



University of Guam Coconut Rhinoceros Beetle Biological Control Project

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<https://github.com/aubreymoore/rearing3>

Bioassay Report: DUG42

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<https://github.com/aubreymoore/rearing3/raw/master/bioassay-DUG42.pdf>

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1 Summary

2 Description

Adult beetles incubated at 30°C and 80% RH for more than 2 weeks to observe possible contamination from green muscardine fungus infection were employed in a bioassay to determine the susceptibility of adults to infection by a virus isolate collected from the Philippines (**Dug42**). Treatment 1 consisted of 10-20 µl sterile filtered water injected at a point on the ventral surface at the junction of the hind leg and the thoracic using a sterile needle. Treatment 2 consisted of 10-20 µl heat-inactivated virus injection while in the treatment 3, beetles were injected with 10-20 µl of untreated virus preparation. Adults were then placed in clean glass mason jars (bleach-treated) with a piece of banana added for food. Beetles were incubated at 30°C and 80% RH in a Percival incubator. All beetles were weighted every other day but monitored daily for four weeks to observe any possible signs of mortality.

2.1 Replicate 1

A total of 10 adult females and five adult males distributed among the three treatments were employed in this replicate.

2.2 Replicate 2

A total of seven adult females and eight adult males distributed among the three treatments were employed in this replicate.

3 Mortality

Table 1: Mortality summary.

| | bioassay_treatment | ntotal | ndead | mortality | adjusted_mortality | significance |
|---|--------------------|--------|-------|-----------|--------------------|--------------|
| 0 | control | 10 | 5 | 0.5 | 0.0 | 1.000000 |
| 1 | heat inactivated | 10 | 7 | 0.7 | 0.4 | 0.649917 |
| 2 | virus | 10 | 7 | 0.7 | 0.4 | 0.649917 |

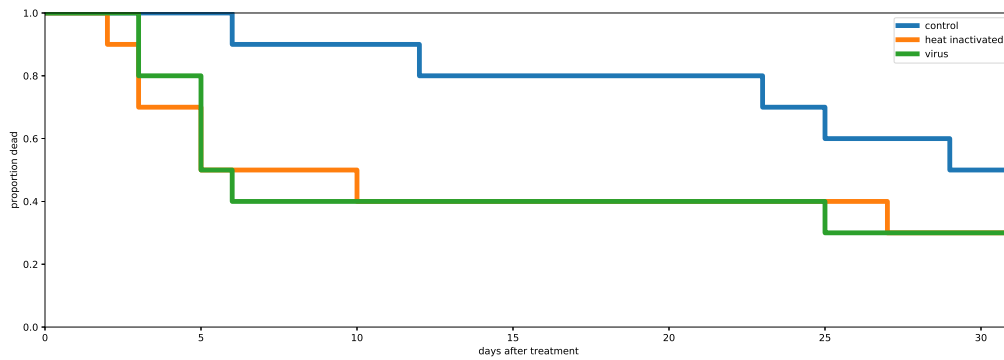


Table 2: Pairwise differences among mortality curves.

| | | test_statistic | p |
|------------------|------------------|----------------|----------|
| control | heat inactivated | 1.920677 | 0.165782 |
| | virus | 2.020872 | 0.155150 |
| heat inactivated | virus | 0.007094 | 0.932879 |

