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Table: 5, Figs : 8

The potential of plant allelochemicals to control *Parthenium hysterophorus* L. weed

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ABSTRACT

A phytosociological study was conducted for three years (2021 to 2024) in Mumbai region. Four sites were selected which were heavily infested with *Parthenium hysterophorus* L. (test weed). Along with *Parthenium*, some other weeds were cohabiting like *Calotropis procera*, (Aiton) W.T. Aiton, *Withania somnifera,* (L.) Dunal, *Cassia occidentalis* L., *Tephrosia purpurea* L. and *Solanum xanthocarpum* Schrad and Wendl. Results revealed that *Cassia occidentalis* was dominant over all other weeds including test weed, at three sites out of four selected sites. The relative frequency, relative density, relative dominance, and importance value index of *Cassia occidentalis* was found to be maximum i.e. 30.30, 53.04, 40.78 and 124.12 at site III followed by sites IV and I. To confirm this field observation, an *in-vitro* study was also conducted to observe the allelopathic potential of *Cassia occidentalis* on *Parthenium hysterophorus*. Efficacy of shoot and root aqueous leachates of 100 % concentration of 3rd and 9th day of *C. procera, W. somnifera, C. occidentalis, T. purpurea* and S. *xanthocarpum* were assessed on *P. hysterophorus* weed. Significant inhibition (P> 0.05) was observed at 100 % concentration of 9th-day shoot aqueous leachates of *C. occidentalis* on *Parthenium* weed over control followed by root aqueous leachates. Significant inhibition on Parthenium seedlings was also found to be maximum by *Cassia occidentalis*. Thus, the allelopathic potential of *Cassia occidentalis* as biological agents can be used to control Parthenium weeds population.

Key words: Allelopathy, Botanic agents, *Calotropis procera, Cassia occidentalis,* Congress grass, Molecular interactions, *Parthenium hysterophorus,* Plant sociability, *Tephrosia purpurea, Solanum xanthocarpum*, Weed biology, *Withania somnifera,*

INTRODUCTION

The word allelopathy was first used by Hans Molish in 1937 when describing the beneficial and deleterious chemical interactions of plants and microorganisms (35). Allelopathy is derived from the Greek words *Allelon* meaning "each other" and *Pathos* meaning "to suffer" (30). It, therefore, translates literally as mutual suffering. Allelopathy is defined as any direct or indirect effect by one plant, including microorganisms, on another through the production of chemical compounds (allelochemicals) that escape into the environment and subsequently influence the growth and development of neighbouring plants (26). Allelopathy is an important molecular interaction mechanism of plant interference, mediated by soil rhizosphere which affects the growth of neighbouring plants. So, this concept of biochemical interaction can be used to control the weed population.

Carrot weed, *Parthenium hysterophorus* L. (Asteraceae), is an annual herbaceous plant supposed to have originated in Northeast Mexico and during the last hundred years, it has spread to Australia, Africa, and Asia (Fig. 1 and 2). It has widely spread across Ethiopia, Kenya, South Africa, Uganda and Tanzania. This weed grows luxuriantly along roadsides, leading to substantial ecological and economic repercussions (31). This weed is now

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dominant in India, Pakistan, Nepal and China. Initially reported in India in the 1950s, it has since emerged as a significant weed problem, especially in agricultural areas and urban settings (2). *Parthenium hysterophorus* entered Australia in the 1950s, possibly via contaminated agricultural goods. Currently, it is widespread in Queensland and New South Wales, presenting a risk to pastures and agricultural areas (13).

Figure 1. Growth stages of *Parthenium hysterophorus* in experimental plot

Figure 2. Mature plant of *Parthenium hysterophorus*

Until 1977, the weed did not find any place in the world's worst weeds. Within the last 10 years, it has become one of the seven most dangerous weeds in the world (20). It is an aggressive invader of disturbed and degraded land, rapidly displacing native and planted pastures (Fig. 3). In addition to reducing land productivity, the plant causes acute allergic dermatitis and rhinitis in susceptible humans. In Australia, *Parthenium* is a serious problem in perennial grassland in Central Queensland where it can reduce beef production by as much as AU$ 16.5 million annually. In India, this weed causes yield losses of up to 48 % in several crops and is reported to reduce forage production by up to 94 %. *Parthenium* contains sesquiterpene lactones which induce several allergic reactions in susceptible individuals who are continuously exposed. It has been declared a national health hazard because of its serious skin allergies to man and animals. In India, 4-7 % of the human population suffers from recognizable clinical symptoms associated with *Parthenium*, while 43-55 % is sensitized without showing symptoms. We have observed that the local people are affected by the continuous exposure of this weed and also infestation in the cropland area.

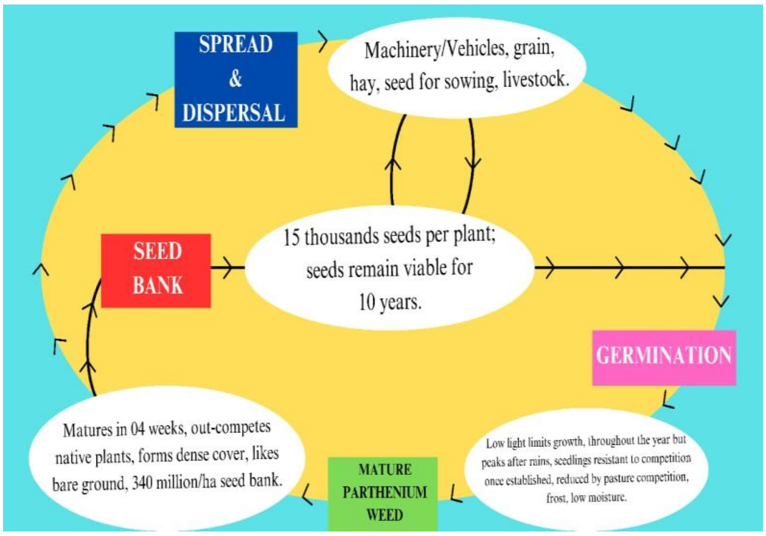

Figure 3. Spread of *Parthenium* weed after germination and quick maturation of seeds

