Final percentage germination was noted and root and shoot lengths were measured on 10th day.

h) *Artificial rain drip and leaching of phytotoxins:*

The technique employed for this experiment was essentially that described by Naqvi & Muller (1975). Airdried leaves of *I. grantioides* were chopped into small fragments and placed in a large funnel attached to conical flask, 500 ml of deionized distilled water was then sprayed with a sprayer on the plant material and the leachate collected. The spraying was done gradually and lasted for 1 h. The leachate (or the "Artificial rain drip") was filtered and a portion of it reduced to one fourth in a rotary vacuum evaporator. Phytotoxicity of the leachate was then assayed using *C. pennisetiformis* and *P. americanum* as out-lined above (section "e" of "Material and Methods").

i) *Partial characterization of the phytotoxins:*

Ten g air dried material of either shoot or root of *I. grantioides* was crushed in

Table I. Summary of phytosociological data of *Inula* dominated stands

Species	No. of stands present	Average I.V.I.	Maximum I.V.I.	Minimum I.V.I.	No. of stands lst dominant	No. of stands 2nd dominant
Inula grantioides Boiss.	5	82.2	95.5	76.30	5	0
Aristida adscensionis L.	5	35.4	43.7	28.1	0	3
Hibiscus micranthus L.f.	5	31.6	47.1	25.7	0	1
Salvia santolinifolia Boiss.	5	26.2	32.7	22.4	0	0
Dactyloctenium scindicum Boiss.	5	28.0	36.0	21.3	0	0
Cenchrus setigerus Vahl	5	15.8	45.4	5.6	0	1
Cenchrus pennisetiformis Hochst. & Steud.	4	29.5	37.2	18.7	0	0
Chrysopogon aucheri (Boiss.) Stapf.	4	9.75	20.7	5.7	0	0
Blepharis sindica Stocks ex T. Anders.	4	29.33	37.3	14.2	0	0
Aristida mutabilis Trin. & Rupr.	3	13.33	15.3	10.0	0	0
Heliotropium strigosum Willd.	3	11.0	15.6	5.0	0	0
Cassia holosericea Fresen.	2	11.0	11.9	11.3	0	0
Fagonia indica Burm. f.	2	17.0	19.5	15.4	0	0

200 ml distilled water, the homogenate centrifuged and adjusted to pH 3 with 0.5 N H_2SO_4. The homogenate was then extracted three times with peroxidase-free ether and the pooled ether fraction evaporated to dryness over $CaCl_2$ in a dessiccator. To the dry material, 2 ml of absolute ethanol was added and streaked on Whatman No. 1 filter paper. Duplicate 10 cm wide chromatograms were developed by descending chromatography in iso-propanol: liquid ammonia: water (10:1:1 v/v/v). When the solvent had moved 30 cm from the origin, the chromatograms were taken out, dried and 10 equal width strips were cut and assayed for growth regulators using wheat coleoptile straight growth test (Nitsch & Nitsch, 1956).

After discarding the upper 3mm coleoptile segments, 5 mm segments of 3 day old dark grown wheat (*Triticum aestivum* var. Pak 70) were excised and floated in distilled water for 1 hr. Ten coleoptile segments were placed in between two strips of the same Rf value, cut from duplicate chromatograms and kept in 11.5 cm diameter Petri plates over two layers of tissue paper moistened with 4 ml 0.02 M citrate phosphate buffer (pH 4.8). After 48 h of growth in dark at 22 ± 2 C^{o} the length of coleoptile segments was measured.

Results

a) *Vegetational composition and structure of* **Inula grantioides** *dominated stands:*

Thirteen species were encountered among 5 *Inula* dominated stands. *I. grantioides* was leading dominant in all the stands with a high average importance value of 82.2 (Table 1). Besides *I. grantioides* four species, viz. *Aristida adscensionis, Salvia santolinifolia, Hibiscus micranthus, Dactyloctenium scindicum* and *Cenchrus setigerus* were present in all the five stands generally with a high importance value.

Table 2. Species diversity ($\bar{H}$), equitability (e) and dominance (c) of five *Inula grantioides* dominated stands.

Stands	$\bar{H} = \Sigma$ pi. log2pi	$e = \bar{H}/\bar{H}max.$	$C = \Sigma pi^2$
A	0.7800	0.7800	0.1436
B	0.8579	0.8990	0.1685
C	0.9879	0.9145	0.1446
D	0.9655	0.9272	0.1284
E	0.9240	0.8873	0.1443

Species diversity ($\bar{H}$) of these communities was low ranging from 0.7800 to 0.9879 (Table 2). Equitability ranged from 0.7800 to 0.9272. Dominance was substantially high ranging from 0.1284 to 0.1686. The high value of dominance indicates the predominance of *I. grantioides* in these stands.

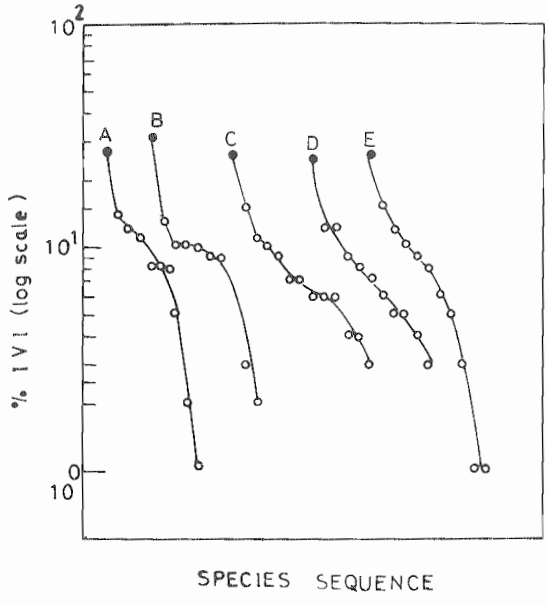

Fig.1: Dominance diversity curves for five *Inula grantioides* dominated communities.

The dominance diversity curves for all the stands except stand B follow geometric distribution (Fig. 1). Stand B seems to folow the relative abundance pattern intermediate between geometric and lognormal.

b) *Detection of pattern and inter-species correlations:*

The graphs of mean square against block size for the selected species at two different sites are given in Fig. 2. The assessment of significance of peaks depends upon the conistency with which they occur in replicated observations.

