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Submission date: 01-Jul-2020 01:41PM (UTC+0700)

Submission ID: 1352122965

File name: The_autoallelopathic_potential_of_the_Siam_weed.pdf (706.58K)

Word count: 2396

Character count: 12401

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To cite this article: D Ziadaturrif'ah *et al* 2019 *J. Phys.: Conf. Ser.* **1217** 012148

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The autoalelopathic potential of the Siam weed (*Chromolaena odorata* L.) leaf extract as a natural herbicide

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1. Introduction

Siam weed (*Chromolaena odorata* L.) is very detrimental because it can reduce the production of crops in plantations such as oil palm, rubber, cashew nuts and the growth of other plants around it Sindel and Coleman [14]. Siam weeds are thought to have a fairly high defense because it is very easy to grow even though it has been, so it is difficult to control [20].

Weed control methods can be carried out physically using manual and chemical removal using herbicides. Both methods of control have advantages and disadvantages. The weakness of manual weed control is that it requires many workforces, especially on large areas that have high weed populations besides it must be done more than once because the manual control does not kill weeds, so the production costs are higher. Manual weed control also takes a long time to encourage farmers to use synthetic herbicides that are faster and easier to use. However, on the other side, synthetic herbicides have a negative impact in the form of environmental damage such as environmental pollution, increase the weed resistance, increase residues in agricultural products, make some natural enemies die and damage nature both temporarily and permanently [17].

Environmental friendly weed control is an alternative treatment by utilizing the potential of allelochemical compounds derived from plants. These allelochemical compounds can suppress the



growth of plants around them so that they can be used as bioherbicides. One of the plants which are thought to have potential as a bioherbicide with the principle of allelopathy is the siam weeds because it contains active compounds such as alkaloids, tannins, flavonoids, saponins, and phenolics [5]. The results of Sari et al. [12] showed that the siam weeds extract can be used as an alternative pre-grown bioherbicide material to control weeds in oil palm plantations. The administration of pre-grown bioherbicides extracts significantly affected the growth of weeds and biomass. The concentration of the best siam weed extract was found in the treatment of 3% extract and was significantly different from herbicide glyphosate and control treatments.

The mechanism of inhibition by allelochemical starts from the entry of allelochemical on the plasma membrane of the target plant which causes loss of membrane permeability and the function of the ATPase enzyme. This process will affect the absorption of certain ions and water which then affect the opening of the stomata and photosynthesis process. The next obstacle occurs in the process of synthesis of proteins, pigments, and other carbon compounds, as well as the activity of several phytohormones. Some or all of these obstacles disrupt cell division and enlargement which ultimately inhibit the growth and development of plants Sharma et al. [13]. According to Singh et al. [15] and Sodaeizadeh et al. [19] the most fundamental allelochemical effect is disruption of the permeability plasma membrane that will cause a decrease in H⁺ + ATPase enzyme activity, degradation of the protein sulfhydryl group, decrease in transmembrane electrochemical potential, and membrane depolarization. This membrane depolarization causes changes in membrane structure. The non-specific changes in anion and cation exchange may increase lipid peroxidation and loss of membrane integrity electrolytes so that the cell membrane will shrink, followed by absorption inhibition of certain ions, disrupt pigment synthesis, affect mitochondrial function for respiration, and disrupt hormone activity in the growth of target plants.

Autoallelopathy is an interaction between similar living things with the intermediaries of allelochemical compounds, which can kill or inhibit their own saplings or other plants of the same type (autotoxin) [8]. Djazuli [3] also mentions that weed roots produce organic acid compounds that are toxic in the allelopathic process which can inhibit the growth of weeds themselves or so-called (autotoxin). This study examined the efficacy of autoallelopathy of Siam weed extract on germination and initial growth of weed seeds so that it can be known the potential of siam weed extract as a natural herbicide to eradicate weeds of the same type.

2. Research Method

2.1. Tools and Materials

Tool and materials used are siam weed and seeds were obtained from Tembalang, Semarang, Central Java, Petri dishes, measuring cups, funnels, test tubes, test tube shelves, drop pipettes, analytic scales, blenders, label paper, cutters, and rulers.

2.2. Preparation of Siam Weed Extract

The extraction was carried out using the method of Darmanti et al. [2], the Siam weed was dried under room in dark conditions for 24 hours. The siam weed was mashed using a blender, then extracted with water at a ratio of 1: 1 weight/volume. The extract was filtered two times using a cloth and filter paper in a row to obtain a concentration of 100%; the extract was diluted with distilled water To achieved at the concentration of 5%, 10%, 15%, 20%, the extract was stored at low temperature. Control was prepared using tap water without any extract.

