Allelopathy of Ethanol, Ethyl Acetate, and Aquadest Extracts of Kirinyuh (*Chromolaena odorat*a) Leaves as a Bioherbicide in Controlling *Spenochlea zeylanica*

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Abstract. *Spenochela zeylanica* is the most prevalent weed of paddy rice in Indonesia, and therefore its presence necessitates control in order to reduce the potential for yield losses. The use of chemical herbicides as a control method is prevalent, but their environmental impact and potential for inducing weed resistance are significant drawbacks. It is therefore necessary to identify control methods that are environmentally friendly and capable of overcoming weed resistance. The leaves of *Chromolaena odorata* have allelopathic properties that could be harnessed for use as a bioherbicide. The objective of this study is to evaluate the efficacy of *C. odorata* leaf extractions using diverse organic solvents as a bioherbicide for the management of *S. zeylanica*. This study used a randomized group design (RBD) with 11 treatments and 3 replications, including: T0: no treatment; 96% ethanol extraction: T1(5%), T2(10%), T3(15%); ethyl acetate extraction: T4(5%), T5(10%), T6(15%); aquadest extraction: T7(5%), T8(10%), T9(15%); T10: 2,4-D herbicide (648 g a.i./ha). The findings indicated that the extraction of *C. odorata* using various organic solvents exhibited promise as a bioherbicide, displaying the capacity to poison weeds up to 100% (T2, T3, T6, T8, T9) and effectively suppressing the height, root development, and chlorophyll content of *S. zeylanica*.

Keywords: Allelopathy, bioherbicide, organic solvents, weeds

INTRODUCTION

Weeds can negatively affect rice production and the quality of the harvested rice when they are present in paddy fields. Plant pests known as weeds can create competition for resources such as light, water, nutrients, and growing space. The absorption and usage of these resources may be impacted by this competition, which could ultimately lower the rice crop's productivity and quality [1]. The results of [2] show that crop output can drop by as much as 45–60% when weed control strategies are not used when growing rice. Thus, in order to sustain ideal output levels, it is imperative to put into practice efficient weed management measures. According to the findings of the dominant weeds identification projects carried out by [3], [4], *Spenochloa zeylanica* is a common broadleaf weed in paddy rice fields. Controlling its presence is necessary to avoid any possible losses in rice production.

Herbicides are the most widely used chemical control approach since they are time- and money-efficient, as well as very effective. Herbicide use for weed control is a technique that is spreading throughout the world [5]. Herbicide use is a key tactic in reducing possible yield losses in plantation and agricultural operations. On the other hand, long-term use of herbicides can lead to the development of weed resistance, damage soil structure [6], cause pollution of the environment, and endanger the health of the main crop [7].

Herbicide residues might interact with soil particles and plant roots due to their persistence in the soil. The surrounding environment may suffer as a result of this interaction [7]. Moreover, consumers of agricultural products may be at risk for health problems due to herbicide residues. The research of [8] show that eating food exposed to glyphosate herbicides can have long-term impacts on people, including the occurrence of biochemical reactions from oxidative stress that damages DNA and causes cancer and other disorders. Finding substitute weed-control technologies that are safer for people and the environment is therefore essential.

One potential solution for the control of weeds in a manner that is both safe and environmentally friendly is the utilization of bioherbicides, which employ allelopathic compounds. Allelopathy is a secondary metabolic process that can inhibit the germination, growth, and development of a plant. The detrimental effects of allelopathic chemical compounds (allelochemicals) on plants can be attributed to disturbances in various physiological processes, including cell membrane permeability, water and nutrient transport, respiration, protein, nucleic acid synthesis, and photosynthesis rates [9]. Consequently, plants with allelopathic compounds may serve as bioherbicides in environmentally conscious weed management [10].

Indonesia has a huge opportunity for the development of plant allelopathy-based bioherbicides due to its tropical climate. A vast range of plants with allelochemical substances that can be utilized as raw materials to make ecologically benign pesticides are made possible by Indonesia's great biodiversity [11]. *Chromolaena odorata*, often known as kirinyuh, is one plant that shows promise as a bioherbicide. Strong allelopathic qualities of this plant are known to stunt the growth of nearby plants [12]. With this characteristic, natural herbicides that are less harmful to the environment than synthetic ones can be created.

