

Use of elicitors weeds to increase the stability of the microenvironment and chili resistance to main pests and yield

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Abstract. Chili productivity is still relatively low ($t\ ha^{-1}$). One of the causes of chili crop failure is pest attacks, as well as extreme environmental changes. On the other hand, in the chili planting environment, a robust ecosystem of various weeds often grows that is not shaken by extreme environmental changes. These weeds contain various elicitors that act as biostimulants to help increase plant productivity and provide defense mechanisms against various stresses. The research aims to increase the resistance of chili plants to major pest attacks and yield by applying elicitor compounds in several indigenous weeds and evaluating the stability of the chili microclimate. The research used a randomized block design with elicitor treatments of *Alternanthera sessilis* leaves, *Cleome ruidosperma*, *Bidens pilosa*, *Tithonia diversifolia*, a combination of these four leaves, and control (Water). The research results show that applying elicitors for various weeds can increase red chili yields and resistance to significant pest attacks, as well as the stability of the microenvironment of chili plantings. Chili production under condition six harvesting with the application of the *Cleome ruidosperma* elicitor was 3.8 times greater than the control treatment or an increase in yield of 280%.

1 Introduction

Chili cultivation (*Capsicum annum L.*) is a profitable farming activity. This is due to the high market demand [1]. National chili production in each planting season often experiences fluctuations in Indonesia, with an average of 1,187,445 in 2016-2020. Chili plants are essential crops that influence inflation and the national economy [2]. Increasing production can be achieved with several breakthroughs implemented, including improving policy aspects, infrastructure development, improving aspects of cultivation activities until post-harvest, handling pest and disease attacks, and marketing [2].

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In the process of chili cultivation, there are main obstacles that must be faced, namely pest attacks that result in damage to quality and crop failure. One of the main pests of chili peppers is aphids (*Myzus persicae* and *Aphis gossypii*) [3]. Aphids generally attack from the lower leaves, leaves of plant shoots, and flower buds to young stems [4]. Aphid infestation results in abnormal and curly leaves; the affected leaves become brittle and yellowish and can indirectly become a vector for viral diseases [5].

Synthetic pesticides are still widely used to control aphid pests. The use of synthetic pesticides causes many severe problems, including decreased plant resistance and microclimate shock, increasing populations of secondary pests, death of natural enemies, and pesticide residues that are harmful to human health and the environment [6].

Effective and environmentally friendly pest control is essential to be implemented. The use of weeds is one of the solutions to eradicate pests. Weeds are a source of elicitor molecules, which are extrinsic molecules related to resistance to pest and disease attacks. Elicitor molecules can attach to particular receptor proteins located on plant cell membranes [7]. These receptors are able to recognize the molecular pattern of elicitors and trigger intracellular defense signals via the octadecanoid pathway. This response results in increased metabolite synthesis that reduces damage and increases resistance to pests, diseases, or environmental stress. [8] This immune response is called pattern trigger immunity [8]. Elicitors can protect plants from attacks and damage caused by pests and diseases. On the other hand, elicitors do not have to be directly toxic to plant-disrupting organisms in order to benefit.

Weeds are available in abundance in nature and around cultivated plantations, including sessile joy weed (*Alternanthera sessilis*), purple cleome (*Cleome rutidospermae*), Cobbler's pegs (*Bidens pilosa*), and Mexican Sunflower (*Tithonia diversifolia*) [8]. More than 2400 types of weed plants belonging to 235 families contain pesticides [9]. The use of weeds as a source of elicitors for resilience generation still needs to be researched. Therefore, elicitor-based pesticides are an alternative to conventional chemical pesticides that are often harmful to farmers, the environment, and consumers. Consumers like healthy and safe products [10]. The research aims to increase the resistance of chili plants to major pest attacks as well as yield through the application of elicitor compounds in several indigenous weeds and evaluating the stability of the chili microclimate.

2 Research methods

The research was carried out on the Agrotechnology land of the University of Muhammadiyah Malang, Jl. Karyawiguna No. 234, Tunggulwulung, Lowokwaru district, Malang City, East Java 65143, in July - November 2023.

