Non-lethal DNA extraction methods for genetic analyses of endangered bumble bee species

Authors: Eiley Kuhlmey, Michelle Boone, Amelia Lindsey, and Sujaya Rao

Question: Does a passive or direct DNA collection method work more effectively for gathering DNA from bumble bees while minimizing harm inflicted on threatened and endangered species?

Introduction

- **Background**
  - Small or endangered populations are more susceptible to low rates of genetic diversity due to an absence of gene flow[1]
  - Low genetic diversity is harmful since organisms within this population are less apt to withstand environmental changes[1]
  - Native bumble bees already experience a number of factors threatening their existence, such as pollution, urbanization, climate change, and pesticide usage[2,3,4], often leading to rapid rates of population decline
- **Significance**
  - Pollinators are responsible for pollinating 35 percent of crops consumed globally[4,5]
  - Up to 90 percent of wild flowering plants rely on insect driven pollination[5]
  - One in four species of North American bumble bees is faced with population decline[4,6]
  - Many commercial plant species require buzz pollination, necessitating bumble bees as pollinators[7]

Methods

- **Objective**
  - Collect 60 *Bombus impatiens* samples
  - 30 samples receive passive DNA collection technique
  - 30 samples receive active DNA collection technique
- **Passive DNA Collection Technique**
  - Collect *Bombus impatiens* sample in 50mL conical tube fitted with a sugar-soaked cotton swab (Fig. 1)
  - Bee remains in tube for two hours in room temperature environment
  - Swab is collected, and DNA is amplified using PCR and analyzed using gel electrophoresis
- **Active DNA Collection Technique**
  - Collect *Bombus impatiens* sample in sweep net and transfer into a 50mL conical tube
  - Place 50mL conical tube on ice for 10-15 minutes
  - Use a sterile swab and aseptic technique to swab the bee on the thorax and abdomen (Fig. 2)
  - Swab is collected, and DNA is amplified using PCR and analyzed using gel electrophoresis

Figures

Figure 1. Passive swabbing method. *B. impatiens* sample was collected in a 50mL conical tube for two hours, allowing DNA to gather on the swab through interactions with the specimen.

Figure 2. Active swabbing method. *B. impatiens* sample was collected, immobilized using ice, and gently swabbed along the thorax and abdomen.

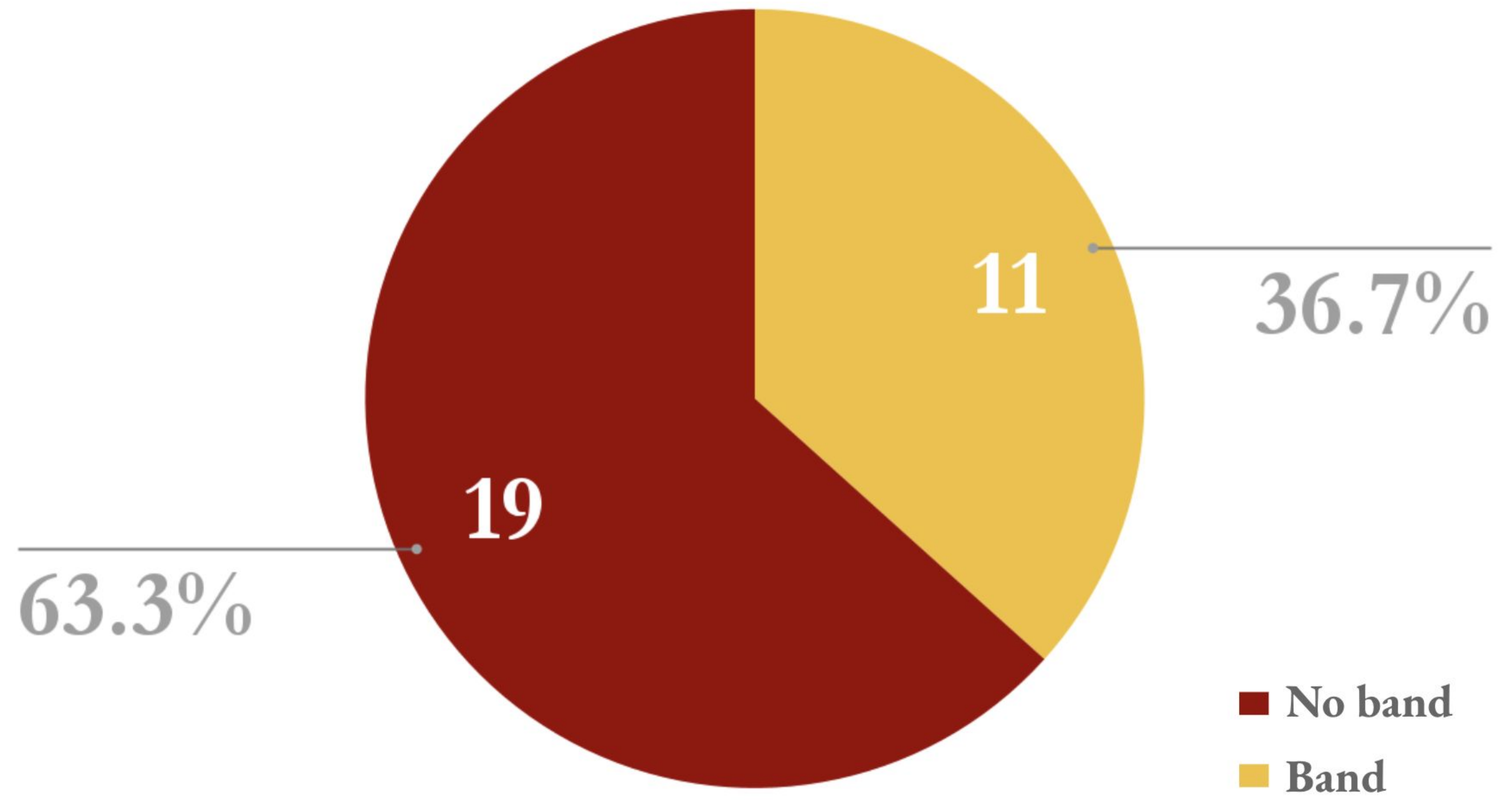

Figure 3. Presence of DNA in Passive Method Specimens. The DNA samples were amplified using PCR and underwent gel electrophoresis to determine the presence of DNA as indicated by the appearance of a band on the gel.

