Fungal Pigments: An Overview

26

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Abstract

Since ages, colors have been an integral part of humankind whether it belongs to foodstuff, clothing, or day-to-day living. Long back in history, various pigments are used by all the races. Earlier the colors that were in use were natural in origin, but due to rise in demand mankind shifted to manufacturing of synthetic colors. With the passage of time, it has been now proved that these synthetic colors have many side effects like being immunosuppressive, carcinogenic. Due to deleterious health effects, the need for some alternative has emerged that can be used as a color. Plants, insects, and other microorganisms have started taken place of synthetic colors. As there are many factors that limit the usage of plants and insects, research turned toward the microorganism. There are many fungi whose pigments are now considered as safe and economical. Fungi like *Aspergillus, Fusarium, Penicillum, Monascus, Trichoderma, and Laetiporus* are reported to produce quinones, anthraquinones, Rubropuntamine, Rubropuntatin, Ankaflavin, Monascin, β-carotene, and many other pigments responsible for various colors, viz. red, purple, yellow, brown, orange, and green. In addition to providing natural colors, these pigments possess many therapeutic applications like immune modulators, anticancer, antioxidant, antiproliferative. These pigments are produced as secondary metabolites by utilizing one of the pathways: polyketide, mevalonate, and shikimate pathways. The pigments are fermentative products so are affected by temperature, pH, carbon source, aeration, and type of fermentation (solid or submerged). There are many

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agencies that approve the usage of pigments for humankind. Fungi can work as cell factories for color production that is economical and human friendly.

Keywords
Pigments · Polyketide · Food colorant · Fermentation

Introduction

Since time, immemorial colors have always fascinated the humankind. Usage of colors dates back to the Bronze Age in European culture. In the world, the earliest written record for usage of natural colors found in Chinese culture that dates back to 2600 BC. In India, it records back to 2500 BC in Indus valley period and has been substantiated by the colored garments of Mohenjo-Daro and Harappa civilization. In central and North America, cochineal dye was used by the Aztec and Maya culture (Aberoumand 2011; Gokhale et al. 2004). The usage of biocolor in food was done by Japanese people shown in shosoin text of Nara period (eighth century). Color also decides the appeal to food. Food colors are divided into four categories: (1) natural colors, (2) naturally identical colors, (3) synthetic colors, and (4) inorganic colors. Natural colors or pigments are the metabolites produced by the living organisms; it includes carotenoids, anthocyanins, etc. Natural identical colors are manmade pigments that are found in nature like *β*-carotene, canthaxanthin. Synthetic colors are also manmade colors that are not found in nature, and it usually includes azo dyes while the inorganic colors are also manmade colors and includes titanium dioxide, gold, silver, etc.

With the passage of time, and due to increase in population and thus the rise in requirements, it was realized that it is not possible to be completely dependent on the natural colors. This promoted the formulation and usage of synthetic colors. There are many synthetic colors that are used in foodstuff, dyestuff, cosmetics, and in pharmaceutical industry. But over the period of time and advancement in technology, it was conferred that synthetic colors have many harmful effects. The precursors that are used for the production of synthetic colors are carcinogenic, some are immunosuppressive and imparting many non-environment friendly and non-biodegradable impact. Due to negative impact of synthetic colorants, the research has turned toward the production of natural pigments and has become significant worldwide. Over a decade, many metabolites have been discovered from diverse sources of nature including plants, animals, insects, and microorganisms. Among these, the pigments produced by the microorganisms are holding special place (Dufosse 2006).

Among the many microorganisms, fungi play an important role in pigment production that can be used safely; moreover, fungi are reported to produce larger amount of pigments (Kirti et al. 2014). Fungi produce many primary metabolites (require for its own metabolism) and secondary metabolites (not required for its own maintenance), organic acids, enzymes, pigments, and other food additives. These products of fungi possess many therapeutic applications like immune modulators, anticancer, antioxidant, antiproliferative. In addition to this, natural colorants possess antimicrobial activity and lesser chance of being allergenic. They are even more stable than that of synthetic colorants and are more eco-friendly (Velmurugan et al. 2010).

