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Research Article

ALLELOPATHIC EVALUATION OF TWO MEDICINAL PLANTS USING FRACTIONATION GUIDED BIOASSAY

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ABSTRACT
Chlorophytum borivilianum Santapau & R.R.Fern and *Asparagus racemosus* Willd were analysed for their phytotoxicity. All the plant parts were screened for their allelopathic potential. In both the plants, root was most phytotoxic where in aqueous root extracts were found to inhibit Radish germination and growth. For fractionation guided bioassay, methanolic extract of roots were subjected to fractionation and fractions so obtained were analysed for their phytotoxicity against *Chloris barbata* (SW.) using agar bioassay. The most allelopathic fraction was analysed for its metabolites. In Chlorophytum, the chloroform fraction from root was the most allelopathic fraction, inhibiting Chloris germination and growth. Phytochemical analysis of the fraction could detect presence of terpenoid and phenolic compounds. In Asparagus, chloroform: methanol fraction was the most allelopathic of all three fractions and was suppressive to the germination and growth at all the applied rates. The fraction showed presence of compounds that were alkaloidal in nature. Thus, Chlorophytum offers allelopathy by water soluble metabolites that are terpenoids or phenolic in nature and Asperagus exhibits allelopathy through water soluble metabolites that may be alkaloids by nature. Fractionation guided bioassay followed by allelopathic analysis of both the medicinal plants gave promising results and aided in identifying the allelopathic potential of the two important medicinal plants. Detailed study on employing the plants as smother crop, rotation crop or companion crop may lead to natural weed mitigation alternative and encourage the medicinal plants cultivation on large scale.

Keywords: *Chlorophytum Borivilianum Santapau & R.R.Fern, Asparagus Racemosus Willd., Raphanus Sativus L., Bioassays, Fractionation, Phytochemical Analysis, Chloris Barbata Sw*

INTRODUCTION
Commercial demand of medicinal plants in pharmaceuticals is met either by collection from wild or by their cultivation. Number of medicinal plant species that are under cultivation as well as their scale of cultivation is very petite. One of the significant reasons behind this is higher inputs and low benefits that are involved in their cultivation. Increasing the benefits of medicinal plant cultivation by any means will lure more ventures into their cultivation. Exploring allelopathic potential of medicinal plant is one such value addition to the benefits of medicinal plants cultivation. “Allelopathy” an ecological phenomenon, is defined as a chemical interaction that may exist between various living organisms in an ecosystem as between plant-plant. Chemicals involved in exhibiting allelopathy are mostly secondary metabolites (Whittaker and Feeny, 1971) and are termed as allelochemicals. Medicinal plants are treasure houses of secondary metabolites and so it is more likely to screen the medicinal plants for allelopathic potential (Patel and Pandya, 2013). Allelopathic potential of the medicinal plants/ non medicinal plant can be utilized to eradicate agricultural weeds when grown as cover crop, rotational crop or as intercrop (Singh *et al.*, 2003; Khanh *et al.*, 2005) which can facilitate large scale production of medicinal plant crop in addition to production of the main crop. Cultivating them as intercrop with agricultural crop of interest will help to eradicate the weeds from the crop field. Increased benefits available by cultivating medicinal plants will also promote their cultivation on larger scale and this will help to meet the increasing pressure on existing demand of medicinal plants.
Large number of molecular discoveries especially in field of medicine and agriculture has been an inspiration, derived from existing natural metabolites. Medicinal plants are known to harbour large

number of active metabolites, screening them for allelochemical with a particular structure and corresponding targeted function is almost obvious. Identifying an allelopathic molecule from a medicinal plant may inspire a future herbicide. Thus, the current study aims to evaluate the allelopathic potential of two important medicinal plants such as *Chlorophytum borivilianum* Santapau & R.R.Fern and *Asparagus racemosus* Willd. Objective of the present study were 1) to identify most effective plant part 2) isolate and identify most effective chemical fraction and 3) to identify phytoconstituents of the most effective fractions. Radish *(Raphanus sativus* L.) was selected as a test plant for preliminary screening, owing to its high sensitivity, high germination rate. *Chloris barbata* Sw., a monocot weed plant was selected as test plant for fractionation guided bioassays.

MATERIALS AND METHODS

Collection of Plant Material: Material for both the medicinal plants was collected from the university arboretum (The M.S. University of Baroda, Baroda, Gujarat, India) in their vegetative state and were brought to laboratory. Leaves, stem and roots of both plant were separated, cleaned dust free and washed separately using distilled water. For preliminary analysis only the freshly collected plant material was used. For the fractionation guided bioassay only the allelopathic part of both the medicinal plant (inferred from the results of preliminary analysis) was selected, dried (air and oven), powdered and stored for future use.

Preliminary Allelopathic Analysis: In preliminary screening allelopathic potential of the medicinal plants was analysed using aqueous extract/ filter paper bioassay. Radish *(Raphanus sativus* L.) was selected as a test plant for these bioassays.

