

ALLELOPATHIC EFFECT OF *Cinnamomum verum* ON EMERGENCE AND SEEDLING GROWTH OF RADISH

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ABSTRACT

Crop plants release biochemicals into surrounding environment that help in obtaining maximum yield of crop by acting as bioherbicide at high concentration and plant growth hormone at low concentration. Therefore, this study was planned at the Department of Agronomy in Weed Science Laboratory, University of Agriculture Faisalabad during Winter season of 2018 to evaluate the hermetic effects of *Cinnamomum verum* (Dalchini) on vegetable crop radish (*Raphanus sativus* L.) and determination and quantification phenolic compound present in aqueous extract of bark of *C. verum*. The experiments were laid out under completely randomized design with three replications. The aqueous extracts of *C. verum* was used on radish seeds at different concentrations (2.5%, 5%, 10%, 20%, 40% and 80%). Data regarding seed germination and seedling growth (shoot length, root length, shoot fresh weight, root dry weight) of radish were recorded following standard procedures. Results of our study revealed that the higher concentration acted as bioherbicide and exhibited inhibitory effects on *R. sativus* while the low concentration it showed hermetic effect and promoted the seedling growth. Among different phenolic compound (syringic acid, *p*-cumeic, ferulic acid, quercetion and gallic acid) were determined in aqueous extract of bark of *C. verum*, maximum quercetion (12.3%) and minimum syringic acid (0.60%). It was concluded from this study that aqueous extract of *C. verum* can be used as bioherbicide at higher concentration to control weed while at low concentration as plant growth regulator.

Keywords: Allelopathy, *Raphanus sativus*, Lower concentration, Germination and Seedling growth.

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Introduction

Crop plants release chemicals into surrounding environment that have ability to either suppress or promote the growth of target crop plant or weed. This phenomenon is very important in biological control of weeds by releasing secondary compounds. These secondary metabolic compounds such as tannins, alkaloids, glycosides, cyanogen, sesquiterpenes, phenolic acids, flavonoids, and many others having allelopathic activity (King and Ambika, 2002; Javaid *et al.*, 2010a). Weeds are unwanted plants in main crop that pose aggressive, competitive, troublesome, and multifaceted effect on crops and reduce the yields of crop (Nadeem *et al.*, 2020a). Herbicides are chemicals compounds that are used for controlling weeds, but they cause several environmental risks. Therefore, finding the harmless and potent systems for controlling weeds was of main attention for maintaining the agricultural yields (Hazrati *et al.*, 2017). Usual product escape out from the remains of medicinal plant might assist to decrease the usage of artificial made herbicides for weed management (Sodaeizadeh *et al.*, 2010; Javaid *et al.*, 2010b).

The biochemical compounds manufactured in plant as secondary compounds have been nonsignificant role in plants but act as defending agent in plant. Phenolic as well as terpenoids usually characterized as allelochemical display chemical diversity and number of metabolic and physiological biochemical process. The phenomena of allelopathy in crop plant may increase the yield as well as growth of allelopathic plant via suppressing the weed growth, with the usage of allelochemicals as natural herbicides as well as growth promoter (Maqbool *et al.*, 2021a; Javaid, 2010). These allelochemical can pass into the atmosphere via different ways such as leakage volatilization, root exudations, seed coat exudations by decaying of diverse part of plant (Rice *et al.*, 2007).

A number of researchers, such as Nadeem *et al.*, 2021a; Maqbool *et al.*, 2021b; Nelson, 1996 and Rice *et al.*, 2007 had documented the interference effects of some plants on other crops, prominent of which the inhibition of

germination (Djurdjevic *et al.*, 2004), reduction in the growth lengths of plumule and radicle (Tobe *et al.*, 2000), retardation of seedling growth (Bhatt and Todorica, 1990) and poor seedling survival (Smitt, 1990). Allelochemicals inhibits seed germination by blocking of nutrient reserve and cell division thereby caused significant reduction in the growth of plumule and radicle of many crops (Tobe *et al.*, 2000). Allelochemicals formed in plant might be escaped out from the tissue of plant and different part of plant into soil atmosphere and environment through leaching exudation of root, decaying of plant remains and volatilization and effect the growth of neighboring plant (Golisz *et al.*, 2007).