The spread of the weed has extended to numerous Pacific Island nations, such as Fiji and Papua New Guinea. Its establishment in these areas jeopardizes indigenous biodiversity and agricultural output (4). The dissemination of *Parthenium hysterophorus* is chiefly driven by human activities, encompassing the transport of contaminated agricultural goods, vehicles and machinery. Moreover, natural dispersal agents like wind, water and animals play a role in its propagation. *Parthenium hysterophorus* displays remarkable adaptability to diverse climatic conditions, spanning tropical to temperate regions. Anticipated climate change and the rise in global trade are poised to augment its expansion into novel territories (27).

Pollen grains of *Parthenium* weed are dispersed when a flowering plant is pulled, which can cause allergy responses. It could be useful to sow pastures or other plants and to plough up weeds before the plants blossom (11). In the Dictionary of Economic Plants in India, *Parthenium hysterophorus* is described as a weed found in Poona and is reported to be used as tonic, febrifuge and emmenagogue. *Parthenium* weed may be a useful source of potash and oxalic acid (19). The weed is also a good source of easily extractable, high-quality protein that can be used in stock feeds and resembles products made from conventional forage species. *Parthenium* also has many medicinal properties. It was also demonstrated that a sublethal dose of *Parthenium* exhibited antitumor activity in mice and that the drug could either cure mice completely or increase their survival time after they had been injected with cancer cells. Other authors have found its anti-amoebic activity to be comparable to standard drugs in fighting hepatic amoebiasis. In the Caribbean and Central America, *Parthenium* weed is used as a folk remedy. It is applied externally on skin disorders and the bitter decoration of the plant is often taken internally as a remedy for a wide variety of ailments. In Jamaica, the decoction is prized as a flea-repellent bath for dogs and other animals (12).

Although herbicide application and disposal with mechanical devices like flame throwers have considerable merits, being cost-effective and providing immediate solutions to most weed problems, they are not the best solution. Besides chemical pollution, enormous cost, danger of toxicity to non-target organisms, treatment in non-agricultural areas and rapidity of reinvasion of treated areas soon after the effect is diminished, are known drawbacks of chemical control. It is still considered however, that the long-term solution will lie in releasing a guild of specific agents to hit as many of the plant organs as possible and thus gradually reduces the test weed vigour overtime. Given the urgency to develop suitable alternatives/supplements to chemical weedicides for control of this obnoxious test weed, where the pesticides are not only non-judiciously and indiscriminately applied but are also expensive, the use of botanic agents could be of great help in effectively suppressing this test weed.

It was observed during field survey that *C. occidentalis* shows dominance over test weed and neighbouring flora. *Cassia occidentalis* Linn. commonly called as coffee weed belongs to the family Caesalpiniaceae. It is an erect, annual shrub. The leaves are pubescent and are 3 to 6 cm long. The inflorescence is axillary cyme. The flowers are yellowish and its flowering time is July to September. Fruit is a legume. It was observed during field survey that *C. occidentalis* shows dominance over test weed and neighbouring flora because of its allelopathic potential. It has also been observed that grazing animals dislike *Cassia* due to its pungent odor and this is considered as a desirable trait, as it repelled cattle and successfully replaced *Parthenium* even in unprotected land. *Cassia* weed has been known to possess antibacterial, antifungal, antidiabetic, anti-inflammatory, anti-cancerous, antimutagenic and hepatoprotective activity. *Cassia occidentalis* is regarded as 'Edible weeds of Agriculture' or 'Famine food'. Its infusion is given against the white discharge. In Mali, *C. occidentalis* is used as an ingredient in a malarial formulation based on a traditional recipe comprising leaf of *C. occidentalis*, leaves of *Lippia chevalieri* and flower heads of *Spilanthes oleracea* (34). Decoction of *C. occidentalis* roots with black pepper is useful in filaria (4). In the Malyagiri hills, a decoction is made from 15 leaves each of *C. occidentalis*, *Glycosmis pentaphylla* and *Vitex* *negundo* and used for bathing the new born baby to make the baby immune to skin diseases (37).

To ascertain the probable reason, the phytosociological analysis was conducted and allelopathic potential of *Calotropis procera* (Aiton) W.T.Aiton (Asclepiadaceae), *Withania somnifera* (L.) Dunal (Solanaceae), *Tephrosia purpurea* (L.) Pers. (Leguminosae) and *Solanum xanthocarpum* Schrad and Wendl (Solanaceae) along with *Cassia occidentalis* L. (Caesalpiniaceae) were assessed on *Parthenium hysterophorus* L. (Asteraceae).

MATERIALS AND METHODS

Four sites were selected for phytosociological studies from January, 2021 to October, 2024 in Mumbai region (**Site I- Matunga**: Geographical location-19.0269 ^{O}N and 72.8553 ^{O}E, msl- 2.51 m, annual rainfall 2213.4 mm with 30 ^{O}C maximum and 24 ^{O}C minimum temperature, **Site II- Panvel**: Geographical location-18.9894 ^{O}N and 73.1175 ^{O}E, msl- 28 m, annual rainfall 1915 mm with 33 ^{O}C maximum and 23 ^{O}C minimum temperature, **Site III- Borivali**: Geographical location-19.2307 ^{O}N and 72.8567 ^{O}E, msl- 14m, annual rainfall 2225.6 mm with 29 ^{O}C maximum and 24 ^{O}C minimum temperature and **Site IV- Virar**: Geographical location-19.4564 ^{O}N and 72.7925 ^{O}E, msl- 11 m, annual rainfall 1800 mm with 27.9 ^{O}C maximum and 25.5 ^{O}C minimum temperature.