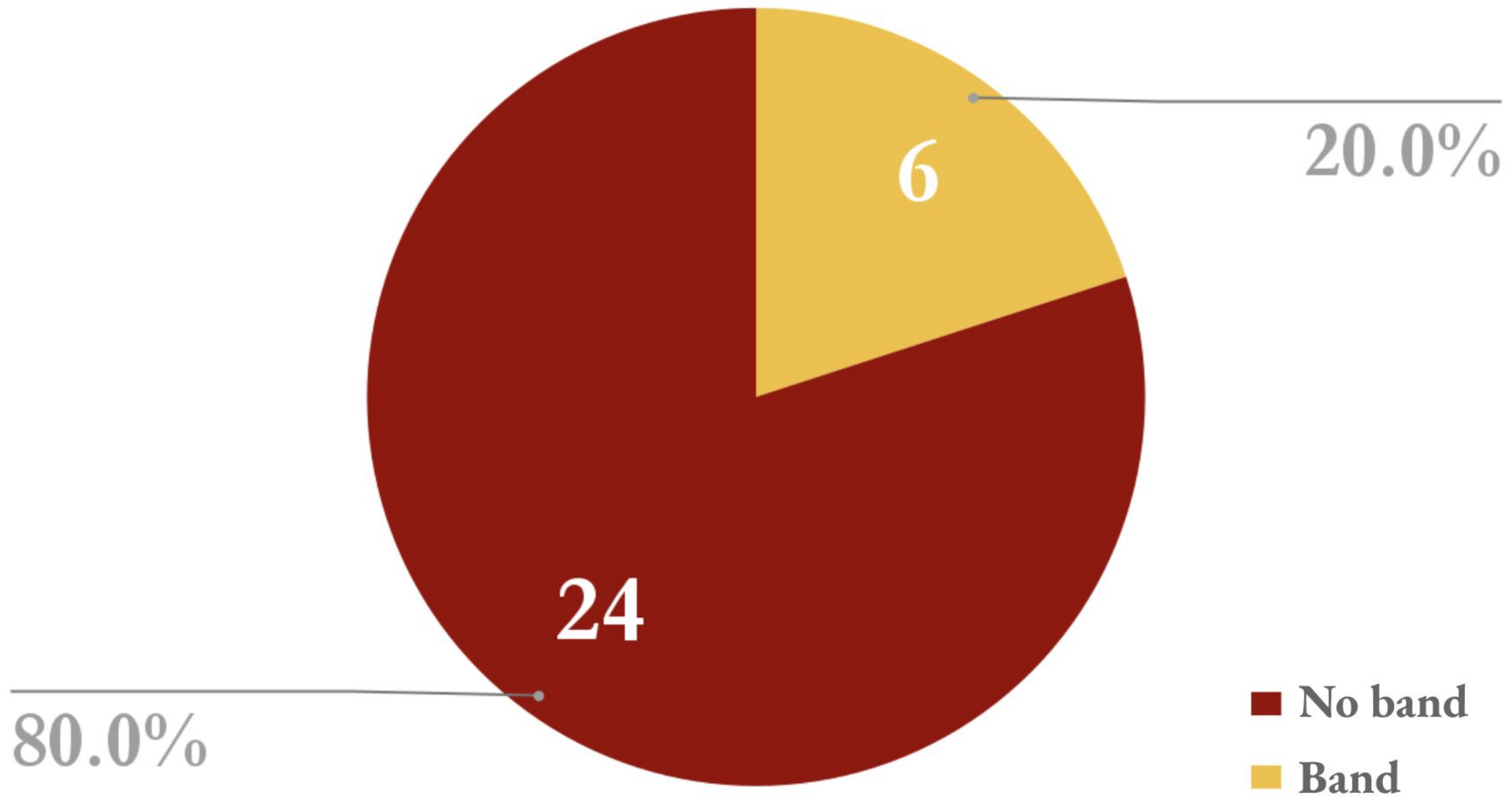

Figure 4. Presence of DNA in Active Method Specimens. The DNA samples were amplified using PCR and underwent gel electrophoresis to determine the presence of DNA as indicated by the appearance of a band on the gel.

Results

- **Passive DNA Collection Technique**
  - Presence of DNA in 11 of 30 specimens, making up 36.7% of the samples (Fig. 3)
  - No DNA band present for 19 specimens, meaning the samples lacked sufficient DNA for amplification and interpretation
- **Active DNA Collection Technique**
  - Presence of DNA in 6 of 30 specimens, making up 20.0% of the samples (Fig. 4)
  - No DNA band present for 24 specimens, meaning the samples lacked sufficient DNA for amplification and interpretation

Conclusions

- **Connection to Research Question**
  - Passive sampling provides a distinguishable amount of DNA about 1.8 times more consistently than active sampling, thus passive sampling is a better method for DNA collection of vulnerable bee species than active sampling
  - Neither method worked exceptionally well, indicating that these techniques should be reserved for threatened species of bees, and traditional tarsal clips or euthanization of bees may be better options for well-established bee species

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