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Comparative allelopathic effects of six weed extracts on seed germination and seedling growth of four pulses

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Abstract

The present scientific investigation deals with the allelopathic potential of *Cayratia* sp., *Mikania* sp., *Acalypha* sp., *Croton* sp., *Jatropha* sp., and *Sanchezia* sp. weeds on seed germination and seedling growth of *Cicer arietinum* (chickpea), *Vigna mungo* (black gram), *Pisum sativum* (pea) and *Lens culinaris* (lentil). The experimental results of *C. arietinum* and *V. mungo* in petridish experiment revealed that the lowest germination percentages 30 % and 47.6 % shown in *Mikania* sp. treated plate where control was 100 % but in case of *P. sativum* shown lowest germination 30 % treated with *Acalypha* sp. In the case of *L. culinaris*, the lowest germination 40 % treated with *Croton* sp. *C. arietinum* seeds in pot experiment show the lowest germination percentages 37.5 % recorded in treated with *Cayratia* sp. and *Acalypha* sp. extracts. In *V. mungo* seeds documented the rate of germination 40 % treated with *Acalypha* sp. and *Croton* sp. extracts whereas *P. sativum* and *L. culinaris* seeds recorded germination rate 30 % treated with *Acalypha* extract. Shoot length of *C. arietinum* recorded 1.76 cm and root length 1.99 cm treated with *Mikania* sp. and *Croton* sp. In the case of *V. mungo* shoot length recorded 3.6 cm and root length 0.7 cm treated with *Mikania* sp. In case *P. sativum*, shoot length and root length recorded 1.3 cm and 1.03 cm treated with *Mikania* sp. and *Croton* sp. In case of *L. culinaris*, shoot length and root length recorded 2.43 cm and 0.93 cm treated with *Mikania* sp and *Croton* sp.

Keywords

allelochemicals; pulses; seed germination; seedling growth; weeds

Introduction

The Fabaceae or Leguminosae family, also referred to as the legume, pea, or bean family, is the third-largest group of flowering plants, with more than 20000 species (1). The term “pulse” is limited to the annual legume crops that are specifically grown for dried and edible seeds. Chickpea, cowpea, pigeonpea, faba beans, lentils and dry beans are some of the types of pulses (2). Pulses are the second-most consumed food crops in the world, right behind cereal grains. They are a crucial source of food for the poor, particularly in developing and underdeveloped countries. Moreover, pulse-based products are in high demand among consumers around the world due to the significant nutritional value for the human diet they offer in terms of protein and mineral quality and bioavailability (3). Incorporated into cropping systems, pulses increase the efficiency of both water and nutrient use, as they can fix atmospheric nitrogen into soils and allow companion crops to use stratified soil water, thereby contributing to sustainability in crop production (4). A total of 89.8 million metric tons of pulses were produced worldwide in 2020, with India being the largest pulse producer (5). A wide range of pulse crops are cultivated around the world, including

chickpea (*Cicer arietinum*), pigeonpea (*Cajanus cajan*), mungbean (*Vigna radiata*), urdbean (*Vigna mungo*), cowpea (*Vigna unguiculata*), lentil (*Lens culinaris Medikus ssp. culinaris*), horse gram (*Macrotyloma uniflorum*), French bean (*Phaseolus vulgaris*) and lathyrus (*Lathyrus sativus*). There have been major challenges in increasing total pulse production to meet its global demand due to both biotic and abiotic stresses. Since pulses take so long to reach maturity, weeds often get a head start on the crops and end up smothering them. Furthermore, most pulses are grown in conjunction with nonlegume crops and 84 % of that area is grown under rain-fed conditions. For this reason, pulses are vulnerable to a wide range of biotic and abiotic stresses (6). Weeds are one of the most challenging problems facing agricultural production all over the world. Weeds compete for light, nutrients, water and space that reduce crop growth and yield. Weed infestation in crops accounts for the highest yield loss, i.e., 34 %, compared to the losses associated with any pests, such as insects and pathogens, depending upon crops and weed's emergence time, density and nature (7,8). Weeds not only reduce crop yields but also impede other agricultural operations and serve as an alternative host for a wide variety of pests and diseases. It is vital to bring the weed density below the threshold level and maximize the crop yield and quality. Additionally, weeds also harbor insect pests, bacterial, fungal and viral pathogens, further reducing the crop yield. Allelopathy is a direct or indirect effect caused by one plant (including microorganisms) on another through the production of chemical compounds that escape into the environment (9,10). Allelochemicals are plant metabolites or their products that are released into the environment, which may affect plants at different stages of plant growth and development, such as seed germination, seedling growth and development, flowering and fruiting, vegetation formation and succession, species regeneration (11-14). The huge number allelopathic interactions are typically negative in character, with positive relations being rare (9). Allelochemicals affect germination and growth of neighboring plants by disruption of various physiological processes including photosynthesis, respiration, water and hormonal balance (15).

Seed germination is the most important stage in plant growth and development for all crop plants (16,17). Allelochemicals can be presented in every organ of plant parts such as roots, rhizomes, leaves, stems, bark, flowers, fruits and seeds (9,12,15). The allelochemicals of sesame released to the environment through stem and leaf leachates, root exudates and residue decomposition liquid, may have allelopathic effect on seed germination and seedling growth of recipient plants (17). Numerous studies were accomplished about the allelopathic effects of sesame on the germination and growth of other plants. Duary reported that sesame leaf extracts in different concentrations had allelopathic influence on germination, seedling growth and dry matter production of black gram (*V. mungo* L.) and rice (*Oryza sativa* L.) (18). Soleymani and Shahrajabian found that sesame extracts caused the delay in the growth and germination of canola (*Brassica napus*) (19). The aim of this study is to investigate the effects of six weeds extracts on seed germination and seedling growth of four pulses.

Materials and Methods

Collection and identification of weeds

To conduct the experiment, leaves of four common types of largely growing weeds were collected from the areas near the local agricultural fields of Raghunathpur, Balurghat block, Dakshin Dinajpur, West Bengal. The weeds were brought to the Department of Botany, Balurghat College, where they were identified and authenticated at the beginning of the experiment. The identified weeds were namely *Cayratia* sp., (W1) *Mikania* sp. (W2), *Acalypha* sp. (W3), *Croton* sp. (W4). Some leaves of *Jatropha* sp. and *Sanchezia* sp. which are not commonly referred to as weed were also collected for the experiment from the field near Balurghat forest, Raghunathpur.

Preparation of leaf extracts of weeds

Leaves of all the six above-mentioned plants were first washed with distilled water and then desiccating them in an open area for several days with the help of sun until each leaf became brittle, after that the leaves were pulverized separately with the help of an electric blender. These leaf dusts were weighed and kept separately for the subsequent steps of experiment. Leaf dust of all the above-mentioned plants were then mixed with 500 mL methanol separately and kept in separate flasks covered with cotton plugs. The flasks were kept closed for several days with little boiling and shaking periodically. These solutions were then filtered with the help of Whatman no -1 filter paper to get rid of the large particles of the dried leaves.

In vitro seed germination and vigor index

Seeds of *C. arietinum*, *V. mungo*, *P. sativum* and *L. culinaris* were taken from a local seed store of Balurghat, Dakshin Dinajpur and sterilized with 5 % sodium hypochloride solution. Ten seeds were taken in 28 petridishes marked by control and treated plates and soaked with sterile distilled water and plant extract. Seeds soaked with sterile double distilled water were set as a control and moistened with sterile distilled water in every alternate day. The effective germination was considered to happen when the radicals achieved half of the seed length. The percentage of seed germination was recorded at every 24 hr interval for 10 days. The rate of seed germination, root and shoot length was measured after 10 days. The experiment was repeated thrice. The germination rate [GR (%)] and vigour index [VI] were calculated following the formula (20):

$GR (\%) = (\text{number of seeds germinated} / \text{total number of seeds}) \times 100$

$VI = \% \text{ germination} \times \text{total plant length}$

In vivo pot experiment

The experimental soil collected from 0 to 15 cm soil depth from the university campus was air-dried, ground, sieved (2 mm mesh), and autoclaved at 121 °C for 20 min. About 2 kg of sterile dry soil taken in a plastic pot (12 cm diameter; 20 cm deep). Ten seeds of similar size and shape were sowed in each pot and labelled properly. Twenty eight pots were used for each of the five different sets of treatment and all the experimental pots were maintained in a sunny area during the spring season and watered regularly using sterile water and plant extract. The different growth promotion parameters viz. shoot length, root length, shoots dry weight;

root dry weight were recorded from 30 days to 75 days from each pot at 15 days interval. The dry biomass of shoot and root was done by taking the weight before and after oven drying at 65 °C for 72 hr.

Results

In vitro assay on seed germination

From the above experiment, it was observed that the rate of germination has decreased in all the plants having the influence of the weeds. In the plant of *C. arietinum*, the rate of germination has decreased from 100 % in control, 80 % in *Jatropha* sp. and *Sanchezia* sp. extracts, 50 % in *Cayratia* sp. and *Croton* sp. extract and 40 % in *Acalypha* sp. extract to 30 % in *Mikania* sp. extract in the seventh day which is shown in Fig. 1.

In *V. mungo* plant, the weeds minimized the rate of germination, 100 % in control and *Jatropha* extract, 94.12 % in *Sanchezia* sp. extract, 64.7 % in *Cayratia* extract, 58.82 % in *Acalypha* extract and 52.94 % in *Croton* extract to 47.6 % in *Mikania* sp. extract which is shown in Fig. 2.

For the plant of *P. sativum*, the rate of germination has decreased from 100 % in control, 90 % in *Jatropha* sp. extract, 80 % in *Sanchezia* sp. extract, 50 % in *Cayratia* sp. extract and 40 % in *Mikania* sp. and *Acalypha* sp. extract to 30 % in *Croton* extract which is shown in Fig. 3.

