

植物挥发物的筛选及其与硅藻土联合对绿豆象防效和酶活性的影响



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摘要: 为寻求绿色高效防治绿豆象 *Callosobruchus chinensis* 的方法, 采用密闭熏蒸法测定茴香烯、柠檬醛、4-烯丙基苯甲醚、丁香酚 4 种植物挥发物对绿豆象成虫的熏蒸效果, 选择熏蒸效果最佳的植物挥发物, 将其与硅藻土联合, 测定联合后对绿豆象成虫的室内毒力、防效及其对绿豆象体内多种酶活性的影响。结果显示: 在 4 种植物挥发物中茴香烯对绿豆象成虫的熏蒸效果最佳, 5 $\mu\text{L/L}$ 茴香烯处理 48 h 时, 绿豆象成虫的校正死亡率达到 100.00%; 茴香烯和硅藻土联合处理表现出显著的协同增效作用, 其对绿豆象成虫的半致死剂量 LC_{50} 为 3.040 $\mu\text{L/L}$, 而茴香烯对绿豆象成虫的 LC_{50} 为 6.979 $\mu\text{L/L}$; 硅藻土对绿豆象成虫体内过氧化氢酶、乙酰胆碱酯酶、琥珀酸脱氢酶、过氧化物酶、谷胱甘肽-S-转移酶和超氧化物歧化酶活性均无影响。表明茴香烯与硅藻土联合可用于绿色防控绿豆象。
关键词: 绿豆象; 茴香烯; 硅藻土; 熏蒸; 酶活性

Screening of plant volatiles and their combined effects with diatomaceous earth on control efficacy and enzyme activities in Chinese bean weevil *Callosobruchus chinensis*

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Abstract: To identify an environmentally friendly and efficient method for controlling Chinese bean weevil *Callosobruchus chinensis*, a sealed fumigation method was used to evaluate the fumigation effects of four plant volatiles (*trans*-anethole, citral, 4-allylanisole, and eugenol) on adult *C. chinensis*. The plant volatile with the best fumigation effect was selected and combined with diatomaceous earth to determine its laboratory toxicity, control efficacy, and effects on multiple enzyme activities in *C. chinensis*. The results showed that among the four plant volatiles, *trans*-anethole exhibited the strongest fumigation effect. At a concentration of 5 $\mu\text{L/L}$, *trans*-anethole achieved a corrected mortality rate of 100.00% against for adult *C. chinensis* after 48 hours of treatment. The combination of *trans*-anethole and diatomaceous earth demonstrated a significant synergistic effect, with an LC_{50} of 3.040 $\mu\text{L/L}$ against adult *C. chinensis*, compared to 6.979 $\mu\text{L/L}$ for *trans*-anethole alone. Diatomaceous earth alone had no effect on the activities of catalase, acetylcholinesterase, succinate dehydrogenase, peroxidase, glutathione-S-transferase, and superoxide dismutase in *C. chinensis*. These findings indicate that the

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combination of *trans*-anethole and diatomaceous earth can be used for the green control of *C. chinensis*.

Key words: *Callosobruchus chinensis*; *trans*-anethole; diatomaceous earth; fumigation; enzymatic activity

绿豆象 *Callosobruchus chinensis* 属鞘翅目叶甲科豆象亚科,是一种严重为害豆类的田间和仓储害虫(Tuda et al., 2005)。绿豆象的初侵染发生在田间,雌成虫在嫩豆荚上产卵,幼虫孵化后蛀入豆粒进行为害,并随绿豆的收获进入粮仓,对仓储绿豆进行二次侵染,为害严重时损失率可达30%~56%(仲建锋等,2013)。目前针对仓储害虫主要采用磷化铝、甲基嘧啶磷等熏蒸剂进行防治(Valmas & Ebert, 2006),但该方法会引发抗药性、环境污染和食品安全风险等问题。因此,寻求生态友好和可持续的替代方法用于防控储粮害虫迫在眉睫(王淳静等,2021)。

植物挥发物对害虫具有胃毒、熏蒸、触杀和拒食等作用,且易于降解,对环境无污染,对非靶标物种为害小,长期施用不易使害虫产生抗药性(宋程飞等,2022)。例如,茶皂素显著影响东亚飞蝗 *Locusta migratoria manilensis* 的取食、消化及生长发育(王铭铭等,2023);天竺葵 *Pelargonium hortorum* 提取物芳樟醇、香茅醇和香叶醇对赤拟谷盗 *Tribolium castaneum* 和谷蠹 *Rhyzopertha dominica* 均有较好的熏蒸效果(Abouelatta et al., 2020);低浓度的香豆素能显著影响米象 *Sitophilus oryzae*、赤拟谷盗和绿豆象这3种储粮害虫的生长发育(Rajashekar et al., 2013);0.04 $\mu\text{L}/\text{cm}^2$ 海茴香 *Crithmum maritimum* 精油对赤拟谷盗驱避率和触杀致死率分别为93%和50%(Mustapha et al., 2020);4%水黄皮 *Pongamia pinnata* 提取物处理48 h时对橘小实蝇 *Bactrocera dorsalis* 1龄幼虫的胃毒活性最强,校正死亡率为95.9%(任军华等,2022)。此外,在外源有毒物质胁迫下,昆虫通过调节自身保护酶、解毒酶等多种酶系活性来维持正常的生长发育,从而降低植物挥发物对自身的为害。例如,乙酸乙酯熏蒸后赤拟谷盗、谷蠹体内超氧化物歧化酶(superoxide dismutase, SOD)、过氧化氢酶(catalase, CAT)活性均被激活;茴香烯熏蒸处理后米象体内羧酸酯酶(carboxylesterase, CarE)和谷胱甘肽-S-转移酶(glutathione S-transferase, GST)的活性被激活,但乙酰胆碱酯酶(acetylcholinesterase, AchE)活性被显著抑制(Bhavya et al., 2018; Adesina, 2023)。本课题组前期研究发现茴香烯、柠檬醛、4-烯丙基苯甲醚、丁香酚这4种植物挥发物对绿豆象有显著的驱避效果(张苗等,2025),但这4种物质对绿

