



The Role of Melanin in Plant Pathogenic Fungi: Insights into Structure, Biosynthesis, and Function

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Abstract

Melanin, a distinctive natural pigment produced by various microorganisms, including fungi and bacteria, plays vital roles and serves a wide array of functions. Melanisation as a 'fungal armour' significantly enhances the virulence of these microorganisms, offering protection against a range of environmental stressors such as UV radiation, heavy metals, desiccation, hydrolytic enzymes, oxidative agents, heat, and cold. The current review provides insights into the structure, biosynthesis, and multifunctional attributes of melanin in plant-pathogenic fungi and sheds light on how melanin contributes to the pathogenic virulence of fungi by shielding against host defence mechanisms, facilitating host tissue penetration and colonization, and promoting the establishment and dissemination of the pathogen within the host.

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Introduction

Melanin, a naturally occurring pigment, is widely found in various living organisms and possesses a range of functional properties and biological activities. The name "melanin" derives from the ancient Greek word "melanos," meaning black, due to its characteristic black or dark brown appearance [14]. Melanin is chemically stable and insoluble in most solvents, making it resistant to chemical degradation. It exhibits heterogeneity in composition, size, colour, function, hydrophobic character, and large molecular weight, with a significantly negative charge, often pigmented and is formed by oxidative polymerization of indolic and phenolic compounds [117].

The unique molecular structure of melanin contributes to its exceptional stability, rendering it resistant to various destructive physicochemical processes. Melanin is described as

a heterogenic polymer of phenolic or indolic nature. It is synthesized through the oxidative polymerization of phenolic compounds, primarily by two pathways. One pathway involves the use of a 1,8-Dihydroxynaphthalene (DHN) intermediate, while an alternative pathway utilizes L-3,4-dihydroxyphenylalanine (L-dopa) in certain fungi, resulting in different types of melanin such as eumelanin, pheomelanin, allomelanin, pyromelanin, and neuromelanin, based on the chemical composition of the monomer subunit structure [43,100]. Enzymes responsible for melanin synthesis mainly belong to the tyrosinase, laccase, and polyketide synthase families [118]. Melanin can be found in the cell walls of certain fungi, appearing as a distinct layer on the outside or associated with the fibrillar matrix of the cell walls. Additionally, extracellular melanin exists outside fungal cells and is separate from the cell wall-bound melanin [140].



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Melanin plays diverse and crucial roles, serving as a survival strategy for organisms inhabiting unfavourable environmental conditions. It affects fungi throughout their life cycle, providing protection against UV radiation, heavy metals, desiccation, hydrolysis enzymes, oxidative agents, heat, cold, and fungicides. Melanin also acts as an effective chelator of metal ions, safeguarding cells from potentially toxic ions and as a thermoregulator in many fungi [115]. In fungi, melanization is observed in cell walls, spores, vegetative hyphae, and fruiting bodies. Melanin deposition protects pigmented cells from physical and biological stress, prevents toxin entry, and minimises metabolite leakage [121]. Importantly, melanin contributes to the virulence of pathogens by reducing susceptibility to host antimicrobial mechanisms and influencing the host's immune response to infection [77].

This review will commence with an introduction to melanin, encompassing its chemical composition, biosynthesis process, and distribution within organisms. Subsequently, our attention will be directed towards elucidating the pivotal role of melanin in bolstering the functional and biological aspects of fungal pathogens, ultimately enhancing their virulence.

Structure and types of melanin

Melanin is a pigment that is found in various forms in different organisms, including fungi. Fungal melanins can be classified into different types based on their sources and chemical properties, such as eumelanin, pheomelanin, neuromelanin, allomelanin, and pyomelanin. These melanins share common characteristics, including broad optical absorption, resistance to strong acids, insolubility in most solvents, and stable free-radical populations.

