First report of *Metharizium* melanins: Preliminary evidence of 1,8-dihydroxynaphthalene-melanin-like compound in *Metarhizium robertsii*

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Microsclerotia of Metarhizium robertsii, fungal aggregates consisting of compacted, pigmented hyphae, exhibit high desiccation tolerance and produce infective conidia, rendering them promising candidates for bioinsecticide formulation. Despite their potential, the pigments' nature within these structures remains elusive. This study supplements routine culture media for M. robertsii microsclerotia differentiation with four melanin inhibitors. Results show significant phenotypic and molecular alterations induced by inhibitors of the 1,8-dihydroxynaphthalene (DHN)-melanin biosynthetic pathway, such as tricyclazole and guaiacol. These modifications include a more spherical shape, reduced size, and heightened susceptibility to desiccation, heat, and oxidative stress compared to inhibitor-free microsclerotia. Moreover, upregulation of genes encoding a polyketide synthase (Mrpks2) and a putative 1,3,6,8-tetrahydroxynaphthalene reductase (Mrthnr) in inhibitor-supplemented media suggests their involvement in the DHN-melanin biosynthetic pathway. Importantly, this study represents the first report of melanins in the genus Metarhizium, implicating their role in oxidative and thermal stress tolerance alongside microsclerotia differentiation.

Keywords: Melanin inhibitors; tricyclazole; guaiacol; thermotolerance; oxidative stress