Smelling good enough to kill! Plant essential oils and their insecticidal activity

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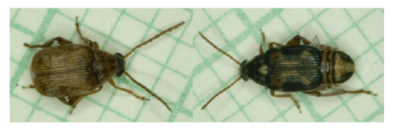
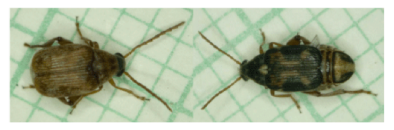




Why use this system?

- Bean beetles are easy to maintain in the laboratory
- There are concerns about chemical insecticides due to effects on non-target organisms and development of resistance
- "Natural" insecticides are locally available, biodegradable, and presumably less toxic to vertebrates (Rozman et al. 2007)
- Ties in several topics and lab skills: plant-insect interactions, evolution of chemical defenses, ethnobotany, agroecology, and ecological consequences of insecticide use

Materials

- Bean beetles, *Callosobruchus maculatus*
- Incubator with light and temperature control
- Plant essential oils
- Micropipettors and tips
- Lidded plastic containers
- Insect netting fabric, 2.5 cm filter circles
- Small paint brushes, paper clips

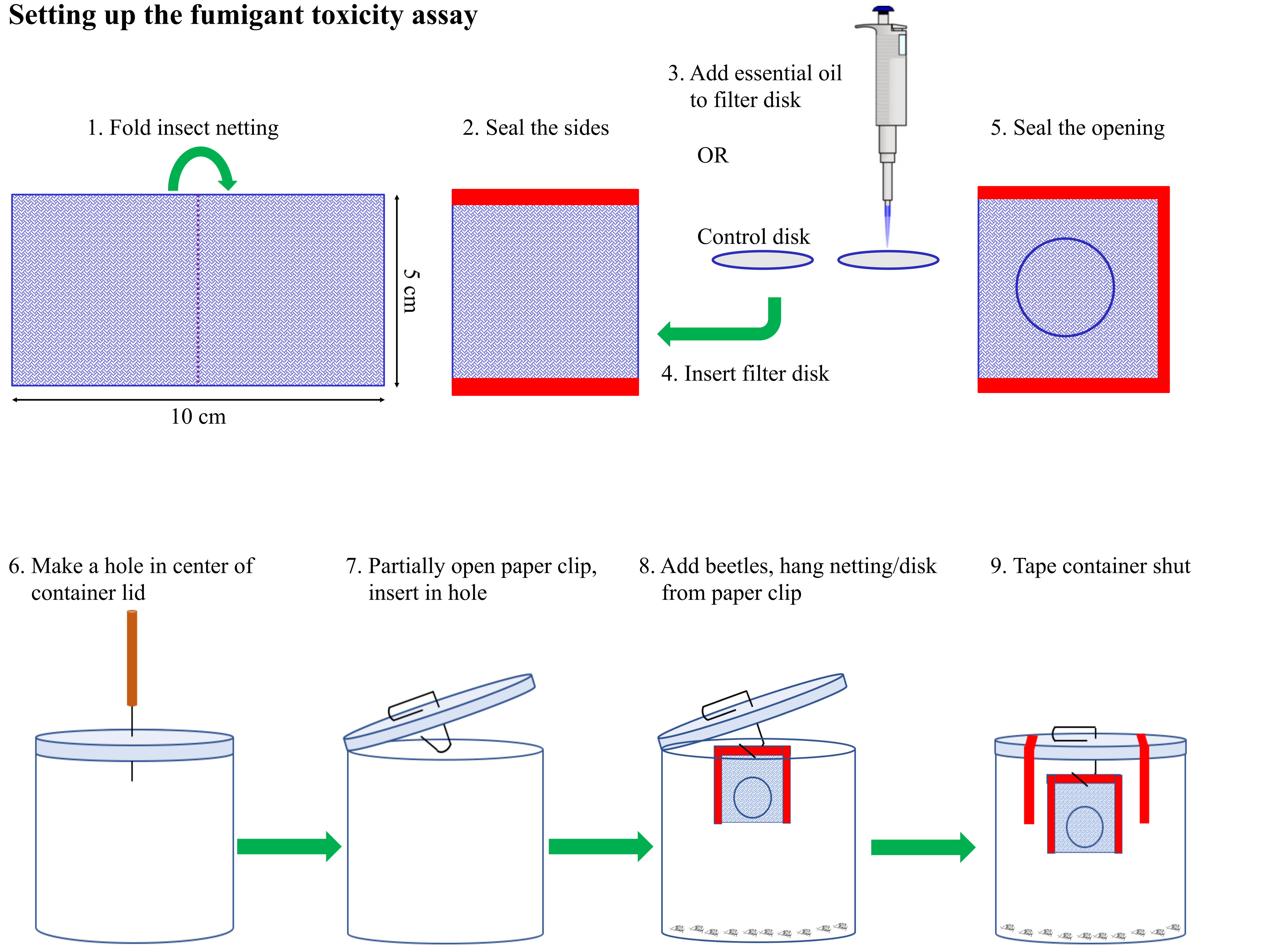
What the students do in the lab

- Learn to identify male and female bean beetles
- Carry out library research on plant essential oils and their potential insecticidal activity
- Set up the fumigant toxicity assay
- Count number of dead/live beetles daily
- Analyze their data and present their findings

What the students should be able to do at the end of the lab exercise