In fungi, melanin biosynthesis can occur through various biochemical pathways or involve different precursor components. Some of the classes of fungal melanins that have been described include γ -Glutaminyl-3,4-Dihydroxy-Benzene (GDHB), L-DOPA, 1,8-Dihydroxynaphthalene (DHN), catechol-melanin, pnnnnnnnnn, paminophenol (PAP)-melanin, as well as heterogeneous melanins. Two recently reported melanins are the *Fusarium graminearum* periderm melanin and the Asp-melanin found in *Aspergillus terreus*. The melanin in the periderm of *Fusarium graminearum* perithecia is based on 5-deoxybostrycoidin, which is synthesized from the reaction of anhydrofusarubin derivatives and ammonia. This melanin contributes to the protective function of the perithecia. Asp-melanin is present in the conidial wall of *Aspergillus terreus*, and its precursor is 4-hydroxyphenylpyruvate. This precursor is oxidized to generate polymerized aspulvinone E derivatives, resulting in the formation of Asp-melanin [48]. These different types of melanins in fungi play important roles in various biological processes, including protection against environmental stresses, pathogenicity, and virulence [40].

- a. Eumelanin is a black-to-brown pigment found in human black hair and cuttlefish ink. It is formed by the oxidation of l-tyrosine or l-dopa through 5,6-Dihydroxy Indole (DHI) or 5,6-Dihydroxyindole-2-Carboxylic Acid (DHICA) [36].
- b. Pheomelanin consists mainly of sulphur-containing benzothiazine and benzothiazole derivatives. It is found in red hair, feathers, and freckles. L-cysteine is the chief source of sulphur, which is essential for the synthesis of pheomelanin [93].

- c. Neuromelanin is believed to be formed by the oxidative polymerization of dopamine or noradrenaline, possibly involving cysteinyl derivatives. It is mainly found in the human brain [38].
- d. Allomelanin is formed by the oxidation of polyphenols such as catechols and 1,8-dihydroxy naphthalene. It is often nitrogen-free and is found in most fungi [118].
- e. Pyomelanin is a dark-coloured pigment derived from Homogentisic Acid (HGA). It is found in fungal metabolites, specifically in *Pseudomonas* and *Aspergillus fumigatus*. Pyomelanin is an extracellular red-brown pigment [132].

Location and biosynthesis of melanin in the fungal pathogen

Melanin, a polymerized bio-pigment derived from indole or phenolic compounds, plays a crucial role in the virulence and survival of fungal pathogens. Although fungal melanin is formed inside the cell, it is transported to different parts and deposited with the assistance of specialized vesicles known as fungal melanosomes. The function of melanin varies depending on its deposition site, contributing to the organism's dominance. Additionally, the localization of melanin changes in different organisms as they progress through developmental stages. Melanin's presence in these locations provides structural support and protects fungal cells against environmental stresses and immune responses [30].

Fungal melanin is primarily found in the cell wall and plasma membrane. In the cell wall, it is either embedded within the wall's structure or forms the outermost layer. Melanin can also be present extracellularly, extending from the fungi's cell wall, known as cell wall-bound melanin. Studies have revealed that fungal cell wall melanin can be either granular or fibrillar in nature. For example, *Cryptococcus neoformans* has been observed to have a complex layer of granular melanin, measuring 40-130 nm in diameter, in its cell wall, as demonstrated by scanning electron microscopy and atomic force microscopy [32]. *Verticillium* spp.'s microsclerotia are covered with a layer of both granular and fibrillar melanin [143]. *Phaeococcomyces* sp, a black yeast fungus, release cell wall melanin and extracellular melanin as granules.

Fungi produce heterogeneous vesicles containing lipids, carbohydrates, and proteins. These vesicles play a crucial role in localizing intracellularly produced melanin to the cell wall and extracellular space [112]. In *Cryptococcus neoformans*, increased vesicular secretion corresponds to increased virulence [111]. Laccase, a key component responsible for vesicular melanization, is loaded inside the vesicles and can be systematically trapped in the cell wall, forming layers of melanin. These melanin-loaded vesicles, referred to as fungal melanosomes, vary in size and number. *Candida albicans* has been found to have fungal melanosomes that contribute to their virulence and survival [136]. Evidence suggests that fungal melanization occurs in specialized vesicles analogous to mammalian melanosomes [33,37,41,136]. In the appressoria of *Colletotrichum lagenarium*, multiple melanin layers have been identified in the cell wall [123]. In the rice blast fungus *Magnaporthe grisea*, the melanin layer is localized just outside the plasma membrane of appressoria [56]. The deposition of melanin in the appressoria of *M. grisea* helps retain glycerol, generating high osmotic pressure, which aids in penetrating the host cell wall and establishing the fungi successfully [68]. Melanin lines the septa and outer walls of wild-type Altern.