The non-random pattern of the species present at the sites examined is clearly evident from Fig. 2. Generally the scales of species pattern are consistent between sites except where "merging" and "drifting" (Greig-Smith, 1961; Kershaw, 1957) have probably occurred. Some species show two pattern scales of which the smaller one, for the sake of convenience, is referred to as 'primary pattern scale' and the larger one as 'secondary pattern scale'.

I. grantioides shows the primary pattern at Ns 2 in both the sites and the secondary pattern at NS 32 and 8 in sites A and B, respectively (Fig. 2). *S. santolinifolia* like-

wise exhibits primary pattern at Ns 2 in both the sites, while the secondary peaks appear at Ns 8 and 16 in site A and B respectively. The pattern exhibited by *H. micranthus* in the two sites in essentially similar to that of *S. santolinifolia. C. pennisetiformis* shows its primary pattern at Ns 32 in both sites and the secondary pattern at Ns 2 and 8 in sites A and B, respectively. *D. scindicum* shows the primary pattern at Ns 8 and B. *sindica* at Ns 4. The latter exhibits secondary pattern at Ns 32.

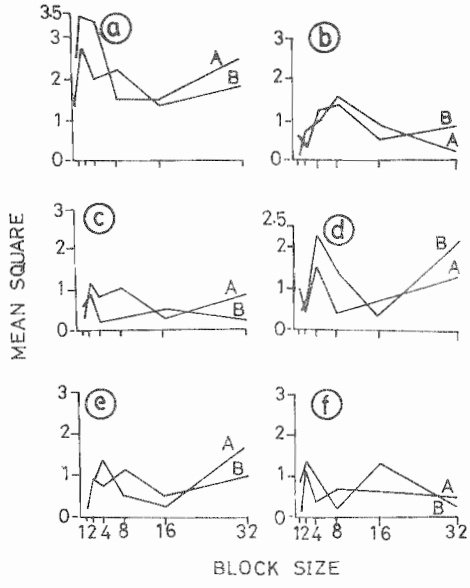

Fig.2: A representative series of mean square; block size graphs. Pattern of *I. grantioides* and its five associates viz. a, *I. grantioides;* b, *Dactyloctenium scindicum;* c, *Salvia santolinifolia;* d, *Blepharis sindica* e, *Cenchrus pennisetiformis,* and f, *Hisiscus micranthus* at two different sites A and B.

Some indications about the inter-species correlations may be obtained from mean square/block size analysis, but more complete information is usually provided by correlation analysis. Interaction of *I. grantioides* with five species as elucidated by pattern correlation analysis is depicted in Fig. 3. The block size at which the correlation peaks occur are generally comparable to those found in the variance analysis. *S. santolinifolia* shows a negative correlation with *I. grantioides* at Ns 2 in site A and at Ns 4 in site B. *H. micranthus* is negatively associated with *I. grantioides* at Ns 2 and 8 in site A and at Ns 2 in Site B. Similar negative association is exhibited by *C. pennisetiformis* with *I. grantioides* at smaller block sizes, viz. at Ns 2 in site B and at Ns 1 and 8 in site A. However, the two species show a tendency of positive association at Ns 16 in site B. Likewise, *D. scindicum* also shows negative correlation with *Inula* at Ns 4 in site A and at Ns 2 and 8 in site B, but are positively associated at Ns 16 and Ns 32 in site A and B, respectively. In contrast, *B. sindica* exhibits a positive association with *I. grantioides* at Ns 4 and 16 in site B and at Ns 8 in site A.

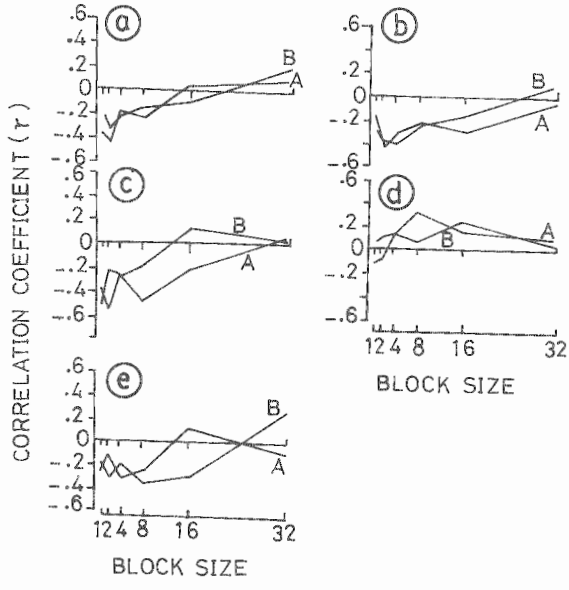

Fig.3.A representative series of correlation coefficients : block size graphs. Correlation pattern between *Inula* and its five associates. a, *I. grantioides* vs. *H. micranthus;* b, *I. granatioides* vs. *S. santolinifolia;* c, *I. grantioides* vs. *C. pennisetiformis;* d, *I. grantioides* vs. *B. sindica;* and *I. grantioides* vs. *D. scindicum* at two sites A and B.

c) *Inter-specific associations:*

The inter-specific associations among the constituent species of the stands where *I. grantioides* was the leading dominant species as analysed by X^2 test (with Yate's correction), are recorded in Fig. 4. *I. grantioides* exhibited negative associations with *S. santolinifolia, H. micranthus, Aristida mutabilis* and *D. scindicum* but was positively associated with *B. sindica*. Apart from associations involving *I. grantioides,* twelve positive and seven negative associations between pairs of species were recorded, whereas rest of the combinations were non-significant.

d) *Density and yield of annuals folowing removal of* **Inula grantioides:**

The complete removel of *I. grantioides* significantly increased the density as well as the biomass ($p < 0.01$ for site A and $p < 0.05$ for site B) of annuals at both the sites of study. Clipping of the shoot of *Inula* also resulted in significant increase of density ($p < 0.05$) and dry matter production of annuals ($p < 0.05$). However, biomass levels of annuals did not differ significantly in plots where *I. grantioides* was uprooted than those where the shoots were clipped but the density of annuals was significantly higher in plots where *I. grantioides* was uprooted (Table 3).