豆象的杀虫活性及生理生化影响尚不清楚。

硅藻土主要成分为二氧化硅,是一种物理化学性质稳定的粉状物质,具有作用效果持久、不会产生抗性、不受温度影响等优点,已被广泛用于防治储粮害虫(Shah & Khan, 2014; 王洋等, 2021)。硅藻土与植物挥发物结合可增强植物挥发物的杀虫活性,减少其使用剂量,延长其作用时间(Zeni et al., 2021)。例如,硅藻土与大蒜精油联合使用对米象有协同作用(Yang et al., 2010);硅藻土与苦霉素联合可增强苦霉素对害虫的毒杀作用(Athanassiou et al., 2009)。为此,本研究采用密闭熏蒸法测定茴香烯、柠檬醛、4-烯丙基苯甲醚、丁香酚这4种植物挥发物对绿豆象成虫的熏蒸作用,筛选出最佳的熏蒸活性物质,研究该物质与硅藻土联合使用对绿豆象成虫的防治效果,并进一步分析联合使用后绿豆象成虫体内保护酶、解毒酶和呼吸酶活性的变化,期为绿豆象的绿色防控提供参考。

1 材料与方法

1.1 材料

供试虫源和植物:2024年9月自山西农业大学农作站带虫卵的绿豆中获取绿豆象种群,并于山西农业大学昆虫重点实验室于温度(27 ± 1) $^{\circ}\text{C}$ 、相对湿度(75 ± 5)%、光周期16 L:8 D的恒温光照培养箱中用绿豆连续饲养多代,挑选12 h内羽化的大小基本一致、健康活跃的成虫供试。绿豆象饲养所用绿豆为晋绿1号,由山西农业大学农作站提供。

试剂和仪器:99%茴香烯、97%柠檬醛、99%丁香酚、98%4-烯丙基苯甲醚标准品,上海阿拉丁生化科技股份有限公司;食品级硅藻土,郑州高晶新材料有限公司;CAT、过氧化物酶(peroxidase, POD)、SOD、GST、CarE、AchE、琥珀酸脱氢酶(succinate dehydrogenase, SDH)活性检测试剂盒及BCA法蛋白质浓度测定试剂盒,生工生物工程(上海)股份有限公司;其他试剂均为国产分析纯。Spectra Max M5酶标仪,美国Molecular Devices公司。

1.2 方法

1.2.1 不同植物挥发物对绿豆象成虫熏蒸效果的测定

采用玻璃瓶密闭熏蒸法测定茴香烯、柠檬醛、4-烯丙基苯甲醚和丁香酚这4种植物挥发物对绿豆象成

虫的熏蒸效果。在500 mL蓝盖试剂瓶瓶口处涂抹聚四氟乙烯防止试虫逃逸,待其挥发后每瓶接入绿豆象成虫50头(1个重复)。茴香烯、柠檬醛、4-烯丙基苯甲醚和丁香酚4种标准品分别吸取1.0、2.5和5.0 μL ,滴加至长2 cm、宽1 cm的滤纸条上,即每种植物挥发物浓度分别为2、5和10 $\mu\text{L/L}$;将滤纸条悬挂于瓶内,距瓶底6 cm,拧紧瓶盖。每个浓度重复3次,以滴加等体积的无水乙醇作为对照。将试剂瓶置于温度(27 \pm 1) $^{\circ}\text{C}$ 、相对湿度(75 \pm 5)%的恒温培养箱内黑暗条件下分别熏蒸24、36、48、60和72 h,散气12 h后检查绿豆象成虫存活情况并记录。用毛笔轻触试虫背部,试虫无运动迹象和肢体伸展者视为死亡。根据死亡情况计算死亡率和校正死亡率。校正死亡率=(处理死亡率-对照死亡率)/(1-对照死亡率) \times 100%。

