Bioactivity and Sublethal Effects of *Ageratina adenophora* (Asteraceae) on *Bactrocera dorsalis* (Diptera: Tephritidae)[1]

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Abstract Local fruit orchards have incurred heavy losses due to invasive insects, including the oriental fruit fly, *Bactrocera dorsalis* (Hendel) (Diptera: Tephritidae), in Guizhou Province, China. In our efforts to manage this invasive pest, a laboratory study was conducted to evaluate the response of adult *B. dorsalis* to an extract of *Ageratina adenophora* (Spreng.) R.M. King & H. Rob (Gunneridae: Asteraceae). We evaluated the contact toxicity and oviposition-deterrent activity of the extract on adult *B. dorsalis*, as well as the sublethal effects on survival and reproduction. We found that the *A. adenophora* extract has significant insecticidal activity, with the 24-h median lethal concentration (LC_{50}) for adults being 26.014 mg/ml. The extract of *A. adenophora* also showed oviposition inhibition. A residual deterrent effect (28.16%) was evident up to 5 d after treatment. Exposure to the LC_{25} concentration of the *A. adenophora* extract caused significant differences in preoviposition and fecundity, decreased adult longevity in the F_0 generation, and decreased egg and pupa survival in the F_1 generation. The levels of carboxylesterase enzyme activity in adults treated with the extract were significantly lower than those in the untreated controls at 12 h and 24 h. This latter phenomenon could perhaps be an adaptive response to the extract that might reduce its toxic effects in *B. dorsalis*.

Key Words *Bactrocera dorsalis*, bioactivity, *Ageratina adenophora*, sublethal effects

The oriental fruit fly, *Bactrocera dorsalis* (Hendel) (Diptera: Tephritidae), is a destructive pest with a wide host range, infesting over 250 fruit and vegetable species from 46 families, including oranges, mangoes, and guavas (Yu et al. 2022, Zeng et al. 2019). Damage by *B. dorsalis* is caused primarily by female oviposition in the fruit. After the eggs hatch, the larvae feed on the fruit flesh, causing the fruit to rot internally and eventually fall before it is ripe, thereby seriously affecting fruit quality and yield and causing massive economic losses in fruit and vegetable production worldwide (Stephens et al. 2007, Wangithi et al. 2021).

Conventional chemical insecticides are effective against *B. dorsalis* (Banks et al. 2017, Li et al. 2021, Zhang et al. 2015); however, their negative effects (e.g.,

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environmental contamination, worker safety, and crop residues) have stimulated research into more environmentally friendly methods of managing the pest. In recent decades, there has been a surge of interest in the study of the potential use of plant-derived natural insecticides, such as plant extracts, which can be relatively ecofriendly for pest control in agriculture (Koul 2008). Many plant extracts have demonstrated biological activity against pests (Afiunizadeh et al. 2022, Phukhahad and Auamcharoen 2021, Wagan et al. 2018). Plant extracts also have demonstrated effective pest control properties in the field (Qari et al. 2020, Siam and Othman 2020).

Ageratina adenophora, an invasive weed species of the Asteraceae family, originated in Mexico and Costa Rica and has since spread throughout Southwest China (Ren et al. 2021, Wan et al. 2010). According to studies, *A. adenophora* and extracts of various parts of some plants exert toxic, repellent, and antifeeding effects on many pests (Mayanglambam et al. 2022, Nong et al. 2015, Samuel et al. 2014, Xu et al. 2009). These plant compounds can also cause detrimental effects on pest reproduction and survival, which are also desirable outcomes in the management of pest insect populations, as is the reduced risk of undesirable environmental effects. Plant extracts have been proposed as a suitable alternative tool for controlling pests (Pineda et al. 2023).

