

Evaluation of the allelopathic capability of *Tinospora cordifolia* (Willd.) Hook. f. and Thoms leaf extract on *Brassica campestris* L., *Brassica oleracea* L., *Oryza sativa* and *Zea mays* L.

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Abstract

Allelopathic potential of *Tinospora cordifolia* (Willd.) Hook. f. and Thoms was tested on seed germination and seedling growth of *Brassica campestris* L. *Brassica oleracea* L. *Oryza sativa* and *Zea mays* L. Aqueous extract of *Tinosporacordifolia* leaves was made by soaking shade-dried leaves in distilled water for 24 hours at room temperature. Sterilized seeds were subsequently treated with extracts at concentrations of 2%, 4%, 6%, 8%, and 10%. Every day, the seeds that germinated were counted to determine the average germination time. It has been discovered that when concentration increases, germination takes a significant amount of time. Additionally, it was shown that germination percentage, root length, shoot length, and seedling vigour value were reduced at concentrations higher than 2% as compare to the control group. The strongest growth inhibiting effect on seedlings growth was demonstrated by the 10% aqueous extract of leaves of *Tinosporacordifolia* extract demonstrated potent inhibitory effects on seed germination and seedling growth in the current study.

Key words : Allelopathy, leaf aqueousextract, seed germination, seedling growth, *Brassica campestris*.

Hans Molisch⁹ originally used and actually defined the term allelopathy in 1937. It is formed from the Greek words allelo- and -pathy, which mean “mutual harm” or “suffering.” Allelopathy is a unique and intriguing field that examines how plants communicate with one another and with the

environment as a result of the chemicals they release into the atmosphere^{2,8,17}. Allelopathy is a process where a plant's (including microorganisms') direct or indirect, positive or negative effects on some other plant through the release of chemicals compound in the environment. Allelochemicals include secondary

plant metabolites that arise from primary metabolic processes in plants. Allelopathic chemicals that the plants release build up in the soil to levels that are physiologically active. Allelochemicals have many diverse impacts on plants, including those on cellular division, hormone secretion, membrane transport and permeability, germination of pollen, mineral intake, stomatal movement, synthesis of protein, respiration, nitrogen fixation, and particular enzyme activities⁶. *Calotropis procera*, also referred to as the “apple of Sodom” or “mudar,” is a plant in the Asclepiadaceae family. According to Zeng *et al.*¹⁹, this plant’s milky sap is known to include the poisonous glycosides calotropin, uscharin, and calotoxin as well as cardiac aglycones, which are steroidal heart toxins. Due of the plant’s extensive use in the control of numerous plants and its allelopathic nature, researchers also paid close attention to it. Previous research has looked into this plant’s phytotoxic and allelopathic impacts on a variety of crops⁶. Its frequent and ongoing presence near fields of wheat, cotton, sugarcane, sorghum, maize, and barley raises suspicions that it might have a deleterious effect on these crops through allelopathic interaction¹⁸. Considering these facts, the present investigation was carried out to assess the phytotoxic effect of *Tinospora cordifolia* on germination and seedling growth of *Brassica campestris* L., *Brassica oleracea* L., *Oryza sativa* and *Zea mays* L.

Fresh *Tinospora cordifolia* (Willd.) Hook. f. and Thoms plants were obtained and carried to the lab from the agricultural area. After that, it was properly cleansed with water and left to air-dry in the shade for 10 days.

Leaf samples were sorted, crushed, and maintained at room temperature in plastic bottles (average 25°C). In 100 ml of distilled water, 10 grams of powdered leaves were soaked for 24 hours at room temperature. The final quantities were adjusted to 100 ml and the aqueous extract was filtered through Whatman No. 1 filter paper, yielding 10% aqueous extract. The extracts were used as a starting point for a series of solutions with varying dilution strengths (2, 4, 6, 8, & 10). Ten healthy seeds were chosen, and each one was surface sterilised with 2% sodium hypochloride for 15 minutes. The seeds were then preserved for germination in sterilised petridishes on two layers of blotting paper, and they were moistened with 10 ml of various aqueous extract concentrations (2%, 4%, 6%, 8%, & 10%). With 10 seeds per replication, 3 different treatments were set up. A different series of controls using distilled water was put up. The petridishes were kept in a lab setting for ten days at an average temperature of 25°C. To preserve moisture content of blotting paper, petridishes were filled with equal parts of distilled water. Daily counts of germinated seeds were made in accordance with the technique for evaluating seedlings. Every 24 hours, the number of seeds that germinated was counted. For each replication of the treatment, after ten days of seeding and germination, the percentage was estimated using the formula (Germinated seed/Total seed × 100). Mean germination time was calculated by the following formula

$$MGT = \frac{\sum(Dn)}{\sum n}$$

Where D is the number of days measured from the start of germination and n is the number of

seeds that germinated on a day. The following formula was used to calculate the germination indexes (GI).