Preparation of Extracts: For the preliminary screening of both the medicinal plants, aqueous extracts of the three plant parts from both the plants were used. Aqueous extract of each plant part i.e. leaf, stem and root were prepared separately at three different concentrations (0.5%, 1%, 2 %). Fresh plant part were finely chopped, weighed and soaked in distilled water for 24 hours (in cold and dark), followed by gently macerating the soaked plant material using mortar and pastel in the same aqueous medium and keeping the same in refrigerator for next 24 hours for further extraction. This aqueous preparation was filtered using whatmann filter paper (no.1) and the filtrate was used as aqueous extract or stored in refrigerator for further use.

Aqueous Extract/ Filter Paper Bioassay: These bioassay's were conducted in randomized complete block design following method by Ben-Hammounda *et al.*, (1995). Petri-plates (Inner diameter- 125 mm) lined with double filter paper were supplied with 10 ml of distil water or 10 ml of aqueous extract of leaf, stem, root of selected medicinal plant in a single bioassay. Three replicates were kept for each treatment. Prepared petri-plates were kept for 24 hours in dark and then were kept in laboratory conditions for incubation. Laboratory temperatures varied from 25 ± 5 ℃ to 40 ± 5 ºC. Relative Humidity varied from 45 to 54 %. Before keeping the radish seeds for bioassay, the uniform sized seeds were surface sterilized with 0.1 % aqueous Sodium Hypochloride solution, washed with distil water atleast for five times and immediately employed for bioassay use. Ten radish seeds were kept in each petri-plate. Coding used for percentage aqueous extracts (leaf, stem and root) for both the medicinal plants is as follow: 1) control (water), 2) 0.5 % leaf. 3) 1.0 % leaf, 4) 2.0 % leaf, 5) 0.5 % stem, 6) 1.0 % stem, 7) 2.0 % stem, 8) 0.5 % root, 9) 1.0 % root and 10) 2.0 % root. Observations were made after one day upon germination. Readings were recorded and measured for germination and seedling parameters like radicle length (cm) and plumule length (cm).

Total germination (G_T): $\frac{N_T \times 100}{N}$

(Where, N_T is the proportion of the seeds germinated for the last time measurement, N is number of seeds used in bioassay) (Chiapusio *et al.*, 1997)

Evaluation of Medicinal Plant Toxicity on Chloris

Considering the results of preliminary screening data only the most potent inhibitory plant part was selected for further use. Respective plant parts were collected, shade dried and finely powdered using

electric grinder. The powdered plant material was subjected to bio-fractionation following Harborne, (1984) protocol and the fraction so obtained was used for conducting allelopathic bioassay. The test plant used for these bioassays was *Chloris barbata* Sw.

Chemical Fractionation of Medicinal Plant Part: Dried root powder was extracted using soxhlet in the ratio 1g powder: 10ml of 70% aqueous methanol. After extraction the methanolic extract was collected and subjected to fractionation as per method given by Harborne, (1984). Upon fractionation three fractions were obtained i.e. A) Chloroform, B) Chloroform-methanol and C) Methanol termed as per the respective solvents used. The three fractions so obtained for both the medicinal plants were analysed for allelopathic potential using bioassays. All the solvents were of analytical reagent grade (Fisher Scientific, India).

Fractionation Guided Allelopathic Analysis of Harborne's Fraction's A, B and C Against Chloris: Modified sandwich method (Fujii *et al.,* 2003) was used for the bioassay**.** Bioassay's studies were conducted using aqueous agar (0.5 %) as a growth medium. The agar based bioassays were carried out in the glass petri-plates (inner diameter - 200 mm). The three Harborne's fraction A, B and C were completely evaporated in water bath (to remove and prevent respective solvent toxicity), dissolved ultimately in the ethanol and these fractions solubilised with ethanol were used for imparting allelopathic treatments.

Three concentrations 0.25, 0.5 and 1 % of each of the three fractions A, B and C were used, constituting nine treatments. A control was always kept where in pure solvent or plant fractions were not added. In addition to control and the fraction treatments, the solvent used to dissolve extracts was also incorporated as a treatment in order to ensure accounting the inhibitory effect of the pure solvent (eventually ethanol) itself, if any.

Required concentrations of the treatments were added to aqueous agar each separately, after the agar was completely melted so as to avoid metabolites degradation. Treatments with percentage of Harborne's fraction for both medicinal plants were coded as follow: **3)** 0.25 A, **4)** 0.5 A, **5)** 1.0 A, **6)** 0.25 B, **7)** 0.5 B, **8)** 1.0 A, **9)** 0.25 C, **10)** 0.5 C and **11)** 1.0 C. Control (aqueous agar) and a treatment with solvent ethanol (added to agar) were coded as treatment **1)** and **2)** respectively. All the treatments were repeated thrice.

For the bioassay, after cooling and solidification of agar, 20 Chloris seeds were uniformly placed in the petri-plates. The setup was kept in dark for an hour and was kept for incubation under the laboratory conditions (at 40±5ºC).

Germination in Chloris was found to commence after a day of incubation. Readings for number of the seeds germinated were recorded at the interval of 24 hour for four days. The radicle length (mm) was measured on the fourth day.

