Abnormal tryptophan pyrrolase and amino acids related to melanogenesis in vitiligo[1]

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Summary. A comparison of the serum tyrosine, DOPA, tryptophan, tyrosinase and tryptophan pyrrolase levels of vitiliginous patients with those of normal subjects show abnormalities in all these parameters. As the clinical diagnosis of vitiligo may be made without difficulty, these parameters appear to be of little diagnostic value in vitiligo. But they may be considered as additional biochemical parameters in vitiligo.

The urinary indoles of vitiliginous patients have been reported to have an abnormal profile, containing 5-hydroxyanthranilic acid and higher amount of 5-hydroxytryptophan[3]. Chakraborty et al.[4] have shown that after oral administration of hydroquinone in *Bufo melanostictus*, there is depigmentation of its skin which recovers after oral administration of psoralen. During these changes, the tyrosinase and tryptophan pyrrolase activities in the skin and liver of *Bufo melanostictus* were found to be inversely related. It has further been observed that oral administration of excess tryptophan causes increased tryptophan pyrrolase activity in both the skin and liver of *Bufo melanostictus* with a concomittent decrease in melanogenesis[5]. These observations prompted us to measure tyrosine, DOPA and tryptophan levels as well as the levels of 2 enzymes, tyrosinase and tryptophan pyrrolase in the blood of vitiliginous patients.

Materials and methods. Only patients with extensive vitiligo were selected. Blood and 24 h urine from 13 normal human subjects and vitiligo 13 patients (age 18–40) irrespective of sex were collected. For the estimation of tyrosine, DOPA and tryptophan, the serum was first deproteinized with trichloroacetic acid (20%), centrifuged and then neutralized. Tyrosine was estimated in the urine and in protein-free filtrate of serum according to Udenfriend et al.[6] using α-nitroso-β-naphthol. DOPA was estimated according to Arnow[7] using nitrite molybdate reagent. Tryptophan was measured with p-dimethylamino benzaldehyde according to Spies[8].

Serum tryptophan pyrrolase activity was measured according to Knox[9] as slightly modified by Spiegel[10]. The enzyme activity was measured under the following conditions: 0.2 M phosphate buffer, pH 7.0 (1 ml), double distilled water (0.7 ml), 0.03 M L-tryptophan (0.3 ml), serum (0.2 ml). The enzyme activity is expressed in terms of µM of kynurenine per ml of serum. Tyrosinase activity was measured according to Pomerantz[11] by measuring the rate of formation of dopachrome from L-DOPA at 37 °C under the following conditions: L-DOPA (1 µM), sodium phosphate buffer, pH 7.4 (35 µM), enzyme (0.2–0.3 units), total 1 ml. The enzyme activity was expressed as µM per ml of serum.

Results and discussion. There is an increase in the levels of tyrosine and DOPA in the serum and urine of vitiliginous patients as compared with those of control subjects (table 1). The excess tyrosine in the serum may represent the tyrosine released by the skin proteins from the depigmented patches[10]. The higher levels in urinary tyrosine may be due to a lesser efficiency of the vitiliginous subjects in metabolizing tyrosine to CO_2 and water; so the increased amounts of tyrosine and DOPA are channelled through the urinary pathway. A higher level of DOPA suggests that DOPA formation is not affected in vitiliginous subjects.

Table 1. Tyrosine, DOPA and tryptophan level in the serum and urine of vitiliginous patients in comparison to those of normal subjects (n = 13)

Subjects	Tyrosine* (mean ± SD)		DOPA* (mean ± SD)		Tryptophan* (mean ± SD)	
	Serum (µg/ml)	Urine (mg/24 h)	Serum (µg/ml)	Urine (mg/24 h)	Serum (µg/ml)	Urine (mg/24 h)
Normal	11.1 ± 4.5	18.2 ± 2.9	12.3 ± 5.1	24.0 ± 3.2	16.3 ± 3.1	15.0 ± 2.5
Vitiligo	24.0 ± 3.7	45.3 ± 4.1	28.0 ± 4.7	90 ± 2.4	32.0 ± 4.3	34.0 ± 3.8

*$p < 0.001$.

Table 2. Tyrosinase and tryptophan pyrrolase activities in the serum of vitiliginous patients in comparison to those of normal subjects (n = 13)

	Tyrosinase* (mean ± SD)	Truptophan pyrrolase** (mean ± SD)
Normal	141 ± 7	118 ± 13
Vitiligo	152 ± 8	183 ± 17

*µM of dopachrome/min/ml of serum ($p < 0.001$); **µM of kynurenine × 10^{-3} ml of serum ($p < 0.001$).

Higher level of tryptophan (table 1) may cause higher tryptophan pyrrolase activity in vitiliginous serum (table 2) which may impair, melanogenesis in the skin[5]. Higher tyrosinase activity in the serum of vitiliginous subjects (table 2) can be considered to be derived from disordered melanocytes by the action of melanocytotoxic agent or any inhibitor at the site of melanin synthesis.

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