I. Phytosociological study- The frequency %, density, abundance relative frequency, relative density, relative dominance and importance value index was estimated, by using 1.0 m x1.0 m (1.0 m^2) quadrat at four selected sites. Sampling was done randomly in triplicate at 10 spots at each selected site. The data were compiled and were analyzed for qualitative and quantitative studies (7,14,21).

II. *In-vitro* allelopathy experiments

(a). Preparation of Aqueous leachates: The upper part of shoot and root tips were collected from the mature and fully grown selected plants. 100 g shoot and root tips were dipped under aseptic conditions for 3 and 9 days and placed under refrigeration at 8 ± ^{0}C. The aqueous leachates were filtered through three layers of muslin cloth/cheese cloth to remove debris. The filtrate was then re-filtered through two layers of Whatman no.1 filter paper. Leachates of 50 % and 100 % concentration were prepared with sterilized distilled water and used for bioassay.

(b). Seed Germination and Seedling growth: Seeds of *Parthenium hysterophorus* were collected in June and germination was recorded in July during monsoon season for three consecutive years from all the study sites. These seeds were thoroughly washed with tap water to remove dirt and dust and rinsed with mild detergent solution for 5-7 min. *Parthenium* seeds were surface sterilized with 0.1 % $HgCl_2$ for 10 min and again washed with sterilized distilled water for 4-7 times and divided into three replicates of 10 seeds each. *Parthenium* seeds were placed on filter paper moistened with 50 % and 100 % of shoot and root leachates. All the seeds lots were allowed to germinate in 12 cm petri dishes. The relative humidity was 82 ± 1 ^{0}C. Petri dishes were covered and placed in sealed polythene bags to prevent further loss of volatile compound and kept undisturbed for 10 days at 24± 2 ^{0}C. Control received distilled water. The seedlings of *Parthenium* were also observed, counted and recorded after one month. Germination %, germination velocity index (G.V.I), speed of germination (SoG) and seedling growth of *Parthenium* weed were calculated with the help of formula given below:

$$\text{Germination \%} = \frac{\text{No. of seeds germinated}}{\text{Total no. of seeds kept}} \times 100$$

Germination velocity Index (G.V.I) = It was calculated by following formula (6).

$$\text{G.V.I.} = \frac{\text{No.of germinated seeds}}{\text{No.of days}} \times 100$$

Speed of Germination (SoG) or Seed Vigour- Index of the speed is calculated by adding, the quotients, of the daily counts divided by the number of days of germination.

$$\text{Germination index} = \frac{n_1}{1} + \frac{n_2}{2} + \frac{n_3}{3} + \ldots\ldots\ldots\ldots + \frac{n_x}{x}$$

where,

$n_1 \ldots\ldots\ldots n_x$ = no. of germinated seeds from 1 to x.

1……… x = no. of days of germination.

Statistical analysis

Statistical analysis of the data recorded was done. The design used is Factorial Completely Randomized Design (FCRD) and conclusion was drawn from the data on the basis of Two-way Analysis of Variance (ANOVA) technique. The software used is INDOSTAT, version 98. The calculated values were compared with tabulated value at 5 % level of significance for the appropriate degree of freedom.

RESULTS AND DISCUSSION

Table-1 revealed that at site IV, III and I, *Cassia occidentalis* is a dominant species and the number of species ranged between 21-61 plants/m^2 but at site II *Parthenium hysterophorus* was dominant with 68 plants/m^2 followed by *C. occidentalis* (47 plants/m^2). Highest sociability of *C. occidentalis* was observed at site III (61 plants/m^2). The relative frequency, relative density, relative dominance and importance value index of *C. occidentalis* was found to be 30.30, 53.04, 40.78 and 124.12 at site III, followed by site I where sociability was found to be 28.57, 36.84, 27.99 and 93.40, respectively. At site IV the relative frequency, relative density, relative dominance and importance value index of *C. occidentalis* was found to be 21.42, 12.96, 8.48 and 42.86, respectively. The relative frequency, relative density, relative dominance and importance value index of *P. hysterophorus* at site II was found to be 22.22, 38.85, 33.54 and 94.61, respectively which is highest at this site followed by *C. occidentalis* (22.22, 26.85, 39.92 and 88.99, respectively). The importance value index % of *C. occidentalis* was found to be highest at site III, IV and I i.e. 41.38 %, 41.24 and 38.42 %, respectively, followed by *P. hysterophorus* in which 36.18 importance value index % was observed at site II.

