The Shape of Beetles: Analyzing Sexual Dimorphisms in Mountain Pine Beetle Using Geometric Morphometrics

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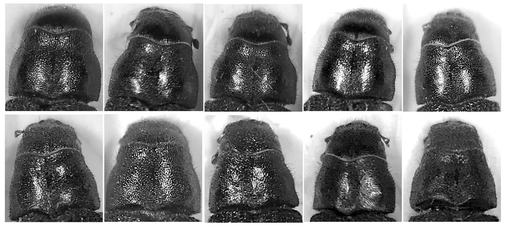
Introduction and Background

- Background: The mountain pine beetle, *Dendroctonus ponderosae* Hopkins, (MPB) relies on flight dispersal to colonize new host trees[1]; however, dispersal of MPB is difficult to study and is therefore not fully understood[2].
- Flight dimorphisms: Male and female MPB fly with similar velocities and have the same wing area proportional to their size, but they possess different propulsion parameters (Strouhal number)[3]. Body shape can affect flight parameters such as propulsion[4].
- **Objective:** Determine if there is a difference in wing and thorax shape between male and female MPB and test the method of geometric morphometrics in bark beetles.
- **Purpose:** Analyzing dimorphisms will be useful to further understand MPB dispersal, and subsequently their ecology, life cycle, and colonization process.



Activity!
- We propose that male and female MPB have different thorax shapes.
- One row shows males and the other females.

Do you see any patterns?

Methods Part 1

Data Collection

Photographing Process:
- Separated > 40 MPB by sex.
- Mounted 10 male and 10 female MPB in plasticine (Fig. 1).
- Dissected the right wing from 10 male and 10 female MPB and mounted the wings onto microscope slides (Fig. 3).
- Photographed MPB and wings with cell phone camera adapter under a stereomicroscope (1.6x).

Measurements of Size and Shape:
- Photos were cropped, rotated, and converted to greyscale in GIMP[5].
- JMorph[6] was used for measurements and placement of landmarks.
- Thorax measurements focused on the size and shape of the pronotum (Fig. 2)
- Wing measurements focused on the size and shape of the wing margin, landmarks placed on important wing venation (Fig. 4).

Pronotum measurements shown in Fig. 2:
- Landmarks were on either side of the anterior and posterior pronotum (A-D).
- Measured width of anterior (E) and posterior (F) pronotum; pronotum length (G).
- Generated Fourier outline (H) around pronotum to obtain area, length, and shape.

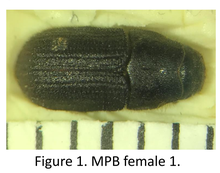


Figure 1. MPB female 1.

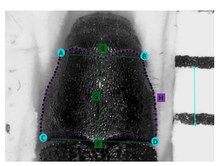


Figure 2. MPB female 1 in JMorph after landmarks and measurements have been taken.

Methods Part 2

Wing measurements shown in Fig. 4:
- Established landmarks (18, green) at the origin and terminus of wing veins and at any branching points.
- Created Fourier outline (purple) around the wing margin to obtain area, length, and shape of the wing.

Figure 3. Wing of female 1.

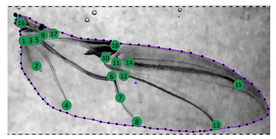
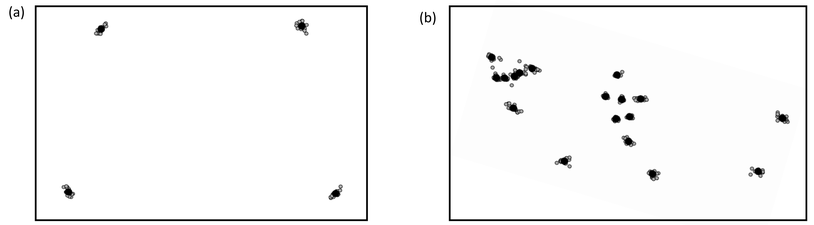


Figure 4. Wing of female 1 in JMorph after landmarks and measurements have been taken.

