

Unveiling the Role of Melanin in Fungal Pathogenicity and Environmental Stress Resistance

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Abstract

Melanin, which is produced by various microorganisms including fungi and bacteria, plays essential roles with a broad range of functions. Melanisation fortifies fungal microorganisms against various environmental stressors including UV radiation, heavy metals, desiccation, hydrolytic enzymes, oxidative agents, heat, and cold amplifying their virulence as protective armour. This study illuminates the roles of melanin in plant-pathogenic fungi, including its function as a shield against host defence mechanisms, a facilitator of penetration and colonization, and a promoter of pathogen establishment and dissemination.

Introduction

Melanin is formed through the oxidative polymerization of phenolic compounds into high molecular weight pigments. A substance qualifies as amelanin if it is dark in color, insoluble in both aqueous and organic solvents, resilient to concentrated acids, and vulnerable to oxidizing agents bleaching action. Fungi, bacteria, and helminths host microbes produce melanin pigments. Ortho-dihydroxyphenols are the general class of monomers for conjugated polymers. Each polymeric melanin residue incorporates two ortho oxygens. Melanins, extracted mostly from fungi, are derived from the precursor 1,8-dihydroxynaphthalene using the polyketide pathway in ascomycetes and deuteromycetes. Basidiomycetes utilize g-glutaminy-4-hydroxybenzene and catechol as additional monomeric precursors for melanin production. It is a stable free radical, as determined by Electro Spin Resonance (ESR) characteristics, defines melanin as a pigment. These microbes' virulence and pathogenicity for their host plants are associated with their melanin production capability. The prominent discussion concerns the role of melanin in enhancing fungal pathogens' virulence, which has been the most extensively investigated mechanism.

Melanin biosynthesis: Many soil fungi produce melanin, which shields them from various

environmental injuries. The production of large quantities of melanin by melanotic fungi indicates its significance as a fungal metabolic product. 30% of *Agaricus bisporus* dry weight comprises melanin. *A. alternata* and *Cladosporium sphaerospermum* among them have been identified in the Chernobyl nuclear reactor site, an environment where radiation levels surpass 10 thousand times the normal limit is mainly due to the presence of melanin in their cell wall. 1,8-di-hydroxy naphthalene and L-3,4-dihydroxyphenylalanine as the precursors of DHN-melanin and DOPA-melanin which are the two types mainly involved in tyrosine and polyketide pathway. (Table 1)

Table 1. Types of melanin biosynthetic pathways in fungus

Synthesis	Enzyme	Example
DHN-melanin	Polyketide Synthase (PKS)	<i>Colletotrichum lagenarium</i> <i>Magnaporthe oryzae</i> , <i>Verticillium dahlia</i> <i>Wangiella dermatitidis</i> <i>Alternaria alternata</i> <i>Aspergillus fumigatus</i>
DOPA-melanin	Tyrosinase or Laccase	<i>Cryptococcus neoformans</i>

Relation of fungal melanin to virulence

Many potentially invasive fungi, not all of which are pathogenic, produce melanin. These are called dematiaceous or phaeohyphomycetous fungi (Table 2). During appressorial differentiation and late stationary mycelial growth, fungal plant pathogens synthesize DHN melanin. An asexual spore's germ tube gives rise to the appressorium as a solitary cell. The appressorium is mostly melanized, with the exception of its pore, through which the fungus interacts with the host plant cell. The lack of melanin at the appressorium pore enables the discharge of cell wall degrading enzymes and intake of nutrients. Melanin synthesis during appressorial growth is controlled by developmental factors. These fungi *viz.*, *Magnaporthe grisea*, *Colletotrichum lindemuthianum*,

Colletotrichum lagenarium require melanin formation in their appressorium for successful invasion of rice, bean, and cucumber, respectively. The penetration peg, rooted in the appressorium pore, pierces the host cell. The immense turgor pressures, over 80 bar, aid in penetrating the host cells epidermal cuticle. Melanin in melanized appressoria acts as a permeability barrier to glycerol. The penetration peg of *Colletotrichum graminicola* exerts about 17 micronewtons of pressure on the host cell.

Table 2. Mechanisms developed by fungus against various host produced chemicals

Mechanism	Target pathogen	Activity
Protection against oxidants	<i>Cryptococcus neoformans</i> <i>Aspergillus niger</i> <i>Alternaria alternata</i> <i>Sporothrixsc henckii</i> <i>Aspergillus fumigatus</i> <i>Magnaporthe grisea</i> <i>Proteus mirabilis</i>	Reduction of Fe^{3+} to Fe^{2+} Redox buffer Against permanganate and hypochlorite Against ROS through redox buffer action ROS quenching mechanism Hydrogen peroxide scavengers
Effect on Phagocytosis	<i>Cryptococcus neoformans</i> <i>Exophiala dermatitis</i> <i>Sporothrixsc henckii</i>	Altered surface hydrophobicity and charge Produce defensins, microbicidal peptides
Resistance to antimicrobial compounds	Yeast <i>Moniliniafruticola</i> <i>Botrytis cinerea</i>	Capsfungin activity Against dicarboximide Conversion of resveratrol by laccase- Fungal toxin

Role of melanin in environmental protection

The melanin production of free-living microbes is linked to an environmental survival advantage. Melanin protects against UV damage by absorbing a wide spectrum of electromagnetic radiation. Compared to non-melanized fungi, melanized fungi exhibited greater resistance. The Chernobyl Reactor no 4 is most notably colonized by black fungi, demonstrating exceptional resilience to

radiation. It is involved in absorption and degradation of environmental pollutants, protection of microorganisms from radiation and oxidation, neutralises the ROS released into the environment. Various mechanisms underlying resistance was listed below in Table 3.

Table 3. Melanin induced resistance against various environmental stresses

Mechanism	Target pathogen	Activity
Resistance to UV light	<i>Monilinia fruticola</i> <i>Phaeococcomyces</i> <i>Alternaria alternata</i> <i>Cladosporium sp.</i> <i>Oidiodendron cerealis</i>	Adding melanin suspensions generated from fungal cell walls
Bind to heavy metals	<i>Cryptococcus neoformans</i>	Binds to silver nitrate
Protection from predators	<i>Cryptococcus neoformans</i>	Less susceptible for cell wall degrading enzymes
Protection from enzyme degradation	<i>Rhizoctonia spp.</i> <i>Sclerotinia spp.</i> <i>Verticillium sp</i> <i>Thielaviopsis basicola</i> <i>Phomopsis sp.</i> <i>Alternaria kikuchiana</i> <i>Phaeococcomyces sp.</i> <i>Magnaporthe grisea</i> <i>Monilinia fruticola</i> <i>Aspergillus sp.</i>	Enzyme sequestration

Conclusion: Melanin synthesis in *C. neoformans* satisfies the stringent conditions for being recognized as a virulence factor and serves as a prime model for grasping the role of melanogenesis in microbial pathogenesis. This treatment can trigger severe tumour reactions, potentially harming the host. The significance of melanization in microbial pathogenesis is ensured by its link to virulence and the widespread synthesis of melanin pigments by various pathogens. Melanotic fungi synthesize and modify melanin within their cell walls during growth. It enhances fungal pathogenicity by providing protection against environmental and host defenses, strengthening the cell wall, and increasing resistance to antifungal agents. Effective cell division and morphogenesis

require synchronization with melanin synthesis and degradation processes. Understanding melanin's role could lead to novel approaches in managing fungal infections. We are hopeful that this will significantly boost research in this mysterious field.

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