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Review

# Diversity, stability and applications of mycopigments

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ABSTRACT

The role of fungi as major pigment producers in the environments has endeared their application as sources of industrially important pigments. Compared to synthetic colorants, fungal pigments are rapidly becoming the preferred choice due to their biodegradability, eco-friendliness and versatility. Besides their uses as colourants, their functions as preservatives and/or bioactive agents have promoted their potential across numerous industries. In the past, more focus has been placed on enhancing the production levels of fungal pigments with little attention to the stabilization of the pigments and other important areas of concern. To this end, this review draws attention to the diverse classes of fungal pigments with emphasis on their existing and future applications, especially in the food and textile industries. Emphasis was also placed on the factors affecting fungal pigment stability and the techniques to efficiently circumvent the instability. Finally, the application of emerging technologies such as copigmentation, microencapsulation, metabolic engineering, and chemo-informatics tools in enhancing the mycopigment industry are highlighted.

## 1. Introduction

The use of colorants dates back to prehistoric times and to the beginning of ancient civilization where they were used as symbols of art and the heritage of human civilization [1]. More recently, the advent of synthetic pigments has promoted the usage of colorants in different industries, hence, they constitute a significant share of the global pigment market [2]. In the food industry for instance, synthetic pigments are added to food products to either enhance the natural colour or restore the colour lost during processing [3]. However, there has been a decline in the popularity of synthetic pigments in recent times, which is primarily due to their negative impacts on human health and the environment. For example, azo dyes, which are mostly used in the food industry, have been identified as potential carcinogens [4]. In other instances, the intake of ponceau, tartrazine and sunset yellow- all permitted synthetic food colours - has been associated with allergic reactions such as urticaria, dermatitis and asthma, even at low levels [5]. Similarly, in the textile industry, synthetic dyes have been noted to produce toxic wastes, contaminate surrounding water bodies, reducing water quality and affecting both aquatic and terrestrial lives [6]. Studies have also shown that exposure to textile dyes, especially through oral ingestion or inhalation, may lead to allergies, cancers, irritations and toxicity in humans [7,8]. In general, toxic wastes generated from different dyes accumulate and persist in different ecosystems for years, causing irreparable damage [9]. Besides the health and environmental damage that has been ascribed to dyes, they also cause significant economic loss due to the expensive operational costs associated with the detoxification of dye-contaminated effluents, especially in textile processing [10].

Against this background, there have been calls for the ban of artificial pigments and a concomitant accelerated search for safer alternatives such as natural pigments. Nature is widely regarded as a very rich store of natural pigments that may also represent useful leads in the development of novel cosmeceutical, nutraceutical, as well as pharmaceutical agents [11,12]. Furthermore, naturally derived pigments from plants, animals and microorganisms are considered safer alternatives to synthetic pigments, triggering consumer preference as well as both industrial and research curiosity [13]. To this end, the demand for natural

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colorants has increased considerably, with its market value expected to reach approximately $1.62 billion by 2023 [14]. Interestingly, microbial-based pigments, especially from bacteria and fungi, have gained more prominence due to their ability to grow under controlled conditions and with limited resources as well as their ease of handling, thus resulting in high productivity without any seasonal limitation [15]. In addition, the genetic diversity in microorganisms has been linked to their ability to produce a plethora of pigments with high yields [16]. Typically, most fungi produce pigments in fulfilment of their ecological functions and in response to prevailing environmental conditions [17]. It was observed that fungal pigment production has a lot of advantages over other natural sources and these include; the ability to produce a wide spectrum of soluble pigments, higher production yields, and relative ease of production optimization [18]. Fungal pigments also exhibit different biological activities which range from antimicrobial, to anti-proliferative [19], metal chelating [20], as well as antioxidant properties [21]. All of these make fungal pigments economically viable and industrially sought after.

Fungi as sources of natural pigments differ widely in terms of their habitats (Fig. 1), a phenomenon which is also directly linked to their various genetic contents, morphologies, taxonomy as well as their ecological roles [22]. In this regard, they are classified according to their various habitats such as halophilic (e.g., *Wallemia ichthyophaga*) in saline environments [23,24], acidophilic (e.g., *Acidomyces acidophilus* and *Acidella bohemica*) in acidic environments, psychrophilic (e.g., *Rhodotorula frigidialcoholis* and *Scelerotina borealis*) in cold environments [24], as well as the thermophilic (e.g., *Thermothelomyces hinnuleu* [25], and *Thermomyces lanuginosus* [26] in regions of high temperature. A significant relationship has since been established between the remarkable survival/tolerance of such fungi in their respective habitats and the huge array of their biosynthesised pigments [27,28]. Furthermore, molecular studies have shown that the regulation of pigment metabolic pathways - such as the melanin-related gene clusters - is largely responsible for the environmental adaptation of different melanin-producing fungi [29]. Fungal pigment biosynthesis has also been noted to be one of the most important protective and coping mechanisms; for example, in some psychrophilic fungi, melanin together with mycosporines and carotenoids protect against desiccation, ionizing radiation, oxidizing agents, and UV radiations [30].

However, despite the progress made in the field of fungal pigments, increasing their production level to a commercial scale is a huge challenge [31]. Furthermore, their low stability, possible contamination (especially by mycotoxins) and undesirable interactions with food matrices have been noted as clogs in the wheels of the industrial growth of fungal pigments. Thus, the optimization of the production and extraction processes has been identified as key in enhancing the utilization and commercialization of fungal pigments [32]. In this regard, this review aimed to draw scientific attention to the immense potential of fungal pigments, by highlighting the diverse nature of the pigments and their sources, their wide array of bioactivities, as well as their current applications in the industry. Furthermore, stabilization techniques to enhance their industrial applications, effective methods aimed at increasing productivity, and areas of future research are also part of the discourse. It is believed that the information presented therein will propel the utilization and commercialization of this class of pigments.


**Fig. 1.** Pigment producing fungi in their diverse ecology (Adapted from Kalra (2020) with modifications.

## 2. Chemical classification of fungal pigment

Fungal pigments are broadly classified based on the biosynthetic pathways as either polyketides or carotenoids. However, they are more specifically categorized into five chemical groups, *viz*., melanins, azaphilones, flavins, phenazines, and quinones (Table 1) [33–35]. The polyketides comprise tetraetides and octaetides, which form the polyketide chain via C2 units, for example, the *Monascus* sp. pigments [36]. On the other hand, carotenoids are made up of terpenoids, which have a forty-carbon backbone in their main chain, e.g. *Neurospora intermedia* pigments [37,38]. However, it is interesting to note that *Fusarium* sp. produces pigments via both the polyketides (e.g., the polyketide-derived pigment) and carotenoids (e.g., neurosporaxanthin) pathways [39].

### 2.1. Polyketide pigments

Polyketides are a major class of secondary metabolites synthesized by microorganisms via a complex multi- enzymatic system consisting of various polyketide synthases (PKSs). Several of these polyketide pigments possess biological activities, making them valuable replacements for chemotherapeutic agents as well as potential adjuncts [40,41]. The most important fungi polyketide pigments are melanins, ankaflavin, anthraquinone, flavins, naphthoquinone and quinones [33,41], however, melanin is noted to be the most prominent being found throughout nature. Melanin is a group of dark brown or black coloured pigments that are mainly negatively charged, hydrophobic and possess high molecular weights. Melanisation is considered a survival strategy for many organisms, especially those inhabiting unfavourable environmental conditions, thus, the pigments are synonymous with antioxidant and radical scavenging activities [42]. Furthermore, melanins have also been shown to be photo-protective [43] as well as metal chelating [44,45]. Melanin synthesis in some ascomycetes, such as *Aspergillus nidulans*, *A. niger*, *A. tamari*, *A. flavus*, *Cladosporium resinae*, *Epicoccum nigrum*, *Hendersonula toruloidea*, *Eurotium echinulatum*, *Holothuria grisea* and *Helichrysum archeri* has been observed to be via the 3,4-dihydroxyphenylalanine (DOPA) pathway [46]; while in *Spissiomyces endophytica* it is via the 1,8-dihydroxynaphthalene (DHN)-pathway [47]. However, *Talaromyces marneffei* has been shown to synthesize both DOPA- and DHN-melanin, depending on the growth conditions and available precursors [48].