$$GI = \frac{\text{No. of germinated seeds}}{\text{Days of first count}} + \frac{\text{No. of germinated seeds}}{\text{Days of final count}}$$

The number of seedlings that emerged from each replication was tallied, and the percentage of emergence was determined using the formula below.

$$\text{Emergence\%} = \frac{\text{Emerged seeds}}{\text{Total seeds}} \times 100$$

From the point where the root and shoot came together at the end of the root to the top of the shoot, the length of roots and shoots were measured in cm. The following formula was used to determine the Seedling Vigor Index (SVI).

$$\text{SVI} = \frac{\text{Germination/Emergence \%} \times \text{Radical length (cm)}}{\text{Radical length (cm)}}$$

For biomass, the entire seedling was divided into roots and shoots, oven dried at 70° C for 48 hours until they attained a constant weight, and then weighed. The treatment means were compared using the Least Significance Difference (LSD) at a probability level of 0.05%, and the data were statistically evaluated using Fisher's analysis of variance using SPSS 16.0 software.

Effect of *Tinospora cordifolia* aqueous leaf extract on seed germination and seedling growth behaviour of four economically important crop plants viz., *Brassica campestris* L., *Brassica oleracea* L., *Oryza sativa* and *Zea mays* L. were studied. Table 1-4 displays the germination percentages for all the tested plants. In all of the studied plants, an ANOVA

showed a significant difference ($p < 0.05$) between treatments. The control had the highest seed germination percentage (93.33 to 100%), and the 8 and 10% extract of *Tinospora cordifolia* had the lowest seed germination percentage (between 33 and 36.67%). At 10% extract, *Brassica oleracea* L., *Oryza sativa* and *Zea mays* L. showed complete inhibition of seed germination. The fact that seed germination percentage significantly decreases with higher doses is quite intriguing. This demonstrated that leaf extract includes compounds that impede growth, resulting in decreased germination of all agricultural crops. Plants that display allelopathy release water-soluble phytotoxins from their leaves, stems, roots, fruits, and seeds; these compounds retard or prevent seed germination. According to Tawaha and Turk¹⁵, allelochemical stress may decrease germination rate by inhibiting water intake, and changing the activity of gibberellic acid^{7,11}. It was observed that the inhibition of seed germination was concentration-dependent^{13,16}. Each crop's germination index was independently recorded in Tables 1-4 along with a F value at a 5% level of significance. All treatment crops'

Table-1. Effect of Leaf aqueous extract of *Tinospora cordifolia* on germination & seedling growth of *Brassica campestris*

Conc. %	Germination%	GI	MGT	Root Length (cm)	Shoot Length (cm)	SVI	Biomass
Control	96.12	6.63	1.5	19.10	12.00	548.11	0.36
2%	63.23	4.04	1.78	11.36	8.42	528.43	0.31
4%	51.10	2.19	2.56	10.56	6.47	534.38	0.28
6%	35.67	2.62	2.77	8.83	4.84	306.10	0.24
8%	33.76	1.64	3.73	6.87	2.78	256.53	0.19
10%	NG	NG	NG	NG	NG	NG	NG
F Value	20.15	16.88	10.88	177	179.7	34.12	64.19
LSD 0.05%	11.65	0.265	0.59	2.16	0.63	245.02	1.35

Table-2. Effect of Leaf aqueous extract of *Tinospora cordifolia* on germination & seedling growth of *Oryza sativa* L.

Conc. %	Germination%	GI	MGT	Root Length (cm)	Shoot Length (cm)	SVI	Biomass
Control	92.33	2.32	3.47	7.8	6.27	680.23	0.586
2%	74.13	1.91	4	7.53	5.18	553.18	0.376
4%	67.68	1.7	4.14	6.82	5.6	455.45	0.281
6%	59.23	1.52	4.28	6.28	4.51	399.16	0.153
8%	50.87	1.26	4.08	5.24	4.27	262.53	0.139
10%	35.17	0.89	4.22	4.7	4.12	175.10	0.081
F Value	18.43	17.6	1.13	38.5	30.7	34.41	176.28
LSD 0.05%	9.53	0.26	NS	0.34	0.29	66.79	0.037

germination indices considerably decreased as extract concentration increased. At 8 and 10% concentrations of extract in *Glycine max* L., it is entirely inhibited. Previous research has shown that GI were entirely suppressed at 10% in *Brassica oleracea* L. and *Brassica campestris*., in contrast to our data. This is in line with Dhole *et al.*,⁴ and Dongre and Yadav⁵. At concentrations between 2 and 10%,

germination mean time was significantly shorter than control. Glycine max 5.21 at 6% has the highest recorded MGT intake, whereas *Brassica campestris* at control has the lowest. In every experiment, MGT is delayed when the concentration is raised. The highest MGT suggests that the aqueous extract of *Tinospora cordifolia* may include a substance that inhibits germination of crop seeds. Results are

Table-3. Effect of Leaf aqueous extract of *Tinospora cordifolia* on germination & seedling growth of *Brassica oleracea* L.

