Brain composition in *Godyris zavaleta,* a diurnal butterfly, reflects an increased reliance on olfactory information

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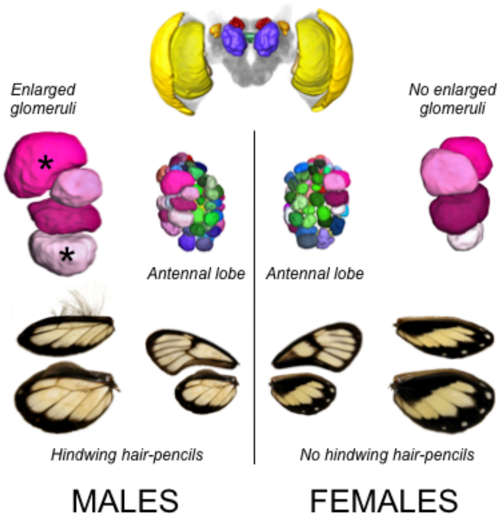
The size and structure of nervous systems are shaped by selection in the context of developmental and functional constraints. Understanding how and to what extent selection negotiates these constraints to bring about adaptive evolutionary change that enhances the fitness of an animal's behaviour is key to understanding the principles of brain evolution. The principal way to tackling these questions has been to compare brain size and structure across multiple species with divergent ecologies. The Lepidoptera have long been a favoured model in evolutionary biology, but descriptions of brain anatomy have to date largely focused on the few species that are commonly used as model organisms in neurobiological research.

We describe the brain of the Zavaleta Glasswing, *Godyris zavaleta*, a member of the Neotropical butterfly sub-family Ithomiinae with enhanced reliance on olfactory information and a derived mating behaviour. This entails sex-specific pheromone communication mediated by hind-wing 'hair-pencils' and a sex-specific motivation to locate specific plant allelochemicals (pyrrolizidine alkaloids) that serve as precursors for pheromone synthesis. We test two hypotheses: i) that the derived mating behaviour of *G. zavaleta* results in sexual dimorphism in the antennal lobes not observed in other butterflies*;* and ii) that the generally enhanced role of olfaction in *G. zavaleta* is supported by shifts in the relative investment in sensory neuropiles. We demonstrate for the first time the presence of sexually dimorphic glomeruli within a distinct macroglomerular complex (*MGC*) in the antennal lobe of a diurnal butterfly.

This presents a striking convergence with the well-known moth *MGC*, prompting a discussion of the potential mechanisms behind the independent evolution of specialized glomeruli. Interspecific analyses across four Lepidoptera further show that the relative sizes of their sensory neuropiles closely mirror interspecific variation in sensory ecology, with *G. zavaleta* displaying levels of sensory investment that are intermediate between the diurnal Monarch butterfly (*Danaus plexippus*), which invests heavily in visual neuropile, and night-flying moths which have a greater investment in olfactory neuropile, and diminished visual neuropile. We identify several traits that distinguish butterflies from moths, and several that distinguish *D. plexippus* and *G. zavaleta*. Our results illustrate how ecological selection pressures mould the structure of invertebrate brains, and exemplify how comparative analyses across ecologically divergent species can illuminate the functional significance of variation in brain structure.


Enlarged glomeruli
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Antennal lobe
Hindwing hair-pencils
MALES
No enlarged glomeruli
Antennal lobe
No hindwing hair-pencils
FEMALES