On the other hand, azaphilones have a highly oxygenated pyranoquinone bicyclic core, usually known as isochromene, as well as a

**Table 1**
Classification of mycopigments.

| Mycopigment | Subclass | Biosynthetic pathway | Reference |
|---|---|---|---|
| Polyketides | | | |
| α-2,5-trihydroxyacetophenone | Hydroquinones | Polyketide pathway | Sun, Huo, Kurtán, Mándi, Antus, Tang, Draeger, Schulz, Hussain and Krohn[67] |
| 1,8-dihydroxynaphthalene (DHN) melanin | Melanin | 1,8-dihydroxynaphthalene (DHN) and the L-3,4-dihydroxyphenylalanine (L-DOPA) pathway | Antipova, Zhelifonova, Zaitsev and Vainshtein[68] |
| 29-methyl ether fusarubin | Naphthoquinones | Polyketide pathway | Eman Mostafa[69] |
| Alterporriol K and L | Anthraquinones | Polyketide pathway | Kanno, Tsurukawa, Kamisuki, Shibasaki, Iguchi, Murakami, Uchiyama and Kuramochi[70] |
| Anhydrojavanicin | Naphthoquinones | Polyketide pathway | Britton[71] |
| Atrorosin | Azaphilones | Polyketide pathway | Caro, Venkatachalam, Lebeau, Fouillaud and Dufossé [72] |
| Aurofusarin | Anthraquinones | Polyketide pathway | Vitale, Coppola, Palma Esposito, Buonocore, Ausuri, Tortorella and de Pascale[61] |
| Catenarin | Anthraquinones | Polyketide pathway | Caro, Fouillaud, Laurent and Dufossé[73] |
| Chrsophanol | Hydroxyanthraquinones | Polyketide pathway | Jiang, Li, Zhang, Jin, Yu, Fang and Wu[74] |
| Citrinin | Azaphilones | Polyketide pathway | Isbrandt, Frisvad, Madsen and Larsen[54] |
| Coniothyrinone A – D | Hydroxyanthraquinones | Polyketide pathway | Jiang, Li, Zhang, Jin, Yu, Fang and Wu[74] |
| Dihydroanhydrojavanicin | Naphthoquinones | Polyketide pathway | Britton[71] |
| Erythroglaucin | Anthraquinones | Polyketide pathway | Caro, Fouillaud, Laurent and Dufossé[73] |
| Eumelanin | Melanin | 1,8-dihydroxynaphthalene (DHN) and the L-3,4-dihydroxyphenylalanine (L-DOPA) pathway | Bayram[75] |
| Glutaminylhydroxybenzene (GHB) | Melanin | γ-L-glutaminyl-4-hydroxybenzene pathway | Bayram[75] |
| Javanicin, | Naphthoquinones | Polyketide pathway | Britton[71] |
| Monasfluores | Azaphilones | Polyketide pathway | de Oliveira, Rocha, Pinto, Ventura, Dos Santos, Crevelin and Ebinuma[76] |
| N-glutaryl-monascorubramine | Azaphilones | Polyketide pathway | Christiansen, Isbrandt, Petersen, Sondergaard, Nielsen, Pedersen, Sørensen, Larsen and Frisvad[77] |
| N-glutaryl-rubropunctamine | Azaphilones | Polyketide pathway | Christiansen, Isbrandt, Petersen, Sondergaard, Nielsen, Pedersen, Sørensen, Larsen and Frisvad[77] |
| Pestalotioquinols A and B | Hydroquinones | Polyketide pathway | Liu, Yan, Li, You and She[78] |
| Pyomelanin | | 1,8-dihydroxynaphthalene (DHN) and the L-3,4-dihydroxyphenylalanine (L-DOPA) pathway | Antipova, Zhelifonova, Zaitsev and Vainshtein[68] |
| Rubrocristin | Anthraquinones | Polyketide pathway | Caro, Fouillaud, Laurent and Dufossé[73] |
| Solaniol | Naphthoquinones | Polyketide pathway | Britton[71] |
| Talanaphthoquinones A and B | Naphthoquinones | Polyketide pathway | Britton[71] |
| **Carotenoids** | | | |
| α, β-carotenes | Carotenes | Mevalonate (MVA) pathway | Christiansen, Isbrandt, Petersen, Sondergaard, Nielsen, Pedersen, Sørensen, Larsen and Frisvad[77] |
| Astaxanthin | Xanthophylls | Mevalonate (MVA) pathway | Wang, Zhang, Chen, Sun, Yan, Shen and Yuan[79] |
| Cantaxanthin | Xanthophylls | Mevalonate (MVA) pathway | Yang, Zhang and Tsao[80] |
| Fucoxanthin | Xanthophylls | Mevalonate (MVA) pathway | Yang, Zhang and Tsao[80] |
| Lutein | Xanthophylls | Mevalonate (MVA) pathway | Charalampia and Koutelidakis[81] |
| Lycopene | Carotenes | Mevalonate (MVA) pathway | Morales-Oyervides, Oliveira, Sousa-Gallagher, Méndez-Zavala and Montañez[82] |
| Neoxanthin | Xanthophylls | Mevalonate (MVA) pathway | Yang, Zhang and Tsao[80] |
| Pirilloxanthin | Xanthophylls | Mevalonate (MVA) pathway | Yang, Zhang and Tsao[80] |
| Violaxanthin | Xanthophylls | Mevalonate (MVA) pathway | Charalampia and Koutelidakis[81] |
| Zeaxanthin | Xanthophylls | Mevalonate (MVA) pathway | Charalampia and Koutelidakis[81] |

quaternary carbon centre [49,50] and they are biosynthesized by numerous ascomycetes species, including *Aspergillus, Penicillium, Chaetomium, Talaromyces* and *Xylariaceae* [51,52]. The monascus pigments are a well-studied azaphilones subgroup which includes the important industrial pigments, citrinins [53] and, a group of red-pigmented compounds produced by the filamentous *Aspergillus neoglaber*, the atrorosins [54]. The azaphilone, hypocrellone A, produced by *Hypocrella* sp., was reported to have applications in natural medicine and healthy foods [55]. Similarly, the quinones are a group of organic compounds formed through the rearrangement of double carbon bonds to produce a fully conjugated cyclic dione structure and are further grouped based on their aromatic carbon skeleton into benzoquinones, naphthoquinones and anthraquinones [49]. Specifically, anthraquinones are regarded as one of the most prominent both in nature and in the industry, making the red-coloured anthraquinones from *Penicillium oxalicum* very popular [56]. In addition, some strains of *Trichoderma* such as *T. aureoviride, T. harzianum, and T. polysporum* are known to produce the orange-red anthraquinone pigment, chrysophanol, which is a very potent food colorant [57]. Other anthraquinone red pigments, such as catenarin and erythroglaucin have been isolated from strains of *Drechslera* species and *Curvularia lunata*, respectively [56] while some *Aspergillus* species were also found to synthesize the red pigments, erythroglaucin, catenarin, and rubrocristin [56,58]. The anthraquinones are also noted for their bioactivities; a notable example is aurofusarin, a mycotoxin which inhibits the growth of some molds and yeasts [59,60].

