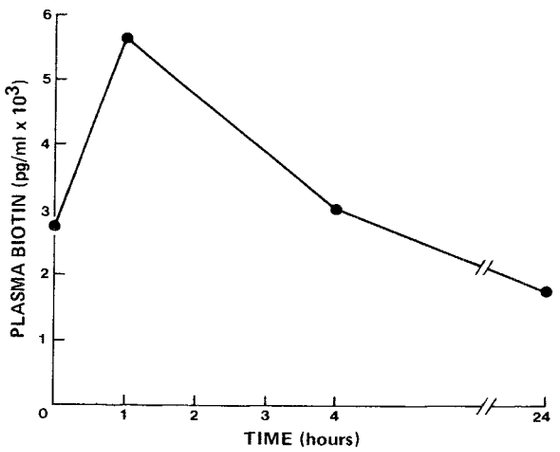
BIOTINIDASE DEFICIENCY IN JUVENILE MULTIPLE CARBOXYLASE DEFICIENCY

SIR,—Juvenile biotin-responsive multiple carboxylase deficiency (MCD) is characterised by onset, after the neonatal period, of lacticacidosis, alopecia, ataxia, seizures, and erythematous rash. Plasma and urinary biotin concentrations are subnormal and all abnormal clinical and biochemical findings are reversed by *d*-biotin.[1] This form of MCD is thought to be caused by a defect in biotin absorption or transport;[2,3] the neonatal form is due to defective biotin holocarboxylase synthetase activity.[4] Recently, deficient activity of serum biotinidase was demonstrated in several patients with the juvenile phenotype, and intermediate activities were found in the serum of their parents. This suggests that biotinidase deficiency is the primary enzyme defect in some patients with juvenile MCD.[5] Moreover, patients with biotinidase deficiency whose tissues are biotin-depleted might fail to exhibit a normal plasma biotin response when given oral biotin,[6] explainable by the rapid entry of biotin into the tissues. To test this hypothesis we evaluated the plasma biotin response after administration of a small amount of biotin to a patient with biotinidase deficiency whose initial plasma biotin concentration was not subnormal.

This 6½-year-old girl with biotin-responsive MCD[1] and defective absorption of biotin[2] had normal fibroblast holocarboxylase



Response of plasma biotin to ingestion of 5 μg/kg of biotin when patient was not biotin deficient.

synthetase activity. She has been on oral biotin (10 mg daily) since 31 months of age and at the time of this study she weighed 25 kg (90th percentile) and was 118 cm tall (50th percentile). 8 days before this study, while the child continued to ingest a normal diet, biotin supplements were stopped. She then received a low oral dose of biotin (5 μg/kg) and her plasma biotin concentrations were measured over 24 h. Urine was collected to determine renal biotin clearance and fractional biotin excretion. Biotin concentrations were measured by competitive protein binding assay.[7] Biotinidase activity was assessed colorimetrically by measuring *p*-aminobenzoate liberation from N-biotinyl-*p*-aminobenzoate.[8]

Previously,[2] after 42 days of biotin deprivation, an erythematous rash had developed with very low plasma biotin concentration (143 pg/ml, mean normal concentration 520±220 SD), and she had excreted in her urine only 4·3–15% of the ingested biotin dose in 24 h (normal value 25–28%) and had a renal biotin clearance of 54·9 ml/min/1·73 m^2 (normal 38·8±6·5). These findings were interpreted as indicating that biotin absorption both in the intestinal tract and in the renal tubules was abnormal. However, when the patient was tested at a time when her tissues were biotin replete (achieved by withholding biotin for only 8 days) her biotin response was normal. The figure shows that at the start of the study her plasma biotin was slightly above normal range (2720 pg/ml). However, 1 h after ingestion of 5 μg/kg of biotin, her plasma biotin concentration rose to 5650 pg/ml, as in similarly treated biotin-replete controls.[2] Biotin clearance was also normal (34·4 ml/min/1·73 m^2). The fraction of biotin excreted in her urine in the 24 h after the biotin dose was 50·8%, twice as high as the amount excreted by the controls and more than three times her previous maximum excretion (15%).

BIOTINIDASE ACTIVITY IN PATIENT AND MOTHER

—	Biotinidase activity (nmol/min/ml serum)	% of control mean
Patient	Not detectable	0
Mother	2·66	46
Controls (n = 18): mean (range)	5·80 (4·30–7·54)	100

Biotinidase activity in the patient's serum was undetectable (table). The mother, who does not have clinical biotin deficiency, had 46% of mean normal biotinidase activity and a plasma biotin of 248 pg/ml, which is below normal.

Dietary biotin exists primarily as biotinyl groups linked to the ε-amino groups of lysine residues of proteins.[9] Biotinidase hydrolyses this bond, releasing free biotin. Biotinidase activity is widely distributed, particularly in liver, kidney, serum, and gut mucosa.[9] Although this enzyme's primary site of action on dietary protein is not known, deficiency could impair ability to maintain normal body stores of free biotin, leading to a hypobiotinaemic state. Whether biotinidase is also involved in the absorption of free biotin is not known.

The patient's normal plasma biotin response to the ingestion of 5μg/kg of biotin when her tissues were replete with biotin, but her failure to respond when she was biotin deficient is probably caused by rapid entry of biotin into the depleted tissues. Similar results have been obtained in thiamine-deficient patients.[10,11] Rapid saturation of deficient tissues with the vitamin results in subnormal increases in the plasma concentrations, mimicking an apparent defect in intestinal absorption.

Juvenile MCD probably results from impaired generation of free biotin from biotinyl residues of dietary protein. Presumably the child is born with normal stores of free biotin but, once dependent on dietary protein-bound biotin, a child with biotinidase deficiency would become hypobiotinaemic and clinical symptoms of biotin deficiency would ensue. This mechanism could account for the late onset of acute symptoms (as opposed to the neonatal onset of symptoms in holocarboxylase synthetase deficiency) and, for some of the clinical variability.

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