In the case of *L. culinaris*, the weeds have retarded the rate of germination from 100 % in control, 90 % in *Sanchezia* sp. and *Jatropha* sp. extract, 60 % in *Mikania* sp. extract and 50 % in *Acalypha* sp. and *Cayratia* sp. extract to 40 % in *Croton* extract which is shown in Fig. 4.

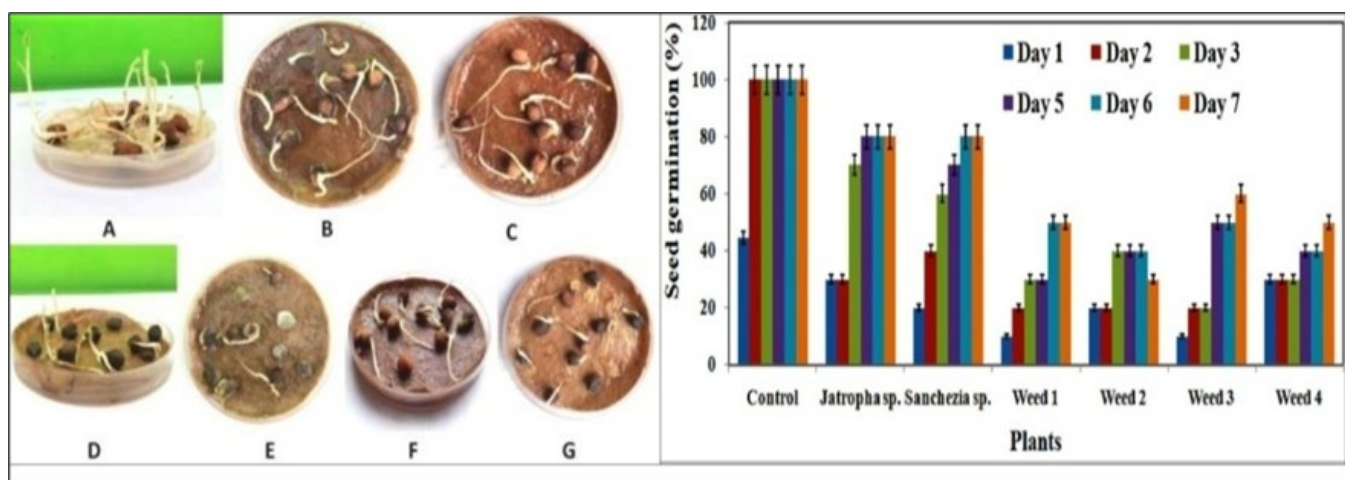


Fig. 1. Seed germination of *Cicer arietinum* against six weeds extracts where (A) Control; (B) *Jatropha* sp.; (C) *Sanchezia* sp.; (D) *Cayratia* sp.; (E) *Mikania* sp.; (F) *Acalypha* sp.; (G) *Croton* sp. up to seven days.

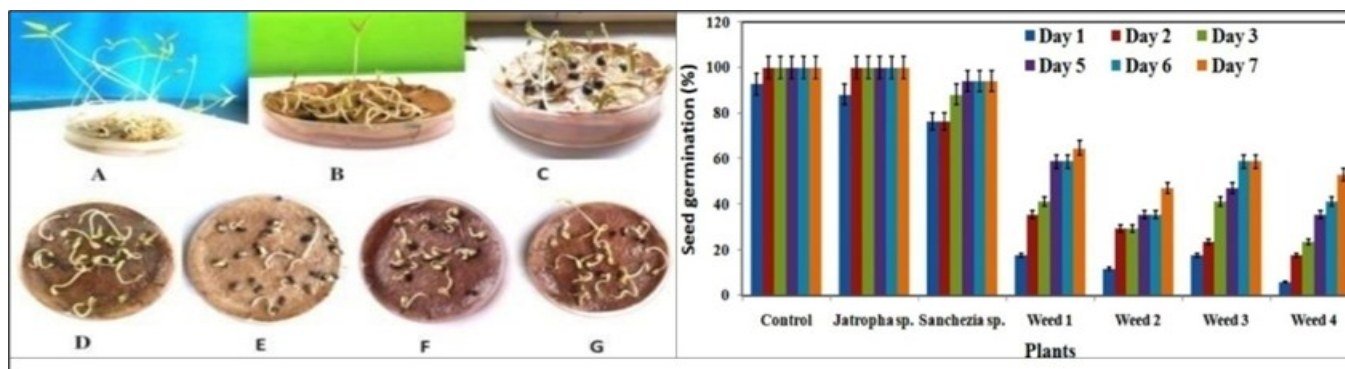


Fig. 2. Seed germination of *Vigna mungo* against six weed extracts where (A) Control; (B) *Jatropha* sp.; (C) *Sanchezia* sp.; (D) *Cayratia* sp.; (E) *Mikania* sp.; (F) *Acalypha* sp.; (G) *Croton* sp. up to seven days.

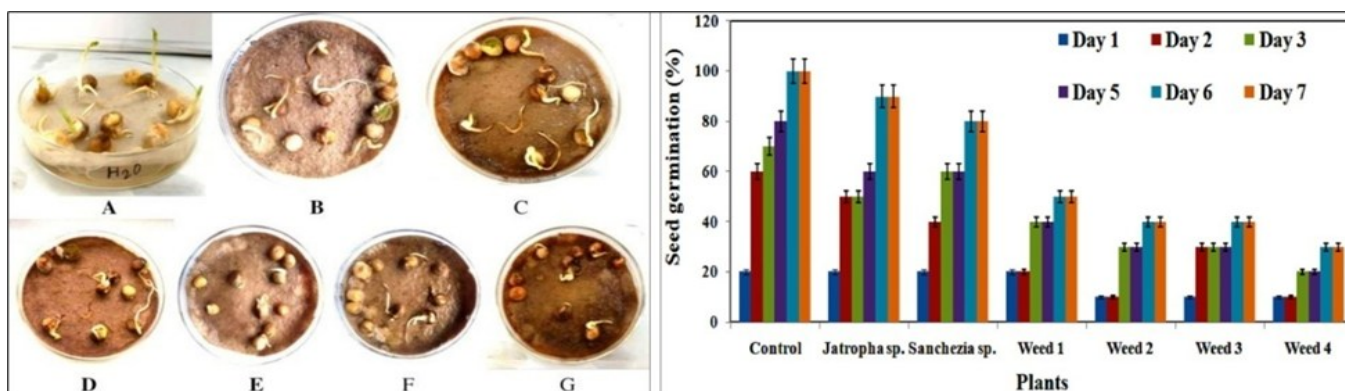


Fig. 3. Seed germination of *Pisum sativum* against six weed extract where (A) Control; (B) *Jatropha* sp.; (C) *Sanchezia* sp.; (D) *Cayratia* sp.; (E) *Mikania* sp.; (F) *Acalypha* sp.; (G) *Croton* sp. up to seven days.

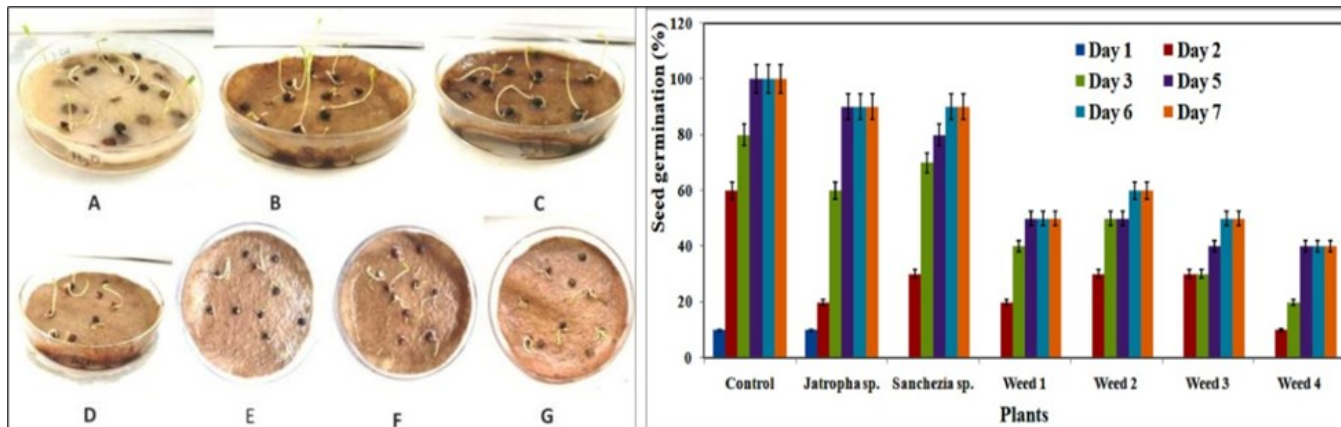


Fig. 4. Seed germination of *Lens culinaris* against weeds extract where (A) Control; (B) *Jatropha* sp.; (C) *Sanchezia* sp.; (D) *Cayratia* sp.; (E) *Mikania* sp.; (F) *Acalypha* sp.; (G) *Croton* sp. up to seven days.

In vitro assay on vigor index

Vigour index (VI) of all the seedlings belong to different pulses were calculated and it was found that the VI of control unit varies significantly in comparison to the plants watered with weed extracts which are shown in Fig. 5. On the seventh day the VI of *C. arietinum* (chickpea) under control unit was 1320, 762.4 in *Jatropha* sp., 663.2 in *Sanchezia* sp., 221.5 in *Cayratia* sp., 121.5 in *Mikania* sp., 175.6 in *Acalypha* sp. and 194.5 in *Croton* sp. extracts. For the plant of *V. mungo* (black gram), VI of control unit was 2073, 983 for *Jatropha* sp., 905.43 for *Sanchezia* sp., 342.26 for *Cayratia* sp., 202.31 for *Mikania* sp., 303.51 for *Acalypha* sp. and 194.5 for *Croton* sp. In the case of *P. sativum* (pea), the VI was found to be 972 for control, 513 for *Jatropha* sp., 388.8 for *Sanchezia* sp., 169.5 for *Cayratia* sp., 99.6 for *Mikania* sp., 128 for *Acalypha* sp., and 78 for *Croton* sp. Calculation shows that the *Lens culinaris* (lentil) under control has the VI of 1132, *Jatropha* sp. has 665.1, *Sanchezia* sp. has 437.4, *Cayratia* sp. has 243, *Mikania* sp. has 203.4, *Acalypha* sp. 194.5 and *Croton* sp. has the VI of 151.6.