### 2.2. Carotenoids

Carotenoids are terpenoid-based pigments synthesized via the isoprenoid pathway from isopentenyl pyrophosphate (IPP), a 5-carbon precursor derived from the parent compound, mevalonate [61]. Based on their degree of oxygenation, carotenoids are classified as either carotenes, which are oxygen-free carotenoids (α-carotene, β-carotene) or xanthophylls, the oxygen containing carotenoids (astaxanthin, lutein, zeaxanthin) [21,62,63]. The presence of an aliphatic polyene chain which comprises eight isoprene units alongside light-absorbing conjugated double bonds, provides their characteristic orange, yellow, and red colours [61]. In general, carotenoids are largely useful in both folk and modern medicine; as nutraceuticals, supplements, feed and food colours in the food industry, as well as cosmetics, perfumes, and dyes for clothing [21,63]. Carotenoids act as antioxidants against free radicals and photoreceptors against lethal or strong light intensities and radiations and maintain membrane fluidity and stability [21]. Furthermore, carotenoids act as precursors of vitamin A and as an antioxidant in the management of several human diseases [11,21]. Astaxanthin, a xanthophyll carotenoid, has been described as one of the most powerful antioxidants due to its biochemical structure and remarkable radical scavenging activity [64]. Various studies have shown the production of carotenoids by fungal species, including yeast. Specifically, β-carotene production has been described extensively, in basidiomycetes, such as *Rhodosporidium* sp., *Sclerotium rolfsii*, *Sclerotinia sclerotiorum*, *Sporidiobolus pararoseus* [65] as well as in ascomycetes such as *Aschersonia aleyroides, Aspergillus giganteus*, *Cercospora nicotiana* and *Penicillium* sp. [66]. In addition, yeasts such as *Phaffia rhodozyma* as well as *Rhodosporidium*, *Rhodotorula*, *Sporobolomyces*, and *Sporidiobolus* species, have been described as notable producers of carotenoids [63].

## 3. Current and potential applications of mycopigments

The relevance of fungal pigments in various industries has been attributed to their high yields, low production cost, pigment characteristics, and stability with respect to environmental factors such as temperature and light [19,82]. For example, the utilization of agricultural residues as low-cost substrates for fungal growth has led to significant increases in mycopigments production, decreasing the production cost while also addressing waste disposal issues [83,84]. Thus, the applications of fungal pigments transverses the food and beverage, textile, pharmaceutical, cosmetic, bioremediation, as well as the electronics industry.

### 3.1. Food and beverage industry

Natural food colours add an appetizing appeal to food products when used as additives and also impart nutritional benefits to consumers, both humans and animals alike [85,86]. More specifically, the consumption of these natural-coloured compounds, which typically possess significant antioxidant and free radical scavenging ability, has been associated with different health benefits such as the amelioration of disease conditions such as obesity, inflammation, diabetes, and tumours [87]. Carotenoids, for example, have been described to alleviate age-related diseases, such as cataracts and muscular degeneration, reduce the incidence of coronary heart disease and carcinomas, including lung, breast, prostate, and colorectal cancers [88]. In the last few decades, different fungal pigments have been studied and identified for their usefulness as antioxidants, natural colorants, preservatives, as well as active and smart packaging components (Table 2).

### 3.2. Textile industry

The textile industry is considered the largest consumer of synthetic organic pigments and dyes [34], and this consumption has various negative impacts on human health and the environment in general. In different parts of the world, effluents from this industry, which are laced with used synthetic dyes, cause irreparable contamination of nearby water bodies or leach into underground water systems [89,90], posing both direct and indirect health risks to all life forms [91,92]. As a result, mycopigments, along with other natural pigments are currently being explored as replacements for synthetic dyes in the colouring of silk, wool, leather, cotton and other textile materials (Table 2). As pointed out earlier, mycopigments are eco-friendly, affordable and easy to produce; in addition, they are also stable, consistent, biodegradable and exhibit excellent colour fastness [61]. All of these properties have promoted the use of carotenoids, flavins, melanins, phenazines, quinones, monascins, violaceins -all fungal dyes- as eco-friendly alternatives in the textile industry [93,94]. For example, the use of *Monascus* pigments as textile dyes have been propagated by their unique functional properties, enhanced biodegradability, and better compatibility with the environment [95], while the anthraquinones' antimicrobial properties have been a major selling point [96].

### 3.3. Other applications

Besides their wide applications in the food and textile industries, mycopigments have also been shown to be quite useful in many other industrial processes, as shown in Fig. 2 and highlighted in Table 2. For example, in bioremediation, fungal melanin has been useful as an eco-friendly biosorbent to remove divalent metal ions contaminated effluents due to the presence of its different functional groups which enable metal ion adsorption [97]. As expected, the biocompatibility of melanin, its cost-effectiveness and its reusability without any excessive degradation along with its reduction in adsorption potential, are the reasons for its significant functionality in bioremediation [98]. In the medical field, it is also used as a powerful antimicrobial agent, a component of effective anticancer therapy [99], and as a hypoglycaemic agent [100]. Similarly, mycopigments are applied as printing inks, paints, and coating agents in the printing industry as chromophores exhibit remarkable chemical light, and thermal stability [34]. Notable examples in this regard include the decolorable ink which contains *Monascus* pigments [101] and the red crystallizing pigment from *Scytalidium cuboideum*, which have both been noted to possess remarkable potential in inkjet printing. In addition, in opto/electronics, fungi-derived pigments are now being explored in producing organic semiconductors,

