RESEARCH

54(265), January 1, 2018

ISSN 2278–5469 EISSN 2278–5450

Discovery

Comparative study of allelopathic effects of aqueous extract of *Lycopodium clavatum* (L.) and *Selaginella uncinata* (Desv ex. Poir) on the seedling growth of *Zea mays*

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Article History
Received: 03 November 2017
Accepted: 20 December 2017
Published: 1 January 2018

Citation
Akpan EN, Denise EM, Ezendiokwelu EL, Anyadike MC. Comparative study of allelopathic effects of aqueous extract of *Lycopodium clavatum* (L.) and *Selaginella uncinata* (Desv ex. Poir) on the seedling growth of *Zea mays*. *Discovery*, 2018, 54(265), 23-28

ABSTRACT

Allelopathic effects of aqueous extracts of *Lycopodium clavatum* and *Selaginella uncinata* were studied on the seedling growth of *Zea mays*. To investigate the effects of the extracts on seedling growth, greenhouse experiment was conducted. The plumule length and radicle length were measured for both extracts using a ruler. The result showed that the two extracts had both inhibitory and stimulatory effect on *Zea mays* radicle growth at different concentrations. *Selaginella uncinata* extract showed stimulatory effect at 25% and 50% concentrations and inhibitory effect at 75% and 100% concentrations. *Lycopodium clavatum* on the other hand showed stimulatory effect at 25% concentration only and inhibitory effect at 50%, 75% and 100% concentrations. Radicle length decreased in both extracts with increase in concentration whereas the plumule length increased over the measurement period and also decreased with increase in concentration. The extracts had strong inhibitory effect on radical elongation of seedling than on plumule elongation.

Key words: Allelopathy, *Lycopodium clavatum*, Seedling growth, *Selaginella uncinata*, *Zea mays*

1. INTRODUCTION

The phenomenon of allelopathy has received increasing attention as a means of explaining vegetation patterns in plant communities (Miller, 1996). Although allelopathy has been observed for over 200 years and the phenomenon reports as early as 300BC document that many crop plants inhibited the growth of other plants and destroyed its field weeds. In 1974, after the publication of first book of allelopathy by Elory L. Rice the phenomenon got a new attention in science community, who later reinforced this definition of allelopathy (Olofsdotter *et al.,* 2002). The effects of one plant to another plant may either be both stimulatory and inhibitory that depends on the concentration of the released compounds (Bhowmik and Inderjit, 2003).

Allelochemicals, which are non-nutritive substances mainly produced as plant secondary metabolites or decomposition products of microbes, are the active media of allelopathy. Allelochemicals can stimulate or inhibit plant germination and growth, and permit the development of crops with low phytotoxic residue amounts in water and soil, thus facilitating wastewater treatment and recycling (Macias *et al.,* 2003; Zeng *et al.,* 2008).

Lycopodium clavatum, commonly known as Club moss, Clubfoot Moss, Foxtail, Ground Pine, Sulfer or Wolfs Claw is one of the most widespread species belonging to family Lycopodiaceae. It is a pteridophyte which is abundantly found in tropical, subtropical and in many European countries. This spore bearing vascular plant is used in various traditional system of medicines viz; stomach pain, against rheumatic disease, muscle pain, Alzheimers disease etc.

Selaginella uncinata 'Peacock Spikemoss is not a true moss but rather a type of lycophyte, which are the oldest living vascular plants that date back to 410 million years ago, making them somewhat prehistoric and unlike most plants by design. This shrub-like tropical plant has uniquely iridescent green and bronze, scaly, coarse foliage that tends to trail upwards and out, creating a nice groundcover.

Maize is the domesticated variant of teosinte (ISAAA, 2011). The two plants have dissimilar appearance, maize having a single tall stalk with multiple leaves and teosinte being a short, bushy plant. The difference between the two is largely controlled by differences in just two genes (ISAAA, 2011). The six major types of corn are dent corn, flint corn, pod corn, popcorn, flour corn, and sweet corn (Linda, 2013).