Various Pigments

Pigments from natural sources have been obtained since longtime and with time, interest in production of natural colorants have been increased due to toxic effects of synthetic colorants. Natural pigments like carotenoids, flavonoids (anthocyanins), chlorophylls, phycobiliproteins, betalains, and quinones are common pigments that are in use. Among these, due to the ease of cultivation, extraction, and genetic diversity, microorganism are most promising. Microorganism such as *Bacillus*, *Achromobacter*, *Yarrowia*, *Rhodotorula*, *Phaffia*, *Monascus* produces a large number of pigments. Carotenoids that are yellow, red, and orange are widely used as food and feed supplements and as antioxidants in pharmaceutical industry. *Phaffia rhodozyma*, *Haematococcus pluvalis*, *Agrobacterium aurantiacum* are widely known for the production of astaxanthin, which is added to poultry feed. Astaxanthin consumption has also been found to be beneficial in case of cardiovascular diseases prevention, immune enhancer, and cataract prevention (Ciapara et al. 2006). Prodigiosin-like pigment which is red in color is found to be produced by *Serratia rubidaed* (Moss 2002). Similarly, Cyanobacterium *Nostoc muscorum* produces Phycoerythrin pigment (Ranjitha and Kaushik 2005). The red mold *Monascus purpureus* that is traditionally used for the production of red rice is a promising source of red pigment (Mukherjee and Singh 2011). But as mentioned earlier, fungi produce higher amount of pigment and due to ever-rising demand by the consumers, natural colorants from fungi have replaced the use of synthetic dyes in food industry as well.

Fungal pigments are secondary metabolites that are sometimes produced due to scarcity in the nutritional value. When the nutritional supply of essential nutrients decreases or there is some disfavoring environmental condition, mycelium produces secondary metabolites (Gupta and Aggarwal 2014). There are some fungi including *Aspergillus*, *Fusarium, Penicillum,* and *Trichoderma* that produce various pigments as intermediate metabolites during their growth (Atalla et al. 2011). Fungal pigments are classified as carotenoids and polyketides. Fungal polyketides are made up of tetraketides and octaketides having eight C_2 units forming polyketides chain.

Anthraquinone is most common class that is proved to be potentially safe (Mapari et al. 2010). Pigment anthraquinone is widely used in dyestuff industry and most commonly produced by *Trichoderma*, *Aspergillus,* and *Fusarium* (Duran et al. 2002). It is now known that single fungal species can produce mixture of different pigments, having various biological properties. Production of theses pigments plays an important role in fungi. Like melanin production helps the fungi to survive in severe environmental stress, helps to cope up with UV light. Study conducted by Kunwar et al. (2012) showed that its consumption by BALB/c mice increases the survival time in radiation-exposed mice. *Monascus* produce six various pigments that are polyketide in origin and imparts yellow, orange, and red colors. Monascin and ankaflavin are yellow pigment, while monascorubrin and rubropunctatin are orange pigment and monascorubramine and rubropuntamine are red pigment (Feng et al. 2012). There are four species of *Monascus,* namely *M. pilosus*, *M. purpureus*, *M. rubber,* and *M. froridanus* that account for majority of pigments isolated so far. *Monascus* pigments are believed to be sensitive to heat and fade with light, unstable at low pH and also have low water solubility although upon reacting with amino containing compounds their stability increases (Dufosse 2009). In addition to this, even the higher fungi, mushrooms, have also been reported for production of various pigments. Gupta et al. (2013) utilized *Trichoderma* sp. for dyeing of silk and wool as it imparts yellow color to silk and wool. Wood-rotting edible mushroom, *Laetiporus sulphurous*, contains non-isoprenoid polyene known as laetiporic acid A and 2-dehydro-3-deoxylaetiporic acid A as the main pigments imparting yellowish or orange color in the fruiting bodies (Davoli et al. 2005).

Similarly, there are many other species of mushrooms that are produce various pigments imparting different colors. There are more than 100 pigments that have been reported in fungi, and it holds place after the plants. Some of the fungal pigments are summarized in Table 26.1.

Various colors of the fungi are one of the very important characteristics that help in their identification. Green color of *Penicillin*, violet color of *Cortinarius*, yellow (Chen et al. 1969), orange, and red color of *Monascus* (Feng et al. 2012) are their distinct feature. Their pigments provide them protection against UV light and may also from the bacterial attack. The pigments of fungi differ greatly from higher plants being not possessing chlorophyll or the anthocyanins that impart various colors to flowers. Many of the fungal pigments are quinones or similar conjugated structures. The pigmentation in fungus sometimes varies with its age. As observed in *Penicillium chrysogenum* that initially, their colonies appear white in color and later that changes to blue-green (Tiwari et al. 2011).

