International Journal of Chemical and Biochemical Sciences
(ISSN 2226-9614)

Journal Home page: www.iscientific.org/Journal.html

Allelopathic property of parthenin on seed germination and seedling growth of wheat (*Triticum aestivum*) and barley (*Hordeum vulgare*)

Solomon Sime Tessema* and Alemu Mekonnen Tura

Department of Chemistry, Arbaminch University, Arbaminch, Ethiopia

Abstract

Allelopathic effect of *Parthenium hysterophorus* L. on seed germination and seedling growth of two crops wheat (*Triticum aestivum*) and barley (*Hordeum vulgare*) was studied at the concentrations level of 0%, 1%, 5% and 10% leaf, stem, flower and root extracts of *Parthenium hysterophorus*. Seed germination of wheat and barley was completely inhibited at 10% of all extract of *Parthenium hysterophorus* especially on flower extract. The seed germination, plumule, radicle length production were reduced with increasing concentration of aqueous solution. As per this study, increasing concentration of extract of *Parthenium hysterophorus* has adverse effect on germination, radicle length, and plumule length. Root and shoot length of seedlings decreased significantly ($p < 0.05$) with increasing concentration of Parthenium. Maximum root (7.00 cm) for wheat and (5.5 cm) for barley and shoot length (6.00 cm) for both wheat and barley were obtained for control (0%) in which the value reduced to 0 cm for both of them at a higher Parthenium concentration 10%, respectively. Qualitative analysis of aqueous Parthenium hysterophorus leaf, root, stem and flower extract was studied for the presence of carbohydrate, alkaloid, steroid, sterols, glycosides, tannin, phenolic compounds, flavonoid, oil, carbohydrate, protein, amino acid and were determined by testing with chemical laboratory in the presence of reagent.

Key words: Aqueous Extract, Chemical Extract, Phytochemicals, Allelopathy, Seed Germination, Seedling Growth

Full length article **Corresponding Author*, e-mail: solosime@gmail.com/solomon.sime@amu.edu.et and kiyyach@gmail.com/alemu.mekonnen@amu.edu.et

1. Introduction

1.1. Background of the Study

Parthenium hysterophorus, enlisted in Global Invasive Species database, is a highly prolific and malicious weed, which was first seen growing in Northeast of Mexico when *Parthenium confertum* and *Parthenium bipinnatifidum* were naturally hybridized [1, 2]. Being introduced for the first time into Ethiopia during the Ethio-Somali war in 1978, it has now been widely disseminated to every corner of the country and currently, it is the dominant weed in eastern Ethiopia [2, 3]. It is one of the ten worst weeds in the world, popularly known as Congress weed, Gajar Ghas, Carrot weed, Star weed, Fewer few, White top, Chatak Chandani, Bitter wood and Ramphool etc. The plant is known to be allergenic and is poisonous to animals [4]. It grows abundantly as a weed in India and was collected from Machillipatnam region, Andhar Pradesh.

Chemicals released from plants and imposing allelopathic influences are termed as allelochemicals or allelochemics and they can exist in several parts of plants including roots, rhizomes, leaves, stems, pollen, seeds and flowers. They are released into the environment by root exudation, leaching from above ground parts and volatilization and/or by decomposition of plant material [5].

Invasive species are recognized as one of the major threats to native species and ecosystem around the world. They are of major concern because of their capability of spreading fast, high competitiveness and ability to colonize new areas within short periods. Particularly, Parthenium has got main concern due to its invasive and allopathic properties [6]. The fast expansion of Parthenium to wheat and barley growing areas is observed to be a great menace for wheat and barley production. The mechanism of its spread in Ethiopia is not yet known. Furthermore, information on the allelopathic potential of Parthenium weed on wheat and barley seed germination and seedling growth is lacking. The present study gives reports on the effect of aqueous extracts of leaves, stems, roots and flowers part of Parthenium weed on wheat and barley seed germination and seedling growth under laboratory conditions.

2. Materials and Methods

2.1. Sample Collection

The sample *Parthenium hysterophorus* leaf, root, stem and flower were collected from Arba Minch University Abaya campus. The collected leaves, roots, stems and flower was cleaned thoroughly allow to shade dry and ground to powder using blender for further investigation. The gummy residue obtained was used for the analysis of percentage yield, behaviour of leaf powder and the remaining marc left was extracted with water and used for qualitative analysis and ground to powder using blender for further use.

