



RESEARCH ARTICLE

Allelopathic effects of bitter gourd (*Momordica charantia* L.) whole plant extract and dry biomass on germination and growth of vegetable cowpea, okra and amaranth

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Abstract

The study was carried out to evaluate the allelopathic potential of bitter gourd on vegetable crops such as vegetable cowpea, okra and amaranth. The research comprised a germination bioassay (September 2023 to October 2023), to assess the allelopathic effect of whole plant extract of bitter gourd on germination and early seedling growth of vegetable cowpea and okra and a soil incubation study (November 2023 to December 2023) to evaluate the effect of bitter gourd dry biomass on the growth of amaranth seedlings. Both experiments were laid out in a Completely Randomised Design (CRD), with each treatment replicated five times. Statistical analysis ($p < 0.05$) revealed that the aqueous extract of the whole plant of bitter gourd at a concentration of 5 w/v (T_1) and 10 w/v (T_2) caused a delay in seed germination in both okra and vegetable cowpea and a reduction in root and shoot growth in vegetable cowpea at the early growth stages compared to the control (T_3 -distilled water). The treatment T_2 recorded the lowest seed germination (78.00±8.37 % in vegetable cowpea on day 5; 78.00±4.47 % in okra on day 4 of the study) but was statistically on par with T_1 . Soil incubation, with lower concentrations of bitter gourd dry biomass (I_1 -5g and I_2 -10 g), enhanced amaranth seedling dry weight at 20 and 30 days after transplanting (DAT), while higher concentration (I_3 -15 g) exhibited an inhibitory effect at 20 DAT compared to the control without incubation (I_4). Through the adoption of proper management strategies to minimise the initial inhibitory effect of bitter gourd, crops such as vegetable cowpea, okra and amaranth can be successfully integrated into bitter gourd-based cropping systems.

Keywords: allelochemicals; bitter gourd; germination; inhibition; whole plant extract

Introduction

Allelopathy is an ecological phenomenon where in organisms interact biochemically through the release of chemical compounds (allelochemicals) into the surrounding environment, further influencing the growth, survival and reproduction of other organisms (1). Allelopathic interactions can be either inhibitory or stimulatory and play a key role in shaping ecosystems. Allelochemicals are typically secondary metabolites from plants and microbes which regulate numerous environmental processes (2). Leaves, stems, roots, rhizomes, seeds, flowers and even pollen from plants serve as sources of allelochemicals. Several types of allelochemicals have been identified, including flavonoids, benzoxazinones, alkaloids, cinnamic acid derivatives, ethylene and cyanogenic compounds (3). The impact of allelochemicals on the target organism is determined by their concentration, chemical composition and existing environmental conditions. Allelochemicals influence physiological and biochemical processes in plants such as cell division, cell elongation, enzymatic activity, hormone balance,

respiration, photosynthesis, water relations and mineral availability, leading to reduced growth, vigour and productivity (4).

Allelopathy has a prominent role in determining the success and sustainability of cropping systems. Autotoxicity and soil sickness are the most frequent problems related to allelopathy. The continuous cultivation of the same crop over the years can lead to the buildup of allelochemicals or pathogenic microbes, resulting in autotoxicity or soil sickness. In crop rotation, the allelochemicals from one crop can either inhibit or stimulate the subsequent crop. Incorporating crop residues into the soil is a common practice in cropping systems for promoting sustainable resource management. However, these residues can sometimes release allelopathic compounds, potentially influencing the germination and growth of subsequent crops. Similarly, the success of intercropping or mixed cropping systems relies on the allelopathic interaction among the component crops. Vegetable species with strong allelopathic effects have been extensively studied (5). Several studies have documented reduced yields in

vegetables in monoculture and cropping systems largely due to allelopathic effects originating from crops, weeds and trees (5-8). In vegetable-based cropping systems, allelopathic interactions may be more evident and significantly affect overall crop performance. Thus, identification and understanding of such interactions are crucial in determining suitable crop combinations and system productivity.

