

## One or two species? Genomics provide insight in the systematics of the Green Whip Snake (*Hierophis viridiflavus*)

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### Abstract:

The two subspecies of the Green Whip Snake *Hierophis viridiflavus viridiflavus* and *H. v. carbonarius* represent an interesting model system to study speciation. Indeed, they likely evolved during glacial periods due to isolation by distance. Recent studies show that even though these subspecies belong to different mitochondrial lineages, a single tested nuclear gene and morphological traits display overlapping character states. Moreover, little is known whether these two subspecies are reproductively isolated from each other. Furthermore, it is unknown how the complete or partial melanism of the subspecies *carbonarius* is regulated and why it is restricted to this eastern subspecies only. Therefore, we evaluated the amount of gene flow within two transects across the contact zone of the subspecies with a genomic approach using double-digest restriction site-associated DNA (dd-RAD). Our molecular samples, including 148 individuals from the contact zones and further away, were genotyped on 24,817 single nucleotide polymorphisms (SNP). Admixture analysis supports the existence of two well-defined clades corresponding to both subspecies. However, a high gene flow between both subspecies was observed within a cline of 300 km. Moreover, associations between phenotypic characters and genomic markers were investigated by quantifying the amount of yellow coloration and by applying geometric morphometrics on the head shape and pholidosis. We conclude that the two subspecies represent evolutionary significant units (ESU). Due to the extensive hybridization zone, it seems that reproductive barriers have not evolved (yet). On a taxonomic basis, we suggest continuing treating the two ESU as subspecies.