The tools used were tractors, hoes, shovels, padlocks, digging sticks, measuring cups, hand sprayers, mulch, rulers, sample labels, bottles, strainers, green nets, plastics, and cameras. The materials needed include large chili of the Columbus variety, compost, urea, TSP, ZA, KCl, water, *methyl eugenol*, cotton, *A. sessilis* leaves, *C. rutidospermae* leaves, *B. pilosa* leaves, and *T. diversifolia* leaves as a source of elicitor materials.

The study used a Randomized block design (RBD) with seven treatments including control (without elicitor, spraying by water (P0), *A. sessilis* leaf elicitor (P1), *C. rutidospermae* leaf elicitor (P2), *B. pilosa* leaf elicitor (P3), *T. diversifolia* leaf elicitor (P4), a combination of four-leaf elicitors (P5), and chemical pesticides with profenofos active compound as positive control (P6). Each treatment was repeated four times so that there were 28 units of experimental units. Each unit (1.2 m²) of the experiment uses eight plants.

The research procedure was carried out in several stages, including the manufacture of elicitors and weed extracts, which were carried out by extracting several types of leaves. The application is carried out in the afternoon by thoroughly spraying plants that were infested

with aphid pests (starting from the bottom to the top evenly). Elicitors and weed extracts were applied when they were 21 – 70 d after planting (DAP), with an interval of 7 d at a dose of 50 mL plant⁻¹.

The investment in aphid pests was carried out by preparing 16 large chili plants that were used as target pest traps. If the target has multiplied, then imago initiation is given when the plant is 14 old. Imago is left for 7 d so that it can adapt, breed, and lay eggs on the leaves of chili plants. Plant cultivation activities include land preparation, seed preparation, transplanting, installation of stilts, plant care, and harvesting.

Observations include analysis of active compounds of elicitor plants, plant height (Cm), number of leaves, leaf area per plant (Cm²), aphid pest population (individual), percentage of infested leaves (%), fruit number, fruit weight (g), to fruit weight per plot (1.2 m²)(g). The estimated yield in t ha⁻¹ is calculated using the formula: Fruit weight in kg/m² multiplied by 10. Data analysis was carried out using the F test to determine the effect of the treatment given. Test of *Duncan's Multiple Range Test* (DMRT) with a level of $\alpha = 0.05$ to determine the difference in each treatment.

3 Results And Discussion

3.1 Number and area of leaves of red chili plant

The results of the analysis showed that the application of several weed elixirs had a natural effect on the number and area of leaves of red chili plants at the age of 40 days after planting. The average is presented in Figure 1.

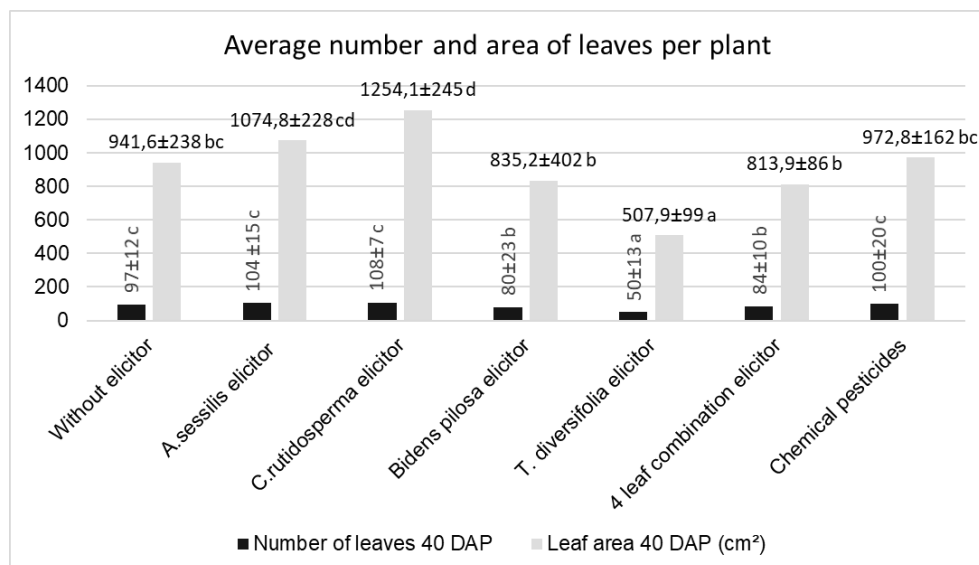


Fig. 1. Average number and area of leaves per plant 40 days after planting (DMRT $\alpha = 0.05$).

