II. Melanins, Melanogenesis and Skin Photoprotection

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IN RECENT years, investigations concerning the nature and origin of epidermal melanins have advanced materially, and through numerous channels, so has our knowledge and appreciation of the field [1]. Although many things remain to be explained, it now seems clear that normal skin pigmentation is the result of a complex interplay of enzymes and co-factors, some of which are genetically encoded in melanocytes, while others appear to act extracellularly and to control the amount of melanin formed [2, 3]. Amazingly, it is mainly because of the action of these epigenetic factors, especially the redox state of the glutathione system, that normal human skin may vary in colour from what is called a flesh colour to a very dark brown or black.

Experimental data in support of this view were first reported by Halprim and Ohkawara [4] who found that the levels of glutathione (GSH) and of the enzyme GSH reductase are significantly lower in black skin than in white skin. They also went on to show that in normal people hyperpigmentation following UVB irradiation is accompanied by a fall in the GSH and GSH reductase levels, and a corresponding rise in the level of oxidised glutathione. At that time, however, little was definitely known about the chemistry of melanogenesis, and the reciprocal relationship between skin pigmentation and GSH level was ascribed to the ability of the SH group to inhibit tyrosinase by combining with the copper atoms at the active site. Such a mechanism, which is still alive in recent literature [5], has been challenged by the demonstration that the mode of action of GSH is not related to inhibition of tyrosinase but rather to its ability to react with enzymically generated dopaquinone to form colourless glutathionyldopas (GSHdopa), so that little or no melanin can be formed [6, 7]. The mechanism of this reaction is schematically illustrated in Fig. 1.

There are grounds to believe that the same pattern of GSHdopas is produced in melanocytes and that an effective

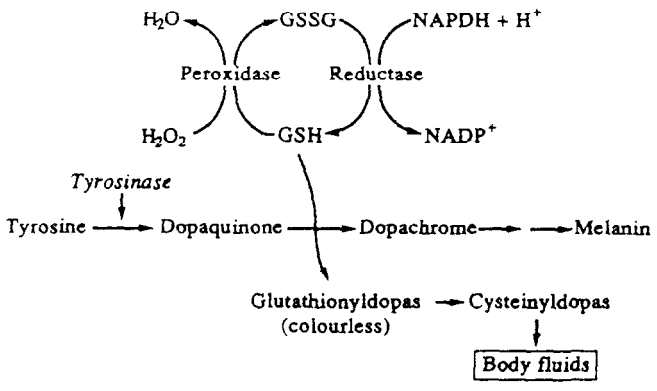

Fig. 1. Regulatory role of the glutathione system in melanin pigmentation.

mechanism exists which converts them into the corresponding cysteinyldopas, also found in pigment cells as well as in blood and urine [8–10]. Overall, formation and metabolism of GSHdopas provide a most interesting mechanism for controlling the total amount of pigment formed in melanocytes. However, other factors must also be responsible for the marked variation in melanogenic activity observed in different skin and the tendency to sunburn and to tan. Clearly, we need to learn more about the role of the GSH in skin photoprotection, and what other enzymes, in addition to GSH reductase, control the later stages of the melanisation process. Reports have appeared suggesting that peroxidase, superoxidase dimutase (SOD) [11], catalase [12] and the thioredoxin reductase/thioredoxin system [13] may all play a critical role in both constitutive and facultative skin pigmentation, but this awaits confirmation.

Another and perhaps more important factor to consider is the type of melanin that may be in the epidermis of people with different skin colour. The general view [14] is that whatever the phenotypic expression, epidermal melanocytes can only form a single type of black to brown melanin by oxidative polymerisation of various indolic precursors, mainly 5, 6-dihydroxyindole (DHI) and to a lesser extent 5, 6-dihydroxyindole-2-carboxylic acid (DHICA). It is also believed that such a polymeric material has a constant composition and is extremely stable so that, once formed, it is transferred to the cluster of keratinocytes without significant changes in its chemical properties. Hence, the view that the polychromy of skin pigmentation is mainly dependent on variation in the quantity, size and distribution of pigment granules produced and transferred to the cluster of keratinocytes forming the epidermal melanin units.