2. LITERATURE REVIEW

Anita and Anjana (2013) studied the allelopathic effects of aqueous extract of leaves of *Mikania micrantha* on seed germination and seedling growth of *Oryza sativa* L. and *Raphanus sativus* L. Seed germination and seedling growth were inhibited by concentrated aqueous extract of *M. micrantha*, *R. sativus* was more sensitive to inhibitory effects of leaf aqueous extract of *M. micrantha*. The extract had strong inhibitory effect on root enlongation of seedling than in shoot enlongation.

Arpana (2014) study was carried out on *Lantana camara* which is often regarded both as a notorious weed and a popular ornamental garden plant. *Lantana* allelopathic effect studies have been done with many crops, trees, shrub and weeds under both laboratory and field conditions to determine their allelopathic potential and its use. Allelochemicals of *Lantana* inhibited the germination, growth and metabolism of crops, weeds and bryophytes and vegetables.

Hegab (2008) studied the allelopathic effect of aqueous whole mature chard plant extract (*Beta vulgaris* L. var. Cicla) on wheat (*Triticum vulgare* L. var. Sides 1) and an associated weed (chard). The aqueous extract retarded the germination of chard more effectively than that of wheat and the effect was concentration dependent. The lowest concentration stimulated the germination of both wheat and chard; on the other hand, the germination was retarded under the application of concentrations above 1%.

Lawan *et al.* (2011) conducted an experiment to observe the inhibitory effects of the leaf extract derived from neem (*Azadiracta indica*) on germination and growth behavior of some cowpea varieties (receptor). he aqueous extracts of leaf caused significant inhibitory effects on germination, root and shoot elongation and development of lateral roots of treated plants. Bioassays indicated that the inhibitory effect was proportional to the concentrations of the extract, as concentration increased the extent of inhibition also increased. The study also revealed that inhibitory effect was much pronounced in root and lateral root development rather than germination and shoot growth.

3. MATERIALS AND METHODS

PLANT MATERIAL:

The leaves of *Selaginella uncinata* and *Lycopodium clavatum* were collected from Ita-Enang Dam-Ididep Ibiono Ibom on 25th of May, 2017. The plants were identified in the herbarium of Botany and Ecological Studies University of Uyo. The plants were taken to the Chemistry Laboratory University of Uyo where the extraction processes was conducted.

AQUEOUS EXTRACTION AND PURIFICATION:

The leaves were spread on a ply wood and were air-dried for three days at room temperature. The dried leaves were poured into two different transparent buckets and 3 litres of distilled water was added into each bucket. It was allowed to stand for a week and three days so that the contents will be properly extracted. Each extract was filtered through a separating funnel stuffed with enough cotton wool and hung on a retort stand to remove debris. The stock concentration was gotten by evaporating the extracts using a water bath.

SEED CULTURE AND TREATMENT:

Seeds of *Zea mays* were surface sterilized with water: bleach (10:1 v/v) solution for 5 minutes to avoid contamination and were thoroughly rinsed four times with sterile distilled water. For testing, 30 Petri dishes (9 cm in diameter), 15 for the first plant and the other 15 for the second plant was washed and sterilized with water: bleach (10:1 v/v) solution for 3 minutes to avoid contamination, each Petri dish were well labeled to avoid mixing up. Whatman No.1 filter papers were kept in each Petri dish, and ten sterilized seeds of Zea mays were placed in each petri dish at equal distance. Concentrations of 25%, 50%, 75% and 100% were prepared from the stock solutions for the two extracts. 10 ml of each concentration was added to the petri dishes separately for the two extracts. Seeds soaked in distilled water were used as control and each of the experiment was repeated two times. 10 ml of each extract and 10 ml of distilled water for control was applied daily during the experiment to keep the Whatman paper moist for seedling development for a period of 14 days. The experiment was conducted at the Botany Green House, University of Uyo, Uyo Akwa-Ibom State Nigeria.

