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Saccharopinuria

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Simell, O., Visakorpi, J. K., and Donner, M. (1972). *Archives of Disease in Childhood*, **47**, 52. **Saccharopinuria.** A girl with spastic diplegia excreted large amounts of lysine and saccharopine in the urine, and had more than 10 times the normal plasma lysine concentration and a saccharopine peak in the plasma amino acid chromatogram. Unlike the earlier reported case of saccharopinuria, this patient had normal plasma and urine levels of citrulline. This case affords further evidence that the main degradative pathway of lysine metabolism in man is via saccharopine and α-aminoadipic acid.

The fact that in this patient there is no other known cause of the spastic diplegia and that the diplegia seems to be progressing suggest a connexion with the metabolic disturbance.

In 1968, Carson *et al.* described the first patient with a new inborn error of lysine metabolism, saccharopinuria. This report presents a further case of this abnormality. A girl, aged 3 years and 6 months, was admitted to the hospital for study of spastic diplegia and was found to excrete large amounts of lysine and another substance, which was shown to be saccharopine. In this patient no excess citrulline could be detected in the plasma or urine.

Case Report

The patient was a girl aged 3 years and 6 months. She was born normally at term, Apgar score 9. Birthweight was 3640 g. The child was mildly yellow for the first few days, and the serum bilirubin was 13·6 mg/100 ml on the fourth day. Her motor development was a little slow, and she walked at the age of 16 months. From the beginning it was noticed that she had difficulty in walking, and from the age of 2 years this has grown worse; she only walks a few steps without support.

Physical examination. At presentation, the patient had spastic diplegia with spasticity especially in the calf muscles and hip adductors. Dorsiflexion of the feet was difficult and the feet were in the valgus position. She walked with knees and hips somewhat flexed (Fig. 1). There was some clumsiness in the upper extremities and some involvement of the throat muscles, with dribbling, difficulty in swallowing, and mild dysarthria. The tendon reflexes were exaggerated and there was ankle clonus; the abdominal reflexes were normal. At the same time there was some weakness of the trunk and hip muscles and possibly slight ataxia.

Somatically she was otherwise normal for her age. Her hair was a beautiful red colour and the skin and nails normal. The liver and spleen were not palpable and the height, weight, and head circumference had followed the line of the 50th centile of the tables for Finnish children since birth. Her mental development was slightly retarded and she had difficulty in concentrating.

Laboratory findings. The following laboratory tests gave normal results: haematological examination,

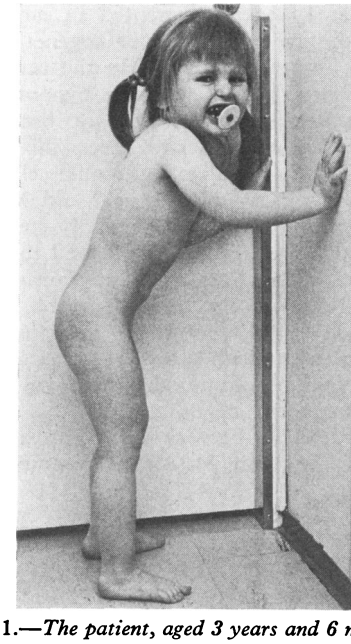

FIG. 1.—*The patient, aged 3 years and 6 months.*

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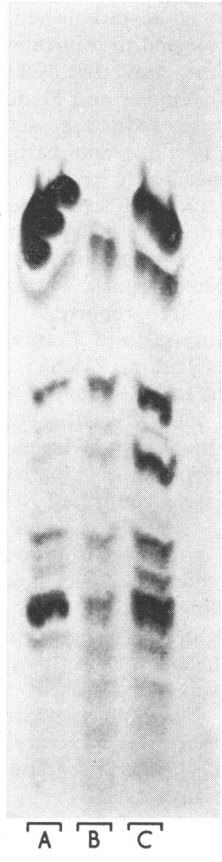

FIG. 2.—*High voltage electropherogram from the urine of (A) saccharopinuric patient, (B) normal control, and (C) patient with lysinuric protein intolerance (Perheentupa and Visakorpi, 1965). The pH of the buffer was 2·1. The symbols are: (1) tyrosine, (2) glutamine, glutamic acid, citrulline, and saccharopine, (3) threonine, (4) serine, (5) alanine, (6) glycine, (7) histidine, (8) lysine and ornithine.*

serum electrolytes, calcium, inorganic phosphate, alkaline phosphatase, blood sugar, serum protein and electrophoretic pattern of the serum proteins, aspartate aminotransferase, alanine aminotransferase, protein-bound iodine, capillary *p*H, and urinanalysis. Blood ammonia and urea were normal both after fasting and postprandially.