4 Change in Mass

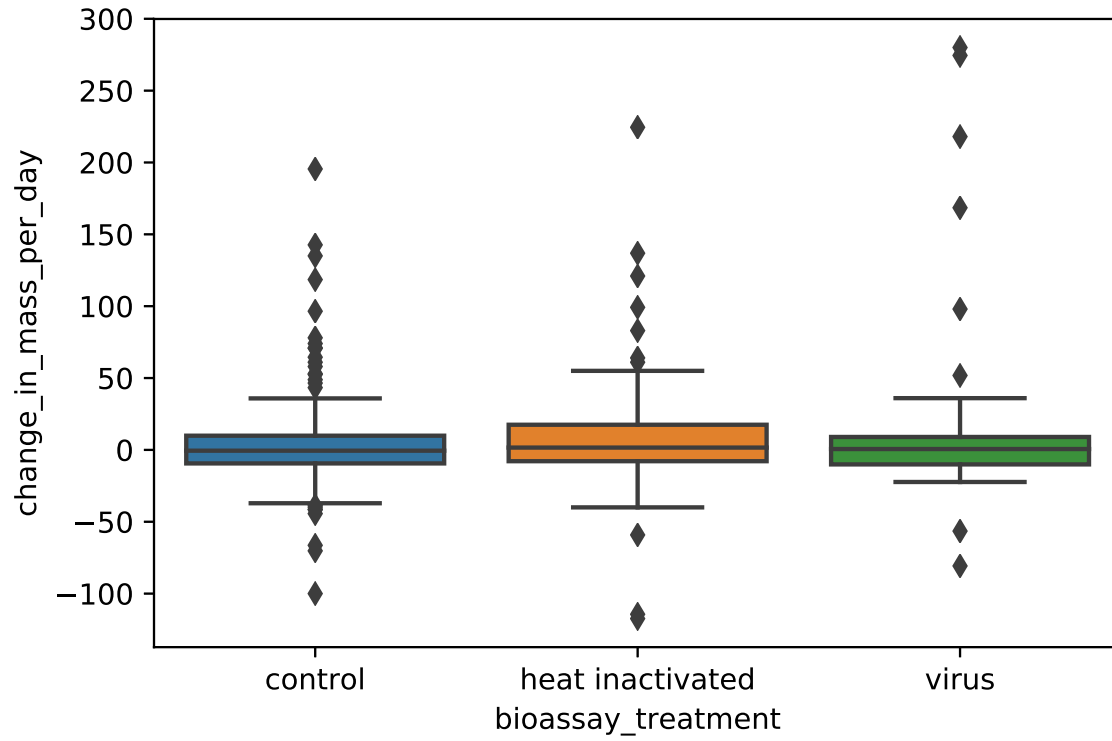


Table 3: Results of pairwise significance tests for differences in change in mass.

| | control | heat inactivated | virus |
|------------------|-----------|------------------|-------|
| control | -1.000000 | 0.901325 | 1.0 |
| heat inactivated | 0.901325 | -1.000000 | 1.0 |
| virus | 1.000000 | 1.000000 | -1.0 |

