**Comparing *Canis* Skull Morphology in R**

**1. Tutorial Overview**

*Biological significance.* Morphometrics is the branch of biology that quantitatively examines biological shape, shape variation, and covariation of shape with additional factors (Webster and Sheets 2010). Landmark-based 3D geometric morphometrics involves the placement of Cartesian coordinates (*x*, *y*, *z*) at specific loci on an anatomically relevant structure (Webster and Sheets 2010; Theska et al. 2020). The goal of comparing landmarks between skulls is to infer the biological relevance of differences in a structure’s shape. This technique has been utilized across Canidae to distinguish populations, determine past hybridization, delineate taxonomy, and evaluate conservation status (e.g., Milenkovic et al. 2010; Schmitt and Wallace 2014; Machado and Teta 2020; Zdjelar et al. 2021, etc.).

*Data readying.* The goal of this tutorial was to create a framework to reliably perform geometric morphometric analyses from initial data collection to final visualization. To accomplish this, I utilized a raw dataset containing 148 individual canids across seven taxonomic delineations. The raw data was imported into 3D Slicer 5.6.1 (Fedorov et al. 2012) via the SlicerMorph extension (Rolfe et al. 2021). Each skull was pseudo-landmarked with 268 x, y, and z points, which were then subject to a Generalized Procrustes Analysis (GPA). The GPA removes any variation due to allometric variables, such as size of structure, to allow for comparison between skulls. Following GPA alignment, the translated data is exported as a CSV containing each skull’s x, y, and z landmark coordinates, Procrustes distance, and centroid. A separate CSV contains classifier information (e.g., group, species, sex, etc.).

*R analyses.* Once the CSVs are imported into R, they are tidied up and combined into a 3D array via the *abind* package. Once in proper structure, the data once again undergoes a GPA (to prevent translation errors that sometimes occur when importing to R) via the *geomorph* package (Adams and Otárola-Castillo 2013). A principal components analysis (PCA) is then performed on the data to assess which combination of anatomical structures accounts for the most variation in the dataset (*geomorph*). The results of the PCA are then displayed visually with a custom legend that shows each included canid group in the analyses.

**2. Selected References**

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