Botanicals (or their constituents) have various modes of action, including contact toxicity, repellency, antifeedant properties, and ovipositional deterrence (Carlini and Grossi-de-Sá 2002, Isman 2006). Several studies have reported the insecticidal and deterrent efficacy of various botanical extracts against different *Bactrocera* flies, such as *Bactrocera dorsalis* (Guo et al. 2020, Jaleel et al. 2020, Li et al. 2017), *Bactrocera zonata* (Saunders) (Ilyas et al. 2017, Khan et al. 2016, Rehman et al. 2009, Tajdar et al. 2020), and *Bactrocera correcta* (Bezzi) (Jaleel et al. 2020). Magrini et al. (2014) discovered that the extract of *Diospyros sylvatica* Robx. (Dilleniidae: Ebenaceae) was repellent and toxic to the tephritid *Anastrepha fraterculus* (Wiedemann). Currently, one study has investigated the effect of sublethal plant extract doses on *B. dorsalis*. (Zou et al. 2010). After the long-term exposure of *B. dorsalis* to an insecticide, detoxification enzymes, such as glutathione S-transferases (GSTs) and carboxylesterase (CarE), showed high levels of active responses, indicating that they were related to insecticide resistance (Meng et al. 2020, Sukhirun et al. 2011).

The objective of our study was to evaluate the toxicity and oviposition-deterrent activities of ethanol extracts from *A. adenophora* against adult *B. dorsalis*. We also examined the sublethal effects of the *A. adenophora* extract on *B. dorsalis* to further evaluate its potential for *B. dorsalis* control. Overall, our study aimed to explore new agents for controlling *B. dorsalis* as well as new ways to use invasive plants effectively. Because these weedy species are widely available, they can help reduce pest control costs. More botanical pesticides will be identified for use in pest biological control as our research continues.

Materials and Methods

Insects. The *B. dorsalis* adults used in this study were from a laboratory colony initially collected as adults from infested kiwifruit, *Actinidia deliciosa* (Chevalier) Liang & Ferguson, orchards in Shuicheng County, Guizhou Province, China (104°97′E, 26°42′N). Adults were housed in screen cages (30 × 30 × 30 cm) and

fed an artificial diet and water (Yuan et al. 2006). A disposable plastic cup (top diameter, 5 cm; bottom diameter, 4 cm; height, 6 cm) with a lid with several small oviposition holes for collecting the eggs (orange juice was placed in egg collection cups) was used as an artificial oviposition and egg collecting device. The larvae were transferred to a glass measuring cup (400 ml) containing larval artificial diets (Yuan et al. 2003). When the larvae exhibited jumping behavior, moist sand was provided as a pupation substrate. The colony was maintained in a growth chamber at 27±1°C, 70 ± 5% relative humidity, and on a light:dark photoperiod of 14:10 h). All assays were conducted under these same conditions.

Plant material and crude extract preparation. *Ageratina adenophora* plants used in this study were harvested from the Guizhou University Teaching Experiment Farm (106°68′E, 26°68′N). We obtained the crude extracts following methods described by Lima et al. (2022). Leaves were washed and air-dried at room temperature before being dried in an oven (DH-101, Beijing Ever Bright Medical Instrument Factory, Beijing, China) with forced air circulation at 50°C for 48 h. The dry plant material was then ground into a fine powder by using a disintegrator (Tianjin Taisite Instrument Co., Ltd., Tianjin, China). Cold maceration in ethanol (99.5%) solvent in a 5:1 (v/w) ratio was used for extraction. For this step, the powder was subjected to constant stirring with ethanol for 5 min before being kept at rest for 3 d. The solution was then filtered with filter paper no. 42. The aforementioned procedure was repeated three times to maximize gains. The solvents from the filtered samples were removed using a rotary evaporator (RE-52, Shanghai Yarong Biochemistry Instrument Factory, Shanghai, China) at 50°C to obtain a homogeneous and concentrated paste. Finally, the crude ethanolic extract of *A. adenophora* was obtained, taking the initial amount of dried plant material submitted to extraction into account. The extracts were then stored at 4°C until used.