Total germination was calculated as per the given formula:

Total germination (G_T): $\frac{N_T \times 100}{N}$, [Where N_T is proportion of the seeds found germinated on the last day, N is number of seeds used for bioassay]

Phytochemical Analysis of the Allelopathic Fraction:

For both the medicinal plants, out of the three analysed fractions the most allelopathic Harborne's fraction was subjected to phytochemical analysis. This fraction was analysed for chemical nature of its secondary metabolites to be whether it is a phenolic, alkaloid and/ or terpenoid. The chloroform extract was used for analysis of terpenoids and phenolics, while the chloroform: methanol extract and the methanolic extract were analysed for presence of alkaloids. The analysis was done using planar chromatographic techniques like paper chromatograph (PC) and thin layer chromatography (TLC). The phytochemical analysis was performed solely for qualitative purpose. For paper chromatography, whatmann no.1 filter paper was used as a stationary phase. For TLC, the plates were prepared using silica gel G and only activated plates were used for the TLC analysis.

Phenolics: Phenolics were analysed using 2 D chromatography (Daniel, 1991). The fractions were dissolved in ethanol and spotted at the corner of square cut whatmann no. 1 filter paper and was allowed to dry. The chromatogram was developed in organic layer of the solvent system Benzene: Acetic acid:

Water (6:7:3).

After the run was over the chromatograms were removed and allowed to dry completely. Then the side of chromatogram was flipped with the side having vertically separated metabolite. This was kept dipped in the second solvent system (Sodium formate: Formic acid: Water- 10:1:200) and the run was allowed to progress in the second direction. The chromatogram was removed from solvent system after the run was over and was air dried. The spray reagents, p-Nitra aniline and Sulfanilic acid were prepared as reported by Daniel (1991). The spray reagents were kept in Refrigerator at 4ºC and always used in chilled condition to favour the diazo test. The chromatograms for an extract were always kept in duplicate, for each of the two diazo spray reagents. The chromatogram were dipped in spray reagent and immediately dipped in 10 % Na_2CO_3. The chromatograms sprayed with both the reagent separately were observed for the colour developed. A single phenol compound, takes different colour in both the spray reagents. Phenolic compounds were identified confirming their colour developed in the spray reagent (Daniel, 1991) respectively.

Terpenoids: Harborne's fraction A was analyzed for presence of terpenoid's using thin layer chromatography (Daniel, 1990). Fraction was spotted in form of bands or spots solubilising each in ethanol. Hexane: Acetone (4:1) was used for developing the spotted TLC plates. For colour development 50% H_2SO_4 was used as a spray reagent. After being sprayed with the spray reagent the TLC plates were heated using hot plate. The plates were observed for the colour developed.

Alkaloids: Fraction B and C were analysed for presence of alkaloids. The alkaloids were analysed as per method given by Daniel, (1990), using TLC plates. Fractions were spotted on TLC plates using ethanol. Developing solvent system used for primary alkaloids was Chloroform: Methanol: Water (50:45:5) and for quaternary alkaloids was Methanol: 2M Ammonium nitrate (3:2). Developed TLC plates were sprayed using Dragendorffs reagent. The sprayed TLC plates were observed for alkaloids which assumes orange to brown colour.

Statistical Analysis: All the bioassay experiments were conducted using a complete randomized design. For germination and seedling growth, data from bioassay results were analyzed using SPSS 16. In order to find significance of differences between control and treatment means, results were subjected to one way ANOVA followed by separation of means using Tukeys post hoc test ($P < 0.05$).

RESULTS AND DISCUSSION

Chlorophytum borivilianum Santapau & R.R.Fern and *Asparagus racemosus* Willd. are two most important medicinal plants. Owing to the secondary metabolites and the medicinal properties they possess, the plants have high application value. Allelopathic analysis of these two plants gave promising results.

Preliminary Analysis: The preliminary analysis was performed using aqueous extract/ filter paper bioassay. Aqueous extract of both the plants imposed various inhibitory effect on the growth parameters of the selected test plant that is radish.

Chlorophytum borivilianum Santapau & R.R.Fern:

In control the radish seed germination was 90%. Total germination of radish in treatment with leaf and stem aqueous extracts was even higher than the control, at all the studied concentrations. Aqueous extract of root at lower concentration viz. 0.5 % and 1 %, inhibited the germination where in percentage germination was 88 % and 78 % (Figure 1). However, the aqueous root extract at 2% concentration was found to impart stimulation to the germination of radish. Radicle length in control was 3 cm. The radicle length was reduced in 2 % of leaf and 2 % of root extracts (Figure 2). All the other extracts at all the concentration were found to stimulate the radicle growth. Plumule length in control was 0.91 cm. The stem extracts at 1 % was most inhibitory to the plumule growth where in plumule length was 0.87 cm (Figure 2). All the other extracts had stimulatory effect on the plumule length. Thus aqueous root extract of Chlorophytum was the most inhibitory suppressing both germination and radicle length of Radish. Root is the principle plant part for metabolite localization in *Chlorophytum* and the inhibitory effect on germination and growth must be owing to the water soluble root metabolites.

