

Altered Feeding Behavior and Viral Detection in Honey Bees Exposed to Organosilicone

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Introduction

Honey bees play an important role in agriculture and the decline of honey bee populations worldwide has generated concern. While the application of pesticides in agricultural settings is often implicated in the deterioration of honey bee population health, pesticide applications contain more than just pesticides; they also contain adjuvants that may have detrimental effects to bee health. One known effect of these adjuvants is the increase of viral loads in honey bee larvae (Fine et al. 2017). We are investigating the effects of one class of inert pesticide adjuvant, organosilicone surfactant (OSS), on adult honey bee health.



Fig. 1 Honey bee hives used in an agricultural setting

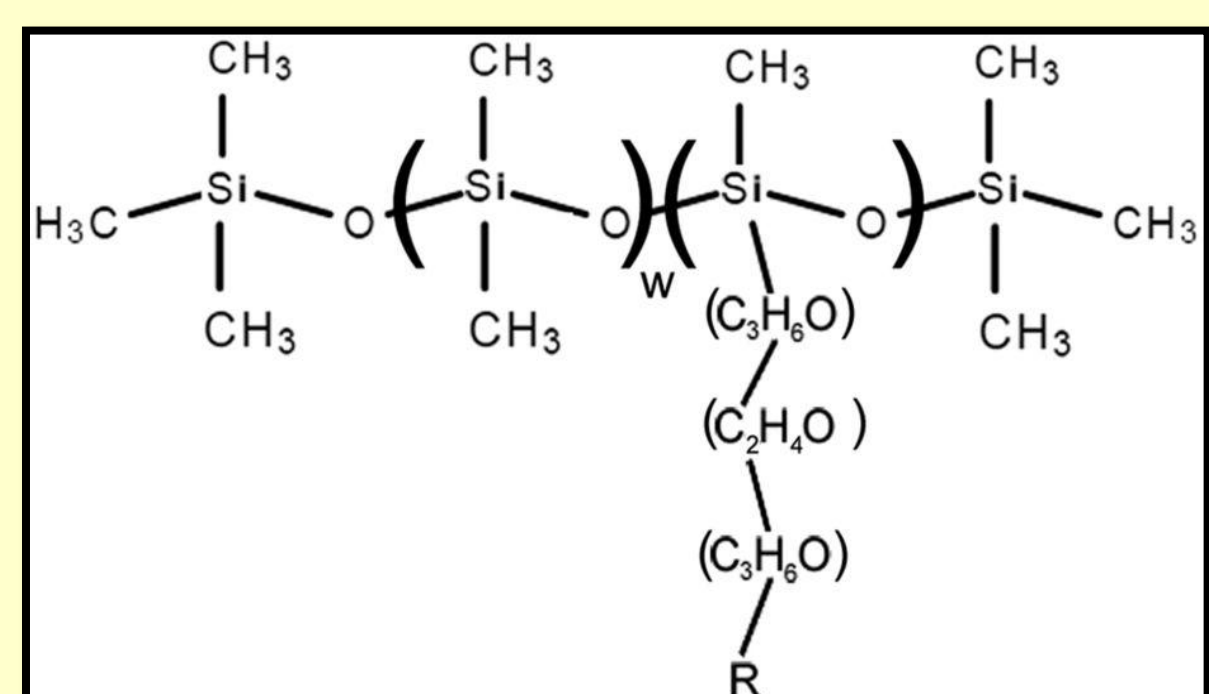


Fig. 2 General organosilicone surfactant structure

Viruses are thought to be a major culprit in the decline of honey bee populations. Consequently, viral presence and activity are important parameters to consider when evaluating the effects of OSS exposure in honey bees. Viral presence can be readily determined by PCR and gel electrophoresis, but determining viral activity requires knowing the quantity of replicating strands present—this knowledge is obtained via stranded RNA sequencing.



Fig. 3 A selection of common honey bee virus symptoms. Top to bottom: Deformed Wing Virus, Black Queen Cell Virus, Sacbrood Virus

Goals

- Determine if OSS exposure alters honey bee feeding behavior
- Determine if OSS synergizes the activity of viral honey bee pathogens via stranded RNA sequencing

How does honey bee feeding behavior change in response to OSS exposure?