Table 3. The density and dry matter yields of annual plants in variously treated 1.5 m^2 plots: October 1978 (mean of five plots ± standard error).

Treatments	Site A		Site B	
	No. of annual plants/m^2	Total dry matter (g/m^2)	No. of annual plants/m^2	Total dry matter (g/m^2)
Control	9.71±1.13	4.16±1.71	8.23±2.33	3.85±1.46
I. grantioides uprooted	17.65±3.12	8.04±1.44	13.08±1.82	6.27±0.86
I. grantioides clipped	14.48±1.86	7.63±2.29	18.17±2.58	7.04±1.90

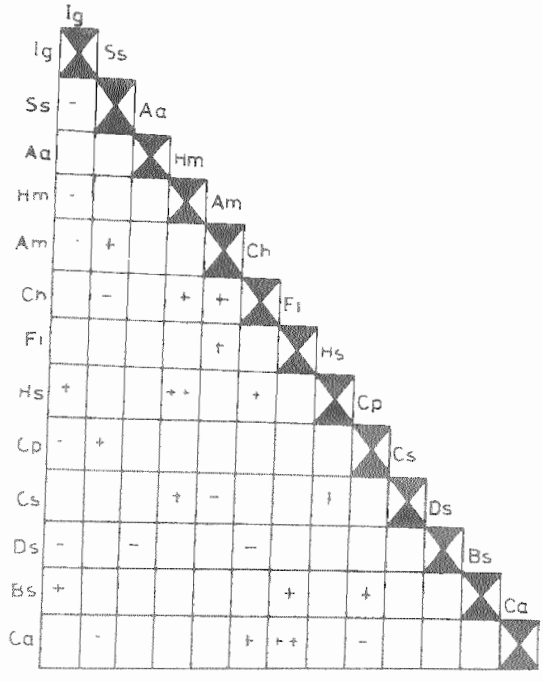

Fig.4 Interspecific association among the species encountered in the sampling of *Inula* dominated communities, as detected by X2-analysis (with Yates, correction). Key to the species: Ig, *I. grantioides*. Ss, *S. santolinifolia:* Aa, *A. adscensionis*. Hm, *Hibiscus micranthus;* Am, *A. murabilis*. Ch, *Cassia holosericea;* Fl, *Fagonia indica;* Hs, *Heliotropium strigossum;* Cp, *C. penniseriformis*. Cs, *C. setigerus;* Ds, *D. scindicum*. Bs, *B. sindica;* Ca, *C. aucheri*.

e) *Phytotoxicity of aqueous extracts of* **Inula grantioides.**

i) *Effect on germination:* The germination percentage of both the grasses, viz. *C. pennisetiformis* and *D. scindicum* was drastically suppressed by the shoot extract and to a somewhat lesser degree by the root extract of *I. grantioides* (Fig. 5). The inhibitory effect increased with increasing concentration of the extracts. Shoots extract of *I. grantioides* impeded the germination of *H. micranthus* at 50% S but suppressed it at 75% and 100% S. Whereas in the root extract the germination of *H. mucranthus* was delayed in 75% S and was inhibited in 100% S. The germination of *C. holosericea* seeds was strongly inhibited by the shoot extract of *Inula* at 50% to 100% S but was delayed by the root extract in 50% and 75% S and suppressed at 100% S.

ii) *Effect on seedling growth:* The shoot extract of *I. grantioides* significantly stimulated the shoot growth of *C. pennisetiformis* at 25% S but significantly inhibited it at 75% and 100% S ($p < 0.01$ at 75% S and $p < 0.001$ at 100% S) (Fig. 6a), whereas root growth was significantly suppressed at 75% and 100% S ($p < 0.001$). Root extract of

Inula significantly retarded the shoot growth of *C. pennisetiformis* only at 100% S but the root growth was depressed at 75% and 100% S ($p < 0.05$) (Fig. 6b). Both root and shoot growth of *D. scindicum* was significantly inhibited by the shoot extract of *I. grantioides* (p at least < 0.05) at 75% and 100% S (p at least < 0.05) (Fig. 6a). Likewise the root extract of *Inula* also suppressed the roots and shoot growth of *D. scindicum* at 75% and 100% S (p at least < 0.05) (fig. 6b).

Root growth of *H. micranthus* was retarded by the shoot extract of *I. grantioides* only at 100% S ($p < 0.001$) but shoot elongation was suppressed by the extract at 75% S and 100% S solution (p at least < 0.05) (Fig. 6a). On the other hand, root extract of *Inula* inhibited both root and shoot growth of *H. micranthus* only at 100% S solution ($P < 0.05$) (Fig. 6b).

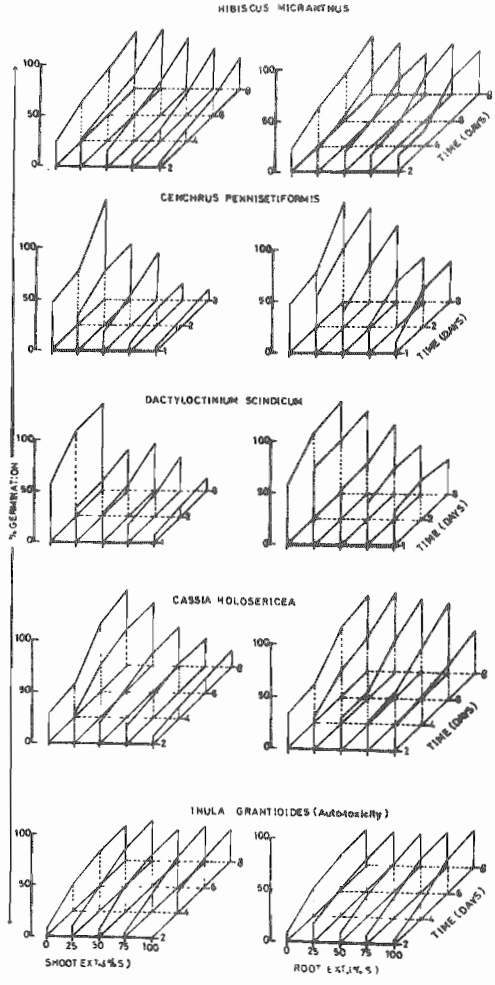

Fig.5: Effect of aqueous extracts of *Inula* on the germination of seeds of *H. micranthus*, C. *pennisetiformis*, *D. scindicum*, C. *holosericea* and *I. grantioides* itself (autotoxicity).