1.2.2 茴香烯与硅藻土联合对绿豆象成虫毒力的测定

根据1.2.1试验结果,茴香烯对绿豆象成虫的熏蒸效果最好,故选择茴香烯进行毒力测定。茴香烯单独处理时,将100 g绿豆放入500 mL蓝盖试剂瓶中,在瓶口涂抹聚四氟乙烯防止试虫逃逸,待其挥发后,每瓶接入50头绿豆象成虫(1个重复),分别吸取2.50、3.25、4.00、4.75和5.50 μL 茴香烯标准品,滴加到长2 cm、宽1 cm的滤纸条上,即茴香烯浓度分别为5.0、6.5、8.0、9.5和11.0 $\mu\text{L/L}$;将滤纸条悬挂于瓶内,距瓶底6 cm,拧紧瓶盖。茴香烯与硅藻土联合处理时,按照每千克绿豆拌入1.2 g硅藻土(该剂量在预试验中杀虫效果最好且不超过国标规定剂量),使绿豆均匀粘附硅藻土粉末(Athanassiou et al., 2003);将100 g绿豆放入500 mL蓝盖试剂瓶中,在瓶口涂抹聚四氟乙烯防止试虫逃逸,待其挥发后,每瓶接入50头绿豆象成虫,用微量进样器分别吸取1.25、1.50、1.75、2.00和2.25 μL 茴香烯标准品,滴加到长2 cm、宽1 cm的滤纸条上,即茴香烯浓度分别为2.5、3.0、3.5、4.0和4.5 $\mu\text{L/L}$;将滤纸条悬挂于瓶内,距瓶底6 cm,拧紧瓶盖。以滴加等体积的无水乙醇为对照,每个浓度处理重复3次。将试剂瓶置于温度(27 \pm 1) $^{\circ}\text{C}$ 、相对湿度(75 \pm 5)%的恒温培养箱内黑暗条件下培养,48 h后检查绿豆象成虫存活情况并记录,计算死亡率和校正死亡率,方法同1.2.1。以茴香烯浓度对数值为自变量,以死亡概率值为因变量进行毒力回归方程拟合,根据毒力回归方程求出半致死剂量 LC_{50} 及95%置信区间,并对回归方程进行 χ^2 检验。

1.2.3 硅藻土对茴香烯杀虫活性的增效作用

根据1.2.2试验结果,选择 LC_{50} 茴香烯与1.2 g/kg硅藻土联合处理绿豆象成虫, LC_{50} 茴香烯浓度为3.040 $\mu\text{L/L}$,分别以 LC_{50} 茴香烯单独处理和1.2 g/kg硅藻土单独处理为对照,试验方法同1.2.2。每隔12 h检查绿豆象成虫存活情况并记录,计算死亡率和校正死亡率,方法同1.2.1。

1.2.4 绿豆象成虫体内酶活性的测定

根据1.2.2试验结果, LC_{50} 茴香烯与1.2 g/kg硅藻土联合处理,并设 LC_{50} 茴香烯单独处理、1.2 g/kg硅藻土单独处理及滴加等体积的无水乙醇对照,方法同1.2.2。每瓶放入50头绿豆象(1个重复),每个处理时间设3个重复,处理后12、18、24、30和36 h,每个处理随机收集15头存活试虫,置于1.5 mL离心管中,液氮速冻后研磨并加入1 mL预冷的1 \times 磷酸盐缓冲溶液,充分混匀后,于4 $^{\circ}\text{C}$ 、12 000 r/min条件下离心15 min,使用移液枪吸取上清液,置于-80 $^{\circ}\text{C}$ 超低温冰箱中保留,即待测酶液。取制备的待测酶液0.1 mL,用1 \times 磷酸盐缓冲溶液稀释10倍,根据BCA法蛋白质浓度测定试剂盒说明书测定蛋白质浓度。取制备的待测酶液作为样品,分别按照CAT、POD、SOD、AChE、CarE、GST和SDH活性检测试剂盒说明书测定吸光度值,按照说明书提供的公式计算各酶活性。