Quinones are very common polyketide fungal pigments that are produced by following polyketide pathway. As its reduction product usually accompanies quinone, this is not necessary that it will show the color of the fungus from which it has been isolated (Feng et al. 2015).

Fumigatin (1) (Fig. 26.1) is isolated from *Aspergillus fumigatus* (Anslow and Raistrick 1938). It was observed that solution in which *Aspergillus fumigatus* grown was initially yellowish-brown and later changed its color to purple when treated with alkali (Hanson 2008). Auroglaucin (2) and Flavoglaucin (3) (Fig. 26.1)

Table 26.1 Various fungal pigments and their sources

Fungi	Pigment	Color	Reference
Penicillium herquei	Atronenetin	Yellow	Takahashi and Carvalho (2010)
Penicillium purpurogenum	Purpurogenone	Orange to yellow	King et al. (1970)
	Mitorubrinol	Red	Teixeria et al. (2012)
	Mitorubrin	Yellow to orange	Martinkova et al. (1995)
Penicillium oxalicum	Anthraquinone	Red	Atalla et al. (2011)
Trichoderma virens	Viridol	Yellow	Mukherjee and Kenerley (2010)
Monascus sp.	Ankaflavin	Yellow	Mostafa and Abbady (2014)
	Monascin	Yellow	Juzlova and Martinkova (1996)
	Rubropuntamine	Red	Yang et al. (2014)
	Monascorubramine	Red	Babula et al. (2009)
	Rubropuntatin New pigment	Orange Red	Moharram et al. (2012) Mukherjee and Singh (2011)
Fusarium oxysporum	Anthraquinone	Pink/violet	Gessler et al. (2013)
Fusarium verticillioides	Naphthoquinone	Yellow	Boonyapranai et al. (2008)
Aspergillus sclerotiorum	Neoaspergillic acid	Yellow	Teixeria et al. (2012)
Aspergillus niger		Brown	Atalla et al. (2011)
Aspergillus versicolor	Asperversin	Yellow	Miao et al. (2012)
Phycomyces blakesleeanus	β-carotene	Yellow-orange	Malik et al. (2012)

are the pigments first studied in 1930s and 1940s in *Aspergillus, Penicillia,* and *Helminthosporium* species (Raistrick 1940; Quilico et al. 1949). Species of *Aspergillus glaucus* series was characterized by green conidial heads and hyphae with varying colors of bright yellow to red. These organisms are found as spoilage organism. Dried form of these organisms gave various pigments like auroglaucin (orange-red needles), flavoglaucin (lemon-yellow needles), and rubroglaucin (ruby-red needles) (Gould and Raistrick 1934). Studies on pigments from Rubroglaucin was eventually shown to be a mixture of hydroxyanthraquinones physcion (4) and erythroglaucin (5) (Fig. 26.1). *Helminthosporium germineum* a causative organism of leaf stripe diseases of barley yielded deep red color when grown in Czapek medium. Main constituent was trihydroxy anthoquinone helminthosporin (6) (Fig. 26.1) (Charles et al. 1933; Hanson 2008).

Later, 40 various species of *Helminthosporium* were studied and various pigments and their isomers were established, it includes catenarin (7) (Raistrick et al. 1934) (Fig. 26.1). An isomer of helminthosporin, Islandicin (8) (Fig. 26.1) was isolated from *Penicillium islandicum* (Howard and Raistrick 1949). Studies reveled that all the red pigments were not anthraquinones. Another pigment Xanthone,

