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Allelopathy effect of aqueous extract of *Clerodendrum viscosum*, *Ageratum conyzoides* and *Parthenium hysterophorus* on the seed germination and seedling vigour of Chickpea seeds (*Cicer arietinum L.*) in vitro

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Abstract: In present study the allelopathic effect of aqueous leaf extracts of *Clerodendrum viscosum*, *Ageratum conyzoides* and *Parthenium hysterophorus* was observed on the seed germination of Chickpea *in vitro* (*Cicer arietinum*.L.).Seed germination was found to be inhibited in all the leaf extract concentrations used i.e. 2%,4%,6%,8% and 10%. However higher inhibition was observed in the 10% leaf extract of the test plants (i.e. *Cl. viscosum*, *Ag. conyzoides* and *P. hysterophorus*). In compare to all allelopathic plants extracts more inhibition was found in *P. hysterophorus* leaf extract followed by *Cl. viscosum* and *Ag. conyzoides*. Higher inhibition of radical and plumule growth of the Chickpea seeds was found with the increase in concentration of the leaf extracts used. Percentage of germination of Chickpea seeds was found to be low in 10% concentration whereas in untreated seeds it was found to be more.

Keywords: Ageratum conyzoides, Allelopathy, Cicer arietinum, Clerodendrum viscosum, Parthenium hysterophorus.

INTRODUCTION

Allelochemicals which inhibit the growth of some species at certain concentrations may stimulate the growth of the same or different species at a lower concentration.Allelopathies are generally associated with the interactions between living plants and have been observed in agricultural lands. Some crops exert allelopathic effects on other crop and weeds. They may inhibit (negative effect) or stimulate (positive effect) the germination and growth of weeds in agro ecosystems. Weeds are the plants which grow where they are not wanted and they interfere with seed germination, growth, biomass production and yield of the cultivated crops. Weeds are an important factor in the management of all land and water resources but their effect is greatest on agriculture (Rao, 1992 and Rao, 2000).

Keeping the above in view the present work was made on the allelopathic potential of some locally available weed species (*Cl. viscosum, Ag. conyzoides* and *P. hysterophorus*) on the germination and growth of seeds of the economically important crop plants (*i.e. Cicer arietinum* L.).

MATERIALS AND METHODS

Collection and screening of plant material: Fresh leaves of *Clerodendrum viscosum, Ageratum conyzoides* and *Parthenium hysterophorus* in its vegetative stage were

collected from near the agricultural fields around the Assam University Campus.

From the preliminary screening it was observed that leaf extract had the strongest allelopathic effect on the seed germination; thus leaf was selected for detailed experimentation. Ten gram of fresh leaves of allelopathic plants (i.e. Cl. viscosum. Ag. conyzoides and P. hysterophorus) was ground, mixed with 100ml distilled water, aqueous extract was obtained as filtrate of the mixture and final volume was adjusted to 100ml; this gave 10% aqueous extract. The extract was considered as stock solution and a series of dilutions with (Sodium hypochlorite for 15 min) of the selected seeds of C. arietinum.L) were kept for germination in sterilized Petridishes lined with double blotting paper and moistened with 10ml of different concentrations of the aqueous leaf extracts (2 to 10%). Each treatment had three replicates (total no of test seeds; 10x3=30). One treatment was run as control and treated with distilled water only. The Petridishes were maintained under the laboratory conditions (room temperature 25°C at mid day, and diffused light during the day) for one week. Equal volume of distilled water was added in the experimental Petri-dishes when moisture content of the blotting paper declined. After one week, percentage of germinated seeds was counted and the radical and the plumule length were measured with the help of a slide caliper and the; observations were made five times, in two days interval. At the end, all

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the radical and plumule were cut separately and oven dried at 70°C or 48hr to get dry biomass of the same; total seedling biomass was calculated as the sum of biomass of radical and plumule.

Significance of the difference in radical and plumule growth of seedlings under different treatments was tested and compared using analysis of variance (ANOVA) and critical difference at 1% and 5% were also tested.