2.2. Chemicals and Apparatus

Apparatus: soxhlet extractor, rotary evaporator, beaker, test tube, volumetric flask, filter paper, water bath, evaporating dish, electrical beam balance, droper, measuring cylinder, petri dish, distiller, conical flask, boiling chips, strirrer, iron stand, tample, grinding machine, ruler, condutometry, pH metre, spatula, labeling paper and test tube holder.

Chemicals: n-hexane, ethanol, distilled water, hydrogen peroxide, sulphuric acid, hydrochloric acid, sodium hydroxide, zinc dust, ferric chloride, potassium dichromate, copper sulphate, ninhydrin, sodium nitropreside solution, magnesium, iodine solution and α-naphtanol.

2.3. Extraction

The Parthenium plants grown naturally in Arbaminch University campus was uprooted and collected during their flowering stage. The plant was brought into laboratory and was immediately separated into leaf, stem, root and flower parts. Each part of the fresh plant will be cut into 2-3 cm and ground separately with a pestle and mortar. Aqueous extraction was prepared by dissolving 30g of powdered *Parthenium hysterophorus* of leaf, flower, stem and root in 200ml of hexane. The mixture of each powder was extracted by soxhlet extraction for 3hr. Then n-hexane was separated by rotary vapour. The standard solutions were prepared by taking 0, 1, 5 and 10g of each extracted aqueous solution and filled upto 100ml with distilled water and filtrates. Then solutions were kept in a volumetric flask and immediately allowed to test barley and wheat for five days.

2.4. Phytochemical Analysis

The extract was tested for the presence of bioactive compounds by adopting standard procedures of fluorescence analysis, behavior of drugs powder with different chemical reagents. The preliminary phytochemical analysis of n-hexane and aqueous extract of Parthenium was carried out using standard procedures to identify the phytochemical constituents (alkaloids, flavonoids, phenols, glycosides, cardiac glycosides, terpenoids, saponins, steroids, tannins, carbohydrates, proteins and amino acids) [7].

2.5. Application of Parthenin (on seed germination and seedling growth)

There were 16 treatments made at four concentration levels (0%, 1%, 5% and 10%) for each leaf, stem, root and flower extracts. Seeds and filter papers were moistened with 10ml of each of 1%, 5% and 10% aqueous extracts. 10ml of distilled water was added to the untreated control (0%). The treatments was arranged in completely randomized design (CRD) with three replications kept at room temperature on a laboratory bench with 12 h supply of fluorescent light during the night. The experiment was made in three replicates [3].

2.6. Data Collection and Analysis

Nine days after plating, data on germinated seeds (number of seedlings with visible shoot and root growth), non-germinated seed (number of seeds showing neither root nor shoot), and shoot and root length (mm) was recorded. The average data obtained from the two experiments was subjected to Origin 8.

3. Result and Discussion

3.1. Phytochemical Screening

The n-hexane of *Parthenium hysterophorus* (leaf, stem, root and flower) were subjected to various qualitative tests for phytochemical constituents which revealed the presence of diverse constituents that are shown in Table 1. The result revealed positive result for carbohydrate, alkaloids, steroids, sterols, saponin, tannin, phenolic compound, amino acids and oil and negative result for glycosides, flavonoids, protein and amino acid in n-hexane. The phenolic compounds are one of the largest and most ubiquitous groups of plant metabolites. Earlier workers reported that Parthenium has both negative and positive allelopathic effects on many agricultural crops and other plant species [8]. The allelochemicals responsible for affecting many plant species are sesquiterpene, lactone and phenolics. The allelopathic effect of Parthenium on other plants is largely attributed to the presence of Parthenin which is found in various parts of the weed which can be leached out from this plant parts when alive or dead. It has been reported that Parthenium also released phenolic acid from its roots and leaves.

Table 1: Qualitative Analysis of Phytochemical in Aqueous Parthenium hysterophorus Extracts (Root, Leaf, Stem and Flower)

Sr. No	Name of the Test	Result

1	Test for Carbohydrate:	
	Root	+++
	Leaf	-----
	Steam	+++
	Flower	+++
2	Test for Alkaloid:	
	Root	+++
	Leaf	+++
	Steam	+++
	Flower	+++
3	Test for Glycosides:	
	Root	-----
	Leaf	-----
	Steam	-----
	Flower	-----
4	Test for Saponins:	
	Root	+++
	Leaf	+++
	Steam	+++
	Flower	+++
5	Test for Flavonoids:	
	Root	----
	Leaf	----
	Steam	----
	Flower	----
6	Test for Tannin and Phenolic Acid Compound:	
	Root	+++
	Leaf	+++
	Steam	+++
	Flower	+++
7	Test for Protein and Amino Acid	
	Root	+++
	Leaf	----
	Steam	----
	Flower	----
8	Test for Fixed Oil:	
	Root	+++
	Leaf	+++
	Steam	+++
	Flower	+++

3.2. Germination Percentage

As is shown in table, concentrations of Parthenium significantly affected the germination of all the test species. There was slight inhibitory effect of Parthenium extract concentration on the wheat and barley seed germination while inhibitory effect on rest of species was comparatively greater.