The number of chili leaves with *A. sessilis* and *C. rutidosperma* leaf elicitor treatment is as good as chemical pesticide treatment, but the application of *C. rutidosperma* elicitor gives a better leaf area response per plant, which is 1254.1 cm². The results of the gas chromatography spectrometry (GCMS) test of *C. rutidosperma* leaf extract contain many compounds and phytochemical molecules. Among these phytochemical compounds, there were those that function as biostimulants (elicitors) that were indirectly able to increase the

growth of chili plants [13]. The elicitor treatment of *T. diversifolia* leaves on chili plants gave the lowest average response of the number and area of leaves compared to other treatments, namely 50 leaves with an area of 507.9 cm². This is suspected to be the result of the concentration of *T. diversifolia* leaf extract, which makes the plant abnormal. Because some plant extract content can be allelochemical compounds at specific concentrations. Allelochemical compounds at certain concentrations have the potential to be plant-based herbicides. These compounds include flavonoids, tannins, phenolic acids, ferulic acid, coumarins, terpenoids, cyanohydrins, quinones, cyanuric acids, steroids, and their derivatives. *T. diversifolia* leaf extract contains all of these allelochemical compounds [11].

3.2 Aphid pest population

The results of the analysis showed that the treatment of various weed elicitors had a natural effect on aphid populations at various observation ages. The average is presented in Table 1.

Table 1. The average population of aphid pests at various observation ages.

| Treatment | Mean aphid population at observation age | | | Pest types number |
|---------------------------------|--|-------------|--------------|-------------------|
| | 42 DAP | 56 DAP | 70 DAP | |
| Without elicitor | 43,0±15 d | 50,2±7,6 d | 166,7±16,4 c | 8 |
| <i>A.sessilis</i> elicitor | 33,0±10,6 c | 18,0±9,3 bc | 6,5±4,5 b | 2 |
| <i>C.rutidosperma</i> elicitor | 30,0±13,3 bc | 14,0±5,5 b | 3,5±2,9 ab | 2 |
| <i>Bidens pilosa</i> elicitor | 27,5±8 bc | 16,2±8,7 b | 7,5±5 b | 2 |
| <i>T. diversifolia</i> elicitor | 25,0±16,7 b | 0,5±1 a | 0,0±0 a | 0 |
| Four-leaf combination elicitor | 26,5±5,2 bc | 16,7±7,7 b | 8,5±6,6 b | 2 |
| Chemical pesticides | 14,7±1,7 a | 24,7±2,6 c | 10,75±6,2 b | 5 |

Note: Numbers followed by the same letter in the same column are not significantly different according to the 5% DMRT test. DAP = days after planting.

The application of the *T.diversifolia* leaf elicitor resulted in the death of the entire aphid pest population, and the plants were free from pest attacks. Likewise, treatment with foliar elicitors *C. rutidosperma*, *A. sessilis*, *Biden pilosa*, and a combination of 4 leaves, resulting in a low aphid population. Without elicitor application, the aphid population continues to increase. Each plant generally contains phytochemical compounds that are specific in both type and function. Often, the compound is a secondary metabolite that is produced with a specific physiological function and at a specific time during plant growth. Among these phytochemical compounds, there are those that function as biostimulants (elicitors), which are indirectly able to increase the growth of chili plants and plant resistance to pest and disease attacks [12]. Some other compounds act as biopesticides (bioinsecticides and biofungicides) that can kill or limit the development of certain pests and diseases. Leaf extract *C.rutidosperma* It contains three metabolite compounds that are organic pesticides, namely propanoic acid, phenol, and isopropyl myristate [12]. The qualitative phytochemical of *C.rutidosperma* screening-detected alkaloids, flavonoids, phenols, sugars, proteins, saponins, sterols, tannins, and terpenoids [13].