Effect on shoot growth of plants grown in vitro plate experiment

At the end of day seven, the seeds grown with the influence of different weeds have shown a little difference with each other and a greater difference with control unit which is shown in Fig. 6. *Cicer arietinum* plant has grown its shoot to an extent of 6.23 cm in control unit, 4.6 cm in *Jatropha* sp., 4.33 cm in *Sanchezia* sp., 2.13 cm in *Cayratia* sp., 1.76 cm in *Mikania* sp., 2.3 cm in *Acalypha* sp. and 1.9 cm in *Croton* sp. extracts. It was measured that *V. mungo* has grown its shoot to the length of 16.63 cm in the control unit, 6.8 cm in *Jatropha* sp., 6.63 cm in *Sanchezia* sp., 3.63 in *Cayratia* sp., 3.6 cm in *Mikania* sp., 3.83 cm in *Acalypha* sp. and 3.66 cm in *Croton* sp. extracts. *Pisum sativum* plant has extended its shoot to 4.06 cm in control unit, 2.5 cm in *Jatropha* sp., 2.3 cm in *Sanchezia* sp., 1.83 cm in *Cayratia* sp., 1.46 cm in *Mikania* sp., 1.6 cm in *Acalypha* sp. and 1.3 cm in *Croton* sp. extracts. *Lens culinaris* plant has shoot to an extent of 8.06 cm in control unit, 4.63 cm in *Jatropha* sp., 5.23 cm in *Sanchezia* sp., 2.96 cm in *Cayratia* sp., 2.46 cm in *Mikania* sp., 2.86 cm in

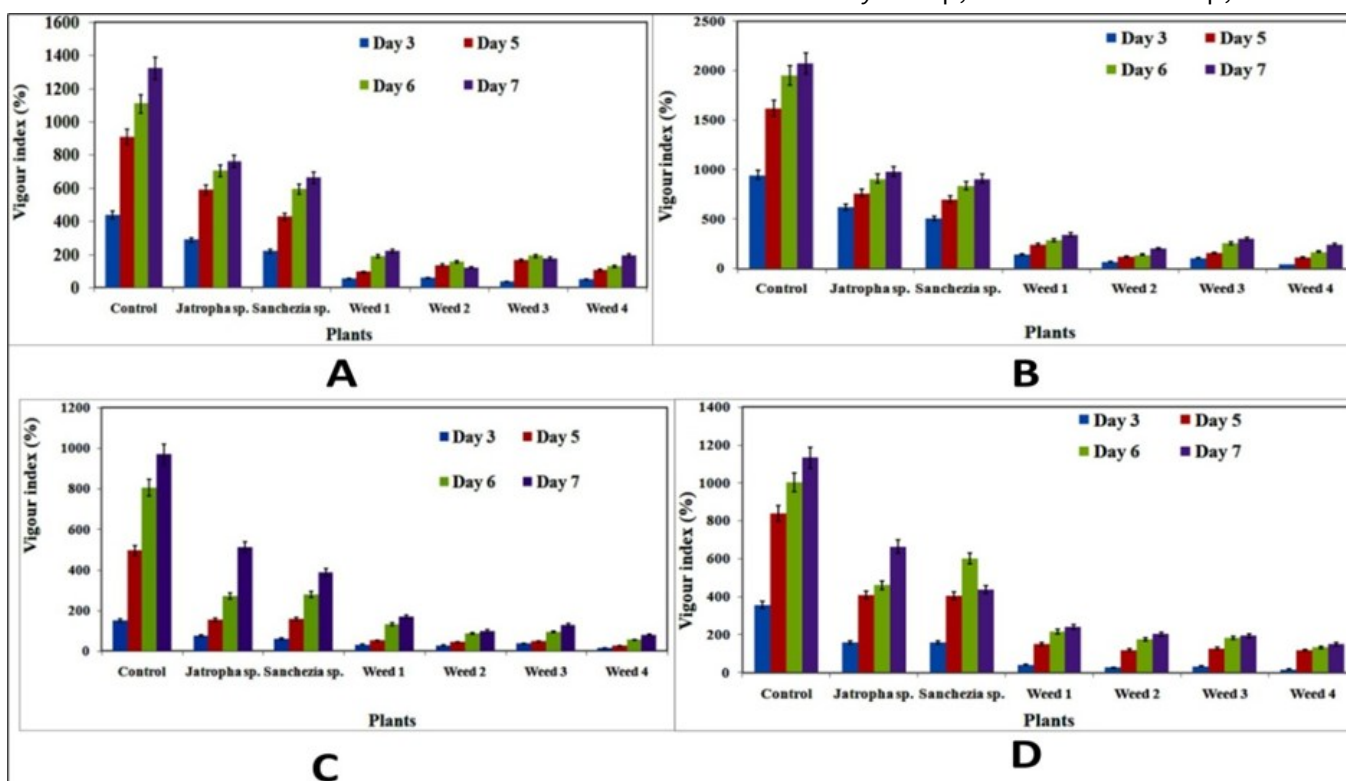


Fig. 5. Vigor index of (A) *Cicer arietinum*; (B) *Vigna mungo*; (C) *Pisum sativum*; (D) *Lens culinaris* against six weeds extract up to seven days.

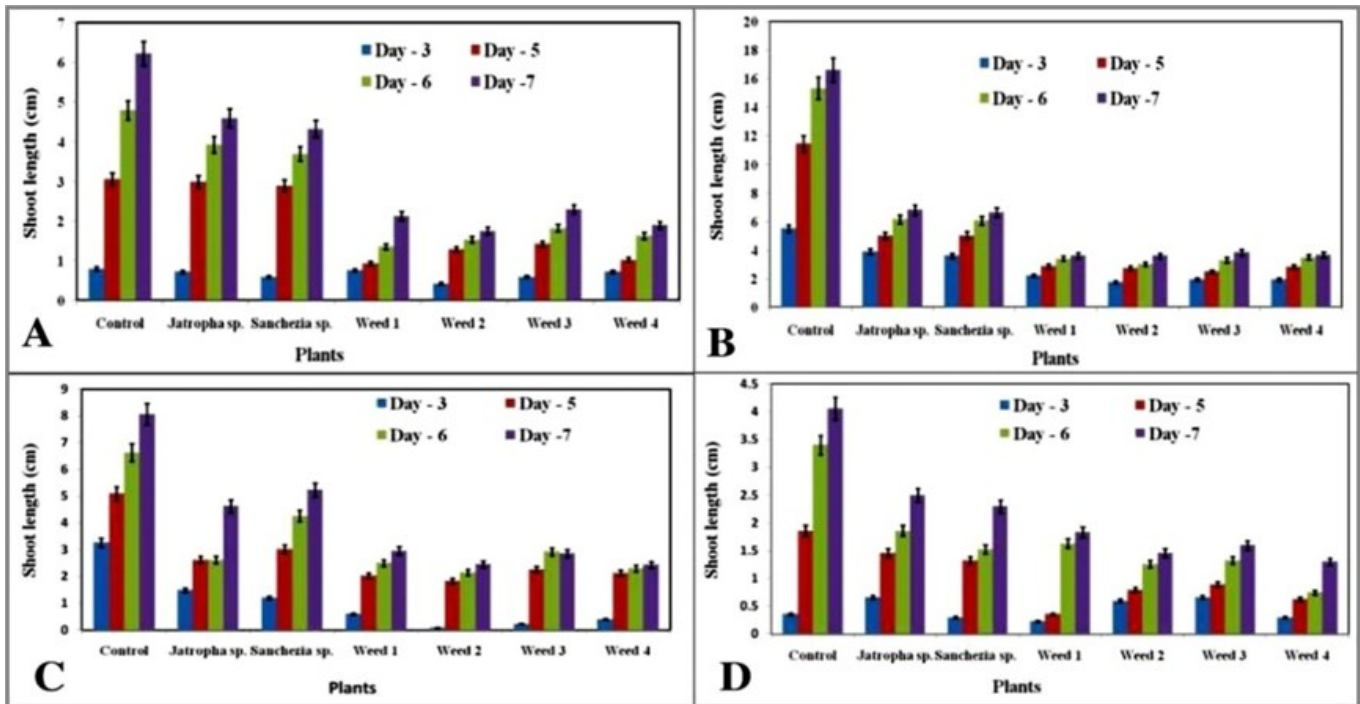


Fig. 6. Shoot length growth of (A) *Cicer arietinum*; (B) *Vigna mungo*; (C) *Pisum sativum*; (D) *Lens culinaris* against allelochemicals extracted from *Jatropha* sp., *Sanchezia* sp., *Cayratia* sp., *Mikania* sp., *Acalypha* sp., and *Croton* sp. for seven days.

Acalypha sp. and 2.43 cm in *Croton* sp. extracts.