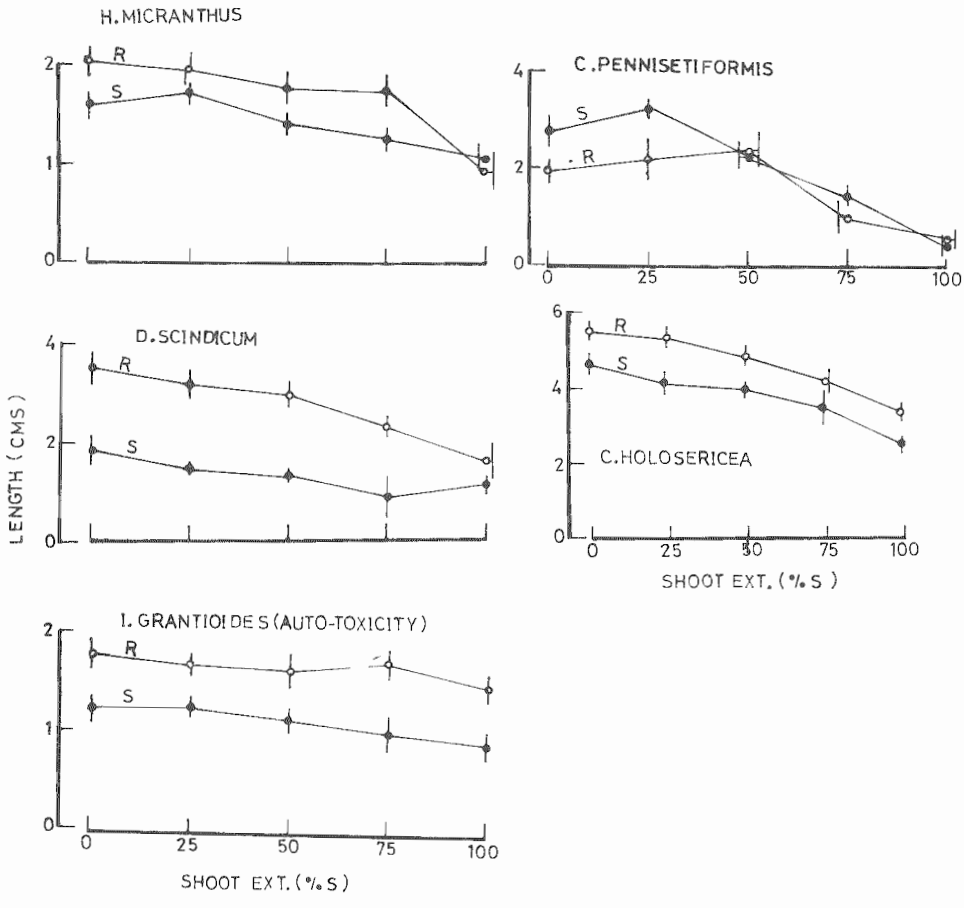

Fig.6a: Effect of aqueous shoot extract of *Inula* on the seedling growth of *H. micranthus*, *C. pennisetiformis*, *D. scindicum*, *C. holosericea*, and *I. grantioides* itself (autotoxicity). Vertical bar at the mean indicates the standard error of the mean.

The shoot extract of *I. grantioides* significantly retarded the root growth of *C. holosericea* only in 100% S solution ($p < 0.001$) but shoot growth was suppressed by this extract at 75% and 100% S (p at least < 0.05) (Fig. 6a). In contrast to the shoot extract, the root extract of *Inula* significantly promoted the root and shoot elongation of *C. holosericea* at 25% S but significantly retarded the root growth at 100% S ($p < 0.05$) (Fig. 6b). The study of the influence of shoot and root extracts of *I. grantioides* on the seedling growth of *Inula* itself revealed that the growth suppression was very little (i.e., there was very little autotoxicity) and only the shoot extract slightly but significantly ($p < 0.05$) inhibited the root and shoot growth at 100% S solution (Figs. 6a and b).

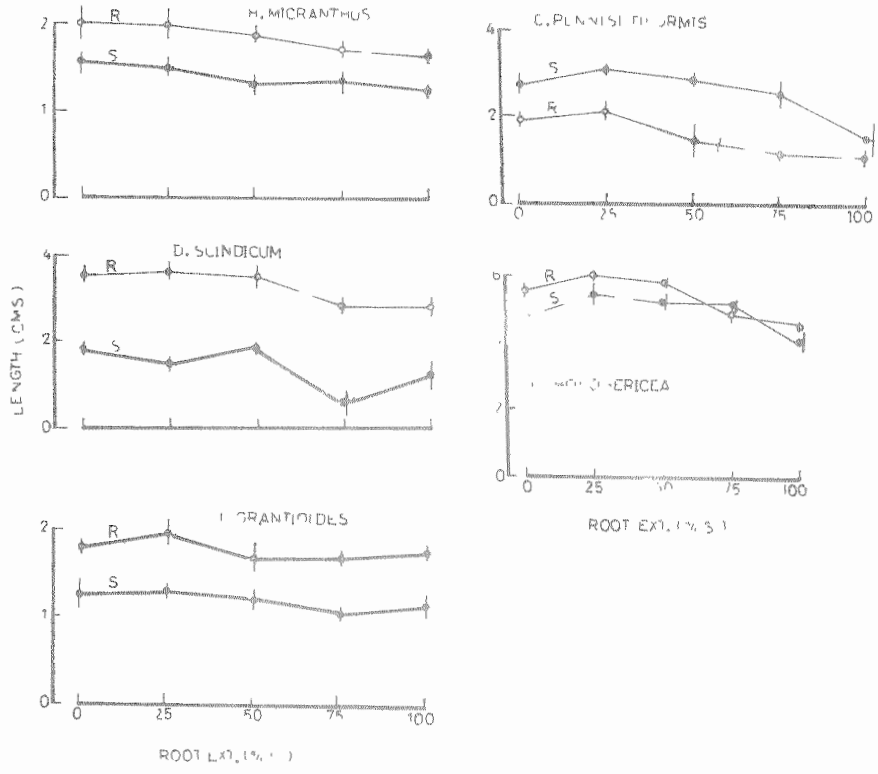

Fig.6b. Effect of aqueous root extract of *Inula* on the seedling growth of *H. micranthus*, C. *pennisetiformis*, *D. scindicum*. *C. holosericeo* and *I. grantioides* itself (autotoxicity). Vertical bar at the mean indicates the standard error of the mean.