**Table 2**
Industrial application of mycopigments.

| Classes | Pigment | Pigment producing fungi | Function/Bioactivity | Reference |
|---|---|---|---|---|
| Food/beverage | | | | |
| Anthocyanin | Aspergiol A, aspergiol B, averythrin, averantin, and methylaverantin | *Aspergillus versicolor* | Colorant in dairy products (cream cheese, fermented milk, milkshakes), low-pH beverages, solid food matrices (pancakes and omelettes) | Pineda-Vadillo, Nau, Guerin-Dubiard, Jardin, Lechevalier, Sanz-Buenhombre, Guadarrama, Tóth, Csavajda and Hingyi[108] |
| Anthraquinone | Anthraquinone and cinnamic acid | *Penicillium chrysogenum* | Anticancer and antimicrobial agent | Pagano and Dhar[109] |
| Anthraquinone | Red | *Penicillium oxalicum* | Food grade colorant | Caro, Anamale, Fouillaud, Laurent, Petit and Dufossé[110] |
| Anthraquinone | Yellow | *Thermomyces* sp. | Colorant in cookies, rice, wine orange, squash, guava jelly | Poorniammal and Gunasekaran[111] |
| Anthraquinone | Yellow | *Thermomyces* sp. | Colorant in dairy products. | Poorniammal, Gunasekaran and Murugesan[112] |
| Anthraquinone | Yellow virone | *Trichoderma virens* | Antifungal agent. | Kamala, Devi, Sharma and Kennedy [113] |
| Azaphilone | Azaphilone - Violet, PP-V | *Aspergillus niger* | Colorant in cookies and lemon juice | Toma, Nazir, Mahmud, Mishra, Ali, Kabir, Shahid, Haque, Siddique and Alim[114] |
| Carotenoid | Yellow-tinted riboflavin | *Ashbya gossip* | Food preservative | Tuli, Chaudhary, Beniwal and Sharma [11] |
| Carotenoid | Orange carotenoid/antibiotic | *Penicillium* sp. GBPI_P155 | Antibacterial agent | Pandey, Jain, Pandey and Tamta[115] |
| Carotenoid | Orange colored carotenoid | *Penicillium* sp. GBPI_P155 | Food colorant | Pandey, Jain, Pandey and Tamta[115] |
| Carotenoid | Fucoxanthin, Violaxanthin | *Penicillium purpurogenum* (SR2) | As fish feed coat and fed to Golden Koi (*Cyprinus carpio*) fish | Patil and Thakare[116] |
| Carotenoid | Carotenoid | *Rhodosporium paludigenum*, and *Rhodotorula glutinis* | Food and beverages colorant. | Ramesh, Vinithkumar, Kirubagaran, Venil and Dufossé[117] |
| Carotenoid | Carotenoid | *Sporidobolus salmoncolor* | Colorant in confectionaries, fruits, breakfast cereals, pasta, sauces, processed cheese, vitamin-enriched milk products, energy drinks and beverages | Narsing Rao, Xiao and Li[34] |
| Carotenoid | Canthaxanthin | *Stemphylium lycopersici* | Inhibition of the oxidation of lipids in liposomes | Gajalakshmi[118] |
| Polyketide | Orange monascorubrin | *Monascus ruber* CCT 3802 | Antimicrobial agent against *S. aureus*, *E. coli*, and *S. enteritidis* | Vendruscolo, Tosin, Giachini, Schmidell and Ninow[119] |
| Polyketide | Red monascorubramine | *Monascus ruber* CCT 3802 | Antimicrobial agent against *S. aureus* ATCC 25923 and *E. coli* ATCC 25922 | Vendruscolo, Tosin, Giachini, Schmidell and Ninow[119] |
| Polyketide | Red mitorubrinol | *Penicillium purpurogenum* | Antimicrobial agent against *P. aeruginosa* and *S. aureus* | Patil, Sivanandhan and Thakare[120] |
| Polyketide | Violet, PP-V | *Penicillium purpurogenum* IAM15392 | Food colorant | Kojima, Arai, Matsufuji, Kasumi, Watanabe and Ogihara[121] |
| Polyketides | Dark brown to black melanin | *Agaricus bisporus* | Incorporated into gelatin coatings for packaging and oxidative stability of pork lard | Łopusiewicz, Jędra and Bartkowiak [122] |
| **Textile** | | | | |
| Anthraquinone | Yellow brown | *Aspergillus* sp. MBYP1 | Dyeing of cotton and silk. | Pandiyarajan, Premasudha and Kadirvelu[123] |
| Anthraquinone | *Yellow pigment, asperyellone* | *Aspergillus niger sp. AN01* | Dyeing of silk and wool fabric | Iswarya, Shanuja, Giri Dev and Gnanamani[124] |
| Anthraquinone | Reddish brown | *Alternaria alternata* | Potent antimicrobial property against *S. epidermis* and *S. pyogenes* | Devi and Karuppan[125] |
| Anthraquinone | Yellow pigment | *Thermomyces* sp. | Antimicrobial agent and natural mordant in silk fiber | Poorniammal, Gunasekaran and Murugesan[112] |
| Carotenoid | Blue pigment, indigoidine | *Rhodosporidium toruloides* | Potential dyeing agent | Wehrs, Gladden, Liu, Platz, Prahl, Moon, Papa, Sundstrom, Geiselman and Tanjore[126] |
| Phenoxazine | Orange - 1,2-dimethoxy-3 H-phenoxazin-3-one | *Gonatophragmium triuniae* | Antioxidant, antibacterial, and dyeing potential | Lagashetti, Dufossé , Singh and Singh [2] |
| Polyketide azaphilone | Acid yellow dye | *Penicillium minioluteum* | Dyeing of wet blue goat nappa skin | Gupta, Tyagi, Agarwal, Singh, Chaudhary, Harit and Kushwaha[127] |
| Polyketide azaphilone | Red pigment | *Penicillium purpurgenium* | Dyeing of wool fibre | Ali and Abd-Elsalam[128] |
| Polyketide azaphilone | Red | *Talaromyces verruculosus* | Dyeing of cotton fabric | Chadni, Rahaman, Jerin, Hoque and Reza[93] |
| **Other applications** | | | | |
| Hydroxyanthraquinone | Emodin, dermocybin, dermorubin | *Dermocybe sanguinea* | Hair dye component | Aishwarya[129] |
| Polyketide | Dark brown to black melanin | *Amorphotheca resinae* | Biosorbent for treatment of Cu (II) and Pb (II) metal-contaminated effluents | Oh, Kim, Kim, Kim and Kim[97] |
| Polyketide | Dark brown to black melanin | *Talaromyces amestolkiae*, *Penicillium citrinum* | Biosorbent for remediation of uranium-contaminated sites | Coelho, Reis, Cotrim, Mullan and Corrêa[107] |
| Polyketide | Dark brown to black melanin | *Sporisorium reilianum* | Hypoglycaemic agent | Lu, Yu, Shi, Ma, Fu, Meng and Shi[100] |
| Quinone | Red crystallizing pigment | *Scytalideum cuboideum* | Inkjet printing on textiles | Vega Gutierrez, He, Cao, Stone, Walsh, Malhotra, Chen, Chang and Robinson [130] |

(*continued on next page*)

**Table 2** (*continued*)

| Classes | Pigment | Pigment producing fungi | Function/Bioactivity | Reference |
|---|---|---|---|---|
| Food/beverage | | | | |
| Quinone | Xylindein | *Chlorociboria aeruginosa* | Semi-conductors design | Giesbers, Van Schenck, Quinn, Van Court, Vega Gutierrez, Robinson and Ostroverkhova[131] |
| Quinone | Xylindein | *Chlorociboria aeruginascens* | Semi-conductors design | Giesbers, Van Schenck, Quinn, Van Court, Vega Gutierrez, Robinson and Ostroverkhova[131] |

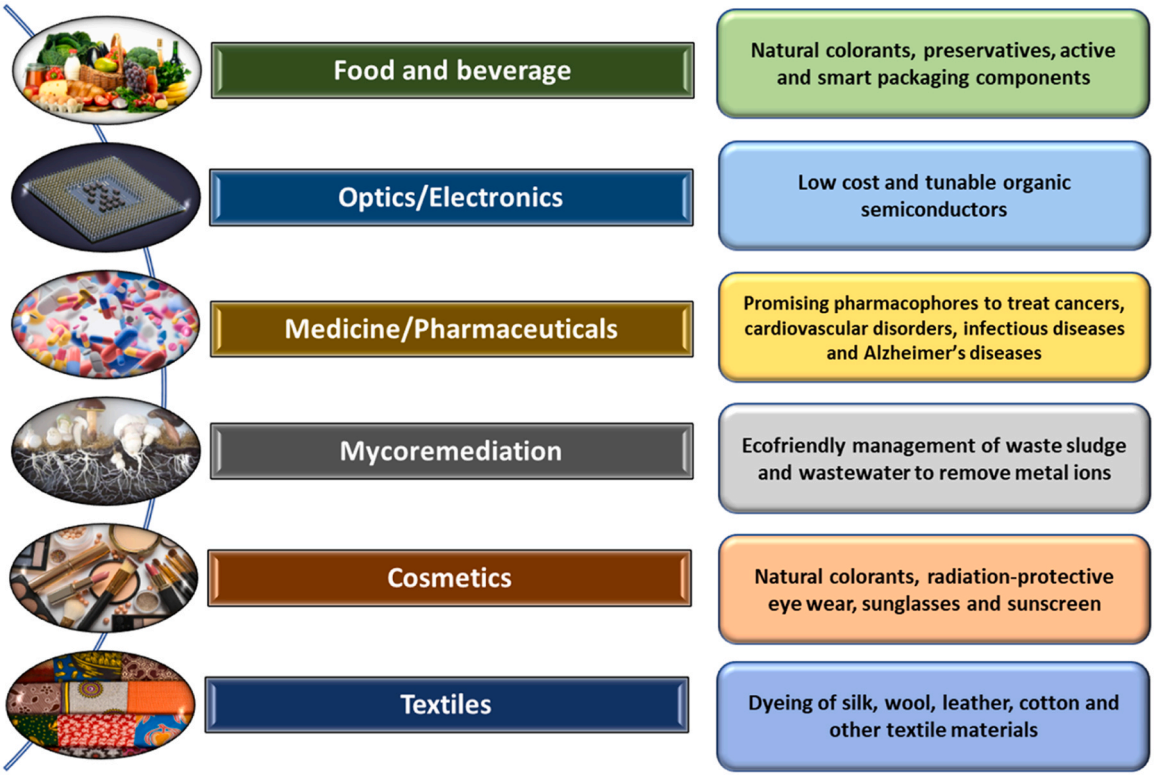


**Fig. 2.** Applications of mycopigments in various industries.

owing to their low cost and tunability [102,103]. Furthermore, the pigment compounds are made up of polar substituents which form strong quadrupolar or even dipolar molecules and consequently larger structural space which is immensely useful in semiconductor design [104]. For instance, xylindein from the wood-eating fungi, *Chlorociboria aeruginosa* and *Chlorociboria aeuriginascens,* is now being incorporated into poly (methyl methacrylate), an amorphous polymer, to form xylindein:PMMA blends [102]. In the cosmetic industry, hydroxyanthroquinones such as emodin, dermocybin and dermorubin from ectomycorrhizal fungus *Dermocybe sanguinea* have been used as hair dye constituents [105]. In addition, fungal melanin is being utilized in the development of radiation-protective eyewear, sunglasses and sunscreen due to its special anti-radiation property [106]. Furthermore, fungal melanin has shown potential in space aeronautics as it can be incorporated into composite material to protect equipment from space radiation hazards [20]. The pigment has also been demonstrated to be a better uranium adsorbent than commercial resins for cleaning uranium-contaminated sites due to its hydrophobicity and negatively charged surface [107].