Methods

- Bioassay conducted with locally collected adult honey bees
- 36 cohorts of 10 bees were given one of 6 treatments (6 reps/treatment)
- Bioassay ran for 7 days
- Feeding and mortality data collected

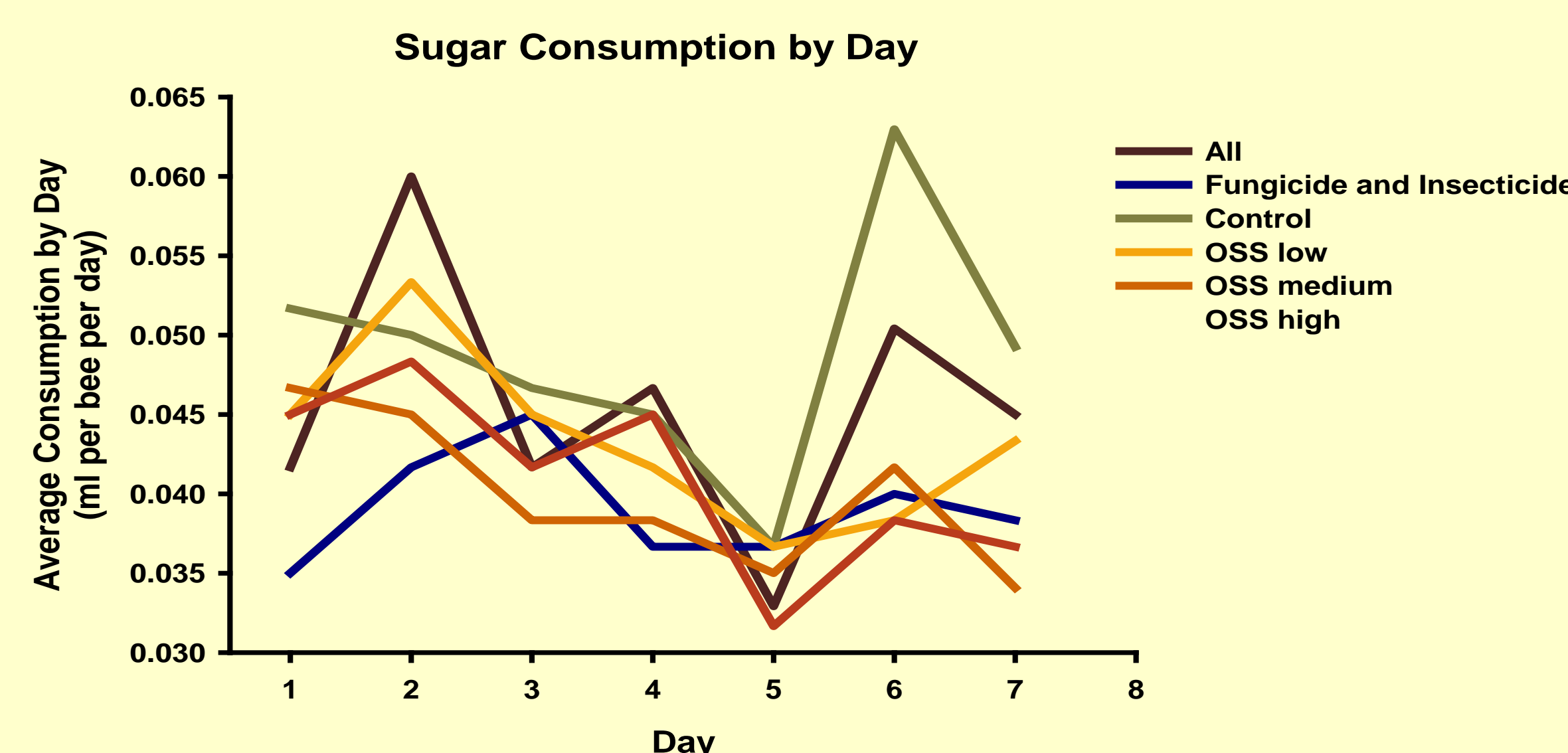
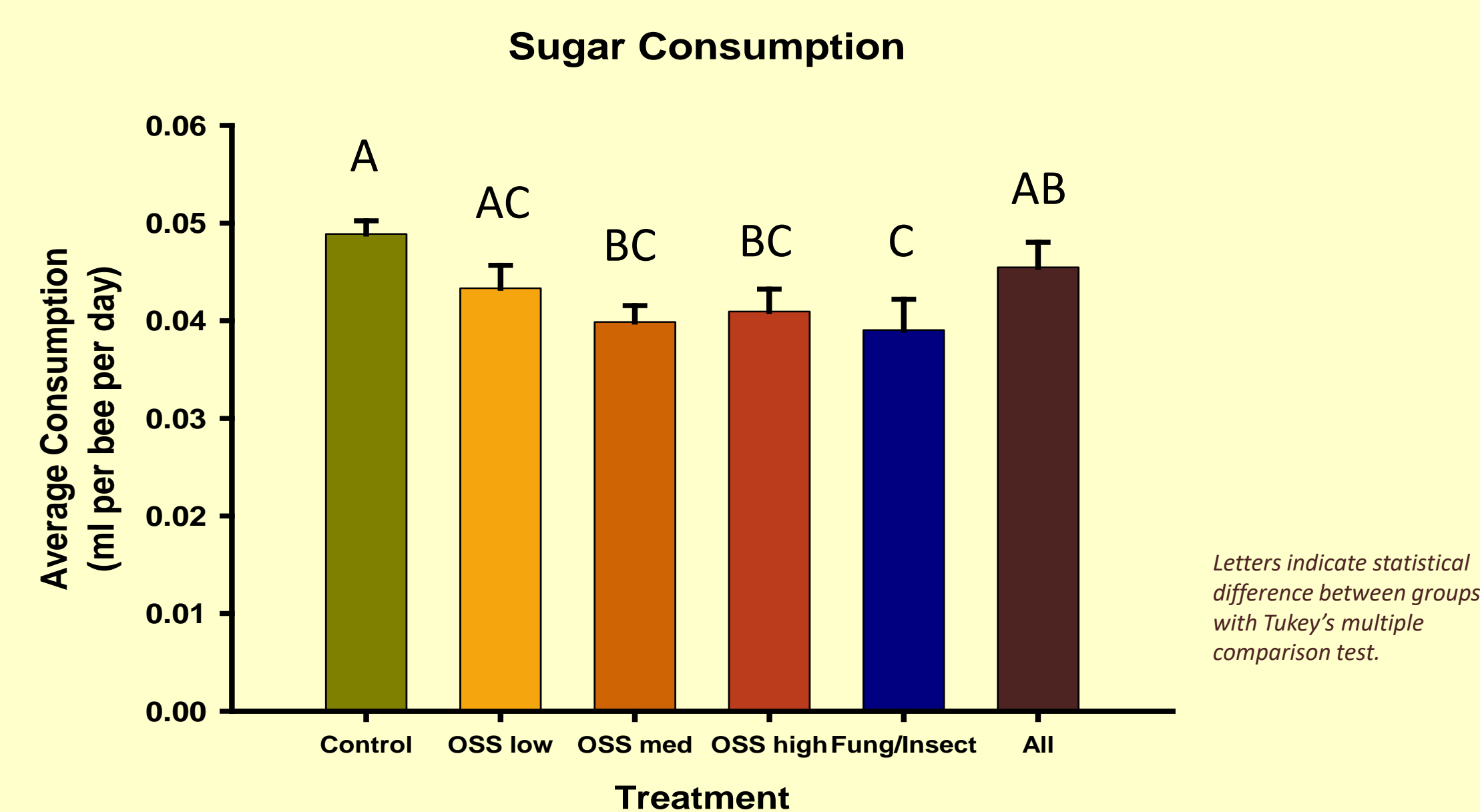
Treatment	Ingredients
Control	N/A
OSS Low	OSS (40ppb)
OSS Medium	OSS (1ppm)
OSS High	OSS (10ppm)
Fungicide & Pesticide	Tilt (150 ppb), Altacor (3ppm)
All (Fungicide, Pesticide, & OSS Medium)	Tilt (150ppb), Altacor (3ppm), & OSS (1ppm)



Fig. 4 Bioassay was conducted with adult honey bees over a period of seven days. Treatments were delivered via sugar water solution in a syringe.

