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Allelopathic effect of *Lantana camara* aqueous leaf extract on the seedling germination of *Zea mays* (maize)

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Abstract. *Mistica DJP, Magsino NJS, Tanjusay ZC, Paclibar GCB. 2023. Allelopathic effect of* Lantana camara *aqueous leaf extract on the seedling germination of* Zea mays *(maize). Asian J Agric 7: 83-87. Lantana camara* L. is an invasive weed rampant in the Philippines and threatens agricultural crops. This study was conducted to evaluate the growth inhibitory effects of *L. camara* aqueous leaf extract in seedling germination of *Zea mays* L. Different concentration levels (25%, 50%, 75%, and 100%) of the *L. camara* leaf extracts as compared to the control were administered to the hypochlorite-treated seeds for two weeks. Results showed a significant difference among the treatments in the number of germinated seeds and main shoot and primary root elongation using ANOVA. Furthermore, the t-test revealed that the presence and absence of the aqueous leaf extract in seed germination are significantly different, showing significant inhibitory effects in the number of germinated seeds and the length of the shoot and root when the aqueous leaf extract was present. Moreover, in vitro assay also indicated that the concentration of the aqueous leaf extract is directly proportional to the growth inhibitory effects on seed germination and root and shoot elongation. Therefore, it is recommended to test the allelopathic effect of *L. camara* aqueous leaf extract in other agricultural crops and its effect on the soil and crop nutrition.

Keywords: Growth inhibitory effect, invasive weed species, *Lantana* bioassay, maize, root and shoot elongation

INTRODUCTION

One of the major threats to biodiversity is the invasive alien species. *Lantana camara* L. is one of the most widespread invasive weed species (Taylor et al. 2012; Enyew and Raja 2015) which is rampant in the Philippines and were already established in disturbed areas (Paclibar and Tadiosa 2019; Paclibar and Tadiosa 2020) and many agricultural areas. The *L. camara* is considered both a pest abs and an ornamental plant (Mishra 2014; Tadele 2014), while some consider it to help in bioremediation and for medication due to its medical properties (Arbiastutie et al. 2017; Ved et al. 2018). For example, lantana could be used to treat gastric and duodenal ulcers in rats (Sathish et al. 2011), as an agent for larvicidal activity (Chavan and Nikam 1982), as an antibacterial agent against Gram-negative and Gram-positive bacteria (Barreto et al. 2010).

Studies about the allelopathic effect of *L. camara* were recorded in agricultural crops (Sahid and Sugau 1993; Ahmed et al. 2007; Choyal and Sharma 2011; Gantayet et al. 2014; Kar et al. 2014; Mishra 2014; Tadele 2014; Enyew and Raja 2015; Rusdy and Ako 2017; Anwar et al. 2018; Mawal and Patil 2019; Gautam et al. 2021), bryophytes (Mishra 2014), forest crops (Hossain and Alam 2010) and communities (Gentle and Duggin 1997) and even to other weeds (Saxena 2000; Mishra 2014). In line with this, stimulatory effects of *L. camara* were also recorded in some agricultural crops (Tadele 2014; Enyew and Raja 2015; Ming et al. 2020).

Zea mays L. or corn, is one of the Philippines' most important food sources and agricultural crops. *Zea mays* is recognized as the second principal agricultural crop next to rice and play an important role as one of the major sources of livelihood for Filipino farmers (Gerpacio et al. 2004). Several studies have been conducted showing the significant inhibition of seed germination and seedling growth of *Z. mays* due to allelopathic effects of plants such as *Castanea henryi* (Skan) Rehder & E.H.Wilson (Ming et al. 2020), *Sonchus arvensis* L. (Bashir et al. 2018), *Ambrosia artemisiifolia* L. (Bonea et al. 2017), *Gliricidia sepium* (Jacq.) Kunth and *Acacia auriculiformis* A.Cunn. ex Benth. (Oyun 2006) among others. Furthermore, the allelopathic effect of *L. camara* in maize was studied by Tadele (2014), Enyew and Raja (2015), Mawal and Patil (2019), and Gautam et al. (2021). However, few accounts were available for full concentration, and few allelopathic effects study was conducted in the Philippines.

Weed infestations are recognized as a serious biological constraint in many different crop plantation types, either lowland or highland. The manual weeding is reduced because of high labor costs has been accompanied worldwide by the intensive use of synthetic herbicides (Tadele 2014). Since weed infestations are recognized as a serious biological constraint in crop plantations, further studies about the allelopathic effect on agricultural crops must be conducted. Therefore, this study was conducted to evaluate the allelopathic effect of *L. camara* in the seedling germination and root and shoot elongation of *Z. mays*; to determine the significant differences of the effect of aqueous leaf extracts concentrations of *L. camara* to the seedling germination, and root and shoot elongation of *Z. mays*; identify the concentration with the most inhibitory activity; and determine the effect of the presence and absence of aqueous leaf extracts to the *Z. mays*. Farmers, governing bodies, and agriculture and weed management organizations can use the findings as a baseline.

