ON THE MECHANISM OF MELANIN FORMATION BY THE ACTION OF ULTRAVIOLET RADIATION*

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Raper (1) and his associates showed in a comprehensive study of the tyrosine-tyrosinase reaction that the enzyme tyrosinase obtained from plant sources catalyzes the conversion of tyrosine to dopa (3,4-dihydroxyphenyl-l-alanine). Dopa is then oxidized, in the presence of the same enzyme, to a red indole derivative which changes spontaneously to melanin.

Melanin formation in mammals has remained somewhat of an enigma, despite the demonstration by Bloch (2) of an intracellular oxidase which catalyzes the oxidation of dopa to melanin but is without apparent effect on tyrosine. The presence of dopa in mammalian tissue or fluids has never been demonstrated and it has not been known how tyrosine is converted into dopa. Recently, however, the enzymatic formation of melanin from tyrosine has been conclusively demonstrated in several types of mammalian tissue, including beef ciliary bodies (3), and the human (4), horse (5) and mouse (6-8) melanomas. The enzyme has been shown to require copper for its activity (9) and to be firmly bound to cellular particulates (8). With the demonstration, therefore, of tyrosinase activity in various mammalian tissues, a scheme such as the one worked out by Raper and outlined in the first paragraph may also describe the mechanism of melanin formation in mammals.

Arnow (10) showed that ultraviolet irradiation of aqueous solutions of tyrosine over a period of four hours results in the formation of small amounts of dopa, as detected colorimetrically (11). He postulated that when skin is irradiated with ultraviolet light, tyrosine in the skin is converted to dopa and the dopa-oxidase in the melanoblasts then catalyzes the oxidation of dopa to melanin. Rothman (12) pointed out that the long period of exposure to ultraviolet radiation required for the conversion of significant amounts of tyrosine to dopa is not consistent with the phenomenon of tanning, which occurs after a shorter exposure. He demonstrated in vitro that addition of small amounts of ferrous salts catalyzes the conversion of tyrosine to dopa by ultraviolet irradiation, thereby reducing the necessary duration of exposure. He, therefore, suggested that ferrous salts or some unknown factors are present in the skin to catalyze this photochemical reaction. Recent results that we have obtained with tyrosinase from extracts of the Harding-Passey melanoma (8) indicate the possibility of an alternate explanation for the relatively short time of exposure to ultraviolet radiation required in vivo for significant formation of pigment. We have found that in the presence of trace amounts of dopa, tyrosine itself is rapidly and completely oxidized to form melanin by enzyme preparations which, in the absence of the dopa, have little or no action on tyrosine although they are highly active against dopa itself. It is, therefore, necessary to form only trace amounts of dopa in the presence of the enzyme and an adequate supply of tyrosine, for formation of pigment to ensue at a rapid rate. Experimental evidence for the validity of this hypothesis, based on a study of the effect of ultraviolet irradiation on the ability of mammalian tyrosinase to oxidize tyrosine to melanin will be reported elsewhere.

The recent experiments of Rothman and associates (13) with aqueous extracts of human epidermis suggest a further, and perhaps equally important, factor in the formation of

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melanin by ultraviolet light. They were able to demonstrate inhibition of the enzymatic oxidation of tyrosine to melanin by plant tyrosinase in the presence of aqueous extracts of human epidermis. This inhibition was abolished by compounds which combine with —SH groups and by ultraviolet irradiation of the extracts (14). These experiments indicate that the inhibitory effect of human epidermis is due to the presence of sulfhydryl groups which probably bind the copper required for enzymatic activity, and that ultraviolet irradiation may play a dual role in the formation of skin pigment.

The interrelationships of the various factors discussed in previous paragraphs which are responsible for the formation of melanin from tyrosine by ultraviolet irradiation are schematically illustrated in the figure.

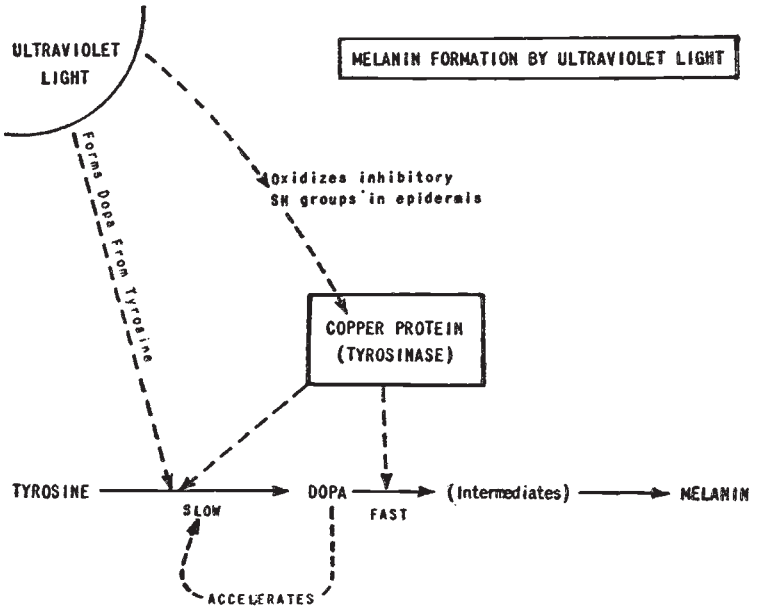

FIG. 1

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