Results

- Using a generalized linear model (SAS 9.4: PROC GLM), we found a significant effect of treatment ($F=6.56, p<0.001$) and day ($F=8.04, p<0.001$) on sugar consumption per bee. The interaction of treatment and day was not significant ($F=1.46, p=0.0656$).
- OSS medium, OSS high, and fungicide/insecticide yielded significantly less consumption than control.

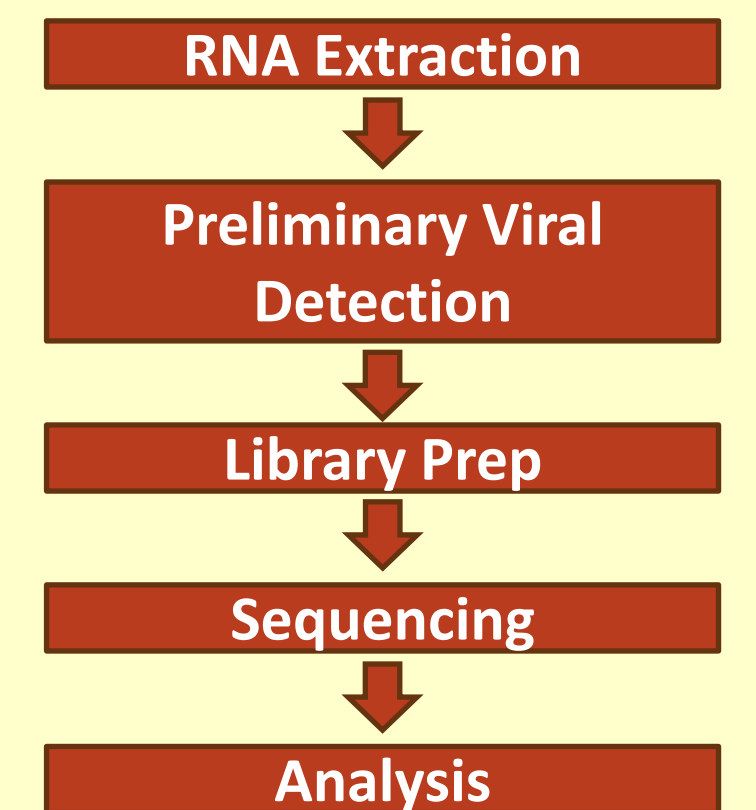


Does OSS exposure increase viral diversity and activity in honey bees?

As a continuation of this multi-semester project, we will sequence viral RNA to determine the variety of viruses present in our bioassay bees as well as the degree of their replication.

Methods

- RNA extracted from bioassay bees
- cDNA created from a sample of RNA and PCR conducted to preliminarily screen for viruses



Results

- We tested for the following viruses in our samples (those in bold were detected in all of our samples):

Deformed Wing Virus
Black Queen Cell Virus
Israeli Acute Paralysis Virus
Sacbrood Virus
Kashmir Bee Virus
Chronic Bee Paralysis Virus
Lake Sinai Virus