MATERIALS AND METHODS

Extract preparation

Mature leaves were collected in Biluso, Silang, Cavite (14.2369° N, 121.0378° E), which had already proliferated in the wide farmland. After the authentication from a qualified botanist, the collected leaves were air-dried for 15 minutes to let the ethanol be off after disinfection. The leaves were then processed using the preparation performed by Tadele (2014) through immersion of 100 grams of leaves of *L. camara* into 1,000 mL (1 L) containers filled with 500 mL (0.5 L) distilled water, which were then sealed in an Erlenmeyer flask, and later shaken and stored at a temperature of 21°C for at least 24 hours. After storing, the aqueous extracts were filtered using filter paper and diluted into 25, 50, 75, and 100% concentrations. The extracts were stored in Erlenmeyer flasks sealed with parafilm and kept in the fridge (5°C).

Germination and growth records

The seeds of maize were selected through fractionation and their viability. Afterward, the maize seeds were washed with distilled water and surface sterilized with sodium hypochlorite aqueous solution (Muzzo et al. 2018) for at least 5 to 10 minutes to prevent contamination and were rinsed thoroughly with distilled water multiple times; while the controls were treated only with distilled water. Next, 15 seeds of maize were evenly placed in 9 cm-diameter petri dishes lined by Whatman No. 1 filter paper (Akpan et al. 2017) with three replicates per treatment. The seeds were then treated with 5 ml of 25%, 50%, 75%, and 100% aqueous extract concentrations and 5 ml of distilled water for the control, which were applied daily with one dosage applied from 8:00 to 9:00 AM and a second dosage at around 3:00 to 4:00 PM to keep the moisture within the filter paper; this allowed seedling development for 14 days (2 weeks) period. Finally, the setup was placed inside the laboratory, wherein the same amount of natural light was treated on all the treatments. The prerequisite for considering a germinated seed is the appearance of the radicle. Therefore, germinated seeds were counted and checked daily while the root and shoot lengths were measured after the experiment (Tadele 2014).

Data analysis

The Germination Capacity (GC) and the root and shoot Relative Elongation Ratio (RER) were calculated using the formula (Bewley and Black 1994; Saxena et al. 1996):

$$\text{GC (\%)} = \frac{(No.\ of\ germinated\ seeds)}{(Total\ No.of\ seeds\ sown)}\ x\ 100$$

$$\text{RER of shoot} = \frac{(Mean\ shoot\ length\ of\ tested\ plant)}{(Mean\ shoot\ length\ of\ control)}\ x\ 100$$

$$\text{RER of root} = \frac{(Mean\ root\ length\ of\ tested\ plant)}{(Mean\ root\ length\ of\ control)}\ x\ 100$$

The significant differences among the treatments were evaluated using Analysis of Variance (ANOVA I) and further assessed using the post hoc Tukey's HSD (Honestly Significant Difference) test. The significant difference between the presence and absence of the aqueous leaf extracts of lantana was calculated using a t-test.

RESULTS AND DISCUSSION

Seed germination of *Zea mays*

The Germination Capacity (GC) of the *Z. mays* seeds was found to be highest at the control (56.67%), followed by 25% (53.33%), 100% (36.67%), and lowest at 50% and 75% concentrations with both GC of 26.67% (Figure 1). The number of seeds grown per treatment was also significantly different ($P<0.05$), wherein the control has the highest number of seeds grown ($\bar{x}$ = 5.67) and lowest at 50% ($\bar{x}$ = 2.67) and 75% ($\bar{x}$ = 2.67) concentrations (Table 1). It is noteworthy that a 25% concentration of lantana leaf extract has no significant difference from the control, then 50%, 75%, and 100% concentrations had no significant differences indicating that high concentrations of lantana aqueous leaf extracts negatively affect the seed germination, and GC of *Z. mays*. The results of the study were aligned with the study of Sahid and Sugau (1993), Ahmed et al. (2007), Hossain and Alam (2010), Kar et al. (2014), Mishra (2014), Rusdy and Ako (2017), Anwar et al. (2018), and Mawal and Patil (2019) stating that *L. camara* leaf extracts had inhibitory effects in the germination of several agricultural crops, forest crops, and other weeds. In line with this, a 75% concentration of *L. camara* leaf extracts had the most inhibitory effect on the germination of *Z. mays*.