Table 1. Phytosociological analysis of test weed along with potential weeds (2021- 2024)

Site No.	Name of weeds	Total no. of individual species (1.0 m^2)	Total no of quadrats in which species occur	Freq. (%)	Den.	Abun.	Rel. Freq.	Rel. Den.	Rel. Dom.	IVI (%)
I	*Cassia occidentalis*	42	10	100	4.2	4.2	28.57	36.84	27.99	38.42
	P. hysterophorus	37	06	60	3.7	6.1	17.14	32.45	40.39	37.01
	Withania somnifera	03	02	20	0.3	1.5	5.71	2.63	0.67	3.70
	Calotropis procera	10	05	50	1.0	2.0	14.28	8.77	14.40	15.40
	Tephrosia purpurea	04	02	20	0.4	2.0	5.71	3.50	4.04	5.45
	Solanum xanthocarpum	03	02	20	0.4	2.0	5.71	2.85	3.45	4.26
II	*P. hysterophorus*	68	10	100	6.8	6.8	22.22	38.85	33.54	36.18
	Calotropis procera	25	07	70	2.5	3.5	15.55	14.28	11.08	15.64
	Tephrosia purpurea	08	04	40	0.8	2.0	8.88	4.57	3.38	6.43
	Cassia occidentalis	47	10	10	4.7	4.7	22.22	26.85	39.92	34.03
	Withania somnifera	08	04	40	0.8	2.0	8.88	4.57	6.65	7.68
	Solanum xanthocarpum	06	03	30	0.6	3.0	6.48	4.12	5.27	5.28
III	*Cassia occidentalis*	61	10	100	6.1	6.1	30.30	53.04	40.78	41.38
	P. hysterophorus	17	09	90	1.7	1.8	27.27	14.78	18.23	20.09
	Withania somnifera	07	06	60	0.7	1.1	18.18	6.08	2.73	8.99
	Calotropis procera	19	05	50	1.9	3.8	15.15	16.52	27.98	19.88
	Tephrosia purpurea	11	03	30	1.1	3.6	9.09	9.56	10.26	9.3
	Solanum xanthocarpum	05	02	20	0.4	2.0	11.24	8.29	5.64	8.92
IV	*Cassia occidentalis*	21	09	90	2.1	2.3	21.42	12.96	8.48	41.24
	Withania somnifera	07	04	40	0.7	1.7	9.52	4.32	5.39	18.50
	P. hysterophorus	16	03	30	1.6	5.3	7.14	9.87	9.59	25.59
	Calotropis procera	05	03	30	0.5	1.6	7.14	3.08	1.67	11.44
	Tephrosia purpurea	01	01	10	0.1	1.0	2.38	0.61	0.34	3.20
	Solanum xanthocarpum	03	01	30	0.7	1.5	5.64	3.42	3.61	5.21

Where, Freq. % = Frequency percentage, Den. = Density, Abun. = Abundance, Rel. Freq. = Relative Frequency, Rel. Den. = Relative Density, Rel. Dom. = Relative Dominance, IVI % = Importance Value Index percentage

Hence, at three out of four selected sites, *C. occidentalis* was found to be dominant over neighbouring flora including test weed, indicating its strong influence on the adjoining species. It was observed (Table-2) that seed germination of *Parthenium* weed was significantly inhibited (at 100 % concentration) by the 9th day shoot leachates of *C. occidentalis*. The inhibitory effect increased with increase in concentration. The 3rd day shoot leachates showed less significant inhibition. Maximum inhibition was observed in *C. occidentalis* that is 98.62 %, where only 1.38 % of germination was recorded at 100 % concentration of 9th day shoot leachates and minimum inhibition was recorded in *S. xanthocarpum* i.e. 39.69 % where only 60.32 % germination at 3rd day shoot leachates was recorded at 50 % concentration. All selected weeds show 90 % germination in control (Fig.4). Similar results were observed in G.V.I and SoG. Maximum inhibition in G.V.I and SoG was observed in *C. occidentalis* treated with 9th day shoot leachates at 100 % concentration and was found to be 9.97 and 9.64, respectively followed by *C. procera, W. somnifera, T. purpurea* and *S. xanthocarpum*. At 3rd day shoot leachates, the severity was observed more at 100 % concentration and was found to be significant. The minimum inhibition of G.V.I and SoG was recorded at 3rd day shoot leachates of *S. xanthocarpum* at 50 % concentration i.e. 8.67 and 8.20, respectively. 9th day was significant over 3rd day of shoot leachates and 100 % concentration is significant over 50 % concentration of shoot leachates.

Table 2. Effects of 3rd and 9th day shoot leachates of potential weeds on germination (%), germination velocity index and speed of germination of *P. hysterophorus*

Name of weeds	**Leachates Conc (%)**	**3rd day shoot leachates**			**9th day shoot leachates**		
		Germination (%)	**G.V.I.**	**SoG**	**Germination (%)**	**G.V.I.**	**SoG**
S. xanthocarpum	50 100	60.32(7.76) 50.21(7.08)	1.33 1.16	1.80 1.66	54.20(7.36) 42.64(6.52)	1.19 1.09	1.50 5.33
T. purpurea	50 100	49.14(7.00) 36.20(6.01)	0.85 0.66	1.53 1.46	38.62(6.21) 31.43(5.60)	0.80 0.63	1.46 1.37
W. somnifera	50 100	30.21(5.49) 22.24(4.71)	0.50 0.33	1.12 0.98	26.00(5.09) 19.26(4.38)	0.33 0.25	1.08 0.83
C. procera	50 100	15.62(3.95) 8.25(2.87)	0.28 0.16	0.87 0.72	14.12(3.75) 6.84(2.61)	0.15 0.09	0.75 0.53
C. occidentalis	50 100	9.34(3.05) 3.46(1.86)	0.14 0.11	0.65 0.51	3.15(1.77) 1.38(1.17)	0.07 0.03	0.48 0.36
Control	-	90.00(9.48)	1.28	5.33	90.00(9.48)	1.28	5.33
C.D (P = 0.05)	-	9.68(3.11)	0.11	0.27	9.00(3.00)	0.10	0.20

Values in parentheses are square root transformed values
Where, G.V. I= Germination Velocity Index, SoG= Speed of Germination, CD= Critical difference, P= Probability