Fungal melanin biosynthesis

Diverse types of melanin found in fungi, the two most significant variants are DHN-melanin and DOPA-melanin. DHN-melanin derives its name from the pathway intermediate, 1,8-dihydroxy naphthalene, while DOPA-melanin is named after one of its precursors, L-3,4-dihydroxyphenylalanine (Hamilton and Gomez 2002) [62].

The DHN-Melanin biosynthesis pathway

In certain plant-pathogenic fungi, such as *Colletotrichum lagenarium* and *Magnaporthe oryzae*, DHN melanin serves as a virulence factor. However, in other fungi, DHN melanin found on conidia, hyphae, and sclerotia primarily functions as a stress protectant [152]. The DHN-melanin biosynthesis pathway has been elucidated in *Verticillium dahlia* and *Wangiella dermatitidis* through genetic and biochemical evidence. This involved the identification of key pathway intermediates and shunt products using techniques like Thin Layer Chromatography (TLC) and High-Pressure Liquid Chromatography (HPLC) [49,142].

The production of DHN-melanin occurs through the pentaketide melanin pathway. The initial step involves the conversion of malonyl-CoA by Polyketide Synthase (PKS1) into the first detectable intermediate, 1,3,6,8-Tetrahydroxynaphthalene (1,3,6,8-THN) [1,44]. A specific reductase enzyme then reduces 1,3,6,8-THN to produce scytalone. Enzymatic dehydration of scytalone results in the formation of 1,3,8-trihydroxynaphthalene [4,5]. This tricyclic compound is further reduced by a reductase enzyme to form vermalone [8,125]. Vermalone, through subsequent dehydration catalyzed by scytalone dehydratase, leads to the production of 1,8-Dihydroxynaphthalene (DHN) [10]. Finally, the dimerization of 1,8-dihydroxynaphthalene and its polymerization by laccase results in the production of melanin [12].

It is important to note that the melanin biosynthesis pathway varies among different fungi, and several byproducts have been detected through TLC and HPLC methods. For instance, the reductase inhibitor tricyclazole causes the accumulation of flaviolin, a shunt product of 1,3,6,8-THN, and inhibits the production of another shunt product, hydroxyjugalone, from scytalone [125,126].

Numerous genes involved in fungal DHN-melanin synthesis have been identified. Scientists have discovered gene clusters responsible for encoding the enzymes involved in DHN-biosynthesis, such as in *Alternaria alternata* and *Aspergillus fumigatus*, where three and six genes were identified, respectively [15,72,76]. The first step in DHN-melanin synthesis, catalyzed by Polyketide Synthase (PKS), is coded by different genes in different fungi. For example, in *Aspergillus fumigatus*, the pksP gene codes for PKS, and a mutation in this gene results in pink-colored conidia, while the wild type produces grey-green conidia [76,129].

DOPA-Melanin biosynthesis

Bell and Wheeler proposed the biosynthetic pathway for fungal DOPA-melanin biosynthesis in 1986. The enzyme tyrosinase or laccase plays a propounding role in the pathway. They catalyse the hydroxylation of L-tyrosine or L-DOPA to dopaquinone which is the first intermediate in the pathway and is highly reactive in nature [98]. In the absence of thiols cyclisation of dopaquinone results in leucodopachrome which is then oxidised to form dopachrome. Dopachrome upon hydroxylation

and decarboxylation forms dihydroxyindoles which polymerise to yield DOPA-melanin [94,145]. This biosynthetic pathway for DOPA-melanin strongly resembles the pathway found in mammalian cells. DOPA-melanin of mammalian consists of both eumelanin which does not contain the thiol group and pheomelanin which contains the thiol group [95]. The DOPA-melanin produced by *Cryptococcus neoformans* contains only eumelanin not pheomelanin and only the enzyme laccase catalysed the initial step in the biosynthetic pathway [144,145].