2.3. Germination and Treatment

The seeds of Siam weed were selected the dry brownish, or black color seed was chosen. The seed germination was carried out on a petri dish that has a cotton pad and filter paper. Into each Petri dish, 10 of siam seed was prepared. The seeds were soaked in the extract that was prepared before. This extract was replaced by everyday. The treatment was stopped after 100% germination occurred At least in one petri dish. All parameters were measured at the end of the study.

2.4 Observations of Parameter

Germination parameters include germination rate and germination percentage, while the initial growth parameters of sprouts include hypocotyl and radicle length and also a fresh weight of sprouts. Germination rate was calculated using the formula according to Matthews and Khajeh [11], while the germination percentage was calculated using the formula according to Akinci and Sermin [1].

$$\text{Germination Rate (LP)} = \frac{N1.T1+N2.T2+N3.T3+\dots+NX.TX}{\text{Total number of seeds}}$$

N: number of sprouts that appear at a certain time unit (once every 24 hours)

T: the amount of time between the start of the test to the end of a certain interval

$$\text{Germination percentage (\% P)} = P / T \times 100$$

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P: Total number of germinating seeds

T: Total number of seeds in the experiment).

2.5 Data Analysis

The resulted data was analyzed using ANOVA Test with a confidence level of 95%. If the results obtained are significantly different, further testing is carried out by Duncan's test of 5% significance level.

3. Results and Discussion

The results showed that the extract of Siam weed reduced the germination rate, the higher the extract concentration been applied, the higher the inhibition on the rate of germination (Figure 1.)

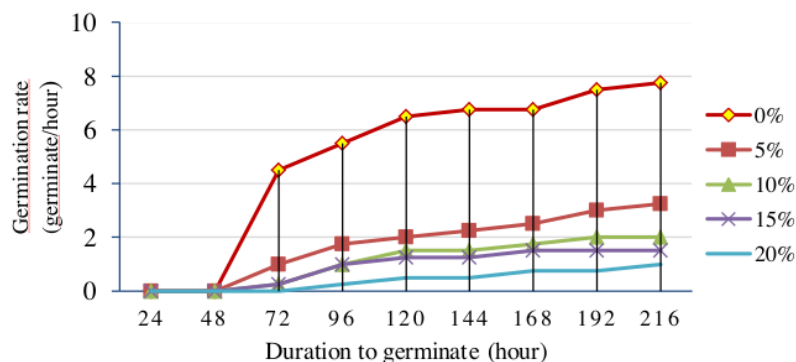


Figure 1. The germination rate of the Siam weed (*Chromolaena odorata* L.) seeds after treated with siam weed extract at different concentration during 216 hours.

It is showed in figure 1 above that there is a difference in the time to germinate the Siam weed seeds in Petri disk. In controls, it was found that the highest rate of germination was resulted compared to siam weed extract treatments. The treatment of 20% extract concentration shows a lower germination rate compared to the treatment of other extract concentrations. Siam weed extracts

application resulted in a reduction of germination percentage addition of higher extract concentration, the germination percentage of siam seed is lower.

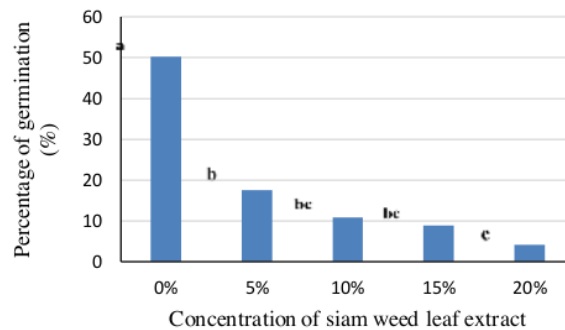


Figure 2. Percentage of siam weed seeds germination due to the treatment of Siam weed extracts during 216 hours.

The initial growth parameters for siam weed seed sprouts after the addition of siam weed extract were shown in Table 1. and Figure 3.

Table 1. Hypocotyl (cm), radicular length (cm) and fresh weight (mg) of Siam weed sprouts treated with siam weed extract for 216 hours.

Extract Concentration	Hypocotyl Length (cm)	Radicula Length (cm)	Fresh Weight (mg)
0%	1.31 ^a	0.68 ^a	1.9 ^a
5%	0.48 ^b	0.16 ^b	0.8 ^b
10%	0.29 ^{bc}	0.08 ^c	0.4 ^{bc}
15%	0.17 ^c	0.07 ^c	0.3 ^c
20%	0.13 ^c	0.04 ^c	0.3 ^c

- Numbers followed by the same letter in the same column show an effect that is not significantly different based on the Duncan test with a confidence level of 95%.