One of the most troublesome invasive weeds is kirinyuh (*C. odorata*), which has an amazing ability to adapt to even the most difficult situations, such as barren agricultural land and plantations. This implies that using *C. odorata* as a bioherbicide would be a good option. Allelopathic qualities of *C. odorata* explain its higher frequency on agricultural land compared to other weeds [13]. Terpenoids, flavonoids, tannins, steroids, and alkaloids have all been identified in *C. odorata* leaves [14], [15]. It has been shown that the leaf extract of *C. odorata* inhibits the growth and germination of a number of weed species, including *Eleusine indica*, *Cyperus iria*, and *Ageratum conyzoides* [16], *Echinochloa crus-galli* and *Amaranthus viridis* [12].

The ability of *C. odorata* leaves to function as a bioherbicide allows for the support of sustainable agriculture initiatives without the use of hazardous chemical residues. This study uses allelopathic kirinyuh (*C. odorata*) leaf extract as a bioherbicide to manage rice weeds (*S. zeylanica*) in order to evaluate the efficacy of a health- and environmentally-friendly method.

METHODS

This research will be conducted at Teaching Farm Polinela Organic Farm (TeFa POF) Politeknik Negeri Lampung (Polinela), Bandar Lampung, Indonesia. The research was conducted from May to September 2024. This study used a randomized group design (RBD) with 11 treatments and 3 replications, including: T0: without treatment; 96% ethanol extraction: T1(5%), T2(10%), T3(15%); ethyl acetate extraction: T4(5%), T5(10%), T6(15%); distilled water extraction: T7(5%), T8(10%), T9(15%); T10: herbicide 2,4-D (648 g a.i./ha). The bioherbicide material of kirinyuh leaves (*C. odorata*) was taken at the Polinela labor field at 112 meter above sea level (masl) (Lat: -5.353577939439514, Long: 105.22842507658335). *S. zeylanica* weeds were taken in the form of seedlings with a height of ± 10 cm and had 4 or more leaves in paddy rice fields in Natar District, South Lampung Regency, Indonesia at 105 masl (Lat: -5.343611, Long: 105.227500).

The preparation of *C. odorata* bioherbicide extracts was carried out in accordance with the methodology outlined by [12], with modifications. The extract was obtained via the maceration method, utilizing solvents including 96% ethanol, ethyl acetate, and distilled water. A total of 500 g of *C. odorata* leaf powder was placed into a dark glass bottle, filled with 2000 mL of each solvent (96% ethanol, ethyl acetate, and distilled water), stirred, and sealed. Maceration was conducted at room temperature for up to 72 hours. Following this period, the maceration results were filtered using the Whatman No. 42 filter paper. All collected filtrates were subsequently concentrated using a rotary evaporator at 60°C, resulting in a thick liquid.

Herbicide application was carried out when the weeds were ± 7-14 days after transplanting or had reached a height of ± 20 cm and had 10 leaves. The application is carried out in the morning using a sprayer at a predetermined concentration. Spraying is done with several dose levels, starting from the lowest dose to the highest dose.