Toxicity bioassays. As per Lin et al. (2013), we used the glass container residual film method. Based on our preliminary testing, we diluted the extract with ethanol into five concentrations (5, 10, 20, 40, and 80 mg/ml). A pipette was used to place 2 ml of the extract solution into a 250-ml conical flask, The flask was turned horizontally so that the extract coated the inner wall to form a residual film, the excess solution was discarded, and then the flask was placed on a rack upside down to air-dry until the ethanol was evaporated. Twenty adult flies (1 male:1 female) that had emerged 3 d earlier were transferred into each flask and fed with 10% honey solution. Each concentration was repeated three times, and ethanol was used as a control. Mortality was recorded after 24 h. Insects were considered dead if they did not respond to mild stimulation with a fine brush.

Oviposition-deterrent activity. Both choice and no-choice bioassays were performed with 30 mature flies (1 male:1 female). These flies were released in a cage (30 × 30 × 30 cm) for 24 h and used for oviposition bioassays the next day. In these assays, four concentrations of 5, 10, 20 and 40 mg/ml of the extract were tested. Ripe mangoes were dipped in the appropriate test solution for 10 s and then allowed to air-dry before being placed into the cages. Control group mangoes were dipped in ethanol only.

In choice bioassays, both control and treated mangoes were placed inside a cage at opposite ends. After 24 h, treated and control mangoes were removed and eggs that were present were counted. A similar procedure was used for the no-choice

bioassays, with the exception that a single mango (either treated or control) was placed at the center of the cage. As in the choice tests, the mango was removed from the cage after 24 h and the eggs on the fruit were counted and recorded. Each treatment in these tests was replicated three times. Percentage effective repellency (%ER) was calculated as described by Li (2017) using the formula %ER = (NC − NT/NC + NT) × 100 for the choice tests, and %ER = (NC − NT/NC) ×100 for the no-choice tests, where NC is the number of eggs in the control and NT is the number of eggs in the treatment.

To determine the residual deterrent activity of the extract, the treated mangoes (concentration: 20 mg/ml, this concentration did not exhibit a significant difference in its repellent efficacy compared to that of 40 mg/ml) were exposed to the adult flies at various intervals, ranging from 1 to 5 d. The treatment intervals were replicated three times.

Sublethal exposure impact to adults. Adults were treated with sublethal doses of LC_{10} and LC_{25} (the estimated dose causing the death of 10% and 25% of the tested population in a given time) of *A. adenophora* extract, as well as ethanol as a control. Following treatment, the surviving F_0 generation adults were chosen. Thirty male and female pairs were caged and fed artificial food and water. After 5 d of rearing, homemade oviposition cups containing orange juice were placed in the boxes to trap the female insects for oviposition. Food and oviposition cups were replaced every day. The survival, reproduction, and longevity of adult insects were observed daily. Each experiment was repeated three times.

For the F_1 generation survival study, 1-d-old eggs were randomly collected from the cup by using an insect-sweeping pen and placed in a petri dish with moist filter paper. Twenty eggs were placed in each petri dish with 10 replications. Next, the hatched larvae were transferred to a glass measuring cup (100 ml) containing larval artificial diets. When the larvae exhibited jumping behavior, they were transferred to a plastic box (15 × 10 × 8 cm) filled with moist sand for the larvae to bore into to pupate. Each day thereafter, the eggs, larvae, and pupa were monitored for development and survival until they turned black or died.

For the F_1 generation development study, many eggs were collected within 3 h and fed in a glass cup (400 ml) containing an artificial diet. Ten fruit fly eggs were placed on glass dishes (diameter = 10 cm) containing a 1.5-cm-thick artificial diet. Six replicates were used for each treatment. The eggs were monitored using a stereoscopic microscope every 6 h. Larvae that hatched at the same time were captured and divided into groups of 20 each and placed on an artificial diet of nearly 1.5-cm thickness in petri dishes. Larvae were monitored every 12 h. Each treatment was replicated three times. After the larvae attained maturity (exhibited bouncing behavior), the same cohorts of pupae were divided into groups of 20 each and placed in boxes (15 × 10 × 8 cm) filled to a depth of 3 or 4 cm of sand. Three replicates were used per treatment, and pupae were observed every 12 h.