During the course of the experiment, data were recorded on the variables mentioned below;

RADICLE LENGTH:

Length of radicle (root) length was measured using a ruler in cm.

PLUMULE LENGTH:

Length of plumule (shoot) length was measured using a ruler in cm.

STATISTICAL ANALYSIS:

Analysis of variance was performed using standard techniques and differences between the means were compared through Duncan's multiple Significant Difference test ($P < 0.05$) using SPSS software package.

4. RESULTS

The results of the allelopathic effects of *Lycopodium clavatum* and *Selaginella uncinata* on the seedling growth of *Zea mays* are shown in table 1, 2, 3 and 4. Table 1: shows that extracts of *Lycopodium clavatum* had inhibitory effect on radicle growth for 75% and 100% compared to control over the measurement period. The extract also showed stimulatory effect at 25% concentration but the treatment with no extract (control) showed the highest radicle length. Table 2: shows there was no plumule growth on day 2 of the start of the experiment. From the table it could be observed that the plumule length increased over the measurement period. This shows that the extract had stimulatory effect on *Zea mays*. It could also be observed that 25% concentration showed the highest plumule growth. Results of the allelopathic effects of *Selaginella uncinata* on the seedling growth of *Zea Mays* are shown in table 3 and 4. Table 3: shows the radicle growth of *Zea mays* while table 4 shows plumule growth of *Zea Mays*. The result for table 3 showed that *Selaginella uncinata* had both stimulatory and inhibitory effects on the radicle length of *Zea mays*. The stimulating effect was observed at 25% and 50% concentrations. Inhibitory effect was also observed at 75% and 100% concentrations. In table 4 it was observed that 25% and 50% concentrations had the highest plumule length compared to control. Decrease in plumule length was observed at 75% and 100% in comparison with control. This shows that *Selaginella uncinata* has both inhibitory and stimulatory effect on *Zea mays*. It could be observed that the plumule length increased over the measurement period.

Table 1

Allelopathic effect of *Lycopodium clavatum* extract on the radicle length of *Zea mays*

Concentration	Day 2	Day 4	Day 8	Day 12	Day 14
Control	0.40cm	1.87cm	1.97cm	2.63cm	2.37cm
25%	0.50cm	1.20cm	2.40cm	2.13cm	2.23cm
50%	0.53cm	0.53cm	0.57cm	0.63cm	0.60cm
75%	0.16cm	0.50cm	0.73cm	1.23cm	1.60cm
100%	0	0.10cm	0.97cm	1.0cm	0.9cm

Table 2

Allelopathic effect of *Lycopodium clavatum* extract on the plumule length of *Zea mays*

Concentration	Day 2	Day 4	Day 8	Day 12	Day 14
Control	0	0.53cm	3.23cm	6.80cm	8.53cm
25%	0	0.50cm	3.57cm	7.80cm	9.17cm
50%	0	0.37cm	0.93cm	1.50cm	2.57cm
75%	0	0.33cm	1.10cm	2.40cm	4.17cm
100%	0	0	0.87cm	1.53cm	1.60cm

Table 3

Allelopathic effect of *Selaginella uncinata* extract on the radicle length of *Zea mays*

Concentration	Day 2	Day 4	Day 8	Day 12	Day 14
Control	0.57cm	0.36cm	0.60cm	1.0cm	1.50cm
25%	0.43cm	0.43cm	1.47cm	1.0cm	0.83cm
50%	0.40cm	0.53cm	1.80cm	1.80cm	1.50cm
75%	0.30cm	0.30cm	1.07cm	0.47cm	0.25cm
100%	0.33cm	0.30cm	0.57cm	0	0

Table 4

Allelopatic effect of *Selaginella uncinata* extract on the plumule length of *Zea mays*

Concentration	Day 2	Day 4	Day 8	Day 12	Day 14
Control	0	0.20cm	1.90cm	3.13cm	3.20cm
25%	0	0.1cm	1.73cm	2.47cm	2.63cm
50%	0	0.47cm	1.90cm	4.13cm	5.10cm
75%	0	0	1.07cm	1.63cm	1.90cm
100%	0	0.36cm	1.07cm	2.50cm	2.53cm