Bitter gourd, a widely cultivated cucurbit in Kerala, is valued for its nutritional and therapeutic properties. It is a rich source of vitamin C, beta - carotene, thiamine, riboflavin, dietary fibre and essential minerals such as calcium, iron, phosphorus and potassium. Bioactive compounds in bitter gourd exhibit anti-diabetic, antioxidant, anti-inflammatory, anti-tumor, hypolipidemic and hypoglycemic activities (9). The rising consumer preference for natural, plant-based and nutrient-rich diets has increased the demand for functional foods like bitter gourd. Reflecting this trend, the cultivation of bitter gourd in Kerala has expanded in recent years, with the area under cultivation increasing from 2258.43 ha in 2018-19 to 2325 ha in 2021-22 (10, 11). Bitter gourd can be successfully introduced into cropping systems; however several studies have identified the allelopathic potential of bitter gourd on other crops. The effect of leaf and stem extracts of bitter gourd on germination and early development of eight vegetable species (*Cucumis sativus*, *Capsicum annuum*, *Vigna unguiculata*, *Raphanus sativus*, *Daucus carota*, *Lycopersicon esculentum* var. *cerasiforme*, *Phaseolus vulgaris* and *Lycopersicon esculentum*) was evaluated and the findings revealed the significant influence of these extracts on both seed germination and seedling growth, although the extent varied with species (12). Notable changes in the growth of tomato seedlings were observed upon exposure to leaf and fruit leachates of bitter gourd (13). The stimulatory effect of bitter gourd was reported earlier (14), noted an increase in dry weight of mung bean after treatment with bitter gourd root extracts. Bitter gourd is rich in phenolic constituents like gallic acid, chlorogenic acid and ferulic acid, which are documented to have allelopathic activity (15). Moreover, the presence of various volatile organic compounds, such as aldehydes and terpenes was found in bitter gourd, which are known for their involvement in allelopathic interactions (16). At present, the validity of introducing okra, amaranth and vegetable cowpea, the major vegetables commercially cultivated in Kerala (17), into the bitter gourd-based cropping system is under consideration. Therefore, it is necessary to investigate the potential effect of bitter gourd on other crops grown along with or after bitter gourd. A comprehensive understanding of allelopathic interactions is essential for the effective design and management of these cropping systems.

Although allelopathy in bitter gourd has been documented, existing research is limited and predominantly focuses on the effect of individual plant parts, such as leaves, stem or fruit on allelopathic activity, while the effect of the entire plant on subsequent crops remains inadequately studied. Incorporating bitter gourd residues into the soil after the final harvest offers a sustainable strategy for nutrient recycling; however, its allelopathic implications require further investigation. The release of phenolic compounds into the soil through processes such as residues decomposition, leaching and root exudation are considered as major contributors allelopathic effects in numerous crops and weeds (18). Therefore,

comprehensive studies assessing the effects of the whole plant are essential. In this context the present study was conducted to assess the allelopathic potential of bitter gourd on vegetable crops, namely vegetable cowpea, okra and amaranth. It was hypothesized that the whole plant extract and dry biomass of bitter gourd would exert a stimulatory effect at lower concentration and inhibitory effect at higher concentration on test crops. The study included a germination bioassay to determine the allelopathic effect of the whole plant extract of bitter gourd on germination and early seedling growth of vegetable cowpea and okra. A soil incubation study was also included to evaluate the effect of bitter gourd dry biomass on the growth of amaranth seedlings.

Materials and Methods

The germination study was conducted under laboratory conditions from September 2023 to October 2023, to determine the possible allelopathic effect of bitter gourd on seed germination and early seedling growth of vegetable cowpea and okra. The experiment included three treatments: aqueous extract of whole plant of bitter gourd at concentration of 5 % (w/v) (T₁) and 10 % (w/v) (T₂) and a control treatment using distilled water (T₃). A separate pot culture soil incubation study was conducted from November 2023 to December 2023 to examine the allelopathic effect of bitter gourd on the growth of amaranth seedlings. The study comprised of four treatments: soil incubation with bitter gourd plant dry biomass at 5 g (I₁), 10 g (I₂), 15 g (I₃) and a control without incubation (I₄). Both germination and soil incubation studies followed a CRD, with each treatment replicated five times. Treatments were randomly allocated to experimental units to reduce bias. The experiments were conducted at College of Agriculture, Vellayani, in Thiruvananthapuram, Kerala, India, situated at 8°30'N latitude, 76°54'E longitude and 29 m above mean sea level.