Detoxification enzyme assay. Adults were treated (12 h, 24 h, and 48 h) with the extract concentration equal to the determined LC_{50}. Treatment with ethanol alone served as the control. Following treatment, the surviving adults were randomly selected from the treatment groups at 12, 24, and 48 h after exposure. These specimens were rapidly frozen in liquid nitrogen and transferred into a centrifuge tube. The activities of GSTs were measured using the GSTs enzyme Assay Kit (Suzhou Michy Biomedical Technology Co., Ltd., Suzhou, China). The homogenates were

Table 1. Lethal toxicity of *Ageratina adenophora* extract to adult *Bactrocera dorsalis*.

Treatment	n	Regression Equation	LC_{50} (95% CL)	LC_{95} (95% CL)	χ^2
Ageratina adenophora	360	$y = 2.004x - 2.836$	26.014 (16.067–47.721)	172.234 (77.714–1699.206)	6.832

n, Total number of adults used; CL, confidence limits.

centrifuged at 8,000 × *g* for 10 min at 4°C, and the supernatants were collected. The reagents were spiked following the manufacturer's instructions. Absorbances were measured at 340 nm by using a microplate reader. Three replicates were used per treatment.

The CarE enzyme activities were measured using the CarE enzyme Assay Kit (Suzhou Michy Biomedical Technology Co., Ltd.). The homogenates were centrifuged at 12,000 ×g for 30 min at 4°C, and the supernatants were collected. Absorbance was determined at 450 nm. Each treatment was repeated three times.

The protein level of the adult insects was measured via the bicinchoninic acid method with the Protein Assay Kit (Suzhou Kemin Biotechnology Co., Ltd.). The homogenates were centrifuged at 12,000 × *g* for 10 min at 4°C, and the supernatants were collected. Absorbances were measured at 562 nm using a microplate reader. There were three replicates in each treatment.

Statistical analysis. SPSS for Windows was used for all data analyses (Statistical Package for the Social Sciences, Version 21.0, Chicago, IL). Probit regression was used to analyze mortality data from adult toxicity bioassays. The *t*-test was used to compare control and treatment groups in oviposition bioassays. Other experimental data were statistically analyzed using one-way analysis of variance. Tukey's test (equal variance) was used to compare means, and significant differences were determined at $P < 0.05$.

Results

Concentration-mortality response. The toxicity bioassay estimated the LC_{50} value of the *A. adenophora* extract against adult *B. dorsalis* to be 26.014 mg/ml and LC_{95} as 172.234 mg/ml (Table 1). Extrapolation of those results showed that the LC_{10} and LC_{25} values were 5.965 and 11.983 mg/ml, respectively. The sublethal effects of the extract on *B. dorsalis* were subsequently examined using the LC_{10}, LC_{25}, and LC_{50} concentrations.

Oviposition-deterrent activity. In our choice tests, the number of eggs laid on the treated mangoes, regardless of the concentration of the extract applied, was significantly ($P < 0.001$) lower than that in the untreated control group (Table 2). Although the test was not designed to compare oviposition response among the various extract concentrations, we observed an increase in the oviposition-deterrent activity of the extract against *B. dorsalis* adults as the extract concentration increased.

We were able to better examine the ovipositional response to extract concentration in the no-choice assays, where we observed significant ($F = 46.308$; df = 4, 10;

Table 2. The oviposition repellent activity of *Ageratina adenophora* extract against *Bactrocera dorsalis* (choice test).

Concentration (mg/mL)	No. of Eggs		Effective Repellency (%)
	Treatment	Control	
5	75.3 ± 10.5**	202.3 ± 3.2	45.74
10	50.3 ± 8.1**	190.3 ± 6.7	58.17
20	22.7 ± 3.5**	142.0 ± 12.2	72.47
40	14.3 ± 7.5**	127.7 ± 7.0	79.81

Data are mean ± SE. The double asterisks indicate significant differences between the control and treatment at the levels of 0.01 (*t*-test).

$P < 0.001$) differences in the numbers of eggs laid among the treatments (Table 3). The highest concentrations (20 and 40 mg/ml) did not differ significantly from each other. The %ER ranged from 47.37% at 5 mg/ml to 84.66% at 40 mg/ml.