There are, however, old and new data suggesting that what is commonly referred to as "melanin" or, according to an improved terminology, eumelanin is by no means a definite chemical species, but a collection of molecules at different degrees of polymerisation and oxidation. This implies that various forms of eumelanins can be found at different sites within the skin which may interact differently with UV radiation. Thus, it is **the quality rather than, or in addition to, the quantity of the pigment in the epidermis which is critical for case–control studies linking skin colour with melanoma risk.** The same arguments could probably explain why not all black skinned people are exempt from sunburn and tend to avoid sunning although their skin is heavily pigmented [15].

To further complicate the picture, there are grounds to believe that two other groups of melanins, with different structural and sunscreening properties, may also be present in human skin:
(1) The sulfur-containing pheomelanins, which are reddish brown polymers or more precisely, mixtures of polymers arising by oxidative polymerisation of cysteinyldopas, and (2) the oxymelanins of similar colour, which are devoid of sulfur and

appear to be structural variants of eumelanins, arising by partial peroxidative cleavage of the 5, 6-dihydroxyindole units [1].

Of the two types of pigments, pheomelanins seem to be less effective than oxymelanins in terms of skin photoprotection, but this is more a matter of surmise than experimental proof. Indeed, we have virtually no data on the photobiological and photochemical properties of isolated native pheomelanins and, especially, of oxymelanins. The difficulties arise from the adverse properties of these pigments which make their isolation and characterisation a most challenging task.

At present, there are no satisfactory tests for distinguishing between pheomelanins and oxymelanins. Pigment colour and solubility in alkali are by no means specific, as well as the ultrastructure of the melanosomes which may be quite similar. It, therefore, seems likely that some of clinical and epidemiological data, relating to pheomelanic phenotypes, might refer in fact to oxymelanic subjects and *vice versa*, which accounts for exisiting confusion regarding skin phototypes, sun exposure and melanoma.

In closing, it can be said that it is a period of renewed inquiry into certain misconceptions and generalisations about melanin skin pigmentation and photoprotection, which have long dominated the field.

WHAT ARE THE DIRECTIONS FOR FUTURE RESEARCH?

1. There is an urgent need to incorporate all the new basic information on melanins and melanogenesis into the current programmes on skin photoprotection and melanoma control.
2. Studies are needed to define more precisely the redox state of the glutathione system in human skin and to evaluate how this relates with the UV susceptibility trait in dark and fair complexioned subjects.
3. A multidisciplinary approach to the development of appropriate technologies for assessment of the amount and type of melanins in human skins of different colour.
4. Pheomelanins versus oxymelanins as risk factors in fair complexioned groups of Anglo-Saxon and Celtic origin.
5. Improved animal models and protocols for studying the putative role of sun exposure in the aetiology of melanoma.

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III. Immunology of UV-Irradiated Skin

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THE OBSERVATION that ultraviolet (UV) B radiation (290–315 nm) is capable of affecting the skin's immune system gave rise to a novel discipline of biomedical research termed photoimmunology, which investigates the interaction of non-ionising electromagnetic radiation, in particular UVB light, with the immune system [1]. Interest in photoimmunology originated from the observation that UVB radiation is capable of suppressing selected cell-mediated immune responses, including immunity to UVB-induced skin cancer, thereby facilitating the growth of UV-induced skin tumours [2]. There is growing evidence that UVB-induced immunosuppression may be of relevance for the development of both non-melanoma skin tumours (e.g. fibrosarcomas, squamous cell carcinomas) and cutaneous melanomas [3, 4]. The capacity to affect cell-mediated immune responses is not specific for immunity against skin cancer, since UVB light was found to alter the immune response to contact-sensitising agents, to host tissue in graft versus host disease, and to certain microorganisms such as viruses, bacteria, fungi or protozoa [1, 2].

Over the last few years, substantial progress has been made to elucidate the mechanisms responsible for UVB-induced immunosuppression [1]. From these studies it appears that UVB light exerts its immunomodulatory effects not just through one, but rather through an array of interacting mechanisms. Specifically, both direct effects on immunocompetent cells at the irradiation site and indirect effects caused by the release of soluble