RESULTS

Seeds germination: In aqueous leaf extracts of *Cl. viscosum, Ag. conyzoides* and *P. hysterophorus* treated seeds of *C. arietinum*.L, germination was less in all the *P. hysterophorus* extracts in comparison to the extracts of *Cl. viscosum* and *Ag. conyzoides*. (Table 1).

Seedling growth: In *Cl. viscosum* leaf extract treatment maximum inhibition was found in 10% concentration i.e. 2.54 cm whereas in control it was found to be 12.60 cm

(Table 2). In *Ag. conyzoides* leaf extract treatment maximum inhibition was found in 10% concentration i.e. 5.98 cm whereas in control it was found to be 9.29 cm (Table 3). In *P. hysterophorus* leaf extract treatment maximum inhibition was found in 10% concentration i.e., no germination whereas in control it was found to be 9.20 cm (Table 4).

Biomass of seedling of radical and plumule of *Cicer* arietinum L., for *Cl. viscosum*, *Ag. conyzoides* and *P. hysterophorus* leaf extract at different concentrations are less energy effecients as compared to control, shown in Table 5.

DISCUSSION

From the present work it was observed that leaf extracts of the three allelopathic test plants (i.e.*Cl. viscosum, Ag. conyzoides* and *P. hysterophorus*) inhibited the seed germination of *C. arietinum* L. *in vitro.* Tefera (2002)

Table 1. Percentage of germination of chickpea seeds treated with the aqueous leaf extract of *Cl. viscosum*, *Ag. conyzoides* and *P. hysterophorus*.

Percentage of germination (%)			
	Cl. Viscosum	Ag. conyzoides	P. hysterophorus
		C. arietinum L.	
Control/Plant extract	83.3	93.6	93.3
2% extract	73.3	83.3	83.3
4% extract	66.6	76.6	73.3
6% extract	60	60.6	53.3
8% extract	60	53.3	43.3
10% extract	50.6	43.3	0

Table 2. Effect of the aqueous leaf extract of Cl. viscosum against the chickpea seed.

No .of days/ treatment	3 rd day	6 th day	9 th day	12 th day	15 th day
		Radi	cle (cm)		
Control	6.48 (±0.86)	11.19 (±0.47)	11.99 (±0.72)	12.03 (±0.42)	12.60 (±0.37)
2% extract	5.25 (±0.79)	9.28 (±0.43)	11.16 (±0.51)	11.42 (±0.45)	11.43 (±0.37)
4% extract	5.05 (±0.89)	8.43 (±0.30)	8.44 (±0.46)	8.78 (±0.57)	8.81 (±0.51)
6%extract	3.5 (±0.28)	6.55 (±0.39)	7.63 (±0.34)	7.86 (±0.14)	7.88 (±0.25)
8% extract	3% extract $3.4 (\pm 0.43)$ $5.12 (\pm 0.11)$		6.5 (±0.16)	6.52 (±0.22)	6.98 (±0.21)
10% extract	1.84 (±0.37)	2 (±0.21)	2.32 (±0.30)	2.53 (±0.36)	2.54 (±0.47)
CD at 5%	3.78	23.56	7.87	8.34	4.87
CD at 1%	7.43	47.32	15.34	16.24	10.65
		Plum	ule (cm)		
Control	2.41(±0.35)	4.88 (±0.36)	5.44 (±0.51)	5.89 (± 0.44)	6.03 (±0.34)
2% extract	0.89 (±0.22)	2.34 (±0.49)	3.38 (±0.046)	3.62 (±0.54)	3.93 (±0.75)
4% extract	0.66 (±0.39)	2.25 (±0.13)	3.36 (±0.33)	3.61(±0.12)	3.71 (±0.57)
6% extract	0.44 (±0.33)	1.79 (±0.42)	2.73 (±0.47)	2.84 (±0.27)	3.33 (±0.24)
8% extract	0.13 (±0.16)	0.46 (±0.20)	2.05 (±0.12)	2.50 (±0.24)	2.97 (±0.19)
10% extract	0.06 (±0.02)	0.08 (±0.01)	1.2 (±0.01)	2.03 (±0.09)	2.53 (±0.25)
CD at 5%	3.54	5.67	7.98	15.76	9.32
CD at 1%	7.45	12.51	15.21	32.78	19.43

