Visible pigmentation of the skin, hair, and eyes depends primarily on the presence of melanin(s) in those tissues. Melanins are produced by specific cells called melanocytes. Not only is the type of melanin produced important, but also its eventual distribution in the tissue dramatically affects visible color, which ultimately determines the functions of the pigment, such as photoprotection (Gilchrest, 2011). Clearly, the specification, migration, and differentiation during development of melanocyte precursors (“melanoblasts”) in specific patterns are essential for eventual pigmentation in adults (Kawakami and Fisher, 2011). Following is a synopsis of critical findings that have led to our current understanding of the biochemical pathways and melanogenic factors involved in melanin synthesis.

The key enzyme involved in the synthesis of all types of melanins from the initial precursor tyrosine is tyrosinase (EC 1.14.18.1). Tyrosinases have been described in many species, including mammals and lower animals, plants, and even fungi; in fact, the earliest observations of the catalytic function of tyrosinase were made in extracts of mushrooms (Bourquelot and Bertrand, 1895), which are still widely used today as a highly enriched source of that enzyme. All tyrosinases depend on the binding of copper for their catalytic function (Lerner et al., 1950; Lerch et al., 1986), although their substrate specificities and physical properties can differ dramatically depending on the species (Lerner et al., 1951; Hearing et al., 1980). The ratelimiting initial step in the biosynthesis of melanin was initially thought to be the hydroxylation of tyrosine to 3,4-dihydroxyphenylalanine (DOPA) and its immediate subsequent oxidation to DOPAquinone (DQ). In melanocytic cells, the DQ formed will be spontaneously converted to an orange-colored intermediate known as DOPACHrome. In vitro, the DOPACHrome will spontaneously lose its carboxylic acid group to form 5,6-dihydroxyindole (DHI), which can then further oxidize and polymerize to form a dense, high-molecular-weight complex now known as DHI-melanin. This was initially reported by Raper (1926), and the pathway was later refined by Mason (1948); hence, the biosynthetic pathway is frequently referred to as the Raper-Mason pathway. Throughout the 1950s, 1960s, and 1970s, the collaborative research groups at Yale (headed by AB Lerner) and Harvard (headed by TB Fitzpatrick) played key roles in defining the involvement of tyrosinase in the human skin pigmentation (Fitzpatrick et al., 1950), how its activities were confined to melanosomes and how those organelles developed (Seiji et al., 1961; Szabo et al., 1969), and the disruptions that occurred in those processes in many skin pigmentation diseases (Breathnach et al., 1965; Kawamura et al., 1971; Lerner and Nordlund, 1978; Rees, 2011; Spritz, 2011). Those findings, plus the training of many post-doctoral fellows and clinicians in their groups, played a major role in establishing research centers in Asia, Europe, and the Americas, which still have a strong influence on studies of skin pigmentation and related pigmentary diseases. As summarized in Figure 1, recent revisions of this melanogenic pathway have shown that DOPA is not a distinct intermediate produced initially from tyrosine, but is in fact produced later in the pathway owing to the paired reduction of DQ (Riley, 1999), and that downstream tyrosinase-related enzymes can rearrange the DOPACHrome to form a carboxylated intermediate (DHI-2-carboxylic acid) known as DHICA, as discussed below.

Analysis of the structures of the high-molecular-weight polymers of melanins depended on the development of new techniques to analyze these intractable pigments, and gradual progress was made in defining those structures, initially by Nicolaus’s group in Naples and Swan’s group in the United Kingdom (Swan, 1963; Nicolaus et al., 1964). In working with melanins found in nature, it quickly became apparent that there were two major types, the brown–black melanins now collectively known as eumelansins, and the yellow–red melanins now collectively known as pheomelansins. Prota’s group in Naples took the lead in defining the structure of pheomelanins and the involvement of sulfur as responsible for its unique color and properties (Prota, 1980). Prota and colleagues, as well as Ito and Wakamatsu in Japan (Ito et al., 1984), developed a series of more sensitive and specific assays for eumelanin and pheomelanin intermediates that gradually formed the basis for our understanding of how they are formed in melanocytes and how they are copolymerized in situ. The critical role of sulfhydryl groups in reacting immediately with DQ upon its formation to form various combinations of cysteinylDOPAs and downstream reactions of those intermediates, via cysteinylDOPA-quinones and benzothiazine intermediates, to produce
Pheomelanins were gradually defined by the groups of Rorsman in Sweden, Ito in Japan, Thody in the United Kingdom, and Prota in Italy (Agrup et al., 1979; Ito and Fujita, 1985; Thody et al., 1991; Napolitano et al., 1994). Studies characterizing the structural and physical properties of melamins in various tissues have also been led by the groups of Simon in the United States, d’Ischia and Zecca in Italy, and Sarna in Poland (Sarna, 1992; Zecca et al., 2001; Liu et al., 2005; Pezzella et al., 2009). The determination to produce eumelanin and/or pheomelanin is regulated physiologically, primarily by the melanocortin 1 receptor (MC1R) as modulated by its opposing ligands, melanocyte stimulating hormone (MSH) and agouti signaling protein (ASIP); this is discussed in more detail by Rees (2011).

