Plasma 5-S-Cysteinyldopa Concentrations in Oculocutaneous Albinism

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Nimmo JE, Hunter JAA, Percy-Robb IW, Jay B, Phillips CI, Taylor WOG. Plasma 5-S-cysteinyldopa concentrations in oculocutaneous albinism. Acta Derm Venereol (Stockh) 1985; 65: 169–171.

5-S-cysteinyldopa concentrations were determined by high-pressure liquid chromatography and electrochemical detection in plasma from normally pigmented patients and patients with oculocutaneous albinism, both tyrosinase-positive and tyrosinase-negative. The plasma 5-S-cysteinyldopa concentrations were similar in all three groups, suggesting that 5-S-cysteinyldopa can be produced by mechanisms which do not involve tyrosinase.
Key words: Tyrosinase; Melanin. (Received September 4, 1984.)

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Human oculocutaneous albinism is of two main types: tyrosinase-positive and tyrosinase-negative albinism. These can be distinguished on the basis of both clinical features, and presence or absence of tyrosinase activity in the hairbulbs (1).

Tyrosinase is thought to be essential for the first two steps in melanin synthesis. These are the hydroxylation of tyrosine to form dihydroxyphenylalanine (dopa) and the subsequent oxidation of dopa to form the highly reactive intermediate dopaquinone. The pathway leading to formation of the red/yellow phaeomelanins involves the spontaneous coupling of dopaquinone with cysteine to form a number of different cysteinyldopas of which quantitatively the major component is 5-S-cysteinyldopa (5-S-CD) (2).

5-S-CD can be detected in the urine of normal healthy individuals, regardless of skin or hair colour (3). Patients lacking demonstrable tyrosinase activity would be expected to be incapable of synthesising melanin or its precursors, including 5-S-CD.

We report the results of a study in which we measured plasma 5-S-CD concentrations in both tyrosinase-positive and tyrosinase-negative patients with oculocutaneous albinism to see if it was possible to distinguish between the two groups on the basis of this measurement.

MATERIALS AND METHODS

There were twenty patients in the study, all with oculocutaneous albinism. They were divided into ten tyrosinase-positive and ten tyrosinase-negative albinos, on the basis of clinical assessment either on its own or, in a few cases, accompanied by a hairbulb tyrosinase test (4).

Controls were ten healthy volunteers of both sexes with a wide range of hair colour.

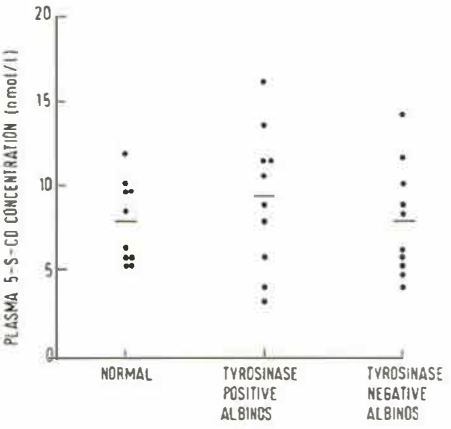

Fig. 1. Plasma 5-S-CD concentrations in albinos and controls.

Sampling from these 3 groups was not restricted to any particular time of year, as we had previously demonstrated that the increases in plasma 5-S-CD levels observed during the summer months in Edinburgh, although significant, were not of sufficient magnitude to invalidate any results obtained during these months (5).

Blood samples were collected into lithium heparin tubes containing dithiothreitol to a final concentration of 5 mmoles/l. 5-S-CD was extracted from the plasma by adsorption onto alumina at pH 8.6 followed by elution with 0.2 M $HClO_4$. 5-S-CD determination was by high-pressure liquid chromatography (HPLC) with electrochemical detection (6), and using 5-S-D-cysteinyl-L-dopa as internal standard.

RESULTS

The plasma concentrations of 5-S-CD found in controls and albino patients are given in Fig. 1.

Statistical analysis of the results using an independent *t*-test, showed that the three groups of results were not significantly different from one another.

DISCUSSION

We have found no significant difference between plasma concentrations of 5-S-CD in normally pigmented controls and in patients with oculocutaneous albinism, or between 5-S-CD concentrations in tyrosinase-positive and tyrosinase-negative albinos.

The finding of measurable, and indeed apparently normal concentrations of 5-S-CD in plasma from tyrosinase-negative albinos is perhaps surprising in the light of a demonstrable lack of active tyrosinase in the hairbulbs of these patients (1). However, Aquaron et al. were able to demonstrate urinary excretion of 5-S-CD in both tyrosinase-positive and tyrosinase-negative albino negroes in the sunny climate of Cameroon (7).

5-S-CD has been shown to be present in non-melanogenic tissues, such as liver, kidney and brain, of mice and rats, as well as in hair of albino mice (8). In these cases the amino acid is present mainly in a protein-bound form, free 5-S-CD being excreted in the urine. The finding of 5-S-CD in these tissues, in which tyrosinase is not detectable, indicates that it can be produced by some oxidation mechanism which clearly does not involve tyrosinase, although the importance of this enzyme in melanin production within melanocytes is not open to question (2).

Similar tyrosinase-independent pathways have also been proposed as a result of work on guinea-pig tissues, including kidney, spleen, heart and sympathetic ganglia, and on the

ganglion stellatum of the cow (9, 10). Ito et al. have, in fact, demonstrated the formation of cysteinyldopas in vitro from dopa and cysteine by a number of systems not involving tyrosinase, including peroxidase-H_2O_2, and superoxide and hydroxyl radicals (11, 12, 13).

It seems likely that the presence of protein-bound 5-S-CD in tissues of mice and rats, including the albino mouse, is due to its synthesis from protein-bound dopa, perhaps by one of the non-specific oxidation mechanisms previously mentioned. Protein-bound dopa is found in high concentrations in these tissues relative to the concentrations of free dopa, and is presumably produced by the action of the enzyme tyrosine hydroxylase on tyrosine. Turnover of the 5-S-CD-containing proteins would account for the excretion of the amino acid in the urine of the rats and mice studied, including the tyrosinase-negative albino mice, in which urinary excretion of 5-S-CD was found to be very similar to that in pigmented animals. Without further investigation in humans it is difficult to explain our observations in tyrosinase-negative albinos, but production of 5-S-CD in these individuals would certainly appear to be by some non-tyrosinase-dependent oxidation pathway, and could perhaps involve one or more of those suggested by 5-S-CD production in non-melanogenic tissues. Reactive oxygens, for instance, are known to be produced in many biological processes.

The findings in human albinos are certainly comparable to findings in albino mice, and it may be that similar mechanisms for production of 5-S-CD are operating in the two species.

ACKNOWLEDGEMENTS

We would like to thank the Melville Trust for their financial support, and Professor Hans Rorsman for advice and the gift of a sample of 5-S-L-cysteinyl-L-dopa.

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