Significant at p=0.05

No .of days/ treatment	3 rd day	6 th day	9 th day	12 th day	15 th day
treatment		Radi	cle (cm)		
Control	3.39 (±0.50)	6.31 (±0.47)	9.06 (±0.72)	9.24 (±0.42)	9.29 (±0.47)
2% extract	3.25 (±0.39)	5.79 (±0.43)	6.36 (±0.51)	6.75 (±0.45)	7.36 (±0.37)
4% extract	3.21 (±0.89)	5.66 (±0.30)	6.29 (±0.46)	6.40 (±0.57)	7.31 (±0.51)
6%extract	3.07 (±0.28)	5.30 (±0.39)	5.90 (±0.34)	6.26 (±0.14)	6.89 (±0.25)
8% extract	2.98 (±0.43)	5.14 (±0.11)	5.39 (±0.16)	6.10 (±0.22)	6.79 (±0.21)
10% extract	1.9 (±0.37)	4.96 (±0.21)	5.02 (±0.30)	5.89 (±0.36)	5.98 (±0.47)
CD at 5%	5.13	31.35	13.86	39.49	38.18
CD at 1%	10.96	67.36	28.29	83.67	80.12
		Plum	ule (cm)		
Control	0.51 (±0.15)	1.73 (±0.16)	4.05 (±0.51)	5.49 (±0.44)	8.78 (±0.34)
2% extract	0.35 (±0.12)	0.65 (±0.49)	2.44 (±0.046)	3.16 (±0.54)	5.17 (±0.75)
4% extract	0.33 (±0.09)	0.60 (±0.13)	2.36 (±0.33)	3.08 (±0.12)	4.80 (±0.57)
6% extract	0.21 (±0.03)	0.58 (±0.42)	2.29 (±0.47)	2.79 (±0.27)	3.82 (±0.24)
8% extract	0.19 (±0.06)	0.50 (±0.20)	2.19 (±0.12)	2.72 (±0.24)	3.55 (±0.19)
10% extract	0.01 (±0.00)	0.22 (±0.01)	1.36 (±0.01)	2.09 (±0.09)	2.6 (±0.25)
CD at 5%	2.16	5.45	9.65	11.23	32.76
CD at 1%	5.34	11.56	20.87	23.42	64.87

Table 3. Effect of the aqueous leaf extract of Ag. conyzoides against the chickpea seed.

Significant at p=0.05

Table 4. Effect of the aqueous leave extract of *P. hysterophorus* against the chickpea seeds.

Kadicle (cm)							
No. of days/ treatment	3 rd day	6 th day	9 th day	12 th day	15 th day		
Control	3.21 (±0.22)	6.35 (±0.32)	8.69 (±0.35)	9.12 (±0.36)	9.20 (±0.37)		
2% extract	2.83 (±0.23)	5.94 (0.35)	8.15 (0.41)	8.37 (0.42)	8.51 (±0.43)		
4% extract	2.81 (±0.19)	5.07 (±0.41)	5.58 (±0.52)	5.82 (±0.45)	5.85 (±0.45)		
6% extract	2.59 (±0.17)	4.73 (±0.54)	5.57 (±0.32)	5.42 (±0.35)	5.48 (±0.35)		
8% extract	2.06 (±0.24)	4.39 (±0.32)	4.79 (±0.25)	4.88 (±0.27)	4.95 (±0.29)		
10% extract	ract No germination of seeds.						
CD at 5%	65.73	90.61	88.02	88.36	89.37		
CD at 1%	138.92	191.49	186.02	186.73	188.87		
Plumule (cm)							
Control	2.12 (±0.25)	3.27 (±0.16)	3.98 (±0.24)	4.36 (±0.21)	4.48 (±0.11)		
2% extract	0.53 (±0.21)	1.77 (±0.16)	2.92 (±0.20)	3.08 (±0.31)	3.21 (±0.21)		
4% extract	0.42 (±0.18)	1.75 (±0.20)	2.86 (±0.15)	3.03 (±0.24)	3.18 (±0.22)		
6% extract	0.42 (±0.16)	1.56 (±0.11)	2.38 (±0.10)	2.54 (±0.18)	2.69 (±0.07)		
8% extract	0.36 (±0.05)	1.4 (±0.13)	2.19 (±0.05)	2.22 (±0.16)	2.32 (±0.03)		
10% extract No germination of seeds.							
CD at 5%	4.84	23.72	39.55	40.57	38.75		
CD at 1%	10.23	47.12	79.58	81.73	81.89		