Under stressful condition the concentration of allelochemical in plants increases while under normal conduction remain stable. The main purpose of allelochemical in plants is to protect the plant form unpredictable environmental stress such as drought, mineral deficiency, temperature, herbivores grazing, water deficiency, etc. Therefore, stress simply referred as any shortage or excess supply of plant essential compounds that hinder to complete the life cycle and retard the usual growth and development of plant. The aqueous extract of different part of allelopathic plant such as roots, stem, leaves and seeds have been used valuable possessions to manage the weed through natural ways (Jamil *et al.*, 2009; Javaid *et al.*, 2020). Ethanolic or methanolic aqueous extracts were used as possible herbicide in mixtures (Cheema *et al.*, 2012). By these methods allelochemicals may be able to manage weeds via weaken weed-plant competition and enhance the crop growth and yield (Murrell *et al.*, 2011).

Water extract application of allelopathy at lower concentrations stimulates development and growth of diverse crops (Nadeem *et al.*, 2020a; Nadeem *et al.*, 2020b). *Cinnamomum verum* are a normal-sized tree (10 to 15 m) native to Sri Lanka and tropical-Asia; the cultivated in the Southern India due to strong scented leaf; bark and the aromatic oils take out from

through steam purification, barks of *Cinnamomum verum* used in experiment to check the allelopathic potential, plane, light brown color and up to 10 mm thickness; the foremost compound gain from the bark of *C. verum* are Eugenol, Cinnamaldehyde, phenolic compounds such as chlorogenic acid, vanillic acid, caffeic acid and Linalool (Kubeczka, 2002). The presence of phenolic in bark exhibit inhibitor possession on the plant germination. Based on the previous idea, the present research was conducted to study the allelopathic effect of *C. verum* on vegetable characteristics of radish crop.

Materials and Methods

Collection *C. verum* plant parts

To make aqueous extract *C. verum* (bark) plant parts were purchased from Ayub Agricultural Research Institute of Faisalabad (AARI).

Preparation of *C. verum* parts aqueous extracts

Plant parts such as bark of *C. verum* were chopped in 2 to 3 cm parts. Then the chopped bark was soaked in distilled water at 1:80 ratio for 2 days (about 48 hours). The aqueous extracts of chopped samples were filtered through filter paper. The concentrated solution was then diluted with distilled water (v/v %) to make different concentrations of solution. Almost seven concentrations (0%, 2.5%, 5%, 10%, 20%, 40% and 80%) were prepared to study the allelopathic activity of the extract. Seven concentrations 0, 2.5, 5, 10, 20, 40, and 80% were prepared by taking extract into 250 ml flask and adding 2.5, 5, 10, 20, 40- and 80-ml stock solution of *C. verum*; whereas controlled solution contained only 250 ml distilled water.

Each dilution of each extract placed in separate tagged bottles by name of each dilution and plant to avoid confusions during utilization in next procedures. The experiment was conducted in 9cm petri dishes lined with filter no.10-filter paper.

To estimate the allelopathic effect, 0%, 0.25%, 0.5%, 1%, 2%, 4% and 8% concentrations of each plant part of *C. verum* were applied on *wild pea* seeds separately. Twenty seeds of *O. punctate* were placed in each Petri

dishes containing filter paper. A 7 mL of all *C. verum* extracts dilutions of each part (leaves, stem, flower and fruit) was added in respective petri dishes having 3 replications of each dilution. One treatment was kept as control and moist with distilled water. To minimize the excess of evaporation petri dishes were covered and rapped with parafilm. The petri dishes were kept at the temperature of 30 °C and were again moistened with 3 mL after one week. The data regarding emergence of the seeds were noted every day for 14 days. After the 14 days, the germinated seedlings of wild pea were taken for different parameters like shoot length, root length, fresh and dry weight. Fresh weight was recorded instantly after harvesting while the dry weight of seedling was observed after oven drying for two days at 60 °C.

Experimental site

Laboratory experiments were conducted at weeds Science Laboratory, Department of Agronomy, University of Agriculture, Faisalabad to check the allelopathic effects of *C. verum* on wild pea (weed) and radish (crop plant).

Laboratory Experiment

This study was carried out using water as extracting medium because allelochemicals are often water soluble and released into the environment through root exudations, leaching by dews and rains or decaying of plant tissue (Turk and Tawaha, 2003). Ten vigorous seeds of wild pea were placed in Petri dishes and *C. verum* prepared aqueous solution were functionally applied at every specific petri dish purified water also cast-off like control treatment. After applying solution, Petri dishes were wrapped with paper tape and placed at room temperature. Petri dishes would keep moisture by applying solution whenever needed. Percentage of germination, mean germination, root length, shoot length and fresh weight of root and shoot were taken afterward the 12th day. Shoot length and root length were measured with measuring scale and fresh weight on weight machine. The diluted extracts of *C. Verum* (0%, 2.5%, 5%, 10%, 20%, 40%, 80%) were applied separately on wild pea.

