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Hair Amino Acids: Normal Values and Results in Metabolic Errors

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van Sande, M. (1970). *Archives of Disease in Childhood*, **45**, 678. **Hair amino acids: normal values and results in metabolic errors.** Hair amino acids have been studied in 10 controls and in 25 patients suffering from metabolic inborn errors. Abnormalities of hair amino acid pattern were found in several cases. In phenylketonuria (7 cases), the hair phenylalanine level was significantly higher than that of the normal subjects. Glutamic acid was decreased. In homocystinuria (5 cases), the phenylalanine level was increased. The results suggest that a detailed examination of the hair proteins could give further information.

Inborn errors of metabolism are often associated with morphological hair changes: for example, the imperfect hair pigmentation in phenylketonuria, the changes in hair texture in Hartnup's disease, the poorly pigmented brittle hair (trichorrhexis nodosa) in several cases of argininosuccinuria, and the sparse, blond, and brittle hair in homocystinuria. The reason for the associated hair abnormalities in these diseases is not always clear.

Relatively little is known concerning the amino acid composition of human hair, either in normal patients or in those with inborn errors. Most data in the literature report isolated findings in pathological cases comparing them with only few control cases. This excludes any statistical evaluation of the results.

The purpose of the present study was to establish normal values and to compare them with the results obtained in 25 patients suffering from metabolic errors.

Material and Methods

Hair specimens were obtained from 10 normal subjects: 9 children (2 boys and 7 girls aged from 4 to 12 years) and 1 adult (a woman of 20 years), and from 25 patients suffering from metabolic errors. Details are given in Table I.

The hair specimens were washed with distilled water, light petroleum (B.P. 60–80 °C.), absolute ethanol, and diethyl ether. Hydrolysis of 10 mg. samples in 6N HCl (22 hours at 110 °C.) was carried out using the method of Moore and Stein (1963). The amino acid composition of the hydrolysates was determined quantitatively on a Technicon Amino Acid Analyzer (column 140 × 6 mm., Chromobeads type A). The elution system was that described by Efron (1965). The values obtained for the labile amino acids are extrapolated to zero time, calculated on companion hydrolysates heated for 70 hours. The concentration of the amino acids is expressed as g. nitrogen per 100 g. nitrogen recovered from column, ammonia excluded.

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Results

The mean values of the hair amino acids from the 10 control subjects and statistical data are given in Table II.

The results obtained on seven patients with phenylketonuria and five patients with homocystinuria have been statistically interpreted by means of the Student t test for small samples. For the other cases, the level of an amino acid is considered abnormal when the value is outside the 95% confidence limit of the control group. The findings are represented graphically in the Fig. Only abnormal values are indicated.

In several hair specimens, abnormal values for some amino acids were obtained.

In the two cases of argininaemia, the microscopical appearance of the hair showed some similarities with the cases of sex-linked neurodegeneration described by Menkes *et al.* (1962): pili torti, trichorrhexis nodosa, and irregular variations in thickness; however, in our two patients these abnormalities were less pronounced. In the oldest patient Case 2, who had been on a low protein diet for several months, no abnormalities of the hair amino acid

TABLE I

Details of Patients Studied

Case No.	Sex	Age (yr.)	Diagnosis	Therapy	Hair colour
1	F	1 6/12	Hyperargininaemia	Low protein diet	Blond, dark
2	F	5	Hyperargininaemia	Low protein diet	Blond, white
3	F	1 2/12	Cystinosis	Low methionine and cystine diet	Blond, yellow
4	F	12	Histidinaemia	None	Blond
5	M	59	Histidinaemia	None	Blond, dark
6	F	2	Glycinaemia without ketosis	None	Blond, dark
7	F	22	Saccharopinuria	None	Dark
8	F	4	Argininosuccinuria	None	Blond, dark
9	F	5	Argininosuccinuria	None	Blond, dark
10	F	3	Ammonia intoxication with ornithine carbamyl transferase deficiency	Low protein diet	Blond, dark
11	M	5	Homocystinuria	Pyridoxine	Blond, yellow
12	F	6	Homocystinuria	Pyridoxine	Blond, yellow
13	F	7	Homocystinuria	Pyridoxine	Blond, yellow
14	F	7	Homocystinuria	Pyridoxine	Blond, yellow
15	F	25	Homocystinuria	Pyridoxine	Blond, dark
16	M	1/12	Phenylketonuria	None	Blond
17	M	8/12	Phenylketonuria	None	Blond, yellow
18	M	2	Phenylketonuria	Diet	Blond, yellow
19	M	2	Phenylketonuria	Diet	Blond, yellow
20	M	5	Phenylketonuria	Diet	Blond, yellow
21	M	5	Phenylketonuria	Diet	Blond, yellow
22	F	6	Phenylketonuria	Diet	Blond, dark
23	M	16	Late infantile amaurotic idiocy (neuronal ceroid lipofuscinosis)	None	Dark
24	F	11	Monilethrix and mental retardation	None	Blond, white
25	M	3	Trichorrhexis nodosa and mental retardation	None	Dark