Bitter gourd variety "Preethi", vegetable cowpea variety "Githika", okra variety "Anjitha" and amaranth variety "Arun" was selected for the experiment. The seeds of bitter gourd were procured from the Agricultural Research Station, Mannuthy, whereas the seeds of the remaining varieties were obtained from the Department of Vegetable Science, College of Agriculture Vellayani. For conducting allelopathic studies, bitter gourd plants were grown during July-August 2023 and during the active growth stage, fresh plant samples were carefully uprooted from the field, ensuring minimal damage to the roots.

Soil required for the incubation study was collected from the field, which had a sandy loam texture, consisting of 67.83 % coarse sand, 17.82 % fine sand, 5.03 % silt and 9.32 % clay (19).

Preparation of bitter gourd whole plant extract

Plant samples were initially washed under running tap water for a few seconds to remove any soil particles adhering to the surface. This was followed by a quick wash with distilled water. The samples were then dried by vigorous hand shaking and gently wiping with a dry, clean cloth. The extraction was carried out immediately after drying. For the preparation of aqueous extract, fresh plant material was weighed first and then chopped into small pieces. Then the biomass comprising roots, stems and leaves of the plant was ground thoroughly using a mortar and

pestle. Distilled water was then added to the homogenized sample at 1/10 (w/v) ratio and mixed thoroughly. The resulting solution was left to stand in the dark at room temperature for 24 hrs before being filtered through Whatman No. 1 filter paper. The filtrate was then diluted to achieve different treatment concentrations (20). To prevent microbial activity and degradation, the extract was stored at 7-8 °C (21).

Germination bioassay

Petri dishes with a uniform diameter of 9 mm were utilised for all the treatments. Prior to use, all the plates were sterilized by autoclaving at 120 °C and under a pressure of 15 atmosphere pressure to eliminate potential microbial contamination. Healthy and uniform seeds of test crops okra and cowpea were selected and were subjected to surface sterilization using 0.1 % sodium hypochlorite (22). For each treatment, ten seeds of each test crop were placed in a petri dish lined with germination paper. The paper was moistened on alternate days with 5 mL of plant extracts, while distilled water was used for the control treatments. All the Petri dishes were incubated in in BOD Incubator at 25±1 °C and a photoperiod of 12 hrs. The optimum humidity of 80±5 % was maintained in the incubator to avoid evaporative loss.

Soil incubation study

Bitter gourd plants at the active growth stage were freshly collected from the field, shade dried and powdered. The powdered samples were then stored in the refrigerator till the experiment began. Uniform-sized mini pots were filled with 500 g of soil and each was incubated with dry biomass of bitter gourd for seven days as per the treatments. A set of pots without any incubation was also maintained as control. Twenty-five-day-old amaranth seedlings raised at the College of Agriculture, Vellayani, were then transplanted individually into each pot. Soil moisture was maintained at field capacity throughout the experiment period. The experiment was laid out with five replications per treatment to ensure the reliability of the data.

In germination bioassay, germination and early seedling growth were assessed by recording the number of seeds germinated, along with the root and shoot length of seedlings at specific intervals. In vegetable cowpea, the germination count was recorded on the 5th, 8th, 14th and 17th day of the experiment and for okra on the 4th, 8th and 21st day of the study. Seeds with a radicle length of at least 2 mm were considered germinated. Root lengths of all germinated seedlings were measured on the same observation days as germination assessment for each crop. Shoot length measurements were done on the 14th and 17th days for cowpea and on the 18th and 21st days for okra.

From the above observations, germination percentage and seedling vigour index were computed. Germination percentage was calculated using the following formula:

$$\text{Germination percentage} = \frac{\text{Total number of seeds germinated}}{\text{Total number of seeds sown}} \times 100 \quad (\text{Eqn. 1})$$

Seed Vigour Index I (SVI) was determined using the formula proposed by (23).

$$\text{SVI} = \text{Seedling length (cm)} \times \text{germination percentage} \quad (\text{Eqn. 2})$$

In the incubation study, the amaranth seedling height was recorded at 10, 20 and 30 days after transplanting (DAT), while seedling dry weight per plant was noted at 20 and 30 DAT. For dry weight assessment, the seedlings were carefully uprooted and oven dried at 65 ± 5 °C until a constant weight was obtained. Based on these observations percentage inhibition or stimulation was computed using the formula.

$$\text{Percentage of inhibition or stimulation} = \frac{T - C \times 100}{C} \times 100 \quad (\text{Eqn. 3})$$

Where: C - dry weight of seedling in control; T - dry weight of seedling in treatment

Statistical analysis

The experimental data were analysed following standard statistical procedures. One way ANOVA was performed to determine whether significant differences existed among treatment means at 5 % level of significance ($\alpha=0.05$). Upon obtaining a significant ANOVA result, a multiple comparison test was conducted using the Least Significant Difference (LSD) test. All the statistical analyses were carried out using grapes Agri 1 software (24).