Conc. %	Germination%	GI	MGT	Root length (cm)	Shoot length (cm)	SVI	Biomass
Control	95.45	2.23	3.79	8.88	7.24	835.65	0.91
2%	81.87	1.83	4.54	7.53	4.66	111.23	0.61
4%	40.32	1.43	4.12	2.17	2.61	277.43	0.51
6%	15.65	1.30	5.43	0.34	0.39	129.42	0.38
8%	NG	NG	NG	NG	NG	NG	NG
10%	NG	NG	NG	NG	NG	NG	NG
F Value	60.14	57.38	74.6	454	549	302	219.4
LSD 0.05%	6.67	0.22	0.51	0.39	0.26	0.137	0.038

Table-4. Effect of Leaf aqueous extract of *Tinospora cordifolia* on germination & seedling growth of *Zea mays* L.

Conc. %	Germination%	GI	MGT	Root length (cm)	Shoot length (cm)	SVI	Biomass
Control	943.67	3.43	2.67	4.38	3.22	417.32	0.89
2%	80	2.82	3.05	3.58	2.68	285.56	0.711
4%	66.67	2.39	3.33	3.26	2.47	198	0.42
6%	53.33	2.01	2.96	2.57	1.8	136.33	0.34
8%	36.67	1.34	3.23	1.67	1.29	60.33	0.31
10%	NG	NG	NG	NG	NG	NG	NG
F Value	46.67	14.13	10.30	213.52	39.29	104	302.3
LSD 0.05%	7.69	0.59	0.64	0.29	0.38	31.11	0.06

corroborated by earlier research¹ that showed chickpea seeds immersed in *Asphodelus tenuifolius* root extract took longer to germinate.

In comparison to control, the root and shoot length of all plants decreases as extract concentration rises. *Brassica oleracea* L had the shortest roots on average while *Brassica*

campestris had the longest roots at 19.15 cm. In contrast to *Zea mays* L, which was completely inhibited at both 8 and 10% aqueous extract in the same experimental condition, *Brassica oleracea* and *Brassica campestris* showed inhibitory action at 2% and no result at 10% aqueous extract. *Brassica oleracea* displayed the longest shoots, followed by *Brassica campestris*, *Zea mays* L. and *Oryza sativa* in

the control group. The same results were reported in *Triticum aestivum*¹⁴, *Zea mays* and *Glycine max*³. In the presents investigation, the results showed that there is an inhibitory effect of the chemicals found in leaves of *Tinospora cordifolia* on the seed germination and seedling growth.

The minimum root and shoot lengths were measured in *Glycine max*, with root and shoot lengths ranging from 2 to 0.43 cm and 0.63 to 0.36 cm, respectively, for concentrations of 2 to 6%. Previous studies in *Oryza sativa*, *Triticum aestivum*, *Zea mays*, and *Glycine max*³. have also revealed that *Tinospora cordifolia* foliar leachates decrease the root and shoot elongation in these plants. This shows that leaves have a higher concentration of the inhibitory compounds available to them. Root growth is very prone to the allelochemicals present in the rhizosphere because of the pint that that root tissues have high permeability for allelochemicals as compare to shoot tissues that results in reduction of root metabolic process and division of cell in root tips. Results demonstrated that, in comparison to the control, seedling vigor index (SVI) decreases with increasing concentration of *Tinospora cordifolia* aqueous leaf extract. Wheat (SVI-1950) had the highest SVI, with soybean (SVI-835) coming in second, followed by rice (SVI-680), pigeon pea (SVI-412) and rice (SVI-680). The findings are corroborated by an earlier finding¹⁰ where *Parthenium hysterophorus* extract had an impact on *Zea mays* germination and growth. Wheat and soybean seedlings (root and shoot) were fully suppressed at 10% extract in dry weight. As the concentration of the leaf extract increased, the biomass of the test crop plants decreased,

demonstrating its potent allelopathic properties. With increasing concentrations of the aqueous extract of *Tinospora cordifolia* the biomass of *Brassica campestris* L., *Brassica oleracea* L., *Oryza sativa* and *Zea mays* L. were reduced.

As a result of its phytotoxic properties, the weed extract hindered the germination and growth of brassica seedlings. Therefore, if present in a field, this weed could prevent brassica plants from establishing a stand. The presence of this weed in agriculture fields and the surrounding areas needs to be taken seriously. The complicated allelopathic processes by which this phytotoxic plant disrupts the nearby plants, and the various allelochemicals found in *Tinospora cordifolia* can be further studied.

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