f) *Effect of* ***Inula*** *shoots at different temperatures on germination and seedling growth of* ***Pennisetum americanum*** *and* ***Cenchrus pennisetiformis*** *(volatile method).*

In general germination percentage of *P. americanum* increased sharply after 24 h. At all the temperatures the germination percentage was impeded and final percentage was significantly reduced in the treatments (with 5 g *Inula* shoots) over the controls (Fig. 7). The inhibitory effect of *Inula* shoots on germination increased with the increment of temperature. Root lenght of *P. americanum* was significantly reduced in the treatments over the controls at all temperatures (Fig. 8). In contrast, the shoot growth of *P. americanum* was inhibited only at 25 and 30°C.

Germination rate of *C. pennisetiformis* was impeded and the final percentage was significantly reduced by *Inula* shoots Fig. 7). The inhibitory effect was more pronounced

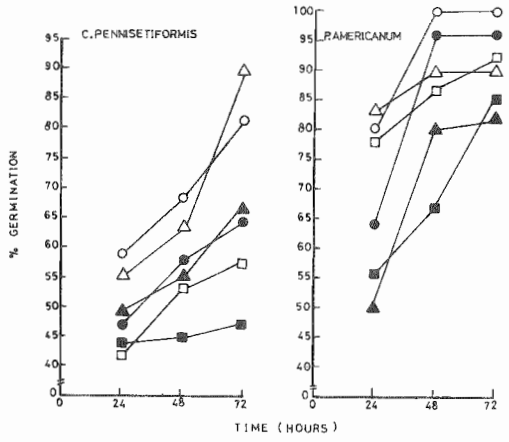

Fig.7: Effect of volatiles substances of *Inula* on germination of *C. pennisetiformis* and *P. americanum* at different temperatures.
□, 25 C; ○, 30 C; Δ, 35 C. Hollow symbols indicates controls and the solids the treatments.

at higher temperatures, viz. at 30 and 35°C. Both root and shoot growth of *C. pennisetiformis* were significantly retarded at all the temperatures and inhibitory effect clearly increased with increase of temperature (Fig. 8).

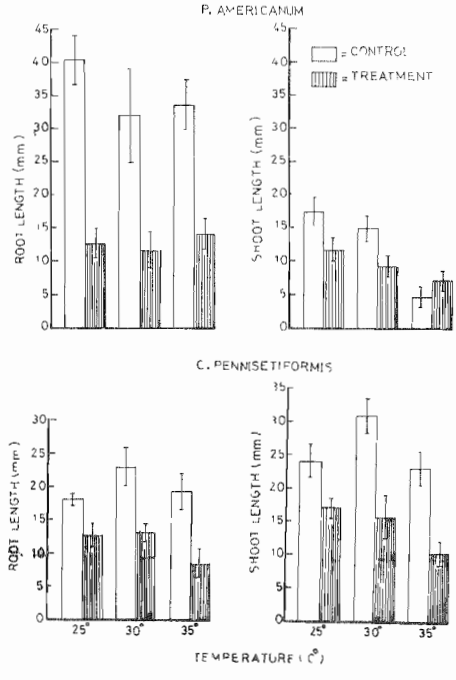

Fig.8: Effect of volatile substance (s) on the seedling growth of *C. pennisetiformis*, and *P. americanum* at different temperatures. Vertical bar at the mean indicates the standard error of the mean.

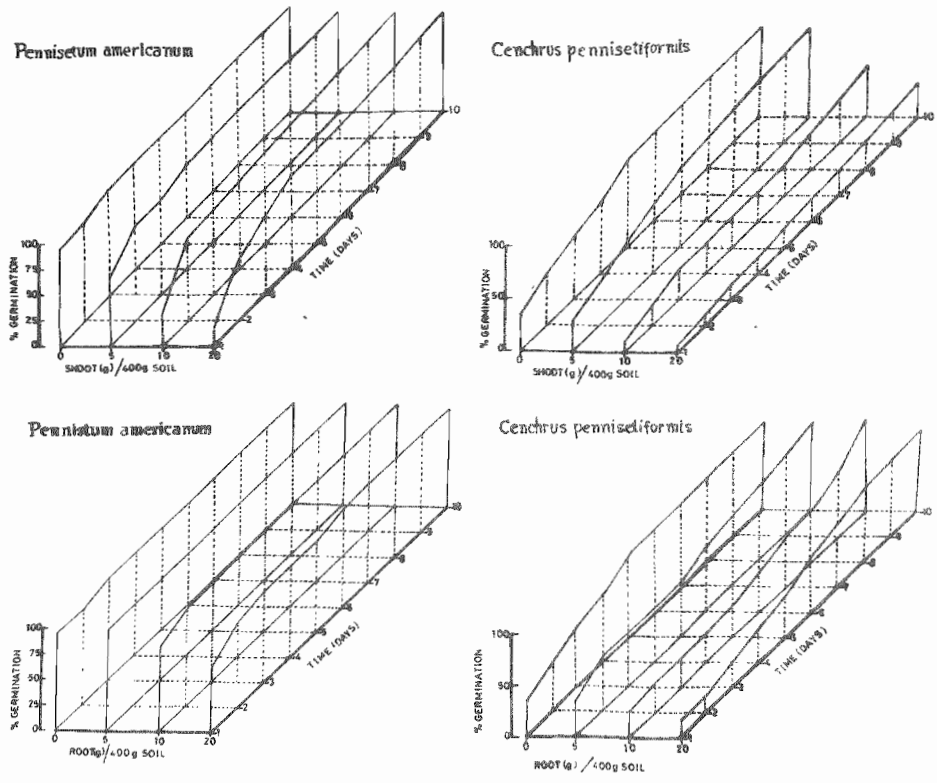

Fig.9: Germination of *P. americanum* and *C. pennisetiformis* in soils incorporated with decaying root or shoot materials of *Inula grantioides.*