Significant at p=0.05

found that the inhibitory allelopathic effect of *P. hysterophorus* leaf extract was more powerful than of other vegetative parts. The phytochemical analysis had already reported higher accumulation of growth inhibitors in leaves of *P. hysterophorus* (Kanchan and Jayachandra, 1979).

The study demonstrated that the percentage of germination was inhibited more when treated with *P. hysterophorus* leaf extract as there was no germination of seeds in 10% concentration of the plant extract used, compared to both *Cl. viscosum* and *Ag. conyzoides*. The higher concentration of the leaf extract showed more

	Cl. Viscosum		Ag. conyzoides		P. hysterophorus		
		C. arietinum (L)					
	Radicle(g)	Plumule(g)	Radicle(g)	Plumule(g)	Radicle(g)	Plumule(g)	
Control	0.34	0.23	0.24	0.22	0.32	0.23	
2% extract	0.23	0.13	0.20	0.22	0.11	0.12	
4% extract	0.13	0.13	0.18	0.21	0.11	0.11	
6% extract	0.5	0.6	0.18	0.21	0.11	0.11	
8% extract	0.03	0.3	0.12	0.15	0.03	0.3	
10% extract	0.02	0.02	0.11	0.13	0	0	

Table 5. Total oven dried biomass of radical and plumule of the seedlings of the test crop plant (i.e. *C. arietinum* L.) treated with different concentrations in *Cl. viscosum*, *Ag. conyzoides* and *P. hysterophorus* leaf extract.

inhibition. According to Kanchan and Jayachandra (1979) and Pandey (1994), P. hysterophorus is one of the best known plant invaders in the world linking allelopathy to exotic invasion. The unique allelopathic effects of some exotic species on native. 'inexperienced' communities (Callaway and Aschehoug, 2000) also contribute to invasive success. Allelopathy is expected to be an important mechanism in the plant invasion process. P. hysterophorus, because of its invasive capacity and allelopathic properties, has the potential to distrupt natural ecosystems (Evans, 1997). It has been reported earlier for causing a total habitat change in native Australian grasslands, open woodlands, riverbanks and floodplains (McFadven 1992, Chippendale and Panetta, 1994).

According to Tiwari et al. (2005) P. hysterophorus has not been used for any purpose in Nepal. Therefore this plant may become a high risk posed invasive species in near future. The present result showed that concentrated aqueous leaf extract of P. hysterophorus inhibited seed germination and seedling growth of C. arietinum.L. seeds tested, the other allelopathic plant Clerodendrum viscosum and Ag. conyzoides leaf extract also showed similar inhibitory effect on the germination and growth of the test crop plant seeds (i.e. Chickpea). According to Rao (2000) Ag. conyzoides aqueous leaf extract showed inhibitory and stimulatory influence on percent seed germination and seedling growth in different varieties of soyabean. Keeping the above in view, it is suggested that these plants should not be allowed to grow in the immediate vicinity of the agricultural fields i.e. Chickpea and other crop grown areas and this should not be used as green manure either.

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