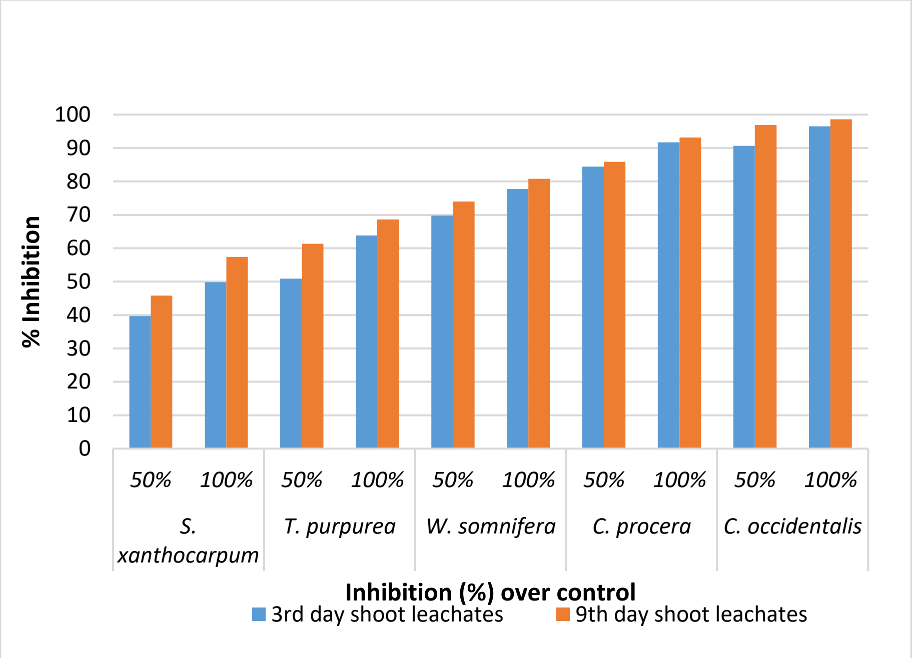

Figure 4. Inhibitory potential of shoot leachates of test weeds on germination (%) of *Parthenium*

Root leachates data depicted in table-3 showed that maximum inhibition in germination % was by *Cassia occidentalis* of 9th day root leachates at 100 % concentration i.e. 81.50 % and germination was found to be only 18.50 %, followed by *C. procera, W. somnifera, T. purpurea and S. xanthocarpum* in which 35.26, 45.65, 54.56 and 65.12 germination % was recorded with an inhibition % of 64.74, 54.35, 45.44 and 34.88, respectively. 100 % concentration is significant over 50 % (Fig. 5). Similar results were observed in G.V. I and SoG in which more severity is caused by *Cassia occidentalis* on *Parthenium* seeds in 100 % concentration of 9th day root leachates and least by *S. xanthocarpum* in which G.V.I. and SoG was found to be 8.54 and 8.05, respectively at 50 % concentration of 3rd day root leachates. Table-4 depicted that maximum inhibition in the seedling growth was by the shoot leachates of *Cassia occidentalis* and was found to be 99 % in 9th day shoot leachates at 100% concentration, followed by 50 % concentration in which 98 % inhibition was observed (Fig. 6). Both the concentrations significantly differ from each other. Minimum inhibition was observed at 3rd day shoot leachates of *S. xanthocarpum*. Similar results were observed by the root leachates of *C. occidentalis* (Table-5). Maximum inhibition was recorded in 100 % root leachates of 9th day and was found to be 98.87 %, followed by 50 % concentration in which 97.22 % inhibition was observed (Fig. 7). 100 % concentration is significant over 50 % and 9th day leachates significantly differ from 3rd day leachates in both shoots and roots.

Table 3. Effects of 3rd and 9th day root leachates of potential weeds on germination (%), germination velocity index and speed of germination of *P. hysterophorus*

Name of weeds	Leachates Conc (%)	3rd day shoot leachates			9th day shoot leachates		
		Germination (%)	G.V.I.	SoG	Germination (%)	G.V.I.	SoG
S. xanthocarpum	50 100	80.62(8.97) 70.16(8.37)	1.46 1.38	1.95 1.80	72.24(8.49) 65.12(8.06)	1.33 1.24	1.85 1.76
T. purpurea	50 100	63.46(7.96) 58.24(7.63)	1.16 0.95	1.68 1.53	60.20(7.75) 54.56(7.38)	1.02 0.88	1.62 1.45
W. somnifera	50 100	53.16(7.29) 48.29(6.94)	0.85 0.66	1.38 1.24	49.88(7.06) 45.65(6.75)	0.75 0.56	1.33 1.10
C. procera	50 100	42.37(6.50) 38.42(6.19)	0.50 0.33	1.99 1.82	40.26(6.34) 35.26(5.93)	0.43 0.28	1.86 1.36
C. occidentalis	50 100	31.12(5.57) 25.28(5.02)	0.21 0.10	1.54 1.42	26.84(5.18) 18.50(4.30)	0.13 0.06	1.43 1.30
Control	-	90.00(9.48)	1.28	5.33	90.00(9.48)	1.28	5.33
C.D (P = 0.05)		9.68(3.11)	0.11	0.27	9.00(3.00)	0.10	0.20

Table 4. Effects of 3rd and 9th day shoot leachates of potential weeds on seedling growth of *P. hysterophorus.*

Name of the plant	Leachates Conc (%)	3rd day shoot leachates	9th day shoot leachates
		Seedling growth	Seedling growth
S. xanthocarpum	50 100	60.12(7.75) 50.01(7.07)	54.00(7.34) 41.88(6.47)
T. purpurea	50 100	48.98(6.99) 35.78(5.98)	38.12(6.17) 31.93(5.65)
W. somnifera	50 100	30.00(5.47) 22.00(4.69)	27.12(5.20) 19.97(4.46)
C. procera	50 100	15.21(3.9) 9.00(3.00)	14.00(3.74) 6.22(2.49)
C. occidentalis	50 100	8.02(2.83) 2.00(1.41)	2.78(1.66) 1.00(1.00)
Control	-	90.00(9.48)	90.00(9.48)
C.D (P = 0.05)	-	8.34(2.88)	8.09(2.84)