- Explain why bean beetles are a good system for studying the effectiveness of plant essential oils as insecticides
- Design a study to investigate whether plant essential oils can be used as potential insecticides
- Collect, analyze, and present the data from such studies
- Discuss the limitations of the study and suggest future steps based on the results obtained

Setting up the fumigant toxicity assay



Sample results

Table 1: Mortality of bean beetles exposed to essential oils from different plant species. Data are means of 3 replications, each with 10 beetles (5 males, 5 females) per treatment. Mortality corrected per Abbott (1925). Amount of essential oil used was 150 μL. Bean beetle strain used was LB grown on mung beans.

	Essential Oil Used								
Day	Clove	Ginger	Thyme	Lemon	Lime	Orange	Cedar Leaf	Tea Tree	Winter-green
0	0	0	0	0	0	0	0	0	0
1	17.0	20.4	81.9	100	100	100	100	100	100
2	52.3	45.2	100	100	100	100	100	100	100
3	88.9	65.0	100	100	100	100	100	100	100
4	92.6	84.1	100	100	100	100	100	100	100
5	100	100	100	100	100	100	100	100	100

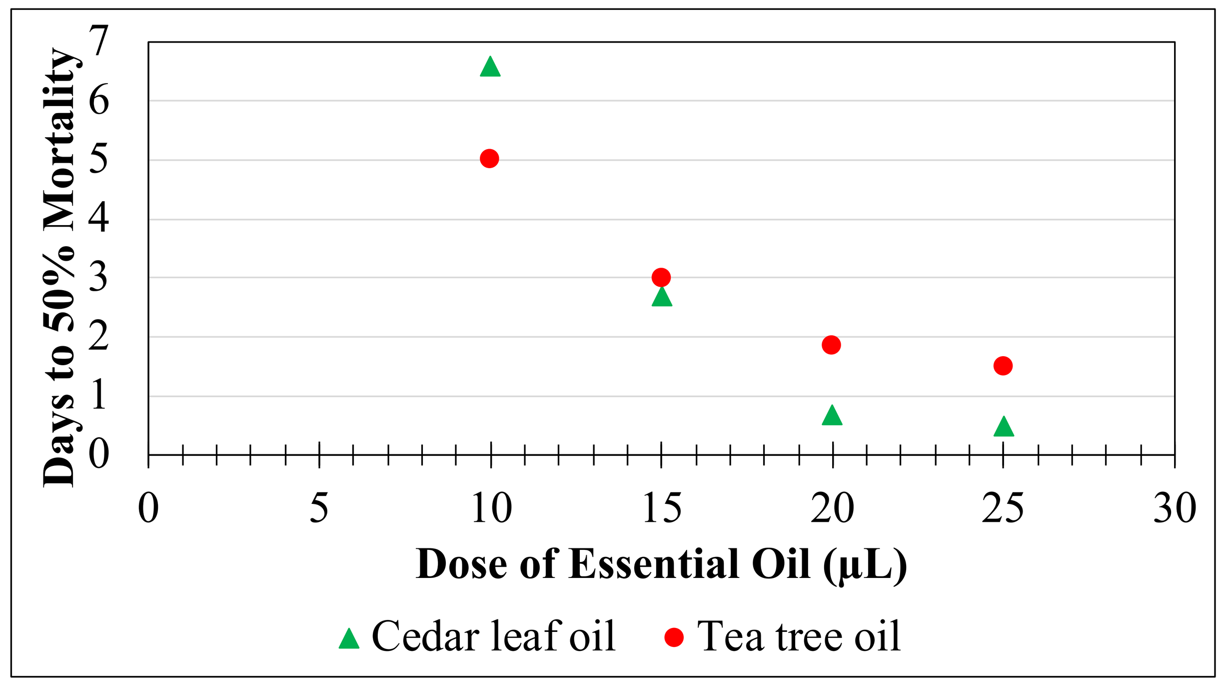


Figure 1: Days to 50% mortality, calculated using regression analysis, for bean beetles exposed to different doses of essential oils from two plant species. Mortality corrected per Abbott (1925). Each treatment included 20 beetles. Beetle strain used was Burkina Faso grown on mung beans.