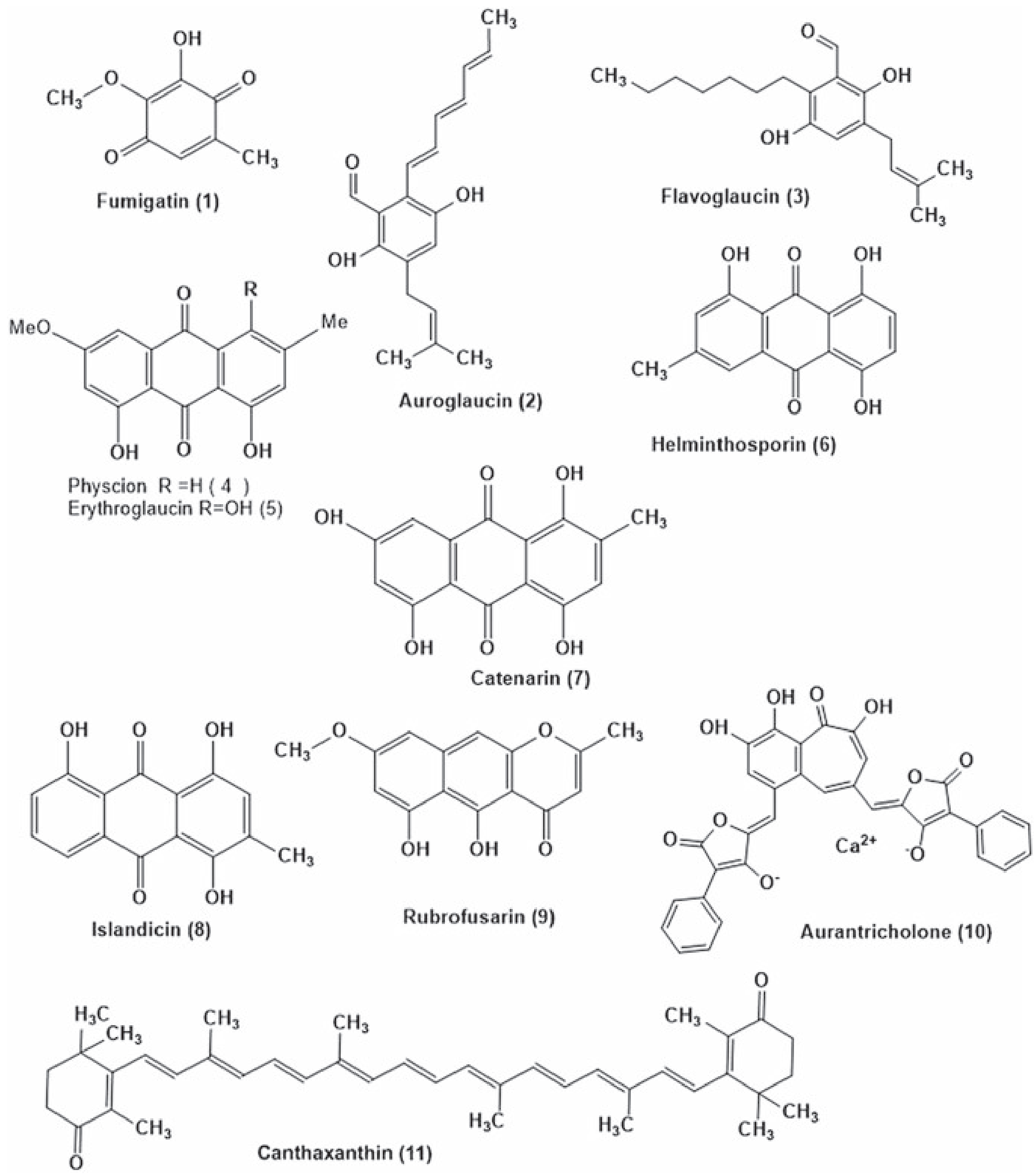

Fig. 26.1 Structure of various pigments

rubrofusarin (9) (Fig. 26.1) was isolated from genus *Fusarium graminearum* (Ashley et al. 1937; Tanaka and Tamura 1962). In 1960s, extended quinone structures were established which were formed by the dimerization process. Fungus *Chlorociboria aeruginosa* imparts green color to the wood, which is due to the extended quinone xylindein. Studies in the field were continued, and later findings suggested that there are many more pigments that are dimerized and occur as complex (Saikawa et al. 2000). Like orange-red cups of *Tricholoma aurantium* contain a pigment aurantricholone (10) (Fig. 26.1) in which pyragallole ring is attached to pulvinic acid and oxidatively dimerized (Klostermeyer 2000). It occurs as calcium complex. Since fungi are non-photosynthetic organism, still there are some species that have been reported to have carotene hydrocarbons. It includes *Blakeslea trispora, Phycomyces,* and *Neurospora crassa.* Yellow pigment canthaxanthin (11) (Fig. 26.1) is isolated from *Cantharellus* sp. And this pigment might have arisen from the carotenoid (Haxo 1950). Studies reveled that fungi due to various environmental conditions produce various pigments also like naphthoquinone pigments are released as a response of fungi under stress conditions. *Monascus purpureus* was selected for the production of food colorant (Lin 1973). Its studies done under submerged conditions reveled that improvement of the O_2 supply increases the production of secondary metabolite (red pigment) and low O_2 transfer coefficient was required to improve the red pigment. Before preferring any strain for production of food colorant, it is important to check the pigment productivity and production of mycotoxins (Hanson 2008).

