Genomic identification and patterns of expression of secondary metabolite gene clusters in the entomopathogen fungus *Metarhizium anisopliae*

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The *Metarhizium* genus harbors cosmopolitan fungi that infect arthropod hosts. Importantly, while some species infect a wide range of hosts (host-generalists), other species infect only a few arthropods (host-specialists). This singular evolutionary trait permits unique comparisons to determine how pathogens and virulence determinants emerge and evolved. Among the several virulence determinants that have been described, secondary metabolites (SMs) are suggested to play essential roles during fungal infection. Nevertheless, genes related to SM production in *Metarhizium* spp. are scarcely described and little is known about their genomic organization, expression, regulation and role during host infection. Here, we have performed a deep survey and description of SM biosynthetic gene clusters (BGCs) in *M. anisopliae* and assessed conservation among the *Metarhizium* genus. RNA-seq data from fungi grown on cattle-tick cuticles (mimicking infection) was analyzed to validate some of the predictions and to access the differential expression of BGCs. Furthermore, our analysis extended to the construction of a phylogeny for the following three BGCs: a tropolone/citrinin-related compound, a pseurotin-related compound, and a putative helvolic acid. Among 73 BGCs identified in *M. anisopliae*, 20% were up-regulated during initial tick cuticle infection and presumably possess virulence-related roles. These up-regulated BGCs include known clusters, such as destruxin, NG39x and ferricrocin, together with putative helvolic acid and pseurotin- and tropolone/citrinin-related compound clusters as well as uncharacterized clusters. Concerning host-range several up-regulated BGCs were not conserved in host-specialist species from the *Metarhizium* genus, indicating possible differences in the metabolic strategies employed by generalist and specialist species to overcome and kill their hosts. These differences in metabolic potential may have been partially shaped by horizontal gene transfer events, as shown in our phylogenetic analysis. In conclusion, several unknown BGCs are described, and their organization, regulation and origin are discussed, providing support for the impact of SM on the *Metarhizium* genus lifestyle and infection process.