g) ***Phytotoxic effects of decaying Inula grantioides on germination and growth of C. pennisetiformis and Pennisetum americanum:***

a) *Cenchrus pennisetiformis:* The germination percentage of *C. pennisetiformis* was significantly reduced ($p < 0.01$) in soils incorporated with 10 and 20 g of decaying *Inula* shoots. Decaying roots, however, did not alter the final germination percentage, though the rate of germination was retarded in soils containing 10 and 20 g root material (Fig. 9). Both root and shoot growth and the dry weights of *C. pennisetiformis* were reduced by the decaying shoots of *Inula* (Fig. 10). Suppressive effects of decaying *Inula* roots on *C. pennisetiformis* were rather weak. Root and shoot growth as well as the respective dry weights of *C. pennisetiformis* were markedly suppressed only at 20 g root material (Fig. 10).

b) *Pennisetum americanum:* The germination percentage of *P. americanum* was significantly depressed by the decaying *Inula* shoots ($p < 0.05$ at 20 g) (Fig. 11) but decaying roots of *Inula* decreased only the rate of germination leaving the final

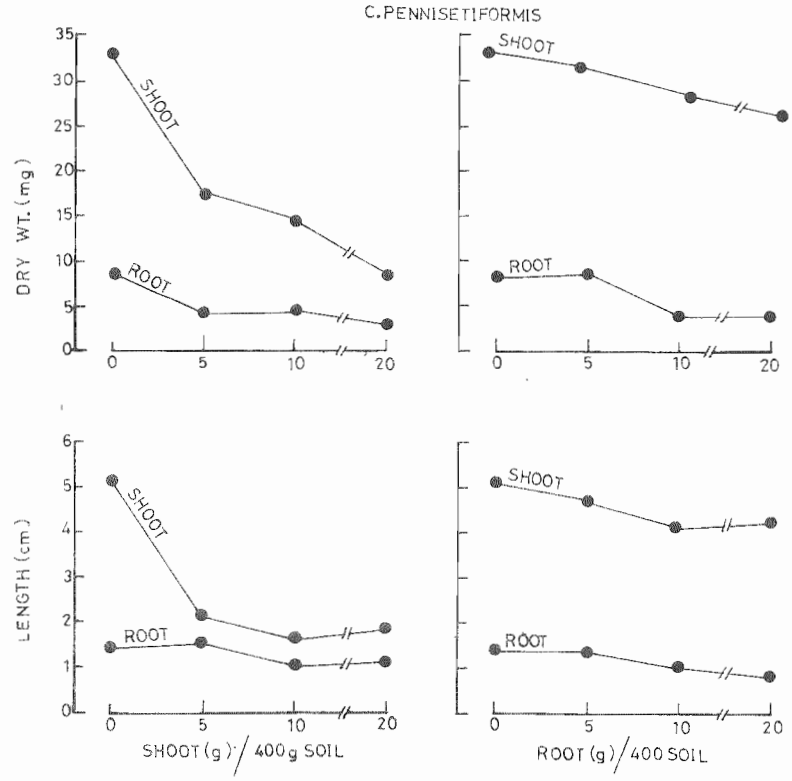

Fig.10: Seedling growth of *C. pennisetiformis* in soils incoporated with decaying root or shoot materials of *Inula grantioides.*

percentage unaffecged. Shoot growth and its dry weight was significantly ($p < 0.01$) reduced by the decaying *Inula* shoots but root length remained unaffected, though the root dry weights significantly decreased ($p < 0.05$). Decaying *Inula* roots (10 and 20g/ 400g soil) suppressed the shoot elongation and shoot dry weight of *P. americanum,* though the root elongation remained almost unchanged by the decaying roots yet the root dry weights were substantially reduced (Fig. 11).

h) *Effect of 'artificial rain drip' on germination and seeding growth of* ***Cenchrus pennisetiformis*** and ***Pennisetum americanum:***

The original (1x) concentration of the leachate did not significantly alter the germination percentage or shoot growth of both test species (Table 4). However, 1x concentration signtificantly inhibited the root growth of *C. pennisetiformis* ($p < 0.001$) and that of *P. americanum* ($p < 0.05$). The 4x concentration of 'artificial rain drip' significantly inhibited shoot growth of both the test species ($P < 0.05$). However, this concentration significantly inhibited root growth only in case of *P. americanum* ($P < 0.05$).

Table 4. Effect of artificial rain drip on germination and early seedling growth of *Cenchrus pennisetiformis and Pennisetum americanum.*

Concentration	*Cenchrus pennisetiformis*			*Pennisetum americanum*		
	% germination	Shoot length cm	Root length cm	% germination	Shoot length cm	Root length cm
Control	85.0 ± 2.88	3.16 ± 0.21	192 ± 0.23	91.66 ± 1.66	1.68 ± 0.19	3.37 ± 0.26
1X	80.0 ± 5.00 n.s.	2.93 ± 0.17 n.s.	1.47* ± 0.10	88.33 ± 1.66 n.s.	1.55 ± 0.27 n.s.	2.32 ± 0.28 n.s.
4X	53.33** ± 4.40	2.44* ± 0.15	1.37 ± 0.20 n.s.	48.33** ± 7.26	1.08* ± 0.07	2.31* ± 0.24