1.3 数据分析

采用SPSS Statistics 27.0软件对试验数据进行统计分析,使用单因素方差分析中Tukey检验法进行差异显著性检验。

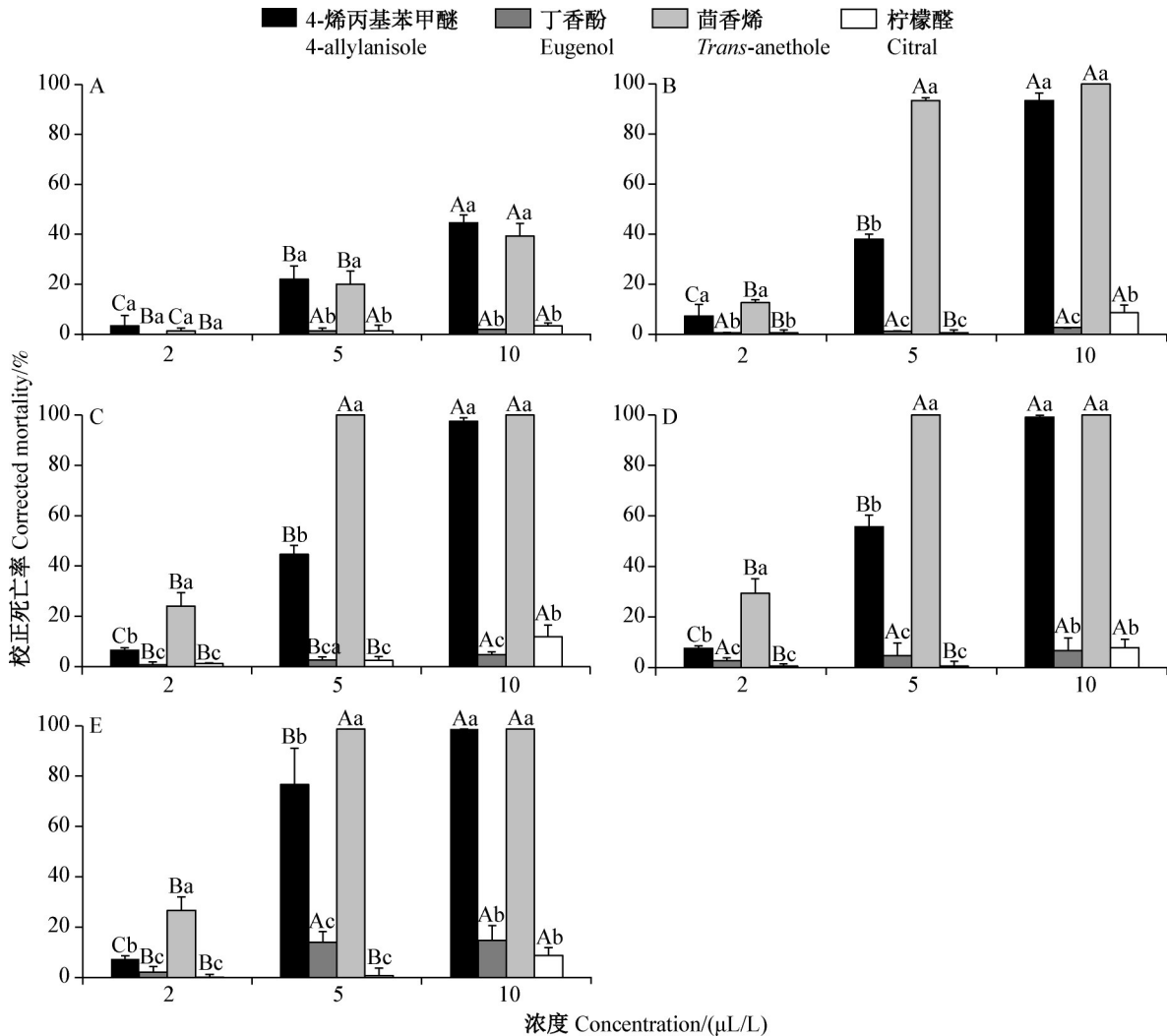
2 结果与分析

2.1 不同植物挥发物对绿豆象成虫的熏蒸效果

4-烯丙基苯甲醚、丁香酚、茴香烯和柠檬醛4种植物挥发物对绿豆象成虫均有一定的熏蒸效果,且随熏蒸浓度增加和熏蒸时间延长,绿豆象成虫的校正死亡率逐渐增加。当熏蒸24 h时,5 $\mu\text{L/L}$ 和10 $\mu\text{L/L}$ 茴香烯熏蒸后绿豆象成虫的校正死亡率与相同浓度4-烯丙基苯甲醚熏蒸后绿豆象成虫的校正死亡率相当,但均显著高于其他处理($P<0.05$,图1-A)。当熏蒸36 h时,5 $\mu\text{L/L}$ 茴香烯熏蒸后绿豆象成虫的校正死亡率为93.33%,显著高于其他处理($P<0.05$),10 $\mu\text{L/L}$ 茴香烯熏蒸后绿豆象成虫的校正死亡率达100.00%,表现出强熏蒸效果(图1-B)。熏蒸48 h时,2 $\mu\text{L/L}$ 和5 $\mu\text{L/L}$ 茴香烯熏蒸后绿豆象成虫的校正死亡率分别为24.00%和100.00%,均显著高于其

他处理($P<0.05$); 10 $\mu\text{L/L}$ 茴香烯和4-烯丙基苯甲醚熏蒸后绿豆象成虫的校正死亡率分别为99.93%和100.00%,两者之间差异不显著,但均显著高于其他处理($P<0.05$,图1-C)。熏蒸60 h时,5 $\mu\text{L/L}$ 4-烯丙基苯甲醚熏蒸后绿豆象成虫的校正死亡率继续较熏蒸48 h时增加,但仍显著低于5 $\mu\text{L/L}$ 茴香烯处理

($P<0.05$,图1-E)。当5 $\mu\text{L/L}$ 4-烯丙基苯甲醚熏蒸时间延长至72 h时,绿豆象成虫的校正死亡率为76.68%,显著低于5 $\mu\text{L/L}$ 茴香烯处理($P<0.05$); 10 $\mu\text{L/L}$ 丁香酚和柠檬醛熏蒸72 h时,绿豆象成虫的校正死亡率仅为14.68%和8.69%,显著低于10 $\mu\text{L/L}$ 茴香烯处理($P<0.05$,图1-E)。因此,选择茴香烯进行后续试验。



A~E: 熏蒸时间分别为24、36、48、60和72 h。A~E: Fumigation times of 24, 36, 48, 60, and 72 h, respectively.