3.3 Percentage of Affected Leaves

The treatment of several weed elicitor treatments had a significant effect on the percentage of infested leaves. The average is shown in Table 2. The lowest average percentage of leaves infested by aphid pests was produced by the treatment of *T. diversifolia* leaf elicitor. The leaf extract not only affects the pest population but also affects the growth and development of

the chili plant as a whole. This can be seen from the beginning of chili growth, where the plant becomes shorter with the lowest number and leaf area compared to other treatments. The concentration and frequency of giving elixirs once every seven days are still often table, so the effect on chili peppers is like poisoning. The plant does not grow optimally, but it is deadly to the pests that attack it. Overall, the application of the elicitor is able to reduce the percentage of leaves infested by pests. Chili fruit production is influenced by the growth of chili plants, especially the number and area of leaves [14].

Table 2. The average percentage of chili leaves infested with aphids at several observation ages.

| Treatment | 42 DAP | 56 DAP | 70 DAP |
|---------------------------------|------------|------------|------------|
| Without elicitor | 13,6±5,5 a | 38,9±2,6 c | 79,4±3,2 d |
| <i>A.sessilis</i> elicitor | 11,9±3,1 a | 4,4±3,3 b | 1,8±1,1 bc |
| <i>C.rutidosperma</i> elicitor | 10,6±5,9 a | 5,0±2,8 b | 1,7±0,9 bc |
| <i>Bidens pilosa</i> elicitor | 12,8±5,3 a | 4,5±2,2 b | 0,9±0,8 ab |
| <i>T. diversifolia</i> elicitor | 14,9±9,3 a | 1,8±2,7 a | 0,0±0 a |
| Four-leaf combination elicitor | 11,4±1 a | 6,3±3,7 b | 2,2±0,8 c |
| Chemical pesticides | 5,4±2,1 a | 2,0±1,7 a | 0,9±0,4 ab |

Note: Numbers followed by the same letter in the same column are not significantly different according to the 5% DMRT test. DAP = days after planting.

3.4 Fruits number, Weight of Fruits per Plant, and per plot

The treatment of elicitor types had a significant effect on the fruit number, fruit weight per plant, and per plot. The average of each treatment can be seen in Table 3.

Table 3. Average fruit number, fruit weight per plant, and per plot.

| Treatment | Fruits number/plant | Fruit weight/plant (g) | Fruit weight/plot (1,2 m ²) (g) | Estimated chili production in ha (t) |
|---------------------------------|---------------------|------------------------|---|--------------------------------------|
| Without elicitor | 9,0±3,6 a | 84,2±39,9 ab | 487,7±52,2ab | 4,1 a |
| <i>A.sessilis</i> elicitor | 29,0±8,2 b | 313,1±48,4 b | 1636,0±44,1 bc | 13,6 bc |
| <i>C.rutidosperma</i> elicitor | 30,7±3,8 b | 334,8±46,4 b | 1860,0±57,3 c | 15,5 c |
| <i>Bidens pilosa</i> elicitor | 26,2±3,3 b | 299,1±31,4 b | 1396,8±66,4 b | 11,6 b |
| <i>T. diversifolia</i> elicitor | 8,5±4,1 a | 65,7±36,6 a | 319,6±91,5 a | 2,7 a |
| Four-leaf combination elicitor | 26,5±10,5 b | 284,3±42,7 b | 1472,4±94,9 bc | 12,3 bc |
| Chemical pesticides | 27,2±13,2 b | 316,1±46,2 b | 1460,33±36,5 b | 12,2 b |

Note: Numbers followed by the same letter in the same column are not significantly different according to the 5% DMRT test. Estimated yield in hectares is calculated using the formula: Fruit weight/plot in kg / m² x 10.

The treatment of *C. rutidospermae* elicitor produced a higher fruit weight per plot than other treatments. Also, the application of elicitors from the leaves of *Alternanthera sessilis*, *Bidens pilosa*, and *Tithonia diversifolia* and their combinations provide a higher fruit number and fruit weights than the controls, as well as chemical pesticide treatments. The estimated production of chili fruits reaches the range of 12.3 - 15.5 t ha⁻¹. This production is 3 – 3.8 times higher than without elicitors, even higher than chemical pesticide treatments. The average production with the application of the elicitor is also higher than the national chili

