Labeled Segmentation of Honeybees Providing 3D Visualizations of Collective Behavior

Caitleen Reyes[1] Danielle Chase[2] Orit Peleg[3]

Abstract

Honeybee swarms demonstrate remarkable social behavior, enabling the range of emergent physiology through collective adaptation. The study explores the morphology and behavior of swarms using 3D visualizations and computational analyses derived from labeled segmented datasets of X-ray imaging. Results reveal a dense internal structure with spatial variations and greater stability at the base due to varied orientations. Ellipsoids were found to be effective for representing individual bees, providing realistic visualizations of swarm structure. The work highlights the potential of advanced labeling and visualization techniques to improve the understanding of honeybee swarm morphology and behavior.

Introduction

Honeybees (*Apis mellifera*) are unique social insects that form a superorganism when they cluster as a swarm (Seeley, 1989). They can accomplish tasks that are greater than what any individual bee can achieve on its own as a result of their cooperative collective behavior (Peleg et al., 2018). In order to communicate, honeybees act as signal amplifiers, making use of chemical cues to communicate pheromone-based information (Nguyen et al., 2021). As individual bees work together, a range of emergent behaviors arise.

In a previous study examining how honeybee swarms withstand dynamic mechanical forces, Peleg et al. (2018) simulates swarms under different loading conditions by creating pendant clusters and exposing them to variations in orientation, frequency, amplitude, and duration. Their findings revealed that swarms actively adapt their morphology via a reversible shape modification to enhance collective stability at the cost of increasing the mechanical load burdened on each individual bee. The individual movement of bees is thus critical to understand as they move from areas of low to high strain in response to local strain gradients. Similarly, Peters et al. (2022) investigates individual bee response to local stimuli caused by fluctuating ambient conditions by analyzing changes in internal temperature, shape, and size of swarm clusters exposed to dynamically changing temperatures. Their results observe clusters that engage in collective thermoregulatory morphing in order to preserve an optimal internal temperature range. Together, these two studies highlight that swarm clusters respond to environmental conditions with multifunctional morphological adaptations that rely on the collective integration of multiple sensory inputs (Peters et al., 2022). However, advancing the understanding of swarm morphology and its collective behavior requires improved methods for analyzing individual bee behavior and the internal structure of clusters.

A study exploring the application of strength-mass scaling laws in the mass distribution of honey bee swarms utilizes 3D reconstructions of bee positions within swarms using X-ray computed tomography (Shishkov et al., 2022). This approach enables detailed 3D visualization of the spatial arrangement of individual bees, facilitating more comprehensive analyses. As a result, this study strives to build on this methodology and improve the tracking of individual bees to better examine individual and collective behaviors. By expanding and refining our training data, we seek to discover new insights into the intricate coordination within honeybee swarms.

Methods

Small dataset analysis

A small cube-sized sample of approximately 70 bees was taken from a bee swarm through X-ray images. The sample was loaded into the image computing software 3D Slicer to inspect its axial, sagittal, and coronal views. Each individual bee in the sample was labeled using these three perspectives. The X-ray imaging data was then imported into a program to generate multiple histograms of the different bee sizes when various thresholds are applied to the X-ray image. Additionally, the labeled segmentation file was processed in another program to produce a histogram showing the distribution of bee sizes.

Multiple neural networks were programmed using a basic architecture with 1000 epochs to accept labeled segmentation files as training data. These networks were designed to evaluate test data, loss on training data, return histograms of torch tensor outputs, and plot residuals. The small dataset segmentation file was used to train several U-Net architectures with different configurations, including basic pathways and variants with halved or doubled channel numbers. Another U-Net model was trained using two different segmentation files derived from small cube-sized samples of the bee swarm.

Large dataset analysis

A full small bee swarm containing about 300-400 bees was acquired from X-ray images to serve as a sample for a large dataset. The sample was loaded into 3D Slicer software, where the labeling of individual bees began. To ensure proper labeling without any disconnected components, code was implemented to detect any disconnected components present in the labeled segmentation file.