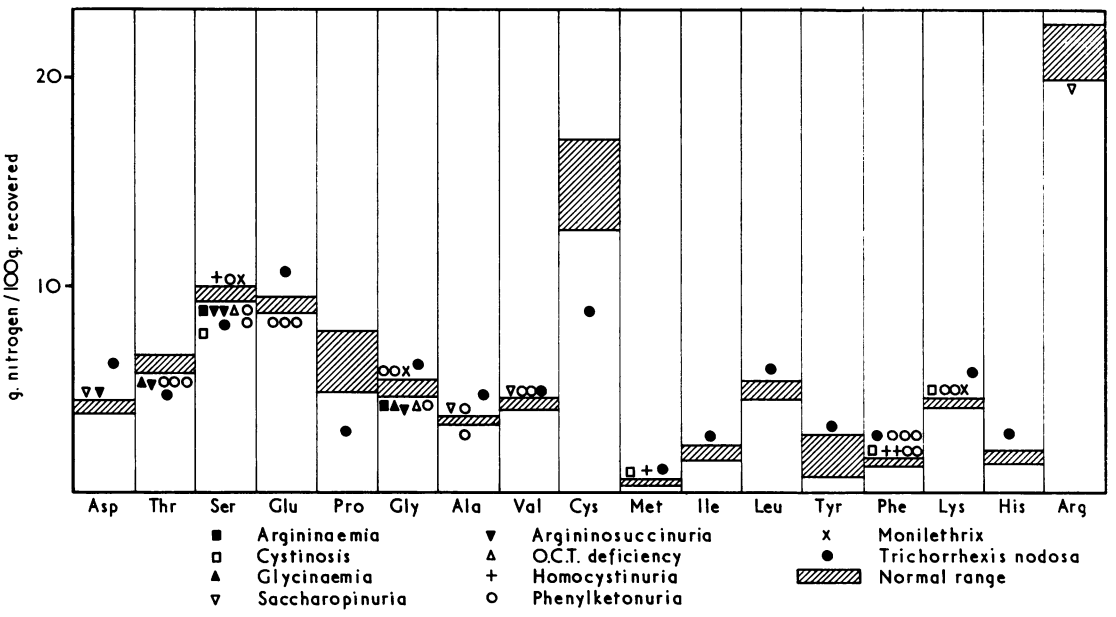

FIG.—*Hair amino acids in metabolic errors. Only the values outside the normal range (mean ± 2 SD) are indicated.*

composition were detected. In the sister (Case 1), who showed more pronounced biochemical abnormalities in the physiological fluids (Terheggen *et al.*, 1969), hair serine and glycine levels were decreased. The hair arginine levels of the two patients were not altered.

In a case of cystinosis with rapid deterioration and fatal outcome, the hair cystine level was normal, though there were pronounced deposits of cystine crystals in the cornea and the bone-marrow.

In a case of non-ketotic hyperglycinaemia, the most striking finding was the lowered glycine level.

In saccharopinuria (Carson *et al.*, 1968), aspartic acid, alanine, and valine were increased, while arginine was decreased.

In the two cases of argininosuccinuria, the only common anomaly was a decreased serine level. No argininosuccinic acid could be detected.

In a case of partial deficiency of ornithine carbamyl transferase activity and consequently ammonia intoxication (Corbeel *et al.*, 1969), threonine, serine, and glycine in the hair specimen were decreased. The biochemical abnormalities of the amino acids in the physiological fluids showed that the arginine level was lowered, and this was accompanied by high glutamic acid levels.

TABLE II

*Hair Amino Acids of 10 Control Cases**

Amno Acid	Mean	SD	SE of Mean	Range Mean ± 2 SD
Asp	4·18	0·10	0·13	3·98–4·38
Thr	6·10	0·18	0·06	5·74–6·46
Ser	9·49	0·18	0·06	9·13–9·85
Glu	9·06	0·23	0·07	8·60–9·52
Pro	6·09	0·65	0·24	4·79–7·39
Gly	5·04	0·16	0·05	4·72–5·36
Ala	3·35	0·08	0·03	3·19–3·51
Val	4·11	0·13	0·04	3·85–4·37
1/2 Cys	14·80	1·08	0·34	12·64–16·96
Met	0·27	0·05	0·02	0·17–0·37
Ile	1·89	0·09	0·03	1·71–2·07
Leu	4·88	0·18	0·05	4·52–5·24
Tyr	1·35	0·35	0·11	0·63–2·07
Phe	1·25	0·03	0·01	1·19–1·31
Lys	4·29	0·14	0·04	4·01–4·57
His	2·59	0·18	0·06	2·23–2·95
Arg	21·36	0·70	0·22	19·96–22·76

*Results expressed as g. nitrogen per 100 g. nitrogen recovered from column (ammonia excluded).