Results and Discussion

Allelopathic effect of bitter gourd on seed germination and early seedling growth of vegetable cowpea

The whole plant extracts of bitter gourd applied at different concentrations significantly influenced the germination and early seedling growth of vegetable cowpea (Table 1.). Significant effects ($p < 0.05$) of bitter gourd whole plant extract on seed germination were observed only on the fifth day of the experiment. The treatment T₂ (10 % w/v) resulted in the lowest seed germination

Table 1. Effect of extracts on germination and seedling growth of vegetable cowpea

Treatments	Number of germinated seeds			Root length (cm)				Shoot length (cm)		Seed vigour index	
	5 th day	8 th day	14 th day	5 th day	8 th day	14 th day	17 th day	14 th day	17 th day	14 th day	17 th day
T ₁	8.60±0.89 ^{ab}	9.80±0.45	10.00±0.00	1.34±0.11 ^a _b	3.73±0.24 ^a _b	7.27±0.57 ^b	7.74±0.53	3.48±0.43	4.91±0.63	1074.60±54.18 _b	1220.50±68.25
T ₂	7.80±0.84 ^b	9.60±0.55	10.00±0.00	1.25±0.07 ^b	3.55±0.29 ^b	6.83±0.41 ^b	7.60±0.73	3.20±0.19	4.47±0.62	1003.10±33.90 _c	1270.60±63.71
T ₃	9.60±0.55 ^a	10.00±0.00	10.00±0.00	1.48±0.14 ^a	4.11±0.36 ^a	7.98±0.50 ^a	8.26±0.31	3.54±0.18	5.11±0.40	1151.20±58.20 _a	1317.00±55.86
SEm (±)	0.35	0.18	0.00	0.05	0.13	0.22	0.25	0.13	0.25	22.32	28.09
CD (0.05)	1.067	NS	NS	0.152	0.414	0.684	NS	NS	NS	68.770	NS

Cell values are mean±SD; Treatments with the same letter grouping are not significantly different, Different letters indicate significance at $p < 0.05$ according to LSD test, NS-non significant.

(7.80 ± 0.84 seeds, 78.00 ± 8.37 %), demonstrating the strongest inhibitory effect. However, T_1 (5 % w/v) showed statistically comparable seed germination parameters with 8.60 ± 0.89 seeds germinated and 86.00 ± 8.94 % germination percentage (Fig. 1). The control treatment (T_3) using distilled water, recorded significantly higher number of germinated seeds (9.60 ± 0.55) compared to that in treatment T_2 . No statistically significant differences in germination were observed among the treatments on the 8th, 14th and 17th day of observation. By day 14, all seeds had germinated in each treatment, suggesting that bitter gourd extracts primarily delayed seed germination rather than causing permanent inhibition. These findings agree with previous studies, documented a delay in germination of mung bean treated with root, stem and leaf extracts of bitter gourd (25). Allelopathic chemicals disrupt enzymatic and metabolic activities in seeds, impairing seed structure and thereby causing seed deterioration and reduction in seed vigour (26).

The whole plant extract of bitter gourd demonstrated a stronger allelopathic effect on root growth than on shoot growth, with the effect being non-significant on shoot length. Similar observations were made by former researchers (27). The inhibitory effect on root length was significantly ($p < 0.05$) evident only during the early growth stages up to the 14th day of the study. The treatment T_2 (10 % w/v) resulted in the shortest root lengths with values of 1.25 ± 0.07 cm on day 5, 3.55 ± 0.29 cm on day 8 and 6.83 ± 0.41 cm on day 14, which were statistically like those of T_1 (5 % w/v). A similar trend was observed in the SVI on the 14th day, with the lowest value (1003.10 ± 33.90) recorded in T_2 , followed by T_1 and the control treatment T_3 .