图1 4种植物挥发物对绿豆象成虫的熏蒸效果

Fig. 1 Fumigation effects of four plant volatiles against *Callosobruchus chinensis* adults

图中数据为平均数±标准误。不同大写字母表示相同植物挥发物不同浓度之间经Tukey检验法检验差异显著($P<0.05$); 不同小写字母表示相同浓度下不同植物挥发物之间经Tukey检验法检验差异显著($P<0.05$)。Data are mean±SE. Different uppercase letters indicate significant differences among different concentrations within the same plant volatile according to Tukey's test ($P<0.05$), and different lowercase letters indicate significant differences among treatments of plant volatiles at the same concentration.

2.2 茴香烯与硅藻土联合对绿豆象成虫的毒力

茴香烯单独处理及其与硅藻土联合处理后,绿豆象的校正死亡率均随其浓度的增加而显著增加($P<0.05$,图2)。对上述浓度的对数值和绿豆象的

校正死亡率进行拟合,得到茴香烯单独处理及其与硅藻土联合处理对绿豆象成虫的毒力回归方程,经计算茴香烯单独处理及其与硅藻土联合处理对绿豆象的 LC_{50} 分别为6.979 $\mu\text{L/L}$ 和3.040 $\mu\text{L/L}$ (表1)。

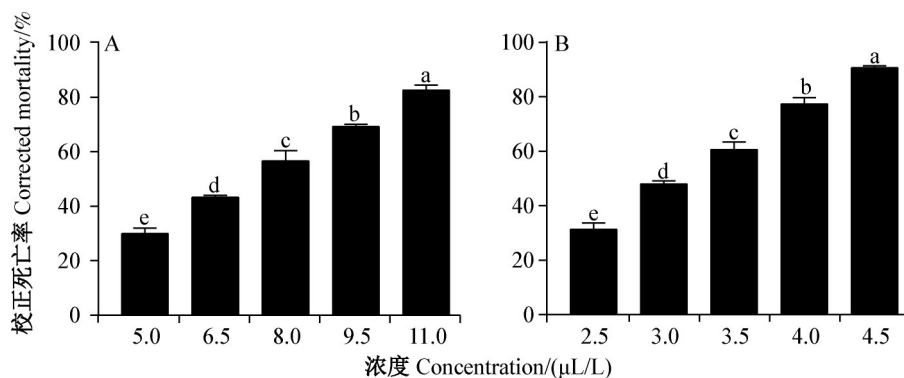


图2 茴香烯单独(A)及其与硅藻土联合处理(B)后绿豆象成虫的校正死亡率

Fig. 2 Corrected mortalities of adult *Callosobruchus chinensis* treated with *trans*-anethole (A) and in combination with diatomaceous earth (B)

图中数据为平均数±标准误。不同小写字母表示不同浓度之间经 Tukey 检验法检验差异显著 ($P < 0.05$)。Data are mean±SE. Different lowercase letters indicate significant differences among concentrations according to Tukey's test ($P < 0.05$).

表1 茴香烯单独及其与硅藻土联合处理对绿豆象成虫的毒力回归方程

Table 1 Toxicity regression equations of *trans*-anethole alone and in combination with diatomaceous earth against adult *Callosobruchus chinensis*

处理 Treatment	毒力回归方程 Regression equation	LC ₅₀ / (μL/L)	95% 置信区间 95% confidence interval/(μL/L)	相关系数 Relative coefficient	χ ²
茴香烯 <i>Trans</i> -anethole	$y=4.18x-3.53$	6.979	6.573-7.359	0.950	5.445
茴香烯+硅藻土 <i>Trans</i> -anethole+diatomaceous earth	$y=6.87x-3.31$	3.040	2.919-3.149	0.952	7.223

y: 死亡概率值; x: 浓度对数值。y: Death probability; x: logarithm of concentration.

2.3 硅藻土对茴香烯杀虫活性的增效作用

硅藻土对茴香烯的杀虫活性有显著协同增效作用。硅藻土与茴香烯联合处理后绿豆象的校正死亡率均显著高于硅藻土单独处理和茴香烯单独处理 ($P < 0.05$), 且处理 36 h 时绿豆象成虫的校正死亡率

达 100.00%, 分别为茴香烯单独处理和硅藻土单独处理的 2.54 倍和 21.41 倍。当绿豆象成虫校正死亡率超过 10.00% 时, 茴香烯单独处理需要 12 h, 硅藻土单独处理需要 48 h, 而茴香烯与硅藻土联合处理则仅需要 6 h (表 2)。

表2 茴香烯、硅藻土单独及其联合处理对绿豆象成虫的校正死亡率

Table 2 Corrected mortality of *Callosobruchus chinensis* adults exposed to *trans*-anethole, diatomaceous earth, and their combination

时间 Time/h	茴香烯 <i>Trans</i> -anethole	硅藻土 Diatomaceous earth	茴香烯+硅藻土 <i>Trans</i> -anethole+diatomaceous earth
6	0.67±1.15 b	0.00±0.00 b	15.33±2.31 a
12	5.33±1.15 b	0.00±0.00 c	70.67±1.15 a
24	23.33±4.16 b	2.67±1.15 c	92.67±1.15 a
36	39.33±2.31 b	4.67±1.15 c	100.00±0.00 a
48	55.33±1.15 b	12.67±2.31 c	100.00±0.00 a

表中数据为平均数±标准误。同行不同小写字母表示不同处理之间经 Tukey 检验法检验差异显著 ($P < 0.05$)。Data are mean±SE. Different lowercase letters within the same column indicate significant differences among treatments according to Tukey's test ($P < 0.05$).