Effect on root growth in *in vitro* plate experiment

A significant change has been observed in the length of roots and shoot of different pulses within and without the influence of different weeds. *Cicer arietinum* has grown 7 cm in control unit, 4.93 cm in *Jatropha* sp., and 3.96 in *Sanchezia* sp., 2.6 cm in *Cayratia* sp., 2.3 cm in *Mikania* sp., 2.09 cm in *Acalypha* sp. and 1.99 cm in *Croton* sp. extract's influence. *Vigna mungo* has

shown the growth of 4.1 cm in control, 3.03 cm in *Jatropha* sp., 2.99 cm in *Sanchezia* sp., 1.66 cm in *Cayratia* sp., 0.7 in *Mikania* sp., 1.33 cm *Acalypha* sp. and 0.96 cm in *Croton* sp. extract. *Pisum sativum* plant has extended its root to 5.66 cm in control unit, 3.2 cm in *Jatropha* sp., 2.56 cm in *Sanchezia* sp., 1.56 cm in *Cayratia* sp., 1.03 cm in *Mikania* sp., 1.6 cm in *Acalypha* sp. and 1.3 cm in *Croton* sp. extracts. It was seen that *L. culinaris* plant has root to an extent of 3.26 cm in control unit, 2.76 cm in *Jatropha*, 2.6 cm in *Sanchezia*, 1.9 cm

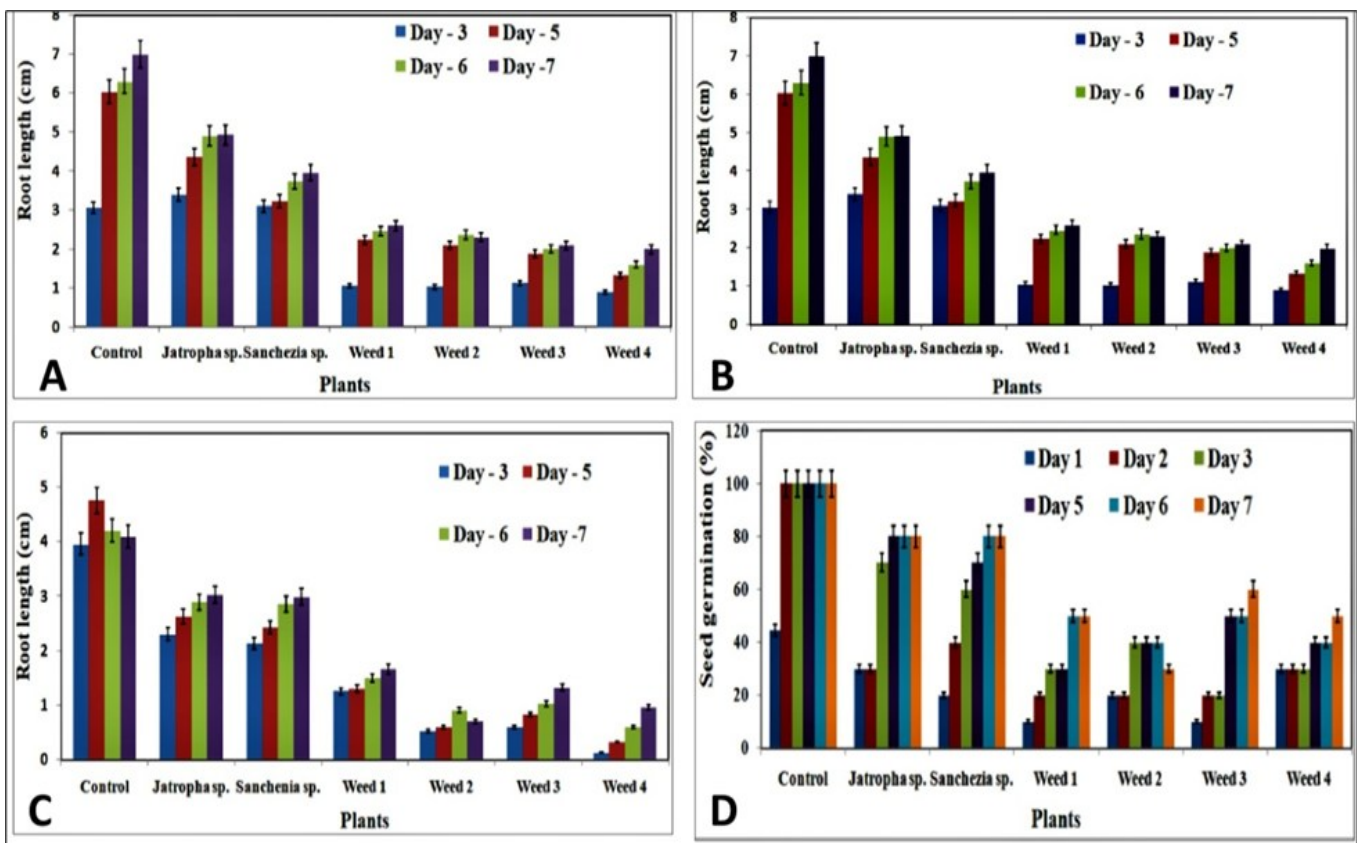


Fig. 7. Growth of root of (A) *Cicer arietinum*; (B) *Vigna mungo*; (C) *Pisum sativum*; (D) *Lens culinaris* against allelochemicals of *Jatropha* sp., *Sanchezia* sp., *Cayratia* sp., *Mikania* sp., *Acalypha* sp., and *Croton* sp. for seven days.

in *Cayratia* sp., 0.93 cm in *Mikania* sp., 1.03 cm in *Acalypha* sp. and 1.36 cm in *Croton* sp. extracts.

Fresh weight of shoot and root in *in vitro* plate experiment

Fresh biomass of *C. arietinum* shoot was observed to be 0.99 g in control, 0.27 g in *Jatropha*, 0.24 g in *Sanchezia* sp., 0.17 g in *Cayratia* sp., 0.09 g *Mikania* sp., 0.1 g *Acalypha* sp. and 0.14 g in *Croton* sp. extract. Fresh biomass of root of *C. arietinum* in control unit is 0.76 g, 0.24 g in *Jatropha* sp., 0.31 g in *Sanchezia* sp., 0.3 g in *Cayratia* sp., 0.17 g in *Mikania* sp., 0.18 g *Acalypha* sp. and 0.21 g in *Croton* sp. extracts. *Vigna mungo* has the shoot biomass of 0.27 g in control, 0.24 g in *Jatropha* sp., 0.21 g in *Sanchezia* sp., 0.08 g in *Cayratia* sp., 0.05 g *Mikania* sp., 0.05 g *Acalypha* sp. and 0.09 g in *Croton* sp. extract. *Vigna mungo* roots were observed to be 0.24 g in control, 0.24 g in *Jatropha* sp., 0.21 g in *Sanchezia* sp., 0.06 g in *Cayratia* sp., 0.05 g *Mikania* sp., 0.07 g *Acalypha* sp. and 0.18 g in *Croton* sp. extract. *Pisum sativum* shoots are measured to be 0.55 g in control, 0.36 g in *Jatropha* sp., 0.32 g in *Sanchezia* sp., 0.23 g in *Cayratia* sp., 0.15 g *Mikania* sp., 0.13 g *Acalypha* sp. and 0.14 g in *Croton* sp. extract. *Pisum sativum* roots were measured to be 0.48 g in control, 0.25 g in *Jatropha* sp., 0.14 g in *Sanchezia* sp., 0.14 g in *Cayratia* sp., 0.14 g *Mikania* sp., 0.14 g *Acalypha* sp. and 0.18 g in *Croton* sp. extract. Weight of *L. culinaris* shoots were measured to be 0.19 g in control, 0.18 g in *Jatropha* sp., 0.16 g in *Sanchezia*, 0.12 g in *Cayratia* sp., 0.1 g *Mikania* sp., 0.09 g *Acalypha* sp. and 0.11 g in *Croton* sp. extract. *Lens culinaris* roots have the biomass of 0.06 g in control, 0.04 g in *Jatropha* sp., 0.03 g in *Sanchezia* sp., 0.04 g in *Cayratia* sp., 0.03 g *Mikania* sp., 0.03 g *Acalypha* sp. and 0.05 g in *Croton* sp. extract which is shown in Fig. 8.

In vitro dry weight of shoot and root

Dry biomass of *C. arietinum* shoot was observed to be 0.52 g in control, 0.14 g in *Jatropha* sp., 0.12 g in *Sanchezia* sp., 0.08 g in *Cayratia* sp., 0.04 g *Mikania* sp., 0.04 g *Acalypha* sp. and 0.07 g in *Croton* sp. extract. For the plant of *Cicer arietinum* the dry weight of roots is 0.04 g in control, 0.03 g in *Jatropha* sp., 0.02

g in *Sanchezia* sp., 0.02 g in *Cayratia* sp., 0.01 g in *Mikania* sp., 0.01 g in *Acalypha* sp. and 0.01 g in *Croton* sp. extracts. *Vigna mungo* has the shoot dry biomass of 0.13 g in control, 0.08 g in *Jatropha* sp., 0.09 g in *Sanchezia* sp., 0.04 g in *Cayratia* sp., 0.02 g *Mikania* sp., 0.02 g *Acalypha* sp. and 0.03 g in *Croton* sp. extract. *Vigna mungo* has a dry root biomass of 0.11 g in control, 0.12 g in *Jatropha* sp., 0.1 g in *Sanchezia* sp., 0.02 g in *Cayratia* sp., 0.02 g *Mikania* sp., 0.01 g *Acalypha* sp. and 0.03 g in *Croton* sp. extract. *Pisum sativum* is measured to have shoot dry mass of 0.26 g in control, 0.15 g in *Jatropha* sp., 0.14 g in *Sanchezia* sp., 0.11 g in *Cayratia* sp., 0.06 g *Mikania* sp., 0.08 g *Acalypha* sp. and 0.06 g in *Croton* sp. extract. *Pisum sativum* roots weighed 0.24 g in control, 0.13 g in *Jatropha* sp., 0.07 g in *Sanchezia* sp., 0.1 g in *Cayratia* sp., 0.06 g *Mikania* sp., 0.06 g *Acalypha* sp. and 0.09 g in *Croton* sp. extract. Dry weight of *Lens culinaris* shoots were measured to be 0.08 g in control, 0.08 g in *Jatropha* sp., 0.06 g in *Sanchezia* sp., 0.06 g in *Cayratia* sp., 0.05 g *Mikania* sp., 0.04 g *Acalypha* sp. and 0.05 g in *Croton* sp. extract. *Lens culinaris* roots have dry biomass of 0.04 g in control, 0.03 g in *Jatropha* sp., 0.02 g in *Sanchezia* sp., 0.02 g in *Cayratia* sp., 0.02 g *Mikania* sp., 0.01 g *Acalypha* sp. and 0.01 g in *Croton* sp. extract which is shown in Fig. 9.