Moreover, the highest concentration of the extract did not greatly inhibit the GC capacity. That aligns with the study of Sahid and Sugau (1993), wherein the full strength of *L. camara* extract did not significantly inhibit the germination of spinach and cucumber seeds. Also, Maharjan et al. (2007) revealed that the highest concentration of *Parthenium hysterophorus* L. had an inhibitory effect on the tested agricultural seeds except on *Z. mays*. Dzafic et al. (2013) discussed that maize has autotoxic effects and can decrease root biomass. Even though the active compounds of both the study plants are not within the scope of the study, it could be considered that the water-soluble compounds interacted with the leaf extract; thus, instead of inhibiting the growth of maize, it has a higher GC than at a lower concentration. As Dorning and Cipollini (2006) explained, allelopathy is most effective in inhibiting plant growth when the plant has resistance to autotoxic effects. It could be observed as well that the GC of all the treatments is low, which can be hypothesized that this is due to the thermal stress, which has been indicated by the study of Farooq et al. (2008), which revealed the optimum temperature for maize germination is between 25-28°C. In addition, Farooq et al. (2008) also added that poor germination could occur under optimal temperatures.

Root and shoot elongation of *Z. mays*

The root and shoot elongation of *Z. mays* were significantly different among the treatments (P<0.05) (Table 2). The longest length of the root was observed in the control ($\bar{x}$ = 28.0), while the shortest length of the root was observed in 75% concentration ($\bar{x}$ = 12.11). The root's Relative Elongation Ratio (RER) reveals that *Z. mays* roots exposed to different aqueous concentrations of *L. camara* had relatively lower RER than the control. Furthermore, the 75% concentration was the lowest (43.25%), followed by 100% (47.82%) and 50% (59.35%), while the 25% concentration was the highest (81.75%) (Figure 2). Based on the findings, the control was significantly different among the treatments (P<0.05) except in 25% concentration in terms of root elongation, wherein the higher the concentration, the inhibitory effect of *L. camara* aqueous leaf extracts was also higher, making the root shorter in length.

The longest length of the shoot was observed in the control ($\bar{x}$ = 37.45), and the shortest was observed in the 75% concentration ($\bar{x}$ = 14.83) (Table 2). The result shows that the shoot of *Z. mays* that were exposed to several concentrations had lower RER compared to the control (Figure 3), in which the 50% (65.13%) had the highest RER followed by 25% (60.16%), 100% (47.69%) and 75% (36.60%) had the lowest RER. That indicates *L. camara* concentrations inhibit the elongation of shoots of *Z. mays*. Based on the results, the control had significant differences among the treatments used, indicating that aqueous leaf extracts of *L. camara* can inhibit shoot elongation.

The *L. camara* leaf extracts significantly negatively affected the elongation of the root and shoot of *Z. mays*, wherein the higher concentrations had higher inhibitory effects. The proportionality of the concentration and the inhibitory effects were in line with the study of Ahmed et al. (2007), Hossain and Alam (2010), and Kar et al. (2014). It is also worth noting that the control differed from the other treatments except for 25% concentration in root elongation. Also, 75% concentration had the lowest length for both root and shoot, with RER of 43.25% and 39.6%, respectively. The inhibitory effects of *L. camara* leaf extracts on the root and shoot of other agricultural and forest crops were observed in several studies (Ahmed et al. 2007; Hossain and Alam 2010; Kar et al. 2014; Enyew and Raja 2015; Rusdy and Ako 2017; Anwar et al. 2018; Mawal and Patil 2019).

Presence of *L. camara* leaf extract

The *Z. mays* germination and root and shoot elongation were significantly different between the presence and absence of *L. camara* leaf extracts indicating that *L. camara* leaf extracts had inhibitory effects on the growth of *Z. mays*, as revealed in Table 3. Furthermore, the absence of *L. camara* leaf extracts had a higher mean ($\bar{x}$) as compared to the presence of *L. camara* leaf extracts. This result coincides with the study of Gentle and Duggin (1997), wherein the removal of *L. camara* significantly increased the germination of *Alectryon subcinereus* (A.Gray) Radlk. and *Cryptocarya rigida* Meisn. in three Australian forest communities. Therefore, adding leaf extract of *L. camara* was found to have a maximum inhibitory effect compared to the stem and root extracts (Choyal and Sharma 2011).

Table 1. Germination of *Z. mays* seeds in response to *L. camara* aqueous leaf extracts at different concentrations after 2 weeks.