2.4 茴香烯与硅藻土联合对绿豆象酶活性的影响

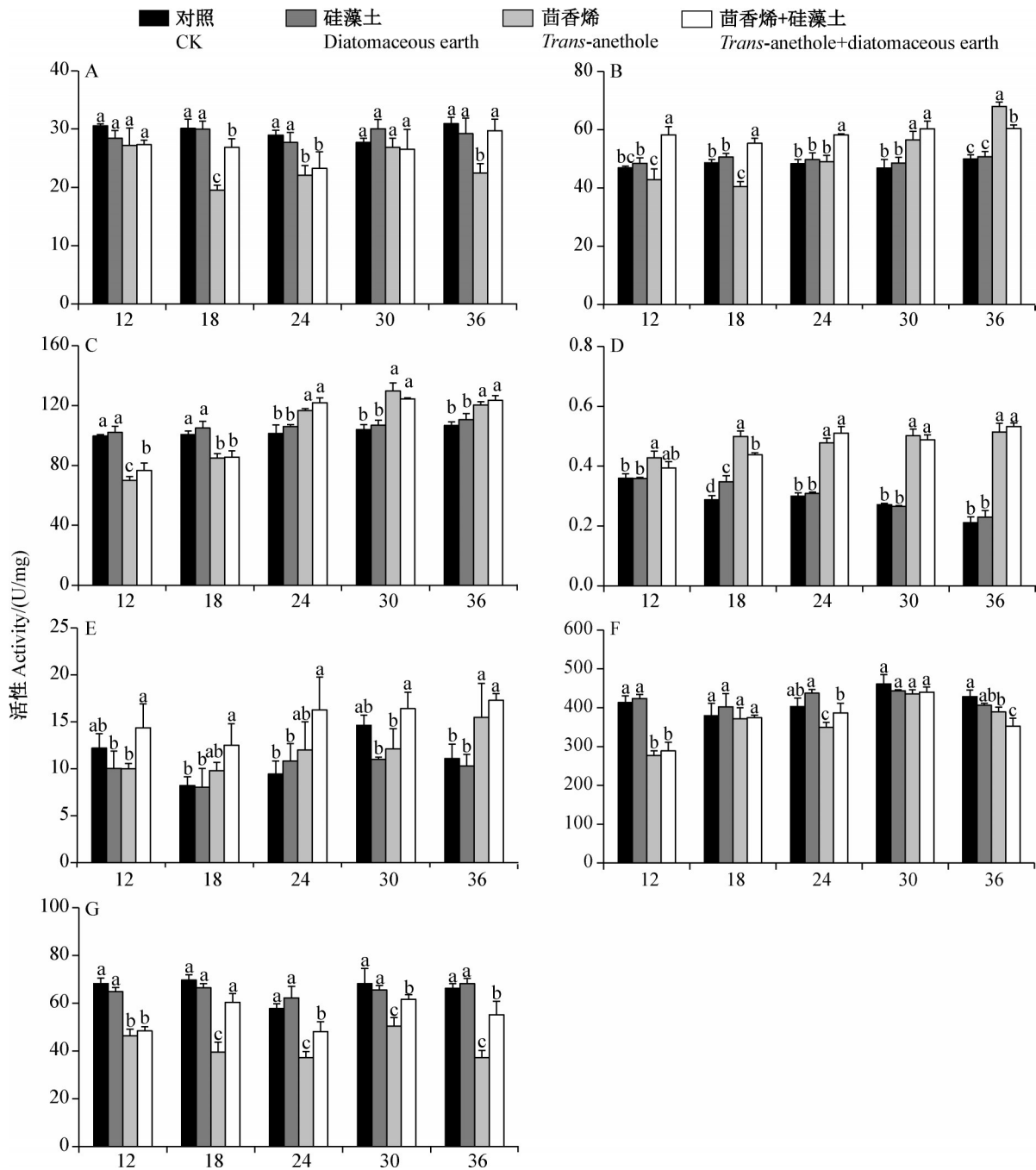
2.4.1 对绿豆象成虫体内 3 种保护酶活性的影响

茴香烯单独处理及其与硅藻土联合处理 18~24 h 时, 绿豆象成虫体内 CAT 活性均显著低于对照 ($P < 0.05$); 茴香烯单独处理 36 h 时绿豆象成虫体内 CAT

活性显著低于其他处理 ($P < 0.05$), 而茴香烯与硅藻土联合处理 36 h 时绿豆象成虫体内 CAT 活性与对照差异不显著 (图 3-A)。茴香烯与硅藻土联合处理后绿豆象成虫体内 POD 活性较对照均显著增加 ($P < 0.05$); 茴香烯单独处理 12~18 h 时绿豆象成虫体内

