Creating artificial beans for bean beetles, *Callosobruchus maculatus*, using a mechanical pill press

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ABSTRACT

The bean beetle, *Callosobruchus maculatus* (Coleoptera, Chrysomelidae), has become a widely used insect species in undergraduate laboratory education. This species is particularly suitable for course-based undergraduate research experiences (CUREs) due to its short generation time, ease of handling and culturing in the laboratory, and sexual dimorphism in its sedentary phase. Bean beetles complete their growth and development inside a host seed (bean) with at least eight different host species. However, conducting manipulative experiments with bean beetles would be enhanced if it were possible to readily prepare artificial beans on which the beetles could complete their life cycle. Here, we report on the use of a mechanical pill press (LFA Machines Model TDP-0) to make artificial beans. We prepared artificial beans by making whole blackeye pea flour (*Vigna unguiculata*) using a coffee grinder. That flour was used in the pill press to make 8mm diameter x 5-9mm thick disk-shaped pills with and without additives. Adult female bean beetles readily laid fertilized eggs on the surface of these artificial beans. Offspring emerged 4-5 weeks later at 25°C, the same development time that would have occurred in natural intact blackeye pea seeds. No special treatments of the artificial beans were required to induce females to lay eggs on them nor for the pills to remain intact during the period of larval and pupal development. This mechanical pill press can produce 30-50 pills per minute, so artificial beans can be produced rapidly in sufficient numbers to conduct meaningful experiments. This simple and effective method for making artificial beans creates the opportunity to conduct studies that have been difficult or impossible in the past. For example, future studies may evaluate treatments such as plant secondary compound concentrations, nutrient content, and antibiotic exposure on bean beetle life history and microbiome communities.

QUESTION

Can artificial beans (pills) be used to culture bean beetles, *Callosobruchus maculatus*?

HYPOTHESIS

If we grow bean beetles on artificial pills instead of their natural host bean, we will see the beetles complete their life cycle as they would on natural whole dried beans.

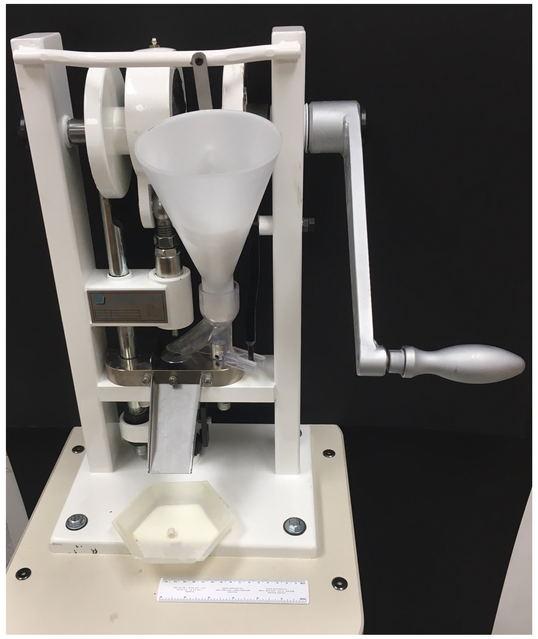

Figure 1. LFA Machines TDP-0 pill press

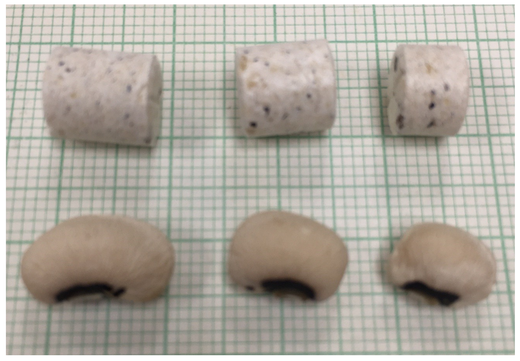

Figure 2. Variations in pill size and blackeye peas (mm squares)

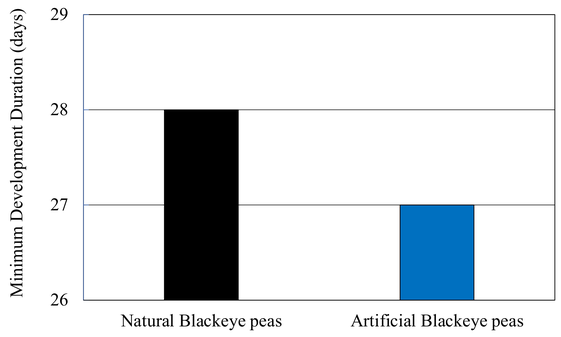

Figure 3. Bean beetle development on natural blackeye peas and artificial bean pills at 25°C and high RH.

METHODS

An LFA Machines Model TDP-0 pill press ($1000 USD, Figure 1) was used to make pill shaped artificial beans. Artificial bean were made by preparing a blackeye pea flour (*Vigna unguiculata*) from whole dried beans to make 8mm diameter pills with thicknesses from 5-9mm, similar to the size of narual blackeye peas (Figure 2). Mature male and female bean beetles (*Callosobruchus maculatus*) were introduced to artificial beans (N=3 replicates of 10 pills each in 100mm Petri dishes) and natural blackeye peas in stock cultures (N=2 replicates) in 150mm Petri dishes, incubated at 25°C and high RH.

RESULTS

Development time was very similar on artificial blackeye peas (pills) compared to natural blackeye peas. Oviposition was first observed on the pills 14-days after the introduction of beetles. Development time was measured from the first day eggs were observed to the first day newly emerged adults were seen. Twenty-seven days after oviposition was observed on pills, adult beetles emerged. Development time on natural blackeye peas is typically 28-days at 25°C (Figure 3).

CONCLUSIONS

Unlike alternative methods for making artificial beans, using a pill press eliminates the need for gelatin capsules or coatings, wet slurries, freeze drying, or casting molds to make reproducible and biologically success bean substitutes. The ability to make artificial beans creates opportunities to conduct a variety of experiments such as manipulations of plant secondary compounds, nutrient content, herbicides, fungicides or antibiotics. We are starting experiments in which partial ablation of the gut bacterial microbiome will be induced by raising beetles on artificial beans containing broad spectrum antibiotics. The effects of partial ablation of the microbiome on development time, adult behavior and reproduction will be evaluated .

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