Values in parentheses are square root transformed values
Where, CD= Critical difference, P= Probability

Table 5. Effects of 3rd and 9th day root leachates of potential weeds on seedling growth of *P. hysterophorus*

Name of the plant	**Leachates Conc (%)**	**3rd day root leachates**	**9th day root leachates**
		Seedling growth	**Seedling growth**
S. xanthocarpum	50 100	80.12(8.95) 68.12(8.25)	71.00(7.75) 64.88(7.75)
T. purpurea	50 100	63.98(7.99) 57.00(7.54)	38.12(7.75) 31.93(7.75)
W. somnifera	50 100	52.14(7.22) 46.24(7.75)	27.12(7.75) 19.97(7.75)
C. procera	50 100	40.21(7.75) 37.00(7.75)	14.00(7.75) 6.22(7.75)
C. occidentalis	50 100	28.02(7.75) 24.56(7.75)	2.78(7.75) 1.13(7.75)
Control	-	90.00(9.48)	90.00(9.48)
C.D (P = 0.05)	-	7.04(2.88)	8.09(2.84)

Values in parentheses are square root transformed values
Where, CD= Critical difference, P= Probability

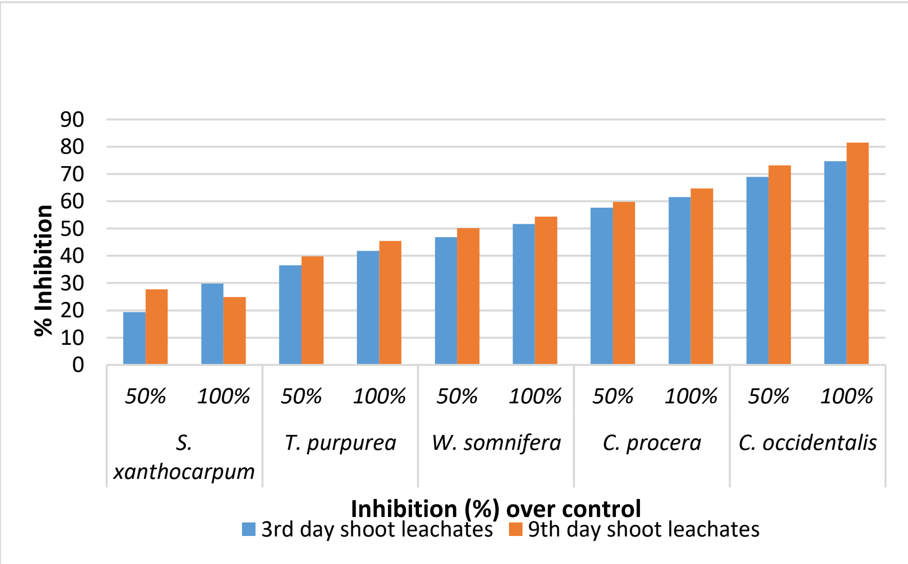

Figure 5. Inhibitory potential of root leachates of test weeds on Germination (%) of *Parthenium*

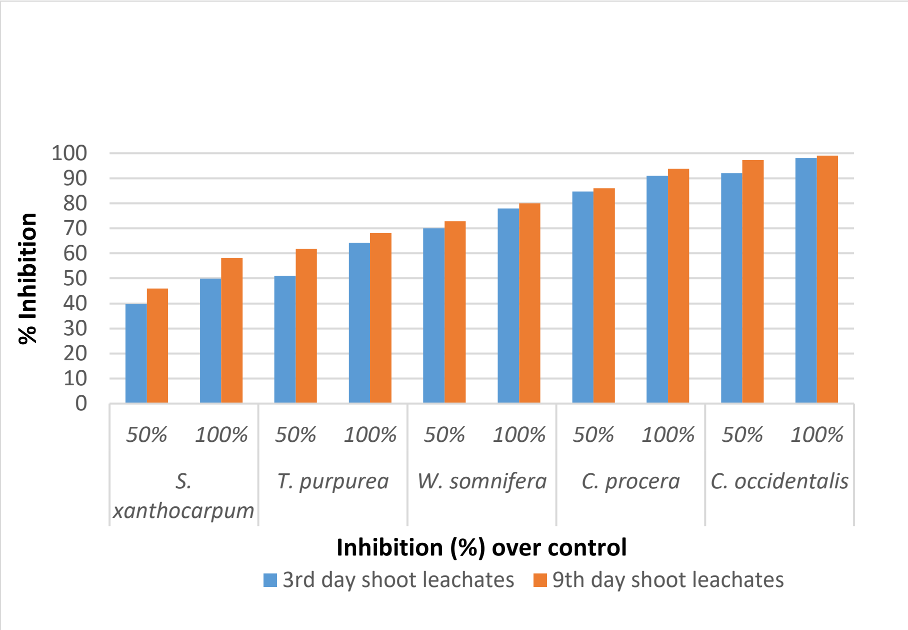

Figure 6. Inhibitory potential of shoot leachates of test weeds on Seedling growth of *Parthenium*