Melanin in fungal virulence

The production of melanin has long been recognized as a virulence factor in both bacterial and fungal pathogens. It serves various functions that contribute to the pathogen's ability to cause disease. Melanin helps prevent plasmolysis, maintain membrane permeability, and sustain high internal solute concentrations, which are crucial for maintaining turgor pressure and cell integrity. In the context of plant pathogens, melanin has been identified as a virulence factor in several fungal species. Melanized strains of fungi exhibit enhanced penetration of host tissues, higher rates of colonization, and increased production of appressoria (in the case of certain fungi) compared to non-melanized strains. These characteristics contribute to the pathogen's ability to establish infection and cause disease in plants. Melanin also plays a role in evading host defense mechanisms. It provides protection against the host's immune responses by interfering with the recognition and response to Pathogen-Associated Molecular Patterns (PAMPs) by the host's pattern Recognition Receptors (PRRs). Additionally, melanin is resistant to defensive compounds produced by the host, such as phytoalexins, defensins, and antimicrobial peptides. It acts as a free radical scavenger, neutralizing Reactive Oxygen Species (ROS) produced by the host as part of its defense response. Melanin-coated spores of some fungal pathogens can evade recognition by the host's immune system and pathogenesis-related proteins, thereby facilitating infection [25,153].

Penetration into the plant host: Melanin plays a crucial role in the penetration of fungal pathogens into plant cells. Appressoria, the infection structures produced by many fungal pathogens, undergo melanization, which is necessary for their attachment to plant surfaces. During appressorium maturation, glycogen and lipids translocate from the conidium to the appressorium, and melanin aids in glycerol accumulation within the appressorium. This accumulation leads to the generation of high osmotic turgor pressure, allowing the penetration peg to puncture the epidermal cuticle and cell wall, facilitating entry into the plant [69,102].

Overcoming plant defense: Melanin also helps fungal pathogens in evading host immune responses. The synthesis of melanin in fungal pathogens like *Alternaria alternata* inhibits the recognition and response of the host defense system. Melanin interferes with the recognition of Pathogen-Associated Molecular Patterns (PAMPs) by host Pattern Recognition Receptors (PRRs), thereby impairing the activation of immune responses. Additionally, melanin is resistant to defensive compounds produced by plants, such as phytoalexins, defensins, and antimicrobial peptides. Melanin acts as an effective free radical scavenger, neutralizing Reactive Oxygen Species (ROS) generated during the defense response. It also exhibits superoxide dismutase and catalase activities, further contributing to ROS detoxification [2,23,101,119].

Associated with the production of hydrolytic enzymes: Melanin is associated with the production of hydrolytic enzymes in some fungal pathogens. Enzymes like ribonuclease, deoxyribonuclease, acid phosphatase, and phenoloxidase have been detected in extracellular melanoprotein secreted by the apple scab pathogen *Venturia inaequalis*. These enzymes interact with melanin in the cell walls and are believed to retain their activity. The concentration of melanin-retained hydrolytic enzymes may facilitate more intensive entry-lesion action, increasing the virulence of the pathogen [52].

Resistance to antagonistic organisms: Melanin provides fungal cells with resistance against antagonistic organisms. Melanized fungal cells are less susceptible to cell wall-degrading enzymes produced by environmental antagonists. The melanin content of fungal cell walls is inversely related to their susceptibility to enzymatic hydrolysis. Melanin-bound chitin, a component of fungal cell walls, is extremely resistant to enzymic degradation. The presence of melanin in fungal walls confers resistance to lysis by hydrolytic enzymes, possibly through sequestration of the enzymes on melanin or steric hindrance. This resistance allows melanized cells to survive longer and be more resilient to environmental challenges, contributing to their dominance in soil ecosystems [10,61,75].

Melanin beyond fungal virulence

The production of melanin in fungal pathogen acts as a protective mechanism against environmental stressors. Melanin acts as a physical barrier to protect pathogens from harmful Ultraviolet radiation (UV), extreme temperatures and oxidative stress [19]. This protection allows pathogens to survive and maintain virulence under adverse conditions.

Cell wall reinforcement: In plant pathogens like *Phytophthora infestans*, melanin deposition in the cell wall reinforces its structural integrity, maintaining cell shape and protecting against mechanical damage from host defense mechanisms [71,91]. Melanin also contributes to the impermeability of the pathogen's cell, making it more resistant to antimicrobial compounds produced by the host [26,50].