We also observed that the number of eggs laid on mangoes treated with ethanol only (control) ranged from a mean ± SE of 242.3 ± 8.4 per day on the fifth day to 324.7 ± 10.4 per day on the first day after treatment (Table 4). The number of eggs laid on the mangoes treated with the *A. adenophora* extract ranged from 63.7 ± 8.0 to 171.3 ± 18.3 eggs per day, of which all were significantly less than the respective controls (0 day: $t = 19.927$, df = 4, $P < 0.001$; 1 day: $t = 12.301$, df = 4, $P < 0.001$; 2 days: $t = 13.858$, df = 4, $P < 0.001$; 3 days: $t = 7.520$, df = 4, $P = 0.002$; 4 days: $t = 6.305$, df = 4, $P = 0.003$; 5 days: $t = 3.541$, df = 4, $P = 0.024$) (Table 4). The %ER declined over time with values of 80.4% on 0 day to 29.2% on day 5.

Sublethal effects of *A. adenophora* extract on the F_0 generation of *B. dorsalis* adults. The exposure of *B. dorsalis* adults to mangoes treated with extract concentrations corresponding to the LC_{25} and LC_{10} levels demonstrated sublethal effects in the F_0 generation *B. dorsalis* adults. The preoviposition period was slightly, but significantly, longer when exposed to either of the two concentrations ($F = 8.000$;

Table 3. The oviposition repellent activity of *Ageratina adenophora* extract against *Bactrocera dorsalis* (no-choice test).

Concentration (mg/mL)	No. of Eggs	Effective Repellency (%)
5	193.3 ± 12.4b	47.37
10	118.7 ± 21.9bc	67.70
20	71.7 ± 16.7c	80.49
40	56.3 ± 6.2c	84.66
Control	367.3 ± 28.1a	

Data are mean ± SE. Different letters in the same column indicate significant differences among different concentrations by Tukey's test ($P < 0.05$).

Table 4. The oviposition repellent activity of *Ageratina adenophora* extract against *Bactrocera dorsalis* with different time.

Time (d)	No. of Eggs		Effective Repellency (%)
	Treatment	Control	
0	63.7 ± 8.0**	324.7 ± 10.4	80.39
1	88.4 ± 8.6**	317.7 ± 16.6	72.19
2	113.3 ± 8.4**	298.3 ± 10.4	62.01
3	139.0 ± 8.9**	260.0 ± 13.3	46.41
4	157.0 ± 10.0**	250.0 ± 9.9	36.93
5	171.3 ± 18.3*	242.3 ± 8.4	29.16

Data are mean ± SE. The single and double asterisks indicate significant differences between the control and treatment at the levels of 0.05 and 0.01, respectively (*t*-test).

df = 2, 6; $P = 0.020$), whereas male and female longevity were significantly reduced following exposure to the LC_{25} concentration (male: $F = 8.056$; df = 2, 6; $P = 0.020$) (female: $F = 19.417$; df = 2, 6; $P = 0.002$) (Table 5). Fecundity decreased as concentration increased, with a significant difference among treatments ($F = 44.761$; df = 2, 6; $P < 0.001$) (Fig. 1A). The survival rate after exposure to the LC_{10} treatment was similar to that of the control group from 0 to 60 d, but from 70 to 110 d, the survival rate was similar to that of those treated with the LC_{25} treatment (Fig. 1B).

Sublethal effects of *A. adenophora* on the F_1 generation of *B. dorsalis*. Compared with the control treatment, the hatching and emergence rates of the F_1 generation treated with LC_{25} were significantly reduced by 11.00% and 7.50%, respectively (hatching: $F = 11.581$; df =2, 27; $P < 0.001$) (emergence: $F = 4.056$; df = 2, 15; $P = 0.039$) (Table 6). However, there was no significant difference in pupation and female ratio between the treatments in F_1.