A significant inhibitory effect of bitter gourd on vegetable cowpea was observed at higher extract concentration relative to the control. Similarly, a marked reduction in root and shoot length was observed in plants treated with leaf and fruit leachates of

bitter gourd, with the degree of inhibition increasing progressively with concentration (13).

Allelopathic effect of bitter gourd on seed germination and early seedling growth of okra

Bitter gourd whole plant extract showed a profound impact on okra seed germination. However, its effect on root length, shoot length and SVI was non-significant (Table 2). A significant effect ($p < 0.05$) in seed germination was found only on the 4th day of the study, with no significant effects recorded at subsequent stages of observation. On day four, the control treatment (T_3) recorded the highest number of seed germination (9.20 ± 0.84), which was on par with T_1 (5 % w/v), with a seed germination of 8.40 ± 0.55 . The lowest number of seeds (7.80 ± 0.45) was germinated in treatment T_2 (10 % w/v), which was statistically at par with T_1 . The germination percentage followed a similar trend, with values of 84.00 ± 5.48 % in T_1 , 78.00 ± 4.47 % in T_2 and 92.00 ± 8.37 % in T_3 (Fig. 2). These results are consistent with the findings of former studies (28), reported a delay in germination of okra seeds at higher concentrations of extracts from sorghum stem, maize root and tassel, with some treatments exhibiting no germination for up to 24 hrs.

The inhibitory effect of the extracts on okra seed germination was found to intensify with increasing extract concentration, like the trend observed in vegetable cowpea. In both the cases of vegetable cowpea and okra a clear inhibitory effect was noted at the initial stages of observation; however, as the experiment advanced, the allelopathic effect was no longer significant. The adverse effect of allelopathy decreased with study duration (14) and the crops may recover from this initial inhibition over time. For instance, even though DTD (4,7-dimethyl-1-(propan-2-ylidene)-1,4,4a,8a-tetrahydronaphthalene-2,6(1H,7H)-dione) can cause significant increase in ABA content in rice roots initially, this effect can decrease after a certain period (96 hrs), indicating that the initial inhibitory effects might not be permanent (29).

Table 2. Effect of extracts on germination and seedling growth of okra

Treatments	Number of germinated seeds		Root length (cm)			Shoot length (cm)		Seed vigour index	
	4 th day	18 th day	4 th day	18 th day	21 st day	18 th day	21 st day	18 th day	21 st day
T_1	8.40 ± 0.55^{ab}	10.00 ± 0.00	1.22 ± 0.20	2.23 ± 0.30	2.74 ± 0.23	3.04 ± 0.21	3.38 ± 0.23	527.00 ± 39.57	612.80 ± 24.52
T_2	7.80 ± 0.45^b	10.00 ± 0.00	1.28 ± 0.16	2.14 ± 0.24	2.51 ± 0.38	2.94 ± 0.17	3.25 ± 0.18	507.20 ± 39.18	576.40 ± 51.17
T_3	9.20 ± 0.84^a	10.00 ± 0.00	1.39 ± 0.37	2.52 ± 0.15	2.95 ± 0.13	3.03 ± 0.20	3.42 ± 0.24	554.80 ± 16.39	636.80 ± 30.58
SEm (\pm)	0.28	0.00	0.12	0.11	0.12	0.09	0.10	14.99	16.64
CD (0.05)	0.872	NS	NS	NS	NS	NS	NS	NS	NS

Cell values are mean \pm SD; Treatments with the same letter grouping are not significantly different, Different letters indicate significance at $p < 0.05$ according to LSD test, NS-non significant.