Unexpectedly, in 1980, Pawelek and colleagues reported the novel finding that a biological factor produced in melanocytes was able to prevent the spontaneous decarboxylation of DOPAchrome to DHI, which led to the production of a more soluble and lighter-colored carboxylated melanin, now known as DHICA-melanin (Körner and Pawelek, 1980). There was some initial controversy about this point, since that activity could be mimicked in vitro by various divalent metal cations (Palumbo et al., 1987), but with the advent of molecular biology and cloning, the race to clone the tyrosinase gene led indirectly to the identification of two tyrosinase-related proteins (now known as TYRP1 and DCT); one of those was quickly shown to have the enzymatic activity of DOPAchrome tautomerase (Tsukamoto et al., 1992). Orlow and colleagues then demonstrated that the three tyrosinase-related melanogenic enzymes polymerized in a complex within melanocytes that facilitated their physiological interactions (Orlow et al., 1994).

Because of its critical role in pigmentation, and the disruptions in normal pigmentation expected to arise from mutations in its encoding gene, Kwon and colleagues initially cloned the gene encoding tyrosinase (Kwon et al., 1987) as well as another key melanosomal protein now known to be critically involved in melanosome structure, Pmel17 (Berson et al., 2001). As noted above, cloning of the tyrosinase gene led to the cloning of two other closely related genes, and a number of groups were instrumental in identifying human pigmentary diseases associated with each of those genes, most notably by the laboratories of King and Spritz in the United States (Getting and King, 1994; Spritz, 1994) and later by many other groups (reviewed in Hearing and Leong, 2005; Nordlund et al., 2006). Interestingly, the four known forms of oculocutaneous albinism result from molecular lesions that disrupt the function of tyrosinase: OCA1, the most severe form, results from mutations in the gene encoding tyrosinase itself, but OCA2, OCA3, and OCA4 (slightly milder forms) result from mutations in genes that affect the processing and trafficking of tyrosinase to melanosomes (P, TYRP1, and MATP, respectively) (Toyofuku et al., 2001; Costin et al., 2003). Many other pigmentary disorders affect other basic processes in addition to their effects on melanocytes and melanin production, most of them by virtue of disrupting intracellular trafficking pathways involved in organelle biogenesis (including melanosomes) and/or the intracellular transport or transfer of melanosomes to neighboring keratinocytes. Such disorders include Hermansky-Pudlak syndrome, Griscelli syndrome, and Chediak-Higashi syndrome. Interested readers are referred to an actively curated web site that lists pigment-related genes and

**Figure 1.** Scheme showing the melanogenic pathway that occurs within melanosomes, leading to the production of eumelanin and/or pheomelanin. Abbreviations used are as defined in the text. Adapted from Expert Review of Dermatology 2011;6:97–108, with permission from Expert Reviews (Kondo and Hearing, 2011).
associated diseases (http://www.espcr. org/micemut/).

One final consideration that is now becoming more widely accepted is the synthesis and function of melanosomes in less obscure tissues than the skin, hair, and eyes. Melanocytes do occur, usually as minor populations, in a wide variety of other tissues, and their functions are currently a matter of conjecture and study. Among those other tissues are the inner ear, the substantia nigra, the heart, and adipose tissue. Evidence is accumulating that the melanosomes play important protective roles in those tissues (Brito and Kos, 2008; Zecca et al., 2008; Randhawa et al., 2009). A recent article summarizes many of those studies and provides hints of interesting directions they might take in the future (Brenner and Hearing, 2009).

Melanizes play important roles in human skin for cosmetic appearance, detoxification, and photoprotection, among other functions. In lower species, melanizes play critical roles in survival (e.g., in camouflage of prey and predator, in thermal regulation in amphibians, etc.) In humans, melanin content (and thus visible pigmentation of the skin) has a dramatic effect on skin’s resistance to UV radiation damage, and the risk of skin cancers in lighter skin is 30–40-fold higher than in darker skin. The roles of melanizes in other tissues are still being studied and will no doubt provide a wealth of information about their various functions in the skin as well.

CONFLICT OF INTEREST
The author states no conflict of interest.

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