Labeling bees accurately is a time-intensive process, so additional programs were developed to analyze the small swarm and create various visualizations during labeling. First, a histogram of bee size distribution was plotted. Then, the center of mass for each connected component was calculated and saved as a list of (x,y,z) centers. The centers were used as data to construct a 3D point cloud of bee positions, both with and without the centroid marked. Four histograms were generated to examine the distribution of bees in the radial (r) and vertical (z) directions: two histograms represented the number of bees, and two represented the number of bees per unit area/height. The average distance between a bee and its nearest neighbors was analyzed. Lists of mean distances to the nearest three and six neighbors for each point were saved, and the overall averages were calculated. These mean distance lists for three and six neighbors were plotted as functions of the Z-coordinate (where the swarm's base is z = 0 and its tip is z = length) and R-coordinate. The visualizations were created using two methods. The first approach involved outputting four histograms using binning to illustrate the average distances to the three or six nearest neighbors as functions of Z or R. The second approach produced four scatter plots using direct plotting of the average distances to the three or six neighbors against the Z or R coordinates. Two 3D plots were created using the centers of mass for each connected component, with points colored according to a scale representing the average distance to their three or six nearest neighbors. Additionally, the principal axis of each bee, represented as a 3D unit vector, was calculated, and the angle between each principal axis and the Z-axis was determined. These angles were visualized in scatter plots as functions of the R and Z coordinates, while a final 3D plot depicted individual bees, colored by the angle of their principal axes with respect to the Z- axis.

Lastly, a 3D visualization of individual bees within the small swarm was generated using ellipsoids in ParaView, where each ellipsoid was color-coded based on the angle between its principal axis and the Z-axis.

Results

A small cube-sized sample of a bee swarm was fully labeled with 67 bees (Figure 1). Out of the three variations of U-Net architectures, the configuration with the basic pathways performed the best with in comparison to the halved and doubled channel numbers when running with the fully labeled sample as training data.

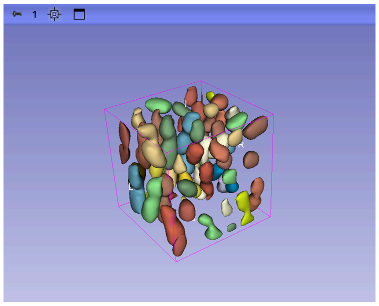

Figure 1. Full 3D segmentation of 67 individual bees from a small cube-sized sample of a bee swarm. The image was created using the software 3D Slicer.

A small swarm with about 300-400 bees was partially labeled with no stray disconnected components (Figure 2). The small swarm sample was used to create visualizations that analyzed its different properties.

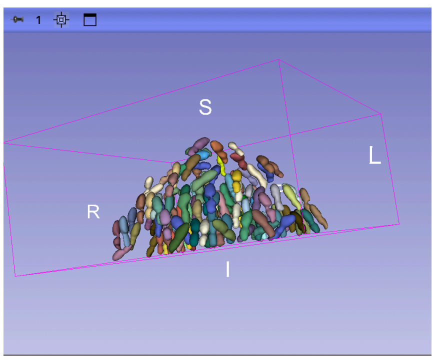

Figure 2. Partial 3D segmentation of individual bees from a 300-400-sized small swarm. The image was created using the software 3D Slicer.

The distribution of bee sizes within the small swarm ranged from 500-1350 pixels, following a pattern resembling a normal distribution, with the most frequent bee sizes occurring near the middle of the range. Bees were more densely packed closer to the middle-range distance from the centroid, while their numbers gradually decreased toward the tip of the cluster. As the distance from the centroid increased, the proportion of vertically and horizontally oriented bees remained relatively consistent, with vertically oriented bees being more prevalent. Moreover, the base of the swarm exhibited greater variation in bee orientation, spanning from nearly vertical to nearly horizontal, compared to the tip of the swarm. All point clouds that were produced reflected the overall shape of a honeybee swarm cluster.

Using ellipsoids to represent individual bees proved to be the most effective approach for creating a 3D visual model of the small swarm. The two most realistic visualizations, closely resembling actual honeybee clusters, were achieved through different approaches. In the first approach (Figure 3), I subjectively adjusted the X, Y, and Z radius ratios of the ellipsoids to approximate the typical proportion of bee sizes. In the second approach (Figure 4), I determined the scaling based on data from the segmented bees in the segmentation file used by calculating the average X, Y, and Z radius dimensions and reducing each dimension by a factor of 0.7 to prevent overlap.

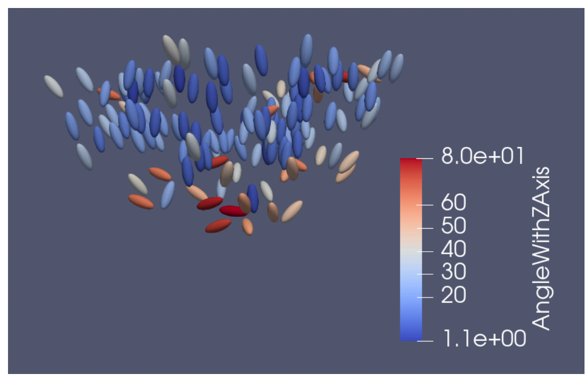

Figure 3. Each 3D ellipsoid's (x,y,z) radius dimensions were set to be (3,3,8). The ellipsoids represent individual bees and the angle between their principal axis and the Z–axis. The image was produced using the computer application ParaView and is based on a partially segmented 300-400-sized small swarm.