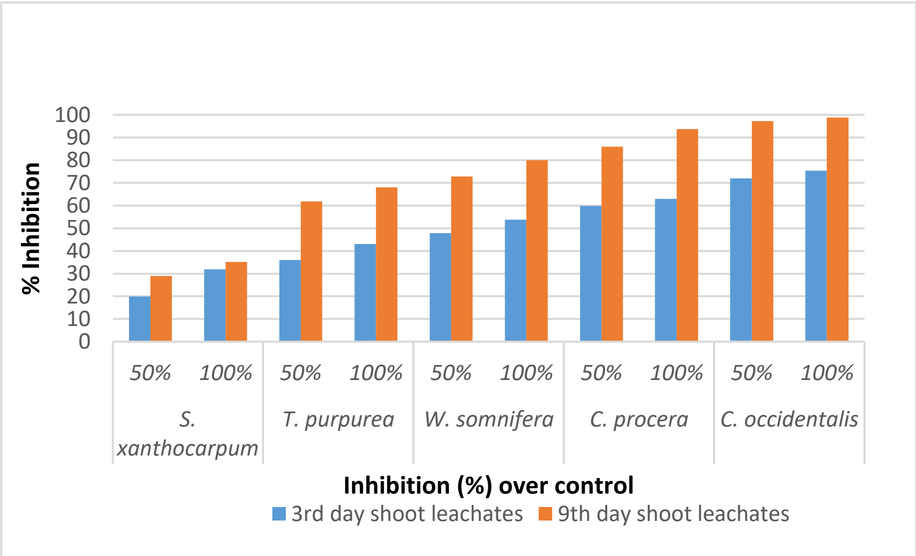

Figure 7. Inhibitory potential of root leachates of test weeds on Seedling growth of *Parthenium*

Phytosociological studies revealed that *C. occidentalis* is dominant over its neighbouring flora including test weed at three out of four sites of Mumbai region. In vitro experiments also showed similar effect where *C. occidentalis* caused marked inhibition in germination %, G.V.I and SoG of *Parthenium* seeds (test weed). Thus, it can be reasonably concluded that *C. occidentalis* has dominating allelopathic effect on germination and subsequent seedling survival of *Parthenium* weed. Seed germination %, germination velocity index, speed of germination and seedling growth were significantly inhibited by selected weeds in the order:

C. occidentalis > *C. procera*> *W. somnifera*> *T. purpurea* > *S. xanthocarpum*

Allelopathy contributes to the competitive advantage of invasive plants, the types of allelochemicals involved, their modes of action, and the ecological impacts on native plant communities (23). Allelopathic effects of methanol and chloroform extracts from two invasive plant species, (*Lantana camara* and *Broussonetia papyrifera*), on radish seed germination and lettuce seedling development was investigated. The experiments employed radish seed germination assays, seedling growth measurements and the sandwich technique to evaluate the phytotoxicity of the extracts. The results revealed a concentration-dependent inhibitory impact on radish seed germination, with *L. camara* methanol extract exhibiting the highest suppression (72.85 ± 2.69 %) at 10,000 ppm (25). Allelopathic potential of methanol extract from flowers of Lantana camara was evaluated against selected weeds viz. *Phalaris minor*, *Chenopodium album*, *Avena fatua* and *Rumex dentatus*. Results provide evidence that methyl palmitate isolated from *L. camara* flowers has herbicidal potential (3). Bio-assay with four fractions at 500 ppm, 1000 ppm and 10,000 ppm indicated chloroform fraction to be most potent phytotoxic against weeds (Monocots: *Phalaris minor*, *Avena fatua*; Dicots: *Chenopodium album*, *Rumex dentatus*). Column chromatography of chloroform fraction with hexane: ethyl acetate (60:40) based on TLC profiling with vanillin visualization stain and subsequent bioassays indicated sub-fraction (iii) of fraction 23 as most growth inhibitory (24). Allelopathic potential of *Carica papaya*, *Rhazya stricta*, *Lantana camara* and *Pinus roxburghii* hexane extracts against weeds was determined at 100 %, 75 % and 50 % concentration on soil, filter paper and agar. The results indicated that *R. stricta*, *C. papaya*, *L. camara* and *P. roxburghii* hexane extracts possesses suppression effects against weeds among which *L. camara* had the most conspicuous inhibition effects on selected weeds. The inhibitory effects of germination and seedling growth were in order *R. stricta* > *L. camara* > *C. papaya* > *P. roxburghii* (22). *Parthenium hysterophorus* is known to cause significant negative impacts on the biodiversity of natural areas, such as natural reserves, national parks, forests, and other protected areas (36). Compounds such as caffeic acid and ferulic acid (present in *Cassia)* (Fig. 8) occur in soil in appreciable amounts and have been shown in laboratory experiments to inhibit the germination and growth of many plants (8). *Calotropis procera* a common problematic weed of wastelands as well as crop fields in India is well known for its lethal allelopathic effect on many agricultural crops including chickpea, mustard, linseed etc (15,16). It also has the potential to disrupt natural ecosystems and has become a curse for biodiversity at many places in India. Aqueous extracts of root, stem and leaves of O*cimum americanum* were tested against seed germination and seedling growth of *Parthenium*. The aqueous extract of *O. americanum* significantly inhibited the germination