Genetic Basis of Pigment Production

Polyketides are most commonly found fungal secondary metabolites. Genetically, yellow *A. nidulans* spore-pigment intermediate naphthopyrone (WA), the carcinogen aflatoxin and the commercially important cholesterol-lowering compound lovastatin are best described. Type I polyketide synthases (PKSs) plays an important role for the synthesis of fungal polyketides. These PKS's are multidomain proteins and are related to eukaryotic fatty acid syntheses. For the synthesis, usually acetyl coenzyme A (acetyl CoA) and malonyl CoA condensed to form carbon chains of varying lengths and reduction of the β-carbon is optional. In addition, ketoacyl CoA synthase, acyltransferase, and acyl carrier domains are also essential for polyketide synthesis, while the ketoreductase, dehydratase, and enoyl reductase domains which are required for ketone reduction in fatty acids are not present in all fungal PKS enzymes. Due to the restriction to one module, they can carry out recurring biosynthetic reactions, and are, therefore, called 'iterative PKSs.' Aromatic polyketide naphthopyrone are formed due to claisen-type cyclization which is performed by the C-terminal region of the enzyme, having thioesterase domain motif. Fungal polyketides structures can perform iteration reactions which results into its diversity. Further, variety is achieved by the introduction of many different post-polyketide synthesis steps (Fujii et al. 2001; Keller et al. 2005). Non-ribosomal peptides are derived from both proteinogenic amino acids and non-proteinogenic amino acids by multidomain, multi-modular enzymes named non-ribosomal peptide synthases (Keller et al. 2005). Fungi are capable of synthesizing many important terpenes, like aristolochenes, carotenoids, gibberellins, indole-diterpenes, and trichothecenes. Terpenes are composed of several isoprene units that can be linear or cyclic, saturated or unsaturated. Utilizing different diphosphates and enzyme terpene cyclase various terpenes are produced. Although terpene cyclases have structural homology, they have little primary sequence similarity and seem to have diverged relatively rapidly from a common ancestor. Several fungal terpene cyclases have been characterized, including a bifunctional terpene cyclase from

Gibberella fujikuroi (Tudzynski et al. 2001), a trichodiene synthase from *Fusarium sporotrichioides* (Rynkiewicz et al. 2001) and an aristolochene cyclase from *Aspergillus terreus* and *Penicillium roquefortii* (Keller et al. 2005). Gene cluster and the arrangement in various domains are one of the major causes of diversity among the fungi and the production of various secondary metabolites.

Factors Affecting Pigment Production

Fungal pigments are secondary metabolites known as polyketides. Although these come under the nonessential metabolites for growth and reproduction, the developmental stage of fungus greatly influence the production of these pigments, and the developmental stages are influenced by the extrinsic as well as intrinsic factors, including pH, substrate, oxygen, temperature, water activity, and light availability. Like any other fermentative production medium composition, aeration rate, agitation rate, nutritional limitation, status of carbon supply affects the production of secondary metabolites. pH of the medium affects the growth and thus the production of secondary metabolites by the fungus. At pH 5 *M. purpureus* produces red pigment, while at the same pH *Penicillum sclerotiorum* produces orange pigment (Mukherjee and Singh 2011; Lucase et al. 2010). The pigments of *Monascus* vary from red-yellow to orange depending upon the culture conditions. Similarly, other growth conditions like aeration and nitrogen source for *Monascus sp.* result production of extra cellular water-soluble pigment (Hajjaj et al. 1998). Temperature is another important factor that affects the pigment production. The optimum temperature for the *Monascus sp.* was found to be within 28–30 °C and pH range of 4.5–8.5. Maximum pigment production has been reported at pH 4.5, which is yellow in color (Dikshit and Tallapragada 2011). The *Penicillium* sp. was found to produce extracellular pigment at pH 9.0 and 30 °C. In the higher fungi, usually, incubation temperature 25 °C is preferred as they require longer time period for mycelial growth. Other sources like carbon, nitrogen, oxygen, phosphorous also play an important role. The higher level of oxygen and lower level of carbon dioxide result in decreased ratio of biomass and thus the pigment production (Han and Mudgett 1992). Even the light conditions also influence the pigment production. It has been reported that incubation of *Monascus* sp. in darkness results in effective production of red pigment while illumination results in loss of pigment and thus postulated the photoreceptor response of fungus. Even the illumination with red and blue light affected the pigment yield (Babitha 2009).