In vivo assay on seed germination

Like the observation done for the *in vitro* experiment, the same was observed for the plantlets growing in pots, the result we get that says, same as the *in vitro* grown plants the rate of germination has decreased in all the plants having the influence of the weeds. The germination rate of the seventh day is described below. In the plant of *Lens culinaris*, the rate of germination has decreased from 100 % in control, 70 % in *Jatropha* sp., 60 % in *Sanchezia* sp., 50 % in *Cayratia* sp. and *Mikania* sp., 40 % in *Croton* sp. to 30 % in *Acalypha* sp. extracts. In the plants of *Vigna mungo*, the weeds have declined the rate of germination, 100 % in control and *Jatropha* sp. extract, 90 % in *Sanchezia* sp. extract, 70 % in *Cayratia* sp. extract, 60 % in *Mikania* sp., 40 % in *Acalypha* sp. and *Croton* sp. extracts. For the plant of *P. sativum*, the rate of

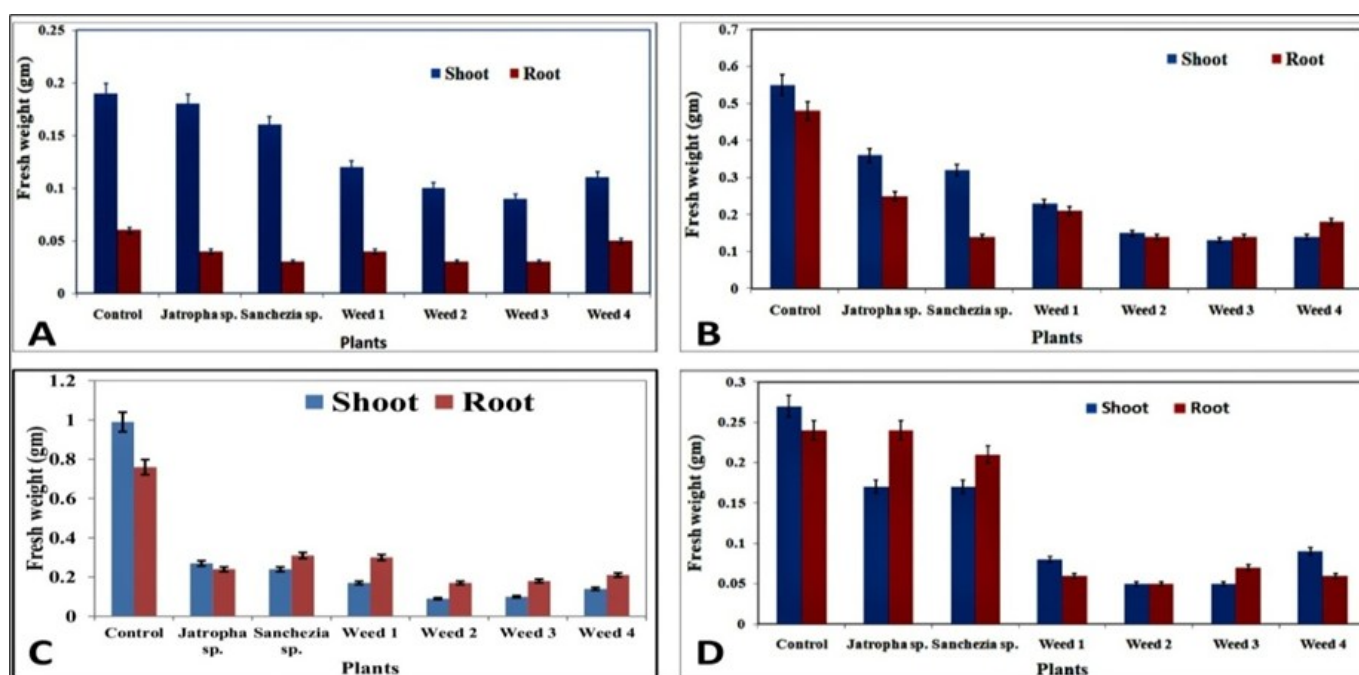


Fig. 8. Shoot and root fresh weight of (A) *Cicer arietinum*; (B) *Vigna mungo*; (C) *Pisum sativum*; (D) *Lens culinaris* against allelochemicals of *Jatropha* sp., *Sanchezia* sp., *Cayratia* sp., *Mikania* sp., *Acalypha* sp., and *Croton* sp. for seven days.

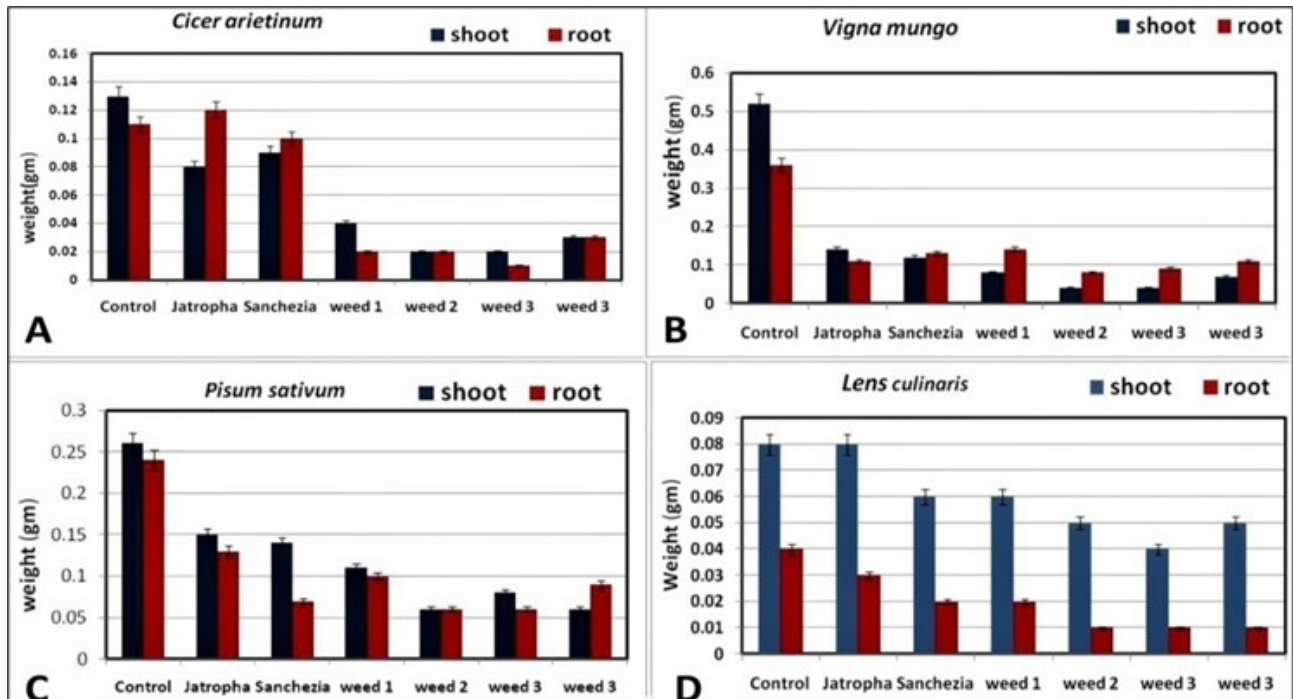


Fig. 9. Shoot and root dry weight of in (A) *Cicer arietinum*; (B) *Vigna mungo*; (C) *Pisum sativum*; (D) *Lens culinaris*.

germination has decreased from 100 % in control, 80 % both in *Jatropa* sp. and *Sanchezia* sp. extract, 50 % in *Cayratia* sp. extract and 40 % in *Mikania* sp. and *Croton* sp. extracts to 30 % in *Acalypha* sp. extract. In the case of *Cicer arietinum*, the weeds have retarded the rate of germination from 100 % in control, 75 % in *Jatropa* sp. extract, 62.5 % in *Sanchezia* sp., 50 % in *Mikania* sp. and *Croton* sp. to 37.5 % in *Cayratia* sp. and *Acalypha* sp. extracts which are shown in Fig. 10 and 11.

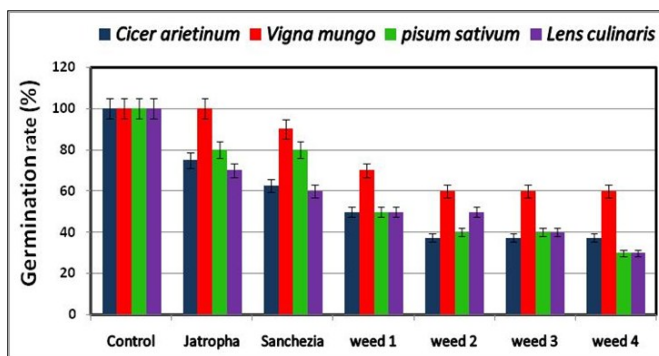


Fig. 10. Percentage of seed germination of different pulses in (A) Control; (B) *Jatropa* sp.; (C) *Sanchezia* sp.; (D) *Cayratia* sp.; (E) *Mikania* sp.; (F) *Acalypha* sp.; (G) *Croton* sp. at day 7.

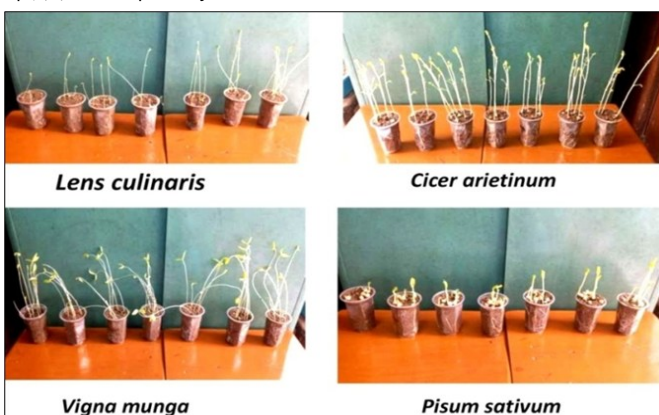


Fig. 11. Photographs showing the growth of pulses in plastic pots over seven days.