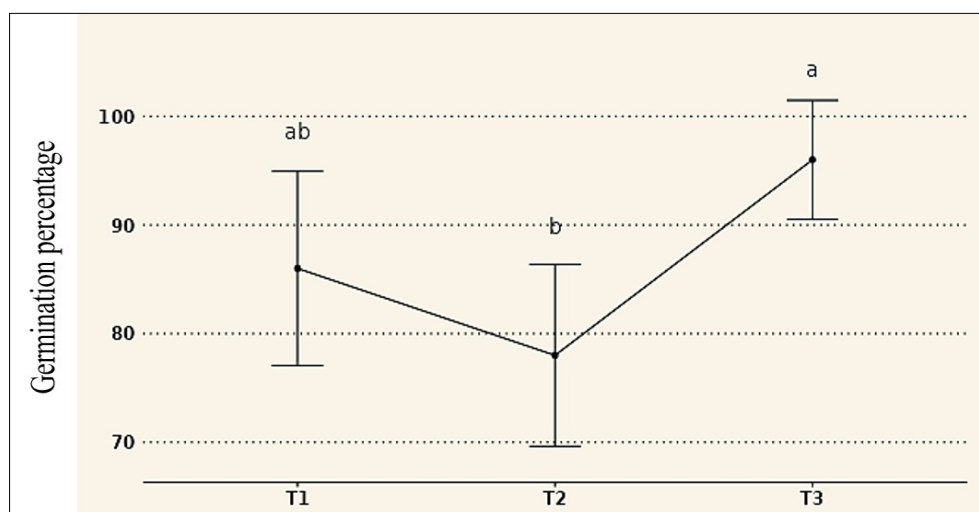


Fig. 1. Allelopathic effect of bitter gourd on germination percentage of vegetable cowpea on 5th day of the study.

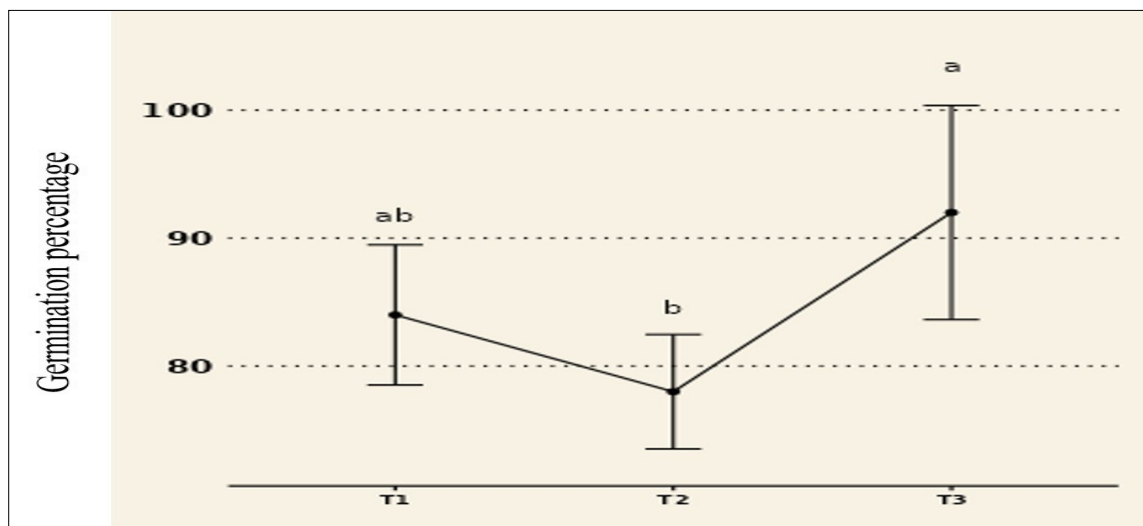


Fig. 2. Allelopathic effect of bitter gourd on germination percentage of okra on 4th day of the study.

Allelopathic effect of bitter gourd on amaranth seedlings

Incubation of bitter gourd dry biomass at various concentrations significantly affected the growth of amaranth seedlings (Table 3). The impact on plant height was significant ($p < 0.05$) only at 30 DAT, with no significant influence at 10 and 20 DAT. At 30 DAT, the treatment I_3 (15 g bitter gourd plant dry biomass) recorded the tallest plants (49.04 ± 1.57 cm), followed by I_2 (10 g bitter gourd plant dry biomass), which was statistically on par with the control I_4 (without any incubation). The shortest plants, measuring 37.72 ± 4.81 cm were observed in I_1 (5 g bitter gourd plant dry biomass), but did not show any significant difference from the control.