The variables observed were as follows: (1) Weed height (cm), *S. zeylanica* was measured with a ruler from the base of the stem parallel to the soil surface to the tip of the highest leaf; (2) Chlorophyll (SPAD Unit), chlorophyll content of *S. zeylanica* due to bioherbicide treatment was observed using Portable SPAD-502Plus; (3) Toxicity (%), determination of percent toxicity was carried out by comparing treated weed with untreated weed (control). Comparisons were made between the observed leaf color, changes in leaf shape, and abnormal growth. From this comparison, the percentage of weed poisoning can be calculated. The observations were conducted using a visual method, with two individuals [1] performing the assessment. The observation of the level of plant poisoning is in accordance with the standards set forth by [17] for the standard method of herbicide efficacy testing (**TABLE 1**). Observations of height, chlorophyll, and percent poisoning variables were conducted every two days, beginning two days after treatment (DAT) and continuing until 14 DAT. (4) Root length (cm), *S. zeylanica* root length was obtained from the final observation at 14 DAT. The roots were initially washed and then measured with a ruler. (5) Wet weight and dry weight (g), wet weight was obtained from the last observation of 14 DAT, weeds were harvested and weighed. Dry weight is obtained by putting weeds and roots that have been weighed wet into a closed paper envelope and then in the oven at 80˚C for 48 hours. (6) Lethal Time 50% (LT_{50}), the time required for each bioherbicide to toxic weeds by 50%. The LT_{50} value is obtained through a simple linear regression equation, namely y = a + bx, where the y value is the probit value of the percent weed toxicity and x is the log DAT of the bioherbicide, then after the x value is known, the LT_{50} can be found by antilog the x value. (7) Effective dose 50% (ED_{50}), the number of bioherbicide doses that cause weed suppression up to 50%. The weed dry weight data obtained was then converted into a percentage value of weed damage. Percentage damage can be obtained through the following equation:

$$\% \text{ Damage} = \left(1 - \left(\frac{P}{K}\right)\right) x\ 100\%$$

Note: P = dry weight value of weeds treated with bioherbicide; K= dry weight value of no treatment weeds.
The percent damage was transformed into probit values with the assistance of probit tables. The dose levels tested were converted into log form. From the probit value of percent damage (y) and log dose (x), a simple regression equation was determined: y = ax + b. From this equation, the value of x was determined for y = 5, as the objective was to ascertain the ED_{50} (the probit value of 50% is 5). The value of x was then log transformed to obtain the ED_{50} of the bioherbicide.

To test the homogeneity of data variance, Bartlett's Test was used and the additivity of data was tested using Analysis of Variance (ANOVA) test. If the assumptions were met, then the data were analyzed by variance analysis and to test the differences in the treatment mean values were tested by Duncan Multiple Range Test (DMRT) at the 5% level. All data were analyzed using Microsoft Excel and Statistical Tool for Agricultural Research (STAR) software version 2.0.1.

TABLE 1. Rating system used to assess weed toxicity [17]

Effect	Rating (%)	Toxicity Description
No Effect	0	No crop reduction or injury
Low	10	Slight crop discolouration or stunting
	20	Some crop discolouration. Stunting. or stand loss
	30	Crop injury more pronounced. but not lasting
Moderate	40	Moderate injury. crop usually recovers
	50	Crop injury more lasting. recovery doubtful
	60	Lasting crop injury no recovery
Heavy	70	Heavy crop injury and stand loss
	80	Crop nearly destroyed. A few surviving plants
	90	Only occasional live crop plants left
Death	100	Complete crop destruction

RESULTS AND DISCUSSION

Height of *S. zeylanica* due to bioherbicide application of *C. odorata*

The results of the study (**FIGURE 1**) demonstrated that the extraction of C. odorata bioherbicide and 2,4-D herbicide using a variety of organic solvents was effective in suppressing weed growth, with the greatest effect observed between 2 and 14 DAT. However, the maximum suppression was not achieved with the 5% C. odorata extraction treatment using ethyl acetate as the solvent. The allelopathic effects of the C. odorata extract in treatments T2, T3, T6, and T9 exhibited the most pronounced suppression of S. zeylanica growth, resulting in a complete cessation of growth at 8, 6, 12, and 10 DAT . The term "allelopathy" is used to describe a biochemical interaction between plants whereby one species releases chemicals that can inhibit the growth of another species. This phenomenon is of particular significance in agricultural and ecological contexts, as it represents a natural method of managing weed populations. These allelochemicals have been demonstrated to affect a range of physiological processes in other plants, resulting in reduced germination rates, stunted growth, and decreased overall plant height [18]. The research conducted by [19] revealed that distilled water extracts of several plants, including *Wedelia trilobata* L., *Artemisia lavandulaefolia* DC. Prodr., and *Ipomoea cairica* (L.), had a significant inhibitory effect on the height of weed seedlings, such as *Bidens pilosa* and *Lolium perenne*. Furthermore, the suppression was more significant at higher extract concentrations, indicating a dose-dependent relationship.