Photoprotection: Melanins also enable organisms to resist UV, solar, or gamma radiation. Organisms like *Monilinia fructicola* [108], *A. alternata*, and *Cladosporium* sp [90,151] produce melanins that absorb a wide range of the electromagnetic spectrum, preventing photo-induced damage [53]. Bacteria such as *Bacillus subtilis*, *Bacillus thuringiensis*, *Bacillus sphaericus*, and *Pseudomonas aeruginosa* produce melanin, which protects them against UV irradiation, hydrogen peroxide, pesticides, and oxygen fluctuations [24,55,109,114]. Melanin synthesis in *B. subtilis*, facilitated by the laccase enzyme CotA, is particularly important in providing resistance to injury [60].

Antioxidant: Melanins possess strong antioxidant properties and can scavenge radicals. They exhibit stable free radical behavior due to unpaired electrons, making them responsive to magnetic fields and paramagnetic in nature. Melanins can also participate in the oxidation or reduction of metals, and fungal melanins have been used as templates for synthesizing metal nanoparticles [6].

Energy harvesting: Fungal melanin exhibits the ability to absorb radiation energy and convert it into metabolic energy [27,110,131,133]. This phenomenon has been observed in studies where irradiation of melanotic fungi with gamma rays or UV/V radiation resulted in enhanced metabolic activity, in-

creased growth rates, and changes in cellular ATP levels [16]. The electroconductive properties of fungal melanin make it attractive for applications in bioelectronics and sustainable electronics [3,147]. Thus, fungal melanin serves as an energy-harvesting pigment and holds potential implications in various fields due to its ability to transduce radiation energy and its electroconductive characteristics.

Protection against heat and cold stress: Melanin plays a crucial role in thermoregulation and protection against heat stress in fungi. Melanin-deficient mutants of certain fungi, such as *Monilinia fructicola*, have shown increased susceptibility to high temperatures and other stressors [108]. Conversely, melanization has been found to enhance tolerance to both heat and cold stress in species like *Cryptococcus neoformans*, potentially by quenching heat-induced Reactive Oxygen species (ROS) or buffering heat flux [113]. Also, melanized endophytes associated with plants contribute to thermoregulation by dissipating heat and absorbing ROS [107]. Furthermore, the synthesis of melanin in fungi is temperature-regulated, highlighting its significance in fungal thermoregulation and stress response.

Metal binding: Fungal melanins possess metal-binding capacity and can bind various metals, including Cu²⁺, Ca²⁺, Mg²⁺, and Zn²⁺, triggering melanogenesis in certain species [20,85,88,149]. This metal-binding property allows fungal melanin to scavenge essential metals from rocks and environmental sources [39,45]. It has been suggested that melanin can protect fungi from heavy metal toxicity in some cases [47,54], although contradictory results have been reported [42]. The metal-binding capabilities of fungal melanin contribute to metal bioabsorption and potentially provide protection against heavy metal toxicity.

Resistance to mechanical and chemical stress: Melanin deposition also enhances cell strength, rigidity, and chemical stability in fungi [91]. It forms crosslinks with macromolecules near the cell wall, influencing cell permeability, turgor forces, and protection against chemical degradation and heavy metal toxicity [47]. Melanization also contributes to higher resistance against hydrolytic enzymes and osmotic stress [14]. However, certain melanolytic fungi can biodegrade melanin using enzymes such as manganese and lignin peroxidases [18,67,81,106]. Notably, certain mould species inhibit enzymatic degradation due to the presence of melanin, emphasizing the complex relationship between melanin, enzymatic degradation, and fungal species, and underscoring the diverse role of melanin in ensuring cell protection and stability.

Protection against desiccation: Additionally, fungal melanization plays a vital role in protecting and adapting to dry conditions in microorganisms such as *Cenococcum geophilum* and *Armillaria mellea* [130,151]. Inhibition of melanin synthesis increases susceptibility to osmotic stress and desiccation. It contributes to the absorption and retention of water, thereby controlling water balance and enhancing resistance to desiccation (Jastrzebska *et al.* 1996). Additionally, melanization alters the porosity of the cell wall, potentially influencing osmolyte exchange and reducing water loss [56,63,73]. Overall, fungal melanin serves as a survival mechanism and contributes to the ability of microorganisms to thrive in dry environments.