production, which is in the range of 8-9 t ha⁻¹. The application of 3 plant extracts, namely *Alternanthera sessilis*, *Cleome rutidosperma*, and *Bidens pilosa*, is feasible to be applied in an environmentally friendly chili plant cultivation system because it can reduce input costs with high yields, and materials are available in the field in abundance. The results of the analysis of the environment around chili plants showed that with the application of the elicitor to the four weed plants, the attack rate and pest types decreased. Even the elicitor *T. diversifolia* causes pests to die entirely or do not develop/colonize chili leaves. The pest type number that appears in treatment without elicitor/control is the most common type. Pests that appear include *Bemisia tabaci*, Thrips, and Aphids; there are 3 types: grasshoppers, leaf-eating caterpillars, and fruit flies. The application of *Alternanthera sessilis* extracts, *Cleome rutidosperma*, and *Bidens pilosa*, on average, only found two types of pests, namely aphids and fruit flies. The number of fewer types of pests shows that the microenvironment between chili plants is more stable/more supportive for plant growth and development so that plants are able to produce better.

Treatment *T. diversifolia* showed the lowest fruit number and fruit weights, even lower than the control treatment (without elicitors). The concentration of *T. diversifolia* leaves is suspected to be the cause of the low number of fruits and yields of chili plants. The concentration of concentrated weed extract solutions with various elicitor compounds produces high allelochemical compounds and poisons plants. As a result of these compounds, plants become abnormal and cannot grow optimally. Some research results show that when plant growth is abnormal, the formation of flowers and fruits produced will also be inhibited. The fruits produced are smaller than other treatments [15]. Leaf extract *T. diversifolia* contains phytochemical compounds alkaloids, flavonoids, tannins, saponins, phenols, sesquiterpenes, monoterpenes, and diterpenes. These active components function as a potent anti-microbial. In the right concentration, it can function as an antioxidant/biostimulant [16 - 18].

4 Conclusion

Application of elicitors from weed plant leaves *Alternanthera sessilis*, *Cleome rutidosperma*, and *Bidens pilosa* able to increase chili yield 3 – 3.8 times higher than without the application of elicitor; able to increase chili resistance to pest attacks, as well as improve the stability of the microenvironment among chili plantations.

References

1. M. T. Sundari, D. Darsono, J. Sutrisno, E. Antriyandarti, Analysis of chili demand in Indonesia, AIP Conf. Proc, The 5th ICALS 2021, Jember, November 3-4, **2583**(1), 110009 (2021) <https://doi.org/10.1063/5.0116855>
2. Ministry of Agriculture of the Republic of Indonesia, Strategic Plan of the Ministry of Agriculture 2020 – 2024. (Ministry of Agriculture, Jakarta, 2020) [https://ppid.pertanian.go.id/doc/1/Draft%20Renstra%202020-2024%20edited%20BAPPENAS%20\(Final\).pdf](https://ppid.pertanian.go.id/doc/1/Draft%20Renstra%202020-2024%20edited%20BAPPENAS%20(Final).pdf)
3. J.J. Gallo-Franco, D.N. Duque-Gamboa, N. Toro-Perea, Bacterial communities of Aphis gossypii and Myzus persicae (Hemiptera: Aphididae) from pepper crops (Capsicum sp.). Sci Rep. **9**, 5766 (2019) <https://doi.org/10.1038/s41598-019-42232-8>