Germination percentage of pulses decreased with increasing concentration of Parthenium. Similar effect is also seen in the two crop species (Table 2). The pulses recorded lowest germination percentage at 10% concentration of extracting flower (0%). The wheat and barley registered highest germination percentage of 28% and 21% at a concentration of 1% of root and flower respectively. The allelochemicals present in the leaf extract prevented the embryo development and its growth and led to death. The extract of *Parthenium hysterophorus* induced a variety of chromosomal aberrations in dividing cells, which increased significantly with increasing concentrations and durations of exposure.

Table 2: Effect of Aqueous Leaf, Stem, Flower and Root Extract of *Parthenium hysterophorus* on Seed Germination of Wheat and Barley

Conc.	Germination (%) (W=Wheat and B=Barley)							
	Leaf		Stem		Root		Flower	
	W	B	W	B	W	B	W	B
0%	36.6	18.8	36.6	20.0	36.66	20	36.66	18.8
1%	15.5	13.3	12	18.8	28	18.8	21	10
5%	8.8	5.5	7.7	15.55	16.6	8.8	7.7	1.1
10%	7.7	4.4	3.3	8.8	2.2	8.8	4.4	0

In this study, reduction in seed germination was observed with increasing concentration of aqueous extract of Parthenium in pulses. As compared to the control (0%), the aqueous extract of Parthenium from leaf and flower parts at 5% and 10% concentration levels exhibited significant ($P < 0.05$) inhibition on seed germination. Complete failure of seed germination was recorded as a result of application of 10% aqueous extract from flower. Aqueous extracts from stem and root had shown no adverse effect on seed germination. Leaf and flower extracts at lowest (1%) concentration had little impact on seed germination.

The significant reduction in wheat and barley germination was observed due to application of Parthenium leaf and flower extracts at intermediate to higher concentrations. This indicates the availability of inhibitory chemicals in higher concentration in leaf and flower parts than in stem and roots. Thus, early removal of Parthenium weed from wheat and barley field is highly desirable if productivity is to be maintained. In the present study, roots appeared more sensitive to the allelopathic extracts than shoots. The stronger inhibitory effects that Parthenium extracts had on roots might have been caused by the fact that roots were in direct contact with extract and subsequently with inhibitory chemicals as described in earlier works with various crops and weeds. The reduction in root length may indicate that cell division was affected as allelopathic chemicals have been found to inhibit gibberellin and indole acetic acid function. This type of growth may expose the crop to lodging, which is a major constraint in wheat and barley cultivation next to weed problems.

3.3. Seedling Growth

There were significant differences ($P<0.05$) between treatments in influencing seedlings shoot and root length. As compared to control, except aqueous extracts from leaves which had a deleterious effect on shoot length, those extracts from stem, flower and root seemed to have a stimulatory effect on plumule length of wheat and barley. Stem extract in low concentration (1%) greatly promoted shoot length. Similarly, stem and root extracts at 5% and 10% and flower extract at 1% and 5% showed a stimulatory effect on shoot length. Aqueous extracts from root at 1% promoted radicle growth of wheat and barley as compared to control. However, as root extract concentration level increased from 5% to 10%, root growth was gradually retarded. Stem extracts in low concentration (1%) retarded

root growth more than at higher concentrations (5% and 10%). In contrast, extracts from leaf and flower parts exhibited higher root growth inhibition even at low concentration levels root parts exhibited allelopathic activity on wheat and barley seed germination and seedling growth. Allelopathic activity depended on the concentration levels of the extracts and the parts of the weed from which they were extracted. Among 1%, 5% and 10% extract concentrations, 10% concentration was generally found to be effective and even more powerful especially when it was extracted from Parthenium leaves, in causing inhibitory allelopathic impact. Leaf aqueous extracts of 10% resulted in complete failure of seed germination and flower extract of 5% and 10% also inhibited germination to a greater extent. Roots appeared more sensitive to allelopathic effect than shoots. Growth of shoot length was promoted by stem extracts in all concentration levels, whereas root extract of 1% promoted root length.