Cell development: Fungal melanin, while not essential for growth, plays a crucial role in normal cell development in various fungal species. It is particularly important in filamentous species that produce melanized structures such as appresso-

ria, sclerotia, conidia, and reproductive structures [10,19,42]. Melanin biosynthesis is associated with healthy conidiation and germination, and disruptions in melanin production can lead to alterations in conidial morphology, cell wall integrity, and surface morphology. Examples include *Pestalotiopsis* microspore [148], *Aspergillus fumigatus* [76,127], *Chaetomium globosum* [58], *Alternaria alternata* (Kawamura *et al.* 1997), and *Bipolaris sorokiniana* (Bashyal *et al.* 2010). Melanin can constitute a significant portion of the dry weight of a cell, such as up to 30% in the spores of *Agaricus bisporus* [104], representing a considerable allocation of material and energy resources. It provides mechanical and chemical resistance to fungal structures, which is crucial during specific developmental stages.

Melanin in some major fungal diseases

Black Sigatoka, caused by the fungus *Mycosphaerella fijiensis*, is a major threat to banana plantations, resulting in significant crop losses. This fungus exhibits a high level of genetic diversity, aggressiveness, and resistance to fungicides and Reactive Oxygen Species (ROS). In culture, the fungus produces a dark-green pigment on the surface of its colony. Interestingly, isogenic mutants of *M. fijiensis* with pink mycelium and reduced melanin production can still penetrate banana leaf tissue, but their infection is halted at an early stage. The appearance of necrotic lesions on leaves is believed to be a result of an overly sensitive defense response by the host plant [29]. It has been suggested that the DHN melanin produced by *M. fijiensis* acts as a virulence factor by generating singlet molecular oxygen, contributing to the pathogenicity of the fungus [11].

Apple scab is caused by the hemibiotrophic ascomycetous fungus *Venturia inaequalis* (Cooke) G. Winter. This pathogen infects apple trees and grows as subcuticular hyphae, deriving nourishment from the host tissue beneath them [9]. When conidia germinate on the leaf surface, they pierce the leaf cuticle and form subcuticular mycelia and stromata. *Venturia inaequalis* produces a dark brown ring structure called a Melanized Appressorial Ring Structure (MARS) at the base of its appressoria, which acts as a sealing ring adhered to the leaf surface. Melanin is deposited in the outer layer of the cell wall of conidia, conidiophores, and the appressorial ring structure. The synthesis of melanin is associated with cell wall stiffness, the aggressiveness of the fungus towards the host plant, and its sensitivity to various xenobiotics. Melanin-deficient mutants of *Venturia inaequalis* exhibit decreased cell wall stiffness, reduced aggression towards the host plant, and increased sensitivity to xenobiotics [122].

Take-all, a root disease affecting wheat and barley in temperate zones, is caused by the fungus *Gaeumannomyces graminis* var. *tritici*. This fungus produces DHN melanin and initiates root infection through melanized ectotrophic "runner" hyphae. The runner hyphae must be melanized to anchor themselves or produce invasive infection hyphae [30]. Melanin deposition in the fungal cell wall is related to the production of intracellular pressure. Melanized wild-type hyphopodia of *G. graminis* var. *graminis* generate significantly higher turgor pressures (1.22 MPa) compared to nonmelanized hyphopodia strains (0.04 MPa). The increase in pressure during hyphopodial development and pigment deposition supports the association between melanin and hyphopodial turgor. Melanization of the cell wall also contributes to its rigidity [91].

Anthracnose, caused by pathogenic fungi such as *Colletotrichum lindemuthianum* and *C. lagenarium*, requires DHN mel-

anin for the appressoria to penetrate and establish within plant cells. In *C. graminicola*, melanin is synthesized through the pentaketide pathway via the polyketide synthase 1 (CgPKS1) gene, which shares similarities with fungal polyketide synthases involved in the synthesis of 1,3,6,8-tetrahydronaphthalene, an intermediate in melanin biosynthesis. Nonmelanized appressoria of *C. graminicola* are sensitive to externally applied cell-wall-degrading enzymes, while melanized appressoria are not affected. Melanin is not necessary for turgor generation but enhances cell-wall rigidity in appressoria of the corn pathogen *Colletotrichum graminicola* [83].