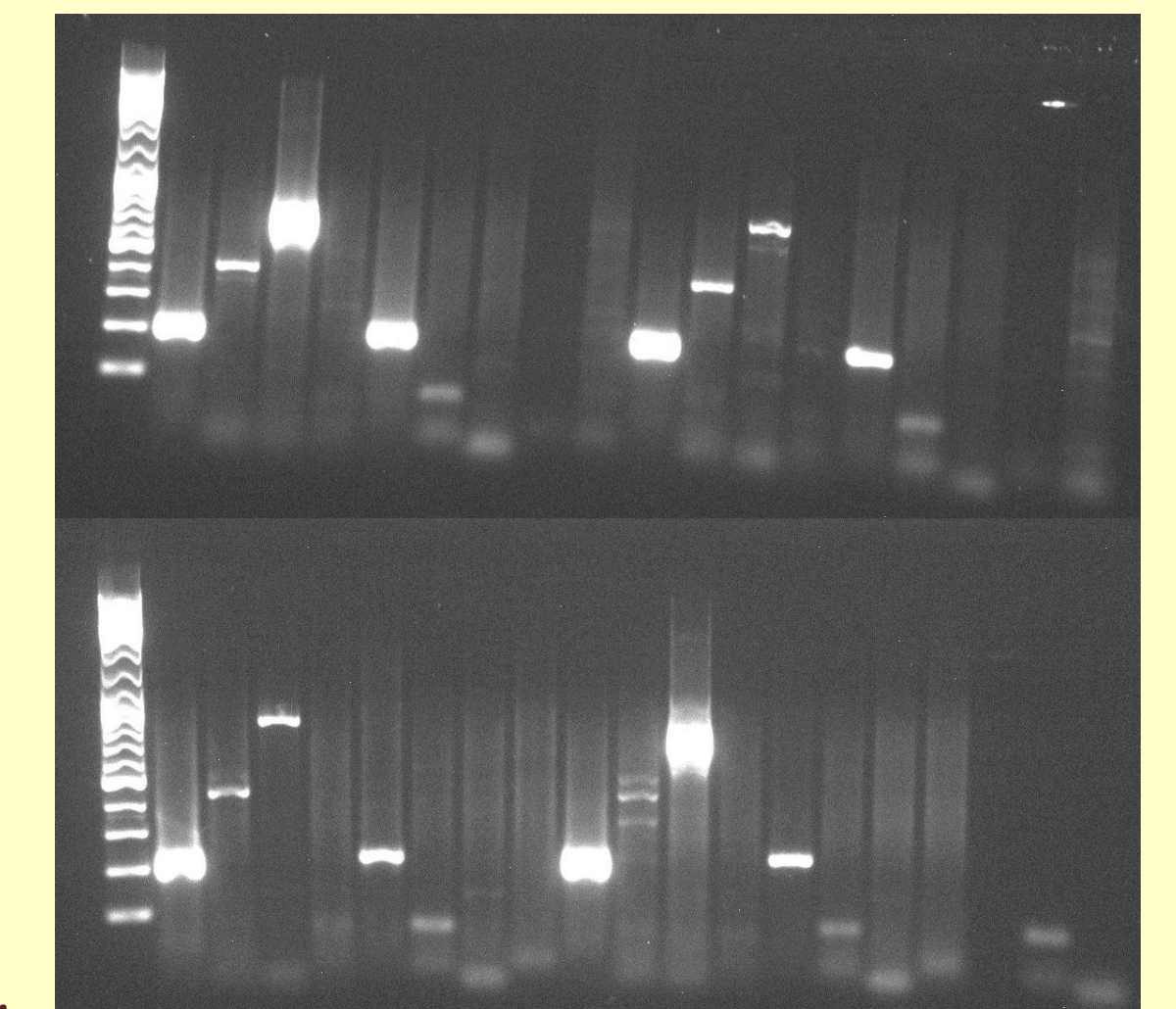


Fig. 5 The similar banding pattern of treatments indicate similar viral presence.

Future work

- Create cDNA with tag primers (tags will indicate strand information) and use targeted PCR to enhance viral signal
- Produce library with Illumina's Nextera DNA library prep kit and sequence.
- Analyze sequence data for virus presence and degree of viral replication (by detecting negative strand synthesis)

Conclusions

- There is evidence that OSS exposure decreases feeding in honey bees. However, the trend is not entirely consistent and further work is needed to elucidate the combined effects of pesticides and OSS.
- Our preliminary virus detection yielded no observed differences in viral presence between treatments. Our future work will examine the potentially large differences in viral activity between treatments.
- Future results will be informative about the impacts of OSS on honey bee health and the potential need to regulate their usage.

References

Fine, J. D. et al. An Inert Pesticide Adjuvant Synergizes Viral Pathogenicity and Mortality in Honey Bee Larvae. *Sci. Rep.* 7, 40499; 2017.