The duration of eggs was the shortest in those who were treated with *A. adenophora* extract at LC_{10} ($F = 12.620$; df = 2, 15; $P = 0.001$) (Table 6), with no difference observed in the durations of eggs in those who were treated with control and LC_{25}.

Table 5. The longevity, preoviposition period and fecundity after treatment of F_0 generation adults with *Ageratina adenophora* extract.

Treatment	Preoviposition Period (d)	Fecundity (Number of Eggs Laid per Female)	Adult Longevity (d)	
			Female	Male
Control	11.00 ± 0.00a	264.68 ± 4.48a	67.08 ± 1.40a	79.28 ± 3.19a
LC_{10}	12.33 ± 0.33b	210.52 ± 13.35b	60.43 ± 2.04a	68.36 ± 4.10ab
LC_{25}	12.33 ± 0.33b	126.22 ± 11.39c	50.54 ± 2.14b	60.74 ± 2.32b

Values (mean ± SE) followed by different letters in a column are significantly different ($P < 0.05$; Tukey's test).

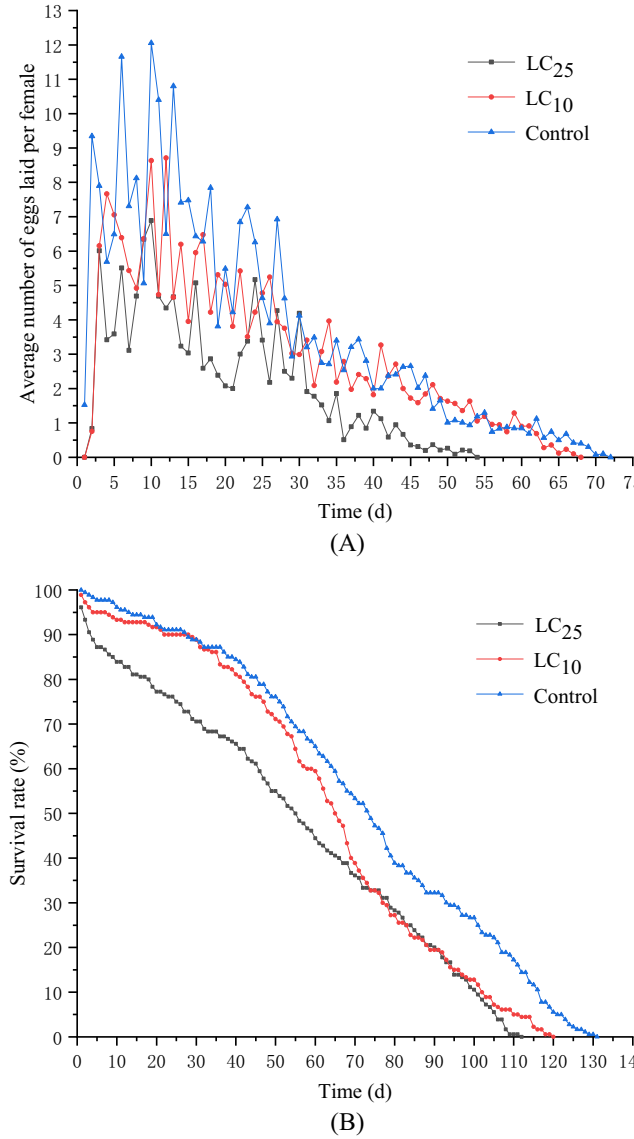

Figure 1. Fecundity dynamics (A) and survival rate dynamics (B) of adult *Bactrocera dorsalis* exposed to *Ageratina adenophora*.

The results of the larval and pupal development time were comparable between larvae treated at LC_{10} and LC_{25} and those in the control group.

Detoxification enzyme activities of sublethal effects of *A. adenophora* in *B. dorsalis*. There was no significant difference in adult GST activity between the results obtained at treatment and those in the control group (Fig. 2A). The responses

Table 6. Developmental duration and survival rate of the F_1 generation of *Bactrocera dorsalis*.