*P <0.05, ** p<0.01, n.s, non-significant

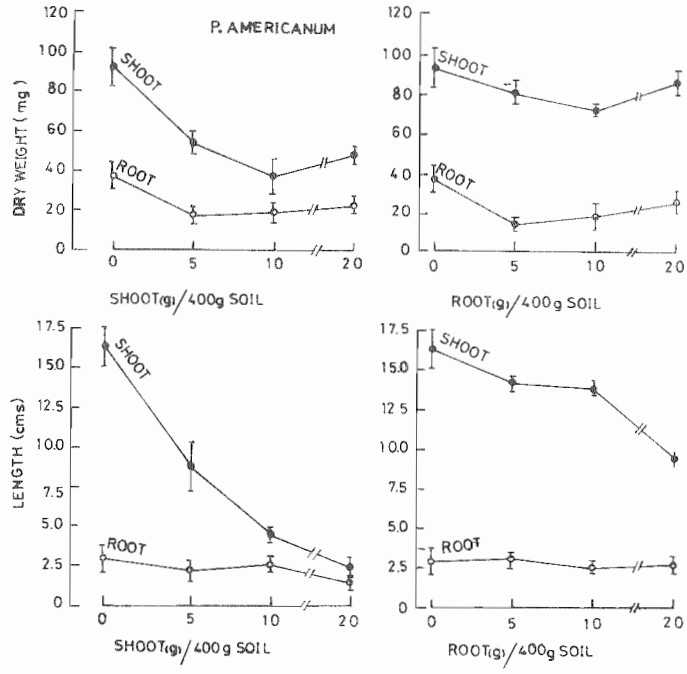

Fig.11: Seedling growth of *P. americanum* in soils incorporated with decaying root or shoot materials of *Inula grantioides*.

i) *Wheat coleoptile bioassay:*

The bioassay of the ether fraction of a queous extract of shoot of *I. grantioides* (Fig. 12) disclosed significant amounts of growth inhibitor at Rf values of 0.1–0.2, 0.2 - 0.3, 0.5 – 0.6, 0.6 - 0.7, 0.7 – 0.8, 0.8 – 0.9 and 0.9 – 1.0. The assay also revealed the presence of growth promoters at Rf value 0.3 – 0.4 and 0.4 – 0.5 in the shoot extracct. Root extract of *I. grantioides* had qualitatively and quantitatively lesser inhibitior than the shoot extract. Inhibitors in the root extract were found at Rf-values of 0.1 – 0.2, 0.2 - 0.3, 0.5 – 0.6, 0.7 – 0.8 and 0.8 – 0.9 (Fig. 12). As in the shoot extract, the growth promoters in the root extract appeared at Rf value of 0.3 – 0.4 and 0.4 – 0.5.

Discussion

The importance of chemical interactions between higher plants is under-rated by ecologists largely because of the paucity of information relating to the allelopathy in natural plant communities in field conditions. In this paper an attempt is made to provide the data to bridge this gap. An examination of dispersion pattern of *I. grantiodies* and its associates revealed that most of the primary peaks appear at block size 2 and 4 indi-

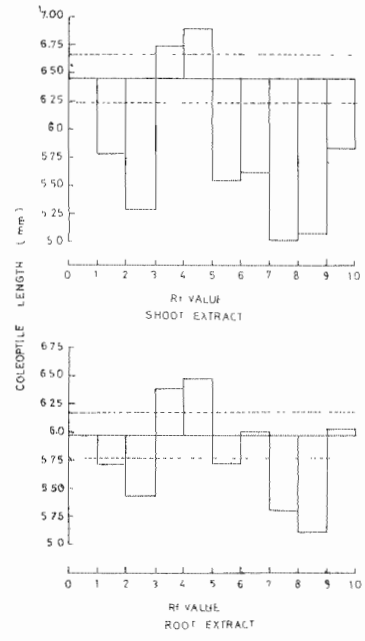

Fig.12: Wheat-coleoptile bioassay histograms of ether fractions of shoot and root extract of *Inula grantioides* chromatographed on Whatman No. 1 filter paper and developed in solvent; isopropanol: Ammonia: Water (10:1:1, V/V/V). Dotted lines represent 95% confidence interval.

cating that the mean linear dimension of the mosaic unit at this scale generally ranges from 1 to 2 meters. None of the species chosen for pattern analysis possess any special mechanism for seed dispersal except *I. grantioides.* The most probable dispersal agency is wind. However, all the herbs are very small plants ($<$ 20 cm in height) and so long distance dispersal ($>$ 40 cm) of more than a very small proportion of seeds by wind is rather unlikely. Even the seeds of *I. grantioides* were seen to be lying below the parent plants. Field observations on other peecies (herbs) also reveal that most seeds remain in the immediate vicinity of the parents. Thus it seems reasonable to suppose that dispersion of seeds in these species is limited to small distance ($\leqslant$ lm). The frequent emergence of their seedlings in clusters around dead parent also supports this. The later phenomenon is particularly very consistent for *I. grantioides.* These considerations suggest that aggregation of individuals at blocksize 2 (corresponding to linear dimension of 1 m) is to a great extent the result of the effect of the limited distance of seed dispersal. The observed primary pattern is. therefore, presumably and predominantly of reproductive origin. Similar interpretation of the small scale pattern has been given by Brook (1969) and Pamadasa *et al* (1974).

The negative correlation between *I. grantioides* and most of its associates, viz.

S. santolinifolia, H. micranthus, C. pennisetiformis and *D. scindicum* at block size 2 indicates that patches (1m in length) of *I. grantioides* and those of its associates alternate with each other. This provides strong evidence of inter-species interference operating between *Inula* and other herbaceous species in the field.

The suppressive effect of *I. grantioides* on other herbs occurring in communities where *I. grantioides* is leading dominant, is also elucidated by the negative interspecific association between *I. grantioides* and other herbs such as *C. pennisetiformis, D. scindicum, H. micranthus, S. santolinifolia* and *A. mutabilis.*

These evidences on the inhibitory influence of *I. grantioides* on other herbaceous species in the natural communities are also corroborated by experiment which evaluated the effect of complete removal or clipping of *I. grantioides* on the density and biomass of herbs. Both density as well as total dry matter production of herbs following removal or clipping of *I. grantioides* increased significantly over the controls. This would suggest that the aromatic plants of *I. grantioides* suppress the germination, establishment and growth of other species in their immediate vicinity, though the possibility of competitive elimination of herbs by *I. grantioides* can not be totally ruled out.