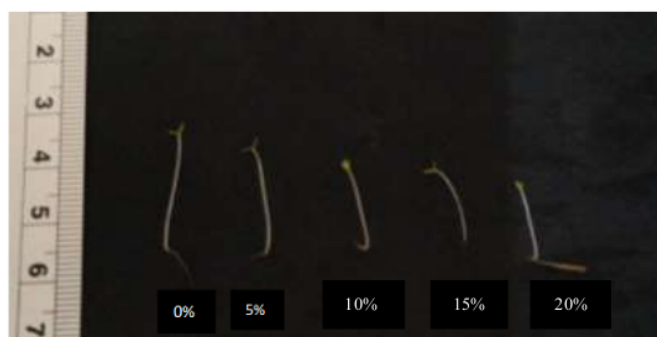


Figure 3. Siam weed sprouts with a concentration of Siam weed leaf extracts (*Chromolaena odorata* L). for 216 hours.

The study results of the initial growth parameters of sprouts show that allelochemical of the Siam weed leaf extract reduced the hypocotyl and, radicle length and fresh weight of Siam weed sprouts. The higher the extract concentration, the inhibition is higher.

4. Discussion

Germination begins with an imbibition process which will trigger the formation of a small amount of the next gibberellin hormone which then activating the enzyme. The enzyme will diffuse into the endosperm and catalyze the food reserves used for the initial growth of the embryo and the rupture of the seed membrane so that the radicle will grow geotropically Song Ai et al. [16].

Allelochemical can reduce seed germination and slow down time to germinate because allelochemical enters the seeds through the plasma membrane then attaches to phospholipids which causes a decrease in fat solubility in the membrane and causes the plasma membrane to shrink so that the membrane permeability becomes damaged Li et al. [10]. Also, Isda et al. [7] stated that the allelochemical would inhibit the subsequent imbibition process, allelochemical compounds disrupted the activity of the gibberellin hormone which resulted in the inhibition of the germination enzymes activity in the form of α amylase enzyme, β amylase, protease enzyme, and lipase enzyme. These enzymes will diffuse into the endosperm and catalyze food reserves into sugars, amino acids and fatty acids that support the growth of the embryo during germination. Disruption of these enzymes results the energy that is used for growth is very little which caused a slow growing so that the ability of the seeds to germinate decreases. It will have an impact on the percentage of siam weed seed that germinates, and the sprout emerges more slowly. It is consistent with the research result of Frastika et al. [5] which stated that the concentration of 15% of Siam weed extracts could inhibit the germination rate of karuilei *Mimosa invisa* and green bean *Vigna radiata*. Also, the research by Hu et al. [6] which stated that siam weed extract start at a concentration of 2 g / L can inhibit the germination percentage, germination index and hypocotyl length of *Pueraria phaseoloides*, *Urena lobate*, *Vitex negundo* and *Caryopteris incana*. ¹¹

The research results showed that the higher the treatment concentration, the lower the average of hypocotyl length, the length of the radicle and the wet weight of the germination. It is consistent with the results of a study by Karim et al. [9], which states that siam weed extract at a concentration of 4% can reduce root length, and hypocotyl length of bean sprouts, peanut, mustard, and rice as much as 42%. Enyi's research [4] showed that the water extract of the Siam weed could reduce the fresh weight and dry weight of tomatoes (*Lycopersicum esculentum* Mill), while the Suwal study [18] showed that the water of Siam weed extract gave the greatest inhibitory effect in reducing germination and initial growth of sprouts rice and duck grass (*Bernyard grass*) compared to stem extracts and root extracts. The decrease in the initial growth parameters of sprouts is due to the presence of phenolic (tannin and coumarin) allelochemical compounds in the weed extract that have an ability to inhibit growth hormone induction such as gibberellin (GA) which plays a role in germination, indole acetate acid (IAA) which plays a role in cell elongation, and cytokines which plays a role in cell division, so the formation of α amylase and β amylase enzymes in the process of hydrolysis of starch into glucose, proteases in the process of hydrolysis of proteins into amino acids, lipases in the process of hydrolysis of lipids into fatty acids are inhibited in the endosperm or cotyledons. It causes inhibition of protein synthesis which will also result in inhibition of protoplasmic synthesis. Therefore the process of cell division and elongation is inhibited, which results in inhibiting hypocotyl and radicle elongation as well as having a direct impact on the reduction in wet weight of Siam weed seeds Yuliani et al. [21].

5. Conclusion

The higher the extract concentration, the higher the inhibition on germination and initial growth of Siam weed sprouts.

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