Data collection

Mean emergence time of *P. sativum*(day)

Ellis and Reberts (1981) equation were used to examine the mean emergence time (MET).

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

Emergence index of *P. sativum*

By using formula of Association of the Official seed Analysis (1990) we recorded the emergence index

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

Emergence percentage of *P. sativum* (%)

Number of emerged seeds were counted daily according to the method of the association of Official Seed Analysis (1990) and converted into emergence percentage by the following formula.

$$\text{Emergence (\%)} = \frac{\text{No. of emerged seeds}}{\text{Total seeds}} \times 100$$

Time taken to 50% emergence of *P. sativum* (day)

The time to the 50% emergence (E_{50}) was recorded by using the formula purposed by Coolbear *et al.* (1984)

$$E_{50} = t_i + \left[\frac{\frac{N}{2} - n_i}{n_j - n_i} \right] (t_j - t_i)$$

Growth attributes of *P. sativum*

All seedlings from each petri dish were separated 14 days after emergence. After that both shoot length and root length were calculated by using meter rod from base level to top of the plants. Seedlings fresh weight was examined by separating seedlings from petri dish and measuring by using digital weight balance.

Phenolic contents

Phenolic contents were determined by using HPLC (Gradient, Reverse Phase made from shimadzu japan detector SPD-10 Av Pump LC-10-AT). Made the (w/v) solution at 1:10 ratio (10 g powdered of *C. verum* and *A. officinalis* and 100 mL methanol) Then wrapped the beaker with aluminum foil and placed for 10 days. After 10 days the material was semidried. 5mg weight with electrical balance taken out for phenolic analysis. In *A. officinalis* quercetin, pcoumaric, ferulic acid, gallic acid and syringic acid were detected.

Statistical analysis

Statistics software (version, 8.1Statistix, Tallahassee, FL, USA) was used to analyses the collected data and least significant difference test (LSD) was used to compare the means of treatment at probability level of 5%.

Results and Discussions

Allelopathic effect of *C. verum* on radish seeds emergence

Time to 50% germination (T_{50})

The aqueous extract of *C. verum* had significant effect on T_{50} of *R. sativus* (Table 1). Maximum T_{50} (5.91 days) of radish seeds was observed at T_3 (5%) concentration. Minimum T_{50} (4.03 days) of radish seeds was observed at T_5 (20%). T_7 (80%) concentration were statistically non-significant with T_6 (40%) concentration which were statistically at par with T_5 (20%), T_4 (10%) and T_2 (2.5%) concentration. T_3 (5%) concentration statistically significant with T_1 (0%) concentration. The time taken to 50% germination of radish seed was increased by (55%) at T_7 (80%) concentration as compared to control treatment T_1 (0%). The previous results revealed that extract of different plant parts of *M. oleifera* affected the rate of germination of *V. radiata* in laboratory condition. The degrees of inhibitory effects of different plant parts on germination were different. Same result reported Cheema *et al.* (1997) that sorghum water extract inhibited germination of certain weed species. sorghum extract could significantly decrease germination percentage in some crops.

Germination index (GI)

The effect of *C. verum* bark aqueous extract had significant effect on germination time of *R. sativus* (Table 1). Maximum germination (8.05) of radish seeds was observed at T_3 (5%) concentration. while minimum germination (4.93) of radish seeds was observed at T_7 (80%). T_7 (80%) concentration were statistically non-significant with T_6 (40%) concentration which was statistically at par with T_5 (20%) concentration. T_3 (5%) concentration statistically significantly with T_1 (0%) concentration. The time taken to 50% germination of radish seed was increased by (55%) at T_7 (80%)

concentration as compared to controlled T₁ (0%). The result was corresponding to Costa and Zimmermann (1988), who stated that according to environmental conditions, some production components may increase or decrease.

Mean germination time (MGT)

The effect of *Cinnamomum verum* bark aqueous extract had significant effect on mean germination time of *Raphanus sativus* (Table 1). Maximum MGT (6.44 day) of radish seeds were observed at T₁ (0%) concentration. Whereas MGT (4.82 days) of radish seeds was observed at T₅ (20%) concentration of extract. T₇ (80%) concentration were statistically non-significant with T₆ (40%) concentration which was statistically at par with T₅ (20%), T₄ (10%) concentration. T₃ (5%) concentration statistically significant when compared with T₁ (0%). The obtained results supported by previous studies that concentrations of allelochemicals at small concentrations of allelochemicals might stimulate the development and growth while increase concentrations suppress or inhibiting the growth and germination of target plant (Hazrati *et al.*, 2017; Tigre *et al.*, 2012).