4. K. Anggraini, K.A. Yuliadhi, D. Widianingsih, Effect of Aphid Population on Large Chili Plants (*Capsicum annum* L.) on Crop Yield. *J. Trop. Agro.* **7**(1), 113 – 121 (2018)
<https://ojs.unud.ac.id/index.php/JAT/article/view/38266>
5. M. Nindatu, D. Moniharapon, S. Latuputty, Effectiveness of Red Chili Extract (*Capsicum annum* L.) Against Aphid Mortality (*Aphis gossypii*) in Chili Plants. *Agrologia.* **5**(1), 10-14 (2016).
<http://dx.doi.org/10.30598/a.v5i1.192>
6. M. S. D. Permadi, H. Fitrihidajati, Effect of Giving Brotowali Stem Extract (*Tinospora crispa*) on Aphid Mortality (*Aphis gossypii*). *Lantern Bio.* **8**(2), 101-106 (2019).
<https://ejournal.unesa.ac.id/index.php/lenterabio/article/view/28621>
7. M.N.A. Abdul, I.S. Kumar, K. Nadarajah, Elicitor, and receptor molecules: orchestrators of plant defense and immunity. *International Journal of Molecular Sciences.* **21**(3):963 (2020). [doi: 10.3390/ijms21030963](https://doi.org/10.3390/ijms21030963)
8. Y. Bektas, T. Eulgem, Synthetic plant defense elicitors, *Front. Plant Sci.* **5**, 804 (2015)
<https://doi.org/10.3389/fpls.2014.00804>
9. H. Haerul, M.I. Idrus, R. Risnawati, The Effectiveness of Plant-Based Pesticides in Controlling Pests in Chili Plants. *J. Agro.* **1**(2), 129 – 136 (2016)
<https://www.neliti.com/id/publications/271888/efektifitas-pestisida-nabati-dalam-mengendalikan-hama-pada-tanaman-cabai>
10. M. Meena, G. Yadav, P. Sonigra, A. Nagda, T. Mehta, P. Swapnil, H. Harish, A. Marwal, Role of elicitors to initiate the induction of systemic resistance in plants to biotic stress. *Plant Stress.* **5**, 100103 (2022). <https://doi.org/10.1016/j.stress.2022.100103>
11. A. Kumar, M. Kumar, A. Jose, V. Tomer, E. Oz, C. Proestos, M. Zeng, T. Elobeid, K. Sneha, F. Oz, Major Phytochemicals: Recent Advances in Health Benefits and Extraction Method. *Molecules.* **16**; **28**(2):887 (2023). [doi: 10.3390/molecules28020887](https://doi.org/10.3390/molecules28020887)
12. A. Jamiólkowska, Natural Compounds as Elicitors of Plant Resistance Against Diseases and New Biocontrol Strategies. *Agronomy.* **10**, 173 (2020).
<https://doi.org/10.3390/agronomy10020173>
13. A. Ikhwan, D. Indratmi, F. Hasanah, M.F.M. Atoum, I. Iqrar, Analysis of Metabolites from Purple Cleome Extract (*Cleome rutidosperma* Linn.) as Potential Organic Fungicides. *J. Sarhad Agric.* **37**(1), 115-121.
<https://dx.doi.org/10.17582/journal.sja/2021/37.s1.115.121>
14. D.P. Soedjarwo, W.A. Tjokrosumarto, Growth and production plant chili pepper (*Capsicum annum*) as a result of the existence pruning leaves. In *Proceedings Jul 13 Vol. 1*, No. 1, pp. 439-449 (2018).
<https://ojs.pnb.ac.id/index.php/Proceedings/article/view/937>
15. M.S. Roopa, S. Ramnath, T. Rhetso, V. Sivaram, Comparative analysis of Phytochemical Constituents, Free Radical Scavenging Activity and GC-MS Analysis of Leaf and Flower Extract of *Tithonia diversifolia* (Hemsl.) A. Gray. *P. Bio. Molec.* **22**(37-38), 211–228 (2021).
<https://ikpress.org/index.php/PCBMB/article/view/6543>
16. R. Arumugam, B. Elanchezhian, J. Samidurai, K. Amirthaganesan, Comparative antioxidant, antibacterial and phytochemical analysis of roots, stems, leaves and seeds from *Cleome rutidosperma* DC. *Nat. Rsc. Human Health*, **2**(4), 479-484 (2022).
<https://doi.org/10.53365/nrfhh/146009>
17. M. Misrahanum, Z. Safarah, Y.S. Ismail, Antibacterial activity of Mexican sunflower leaf *Tithonia diversifolia* (Hemsl.) A. Gray Aqueous extract against methicillin-resistant

-
- Staphylococcus aureus*. *Pharmaciana*. **12**, 1 (2022).
<http://journal.uad.ac.id/index.php/PHARMACIANA/article/view/20469>
18. T. S. Agidigbi, O. Odeyemi, Antibacterial Activities of Crude Extracts of *Tithonia Diversifolia* Against Common Environmental Pathogenic Bacteria. *The Experiment*, **20**(4), 1421–1426 (2014). <http://dx.doi.org/10.13140/RG.2.2.13506.68809>