Musaalbakri et al. (2006) explained that *M. purpureus* FTC 5391 was able to produce red pigment with different carbon sources, viz. glucose, potato starch, and rice starch. And the addition of tryptophan or 6-furfuryl aminopurine as nitrogen source was helpful in production of extra cellular pigment (Zhang et al. 2013). The addition of gibberlic acid, vitamin B2, and other amino acids (L-leucin and glycin) to the liquid medium also enhanced the pigment production (Baneshi et al. 2014). The pigment production was found to increase in *Fusarium verticillioides* with the proportional rise carbon source, glucose, and yeast extract (Boonyapranai et al. 2008). In *Fusarium moniliforme,* KUMBF1201 as compare to others peptone and yeast extract showed the best result for higher pigment production (Pradeep et al. 2013).

Fermentation for Pigment Synthesis

The production of microbial pigments by fermentation is a fascinating area, and nowadays, due to lot of advancements, this biotechnological approach has attracted lot of attention. Based upon the requirement, solid-state fermentation or submerged fermentation is in the practice. Vegetative cells or spore suspension are used as inoculum for the fungal fermentation. The spore suspension is more advantageous over the vegetative cells due to ease of handling, high viability, longer storage, easy maintenance, and preservation (Ajdari et al. 2011). In submerged fermentation, there is utilization of free-flowing substrates (broth, molasses, etc.). The required product is secreted into the fermentation broth. In this method, the continuous supply of substrate and nutrients is required as their utilization is fast. The product is easily purified from the fermentation broth. Although solid-state fermentation is a traditional method used for pigment production (Vendruscolo et al. 2010), for some species submerged fermentation has been developed. Submerged fermentation has been explored for *Monascus* pigments so that the demerits like problems of space, scale-up, and development control of solid culture can be achieved. The submerged fermentation is also helpful in production of many secondary metabolites; it also decreases the cost of production as compared to solid-state fermentation. For the large-scale production of new red pigment from *Monascus purpureus* submerged fermentation would be more economical and beneficial (Mukherjee and Singh 2011). *Penicillium funiculosum* IBT3954 found to yield (0.13 g/L) greater amount of red pigment under the submerged condition (Jens et al. 2012). Similarly, *Paecilomyces sinclairii* showed good yield of 4.40 g/L under the submerged fermentation (Cho et al. 2002). Addition to this, carotenoid production from *Aspergillus sp*. is also favorable through submerged fermentation. *Penicillium purpurogenum* produce color both in solid as well as liquid media while *P. purpurogenum* DUPA 1275 showed good results in submerged culture (Mendez et al. 2011; Santos-ebinuma et al. 2013). Many other pigments with higher commercial value including prodigiosin, monascorubramie, astaxanthin, canthaxanthin, β-carotene, etc. are produced by utilizing submerged fermentation. Solid-state fermentation is the process where fermentation process is performed on a non-soluble substance which acts as support source for growth ad also a nutrient supplier. There are many physical supports and nutrient supplier like rice bran, wheat bran, coconut oil cake, jackfruit seed powder that are used for solid-state fermentation. The main advantage of this type of fermentation is this that it utilizes nutrient-rich waste material. The utilization of nutrients is very slow, and same substrate can be used for longer time period. This is best suited for those fermentation processes where requirement for moisture content is very low. Among so many fungal sp. *Monascus sp.* holds a special place, as since ages, it is utilized in this for production of red pigment. *M. purpureus* KACC 42430 gave the higher yield under the solid-state fermentation when corncob powder is used as a substrate. Corncob supplies higher amount of cellulose and hemicellulose that promotes the pigment production (Babitha et al. 2007). Rice is considered as one of the best substrates for *Monascus* under the solid substrate fermentation, at optimum fermentation time it produces considerable amount of pigment (Singh et al. 2015).

