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CYSTINURIA IN THE SOUTH OF BRAZIL

Roberto Giugliani[1], Maria S.R.B. Figueiredo[1], Adriana Folberg[1], Maria Cristina K. Giugliani[1], Gilberto Padovan[2] and Lewis Joel Greene[2]

ABSTRACT

To determine the prevalence of cystinuria in an area located in southern Brazil, we collected random urine samples from 827 stone-forming patients from the Porto Alegre region, state of Rio Grande do Sul. The urine samples were submitted to the cyanide-nitroprusside test, amino acid thin-layer chromatography and amino acid quantification by ion-exchange chromatography. We detected 7 patients with cystinuria in the sample studied (0.84%). Although this frequency is not higher than that observed in many other studies, the identification of several cases of cystinuria shows that this disease does occur in the south of Brazil and it is not very rare among stone-forming patients. Since it is a treatable disorder and easily detectable by a simple and inexpensive method, we suggest that cystinuria screening should be performed in every stone-forming patient in this region. We think that the protocol carried out in the present study offers a rational approach for cystinuria screening in a country like Brazil.

INTRODUCTION

Cystinuria is a disease that affects the transepithelial transport of cystine and dibasic amino acids (lysine, arginine and ornithine) (Segal and Thier, 1983). It is an autosomal recessive genetic defect for which 3 mutant alleles (I, II and III) have been identified (Rosenberg *et al.*, 1966), and there is some evidence that more genetic heterogeneity exists (Bovee and Segal, 1984). On the basis of the study of urinary

[1] Unidade de Genética Médica, Hospital de Clínicas de Porto Alegre, Rua Ramiro Barcelos 2350, 90210 Porto Alegre, RS, Brasil. Send correspondence to R.G.

[2] Centro Interdepartamental de Química de Proteínas e Departamento de Farmacologia da Faculdade de Medicina de Ribeirão Preto, 14049 Ribeirão Preto, SP, Brasil.

amino acid excretion, homozygotes (I/I, II/II and III/III genotypes) and compound heterozygotes (I/II, I/III and II/III genotypes), which are indistinguishable from one another and are denoted as cystinurics, exhibit a marked increase in urine excretion of all 4 aminoacids (Kelly, 1978). Heterozygotes +/II and +/III excrete slightly elevated ammounts of cystine and lysine in urine, whereas +/I heterozygotes and normal homozygotes show no changes in urine amino acid excretion (Kelly, 1978; Giugliani *et al.*, 1985).

Cystinurics are predisposed to the formation of urinary stones because of the low solubility of cystine at the relatively acid pH of the urine (Dent and Senior, 1955). Diagnosis permits the application of specific therapeutic measures which are generally effective in preventing the recurrence of urolithiasis (Dahlberg *et al.*, 1977).

Epidemiologic data about kidney stone disease in Latin America are scarce (Scott, 1985), and in Brazil we are only aware of our previous study in the Southeastern part of the country (Giugliani *et al.*, 1986). No data about the frequency of cystinuria is available for southern Brazil, a region not only geographically but also ethnically distinct from the southeast and the rest of the country. The objective of this study was to determine the prevalence of cystinuria in stone-forming subjects in southern Brazil, and to compare it with data on cystinuria available from southeastern Brazil and from other countries.

MATERIAL AND METHODS

A total of 827 subjects who had experienced at least one episode of urinary lithiasis (according to the inclusion critaeria described by Giugliani and Ferrari, 1980) were tested. These subjects were from the Porto Alegre region, in the southern Brazil.

An occasional urine sample was collected from each subject and submitted to the cyanide-nitroprusside test (Brand *et al.*, 1930), generally on the day of collection. When this was not possible, the samples were stored at +4°C for a maximum of 5 days. The samples that gave negative results were discarded, while whose that gave positive or doubtful results were divided into 3 aliquots, which were stored at -20°C. The first aliquot was submitted to the colorimetric determination of creatinine (Levinson and MacFate, 1956) for standardization of the urine dilution and to normalize quantitative results of amino acid analysis; the second aliquot was submitted to thin-layer amino acid chromatography (Mehta and Saini, 1974). When the pattern of amino acid excretion determined by thin-layer chromatography was compatible with cystinuria, the third aliquot was submitted to quantitative amino acid analysis by ion-exchange chromatography (Spackman *et al.*, 1958; Alonzo and Hirs, 1968). The criteria for diagnosing homozygous cystinuria was described in a previous paper (Giugliani *et al.*, 1985).