In vivo assay on vigor index (VI)

Vigour index (VI) results of all the plants grown during the time of the experiment which is shown in Fig. 12. In case of *C. arietinum*, VI of control set is 5230 %, 3427.5 % for *Jatropa* sp., 2106.25 % for *Sanchezia* sp., 1136.25 % for *Cayratia* sp., 1620 % for *Mikania* sp., 952.5 % for *Acalypha* sp. and 1570 % for *Croton* sp. influenced plants. *Vigna mungo* plants had the VI of 2150 % at control, 3160 % at *Jatropa* sp., 2412 % in *Sanchezia* sp., 1722 % at *Cayratia* sp., 1014 % at *Mikania* sp., 942 % *Acalypha* sp. and 1134 % at *Croton* sp. extracts. *P. sativum* plants showed the VI of 2360 % in control, 1496 % in *Jatropa* sp., 2232 % in *Sanchezia* sp., 1098 % in *Cayratia* sp., 912 % in *Mikania* sp., 378 % in *Acalypha* sp., 636 % in *Croton* sp. extracts. *Lens culinaris* were observed to have a VI of 2770 % in control, 1785 % in *Jatropa* sp., 1170 % in *Sanchezia* sp., 945 % in *Cayratia* sp., 775 % in *Mikania* sp., 453 % in *Acalypha* sp., 820 % in *Croton* sp.

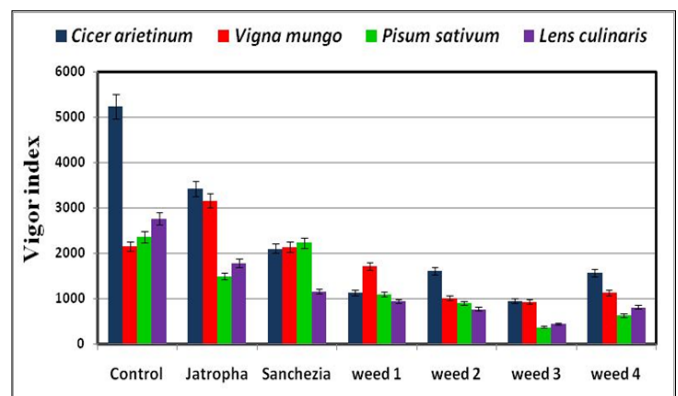


Fig. 12. Vigor index of different pulses in (A) Control; (B) *Jatropa* sp.; (C) *Sanchezia* sp.; (D) *Cayratia* sp.; (E) *Mikania* sp.; (F) *Acalypha* sp.; (G) *Croton* sp. for seven days.

Effect on shoot and root growth of plants grown in vivo pot experiment

Lengths of the roots and shoot were measured for all the varieties of pulses grown in the pots at the seventh day of experiment which is shown in Fig. 13 and 14. *Cicer arietinum*

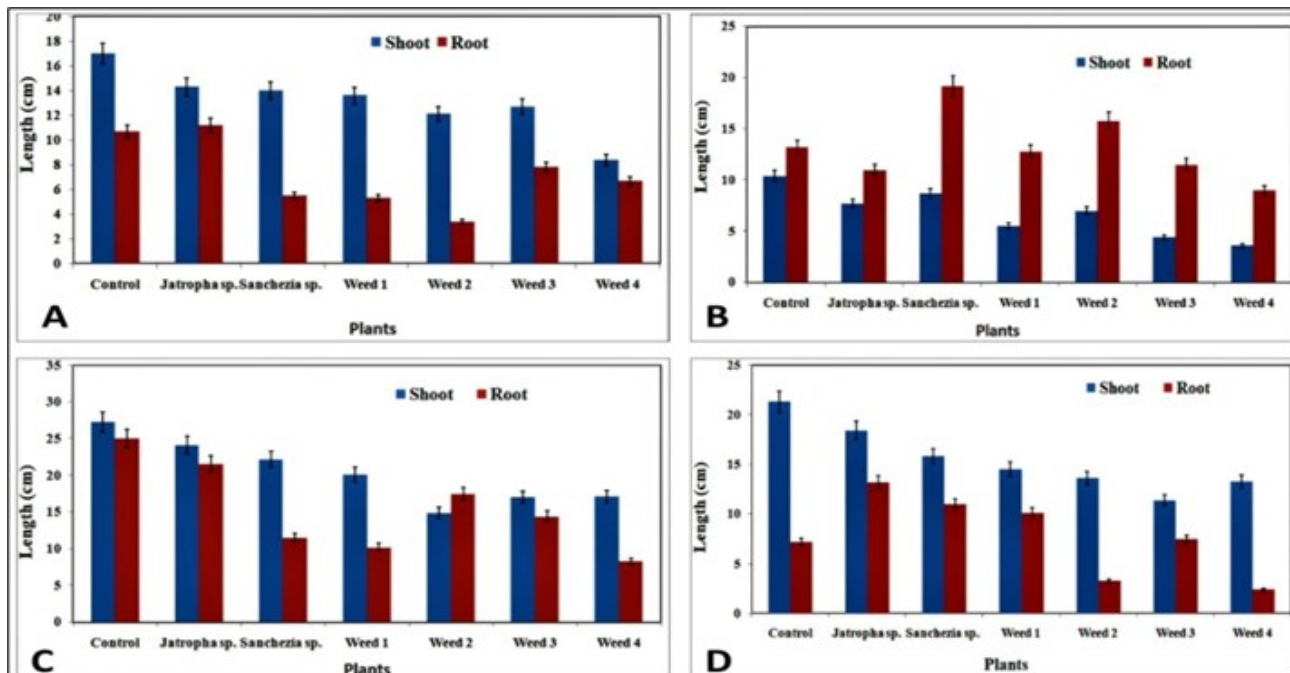


Fig. 13. Growth of shoot and root length of (A) *Cicer arietinum*; (B) *Vigna mungo*; (C) *Pisum sativum*; (D) *Lens culinaris* against allelochemicals of *Jatropa* sp., *Sanchezia* sp., *Cayratia* sp., *Mikania* sp., *Acalypha* sp., and *Croton* sp. for seven days.

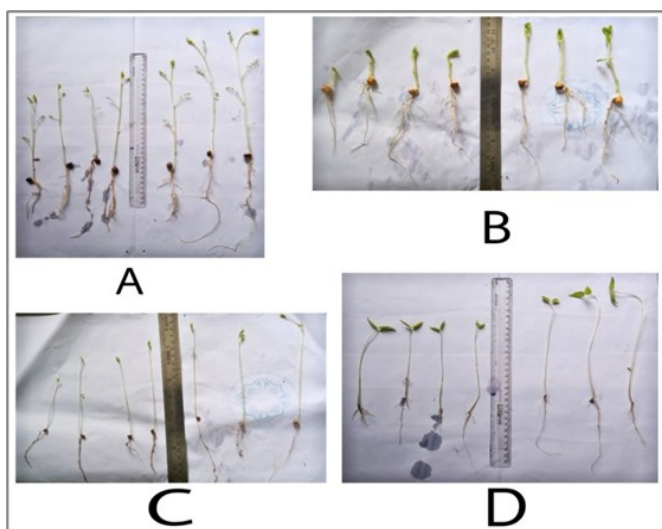


Fig. 14. Length of root and shoot of (A) *Cicer arietinum*; (B) *Vigna mungo*; (C) *Pisum sativum*; (D) *Lens culinaris* grown on different weed extracts under *in vivo* pot experiment.

has developed its shoots to the extent of 27.3 cm in control, 24.1 cm in *Jatropa* sp., 22.2 cm in *Sanchezia* sp., 20.1 cm in *Cayratia* sp., 14.9 cm in *Mikania* sp., 17 cm in *Acalypha* sp. and 17.1 cm in *Croton* sp. extract. *Cicer arietinum* plants show the length of root in control to be 25 cm, 21.6 cm in *Jatropa* sp., 11.5 cm in *Sanchezia* sp., 10.2 cm in *Cayratia* sp., 17.5 cm in *Mikania* sp., 14.4 cm in *Acalypha* sp., 8.3 cm in *Croton* sp. extract. *Vigna mungo* has grown its shoots to the length of 21.3 cm in control, 18.4 cm in *Jatropa* sp., 15.8 cm in *Sanchezia* sp., 14.5 cm in *Cayratia* sp., 13.6 cm in *Mikania* sp., 11.4 cm in *Acalypha* sp. and 13.3 cm in *Croton* sp. extract. It has been observed to be grown its root 7.2 cm in control, 13.2 cm in *Jatropa* sp., 11 cm in *Sanchezia* sp., 10.1 cm in *Cayratia* sp., 3.3 cm in *Mikania* sp., 7.5 cm in *Acalypha* sp. and 2.4 cm in *Croton* sp. extract. Plant of *P. sativum* was observed to extend its shoots to 10.4 cm in control, 7.7 cm in *Jatropa* sp., 8.7 cm in *Sanchezia* sp., 5.5 cm in *Cayratia* sp., 7 cm in *Mikania* sp., 4.4 cm in *Acalypha* sp. and 3.6 cm in *Croton* sp. extract. *Pisum sativum* roots appeared to extend to 23.2 cm in control, 11 cm in *Jatropa* sp., 19.2 cm in

Sanchezia sp., 12.8 cm in *Cayratia* sp., 15.8 cm in *Mikania* sp., 11.5 cm in *Acalypha* sp. and 9 cm in *Croton* sp. extract. In the plant of *L. culinaris*, the shoots have grown to an extent of 17 cm in control unit, 14.3 cm in *Jatropa* sp., 14 cm in *Sanchezia* sp., 13.6 cm in *Cayratia* sp., 12.1 cm in *Mikania* sp., 12.7 cm in *Acalypha* sp. and 8.4 cm in *Croton* sp. *Lens culinaris* plant has root to an extent of 10.7 cm in control unit, 11.2 cm in *Jatropa* sp., 5.5 cm in *Sanchezia* sp., 5.3 cm in *Cayratia* sp., 3.4 cm in *Mikania* sp., 7.8 cm in *Acalypha* sp. and 6.7 cm in *Croton* sp. extracts.