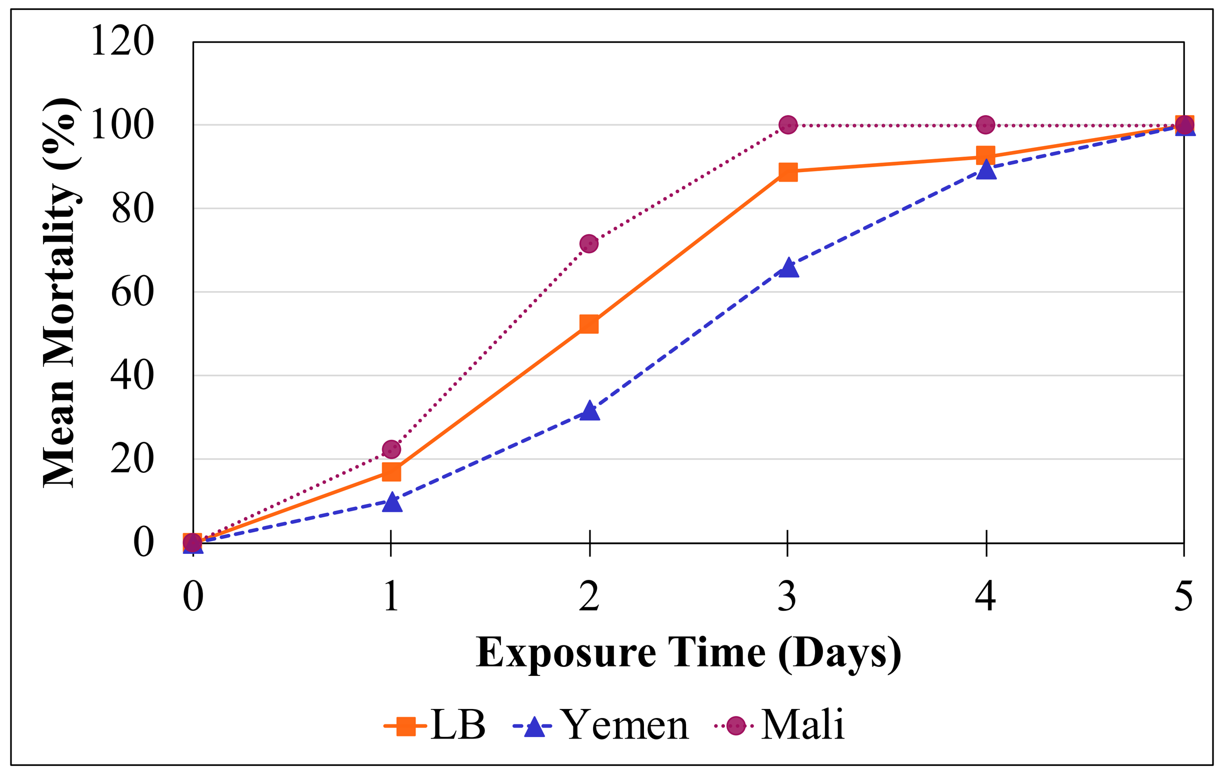


Figure 2: Mortality of different geographic strains of bean beetles exposed to clove oil. Mortality corrected per Abbott (1925). The data are means of 3 replications. Each treatment included 10 beetles (5 males, 5 females). Amount of essential oil used was 150 μL.

Comments

- Correction of mortality data: effectiveness of an insecticide per Abbott (1925):

$$\frac{100 \times (\%\text{ living in control} - \%\text{ living in treatment})}{\%\text{ living in control}}$$

- Set up is quick, requires daily check-up
- Readings from the primary literature such as:
  - Rozman et al. (2007): Toxicity of naturally occurring compounds of Lamiaceae and Lauraceae
  - Yazdgerdian et al. (2015): Insecticidal effects of essential oils against different insects

Current work

- To determine whether beetle strains grown on different legume seeds show different sensitivity to specific essential oils
- To compare essential oils from species in the same family or from different families
- To determine whether insecticidal activity of the oils is due to inhibition of specific enzymes

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