Mycotoxins and Their Replacement

Study on the fungal secondary metabolites is not complete, if we do not discuss the mycotoxins. Mycotoxins are the secondary metabolites produced by many fungi which have the capacity to damage health, productivity and sometimes lead to death also.

Monascus sp. produces yellow and red pigments that are commercially and legally used as food colorant and used in the form of red rice powder in Southeast Asia. But, *Monascus* pigments are not approved in European Union (EU) and the USA (US), mainly due to the risk of the possible contamination by the nephrotoxic, and hepatotoxic metabolite citrinin. Another example is Quorn TM produced by *Fusarium venenatum* which also produces cytotoxic metabolite 4,15-diacetoxyscirpenol. But fungal producers are generally categorized as GRAS, which implies that with continuous monitoring their mycotoxins can be controlled. But the controversy has triggered the scientific community to find out some of the alternatives, either by manipulating culture condition, developing strains incapable of synthesizing citrinin by metabolic engineering or by screening some genera other than *Monascus* that produce polyketide pigments (Dufosse et al. 2014). Rigorous search for some potential strain has resulted in discovery of some strains of *Talaromyces species* (*Talaromyces aculeatus, T. funiculosum, T. pinophilus, T. purpurogenum*) that are producing *Monascus*-like polyketide azaphilone pigments without co-producing citrinin or any other known mycotoxins (Mapari et al. 2009). Attention is now moving toward the marine fungi also; studies have shown that marine fungi can produce more brilliant color with more stability and lesser or no mycotoxins.

Relevance of Pigments in Various Fields

With the advancement in the biotechnological tools, interest in search of natural pigments has also been augmented. The inclination of society for the search of natural ingredients has hard-pressed the scientist to work in the field for the production of more economical products with health benefits. Moreover, nowadays food industry is facing lot of challenges in replacing the synthetic colors with natural colors having antimicrobial and antioxidant properties (Vendruscolo et al. 2013). The red color pigment derived from *Monascus sp.* is very well documented as one of the oldest pigments used in Chinese culture. The red pigment produced by *Monascus purpureus* possesses antimicrobial activity as compared to ciprofloxin (Kumar et al. 2012). *Monascus* sp. producing rubropuntatin (orange pigment) as pigment is found to possess anticancer activity (Moharram et al. 2012). Another pigments Monascorubramine and Rubropuntamine forming red color showed antioxidant activity (Babula et al. 2009; Yang et al. 2014). Similarly, pigment virone produced by *Trichoderma virens* showed antifungal activity (Kamala et al. 2015). In the last decade, much research has been focused to limit the production of citrinin that has limited the use of *Monascus* in food directly. Many strains have been developed showing low or no production of citrinin. Although the gene responsible for pigment production in *Monascus* is still under controversy, only few genes are reported. It has been reported that *MpigE* had great impact on pigment production and its overexpression has led to low concentration of citrinin in fermentation media (Liu et al. 2014). *Penicillium* sp. has been reported to produce many pigments with significant therapeutic value and can be used in agriculture also. *P. oxalicum* var. Armeniaca CCM 8242 produces Ar pink red pigment that possesses anticancer activity.

Mitorubrin pigment (yellow to orange in color) produced by *P. Purpurogenum* plays important role in pharmaceutical and food industry (Mapari et al. 2005). *Penicillium herquei* producing yellow color pigment, Atronenetin showed antioxidant activity and is used in food industry (Takahashi and Carvalho 2010). *Aspergillus sp.* showed the production of secondary metabolite that are useful in agrochemical industry like *Aspergillus sclertiorum* DPUA 585 produces neoaspergillic acid that showed antibacterial activity against *E. coli, Mycobacterium smegmatis*, and *S. aureus* and antifungal activity against *C. albicans* (Teixeria et al. 2012). Astaxanthin produced from *Phaffia rohodozoa* and *H. pluvalis* is a red pigment and is used in feed, pharmaceutical, and aquaculture industries. Textile is another such sector where coloring agents are required and if they are from natural resources they are more preferred, due to better biodegradability and higher compatibility with environment. *Trichoderma virens*, *Alternaria atternata, and Curvularia lunata* have been utilized for pigment production for textile industry (Shrama et al. 2012). Many species of *Aspergillus* are being studied in this field. *A. niger* NRC 95 produces brown pigment that is used for the dying of wool (Atalla et al. 2011). *Fusarium oxysporum* produces anthraquinone compound, which is also used for the dyeing of wool and silk (Nagia and EL-Mohamedy 2007). Poorniammal et al. (2013) purified pigments from *P. purpurogenum* that are helpful in dyeing of cotton fabrics and additionally possess antibacterial activity. Likewise, there are many more fungal pigments that are helpful to us in many ways. The world of fungi is very colorful and still needs rigorous work to isolate more pigments that are friendly to us.