Fresh weight of shoot and root of plants grown *in vivo*

Biomass of *C. arietinum* shoots were measured to be 0.84 g in control, 0.6 g in *Jatropa* sp., 0.75 g in *Sanchezia* sp., 0.58 g in *Cayratia* sp., 0.29 g in *Mikania* sp., 0.36 g in *Acalypha* sp. and 0.34 g in *Croton* sp. extract. Biomass of roots of *C. arietinum* grown in pot, in control unit is 0.41 g, 0.32 g in *Jatropa* sp., 0.4 g in *Sanchezia* sp., 0.41 g in *Cayratia* sp., 0.25 g in *Mikania* sp., 0.31 g in *Acalypha* sp. and 0.14 g in *Croton* sp. extracts. The biomass of *V. mungo* shoots is 0.26 g in control, 0.23 g in *Jatropa* sp., 0.16 g in *Sanchezia* sp., 0.16 g in *Cayratia* sp., 0.1 g in *Mikania* sp., 0.16 g in *Acalypha* sp. and 0.14 g in *Croton* sp. extract. Biomass of *V. mungo* roots were observed to be 0.06 g in control, 0.02 g in *Jatropa* sp., 0.03 g in *Sanchezia* sp., 0.02 g in *Cayratia* sp., 0.02 g in *Mikania* sp., 0.04 g in *Acalypha* sp. and 0.03 g in *Croton* sp. extract. Shoot biomass of *P. sativum* is measured to be 0.55 g in control, 0.36 g in *Jatropa* sp., 0.32 g in *Sanchezia* sp., 0.23 g in *Cayratia* sp., 0.15 g in *Mikania* sp., 0.13 g in *Acalypha* sp. and 0.14 g in *Croton* sp. extract. *Pisum sativum* roots weighed 0.3 g in control, 0.3 g in *Jatropa* sp., 0.19 g in *Sanchezia* sp., 0.32 g in *Cayratia* sp., 0.36 g in *Mikania* sp., 0.17 g in *Acalypha* sp. and 0.26 g in *Croton* sp. extract. Weight of *L. culinaris* shoots were measured to be 0.19 g in control, 0.18 g in *Jatropa* sp., 0.16 g in *Sanchezia* sp., 0.12 g in *Cayratia* sp., 0.1 g in *Mikania* sp., 0.09 g in *Acalypha* sp. and 0.11 g in *Croton* sp. extract. *Lens culinaris* roots have biomass of 0.04 g in control, 0.05 g in *Jatropa* sp., 0.03 g in *Sanchezia* sp., 0.02 g in *Cayratia* sp., 0.02 g in *Mikania* sp., 0.02 g in *Acalypha* sp. and 0.34 g in *Croton* sp. extract which is shown in Fig. 15.

Dry weight of shoot and root of plants grown *in vivo*

Dry biomass of shoot of *C. arietinum* was measured to be 0.46 g in control, 0.33 g in *Jatropha* sp., 0.39 g in *Sanchezia* sp., 0.22 g in *Cayratia* sp., 0.12 g *Mikania* sp., 0.14 g *Acalypha* sp. and 0.15 g in *Croton* sp. extract. Dry roots of *C. arietinum* were measured to be 0.21 g in control, 0.32 g in *Jatropha* sp., 0.19 g in *Sanchezia* sp., 0.2 g in *Cayratia* sp., 0.1 g *Mikania* sp., 0.12 g *Acalypha* sp. and 0.05 g in *Croton* sp. extract. *Vigna mungo* shoots have dry biomass of 0.14 g in control, 0.09 g in *Jatropha* sp., 0.07 g in *Sanchezia* sp., 0.06 g in *Cayratia* sp., 0.04 g *Mikania* sp., 0.06 g *Acalypha* sp. and 0.06 g in *Croton* sp. extract. *Vigna mungo* roots have a dry biomass of 0.06 g in control, 0.04 g in *Jatropha* sp., 0.03 g in *Sanchezia* sp., 0.02 g in *Cayratia* sp., 0.01 g in *Mikania micrantha*, 0.03 g in *Acalypha* sp. and 0.01 g in *Croton* sp. extract. *Pisum sativum* shoots appear to have dry mass of 0.28 g in control, 0.17 g in *Jatropha* sp., 0.15 g in *Sanchezia* sp., 0.14 g in *Cayratia* sp., 0.15 g *Mikania* sp., 0.05 g *Acalypha* sp. and 0.1 g in *Croton* sp. extract. *Pisum sativum* roots were measured to have dry mass of 0.01 g in control, 0.01 g in *Jatropha* sp., 0.1 g in *Sanchezia* sp., 0.13 g in *Cayratia* sp., 0.15 g *Mikania* sp., 0.09 g in *Acalypha* sp. and 0.12 g in *Croton* sp. extract. Dry weight of *L. culinaris* shoots were measured to be 0.05 g in control, 0.04 g in *Jatropha* sp., 0.04 g in *Sanchezia* sp., 0.03 g in *Cayratia* sp., 0.02 g *Mikania* sp., 0.01 g *Acalypha* sp. and 0.01 g in *Croton* sp. extract. The dry weight of *L. culinaris* roots were measured to be 0.02 g in control, 0.02 g in *Jatropha* sp., 0.02 g in *Sanchezia* sp., 0.02 g in *Cayratia* sp., 0.01 g *Mikania* sp., 0.01 g *Acalypha* sp. and 0.01 g in *Croton* sp. extract which is shown in Fig. 16.

Discussion

Growth and yield of plants goes down with the increase of the weed material (21, 22). The experiment conducted by us clearly showed that the weeds like the four used by us i.e., *Jatropha* sp., *Sanchezia* sp., *Cayratia* sp., *Mikania* sp., *Acalypha*

sp. and *Croton* sp. have some allelopathic effects on the crop plants pulses like *C. arietinum*, *V. mungo*, *P. sativum* and *L. culinaris* which significantly reduce germination and growth of the seeds and seedlings. All the parameters used to assess the growth of different pulses in the presence of weed extracts showed significant differences compared to the control set without weed extracts. One of the key parameters for comparison is the germination rate, which represents the proportion of seeds that germinate out of the total number of seeds sown. Germination of seeds is the basis of plant development that directly impacts the growth and survival of plants (23-25). Allelopathic research typically includes seed germination test as a biological detection method (26). The germination rate (GR) is largely affected by the allelopathic effects of weeds. In the *in vitro* experiment, the maximum GR is observed in the control set of all the types of pulses (100 %) and lowest is observed in the *C. arietinum* (chickpea) with the influence of *Mikania* sp. extract and *P. sativum* (pea) with the influence of *Croton* sp. extract (30 %). In the pot experiment, it was again observed that the maximum GR is given by the control unit of all the types of pulses (100 %) and minimum GR is provided by *Acalypha* sp. extracts in both *P. sativum* and *L. culinaris* (30 %). This result justifies the work of Nouri and colleagues which states that the exposure of seeds of any species to allelochemicals leads to a great decline in the germination of those seeds (27). In contrast, the minimum rot percentage is shown by the control sets of all types of pulses, the maximum rot percentage is shown by *C. arietinum* under the influence of *Mikania* sp. extracts and *P. sativum* (70 %) in the *in vitro* experiment. In case of pot experiment again the minimum perishing percentage is given by the control unit of all the pulses where the maximum is given by *Acalypha indica* extracts in both *P. sativum* and *L. culinaris* (70 %). This proves that allelopathic effects of weeds not only retards the rate of germination but also perish the seedlings. The shoot and root