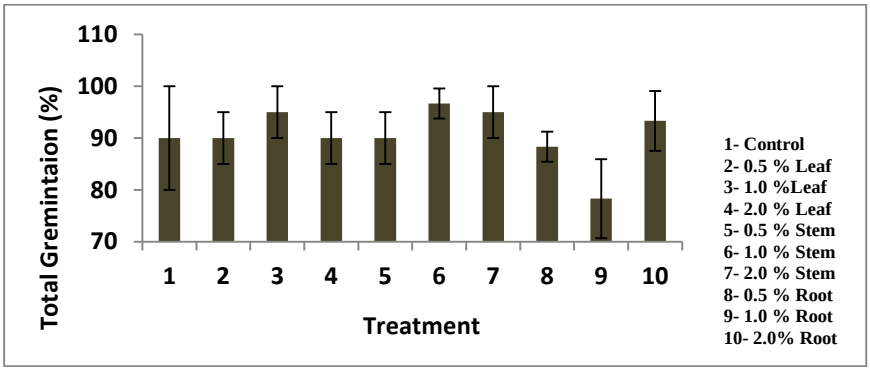

Figure 1: Effect of Chlorophytum Plant Part Extracts on Radish Seed Germination
(Note: Bars represent means ± std)

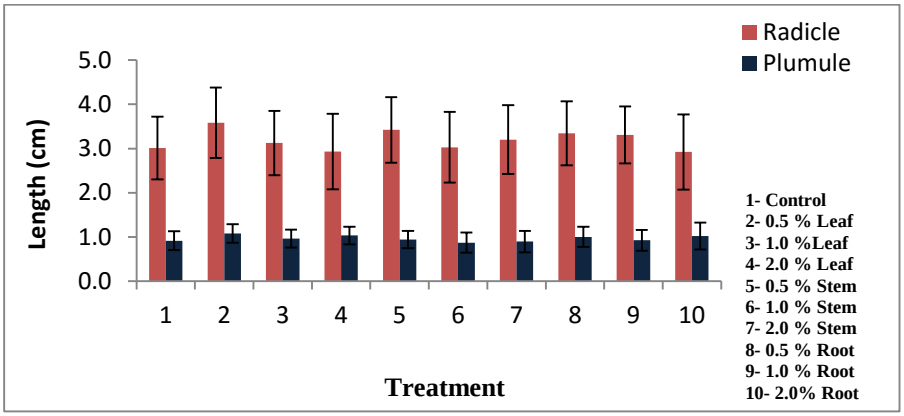

Figure 2: Effect of Chlorophytum Aqueous Extract on Radish Seedling Growth
(Note: Bars represent means ± std)

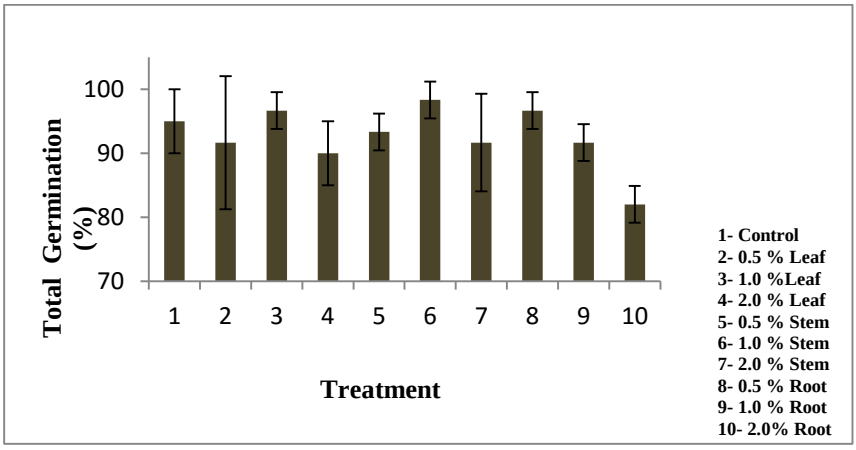

Figure 3: Effect of Asparagus Plant Parts Extracts on Radish Seed Germination
(Note: Bars represent means ± std)

Asparagus racemosus Willd.: Germination of radish was found to be 95 % in the control. Treatment with all the plant part affected the germination to different extent. The least germination that is 82 % was observed to be in treatment with 2 % of root extract and followed by 90 % germination observed in 2 % leaf and 92 % observed in 0.5 % of leaf, 2.0 % stem and 1 % of root extracts (Figure 3). Radicle length

was greatly affected by 2 % of root extracts and all the other extracts that is leaf, stem and root at different concentration had no effect on radicle growth (figure 4). Plumule was not affected by any of the aqueous extract treatments. The allelopathic effect of Asparagus roots on germination and radicle growth of radish must be owing to the secondary metabolites that are extractable in water. Thus, water soluble secondary metabolites from Asparagus are allelopathic in nature.

