## Characterizing the cis-regulatory evolution of the gene WntA in nymphalid butterflies

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Evolutionary changes in wing patterns of Nymphalidae butterflies, such as that of the common buckeye butterfly (Junonia coenia), are mediated by regulatory elements associated with a few master genes. Credit: *Science* (2022). DOI: 10.1126/science.ade5689

A combined team of researchers from Cornell University and The

George Washington University, has characterized the cis-regulatory evolution of the gene WntA in nymphalid butterflies. In their study, published in the journal *Science*, the group used a variety of techniques to better understand how gene regulatory processes in a type of butterfly can allow for both deep homology and speedy adaption to environmental changes.

Marianne Espeland and Lars Podsiadlowski with Leibniz Institute for the Analysis of Biodiversity Change at Museum Koenig, have published a Perspective piece in the same journal issue outlining the means by which gene regulatory agents play a major role in the pattern formation of butterfly wings and the work done by the team in this new effort.

Prior research has shown that patterns in the appearance of creatures such as butterflies and the structures behind them arise due to the influence of transcription factors and cis-regulatory elements (CREs.) But how such factors have evolved is still not very well understood. In this new effort, the researchers sought to gain insight into the regulatory systems that underpin rapidly evolving traits.

To learn more about such systems, the researchers conducted comparative sequence analysis, ATAC-seq on five species of butterflies, focusing specifically on 46 CREs and the WntA gene. They also used CRISPR knockouts to learn more about the role that the WntA gene plays in wing color. They found strong evidence that suggests regulatory genetic elements from the deep evolutionary past in nymphalid butterflies exert a strong influence on the ways that patterns have developed on their wings.

The researchers found that the WntA gene is a master gene, playing a critical role in the development of patterns that appear on butterfly wings. They found that its expression has an impact not just on color and patterns, but on the expression of several other genes located near it in

the butterfly DNA. And by generating deletions using CRISPR-Cas9, they were able to study the impact of multiple transcription factors and CREs on wing pattern and color. They found multiple changes in one species that were not evident in others, suggesting newly evolved CREs that had very quickly become part of the regulatory process.

**More information:** Anyi Mazo-Vargas et al, Deep cis-regulatory homology of the butterfly wing pattern ground plan, *Science* (2022). DOI: 10.1126/science.abi9407

Marianne Espeland et al, How butterfly wings got their pattern, *Science* (2022). DOI: 10.1126/science.ade5689

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