Radiological examination revealed no osteoporosis. The electroencephalogram has been recorded three times between the ages of 2 and 3 years and has been normal or slightly abnormal with some excess of slow activity. No recordings have been made during sleep. Pneumoencephalography revealed dilatation, especially of the middle part of the lateral ventricles and of the pontine cistern, as a sign of moderate central and basal atrophy, while the distribution of the gas over the hemispheres was normal. Neuro-ophthalmological examination gave normal results.

In high voltage electrophoresis of urinary amino acids (Visakorpi, 1960) conspicuous lysinuria was found and what was at first thought to be glutamic aciduria (Fig. 2). In ion exchange column chromatography (Spackman, Stein, and Moore, 1968) this fraction was seen to move exactly like saccharopine.* A peak in the position of α-amino-adipic acid was also seen. In the serum, more than 10 times the normal amount of lysine was seen and traces of saccharopine too. Urinary and plasma citrulline were normal. The amino acid levels in the plasma of the patient are presented in the Table.

TABLE

Plasma Amino Acid Concentrations (μmoles/ml) in Patient with Saccharopinuria

Amino Acid	Patient with Saccharopinuria	Normal Children (range)*
Alanine	0·028	0·173–0·305
α-aminoadipic acid	0·000	0·000
Arginine	0·050	0·023–0·086
Aspartic acid	0·002	0·004–0·023
Asparagine + glutamine	0·526	0·057–0·467
Citrulline	0·015	0·012–0·055†
Cystine	0·091	0·048–0·140†
ε-N-acetyl-lysine	0·000	0·000
Glutamic acid	0·035	0·023–0·250
Glycine	0·173	0·117–0·223
Histidine	0·079	0·024–0·085
Homocitrulline	0·000	0·000
Isoleucine	0·103	0·028–0·084
Leucine	0·200	0·056–0·178
Lysine	1·320	0·071–0·151
Methionine	0·034	0·011–0·016
Ornithine	0·024	0·027–0·086
Phenylalanine	0·061	0·026–0·061
Proline	0·131	0·068–0·148
Saccharopine	Trace	0·000
Serine	0·157	0·079–0·112
Threonine	0·137	0·042–0·095
Tyrosine	0·061	0·031–0·071
Valine	0·028	0·128–0·283

*From Scriver and Davies (1965).
†From Dickinson, Rosenblum, and Hamilton (1965).

Family. The parents and sibs of the patient had normal urinary amino acid patterns and were clinically healthy. The parents are not aware of any consanguinity, but all the grandparents of the patient come from the same rather isolated community.

Discussion

Lysine has some peculiar metabolic features (Fig. 3). It does not participate in reversible transamination like other amino acids (Schoenheimer and Rittenberg, 1940; Weissman and Schoenheimer, 1941) and is incorporated directly into proteins (Clark and Rittenberg, 1951). The inactivation

*We had a standard of saccharopine as a generous gift from Nina Carson.