Analysis

Geometric Morphometrics:
- Data from JMorph were analyzed in R[7] using the *geomorph*[8] package and *vegan*[9]
- ANOVA was used to analyze differences in size and area between sexes.
- Procrustes analysis was used to compare shape as a dimensionless quality.



Procrustes analysis plots pronotum (a) wing (b):
- Black points represent the mean position of the landmarks of all samples.
- Grey points represent the position of the landmark from each beetle.
- If groups of points that separate from the mean can be classified differently (ie. M or F), they may have different relative shapes.

Results: Relative Shape

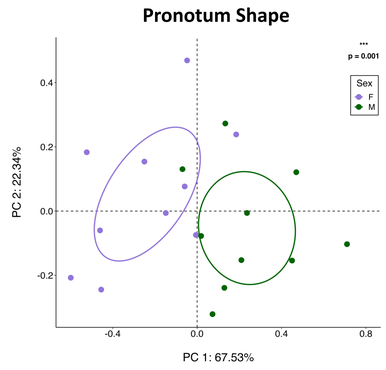


Figure 5. Procrustes analysis of thorax shape. Points are relative shape of each beetle, ellipses are 95% CI of the shape of each sex. P-value from PERMANOVA shows pronotum shapes of male and female MPB are significantly different.

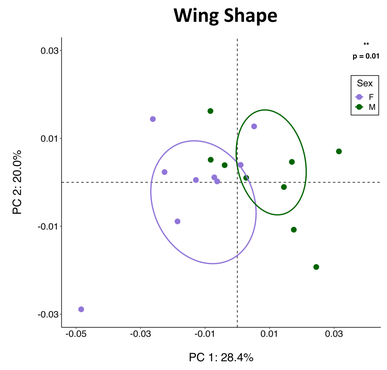


Figure 6. Procrustes analysis of wing shape. Points are relative shape of each beetle, ellipses are 95% CI of the mean shape of each sex. P-value from PERMANOVA shows wing shapes of male and female MPB are significantly different.

Results: Area

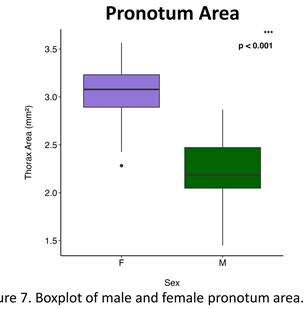


Figure 7. Boxplot of male and female pronotum area. There is a significant difference in area of the pronotum between male and female MPB. This confirms previous knowledge that differences in area exist[9] and validates the results from JMorph.

Figure 8. Boxplot of male and female wing area. There is a significant difference in area of the wing between male and female MPB. This again confirms previous knowledge about wing size[10] and validates the results obtained in JMorph.

Conclusions

- Sexual dimorphisms in pronotum shape and wing shape exist in MPB. Difference in wing shape may be significant in determining why male and female MPB have different Strouhal numbers.
- Geometric morphometrics can be applied to the field of entomology and can accurately measure quantities like area, as well as dimensionless qualities like shape. JMorph, and all programs used throughout this project, are open access and are therefore easily accessible and efficient tools for studies involving geometric morphometrics.

Future Research

- The use of geometric morphometrics was previously utilized almost exclusively by paleontologists. By applying geometric morphometrics to studies of living organisms and confirming the reliability of programs like JMorph, we hope this tool can be exploited across many fields of biology and science.
- Our project inspired many questions about the relationship between pronotum shape, wing shape, and flight dispersal capacities. Previous studies have shown that dimorphisms exist in how MPB expend their energy during flight. Future research could examine the relationship between pronotum shape, wing shape, and flight efficiency in male and female MPB.

References

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Acknowledgements

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Pglelievre: https://github.com/pglelievre/jmorph.git