## 4. Biotechnological strategies to improve mycopigment production

Currently, mycopigment production on a commercial scale is still limited due to the lengthy fermentation period and low product yield. However, various approaches have been identified to overcome these drawbacks and promote the economic viability of mycopigment production (Fig. 3). These approaches are further discussed in this section.

### 4.1. Optimization of production parameters

Production under optimal conditions is regarded as one of the key approaches in the upscaling of bioprocesses. In this regard, the mode of the bioreactor, the composition of the fermentation medium, and the environmental factors can all be optimized using the one factor at a time approach or the statistical and computational approaches. In a recent investigation using Box–Behnken experimental design, it was observed that a pH of 6.4, incubation temperature of 24 °C, agitation speed of 164 rpm, and a fermentation time of 149 h as the optimal factors for the production of orange and red pigments by *Talaromyces albobiverticillius* under submerged fermentation [132]. However, optimization of pigment production by *Monascus purpureus* under solid-state fermentation identified 50% initial moisture content, initial pH of 4.5–7.5, 30 °C, and a fermentation time of 144 h as ideal for maximum production. As expected for many fungal species, moderate temperature and acidic pH ranges were observed to be generally favourable for pigment production. The nutritional composition of the growth medium has also been optimized in various studies to enhance pigment production. Wu,

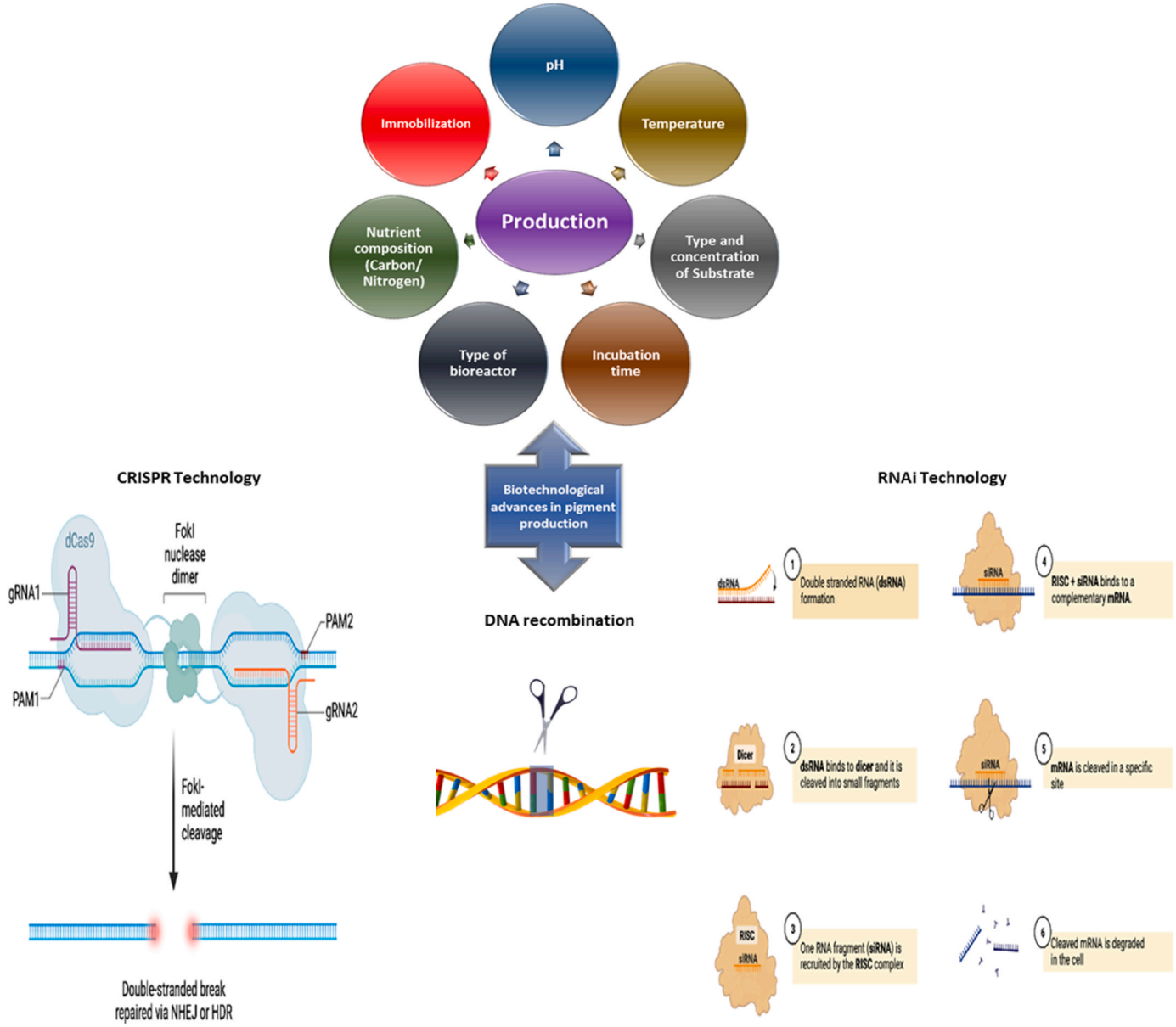


**Fig. 3.** Biotechnological approaches for improving pigment production.

Zhang, Yang, Zhou and Yang [133], optimized melanin production using submerged cultures of *Auricularia auricula*; the study demonstrated 1% methanol, 0.25% peanut oil, 1.0% stearic acid or 0.5% palmitic acid were optimal for heightened melanin yield. Using the central composite design (CCD), maximum production of orange and red pigments by *T. albobiverticillius* 30548 was obtained in the medium containing 3 g/L of yeast extract, 1 g/L of $K_2HPO_4$, and 0.2 g/L of $MgSO_4{\cdot}7\ H_2O$. Recently, the optimized media for enhanced *Rhodotorula* sp. carotenoids production was shown to be made up of 3.7 g/L malt extract, 7.7 g/L fructose, 9 g/L urea, 35 g/L NaCl, and 1 g/L yeast extract [134].

### 4.2. Cell immobilization

Immobilization of organisms has been reported to enhance pigment production as well as cell proliferation [135]. This technique is usually aimed at enhancing the production and stability of pigments while reducing the cost of production associated with the overall fermentation process [95,135]. More so, the use of immobilized cells has been described as more economical and promising since production is typically higher than observed in free cells [95,136]. According to Liu, Guo, Luo, Chai, Wu, Zhao, Jiao, Luo and Lin [137], the production of extracellular monascus pigments (yellow, orange and red) from *M. purpureus* LQ-6 increased significantly to 35.52 U/mL when immobilized in sodium alginate, compared to 14.19 U/mL of the free culture. In another study, immobilized *Talaromyces atroroseus* GH2 cells showed 30% higher pigment production levels relative to the free cells [138]. Similarly, pigment production from immobilized *P. purpurogenum* increased after the first cycle from 250 mg/mL to 264.6 mg/mL in the third cycle [139].