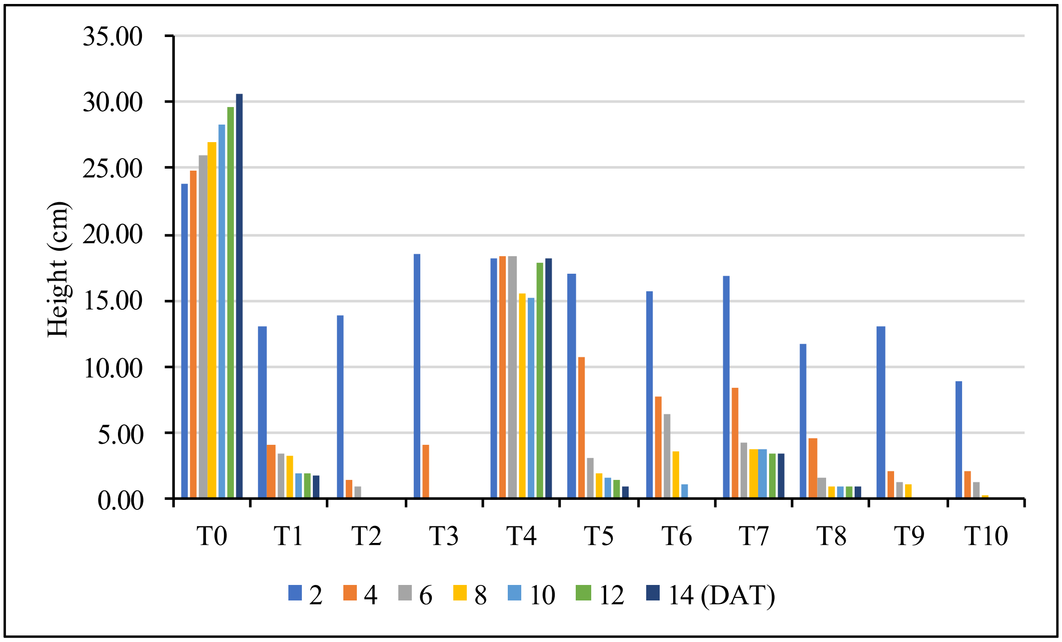

FIGURE 1. Height of S. zeylanica due to C. odorata bioherbicide application. Note: T0: control; 96% ethanol extraction: T1(5%), T2(10%), T3(15%); ethyl acetate extraction: T4(5%), T5(10%), T6(15%); aquadest extraction: T7(5%), T8(10%), T9(15%); T10: 2,4-D (648 g a.i./ha)

Chlorophyll of *S. zeylanica* due to bioherbicide application of *C. odorata*

The results of the study (**FIGURE 2**) showed that extraction using several organic solvents at the dose level of *C. odorata* bioherbicide and 2,4-D herbicide was able to reduce the chlorophyll of weeds starting from observation 2 - 14 DAT, this indicates that bioherbicide *C. odorata* is able to interfere with the chlorophyll performance of weeds so that the growth process does not run optimally. Not optimal results were shown in T4 (5%) extraction treatment of *C. odorata* using ethyl acetate solvent. Without treatment (control) also decreased due to the aging of *S. zeylanica* leaves. Allelopathy can significantly affect chlorophyll content in weeds, leading to reduced photosynthetic capacity and overall plant health. This occurs through the release of allelochemicals by plants that can inhibit the growth and physiological functions of surrounding plants. The presence of allelochemicals can induce a stress response in plants, leading to changes in chlorophyll content [20]. For example, a study conducted [21] that evaluated the effects of *Cyperus esculentus* allelopathy on aggressive weeds such as *Ipomoea tricolor* showed a significant reduction in chlorophyll content alongside other growth metrics. Findings indicated that higher residual concentrations of *C. esculentus* led to decreased levels of chlorophyll a and b, as well as total carotenoids in the target weeds. Another investigation on the allelopathic potential of aquadest extract of *Sorghum halepense* showed a marked decrease in total chlorophyll content and carotenoid levels in lettuce seedlings. This suggests a direct relationship between allelochemical concentration and chlorophyll suppression [22]. The same results have also been reported by numerous researchers, who have found that bioherbicide applications can result in a reduction in chlorophyll content in a number of plant species, including *Tagetes erecta* [23], *Pelargonium radula*, *Persicaria odorata*, *Plectranthus amboinicus*, *Murraya koenigii*, and *Cupressus macrocarpa* [24].