3.4. Plumule Length

Different concentrations of Parthenium had significant effects on plumule length of wheat and barley. Seedling length of wheat and barley was significantly decreased with the increase in concentration of Parthenium extracts from 0% to 10%. The shoot length of all the test species (wheat and barley) at 10% concentration was significantly different from that of control; whereas at the same concentration root length was significantly different from the control (Table 3). In the present study, the reduction in plumule length was observed in increasing concentration of Parthenium. Higher concentration of Parthenium retard the growth of plants which might be due to inhibition of cell division as allelopathic chemicals have been found to inhibit gibberellin and indole acetic acid function which causes reduced plumule length. Reduction in plumule length was due to the presence of allelochemical (Parthenin) in leaf extract. This parthenin content present in aqueous extract leads to phytotoxicity of the emerged plumule growth in seeds. The inhibition of shoot elongation caused by allelochemical leads to reduced plumule length.

Table 3: Effect of Aqueous Leaf, Steam, Flower and Root Extract of *Parthenium hysterophorus* on Plumule Length of Wheat and Barley

Plumule Length of Wheat (cm)					Plumule Length of Barley (cm)			
Conc.	**Leaf**	**Stem**	**Root**	**Flower**	**Leaf**	**Stem**	**Root**	**Flower**
0%	6.00	7.8	7.2	7.4	6	5	4.8	4.8
1%	6.5	5.5	7.7	6.3	4.6	5.5	5	6
5%	3.4	6.3	4.8	1.5	3.6	3.8	6.1	3.1
10%	2.5	4.5	4	0	4	3.2	6	0

3.5. Radicle Length

The root length of crop decreased with increasing concentration of Parthenium. The maximum decrease in root length was recorded in wheat and barley in 10% concentration. The reduction in radicle length (Table 4) was observed in all the test crops with increasing concentration of extracts. The radicle length was affected more, because of reduction in root elongation. This is due to contact of root outer surface to leaf extract. Similar effect of leaf aqueous extract of *Parthenium hysterophorus* was reported in cereals. Among the plumule and radicle length, the radicle length trend showed a rapid reduction than the plumule length in all the crops. Because the radicle had more area of root surface exposed to the allelochemical. The strong inhibitory effects that *Parthenium hysterophorus* had on root elongation might be due to direct contact of root than the shoot with the extract and subsequently with inhibitory chemicals as described in early works with various crops and weeds.

Table 4: Effect of Aqueous Leaf, Stem, Flower and Root extract of *Parthenium hysterophorus* on Radicle Length of Wheat and Barley

Radicle Length of Wheat (cm)					Radicle Length of Barley (cm)			
Conc.	**Leaf**	**Stem**	**Root**	**Flower**	**Leaf**	**Stem**	**Root**	**Flower**
0%	7	6.6	5	6.6	5.5	3	5.5	5.5
1%	5.2	5.8	4.3	7	4.2	4	6.6	5.3
5%	4.9	4.7	6.3	2.5	3.6	4.5	7.2	4
10%	4.3	4.2	5.2	0	2.8	3.6	5.4	0

The inhibitory effects of Parthenium weed on wheat and barley seed germination percentage and seedling growth can probably be attributed to the allelopathic nature of extracts. Different kinds of allelopathic chemicals were reported to be released from Parthenium leaves, stems, roots and flower). Sesquiterpene, lactones and phenolics are thought to be the water soluble compounds involved in allelopathic activities. These chemicals were reported to have had allelopathic potential on various agronomic crops and weeds, vegetable crops and multipurpose trees.

Even though the effects obtained under laboratory conditions are not necessarily significant in the field, it can be concluded that there are compounds in the tissues of Parthenium which may cause allelopathic effects also under field conditions if the compounds are released in some way. The allelopathic mechanisms might have allowed the Parthenium weed to compete strongly with wheat and barley and spread itself widely in wheat and barley fields. The types and levels of concentration of these chemicals in different parts of the weed, however, need to be established.

Parthenium residues have inhibitory effects on the germination and growth of many plants such as radish, mustard and other plants. It has also been reported that a decrease in phytotoxicity was observed with increasing age of residue. The reduction in inhibitory activity of the allelochemicals over time is generally related to degradation mostly by soil microorganisms. Allelochemicals are subject to various biotic and abiotic processes that reduce their persistence, concentrations, availability and biological

activities after they are released into the soil. Such processes embody utilization by soil microorganisms, chemical transformation and polymerization among others [9].