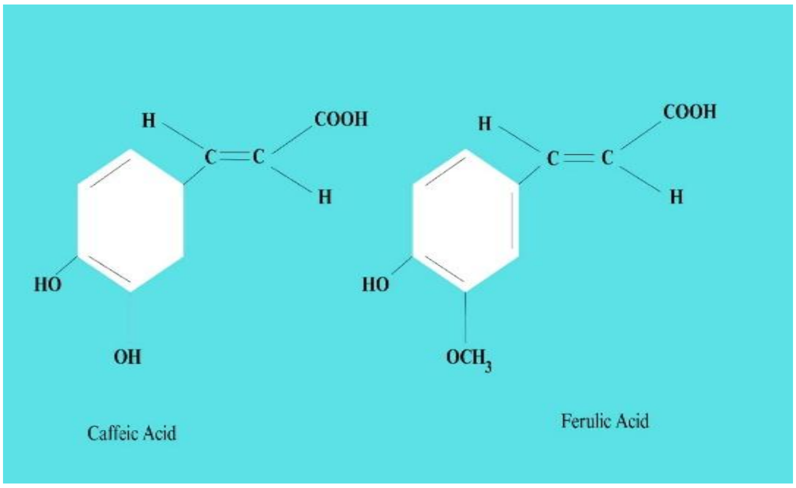

Figure 8. Molecular structure of Caffeic acid and Ferulic acid present in *Cassia occidentalis*

and seedling growth of *Parthenium.* Seed germination and seedling growth were significantly inhibited in the order: leaf > stem> root (17). The allelochemicals present in *Azadirachta indica* have inhibitory effect on the germination and seedling growth of *Parthenium* and *Amaranthus*. There was a variability in germination %, shoot and root growth of seedling due to the treatments. The degree of phytotoxicity of senescent leaves was more as compared to green leaves and bark was more inhibitory to germination of *Parthenium* and *Amaranthus.* The leachates of senescent leaves also reduced the shoot length and root length of both the weeds (28). Herbicidal potential of leaf leachates of plants like *Cymbopogon citratus*, *Withania somnifera* and *Calotropis procera* was assessed on *Parthenium.* Treatment of *Parthenium* shoots with leaf leachates of C*ymbopogon citratus* was much pronounced. Phytotoxic damage rating was found to be 3.66 ± 0.81 in 9th day leaf leachates of 100 % concentration of *Cymbopogon citratus* followed by 3.00 ± 0.00 and 2.00 ± 0.00 in *Withania somnifera* and *Calotropis procera*, respectively (10). On germination, the shoot leachates of *Cassia occidentalis*, *Abutilon indicum* and *Calotropis procera* were more effective in causing inhibition and the weakest effect was exhibited by *Ocimum sanctum.* Germination velocity index was also influenced in the same manner. However, the maximum inhibition in SoG was observed in *O. sanctum* which is otherwise appeared to be the weakest in effect (29). The harmful effect of higher leachates concentration on growth parameters might be due to excess allelochemicals which inhibit gibberellin and IAA induced growth. Chlorosis and necrosis caused the loss of chlorophyll from leaves. Drooping of leaves and twigs also decrease the photosynthetic area exposed to light. Depletion of chlorophyll is due to phytotoxic effects of allelochemicals. It was reported that mycotoxin causes the photobleaching of chloroplasts in plants (32). Polyphenolic compounds and other allelochemicals are known to change the permeability of plasma membrane. Rapid loss of integrity of plasma membrane under the influence of allelochemicals caused the cellular leakage (1). Glyphosate is a systemic, non-selective herbicide that is commonly used to control *Parthenium hysterophorus*. It is particularly effective when applied to young plants, as it inhibits essential plant enzyme pathways. A study conducted in Australia found that glyphosate (1.0 kg a.i. ha^{-1}) controlled *Parthenium* seedlings by up to 90 % when applied during the early growth stages. (13). Paraquat is a fast-acting contact herbicide that destroys green plant tissue on contact. It is often used for the immediate suppression of *Parthenium infestations*, particularly in non-crop areas. In a study conducted in Ethiopia, paraquat (0.75 kg a.i. ha^{-1}) applied to young *Parthenium* plants reduced weed biomass by 85 %. However, because paraquat only affects the plant tissue it contacts, regrowth from the root system was observed within 3-4 weeks, necessitating repeated applications (33). Pre-emergence herbicides such as oxyfluorfen and pendimethalin have been used to control *Parthenium* in non-crop areas by inhibiting seedling establishment. In an experiment conducted in Kenya, the application of pendimethalin (1.0 kg a.i. ha^{-1}) as a pre-emergence treatment reduced Parthenium seedling emergence by 75-80 %, with effects lasting for up to 8 weeks (9). Combining herbicides with other control methods, such as mechanical removal or cultural practices, has shown improved results in managing *Parthenium hysterophorus* over time. A study in India combined the application of 2,4-D with manual removal and mulching, which reduced Parthenium density by over 90 % after two growing seasons. The integrated approach provided more sustainable and long-term control than herbicides alone (18).

CONCLUSIONS

Both beneficial plants and weeds possess allelopathic compounds, yet, but, the weed species often possess much higher levels of these than the crop plants. *Parthenium hysterophorus* release allelopathic compounds from their foliage which prove unhealthy to the nearby plants. The allelopathic compounds derivative of benzoic acid, cinnamic acids, phenolic acids, coumarins, hydroquinones, benzoquinones, cineoles and other C_{15} group chemicals. These allelochemicals offer great potential as pesticides because they are free from problems associated with present pesticides. *Cassia* plants contains anthraquinones. The most widely used species of *Cassia* in herbal medicine are senna (*Cassia senna* L. or *C. acutifolia* L.). In nature, *Cassia* competes well with *Parthenium* and inhibits its growth. *Cassia occidentalis* due to biomolecular interactions, suppresses and subsequently replaced *Parthenium hysterophorus.* Thus, it provides an efficient and environment-friendly alternative to other time-consuming, costly, toxic, physical and chemical methods. Future research focusses on isolating active compounds responsible for allelopathic effects.

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AUTHORS' CONTRIBUTIONS

This work was carried out in collaboration with all authors. All authors finally approved and drafted the manuscript.

DECLARATION

We declare that all authors of this manuscript have made substantial contributions. We have not excluded any author that substantially contributed to this manuscript. We have followed our ethical norms established by our respective institutions.

ETHICAL STATEMENT

In this study, we did not involve any animal and human studies.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest

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