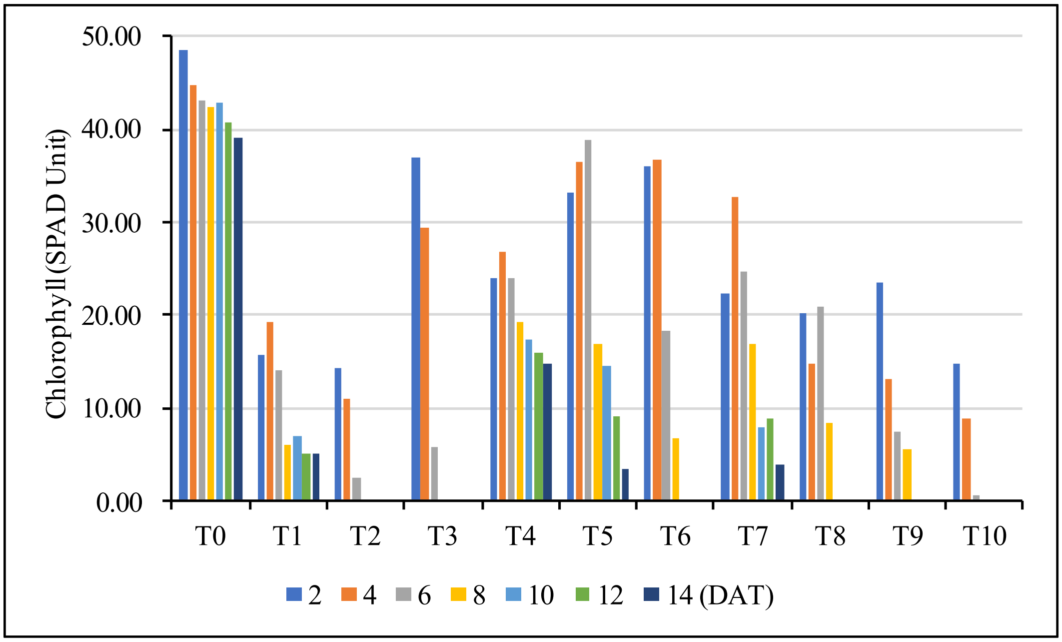

FIGURE 2. Chlorophyll of *S. zeylanica* as a result of *C. odorata* bioherbicide application. Note: T0: control; 96% ethanol extraction: T1(5%), T2(10%), T3(15%); ethyl acetate extraction: T4(5%), T5(10%), T6(15%); aquadest extraction: T7(5%), T8(10%), T9(15%); T10: 2,4-D (648 g a.i./ha)

Toxicity percent of *S. zeylanica* due to bioherbicide application of *C. odorata*