It is clear from the present study that parthenin exhibit an inhibitory effect on the germination and growth of both the test seed species wheat and barley. From the study, it is also clear that parthenin exerted more effect on root than on shoot. Thus, parthenin causes considerable toxicity to test weeds in soil too. However, much needs to be done in this direction as regards its fate and dynamics in soil. As regards the biodegradation of the parthenin in the soil, no study is available. However, in aquatic environment, the phytotoxicity of parthenin is gradually lost in about 30 days under outdoor conditions making lethal dose unlethal. From the present study, it could be concluded that parthenin possesses weed suppressing ability that can be utilized for future weed management strategies [10].

4. Conclusion

The present project work was established that the effect of *Parthenium hysterophorus* on seedling germination and seedling growth of barley and wheat. Among the two seeds, barley was more affected by *Parthenium hysterophorus* extracts compared to that of wheat. The concentration dependent inhibitory activities of the aqueous n-hexane extracts of Parthenium on the germination and seedling growth of the test species suggest that the plant has allelopathic potentiality and possess allelochemicals. These allelochemicals could be the main reason for the restricted growth of other plant species near their colony. In line with this, this study showed that allelopathic weed plants pose threat on seed germination, seedlings radicle and plumule lengths of wheat and barley. Leaf, flower, root and stem extracts of *Parthenium hysterophorus* are found to inhibit wheat and barley seed germination and seedlings growth. Thus, farmers should give special attention to avoid or minimize those weed species from their farm to contain their adverse effects on crops. The present work also established that the *Parthenium hysterophorus* of leaf, flower, root and stem contains most of phytochemical when tested qualitatively and also shows considerable amount of saponin, fixed oil, alkaloids, phenolic acid compounds and absence (gives negative test) in flavonoid and glycosides. Root was the only containing protein and amino acid compared to flower, steam and leaf.

5. Acknowledgments

The authors are grateful to Arba Minch University College of Natural Science, Department of Chemistry for providing equipment and laboratory facilities.

6. References

[1] A. Saini, N.K. Aggarwal, A. Sharma, M. Kaur, A. Yadav. (2014). Utility potential of *Parthenium hysterophorus* for its strategic management. Advances in Agriculture. 2014:16.

[2] A. Netsere, E. Mendesil. (2012). Allelopathic effects of *Parthenium hysterophorus* L. aqueous extracts on soybean (*Glycine max* L.) and haricot bean (*Phaseolus vulgaris* L.) seed germination, shoot and root growth and dry matter production. Journal of Applied botany and food quality. 84(2): 219.

[3] T. Tefera. (2002). Allelopathic effects of *Parthenium hysterophorus* extracts on seed germination and seedling growth of Eragrostis tef. Journal of Agronomy and Crop Science. 188(5): 306-310.

[4] P.J. Biswanath Das, S. Malampati, K. Jajula and D. Ratna. (2013). Pseudoguaianolides from a collection of the flowers of *Parthenium hysterophorus* L. (Compositeae). Journal of Organic Molecular Chemistry. 1: 195-200.

[5] O. Devi, B. Dutta. (2012). Allelopathic effect of the aqueous extract of *Parthenium hysterophorus* and *Chromolaena odorata* on Seed germination and Seedling Vigour of *Zea mays* L. Acade. J. Plant Sci. 5(4): 110-113.

[6] Basarkar and Saoji. (2013). Isolation, characterization of sesquiterpene parthenin and its estimation from *Parthenium hysterophorus* pollen. International Journal of Emerging Technologies in Computational and Applied Sciences. 5(4): 364-368.

[7] M. Krishnaveni and R. Dhanalakshmi. (2014). Phytochemical analysis of *Parthenium hysterophorus* L. leaf. World Journal of Pharmaceutical Research. 3(6): 1066-1074.

[8] D. Nganthoi, B.K. Dutta, R. Sagolshemcha and N.I. Singh. (2014). Allelopathic effect of *Parthenium hysterophorus* L. on growth and productivity of *Zea mays* L. and its phytochemical screening. International journal of current microbiology and applied sciences. 3(7): 837-846.

[9] W. Mulatu, B. Gezahegn and T. Solomon. (2009). Allelopathic effects of an invasive alien weed *Parthenium hysterophorus* L. compost on lettuce germination and growth. African Journal of Agricultural Research. 4(11):1325-1330.

[10] Daizy R. Batisha, Harminder Pal Singh, Ravinder K. Kohlia, Shalinder Kaura, Dinesh B. Saxenac, and Surender Yadav. (2007). Assessment of Phytotoxicity of Parthenin. Journal for Nature Research. 62:367-372.