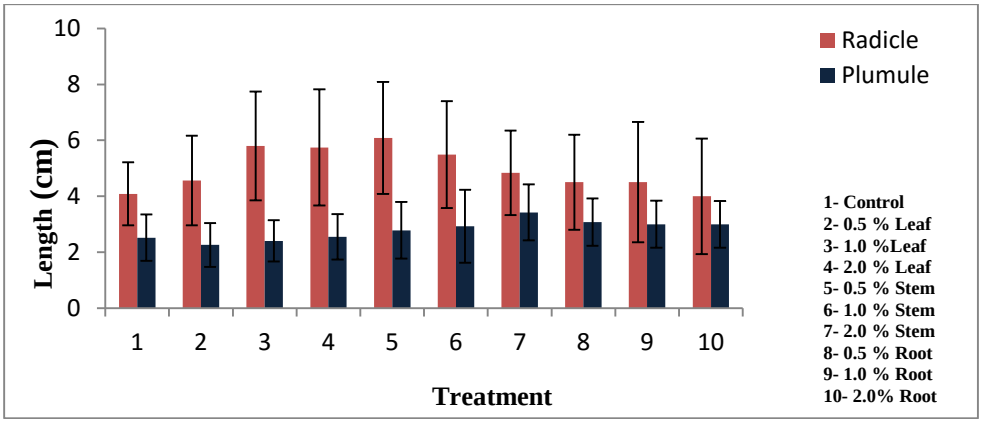

Figure 4: Effect of Asparagus Aqueous Extract on Radish Seedling Growth
(Note: Bars represent means ± std)

Roots of both the medicinal plants, i.e. Chlorophytum and Asparagus exhibited phytotoxicity to the growth of radish in the preliminary analysis. The medicinally important part in both the plant is root. Roots are reported to have number of secondary metabolites. Medicinal property of the root is owing to these metabolites which are pharmacologicaly significant. The aqueous root extracts of both the medicinal plants were found to be more inhibitory than other part. Plant roots were thus selected for further study. The dried root powder was subjected to Harborne's fractionation followed by bioassay and phytochemical analysis of the same. These bioassay studies were conducted on *Chloris* a common monocot weed selected as the test plant.

Evaluation of Medicinal Plant Toxicity on Chloris:

Fractionation Guided Allelopathic Analysis of Harborne's Fraction's A, B and C Against Chloris:

Chlorophytum borivilianum Santapau & R.R.Fern.: Out of the three Harborne's fractions, Chloroform fraction at its highest concentration (2%) was found to be most inhibitory to the growth parameters such as germination, growth and biomass of *Chloris*. Chloroform: Methanol and Methanol fractions at same concentrations offered stimulatory effect on some growth parameters of *Chloris* which was ultimately ceased after 2-3 days. Treatment with solvent ethanol did affect the growth parameters in *Chloris* however fraction addition further amplified the effect.

Germination: The *Chloris* seed germination in control was 82 % and that in solvent alcohol treatment was 83 %. Chloroform fraction was the most inhibitory of all the fractions analysed and was found to inhibit the *Chloris* germination. Chloroform fraction at 2% concentration inhibited the germination to 19 % (figure 5). The *Chloris* germination was affected in a concentration dependant manner, where in germination decreased with the increase in concentration of each fraction.

Seedling growth: Radical length in control was 0.87 cm and that in solvent alcohol was 0.72. The alcohol treatment did affect the radicle length growth but further treatment with plant fractions significantly inhibited it even more. Both Chloroform and Methanol fraction suppressed the radicle growth. Chloroform fraction at 0.5% and 1% showed the major inhibitory effects reducing the radicle length to 0.12 cm and 0.10 cm (figure 6). Methanol fraction at 0.25 %, 0.5 % and 1 % inhibited the radicle length to 0.47, 0.35 and 0.23 cm respectively. The inhibitory effect all the three fractions was observed to be concentration dependent that is increase in the concentration of the extracts decreased the radicle length. Plumule length in control and alcohol was 0.83 and 0.46 cm. Length was most affected by the highest

concentrations of both Chloroform and Methanol fractions that is 1 % (figure 6). The fraction were found to be inhibitory to the growth of Plumule although statistically significant inhibition of plumule growth was observed for 1% concentration of Chloroform extract and 1 % methanol fraction (figure 6) where in the plumule length reduced to 0.1 and 0.2 cm respectively.

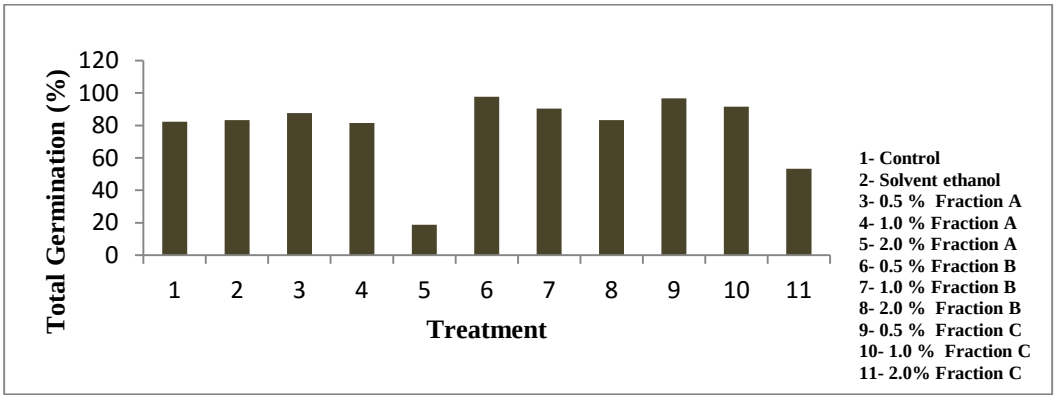

Figure 5: Effect of Different Fractions from Chlorophytum on Germination of *Chloris*