A significant effect ($p < 0.05$) on seedling dry weight was observed at both 20 and 30 DAT. Among the treatments, I_2 (10 g bitter gourd plant dry biomass) recorded the highest dry weight of 0.72 ± 0.07 g at 20 DAT and 1.77 ± 0.09 g at 30 DAT and was statistically comparable to treatment I_1 (5 g bitter gourd plant dry biomass). The treatment I_3 (15 g bitter gourd plant dry biomass) recorded significantly the lowest dry weight (0.40 ± 0.03 g) at 20 DAT, whereas at 30 DAT, the treatment I_4 (control) recorded the lowest dry weight (1.11 ± 0.12 g), although statistically comparable to I_3 (15 g bitter gourd plant dry biomass). Differential responses in plant height and dry weight of amaranth were observed upon incubation with bitter gourd dry biomass. A higher dry weight does not necessarily correspond to greater plant height, as biomass accumulation is influenced by several factors, including stem thickness, leaf number, root development, etc. Since dry weight provides a more reliable indicator of overall growth performance than height alone, it was selected as the basis for drawing conclusions.

While lower concentrations of dry biomass of bitter gourd (I_1 and I_2) exhibited a stimulatory effect relative to the control (I_4), with a dry weight increase of 16.82 % and 22.47 % respectively,

treatment I_3 (15 g bitter gourd plant dry biomass) resulted in an inhibitory response, with a reduction in dry weight by 32 % (Fig. 3). These findings reveal a concentration-dependent dual response of bitter gourd biomass on seedling growth. Similar observations were made by (30), where higher concentration of phenolic compounds such as ferulic acid, p-hydroxybenzoic acid and p-vanillic acid inhibited germination of *Chenopodium album*, while lower concentrations of all phenolics (except p-hydroxybenzoic acid) exhibited a stimulatory effect. This pattern of dose-response, known as hormensis, is well documented in allelopathic studies, where lower concentration of allelochemicals causes a stimulatory effect and higher concentrations lead to inhibitory effects (31). All the treatments showed a stimulatory effect compared to the control (I_4) at 30 DAT (Fig. 3), with the highest increase in dry weight (61.63 %) noted in I_2 (10 g bitter gourd plant dry biomass). Treatments I_1 (5 g bitter gourd plant dry biomass) and I_3 (15 g bitter gourd plant dry biomass) exhibited an increase in dry weight by 43.83 % and 18.94 %, respectively. These findings concur with those of (32), who reported the growth promoting effect of hot pepper root exudates on crops such as tomato and radish, although the stimulatory response was reduced at higher concentrations. At the lower concentration, the plant material needed more time to break down and release enough chemicals to affect the crop growth, whereas at higher concentration, since there was more plant material, the necessary amount of chemical was released much faster (33), which might account for the initial inhibitory effect. However, over time, these compounds may have broken down or become diluted, reducing their inhibitory effect at later stages.

At lower concentrations, allelochemicals may induce mild stress in target plants, triggering adaptive responses that promote growth and development. Allelochemicals, including phenolics and flavonoids at lower concentrations enhanced the activity of

Table 3. Effect of incubation of dry biomass of bitter gourd on seedling growth of amaranth

Treatments	Plant height (cm)			Dry weight of seedlings (g)	
	10 DAT	20 DAT	30 DAT	20 DAT	30 DAT
I_1	12.90 \pm 1.94	25.92 \pm 5.29	37.72 \pm 4.81 ^c	0.69 \pm 0.03 ^a	1.58 \pm 0.14 ^a
I_2	17.26 \pm 4.06	31.20 \pm 5.85	43.45 \pm 1.30 ^b	0.72 \pm 0.07 ^a	1.77 \pm 0.09 ^a
I_3	15.66 \pm 1.85	29.76 \pm 3.17	49.04 \pm 1.57 ^a	0.40 \pm 0.03 ^c	1.30 \pm 0.24 ^b
I_4	16.36 \pm 3.31	27.00 \pm 4.12	38.72 \pm 6.46 ^{bc}	0.60 \pm 0.10 ^b	1.11 \pm 0.12 ^b
SEm (\pm)	1.32	2.11	1.86	0.03	0.07
CD (0.05)	NS	NS	5.570	0.086	0.213

Cell values are mean \pm SD; Treatments with the same letter grouping are not significantly different, Different letters indicate significance at $p < 0.05$ according to LSD test, NS-non significant.