5 Post Mortem Images

5.1 control



Figure 1: Bioassay: DUG42-1; Treatment: control; Beetle ID: 46



Figure 2: Bioassay: DUG42-1; Treatment: control; Beetle ID: 47



Figure 3: Bioassay: DUG42-1; Treatment: control; Beetle ID: 48



Figure 4: Bioassay: DUG42-1; Treatment: control; Beetle ID: 49



Figure 5: Bioassay: DUG42-1; Treatment: control; Beetle ID: 50



Figure 6: Bioassay: DUG42-2; Treatment: control; Beetle ID: 61



Figure 7: Bioassay: DUG42-2; Treatment: control; Beetle ID: 62

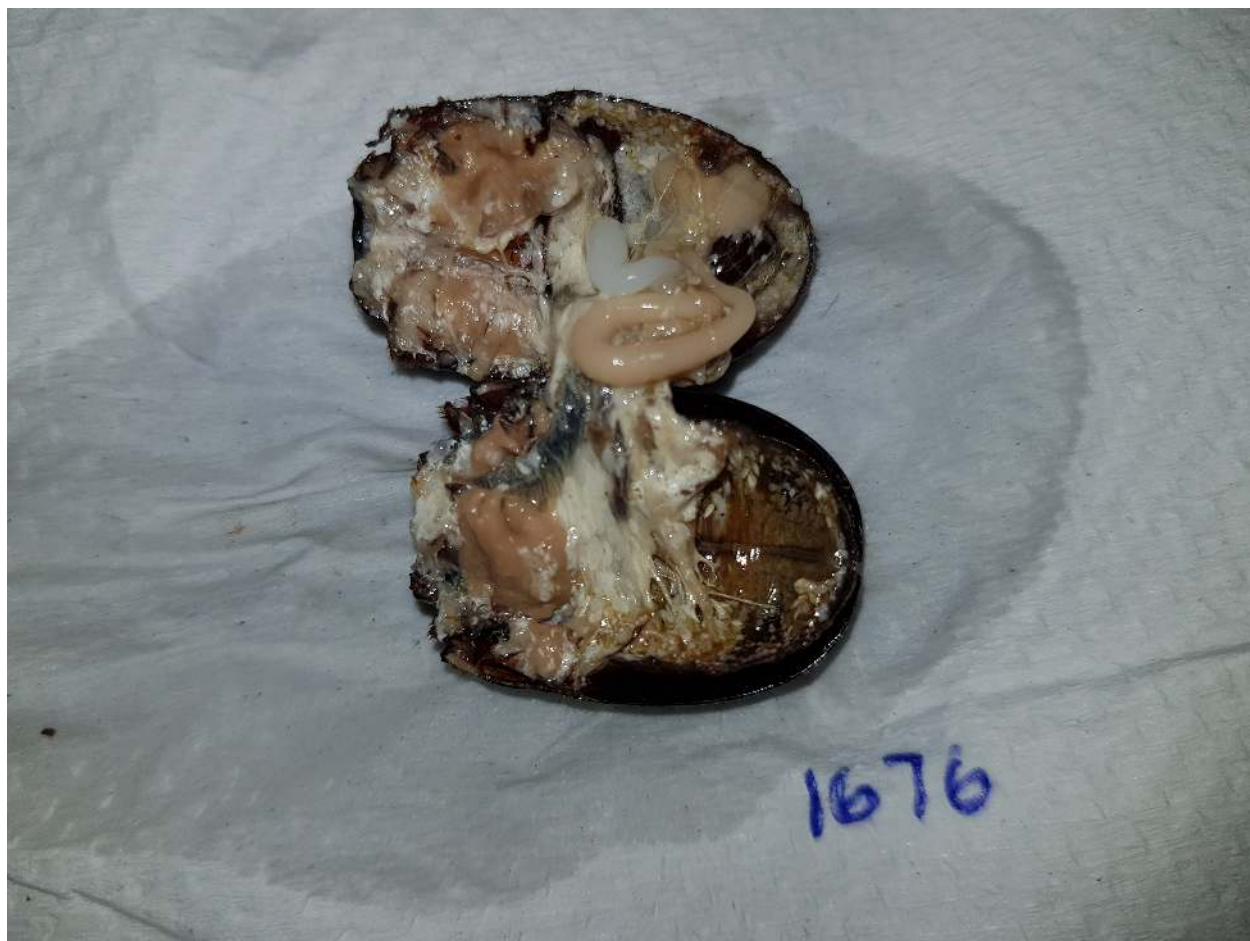


Figure 8: Bioassay: DUG42-2; Treatment: control; Beetle ID: 63



Figure 9: Bioassay: DUG42-2; Treatment: control; Beetle ID: 64



Figure 10: Bioassay: DUG42-2; Treatment: control; Beetle ID: 65

5.2 heat inactivated



Figure 11: Bioassay: DUG42-1; Treatment: heat inactivated; Beetle ID: 51



Figure 12: Bioassay: DUG42-1; Treatment: heat inactivated; Beetle ID: 52



Figure 13: Bioassay: DUG42-1; Treatment: heat inactivated; Beetle ID: 53



Figure 14: Bioassay: DUG42-1; Treatment: heat inactivated; Beetle ID: 54



Figure 15: Bioassay: DUG42-1; Treatment: heat inactivated; Beetle ID: 55



Figure 16: Bioassay: DUG42-2; Treatment: heat inactivated; Beetle ID: 66



Figure 17: Bioassay: DUG42-2; Treatment: heat inactivated; Beetle ID: 67



Figure 18: Bioassay: DUG42-2; Treatment: heat inactivated; Beetle ID: 68



Figure 19: Bioassay: DUG42-2; Treatment: heat inactivated; Beetle ID: 69



Figure 20: Bioassay: DUG42-2; Treatment: heat inactivated; Beetle ID: 70

5.3 virus



Figure 21: Bioassay: DUG42-1; Treatment: virus; Beetle ID: 56



Figure 22: Bioassay: DUG42-1; Treatment: virus; Beetle ID: 57



Figure 23: Bioassay: DUG42-1; Treatment: virus; Beetle ID: 58



Figure 24: Bioassay: DUG42-1; Treatment: virus; Beetle ID: 59



Figure 25: Bioassay: DUG42-1; Treatment: virus; Beetle ID: 60



Figure 26: Bioassay: DUG42-2; Treatment: virus; Beetle ID: 71



Figure 27: Bioassay: DUG42-2; Treatment: virus; Beetle ID: 72



Figure 28: Bioassay: DUG42-2; Treatment: virus; Beetle ID: 73



Figure 29: Bioassay: DUG42-2; Treatment: virus; Beetle ID: 74



Figure 30: Bioassay: DUG42-2; Treatment: virus; Beetle ID: 75