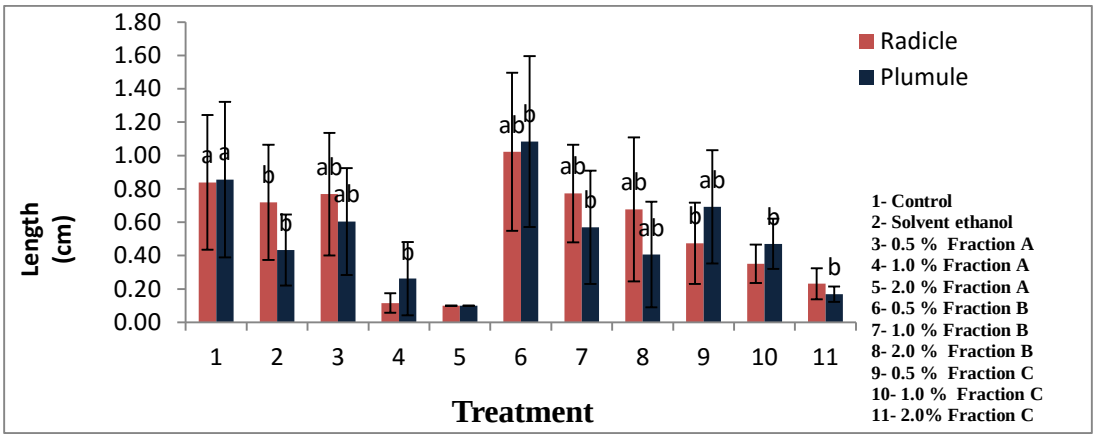

Figure 6: Effect of Different Fractions from Chlorophytum on the Growth of *Chloris*

(Note: Bars represent means ± std, alphabet 'a' denotes the control, 'b' denotes the alcohol treatment, Bars with similar alphabets are not statistically significant)

Phytochemical Analysis of the Harborne's Fraction from Chlorophytum: Fraction A was analysed for terpenoids and phenolic. The TLC analysis of terpenoids gave three bands of purple to blue colour for one spot (Figure 7a) indicating presence of three compounds. The 2D paper chromatogram showed single band appearing purplish pink in colour when developed in the sulphanilic reagent (Figure 7b). Inferring the colour and position of the spot, the compound was identified as sinapic acid. Thus, the fraction A which was also the most allelopathic fraction obtained by fractionation of Chlorophytum root, showed presence of three terpenoids and one phenolic acid. The analysis of fraction B performed using TLC gave positive results for primary alkaloids with one band per spot (Figure 7c) representing one compound. Similarly, the analysis of fraction C for quaternary alkaloids developed one band per spot (Figure 7d). The allelopathy of Chlorophytum can be due to either of two i.e. Terpenoid or Phenolic or a combine effect of both. Number of reviews has discussed the allelopathic potential of both the class of secondary metabolites. A review given by John and Sarada, (2012) has reported the allelopathic activity of phenolic compounds. Mizutani, (1999) discusses different terpenoids to have allelopathic potential. In Chlorophytum, terpenoids are the metabolites that are medicinally important. The promising results of preliminary and fractionation bioassay suggest that further studies can be done to identify the actual

compound which is responsible for the allelopathic activity of Chlorophytum. The allelopathic compound from the plant may be explored as commercial herbicide source.

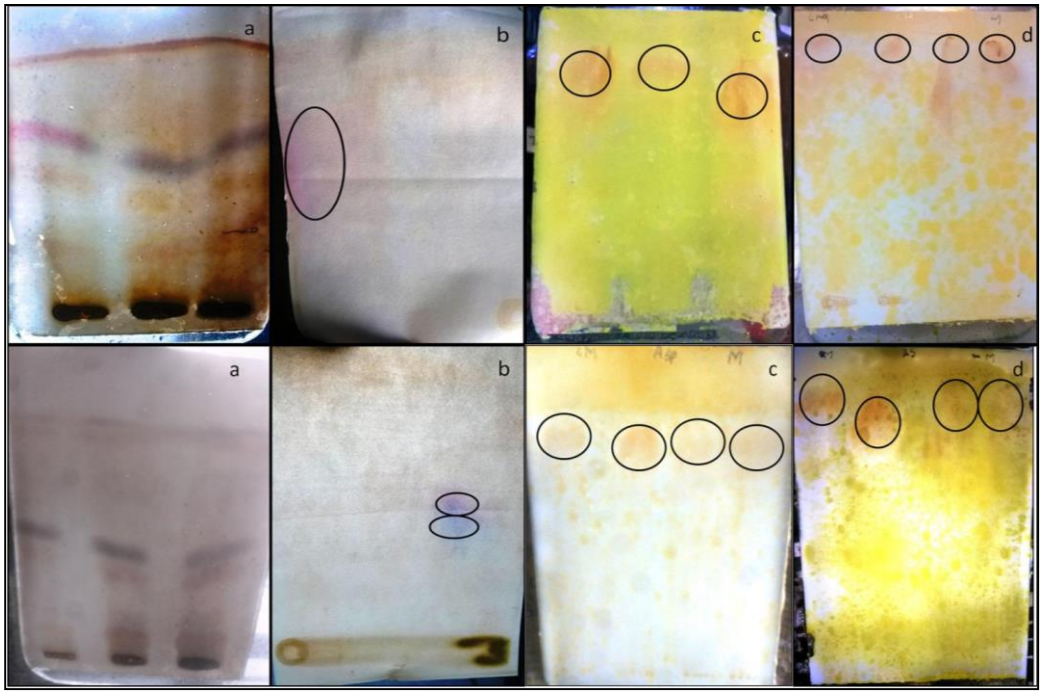

Figure 7: Phytochemical Analysis of Harborne's Fractions
Note: a) terpenoids, b) phenolics acids, 3) primary alkaloids, d) quaternary alkaloids
Upper row: Chlorophytum and lower row: Asparagus