The application of *C. odorata* bioherbicide and 2,4-D herbicide was observed to cause toxicity of *S. zeylanica*, resulting in growth inhibition and subsequent mortality. This is illustrated in **FIGURE 3 & 4**, which depicts the toxicity percent observed in the study. The toxicity percent exhibited a gradual increase from 2 to 14 DAT. The higher the dose administered in each extraction, the greater the level of toxicity observed in *S. zeylanica*. The 96% ethanol extraction treatment with a dose of 15% (T5) and 10% (T4) elicited the most pronounced response, resulting in the death of *S. zeylanica* at 6 and 8 DAT. This outcome surpassed the efficacy of the herbicide 2,4-D, which took 10 DAT to achieve the same result. At 14 DAT, several bioherbicide treatments with various solvents were observed to have a greater than 85% efficacy in poisoning weeds. However, T4 (5%) exhibited relatively low results at 51.67%. These findings suggest that *C. odorata* extract has the potential to serve as an environmentally friendly bioherbicide. Bioherbicides derived from plant extracts have demonstrated promising potential in the management of weeds. Some plant extract compounds have been demonstrated to exhibit specific inhibitory activity against weed growth while simultaneously demonstrating minimal toxicity to the main crop [25]. This phenomenon may be attributed to disparities in sensitivity to the target enzyme or the existence of particular receptors on the weed that are capable of recognizing and reacting with the compound [26]. Specific plant species are capable of secreting a variety of metabolites, collectively known as allelochemicals, which include alcohols, fatty acids, phenolics, flavonoids, terpenoids, and steroids. These compounds have the ability to impede the reproduction, growth, and development of adjacent vegetation, including weed species [27]. [12] extracted several parts of *C. odorata* as a bioherbicide. The results demonstrated that the efficacy of *C. odorata* leaf extracts on weed germination and growth was superior to that of extracts from other parts, which is consistent with the findings reported by numerous researchers [28], [29]. Leaf extracts may be more phytotoxic because leaves are the main site of plant metabolism, and secondary metabolite levels are higher compared to other plant parts [30]. The mechanism of C. odorata leaves in poisoning weeds is a decrease in weed chlorophyll content over time after application, which disrupts photosynthetic metabolism and causes weed death [12].

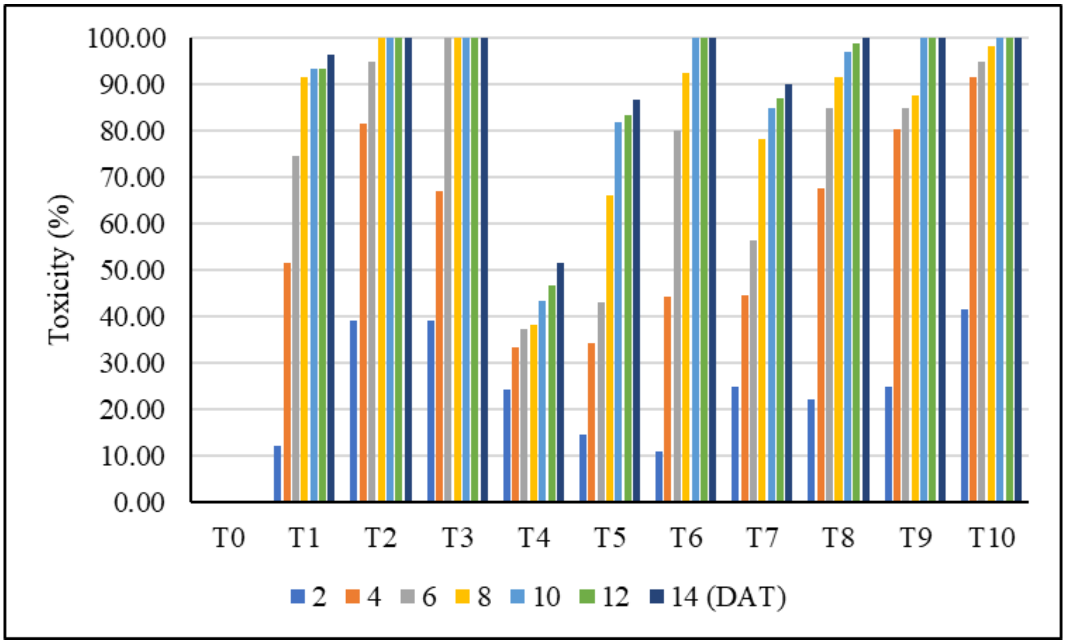

FIGURE 3. Toxicity percent of *S. zeylanica* as a result of *C. odorata* bioherbicide application. Note: T0: control; 96% ethanol extraction: T1(5%), T2(10%), T3(15%); ethyl acetate extraction: T4(5%), T5(10%), T6(15%); aquadest extraction: T7(5%), T8(10%), T9(15%); T10: 2,4-D (648 g a.i./ha)