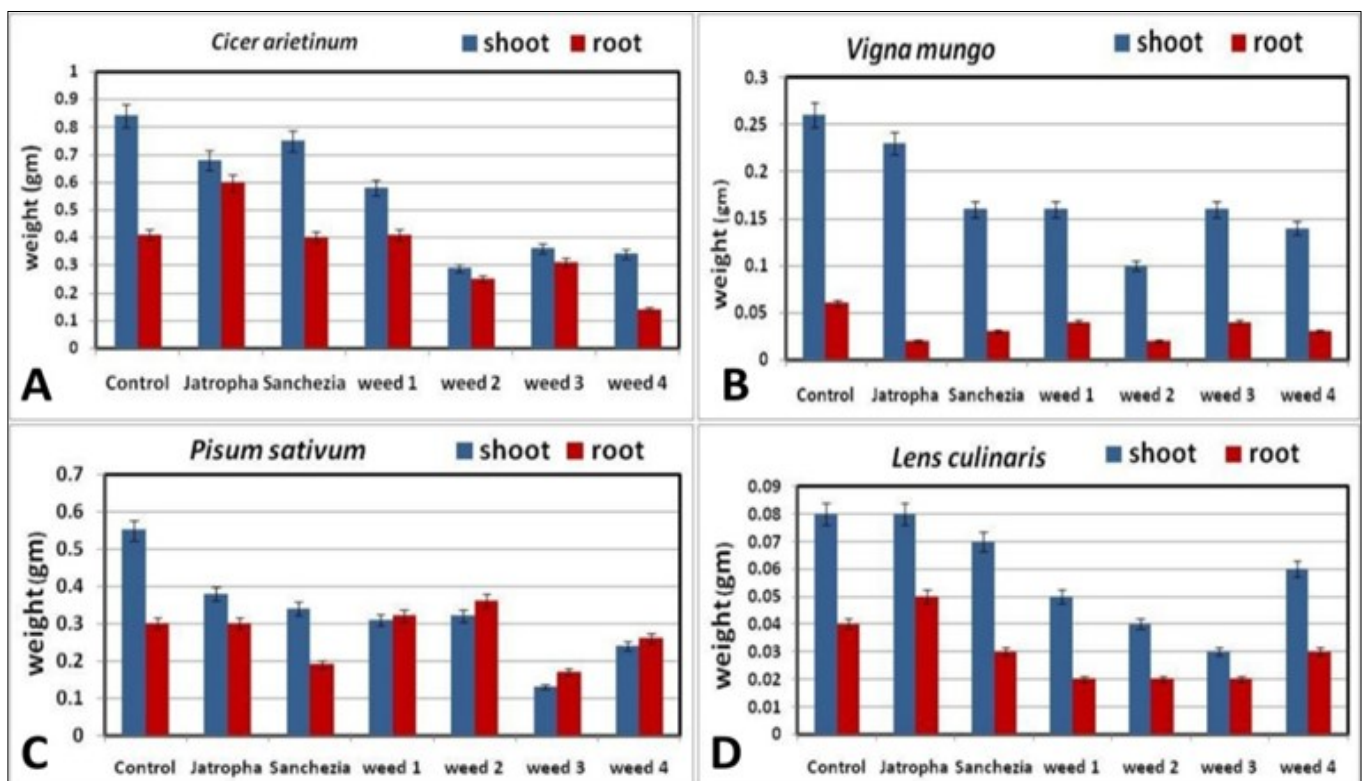


Fig. 15. Shoot and root fresh weight of (A) *Cicer arietinum*; (B) *Vigna mungo*; (C) *Pisum sativum*; (D) *Lens culinaris*.

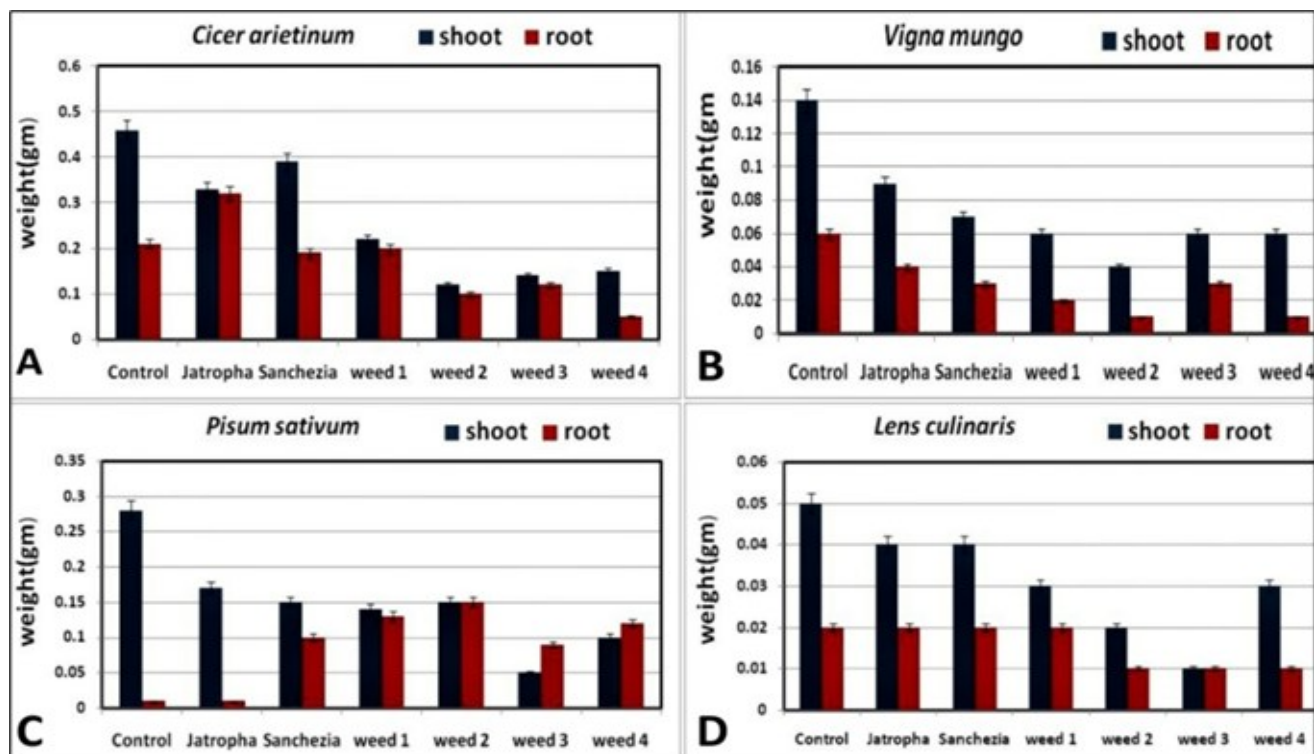


Fig. 16. Shoot and root dry weight of (A) *Cicer arietinum*; (B) *Vigna mungo*; (C) *Pisum sativum*; (D) *Lens culinaris*.

length of the plants are also majorly affected by the allelochemicals of the weeds. In the *in vitro* experiment the longest root is observed in *C. arietinum* (chickpea) under the control unit (7 cm) and shortest is observed in *V. mungo* (black gram) under the influence of *Mikania micrantha* extract (0.7 cm). In the pot experiment, the longest root is observed in *C. arietinum* (chickpea) under the control unit (25 cm) and shortest is observed in *V. mungo* (black gram) under the influence of *Croton* sp. extract (2.4 cm). In the case of shoot length of *in vitro* experiment, the longest shoot is found to be in the control unit of *V. mungo* (16.63 cm) and the shortest is in *P. sativum* (pea) under the influence of *Croton* sp. (1.3 cm). While the pot experiment shows maximum shoot length in *C. arietinum* (chickpea) in control unit (27.3 cm) and minimum in *P. sativum* (pea) under the influence of *Croton* sp. (3.6 cm). The study of length of shoot and root explained to us that the influence of weed can drastically reduce the length of root and shoot of the plant making the plant short and weak limiting the growth of the plant to its fullest. For the parameter of seed VI, the highest value can be obtained in *V. mungo* (black gram) within the control set (2073) and the lowest value can be obtained in *P. sativum* (pea) with the influence of *Croton* sp. (78) for the *in vitro* experiment. In the case of pot experiments, maximum VI can be seen in *C. arietinum* within control (5230) and minimum VI can be obtained in *P. sativum* under the influence of *Acalypha* sp. extract (378). Yan and his co-workers presented a possible explanation to this phenomenon by finding that allelochemicals interfered with both the metabolism of plants and the activity of enzymes during seed germination, which may explain why seeds deteriorate and become less vigorous (28). This outcome is justified by several researches state that allelochemicals begins to affect the plant from the time of germination but the effect is stronger and more effective during the growth of the seedling (29-31). Measuring the fresh biomass of plants we get that in the *in vitro* experiment, the

maximum root fresh weight can be observed in *C. arietinum* (chickpea) under the control treatment (0.76 g) and lowest root fresh weight can be seen in *L. culinaris* (lentil) under the influence of *Acalypha* sp. extract (0.03 g). Highest fresh shoot biomass can be obtained from *C. arietinum* (chickpea) under control (0.99 g) and lowest can be obtained from *V. mungo* (black gram) under the influence of *Acalypha* sp. and *Mikania* sp. extracts (0.5 g). Recording the dry biomass of the plants we come to the statement that in the *in vitro* experiment maximum root dry weight can be observed in *C. arietinum* (chickpea) under the control treatment (0.36 g) and minimum in *L. culinaris* (lentil) under the influence of *Mikania* sp., *Acalypha* sp., *Croton* sp. and *V. mungo* under the influence of *Sanchezia* and *Acalypha Indica* (0.01 g). Now in the pot experiment, maximum root dry biomass is obtained from *C. arietinum* (chickpea) under the control treatment (0.21 g) and minimum is obtained from *L. culinaris* (lentil) under the influence of *Acalypha* sp., *Mikania* sp.; *V. mungo* under the influence of *Mikania* sp. and *Croton* sp.; and *C. arietinum* with the influence of *Mikania* sp. (0.01 g). This result satisfies the studies of Shaikat and Chowdhury which states that the dry and fresh weight of plants will reduce with increase in weed material that the plant is exposed to (32-36). The experimental reports match with some crop plants reported by Chowdhury for chili and tomato (37-39).

Conclusion

Conducting the experiment and recording all the data observed from the growing plants during the time of experiment can conclude that the leaf extracts of all the plant species can largely retard the germination and growth of the plant of interest or the pulse crops. Even the plants which are not commonly referred to as weeds also somewhat affect the germination and growth of the crops but not to the extent of the weed plants (*Cayratia* sp., *Mikania* sp., *Acalypha* sp., *Croton* sp.) as the destruction caused by the four weeds was

much higher than those plants (*Jatropha* sp. and *Sanchezia* sp.). These plants produce allelochemicals which are harmful for the crop, leads to the retardation in rate of germination and growth of the crops primarily promoting rotting of seeds and seedlings, all the above-mentioned characteristics of the weeds collectively referred as the allelopathic effect of weeds. Due to this very reason the growth of these kinds of weeds must be discarded for the sake of good germination of the desired seeds and healthy growth of the plant of interest. Allelopathic species can be manipulated for the sustainable management of weeds in different ways such as the introduction of an allelopathic crop into crop rotation schemes, the use of an allelopathic cover crop, or the identification, isolation and extraction of plant allelochemicals for the possible production of bioherbicides

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Authors' contributions

MB wrote the manuscript draft. SKC designed the review and improved the manuscript. DB reviewed and corrected the manuscript. KSC and TM collected the data. All authors read and approved the final manuscript.

Compliance with ethical standards

Conflict of interest: The authors have no apparent conflicts of interest.

Ethical issues: None

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