Only minor abnormalities were found in 5 hair specimens of homocystinuric patients. However, comparison of the mean values obtained for the control group with those of the 5 patients with homocystinuria shows an increase of the mean phenylalanine level in the hair of the patients ($0{\cdot}01 < p < 0{\cdot}02$). The cystine levels were found to be normal.

In phenylketonuria, the most striking finding was the increased phenylalanine level in the hair amino acids in 5 cases out of 7. In the 2 non-treated cases, this abnormality was accompanied by an increase of the lysine and glycine levels; threonine and glutamic acid were below the 95% confidence limit about the mean value of the control specimens. Statistical evaluation of the results on the 7 patients showed a decrease in the threonine level ($0{\cdot}025 < p < 0{\cdot}05$) and in the glutamic acid concentration ($0{\cdot}001 < p < 0{\cdot}005$). Phenylalanine ($0{\cdot}005 < p < 0{\cdot}01$) and lysine ($0{\cdot}02 < p < 0{\cdot}025$) were increased. On the patients on dietary therapy, a similar statistical study showed only a decreased glutamic acid level ($0{\cdot}01 < p < 0{\cdot}02$), and an increased phenylalanine level ($0{\cdot}025 < p < 0{\cdot}05$). After treatment, the threonine levels become normal. No significant differences were found for the other amino acids.

The hair amino acid patterns of a patient with late infantile amaurotic idiocy and of two patients with histidinaemia were completely normal.

In a patient with monilethrix, associated with mental retardation (Sfaello and Hariga, 1967), serine, glycine, and lysine were increased.

The most abnormal pattern was obtained on a hair specimen, kindly sent us by Dr. Pollitt, of a patient with mental and physical retardation and trichorrhexis nodosa—the male sib of the family described by Pollitt, Jenner, and Davies (1968). Our results agreed well with those published by them.

Discussion

It could be suggested that excess of an amino acid in the biological fluids might result in the production of abnormal proteins. However, it has been shown that in the proteins of various phenylketonuric organs, no abnormalities in the pattern of amino acids could be detected (Block *et al.*, 1940). Further evidence was given for haemoglobin by Allen and Schroeder (1957). These authors showed that the incorporation of phenylalanine into haemoglobin by a phenylketonuric subject was normal. On the other hand, it should be remembered that keratin is an unusual protein, the composition of which can be changed by dietary conditions, deficiency of trace metals, or a fault in lipid metabolism (Pollitt *et al.*, 1968).

The results we obtained on the hair amino acid composition in metabolic errors are difficult to compare with the findings of other authors, as their

number of control samples is too small to permit a statistical evaluation.

Vellan, Gjessing, and Seip (1969) described a normal pattern in two cases of cystinosis. The same authors did not mention any abnormality in a case of homocystinuria, except a relatively low cystine content. However, the value they mention for phenylalanine in this case is high, compared with the values of their two control cases.

The cystine levels of our 5 patients with homocystinuria were normal. As all these patients were treated with pyridoxine, the concentration of cystine may have been influenced by the therapy. Barber and Spaeth (1969) observed a low value for hair cystine in a patient before pyridoxine treatment which was drastically influenced by the treatment. Furthermore, hair pigmentation changed. However, keratinization may have been abnormal in other ways, in this patient, for the arginine values given by these authors are strikingly high before starting therapy.

Concerning phenylketonuria, our results are in opposition to those of Vellan *et al.* (1969), who reported no abnormality in the hair of an adult patient with this condition.

Though more patients need to be studied, we believe that our results indicate a biochemical relation between inborn errors and the frequently associated morphological hair abnormalities.

It is difficult to understand why in some of these diseases the very high concentration of one amino acid in the biological fluids is not mirrored in the hair keratin (e.g. argininaemia, histidinaemia), while in other diseases (e.g. phenylketonuria, glycinaemia) such a relation seems to exist.

Further evidence could be obtained by study of the hair proteins in these patients. The fractionation of hair keratin in high-sulphur, low-sulphur, and high-glycine proteins, further studied by the modern techniques of protein separation, could probably open a new field of investigation in the inborn errors.

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