FIGURE 4. Response of *S. zeylanica* due to application of *C. odorata* bioherbicide at 14 DAT. Note: T0: control; 96% ethanol extraction: T1(5%), T2(10%), T3(15%); ethyl acetate extraction: T4(5%), T5(10%), T6(15%); aquadest extraction: T7(5%), T8(10%), T9(15%); T10: 2,4-D (648 g a.i./ha)

Root length, wet weight, and dry weight of *S. zeylanica* due to bioherbicide application of *C. odorata*

The results, as presented in **TABLE 2**, revealed significant discrepancies in the observations of root length, wet weight, and dry weight as a consequence of the application of the *C. odorata* bioherbicide and the 2,4-D herbicide, with the exception of the T4 (5%) *C. odorata* extraction treatment utilising ethyl acetate as the solvent. These findings indicate that T4 is an ineffective method for controlling *S. zeylanica*, as evidenced by observations of plant height, chlorophyll content, and percent poisoning. The most effective root growth suppression was demonstrated by treatments T2, T3, T9, and T10. Inhibition of root growth results in suboptimal absorption of water and nutrients, which disrupts overall growth. The 96% ethanol extraction treatment (T3, 15%) and the 2,4-D herbicide (T10) were the most effective in terms of observing the wet and dry weights of the weeds. The observation of dry weight is a common practice in weed science, as it provides insight into the efficacy of a given treatment. The results of this study indicate that a 15% dose of *C. odorata* extract, extracted using 96% ethanol solvent, can produce the same level of control over *S. zeylanica* as the 2,4-D herbicide. This suggests that the use of *C. odorata* as an alternative control method may offer a more environmentally friendly approach. The allelopathic compounds present in *C. odorata* leaves may be responsible for this observed effect. The phenomenon can be attributed to the presence of allelopathic compounds in the leaves of *C. odorata*. The results of the content identification conducted by [31] revealed that the *C. odorata* leaf extract contains tridecane, tetracosane, hexadecane, and pentadecane. The identified compounds included nonacosane, octacosane, tetracosanol, eicosane, hexadecane, 1-hexadecanol, spathulenol, caryophyllene, copaene, globulol, eugenol, and cadinal. While essential oil from *C. odorata* was found to contain copaene, trans-caryophyllene, caryophyllene oxide, d-cadinene, germacrene-D, and humulene [32], The combination of several compounds found in C. odorata leaves is believed to be capable of synergizing in order to control weeds. In accordance with these findings, [33] indicated that some allelochemicals are capable of working synergistically or interacting with one another to form new compounds with inhibitory effects.

TABLE 2. Root length, wet weight, and dry weight of *S. zeylanica* as a result of *C. odorata* bioherbicide application

Treatment	Root Length (cm)	Wet Weight (g)	Dry Weight (g)
T0	7.67 a	3.34 a	1.15 a
T1	3.73 bc	0.42 c	0.20 c
T2	2.40 c	0.37 c	0.12 c
T3	1.83 c	0.30 c	0.09 c
T4	5.50 ab	2.33 b	0.91 b
T5	3.00 bc	0.97 c	0.31 c
T6	3.10 bc	0.32 c	0.11 c
T7	2.93 bc	0.42 c	0.21 c
T8	3.80 bc	0.85 c	0.23 c
T9	2.60 c	0.42 c	0.12 c
T10	2.50 c	0.27 c	0.09 c

Note: Based on Duncan Multiple Range Test (DMRT) test at 5% level. Note: T0: control; 96% ethanol extraction: T1(5%), T2(10%), T3(15%); ethyl acetate extraction: T4(5%), T5(10%), T6(15%); aquadest extraction: T7(5%), T8(10%), T9(15%); T10: 2,4-D (648 g a.i./ha)

LT_{50} and ED_{50} values of *C. odorata* bioherbicide in toxicizing *S. zeylanica*