Asparagus racemosus Willd.: The chloroform: methanol fraction (fraction B) at two concentrations i.e. 0.5 % and 1%, imposed highest inhibitory effect on the germination, seedling growth and fresh weight of *Chloris*. Methanolic extract was least inhibitory to *Chloris* growth parameters.
Germination: The germination in control was 93 % and that in alcohol treated seedlings was 93 %. Chloroform: methanol fraction inhibited the germination of *Chloris* at all the concentrations suppressing the germination to 90 %, 51 % and 17 % respectively. The lowest germination was observed in the *Chloris* seeds treated with highest concentration of chloroform: methanol fraction that is 1 % (Figure 8). Both chloroform and methanol fractions had no effect on germination of *Chloris.*

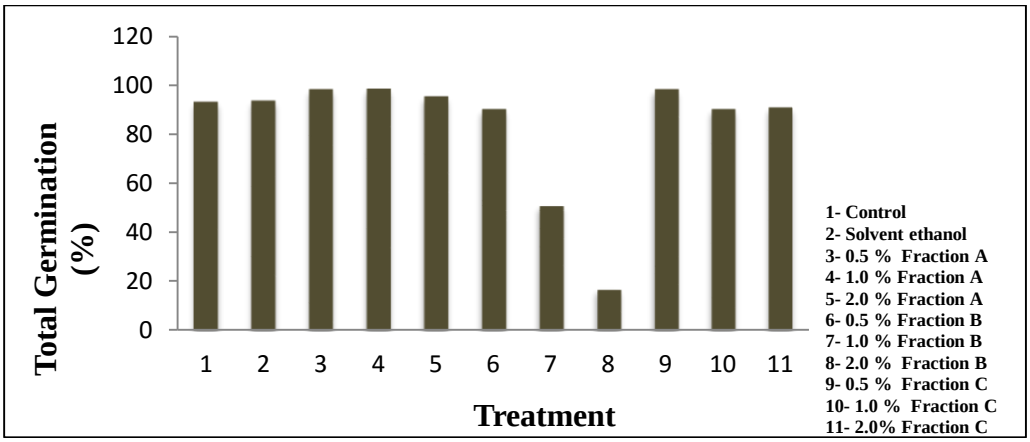

Figure 8: Effect of Different Fractions from *Asparagus* on Germination of *Chloris*

Seedling growth: Radical Length in control was 0.63 cm and in alcohol treatment was increased to 0.98 cm. It was greatly inhibited owing to treatment with chloroform: methanol fraction (figure 9), wherein increase in concentration gradually inhibited the radicle growth. Both 0.5 % and 1% showed inhibitory effect but the highest inhibition was observed in 1% chloroform: methanol fraction where in the radicle length reduced to 0.18 cm. The inhibitory effect was concentration dependant. Methanol fraction also affected the radicle growth at 1 % concentration to 0.9 cm. Plumule growth was highly reduced in all the treatments of chloroform: methanol extract (figure 9), though 1% concentration was the most inhibitory reducing the length to 0.26 cm. Plumule length in control was 0.66 cm and in alcohol treatment was 0.84 cm.

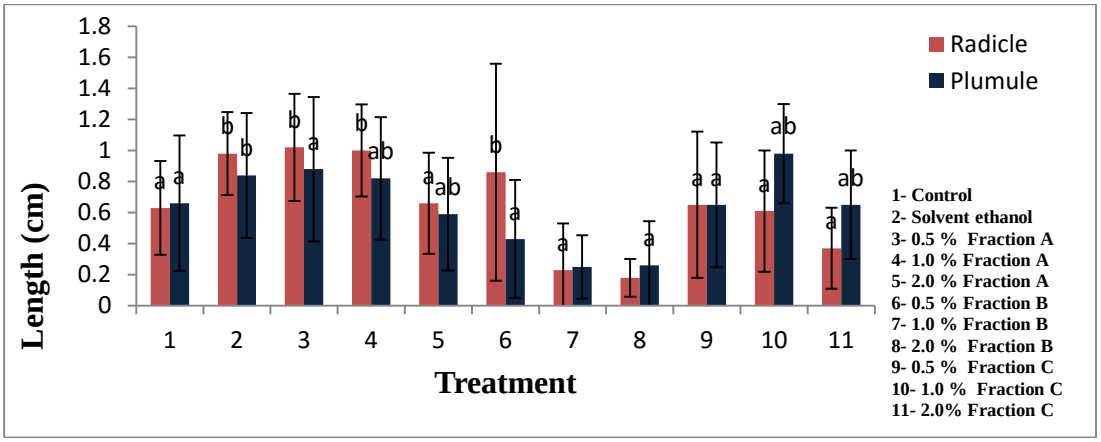

Figure 9: Effect of Different Fractions from *Asparagus* on Growth of *Chloris*
(Note: Bars represent means ± std, alphabet 'a' denotes the control, 'b' denotes the alcohol treatment, Bars with similar alphabets are not statistically significant)