POD 活性均显著低于其他处理($P<0.05$),而后逐渐增加,处理 36 h 时 POD 活性达到最大值,为茴香烯与硅藻土联合处理的 1.13 倍(图 3-B)。茴香烯单独处理及其与硅藻土联合处理后绿豆象成虫体内 SOD 活性均呈上升趋势,其中处理 12~18 h 时绿豆

象成虫体内 SOD 活性均呈抑制状态,处理 24~36 h 时 SOD 活性均呈激活状态(图 3-C)。硅藻土单独处理后绿豆象成虫体内 3 种保护酶活性均与对照差异不显著(图 3-A~C)。



A~G: 过氧化氢酶、过氧化物酶、超氧化物歧化酶、谷胱甘肽-S-转移酶、羧酸酯酶、乙酰胆碱酯酶、琥珀酸脱氢酶。A~G: Catalase, peroxidase, superoxide dismutase, glutathione S-transferase, carboxylesterase, acetylcholinesterase, succinate dehydrogenase.

图3 茴香烯、硅藻土单独处理及其联合处理对绿豆象体内酶活性的影响

Fig. 3 Effects of *trans*-anethole, diatomaceous earth alone, and their combination on enzyme activities in *Callosobruchus chinensis* 数据为平均数±标准误。不同小写字母表示同时间不同处理之间经 Tukey 检验法检验差异显著 ($P < 0.05$)。Data are mean±SE. Different lowercase letters indicate significant differences among treatments at the same time according to Tukey's test ($P < 0.05$).

2.4.2 对绿豆象成虫体内3种解毒酶活性的影响

茴香烯单独处理及其与硅藻土联合处理后绿豆象成虫体内GST活性被显著激活,且随时间延长,活性越高,在处理36 h时绿豆象成虫体内GST活性分别达0.51 U/mg和0.53 U/mg,分别为对照的2.42倍和2.52倍(图3-D)。茴香烯与硅藻土联合处理后,绿豆象成虫体内CarE活性均表现为激活状态;而茴香烯单独处理12 h时,绿豆象成虫体内CarE活性仅为10.02 U/mg,为茴香烯与硅藻土联合处理的69.78%,随后逐渐增加,到处理36 h时显著高于对照($P<0.05$),为对照的1.35倍(图3-E)。茴香烯单独处理及其与硅藻土联合处理12、24和36 h时绿豆象成虫体内AchE活性均处于显著抑制状态($P<0.05$),尤其是处理12 h时AchE活性最低,分别为对照的66.99%和69.82%(图3-F)。硅藻土单独处理后绿豆象成虫体内3种解毒酶活性均与对照差异不显著(图3-D~F)。

2.4.3 对绿豆象成虫体内呼吸酶活性的影响

茴香烯单独处理及其与硅藻土联合处理12~36 h时绿豆象成虫体内SDH活性均处于显著抑制状态($P<0.05$),其中茴香烯单独处理的抑制程度最大(图3-G)。硅藻土单独处理后绿豆象成虫体内SDH活性均与对照差异不显著(图3-G)。

3 讨论

植物挥发物对害虫有较好的熏蒸作用,其通过呼吸作用进入害虫体内,导致其中毒甚至死亡(Hashem et al., 2018; Kovaříková et al., 2025)。目前已发现多种植物挥发物对绿豆象有较强的熏蒸活性(Mario et al., 2023)。例如,大蒜提取物和桉叶*Folium eucalypti*提取物对绿豆象成虫的 LC_{50} 分别为7.73 $\mu\text{L/L}$ 和26.88 $\mu\text{L/L}$ (胡婷婷等, 2019);26 $\mu\text{L/L}$ 罗勒*Ocimum basilicum*挥发物熏蒸处理后绿豆象成虫的死亡率达100%(Abd El-Salam, 2010)。本研究发现茴香烯对绿豆象表现出较强的熏蒸效果,5 $\mu\text{L/L}$ 茴香烯熏蒸48 h或10 $\mu\text{L/L}$ 茴香烯熏蒸36 h后绿豆象成虫的校正死亡率均可达100.00%。茴香烯对其他昆虫也有较强的熏蒸活性。例如,茴香烯对米象和赤拟谷盗的 LC_{50} 分别为76.98 $\mu\text{L/L}$ 和29.10 $\mu\text{L/L}$ (Johnson et al., 2022);20.0 mg/L茴香烯熏蒸72 h后玉米象*Sitophilus zeamais*的校正死亡率为90.26%(Li et al., 2013)。与米象、赤拟谷盗和玉米象等害虫相比,较低浓度茴香烯处理后绿豆象即表现出较高的致死率,推测可能与昆虫本身的生理结构、嗅觉