Germination percentage (GP)

The bark extract of *C. verum* had significant effect on germination percentage of *Pisum sativum sub species elatius* (Table 1). Maximum percentage of germination (96.6 %) of wild pea seeds was observed at T₁ (0%) concentration of extract. whereas minimum percentage of germination (70.0%) of wild pea seed was observed at T₇ (80%) and T₆ (40%) concentration of extract. T₇ (80%) concentration showed statistically significant relationship with T₁ (0%) concentration. T₇ (80%) concentration showed non-significant relationship with T₆ (40%) concentration. The germination percentage of wild pea seed was significantly decreased 27% at T₇ (80%) concentration as compared to controlled T₁ (0%). Same result reported Cheema *et al.* (1997) that sorghum water extracts inhibited germination of certain weed species. Sorghum extract could significantly decrease germination percentage in some crops.

Allelopathic effect of aqueous extract of bark of *C. verum* on seedlings growth of radish

Shoot length (mm)

The extract of *C. verum* (bark) had significant effect on shoot length of radish as described in Table 2. Maximum shoot length (93.8 mm) of radish seedling was observed at T₇ (80%) concentration. Minimum shoot length (61.6 mm) of radish were observed at controlled T₁ (0%) concentration. T₂ (2.5%) concentration showed non-significant relationship with T₆ (40%) concentration which was statically at par with T₃ (5%) and T₅ (10%) concentration. T₇ (80%) concentration showed significant relationship with T₁ (0%) concentration. The maximum shoot length of radish significantly increased (52%) at T₇ (80%) concentration as compared to control T₁ (0%) concentration. Increased in concentration not dramatically reduce the shoot length of radish. Increase in shoot length of radish over control by application of bark extract of *C. verum* showed that stimulatory allelochemicals in these extracts might be the reason. The obtained result was not according to Reigosa *et al.* (2006) that Cinnamon abstract repressed the fungus growth yet 2.0 and 2.5% have the most effective because these concentrations were not permit the disease-causing agent to grow and showed 100% inhibition in bio-mass production. Besides these also proposed that increase concentrations might badly influence the development of the plant. But the results are supported by Rice, (1984) analysis that difference in extract concentration might be shown different results, (both inhibitory and stimulating results).

Root length (mm)

The extract of *C. verum* (bark) had significant effect on *R. sativus* L. as described in Table 2. The extract brought about considerable inhibition in the root length of radish seedling. Degree of inhibition increased with increasing the concentrations of the extracts. This tends to suggest that the effect of the extract is concentration dependent. The maximum root length (129.3 mm) was observed at control T₁

(0%) concentration. Minimum reduction in root length (21.7 mm) was observed at T₇ (80%) concentration. At the highest extract concentration all aqueous extracts significantly, reduced root length as to compared controlled treatment. T₇ (80%) concentration statistically showed non-significant relationship with T₆ (40%) concentration. T₄ (10%) concentrations statistically showed significant relationship with T₃ (5%) concentration. T₂ (2.5%) concentrations showed significant relationship with T₇ (80%). T₇ (80%) concentration showed significant relationship with control T₁ (0%) concentration. The root length of radish was significantly decreasing (83%) at T₇ (80%) which was statically similar at T₆ (40%) concentration as compared to control T₁ (0%) concentration. These results were similar with earlier studies and seedling growth bioassays that water extracts of allelopathic plants were more pronounced effects on radicle growth than on hypocotyl growth and might be the presence of numerous allelochemicals in the abstract solution (Rice *et al.*, 2007). Arooj *et al.* (2021) directed that the atrazine produces hermetic effect at dose of 10 g a.i. (lower dose) while showed herbicidal at higher dose (80 g a.i.) and produce maximum root length (9.99) and minimum root length (7.34 cm) respectively.

Shoot fresh weight

The aqueous extract of *Cinnamomum verum* (bark) had significant effect on shoot fresh weight of *Raphanus sativus* L. as described in Table 2. Maximum fresh weight of shoot (16.3 mg) were noted at T₅ (20%) concentration. Minimum fresh weight of shoot (10.2 mg) were noted at T₄ (10%) concentration treatment. T₇ (80%) concentration showed non-significant relationship with T₂ (2.5%) concentration which was statically similar with T₃ (5%) and T₆ (40%) concentrations. T₅ (20%) concentration showed significant relationship with control T₁ (0%) concentration. The fresh weight of shoot significantly increased (11%) increased at T₇ (80%) concentration as compared to control T₁ (0%). The fresh weight of shoot significantly decreased (21%) at