### 4.3. Genetic engineering

#### 4.3.1. Strain improvement

Strain improvement techniques have ensured that the limitations of low pigment yield by wild microbial cultures can now be circumnavigated. Major developments in industrial strain improvement have been achieved by mutagenesis using chemical or physical mutagens. For example, the use of UV-mutagenesis on *Rhodotorula glutinis* NCIM 3353

resulted in a mutant which produced 120 times more carotene than the parent culture [140]. Furthermore, while gamma radiation with cobalt-60 enhanced hypocrellin A production from mutant *Shiraia* sp. H-4–2 by 415% [141], cobalt-60 gamma-radiated mutant *Phaffia rhodozyma* produced 15 times more astaxanthin than the original strain [142]. Similarly, Fadel and Elkhateeb [143] reported a 50% increase in pigment production from gamma irradiated *T. atroroseus* TRP-NRC compared with the wild type and this was attributed to the effect of mutation via an increase in fungal-membrane permeability and gene copy number or upregulation of gene expression involved in pigment production [144]. Alternatively, chemical mutagens such as N-methyl-N'-nitro-N-nitrosoguanidine (MNNG) have also been used to enhance the production of astaxanthin from *Xanthophyllomyces dendrorhous* with a production fold increase of ~ 110% [145]. The combined effect of physical and chemical mutagens, such as an atmospheric and room temperature plasma method and nitrosoguanidine, has also been demonstrated to be effective in *Rhodosporidium toruloides* np11, as the mutant showed enhanced production when compared to the wild strain [146].

#### 4.3.2. Heterologous expression

Production of microbial pigments via heterologous expression allows either the expression of a metabolic route of synthesis in a microorganism that initially did not produce any pigment or the transfer of transcriptional factors or specific enzymes from one pigment producing microbe to another [147]. For instance, the overexpression of the transcription factor AurR1 in the aurofusarin gene cluster of *Fusarium graminearum* resulted in a production level which was thrice that of the wild type. The overexpression of the inosine-5′-monophosphate dehydrogenase (IMPDH) gene in *Ashbya gossypii* was found to increase the metabolic flux through the guanine pathway enhancing riboflavin production by 40% with respect to the wild strain [148]. Similarly, Lee, Lee and Lee [149] highlighted the overexpression of the laeA gene (a positive regulator of secondary metabolism) and replaced its promoter with alcA (a strong promoter from *Aspergillus nidulans*) in *Monascus pilosus*, which resulted in a 1.5-fold increase in pigment production and a 4-fold increase in antioxidant activity of pigments. However, heterologous production has the potential to not only maximize pigment yields, but it could also be used as an effective strategy to reduce the generation of unwanted by-products during bioprocessing. For instance, the use of monascus red, a food colourant produced by *Monascus purpureus*, is limited due to contamination with the nephrotoxin citrinin, which is also produced by the same fungus. A recent study developed a dual-plasmid CRISPR/Cas system for the deletion of the 15-kb citrinin biosynthetic gene cluster in *M. purpureus* industrial strain KL-001 [150]. Furthermore, the obtained homokaryotic mutants were stable, and citrinin was unambiguously eliminated, along with a 2–5% increase in the production of red monascus pigment [150].

## 5. Factors affecting the mycopigment stability

The stability of fungal pigments is of crucial importance with regard to their successful industrial applications, as during processing and extraction, conditions outside certain limits may render them prone to degradation, colour loss and reduced shelf life [151]. Typically, partial or complete colour loss of pigments occurs under extreme physical and chemical process conditions due to alterations in the chemical structure of the pigment molecule [117]. For instance, an increase in absorption intensity with its concomitant rise in temperature led to rupturing of the hydrogen bonds between the hydroxyl moieties of cochlioquinol II, a pigment from the endophytic fungus *Arcopilus aureus,* while an increase in pH led to the oxidation of the same pigment, contributing to the decrease in the intensity of its yellow colour and a loss in stability [152]. Thus, it is believed that the undesirable instability of these pigments has negated the progress made so far in reducing the high cost of fungal pigment production. Studies have since demonstrated that the major factors responsible for this instability include physicochemical factors, extraction methods, as well as processing methods [34].

### 5.1. Physicochemical factors

Like most natural compounds, the stability and shelf life of fungal pigments is usually affected by varying physicochemical conditions [34]. Although pigment production in some cells has been shown to increase when the cells were exposed to certain stress conditions such as elevated temperatures, osmotic pressures, metabolic inhibition and the presence of heavy metals, however, most often than not, pigment stability may be affected in many organisms [135]. In general, understanding the tolerance of fungi to conditions essential to their growth and function is vital in determining the application of their metabolites, such as pigments [153]. However, many studies have shown that the stability of fungal pigments may be a function of the chemical property of the pigment compound itself rather than the physiological growth conditions of the producing fungi [154,155]. In this case, the functionality and stability of these pigments have been observed in conditions which are not at par with their typical environmental conditions. For instance, pigments from the mesophilic fungi, *Monascus purpureus, Emericella* sp., *Fusarium* sp., *Isaria* sp., and *Penicillium* sp. were more stable at high temperatures and showed dyeing ability at acidic pH of 5.0 [154]. In contrast, a yellow pigment from *Thermomyces* sp., a thermophilic fungus, was used in dyeing cotton, silk and wool fabrics at optimum conditions of 30 °C and pH 3.0 [156]. It was also revealed the thermal stability of red pigment extracted from *Penicillium purpurogenum* GH2 was in the temperature range of 60 – 90 °C and at pH 6.0, thus, suggesting the applicability of the pigment in pasteurized food matrices where high-temperature short time condition is required [157]. It has also been shown that pigments produced by the same organism may differ in terms of response under certain physicochemical conditions, as observed in *Monascus* fungi, where the yellow pigments showed remarkable resistance to photodegradation and tolerance to high temperature as well as pH variations compared with the orange and red pigments [36]. The yellow pigment was found stable to light and tolerated temperatures up to 100 °C for 60 min at a wide pH range of 2.0–10.0, unlike the orange and red pigments [36].

### 5.2. Extraction methods

Extraction of fungal pigments from different sources is a delicate process due to the instability of the pigments at certain physicochemical conditions; thus, the need to use various extraction solvents (e.g., water, alcohols) and physicochemical methods. The general methods for the commercial extraction of mycopigments are widely classified into the conventional and the non-conventional methods, however, the choice of an extraction method depends on the efficiency of pigment released from the matrix into the extractant without altering them [158].

#### 5.2.1. Conventional extraction methods

Pigments can be extracted extracellularly when released into the fermentation medium or intracellularly from the fungal biomass through cell disruption of the dried or wet biomass [58,159,160]. Mechanical disruption is the common disruption method used via sonication, high-pressure homogenization, grinding, and bead-milling, which all cause considerable heat generation [161]. However, active cooling during extraction can minimize the degradation of thermolabile pigment compounds resulting in an efficient process with a high level of consistency in the property of extracted pigments [162]. A common workflow for the extraction of polyketide-derived pigments from mycelial biomass involves the use of organic solvents (such as ethyl acetate, chloroform), or a mixture of solvents, followed by mechanical disruption of the cells for removal of intracellular fungal pigments, and subsequent centrifugation or filtration [160]. These processes involve extended extraction times, exposure to organic solvents, and high

temperatures, with significant loss of bioactive substances due to hydrolysis, photo-oxidation and reduction in pigment stability. Thus, the selection of suitable extraction and purification procedures for mycopigment is highly dependent on the source (ascomycetes, basidiomycetes, yeasts, etc.), localization (intracellular/extracellular), and the final application [163]. For example, during intracellular melanin extraction, fungal biomass is hydrolysed using NaOH followed by pH adjustment of the broth to an acidic level for precipitation and further purification of the pellets [97,133,163]. However, during extracellular extraction from the fermentation broth, the pH is initially adjusted with 6 M HCl. In both methods, the precipitate is usually rinsed with deionized water, and washed with organic solvents (ethanol, ethyl acetate and chloroform to separate lipids and carbohydrates), followed by freeze-drying [164]. However, the use of certain acids and solvents aimed at removing the entire protein fraction, cell debris, and unconsumed nutrients has been found to alter the melanin polymeric skeleton [45,163]. Generally, consideration is also given to the safety, environmental and economic implications of solvents used in extraction bearing the intended use in mind [165].