功能、解毒能力、靶标敏感性等有关。

硅藻土不仅可直接用于防治储粮害虫,而且还与其他药剂联合使用。例如,赤拟谷盗和谷象*Granary weevil*对黄连挥发物和硅藻土的耐受性均较高,而两者联合处理对赤拟谷盗和谷象有较高的杀虫效果(Ziaee et al., 2014);茴香*Foeniculum vulgare*精油与硅藻土联合处理对谷象的毒性较单独处理增加3倍左右(Pierattini et al., 2019);乳香黄连木*Pistacia lentiscus*和小茴香*Foeniculum vulgare*精油与硅藻土复配后对玉米象的杀虫效果较单独处理增强(Bougherra-Nehaoua et al., 2015)。本研究结果显示,茴香烯与硅藻土联合处理对绿豆象成虫的毒力较单独处理明显增加,表明茴香烯与硅藻土具有协同增效作用,在更短的时间对绿豆象有较高的致死率,究其原因可能是硅藻土颗粒通过物理摩擦破坏了害虫表皮酯层,导致其体内水分流失,从而引发害虫生理胁迫并显著降低其抗性(Gad et al., 2023)。

抗氧化保护酶广泛存在昆虫体内,其主要作用是清除体内自由基从而保护机体免受氧化损伤(Dryer et al., 1980),外源有毒物质侵入后昆虫会激活体内保护酶(Wang et al., 2020)。本研究结果显示,硅藻土单独处理后绿豆象成虫体内CAT、POD、SOD活性与对照均差异不显著;而茴香烯可抑制绿豆象成虫体内CAT活性;茴香烯可激活绿豆象成虫体内POD活性,而其与硅藻土联合处理后绿豆象成虫体内POD活性先被抑制而后被激活;茴香烯单独处理及其与硅藻土联合处理后绿豆象成虫体内SOD活性均呈现先被抑制而后被激活的变化趋势,表明茴香烯可激活绿豆象成虫体内3种保护酶的活性,且具有时间效应和浓度效应,即在逆境条件下绿豆象发生一系列防御反应,但当应激反应超过一定强度后,机体无法适应而出现不可逆的损伤(Spinozzi et al., 2023;王泽华等, 2024)。茴香烯熏蒸处理后桃小食心虫*Carposina sasakii*体内SOD和CAT活性呈现初期升高、后期下降的趋势,最后低于对照水平,而POD始终处于激活状态(李晓颖, 2012),这与本研究结果不一致,表明不同昆虫的抗氧化保护酶对同一外源有毒化合物响应规律不同。

昆虫体内解毒酶系主要参与昆虫对外源毒物的解毒代谢(Wang et al., 2014),其中AchE是许多外源有毒物质的主要作用靶标,是参与昆虫神经传导的一种特殊酶,能降解神经递质乙酰胆碱(李飞等, 2003)。本研究结果显示硅藻土对绿豆象成虫体内解毒酶未产生显著影响,而茴香烯是诱导绿豆象成

虫体内解毒酶活性变化的主要因素;茴香烯单独处理及其与硅藻土联合处理后绿豆象体内 GST 活性均显著升高,表明绿豆象成虫通过激活体内 GST 活性来应对茴香烯的胁迫;茴香烯单独处理后绿豆象成虫体内 CarE 活性显著高于其与硅藻土联合处理,表明低浓度茴香烯处理后绿豆象可迅速激活体内 CarE 从而产生解毒反应,但高浓度茴香烯处理可能对绿豆象造成应激损伤。类似地,低浓度茶皂素诱导东亚飞蝗体内 GST、CarE 活性增加,而高浓度茶皂素处理后其体内 GST 活性被抑制(王铭铭等, 2023); α -茄碱能诱导马铃薯块茎蛾 *Phthorimaea operculella* 幼虫体内 GST 活性增加(闫寒等, 2024); α -松油醇密闭熏蒸处理后,米象和赤拟谷盗体内 GST、CarE 活性均显著增加(刘雯雯等, 2025)。本研究结果显示,与 GST、CarE 活性表现不同,茴香烯单独处理及其与硅藻土联合处理后绿豆象成虫体内 AchE 活性均处于被显著抑制状态,表明 AchE 可能是茴香烯作用于绿豆象的关键靶标酶。类似地,茴香烯对地中海斑螟 *Ephestia kuehniella* 幼虫、锈赤扁谷盗 *Cryptolestes ferrugineus*、桑树美国白蛾 *Hyphantria cunea* 体内 AchE 活性均有显著的抑制作用(Shahriari et al., 2018; Wang et al., 2021; Pour et al., 2022)。推测 AchE 活性被显著抑制可能使试虫出现过度兴奋、多动等症状,从而增加了与硅藻土之间的摩擦,导致损伤程度加剧,但其具体作用机制有待进一步验证。SDH 是线粒体内膜上的关键酶,参与腺嘌呤核苷三磷酸合成,是细胞能量代谢核心环节之一(Haran et al., 2024)。本研究结果显示茴香烯处理后绿豆象成虫体内 SDH 活性均处于抑制状态,且其单独处理的抑制程度更强,这可能是造成绿豆象体内活性氧积累以及能量代谢失调的主要原因(Pekny et al., 2018),而硅藻土单独处理后绿豆象成虫体内 SDH 活性与对照无显著差异。本文仅从代谢水平分析了茴香烯对绿豆象体内相关酶活性变化规律的影响,其潜在的分子调控机制需进一步深入研究。

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