The LT_{50} value refers to the time required for a specific treatment to achieve a 50% kill rate for target weeds. This value is of great utility in evaluating the speed of action of herbicides. For example, studies have demonstrated that different herbicides exhibit disparate LT_{50} values contingent on their mode of action and the weed species being targeted. A shorter LT_{50} value indicates a more rapid effect, which can be of particular importance in situations where rapid control is necessary to prevent weeds from competing with the crop [1]. The results of the analysis of the LT_{50} value of C. odorata leaf extract bioherbicide in controlling *S. zeylanica* (**TABLE 3**) demonstrate that the higher the concentration applied, the shorter the toxicizing time of *S. zeylanica*. The most efficacious result was observed with T10 (2,4-D chemical herbicide), with a poisoning time of 2.12 days. However, a similar performance was exhibited by T4, with a poisoning time of 2.15 days. The results demonstrate that the 96% ethanol extract bioherbicide with a 15% concentration is capable of achieving performance outcomes that are comparable to those of the chemical herbicide 2,4-D. Additionally, the T2 treatment exhibited favorable outcomes at 2.28 days, while the distilled water extraction treatment demonstrated the most optimal performance at T9, with a duration of 2.97 days. The bioherbicide extraction using ethyl acetate demonstrated a prolonged effect on weed poisoning, with a 50% longer duration than other treatments. This was evidenced by the analysis of LT_{50} values at varying concentrations of T4, T5, and T6, which exhibited LT_{50} values of 15.05, 5.46, and 3.75 days, respectively, at the lowest to highest concentrations.

TABLE 3. LT_{50} value of *C. odorata* bioherbicide in toxicizing *S. zeylanica*

Treatment	Regression	LT_{50} (days)
T1 (96% ethanol): 5%	y = 3.5238x + 2.8911	3.97
T2 (96% ethanol): 10%	y = 4.4338x + 3.4106	2.28
T3 (96% ethanol): 15%	y = 4.4915x + 3.503	2.15
T4 (ethyl acetate): 5%	y = 0.806x + 4.0509	15.05
T5 (ethyl acetate): 10%	y = 2.7106x + 3.002	5.46
T6 (ethyl acetate): 15%	y = 5.6919x + 1.7295	3.75
T7 (aquadest): 5%	y = 2.4444x + 3.4815	4.18
T8 (aquadest): 10%	y = 4.1436x + 2.8877	3.23
T9 (aquadest): 15%	y = 4.6458x + 2.8055	2.97
T10 (2,4-D): 648 g a.i./ha	y = 4.0306x + 3.6856	2.12

Note: x = log day of observation; y = probit value of toxicity percent; LT_{50} = anti log of x value

The ED_{50} value represents the herbicide dose that is required to achieve 50% control of the target weed. This value is of critical importance for determining the appropriate application rate for effective weed management while simultaneously minimizing the potential for phytotoxicity to the crop [34]. The results of the analysis of the ED_{50} value (**TABLE 4**) demonstrated that the most effective extraction of *C. odorata* leaves as a bioherbicide was achieved using a 96% ethanol solvent, with a weed poisoning dose of 50% requiring a concentration of 5.98%. The distilled water solvent also exhibited promising results at 6.35%, while the ethyl acetate solvent required a higher concentration of 8.08%.

TABLE 4. ED_{50} value of *C. odorata* bioherbicide in toxicizing *S. zeylanica*

Extraction solvent	Regression	ED_{50} (%)
Ethanol 96%	y = 5.7315x + 0.5496	5.98
Ethyl acetate	y = 5.4375x + 0.0672	8.08
Aquadest	y = 5.4926x + 0.5892	6.35

CONCLUSIONS

The extraction of *C. odorata* leaves using a variety of organic solvents has the potential to be developed as a bioherbicide, exhibiting the capacity to toxicizing *S. zeylanica* up to 100% (T2, T3, T6, T8, T9) and the ability to suppress the height, root development, and chlorophyll content of *S. zeylanica*. The optimal extraction solvent was determined to be 96% ethanol, which demonstrated superior control performance. At a concentration of 15%, it exhibited comparable control performance to the 2,4-D chemical herbicide on all observed variables.

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