Rice blast, caused by the fungus *Pyricularia oryzae*, poses a significant threat to global rice and wheat production. Conidiation and appressorium formation are crucial steps in the infection cycle of this pathogen. Blast disease spreads among plants through conidia, and the fungus infects plants via appressoria. The DHN melanin layer acts as an impermeable barrier for appressoria, allowing them to generate the high turgor pressure required for penetration through the plant cuticle, enabling the infection peg of *P. oryzae* to enter the plant fully [56].

Grey mould, caused by *Botrytis cinerea*, is a significant fungal plant pathogen that affects over 200 plant species worldwide, leading to grey mould disease. Due to melanin accumulation in its tissues, this fungus typically produces grey-coloured mycelia, conidia, and sclerotia [22]. Melanin, specifically 1,8-dihydroxynaphthalene (DHN) melanin, is deposited on the cell walls of conidia, hyphae, and sclerotia. In *B. cinerea*, DHN melanin primarily functions as a stress protectant rather than a pathogenicity or virulence factor [116].

Black rot, caused by *Phyllosticta* spp, poses a threat to grapevine and citrus production. All *Vitis vinifera* cultivars are highly susceptible to black rot. This polycyclic disease involves repeated cycles of primary and secondary infections [150]. Melanin-dependent increase in the stiffness of the appressorium cell wall in *Phyllosticta* spp. contributes to their resistance to collapsing during periods of desiccation and osmotic imbalance [74].

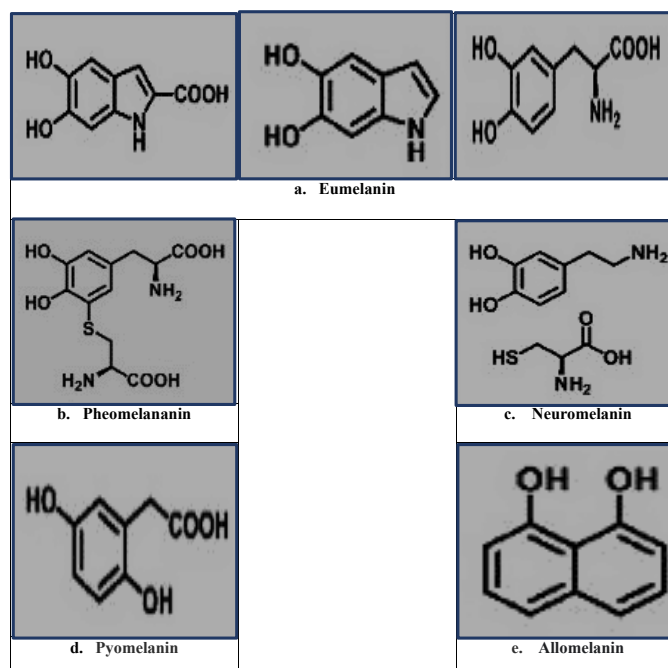


Figure 1: The chemical structure of different types of melanin.