Future Prospects

As already discussed that use of natural colors as food colorants in the form of red rice, wine, etc., dates back to Bronze Age. The place of natural colors was taken by chemically synthesized colors in the nineteenth century. But with the reports of health hazards imposed by these chemically synthesized colors, various regulations throughout the world thus resulted in resurgence of demand for natural colors. Nowadays consumer awareness between diet and health is increasing, that has promoted the food colorant market, and even the demand of natural dyes has also increased. Fungi specifically ascomycetous, basidiomycetous and lichens are known to produce various pigments. Out of these, ascomycetous has taken most of the attention as they are easy to grow under laboratory conditions and thus provides ease for large industrial production. There are many reports showing the large-scale pigment production from ascomycetous fungi in a bioreactor under controlled conditions. Moreover, the usage of fungi does not make producer season dependent. The usage of natural food colorant was earlier confined to the semi-fermentative production of riboflavin, which is a natural yellow food colorant, produced by *Eremothecium ashbyii* and *Ashbya gossypi* (Wickerham et al. 1946). But it has some limitations, as it is light sensitive and readily gets fade away. The production of β-carotene from the *Blakeslea trispora* by DSMTM in the Netherlands was a breakthrough in the world of food colors. Earlier, tomato was the only source of carotenoid lycopene, but now EU legislation has approved *B. trispora* as a potential source of lcopene (Commission regulation 2006). With the production of many natural colors, many synthetic colors have been banned. Import of red colorants of the Sudan series is banned, as there are reports showing the carcinogenic effect of red color.

In the European parliament, it is clearly mentioned that any food article having synthetic color (sunset yellow carmoisine, ponceau 4R, etc.) will require a proper safety label. There is wide range of Polyketide pigments including anthraquinones, hydroxyanthraquinones, naphthoquinones, and azaphilone structure, each of which exhibits a wide range of colors. Polyketide pigments of *Monascus* sp. is traditionally used in Southern China, Japan, and Southeast Asia for making red rice wine, red soybean cheese, and Anka. *Monascus* sp. produces various pigments including ankaflavin and monascin producing yellow color, monascorubrin and rubropunctatin producing orange color, and monascorubramine and rubropunctamine producing purple-red color. Additionally, it produces mycotoxin citrinin, a hepato-nephrotoxic compound, which limits its usage.

Although literature did not show any reports showing death due to consumption of red rice wine, red soybean cheese or Anka. Apart from *Monascus, Penicillium* has also been reported for the production and further utilization of human-friendly pigment. A pink red, anthraquinone-based colorant, derived from *Penicillium oxalicum* has been reported as a safe biocolor. In addition, there are other species of *Penicillium,* viz. *Penicillium aculeatum, P. pinophilum* that are reported to produce azaphilone *Monascus*-like pigment. These strains do not produce any other mycotoxins and are safe for the human use (Mapari et al. 2008). *Penicillium herquei, Cordyceps unilateralis* show a promise for a future, having similar structure like that of plant-derived red pigments shikonin and alkanin.

Food colors from plants are already in the market to a greater extent. But due to their unavailability throughout the year and lesser production per cycle has moved the trend toward the microorganisms. Exploration of fungal diversity for biocolor production and lesser or no mycotoxin production is still going on, with an emphasis on production of water-soluble pigments (Mapari et al. 2010). We can conclude that fungal pigments have quite good future prospects for robust industrial production of various colors. The natural pigments from ascomycetous fungi can serve as a sustainable natural color. Available data indicate that filamentous fungi can be used as cell factories for pigment production. Despite production of mycotoxins by some of the species, studies are still going on to find out other fungal cell factories that are economical and human-friendly.

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