	Egg		Larva		Pupa		Adult
Treatment	Developmental duration (d)	Hatching rate (%)	Developmental duration (d)	Pupation rate (%)	Developmental duration (d)	Emergence rate (%)	Female proportion (%)
Control	2.04 ± 0.03b	81.50 ± 2.12a	8.10 ± 0.13a	98.57 ± 0.92a	10.05 ± 0.08a	95.83 ± 1.54a	46.00 ± 1.98a
LC_{10}	1.82 ± 0.02a	82.00 ± 2.00a	8.33 ± 0.05a	95.71 ± 1.30a	10.16 ± 0.09a	89.17 ± 1.54ab	47.51 ± 3.69a
LC_{25}	2.02 ± 0.05b	70.50 ± 1.57b	8.40 ± 0.10a	96.43 ± 1.43a	10.13 ± 0.05a	88.33 ± 2.79b	47.83 ± 2.02a

Values (mean ± SE) followed by different letters in the same column are significantly different ($P < 0.05$; Tukey's test).

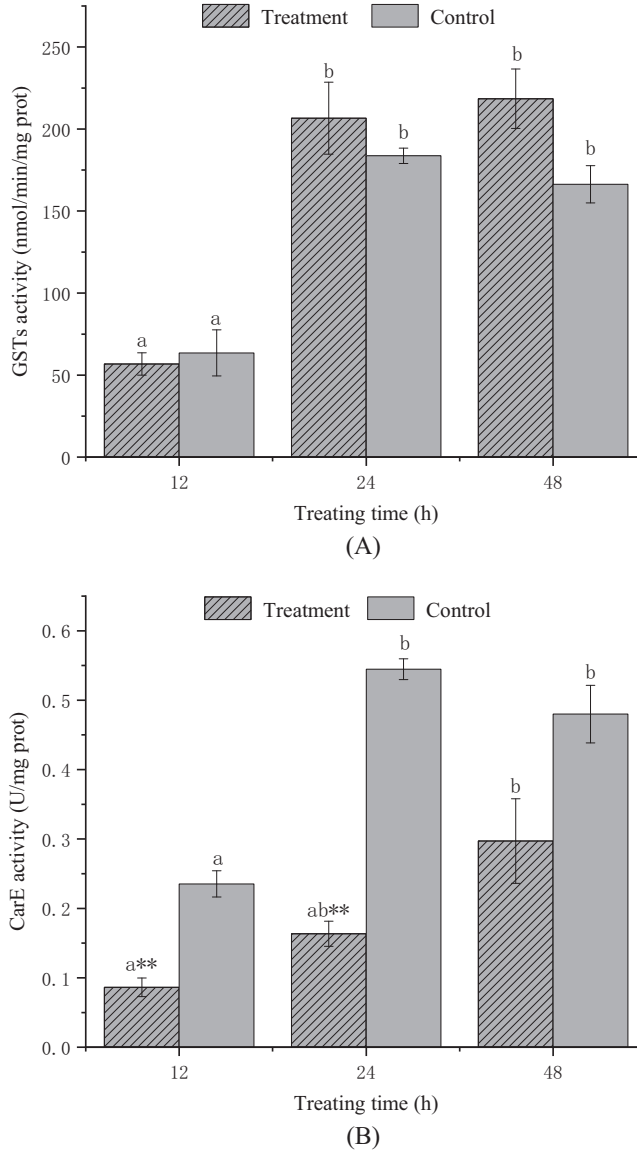

Figure 2. GSTs activity (A) and CarE activity (B). Different letters above bars represent significant differences at the 0.05 level among different time at the same treatment (Tukey's test). "" indicates significant differences at the 0.01 level among different treatments at the same time.**

of the adult CarE enzyme to *A. adenophora* stress appeared to differ, with the CarE activity at 12 h and 24 h being significantly lower than that of the control (Fig. 2B).