Lantana camara leaf extract concentrations (%)	Mean of germinated seeds of *Zea mays*
0	5.67^{a}
25	5.33^{a}
50	2.67^{b}
75	2.67^{bc}
100	3.67^{bcd}

Note: Different letters imply significant differences among the treatments (P<0.05)

Table 2. Root and shoot elongation of *Z. mays* (mean) in response to *L. camara* aqueous leaf extracts in different concentrations after 2 weeks.

Lantana camara leaf extract concentrations (%)	Root elongation of *Z. mays* ($\bar{x}$)	Shoot elongation of *Z. mays* ($\bar{x}$)
0	28.00^{a}	37.45^{a}
25	22.89^{ab}	22.53^{b}
50	16.62^{bcd}	24.39^{bc}
75	12.11^{c}	14.83^{bcd}
100	13.39^{cd}	17.86^{bcde}

Note: Different letters imply significant differences among the treatments (P<0.05) per variable

Table 3. The presence and absence of *L. camara* leaf extracts in response to the growth of *Z. mays* after two weeks

Zea mays growth	Presence of *L. camara* aqueous leaf extract ($\bar{x}$)	Absence of *L. camara* aqueous leaf extract ($\bar{x}$)
Germination	3.58^{X}	5.67^{Y}
Root elongation	16.25^{X}	28.00^{Y}
Shoot elongation	19.90^{X}	37.45^{Y}

Note: Different letters signify significant differences between rows

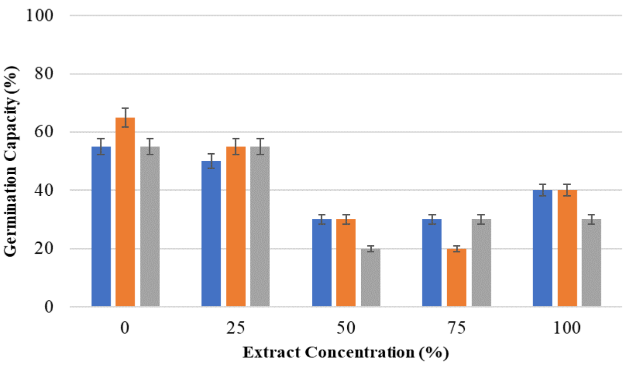

Figure 1. Germination Capacity (GC) of *Z. mays* in response to *L. camara* aqueous leaf extracts at different concentrations. Bars indicate GC with Standard Error (SE)

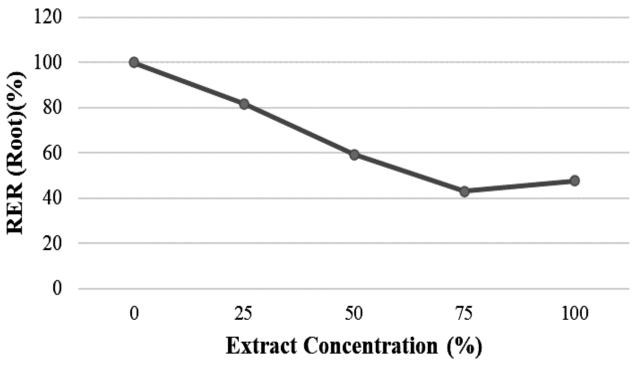

Figure 2. Relative elongation ratio of *Z. mays* roots in response to different extract concentrations of *L. camara* after 2 weeks

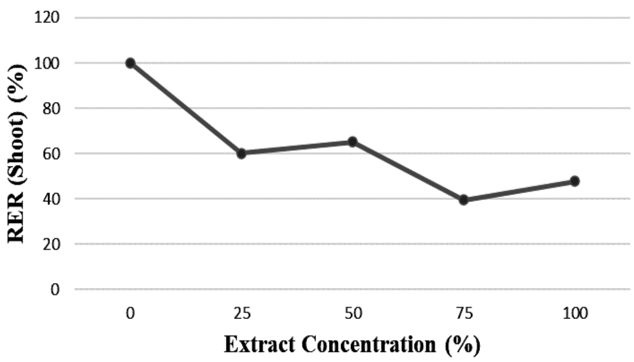

Figure 3. Relative elongation ratio of *Z. mays* shoots in response to different extract concentrations of *L. camara* after 2 weeks

In conclusion, the study concluded that the different concentrations of *L. camara* aqueous leaf extracts had inhibitory effects on the germination and root and shoot elongation of *Z. mays*, wherein the higher the concentration is, the higher the inhibitory effect. Since the thermal stress and autotoxicity affected the lantana's germination capacity, their association with the different extract concentrations should be further studied. Furthermore, though bioassay is deemed important in assessing the allelopathic potential of *L. camara*, however, field investigations should be done to relate and compare the in vitro study to the field results. Also, the effects of *L. camara* aqueous leaf extract on other agricultural crops in the Philippines should be investigated.

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