#### 5.2.2. Non-conventional extraction methods

The non-conventional methods of mycopigments extraction are the several alternative methods whose operation revolve around environmental sustainability, as well as the optimal utilization of energy and chemicals. These methods can be considered "green" alternatives" and are more selective, less time-consuming, and thermally sensitive to the pigments compared to the conventional method. The more commonly utilized non-conventional methods of mycopigments extraction include pressurized liquid extraction, microwave-assisted extraction, ultrasound-assisted extraction, and green solvents. The pressure liquid extraction (PLE) technique is considered a promising eco-friendly extraction process for biological samples and other natural products [166]. Furthermore, PLE has been mostly applied to environmental samples (during recovery) as well as food and biological samples as an analytical method [167]. A report on PLE showed promising outcomes where mycelial biomass from 15 fungal strains was subjected to a six-stage pigment extraction process [58]. Since this method minimizes the exposure of fungal biomass to oxygen, light, higher temperatures and hydrolysis without extending the extraction time, the chances of affecting pigment stability are minimal. According to Lebeau, Petit, Fouillaud, Dufossé and Caro [166], the use of PLE enhanced the selective extraction of red azaphilone pigments from marine-derived *Talaromyces* sp. 30570.

Similar to PLE, microwave-assisted extraction (MAE) is considered an advanced and efficient extraction method requiring less extraction time with minimum solvent. In MAE, pigments within the cell are heated using microwave radiation energy [159,168] and are therefore less prone to alteration in structure or stability, since it is a very fast extraction process performed at highly controlled temperature conditions unlike in conventional thermal processes where heating is non-uniform [169]. The efficiency of MAE extraction is regulated by the nature of the substrate, type of solvents, solid-liquid ratio, pressure, temperature, and particle size for a high pigment yield [170]. Additionally, green solvents could be combined with MAE for efficient extraction as well as PLE. In a study on carotenoid extraction, degradation of carotenoids during microwave heating was observed at an initial temperature above 60 °C for the most thermosensitive molecules (violaxanthin and antheraxanthin) while provitamin A carotenoids (β-carotenes, α-carotenes and β-cryptoxanthin) and lutein were more stable to heat [171]. Hence, to err on the side of caution, MAE temperature should not exceed 60 °C to ensure mycopigment extraction in non-denaturing conditions. During ultrasound-assisted extraction (UAE), on the other hand, waves generated via acoustic energy within the liquid medium result in localized pressure which ruptures the tissue and releases the intracellular pigments into the extracting solvent [159,162,172]. The extraction of most biological compounds by UAE is regarded as a clean process and can be employed in a similar way as MAE. Another advanced method of pigment extraction is the enzyme-assisted technique which involves breaking up the structural integrity of the fungi using cell wall hydrolytic enzymes such as cellulase, hemicellulase and pectinase. This technique ensures higher recovery, faster extraction yield with lower energy requirements and reduced quantity of solvent utilized, although expensive [173,174]. Ultrasonication combined with enzymatic hydrolysis has been employed for the effective extraction of intracellular components including pigments from A*garicus blazei*, *Boletus*, *Shiitake*, and *Agrocybe cylindracea* following a two-stage enzymatic hydrolysis using cellulase, pectinase, and papain [175].

Recently, research on the use of green solvents, *viz*., deep eutectic solvents (DES) and ionic liquids (IL), to extract pigments and retain their intrinsic properties such as colour intensity, stability and bioactivity has become more prominent [8]. It was observed that these green solvents help to overcome the limitations posed by conventional solvents and are therefore considered suitable alternatives due to the reduced amount of solvent required, shorter extraction time, and their environmentally friendly nature [159]. Thus, ILs and DES are suitable for the extraction of bioactive pigments such as aglycones flavonoids [176], anthraquinones [177], astaxanthin [178], hence, they can be adapted for the extraction of fungal pigments since they are highly tuneable [179,180]. However, few studies have been reported on the use of these green solvents specifically in fungal pigment extraction, thus giving room for more research. For example, soluble polyketide pigments of red, orange and yellow hues were extracted from the fermented broth of four fungal strains: *Talaromyces albobiverticillius, Emericella purpurea, Penicillium marquandii,* and *Trichoderma harzianum* using quaternary tetrabutylammonium bromide ([N4444]Br-), 1-butyl-3-methyl-imidazolium chloride ([C4Mim]Cl- and 1-butyl-3-methylimidazolium bromide [C4Mim]Br-) ionic liquids [58]. Similarly, aqueous solutions of 1-alkyl-3-methylimidazolium chloride ILs ([$C_n$mim]Cl) (45% w/v in water) in combination with an ultrasonic-temperature-assisted technique were demonstrated to enhance the recovery of red pigments from filamentous fungi *T. amestolkiae* cell [181].

### 5.3. Processing methods

The processing of fungal pigments are the downstream activities that occur subsequent to the extraction process, after which the consumable mycopigments can be delivered to the end user in whatever form is required. It has been shown that these processing methods significantly affect the stability of mycopigments, and these methods have been classified into thermal and non-thermal methods. In thermal processing methods, heat is generated outside a product by the combustion of fuels or by an electric resistive heater and transferred into the product via conduction and convection [182]. One of the aims of conventional thermal processing methods in the food industry is to ensure the quality of food by enzyme inactivation and destruction of microorganisms [183], however, the heat generated during processing may degrade pigments used as food colorants [160]. For instance, blanching as a pretreatment method is beneficial in inactivating oxidative enzymes, however, it leads to the degradation of carotenoid pigments [32]. Thus, given the higher sensitivity of the pigment to temperature, high temperature and short-time conditions (HTST) are recommended [157]. Microwave irradiation (MI) is one of the highly regarded non-thermal methods employed in processing mycopigments. MI is a powerful non-contact, eco-friendly heating technique applied during various textile processes such as finishing, dyeing, printing and wet processes [184]. Unlike conventional processing, during MI, there is uniform heat distribution with reduced power consumption and operational time compared to conventional heating methods, making it an attractive heating technique [185]. The impact of MI has been reported on both the physical and chemical properties of pigmented fabrics. For example, a study by Elshemy and Haggag [184] reported significant improvement

in wool fabric dyeing with no significant change in its chemical structure when examined via IR spectroscopy, X-ray diffraction and scanning electron microscopy, thus implying that MI is non-destructive and therefore safe for fabric dyeing.

## 6. Stabilization of fungal pigments

Several emerging technologies have been investigated to tackle challenges such as colour degradation, instability and loss of bioactivity, which are encountered with mycopigments during food storage and/or processing. Microencapsulation and co-pigmentation have since been identified as the major approaches with immense potential in this regard (Fig. 4) and they are further discussed in this section.