5. DISCUSSION

RADICLE (ROOT LENGHT)

Statistical analysis of data showed that different concentrations significantly affected the radicle length of maize (P<0.05). With increased concentration, the buds root length reduced in comparison with control in *Lycopodium clavatum* extract *and Selaginella uncinata* extract. It was observed that *Lycopodium clavatum* has more effect on the radicle length with the highest length in control(no extract) and the radicle length decreased with increase in concentration showing that *Lycopodium clavatum* has inhibitory effect on radicle length this is in line with the findings of Sardoei *et al.* (2012) where the use of the *cyperus rotundus* extract reduced the germination, and it significantly decreased by increasing its in density, such that at the 75% and 100% densities, germination was terminated. *Selaginella uncinata* on the other hand showed the highest radicle growth at 50% concentration and least length at 75% and 100% concentrations. The reason for differences in radicle length cannot be explained because allelochemicals can be selective in their actions and plants can be selective in their responses. For this reason, it is difficult to determine the action of these compounds (Seigler, 1996). Many studies have shown changes in germination and growth of target seedlings, but few have evidenced the physiology and method of action of allelochemicals (Reigosa *et al*., 1999; Inderjit & Duke, 2003).

PLUMULE (SHOOT LENGTH)

There was no significant difference in the plumule length of *Zea mays* in both extract (P>0.05). The plumule length was not affected by the extracts of *Lycopodium clavatum* and *Selaginella uncinata*. With time passing, the plumule length increased over the measurement period in the two extracts. The highest plumule length was observed in 25% for *Lycopodium clavatum* and in 20% and 50% concentrations for *Selaginella uncinata.* Allelochemicals are more sensitive to radicle length than plumule length.

6. CONCLUSION

The application of the extracts of *Selaginella uncinata* and *Lycopodium clavatum* had both stimulatory and inhibitory effect on the radicle growth of *Zea mays* different concentrations. It was observed that at 50%, 75%, and 100% concentrations, *Lycopodium clavatum* had inhibitory effect on *Zea mays* radicle growth and caused stimulation at 25% concentration, while *Selaginella uncinata* on the other hand showed stimulatory effect on *Zea mays* radicle growth at 25% and 50% concentrations, inhibitory effect at 75% and 100% concentrations. In conclusion, both extracts had stimulatory effect on plumule growth of *Zea mays.* However, both extracts could be used for growing maize when low concentrations are used.

SUMMARY OF RESEARCH

1. Allelopathy is a natural ecological phenomenon. It has been known and used in agriculture since ancient times. It was defined as the harmful effect of one plant upon another. Currently, a more complete definition includes the positive and negative effects of chemical compounds produced mainly from the secondary metabolism of plants, microorganisms, viruses and fungi that have an influence upon the growth and development of agricultural and biological ecosystems (excluding mammals).
2. Most of the lower plants have inhibitory effect on seedling growth and development of cereal crops but could also act as a stimulator when in low concentration.
3. Roots of plants are more prone to inhibition by allelochemicals from plants than the plumule of plants, since roots are in close contact with the soil and therefore it's the first to be affected by allelochemicals from allelopathic plants.

FUTURE ISSUES

Many scientists have researched on the inhibitory and stimulatory effects of allelopathic and non allelopathic plants. Most of the researches done has always focused on the inhibitory or stimulatory effects of plant extracts on the seedling growth, germination percentage, or germination rate of the test plants but only few have studied the mode of action of allelochemicals on test plants, physiology of test plants and reason why plant extract has severe inhibitory effect on one plant and less on the other plant.

DISCLOSURE STATEMENT

There is no special financial support for this research work from the funding agency.

AKNOWLEDGMENT

We wish to acknowledge Miss Ezinne Ezendiokwelu and the entire research team for their role in this research work and for typing the manuscript. This work would not have been completed if not for their tireless effort. We appreciate you.

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