Phytochemical analysis of the Harborne's fractions from Asparagus: Chloroform: methanol fraction was analysed for the primary alkaloids using TLC. TLC developed one band per spot in the dragendorff spray reagent, indicating presence of single compound (Figure 7c). The TLC analysis of chloroform: methanol fraction showed presence of atleast one quaternary alkaloids attaining dark orange colour in the dragendorff reagent (figure 7d). Our study reports presence of primary and quaternary alkaloids in the roots of Asparagus. The chloroform: methanol fraction was also the most allelopathic fraction and was found to inhibit Chloris growth. Thus the alkaloids from Asparagus can be the metabolite responsible for allelopathic potential of the plant. However, the medicinal utility of Asparagus roots is owing to the pharmacologicaly active terpenoids. The fraction A was analysed for terpenoids using TLC and developed 6 bands of purple to pink colour per spot indicating presence of terpenoids (Figure 7a). Fraction A was also analysed for phenolic compounds using 2D paper chromatography. The 2D chromatograms showed 2 bands per spot, in both the reagents each. The bands developed purple and blue colour in diazo P Nitra-aniline and developed orange and pink colour in the Diazo Sulphanilic acid reagent. The metabolites were identified based on their colour in both the reagent and appears to be vanillic acid and syringic acid, both are simple phenols (figure 7b). Results of our study suggest that alkaloids present in the Asparagus roots are responsible for the inhibitory effect imparted by chloroform: methanol fraction obtained from the same. Many alkaloids are known to exhibit allelopathy. Lovett *et al.,* (1981) have reported allelopathic potential of two alkaloids scopolamine and hyoscyamine. Rizvi *et al.,* (1981) have reported the allelopathic activity of caffeine. The plant residues or the soil from the plant rhizosphere were found to contain the alkaloids and are responsible of the allelopathy exhibited by plant. The study reports the alkaloids present in a terpenoids dominated plant to be allelopathic. However, detailed study on identification of the actual allelochemical responsible for allelopathic potential of Asparagus may increase the utility value of Asparagus. As the plant is important due to the medicinally

important terpenoids which are present in the fraction A, the fraction B that is the chloroform: methanol containing alkaloids fraction can be further explored for its allelopathic potential. Watanabe *et al.,* (2011) have reported the allelopathic effect of Asparagus roots on lettuce seedlings and have found it to be independent of plant sex. Also number of studies has been done to study the replant problems in Asparagus. Yeasmin *et al.,* (2013) have reported that the plant has replant problems and exhibits auto toxicity which can be overcome by selection of proper verities. They have studied the plant residue amended soil to have allelopathic potential. Our study analysed and found the aqueous root extract and the chloroform: methanol faction obtained by fractionation to be allelopathic to the growth of Chloris. The phytochemical evaluation of chloroform methanol fraction could detect presence of alkaloids. Results of the present study suggest that some water soluble metabolites or the alkaloids may be responsible for autototoxicity of the plant. Yeasmin *et al.,* (2013) have reported certain organic acids such as oxalic, succinic and tartaric to be the allelochemicals responsible for the allelopathic potential of Asparagus. The allelopathic activity of aqueous root extract suggest that the plant must release water soluble compounds through roots into the soil or the root debris may release the compounds into the soil and cause soil sickness.

Both the plants were primarily phytotoxic through their aqueous root extracts. As the medium of metabolite transport in agriculture field is aqueous, these plants exhibiting allelopathy by their aqueous root extracts, when grown with proper spacing strategies may be utilized for weed management. Study to assess spread of allelochemicals in the rhizosphere of these plants is in progress. Present study conducted in laboratory may be extended to the field to utilize the allelopathic potential of Chlorophytum and Asparagus for effective weed control through various means. Upadhyay *et al.,* (2011) emphasize importance of Asparagus for weed mitigation through integrated weed management. They suggest that Asparagus, when used as cover crop, rotation crop or in integrated weed management can reduce weeds infestation. Results from the present study show that both the plant can be employed for weed control when grown with other economically significant crop plants. These medicinal plants can be considered for their use as smother crop or companion crop and can be incorporated in to an integrated weed management system. Compatibility of both medicinal plants with that of crop plants should be evaluated before attempting the wide scale application. Also the phytochemical analysis of the plant fractions enabled detection of metabolites in the allelopathic fractions. Thus the plants can be potential source of allelopathic/ herbicidal molecule and can be further studied for identifying actual molecule. Detailed study on the allelopathic molecule, its mode of action and site of action may prove to be a great help to finding the ecological weedicial alternatives. Evaluation of allelopathic potential of medicinal plants has imparted value addition to the existing importance. The added benefits may be considered and encourage increased cultivation ventures for both the plants. Systematic evaluation of further allelopathic potential of the plants may direct an ecologically viable alternative to the synthetic herbicides used in agricultural systems.

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