T₄ (10%) concentration as compared to control T₁ (0%) concentration. The increase in fresh weight of radish seed may be due the stimulatory allelochemicals in these extracts and shoot of radish show more resistant at higher concentration of extract. While lower concentrations of extract were suitable to reduce the fresh weight of radish. That result is not corresponding to Srivastava *et al.* (1996) that water abstracts of plants and inflorescences at higher concentration suppressed the germination seedling growth and reduce the fresh and dried mass of barley wheat and pea. (Arooj *et al.*, 2021) reported that Atrazine at lower dose (10 g a.i.) produce hermetic effect and gave highest fresh weight while at higher dose (80 g a.e.) produce herbicidal effect and produce minimum fresh weight of *Tribulus terrestris*.

Root fresh weight

The effect of *C. verum* (bark) aqueous extract had significant effect on root fresh weight of *R. sativus* as described in table (2). Maximum root fresh weight (57.6 mg) was observed at T₇ (80%) concentration. Minimum fresh weight of root (30.0 mg) was noted at control T₁ (0%). T₇ (80%) concentration had significant relationship with control T₁ (0%). T₃ (5%) concentration had non-significant relationship with T₄ (10%) concentration which was statically at par with T₂ (2.5%), T₆ (40%) and T₁ (0%) concentration. The fresh weight of root significantly increased 92% increased at T₇ (80%) concentration as compared to control T₁ (0%). The fresh weight of shoot significantly decreased 42% at T₆ (80%) concentration as compared to T₇ (80%) concentration. Highest concentration of extract not significantly reduce the root fresh weight of radish. The rise in fresh weight of radish may be due the stimulatory phytochemicals in these extract and root of radish show more resistant at higher concentration of extract. While lower concentrations of extract were suitable to reduce the fresh weight of radish. Similar results were reported by Mubeen *et al.* (2011) who found that leaf extract of *Trianthema portulacastrum* inhibited maximum reduction on shoot and root

length of rice seedling whereas minimum shoot and root dry weight was occurred as a result of interactive effect of different weeds and their water extracts.

Phenolic compounds and their concentration in *C. verum*

Phenolic compounds and their concentration in *C. verum* were presented in Table 3. In *C. verum* quercetion, vanilic acid, pcoumeric acid, caffic acid and chlorogenic acid were detected. Among these phenolic compounds detected in *C. verum* maximum quercetin (12.3%) compound and minimum p-coumaric aid (0.89%) was found.

Conclusion

The results of experiment directed that aqueous extract of bark of *C. verum* showed inhibitory effect on radish germination and seedling growth at higher concentration (80%) and growth regulatory effect at lower concentration. Among phenolic compounds detected in *C. verum*, maximum Quercetin (12.3%) compound and minimum p-coumaric acid (0.89%) were found.

Accordingly, aqueous extract of bark of *C. verum* can be used as potential bio-herbicide to control weed at 80% concentration and growth regulator at lower concentration. Also, further studies are recommended in order to fix and support the obtained findings.

Table 1: Allelopathic effect of *C. verum* on emergence of *R. sativus* seeds

Concentration (%)	Time to 50% germination (days)	Germination Index	Mean germination Time (days)	Germination (%)
0 %	4.59b	6.21b	6.44b	83.3b
2.5 %	4.56b	5.39bc	5.89bc	80.0bc
5%	4.42b	6.08b	5.45cd	83.3b
10%	4.27b	6.14b	5.25cd	86.6ab
20%	4.03b	8.05a	4.82d	96.6a
40%	4.58b	5.06c	5.78bd	70.0c
80%	5.91a	4.93c	7.54a	70.0c
LSD	0.7665	0.8662	1.0029	10.111

Table 2: Allelopathic effect of *C. verum* seedling growth on *R. sativus* seeds

Concentration (%)	Shoot length (cm)	Root length (cm)	Shoot fresh weight (cm)	Root fresh weight (cm)
0 %	61.6d	42.4d	13.0d	30.0d
2.5 %	79.0c	61.7c	14.3bc	33.3c
5%	80.6bc	52.0cd	14.5bc	33.3c
10%	85.0b	86.8b	14.6b	45.3b
20%	93.8a	129.3a	16.3a	57.6a
40%	80.8bc	26.6e	14.0c	30.6cd
80%	77.6c	21.7e	10.2e	30.6cd
LSD	5.6203	10.027	6.0967	3.0274

Table 3: Phenolic compounds and their concentration in *A. officinalis*

Phenolic compounds	Concentration
Syringic acid	0.60
p-crumeic	0.94
Ferulic acid	7.48
Quercetion	12.3
Gallic acid	4.69

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