Discussion

According to the habit of the oriental fruit fly, spraying pesticides on fruit to control the population of *B. dorsalis* can interfere with reproductive behavior or toxicity. Our results showed that *A. adenophora* extract has significant bioactivity against *B. dorsalis*, including the potential for toxicity and deterrent effects on oviposition. *Ageratina adenophora* was chosen because its extracts are known for their richness of bioactive secondary metabolites (Darji et al. 2021, Zhao et al. 2009). The insecticidal activity may be attributed to the presence of the primary components within the extract. Nong et al. (2015) discovered that 9-oxo-10,11-dehydro-ageraphorone (isolate from *A. adenophora* petroleum ether extract) exhibits significant aphicidal activity against *Pseudoregma bambucicola* (Takahashi). At a concentration of 2 mg/ml, this ageraphorone compound caused a 73.33% mortality rate of *P. bambucicola* within 6 h in a laboratory bioassay. There are other reports that also suggest that extracts from plants, such as rosmarinic acid and tobacco leaves, are toxic to *B. dorsalis* (Mughees et al. 2021, Wang et al. 2022).

Females prefer to oviposit in untreated control fruits rather than in *A. adenophora*-treated ones regardless of the application method (choice or no-choice), and the % ER decreased with increasing time. In addition to toxicity, many extracts have oviposition inhibitory activity (Silva et al. 2019). Guo et al. (2020) also discovered that the toxic and repellent properties of citronella oil (7 mg/ml) against *B. dorsalis* resulted in high mortality rates of 87.78% and high repellent effects (80.77%). We found little difference in the %ER at higher concentrations, which corroborated findings of Li et al. (2017) who discovered that an acetone extract of *Aloe vera* (L.) Burm. (1 and 10 mg/ml) had repellent activity (86.40 and 89.43%) against *B. dorsalis*; these results did not differ significantly and the duration of the %ER is consistent with our results. Our results suggest that potential future applications of the extract or their active components for *B. dorsalis* control may exploit more than one mode of action.

The development of new biopesticides based on plant extracts requires a thorough understanding of their potential efficacy against pests. Our findings indicate the potential for sublethal effects of *A. adenophora* on the reproductive physiology of *B. dorsalis*. Treatment with the tested LC_{25} resulted in significant reductions in the total number of eggs produced and adult survival and alterations in preoviposition. Srinivasan et al. (2015) showed that extracts of *Melia azedarach* (L.) seed kernels significantly reduced the fecundity, egg-hatching rates, and pupal weight of *B. dorsalis*. These results are in agreement with a previous study showing pronounced similar effects of *A. adenophora* extract on other insects (Wu et al. 2020). According to the findings of Cheng et al. (2007), feeding the extract of *A. adenophora* may inhibit *Helicoverpa armigera* (Hübner) growth and reproduction, and concentrations of 0.08, 0.4, and 2 g/L showed an attraction effect on the oviposition of the bollworm females, but a concentration of 10 g/L showed a repellent effect for oviposition. Exposure induced a small effect on F_1 generation, causing harmful effects only on the duration of the egg developmental period, hatchability, and emergence. Thus, the results indicate that once fruit fly females can survive the insecticide treatment,

they can still harm the host plants. The results of promoting egg development at low concentrations were similar to the study of Wang et al. (2022). The activity of the adult CarE enzyme used to treat *A. adenophora* appeared different, showing an established inhibition of CarE. The same sublethal effects have been reported in the past. For example, the hexane and ethanol extracts of *Alpinia galanga* (L.) Willd rhizomes were effective against adult *B. dorsalis* (LC_{50} = 4,866 and 6,337 ppm) after 24 h, the activity of the hexane extract against CarE was significantly inhibited, and GSTs were not significantly inhibited (Sukhirun et al. 2011). Considering the excellent activity of *A. adenophora* and the resistance status of *B. dorsalis*, this extract may be a good alternative insecticide for the control of these insects.

The current study discovered that the extract of *A. adenophora* has insecticidal and oviposition-repellent activities. Sublethal concentrations of the *A. adenophora* extract caused significant changes in the life cycle of *B. dorsalis*, and the CarE enzyme might partially alleviate the toxicity damages. The extract was shown to have a high potential for use as botanical insecticides and oviposition inhibitors against the oriental fruit fly. Future research will be needed to purify the active compounds from these extracts and investigate these findings in field testing.

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