### 6.1. Microencapsulation

Microencapsulation is applicable in various fields of science, especially in the food, pharmaceutical, agricultural, textile, pharmaceutical, and biomedical industries [186]. Various methods in the application of microencapsulation include complex coacervation, air coating, pan coating, centrifugal extrusion, solvent evaporation, spray drying, as well as *in situ* polymerization [186,187]. In the food industry, where it is more commonly used, microencapsulation enhances the stability of bioactive compounds and prevents undesirable interactions with other components of food formulation during processing and storage. The mechanism of microencapsulation involves the formation of a membrane wall to enclose droplets or particles of the encapsulated ingredient for protection and enhanced stability [188]. Spray-drying has been noted as one of the simplest and most widely used microencapsulation techniques with regard to mycopigments due to its relatively low cost, efficiency and availability of appropriate equipment [189]. For example, the spray drying of natural pigments produced by *Epicoccum nigrum* (CML2971), *Penicillium flavigenum* (CML2965), and *Aspergillus keveii* (CML2968) encapsulated in maltodextrin, modified starch, and gum arabic led to the production of yellow fine powders with low moisture content and water activity, which enhanced product stability and shelf life as well as product recovery and pigment retention [190]. Similarly, the encapsulation of *Monascus* pigments using the co-polymers, hydroxypropyl cellulose (HPC) and poly (lactic-co-glycolic) acid (PLGA) to enhance the photostability of the mycopigments has also been demonstrated [191]. Complex coacervation is a thermostable method with high dye retention and encapsulation efficiency as well low level of sophistication and lower operational temperature, and it has also been noted to have a lot of potential in mycopigment stability [192]. Similarly, nanotechnology is currently being employed in food processing, where the elaboration of nano-emulsions is utilized to increase stability. Depending on the method of preparation and ingredients used, different structures can be prepared which include nano-capsules, nanocarriers, nanocrystals, and nano-emulsion using biopolymers such as polysaccharides (chitosan, alginate, gum Arabic), protein (casein) either alone or as combined [193]. Thus, the increased application of these strategies to increase the solubility and stability of natural pigments in accordance with food commodity requirements might be a promising area of research for future advancement.

### 6.2. Co-pigmentation

Co-pigmentation, as a stabilization technique has been reported to improve the potential of natural pigments for applications in food, pharmaceutical, and other industries [194]. This phenomenon has been applied in enhancing colour expression and polymeric pigment formation because it causes shifts in the absorption maximum to longer wavelengths via inter-molecular co-pigmentation, self-association, intra-molecular co-pigmentation, self-association of acylated pigments and co-pigmentation in metal-pigment complexes [195,196]. However, the few reports on co-pigmentation as a stabilization technique for pigments are mainly on anthocyanins, which are plant pigments. However, the biosynthesis of anthocyanin by a fungal strain was

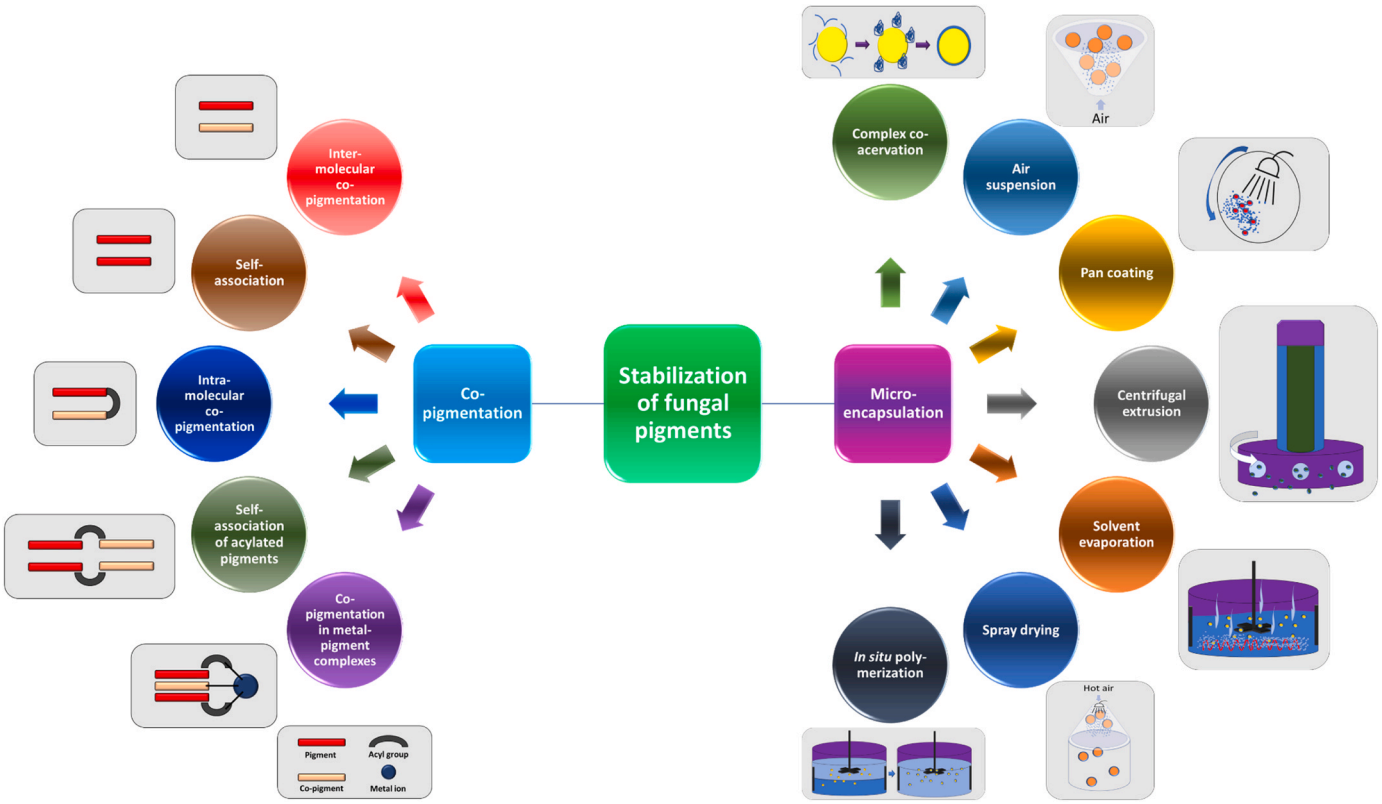


**Fig. 4.** Techniques for stabilization of mycopigments.

recently demonstrated in *Aspergillus sydowii* by Bu, Zhang, Zeng, Cao, Hao, Qiao, Cao and Xu [197], and with this finding it is expected that co-pigmentation will be a useful tool in enhancing the stability of anthocyanins and other related pigments from fungal species. Anthocyanin (ACN) in food products is susceptible to degradation in atmospheric conditions, however, studies have shown an improvement in the stability of these pigments via intramolecular co-pigmentation [198].

## 7. Conclusion

The utilization of mycopigments holds immense benefits over other natural pigment sources and their synthetic counterparts, thus increasing their demands and also opening new research avenues. This increased demand is attributed to their safety, eco-friendliness, and numerous remarkable biological activities. Over time, a lot of effort has been invested in identifying the critical variables affecting the production of pigments with the aim of optimizing yield, eliminating the presence of toxins as well as reducing the cost of production. On the other hand, less attention has been given to maintaining the stability of this pigment either stand-alone or when incorporated within material matrices such as food and textile fabric. In this regard, the limiting factors to mycopigment stability and useful techniques alongside existing approaches in extraction and processing to enhance their industrial applications were discoursed. Future work should be tailored towards the application of biotechnological approaches involving the latest data handling methods and chemoinformatic tools to explore fungal genomes for better production, functionality and stability. In essence, the shift to natural pigments and particularly mycopigments has begun in earnest, it is hoped that the scientific and industrial community will meet up with this shift, which is synonymous with the sustainability of our planet.

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## CRediT authorship contribution statement

**Grace Abel:** Investigation, Formal analysis, Writing – original draft. **Ayodeji Amobonye:** Conceptualization, Writing – original draft, Writing – review & editing. **Prashant Bhagwat:** Conceptualization, Writing – original draft, Writing –review & editing. **Santhosh Pillai:** Conceptualization, Resources, Funding acquisition, Supervision, Writing – review & editing.

## Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

## Data Availability

No data was used for the research described in the article.

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The authors acknowledge the use of Biorender that is used to create the schematic in Fig. 3.

## *Code availability*

No codes were utilized during this study.

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