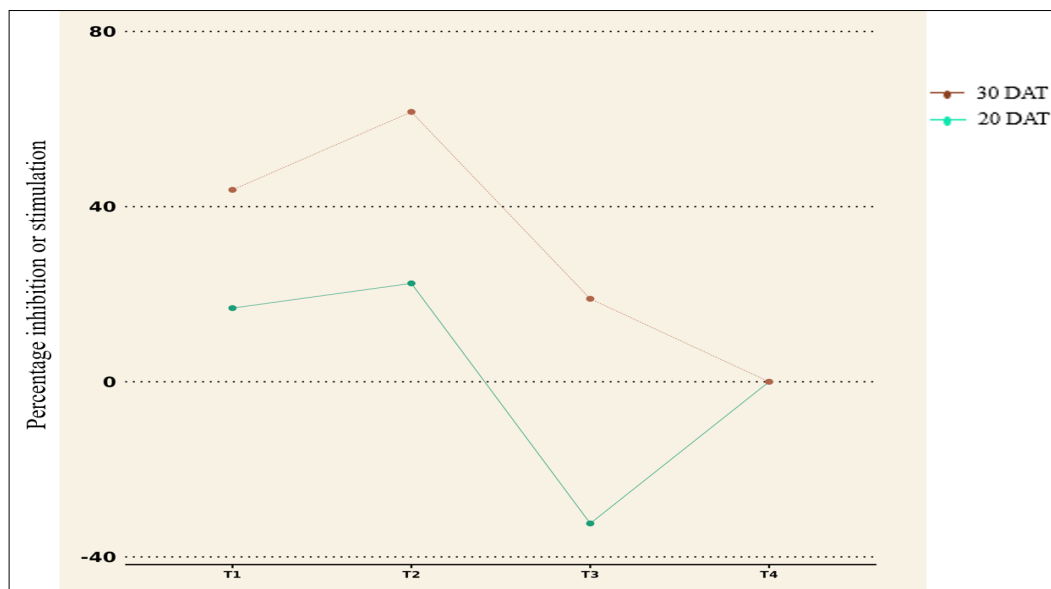


Fig. 3. Effect of incubation of bitter gourd plant dry biomass on the percentage inhibition or stimulation of dry weight of amaranth.

antioxidant enzyme like superoxide dismutase, catalase and ascorbate peroxidase, thereby supporting the redox equilibrium of cell and improving abiotic stress resistance (34, 35). Several studies have reported that applying allelochemicals at sub-inhibitory levels enhances enzyme activity, promotes cell division, improve ion absorption and lead to enhanced plant growth and development (36). In addition, certain allelochemicals at lower concentrations may improve nutrient uptake by the plant, leading to improved growth. For example, diphenylamine at a lower concentration enhanced the absorption of N and K by tomato roots (37).

The differential responses observed with whole plant extract and dry biomass of bitter gourd-specifically, the inhibitory effect of whole plant extract at higher concentration and stimulatory effect of bitter gourd dry biomass upon soil incubation-might be due to the crude aqueous extract containing concentrated allelochemicals that exert a direct phytotoxic effect on test plants, whereas the decomposition of dry biomass incorporated in to soil may have released nutrients, that promote plant growth. Once released into soil, allelochemicals interact with organic and inorganic soil phases, as well as with soil microorganisms, which determines their bioavailability and phytotoxic level (38). Studies have shown difference in the effects of allelochemicals even between dry and fresh leaf extracts (39).

Conclusion

In both vegetable cowpea and okra, the aqueous extract of bitter gourd initially exhibited an inhibitory effect at higher concentration. However, as the experiment progressed, this allelopathic influence became non-significant. Similarly, in the case of amaranth, soil incubation of dry biomass of bitter gourd showed an inhibitory effect at higher concentrations in the early stages. Over time, the effect became stimulatory at all the concentrations. These results suggest that, by adopting appropriate management measures to minimise the initial inhibitory effect of bitter gourd, crop such as vegetable cowpea, okra and amaranth can be effectively integrated into bitter gourd -based cropping systems without adverse effects of allelopathy.

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Authors' contributions

SSR carried out the laboratory experiments, collected the data and drafted the manuscript. RG, SPP, JJ, RVM and GPP participated in design, coordination and draft correction. All authors read and approved the final manuscript.

Compliance with ethical standards

Conflict of interest: Authors do not have any conflict of interest to declare.

Ethical issues: None

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