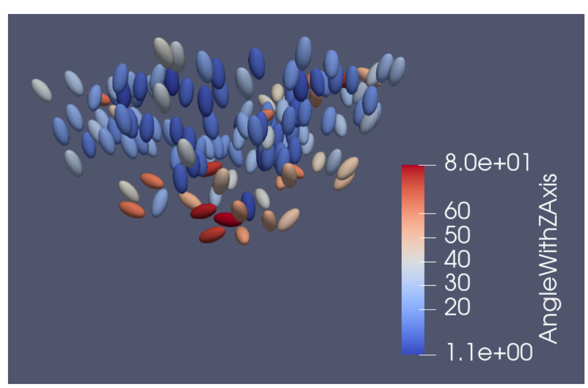

Figure 4. Each 3D ellipsoid's (x,y,z) radius dimensions were set based on the average dimensions of the segmented bees. The ellipsoids represent individual bees and the angle between their principal axis and the Z–axis. The image was produced using the computer application ParaView and is based on a partially segmented 300-400-sized small swarm.

Discussion

When analyzing variations in the U-Net architecture configurations, halved channel numbers resulted in poorer accuracy and performance, while doubled channel numbers had slower running times and required more memory. As a result, the basic pathway performed the best, balancing both efficiency and performance. The swarm exhibited a dense internal structure throughout, except in the middle, where space was allocated for the queen's box, and the outermost layer, where there were bees who were more sparsely distributed acting as stragglers. The higher concentration of bees at the base compared to the tip of the swarm was attributed to the structural integrity provided by the conical shape, where a stronger base was achieved with more bees concentrated at the bottom. The consistent radial distribution of bee orientations can be accredited to the space efficiency provided by uniform orientation. However, the base of the swarm showed greater variation in bee orientation compared to the tip because the variation enhances stability at the base, where structural support is critical. Ellipsoids proved to be the most effective representation of honeybees, providing an intuitive visualization of the swarm's structure. Similarly, in a study on bacteria in biofilms Nijjer et al. (2021) used 3D visualizations to represent the spatial orientation of bacterial cells on biofilms, employing ellipsoid-like shapes. Their methods aligned with our reasoning to utilize ellipsoids as representative shapes. Our results aligned with our expectations, suggesting that improved training data through labeled swarms enhances visualizations and enable more accurate and in-depth analysis. However, the current small swarm is not fully labeled, which introduces the discrepancies in the analysis of bee orientations due to missing labels for bees in the segmentation files. Future work should focus on completing the labeling of the small swarm to support more robust conclusions. Additionally, fully labeling multiple small swarms would provide better training data for improved neural networks and visualizations, allowing the opportunity for more detailed analyses.

References

[1] Nguyen, D. M. T., Iuzzolino, M. L., Mankel, A., Bozek, K., Stephens, G. J., & Peleg, O. (2021). Flow-mediated olfactory communication in honeybee swarms. *Proceedings of the National Academy of Sciences*, *118*(13), e2011916118.

[2] Nijjer, J., Li, C., Kothari, M., Henzel, T., Zhang, Q., Tai, J. S. B., ... & Yan, J. (2023). Biofilms as self-shaping growing nematics. *Nature Physics*, *19*(12), 1936-1944.

[3] Peleg, O., Peters, J. M., Salcedo, M. K., & Mahadevan, L. (2018). Collective mechanical adaptation of Honeybee Swarms. *Nature Physics*, *14*(12), 1193–1198.

[4] Peters, J. M., Peleg, O., & Mahadevan, L. (2022). Thermoregulatory morphodynamics of honeybee swarm clusters. *Journal of Experimental Biology*, *225*(5), jeb242234.

[5] Seeley, T. D. (1989). The honey bee colony as a superorganism. *American Scientist*, *77*(6), 546-553.

[6] Shishkov, O., Chen, C., Madonna, C. A., Jayaram, K., & Peleg, O. (2022). Strength-mass scaling law governs mass distribution inside honey bee swarms. *Scientific Reports*, *12*(1), 17388.