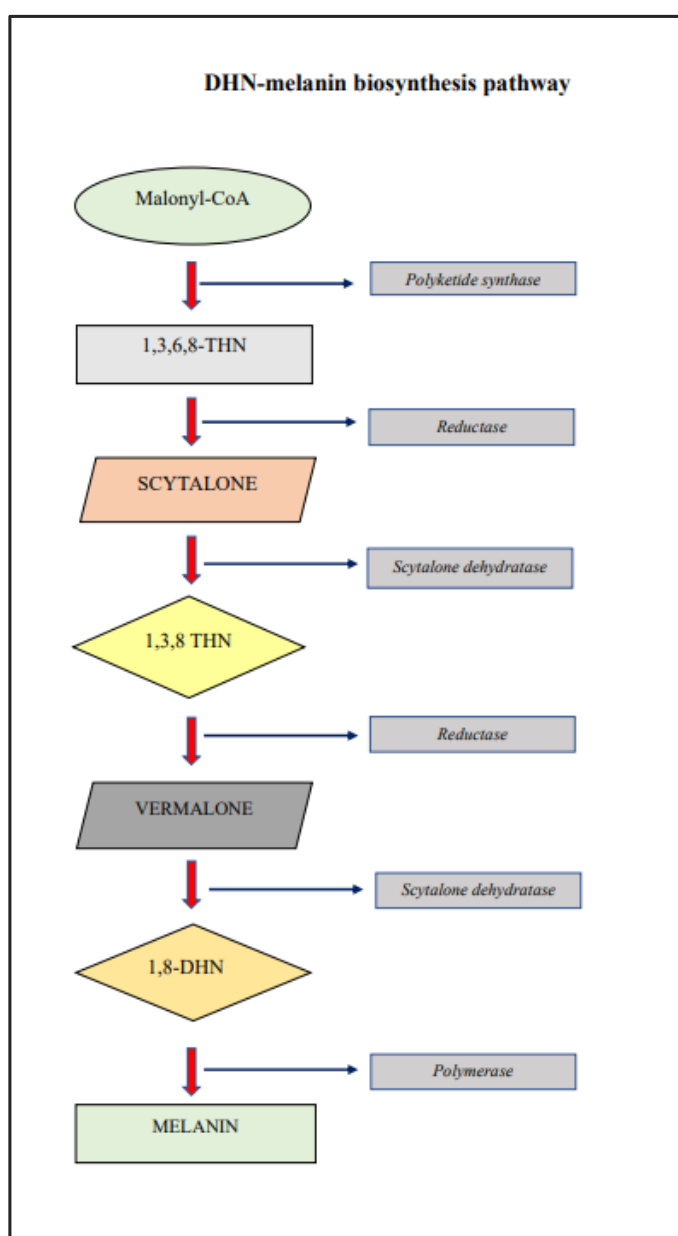


Figure 2: DHN- Melanin biosynthesis pathway.

Conclusion

Melanin plays a significant role in plant pathogenic fungi, contributing to their pathogenicity and survival. Melanin production in these fungi provides protection against various environmental stresses, including UV radiation, oxidative stress, and host defense mechanisms. Additionally, melanin can enhance fungal virulence by promoting adhesion, penetration, and evasion of the host immune response. The synthesis and regulation of melanin in plant pathogenic fungi are complex processes influenced by genetic factors and environmental cues. Understanding the mechanisms underlying melanin production in these fungi can provide valuable insights into developing effective strategies for controlling plant diseases. Further research is warranted to unravel the specific functions and interactions of melanin in plant-fungal interactions, paving the way for the development of targeted management approaches in agriculture.

Author statements

Data Availability Statement

All the required data are included in the article, further inquiries can be directed to the corresponding author.

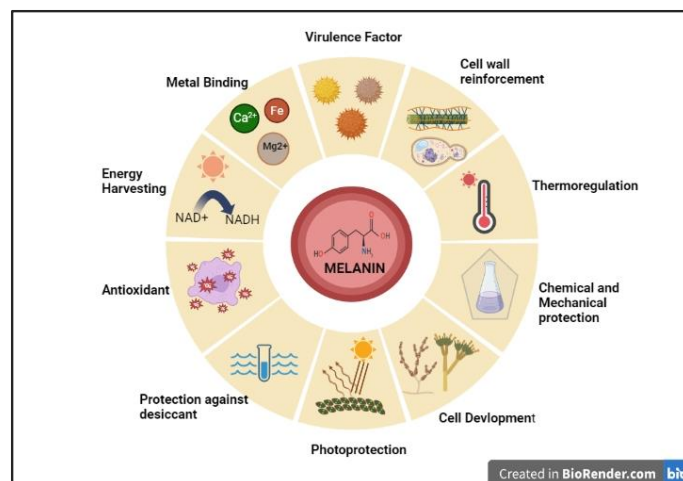


Figure 3: Diverse functions of fungal melanin.

Author contributions

All the authors contributed equally to the article and approved the submitted version.

Conflict of interest

The authors declare that there is no potential conflict of interest.

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