The frequency of cystinuria observed in our study was compared with data

reported elsewhere by using the chi-square test with the Yates correction (Snedecor and Cochran, 1967). The accepted level of significance was 5%.

RESULTS

The cyanide-nitroprusside test applied to 827 urine samples from stone-forming subjects gave negative results in 772, doubtful results in 45 and positive results in 10 samples. When the 55 abnormal samples (doubtful plus positive tests) were evaluated by thin-layer chromatography, 14 of them showed an abnormal pattern of amino acid excretion. When the amino acid pattern of these samples was determined by quantitative ion-exchange chromatography, only 7 were found to be compatible with homozygous cystinuria (Table I).

Table I - Urinary concentrations of lysine, arginine and cystine in the urine of cystinuric patients*.

Patient	Lysine**	Arginine	Cystine
B.L.	624	216	166
M.G.	984	616	469
F.A.S.	157	14	66
F.B.S.	1077	246	191
R.J.	610	242	114
A.M.N.	1529	31	71
E.A.F.	711	437	222
Upper normal limit***	19	2	11

*Amino acid quantification performed by ion-exchange chromatography and reported as mg/g creatine.

**Lysine values also include ornithine.

***Obtained with 20 healthy adults studied simultaneously.

DISCUSSION

The frequency of cystinuria frequency (1:118) detected among sotne-forming subjects in the region of Porto Alegre, south Brazil, was lower than that reported in three studies carried out in the northern hemisphere (Melick and Henneman, 1958; Malek and Kelalis, 1975; Rose, 1977), but similar to two other studies performed in Europe (Ghazali *et al.*, 1973; Marquardt and Nagel, 1977) and to the only study available for southeast Brazil (Giugliani *et al.*, 1986) (Table II). As these studies were

Table II - Frequency of cystinurics among stone-forming subjects in different studies.

Study and Country	Sample studied	Number detected	Comparison with the present study*	
			Chi-square	P
Melick and Henneman, 1958 (USA)	207	6	5.41	<0.05
Ghazali *et al.*, 1973 (UK)	110	2	0.20	NS
Malek and Kelalis, 1975 (USA)	78	5	15.71	<0.001
William and Chisholm, 1976 (UK)	120	3	2.65	NS
Marquardt and Naguel, 1977 (FRG)	131	1	0.17	NS
Rose, 1977 (UK)	108	6	14.48	<0.001
Giugliani *et al.*, 1986 (South East Brazil)	200	1	0.02	NS
Present study (South Brazil)	827	7	–	–

* Probability test by the chi-square test, with Yates correction; NS, non-significant.

made with selected samples, the discrepance in frequencies probably reflects the selection criteria rather than the actual frequency of the disease.

The present results demonstrate that cystinuria does occur in Brazil and it is in fact not very rare among stone-forming patients. Since it is detectable by a simple and inexpensive screening method and as it is a treatable disorder, all patients with urinary stone disease should, in our opinion, be screened for cystinuria.

We also think that the protocol carried out in the present study offers a rational approach for cystinuria screening in a country like Brazil. Every clinical chemistry laboratory is able to perform the cystinuria screening test and send the samples with positive or doubtful results to a regional reference center in order to

perform amino acid thin-layer chromatography. The few samples that will show an amino acid pattern compatible with cystinuria should then be sent to a national or supraregional reference center for diagnostic confirmation by amino acid quantification.

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RESUMO

Com o objetivo de estimar a prevalência de cistinuria numa área localizada no sul do Brasil, foram coletadas amostras ocasionais de urina de 827 pacientes com litíase urinária da região de Porto Alegre, Rio Grande do Sul. As amostras de urina foram submetidas ao teste do cianeto-nitroprussiato, à cromatografia em camada delgada de aminoácidos e à determinação quantitativa dos aminoácidos por cromatografia de troca iônica. Foram detectados 7 pacientes com cistinúria na amostra estudada (0,84%). Ainda que essa freqüência não seja superior a observada em diversos outros estudos, a identificação de vários casos de cistinúria mostra que a doença existe no sul do Brasil, não sendo muito rara entre pacientes com litíase urinária. Como é uma doença tratável e facilmente detectável por um método de triagem simples e econômico, os autores sugerem que o teste de triagem para cistinúria seja realizado, também na nossa região, em todos os pacientes com litíase urinária. Os autores são de opinião que o protocolo empregado no presente trabalho oferece uma abordagem racional para a triagem de cistinúria num país com as características do Brasil.

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