The examination of the influence of the aqueous extracts of root and shoot of *I. grantioides* on germination of a range of associated herbs disclosed that the shoot extract was much more inhibitory to the process of germination in comparison to the root extract indicating that shoot contains greater amounts/Numbers of inhibitors than does the root. The suppression of germination of various species by the shoot extract was in the order: *C. pennisetiformis, D. scindicum, C. holosericea, H. micranthus.* The root extract of *I. grantioides* which was less suppressive than the shoot extract, also inhibited the germination of the above mentioned species in the same order. The aqueous extracts of many species are known to inhibit seed germination (Le Tourneau *et al,* 1956; Ohman & Kommedahl, 1960; Singh, 1968; Datta & Sinha-Roy, 1975; Naqvi & Muller, 1975; Friedman *et al,* 1977). The marked inhibitory effect of the shoot extract on germination was presumbaly due to phenolic compounds and certain volatile substances. Phenolic compounds in the shoot extract were demonstrated in the bioassay of ether fraction of aqueous extract whereas the presence of the volatile substances was disclosed in the experiments where germination and growth inhibition was found in response to the exposure of seeds to fresh shoot of *Inula.* Inhibition of germination by phenolic compounds has ben reported (Massart, 1957; Evenari, 1961; Naqvi, 1976). Germination of seeds of *I. grantioides* in the shoot and root extracts of *I. grantioides* remained more or less unaffected.

The shoot extract of *I. grantioides* was more phytotoxic to seedling development than was the root extract which even promoted seedling development, particularly the root growth in some instances at low concentrations. The root growth of various species

was retarded by the shoot extract of *Inula* in the order: *C. pennisetiformis, D. scindicum, H. micranthus, C. holosericea* whereas the shoot growth was inhibited by this extract in the order: *C. pennisetiformis, D. scindicum, H. micranthus, C. holosericea.* On the other hand, the root extract of *Inula* suppressed the root growth of various species in the order: *D. scindicum, C. pennisetiformis, C. holosericea, H. micranthus.* The shoot growth of these species was inhibited by root extract of *Inula* in the same order. Only the shoot extract of *Inula* (100% S solution) slightly inhibited the root and shoot growth of *I. grantioides* indicating that the autotoxicity was negligible. The effect of phytotoxins released from *Inula* shoot and root on germination and seedling growth is evidently fairly specific as not all species were equally susceptible to the extracts. The species specificity of phytotoxins has also been demonstrated for the extracts of *Croton bonplandiamum* Baill., (Datta & Sinha-Roy, 1975), *Lolium multiflorum* Lamb (Naqvi & Muller, 1975) and *Artemisia herba-alba* Asso., (Friedman et al, 1977). The phenomenon is undoubtedy due to inherent differences in physiological and to a certain extent morphological characteristics of the various species involved.

The decaying shoots and roots of *I. grantioides* were also found to be pernicious to both germination and seedling growth of *C. pennisetiformis* and *P. americanum* but the shoot material was more detrimental than the decaying root as was observed with *Inula* extracts. Le Tourneau & Heggenes (1957) found no evidence of phytotoxicity even after incorporation of 20 g material of various spurges in soil. Wilson & Rice (1968), however, reported both stimulatory and inhibitory effects with as little amounts as 1 g of decaying sunflower leaves in 454 gm soil (2/3 garden loam soil + 16/3 sand). Nevertheless, Datta & Sinha-Roy (1975) obtained significant reduction in percentage germination with 5 and 10 g decaying *Croton bonplandianum* leaves per 250 g soil in 13 out of 15 species tested. In such experiments the texture of soil being used is of considerable importance as the phytotoxins are more effective in coarse-textured soils than in fine-textured soils where they get irreversibly adsorbed on colloidal particles that form high proportion in such soils. The importance of phytotoxic substances as edaphic variable is greater in desert regions where the process of leaching is restricted owing to scanty rainfall. In desert plants, considerable competition for limited water available in the soil would result in the development of many competitive effect including allelopathy as a mechanism for survival under these harsh conditions (Muller & Chou, 1972; Harborne, 1977). The evidence provided above strongly suggests that *I. grantioides* inhibiting the growth of annuals, it ensures for itself the available moisture in the vicinity of its sites of growth.

The presence of toxic residues from various decaying plants (that contain phytotoxins) in soil plays a pre-eminent role in determining the community structure, composition and dynamics (Abdul – Wahab & Rice, 1967; Parenti & Rice, 1969; Muller & Chou, 1972; Harbonre, 1977). All the communities in which *I. grantioides* was a leading dominant had low diversity and high dominance. The relative aboundance pattern of

these communities, as evaluated by importance value curves, was essentially linear on the semi-log plot indicating geometric distribution. Such a distribution is attributed by Whittaker, (1975) to species poor, environmentally extreme situation, e.g., desert plant communities.

The germination and seedling development of *C. pennisetiformis* and *P. americanum* in closed dishes, containing *Inula* shoot material, was drastically inhibited suggesting that the aromatic shoots of *the* aromatic shoots of *Inula* release volatile inhibitory substance (s). The fact that aqueous extracts of *Inula* shoots caused inhibition effects similar to those produced by *Inula* shoots in the close dishes indicate that the later are not merely the result of a specific change in composition of the atmosphere in the sealed dishes, but attributable to the presence of living shoots of *Inula.*

The inhibitory nature of 'artifical rain drip' suggests that the phytotoxins present in *I. grantioides* are highly water soluble and under natural conditions it is very likely that these may be washed out from the leaves by rain, fog or dew into the soil where they would exert inhibitory effects on germination and growth of neighbouring plants.

Wheat coleoptile straight growth bioassays of ether fraction of aqueous extracts of *Inula* revealed the presence of seven phenolic inhibitors in the shoot and five in the root. The volatile toxic principle (s) which inhibit (s) germination and seedling growth is/are presumably (an) aromatic compound (s) whose identifcation was not attempted. Nevertheless, it is possible that the volatile component(s) may be terpenoid or phenolic in nature. Hegnauer (1977) has repored many 'acetophenones' in several composite semi-shrubs. *Inula* being a composite may probably contain such compounds(s).

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