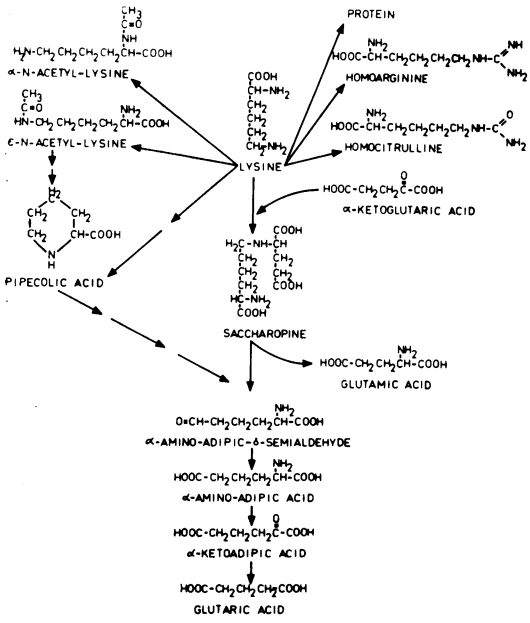

FIG. 3.—*Pathways postulated for lysine metabolism.*

of the ε-amino group of lysine by acylation leads to free deamination of the α-amino group (Paik, 1962). Using isotope methods and large amounts of pipecolic acid as a 'trap', Rothstein and Miller (1954) isolated a relatively high concentration of pipecolic acid from rat urine, and concluded that the pipecolic acid pathway was the major route of lysine metabolism in the rat. Rothstein and Greenberg (1960) showed that pipecolic acid forms α-aminoadipic acid and glutaric acid in the rat, and that rat liver mitochondria oxygenate pipecolic acid and α-aminoadipic acid to CO_2. Later on, it was verified (Grove *et al.*, 1969) by isotope trapping methods that in the rat L-pipecolic acid is formed from D- and L-lysine after removal of an α-amino group, and that D-lysine is a much better precursor of pipecolic acid than L-lysine. The authors stated that, in the metabolism of L-lysine, pipecolic acid might not be the main route. In 1968, Gatfield *et al.* reported a patient with greatly raised serum pipecolate and urinary pipecolic acid. In this patient there was no rise in pipecolic acid in the serum or urine after a peroral L-lysine load.

Saccharopine was first isolated from yeast (Darling and Larsen, 1961). It has been shown to be the precursor of lysine in yeast (Kuo, Saunders, and Broquist, 1964). In mammals, Higashino, Tsukada, and Lieberman (1965) noticed that saccharopine was formed by rat liver mitochondria from lysine on reaction with α-ketoglutarate. In 1967, Higashino *et al.* established that α-aminoadipic acid was formed in mitochondria of rat liver from saccharopine, and that NAD was required in the reaction. Hutzler and Dancis (1968) found lysine-ketoglutarate reductase activity in many human organs *in vitro* and partially purified it. This enzyme specifically requires TPNH for its action. In the human liver the activity of lysine-ketoglutarate reductase many times exceeds the ordinary dietary requirements.

In the published reports 7 cases of persistent hyperlysinaemia are reported (Woody, 1964; Ghadimi, Binnington, and Pecora, 1964; Woody and Ong, 1967; Ghadimi, Zischka, and Binnington, 1967; Armstrong and Robinow, 1967; Woody and Pupene, 1970). Dancis *et al.* (1969) have shown a clear decrease in the activity of lysine-α-ketoglutarate reductase in fibroplasts of 3 patients with hyperlysinaemia, and have confirmed that the metabolic block lies there. In 1968, Carson *et al.* described the first patient with saccharopinuria, and this girl had high levels of lysine, saccharopine, and, surprisingly, citrulline in the serum and urine. In the urine, traces of ε-N-acetyl-lysine were found, which possibly means that in this patient the pipecolic acid pathway serves as an overload route (Carson, 1969). The high citrulline led the authors to suggest that there was a relation between lysine degradation via the saccharopine pathway, citrulline, and the urea cycle.

Some features of our patient differ from those just described. Our patient has no hypercitrullinaemia, and lysine and saccharopine are higher in her plasma. The metabolic block in this patient may be between saccharopine and α-aminoadipic acid or between the latter and α-ketoadipic acid. Yet a third possibility exists, namely that transport of saccharopine to or from the mitochondria in the liver cells is prevented.

The findings in this patient and in the girl described by Carson *et al.* lend added support to the view that the saccharopine-α-aminoadipic acid pathway is the main route of lysine metabolism.

The fact that in this patient there is no other known cause of the spastic diplegia and that the diplegia seems to be progressing suggests its connexion with the metabolic disturbance.

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