

## Macro Photography of Virginia Bee Species

Haden G. VonCanon '27 and Scott M. Starr

Department of Biology, Hampden-Sydney College, Hampden-Sydney, VA 23943

### Background Information

Bees are insects that belong to the order Hymenoptera. They are distinguished from other insects by their two sets of wings and by their hairs covering most of their body (National Wildlife Federation). They only eat nectar and pollen from plants in their habitat (National Wildlife Federation). They can vary in color and size (Odanaka et al., 2019). The hairs on their body are an easy way to distinguish them from other insects like wasps (Odanaka et al., 2019). Bees are classified into seven families which are Apidae, Megachilidae, Andrenidae, Halictidae, Melittidae, Colletidae, and Stenotritidae (Mulhern, 2022). Of these seven families there are over 20,000 different species of bees, 4,000 of which are native to North America (U.S. Geological Survey). In Virginia, 477 species have been documented (Orcutt, 2021). Of the 477 species documented, 19 of them are not native to Virginia (Orcutt, 2021).

Bees have many important uses. They are known for their production of honey. In North America, there is only one species of bee that makes honey, the European Honeybee, *Apis mellifera* (Burlew, 2021). The most important use of bees is pollination. Bees are responsible for pollinating over 30% of the agriculture that we eat and 20% of bees are specialists (Dingwell, 2015). Many native bee species help to pollinate specific species of plants. Knowing the diversity and the abundance of bees within a specific area is important for agriculture. Tragically, data is suggesting that bee populations have been in decline since the 1990s (Zattara, 2021). Because of this, it is even more important to collect and identify bee species within a region to better understand their biodiversity and the roles that individual bee species plays within the environment (Roulston et al., 2007).

A good way to document and identify bees is to take photos of them. Unfortunately, taking close up photos isn't as simple as just pointing a camera at a specimen. To get a closeup image we must use a microscope with an attached camera (Figure 1). These images often have a high depth of field resulting in only certain focal lengths of a sample to be in focus (Figure 2A). Capturing all focal points of the specimen is vital to preserve the details and characteristics that identify specimens. Focus stacking is a technique used by photographers to give photos multiple focal lengths. To do this, a photographer takes multiple shots of the same frame at different focal points (DeGuzman, 2022). These photos are then combined in post-production with all the focal lengths (DeGuzman,

2022) (Figure 2B).



Figure 1. Microscope and camera setup for macro photography.



Figure 2: A) *augochlora pura* macro image, B) *augochlora pura* photo stacked image.

## Methods

Bee specimens were collected from the Hampden-Sydney College campus during the spring and summer of 2024. Nets, bowl traps, and blue vein traps were used to capture specimens. After collection in the field, specimens were stored in the freezer or in ethanol. The specimens were prepared before the photos were taken. If the specimen was stored in ethanol, it was blow-dried under a metal mesh net to get all liquid off the body. From here, freezer and ethanol specimens experienced the same processing steps. Each specimen was pinned through the thorax using insect pins. They were then positioned with all morphological features visible. Specimens were left to completely dry for at least 24 hours, however some specimens required more dry time.

After the specimen were dried, they were ready for imaging. A Labomed Luxeo 6z stereo microscope was used with a Excelis 4k camera (Figure 1). Each specimen was carefully positioned onto a cork platform under the microscope. The cork held the specimen in place. The cork platform was placed out of the way. This allowed the specimen to be placed against a black background. Lights were then placed to fully illuminate the specimen.

Once the specimen was positioned and illuminated, we photographed the part of the bee that was closest to lens. We then took a picture and ever so slightly turned the focus wheel further from the camera lens. This process was repeated until the entire specimen was entirely out of focus. At the end of this process, we were left with anywhere from 50 to 150 photos.

Images were uploaded into the CombineZP software ("CombineZP."). CombineZP automatically analyzes the photos and combines all the focal lengths into one photo. If the focal stacked image was high quality, the next step was to edit. If areas of the specimen were not in focus, more pictures were taken to improve the quality of the focal stacked image.

Editing was done with the built in photo editing software into the Windows 11 operating system. Any editing done to these photos were basic color correction, cropping, and digital sharpening.

## Results

49 species of bees were captured and identified on campus during summer 2024.

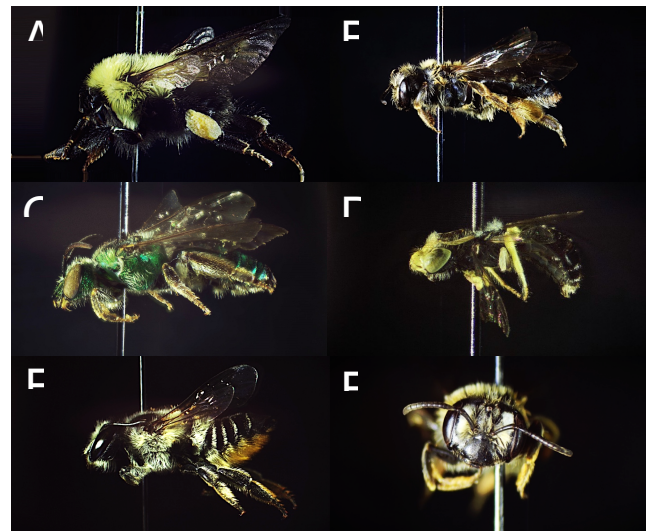


Figure 3: A) *Bombus perplexus*, B) *Andrena imitatrix*, C) *augochlora pura*, D) *calliopsis andreniformis*, E) *Megachile sp.* and F) *Megachile sp.*

## Discussion

These 6 images (Figure 3 A-F) are highlights from the photos that were produced. Focus stacking method was used to get as much of the specimen in focus, however some photos did not require this process. Figure 3 E-F are created using macro photography. Although focus stacking was used to show detail in specimens, we did find that not using focus stacking was useful for pictures of specific specimens (Figure 1A-B). By not using focus stacking, we were able to show the depth of these specimens. Figure 3 A-D used focus stacking and most of the detail on the specimen can be seen.

Because every part of the specimen is in focus, it is hard to see the depth of the specimen. It is difficult to discern the leg and wing placement with no depth. Figure 3F was a single photo from an attempted focus stack. Unfortunately, due to the head on orientation, it had a large depth. When these photos were stacked together, the product looked confusing and not aesthetically pleasing. Not all focus stacking worked perfectly.

It is possible to focus stack manually, yet it is unrealistic due to the volume of photos. That is why we had to use CombineZP. A result of this, was that not all photos were perfect. Many photos had unnatural warping. Almost all the photos had warping on the outside. The problem of warping on the border of the image was solved by cropping. When there was warping on the specimen, the image was not able to be saved.

## Conclusion

In conclusion, the pictures that were produced were high quality photos. Having the black backdrop improved the contrast. The contrast helped the features of the individual specimens stand out. Making these photos scientifically accurate and visually pleasing were the top priorities. The intention of a visually pleasing image is to draw people's attention to education on the bee populations. Many of these bee species ranged from 2mm in length up to 39mm in length. Bees of this size are often overlooked when considering bee education and population.

These photos were a helpful resource when identifying the bee species that were captured on campus. These images could also be uploaded to websites like iNaturalist.com where identification is made by the website users. A detailed photo is able to provide a lot more details for someone trying to identify a bee rather than a photo taken from a phone.

The photos stacking process can be used for more than just taking pictures of bees. Focus Stacking can be used for documenting smaller insects. By documenting and sharing the results of this process, many ecologists can learn how to use focus stacking for documenting smaller insects. In this digital age we live in, having detailed images of species is an important resource for scientists.

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