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# DETECTION OF METABOLIC DISORDERS IN HIGH-RISK PATIENTS

## A PILOT STUDY IN SALVADOR, BAHIA

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ABSTRACT - The purpose of this pilot-study was to evaluate the applicability of a screening protocol for the detection of inborn errors of metabolism (IEM) in high-risk patients. The protocol was applied in 65 patients referred to the Medical Genetics Laboratory of the University Hospital Professor Edgard Santos due to the suspicion of an IEM. Eight of these patients (12.3%) displayed an abnormal result in the screening protocol. These patients, along with 22 who displayed normal results in the screening protocol but who presented clinical symptoms or signs suggestive of an IEM not detectable by the tests applied, were selected for a further diagnostic investigation. In 5 of these 30 patients (7.7% of the total sample) it was possible to establish the diagnosis of an specific IEM. The results indicate that the designed screening protocol was sucessfully applied, allowing the detection of affected patients in a frequency comparable to that observed in larger studies performed elsewhere. The continuation of this study and the enlargement of the sample will help to delineate the profile of IEM in northeast of Brazil and will allow the identification of a significative number of patients and families, who could benefit from the therapeutic and preventive measures available for these diseases.

KEY WORDS: Mental deficiency, inborn errors of metabolism, metabolic disorders, amino acid disorders, lysosomal disorders.

**Detecção de doenças metabólicas em pacientes de alto-risco: estudo piloto em Salvador, Bahia**

RESUMO - O objetivo deste estudo-piloto foi avaliar a aplicabilidade de protocolo de triagem para a detecção de erros inatos do metabolismo (EIM) em pacientes de alto risco. O protocolo foi utilizado em 65 pacientes encaminhados ao Laboratório de Genética Médica do Hospital Universitário Prof. Edgard Santos por suspeita de apresentar um EIM. Oito desses pacientes (12,3%) apresentaram resultado anormal no protocolo de triagem. Estes, junto com outros 22, cujo protocolo de triagem teve resultados normais mas que apresentavam sinais ou sintomas clínicos sugestivos de um EIM que poderia passar desapercebido pelos testes realizados, foram selecionados para investigação diagnóstica adicional. Em 5 desses 30 pacientes (7,7% da amostra total) foi detectado um EIM. Estes resultados indicam que o protocolo de triagem planejado foi aplicado com sucesso, permitindo a detecção de pacientes afetados em uma frequência comparável à observada em estudos mais amplos realizados em outros locais. A continuação do estudo e a ampliação da amostra vão ajudar a delinear o perfil dos EIM no nordeste brasileiro, bem como a identificação de número significativo de pacientes e de famílias com EIM, que podem se beneficiar das medidas preventivas e terapêuticas disponíveis para essas doenças.

PALAVRAS-CHAVE: retardo mental, erros inatos do metabolismo, doenças metabólicas, aminoacidopatias, doenças lisossômicas.

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Inborn errors of metabolism (IEM) may be defined as a large and heteregenous group of genetic disorders which affect synthesis, degradation, transport and storage of molecules[9]. More than 400 genetic traits may be designated as IEM, most of recessive inheritance[8]. The overall frequency is estimated in one case in every one thousand newborns[7]. Most of these disorders have severe implications for affected individuals, causing mental deficiency and/or other neurological symptoms[10]. However, the clinical manifestations of IEM are extremely diverse, and usually result from the lack of essential products and/or from the accumulation of toxic substances.

The diversity of metabolic effects has imposed the need to systematize their diagnostic evaluation. The investigation of IEM usually starts with the use of simple and qualitative screening tests. When these tests are positive, a more detailed investigation is indicated, which lead to diagnostic confirmation by special methods. In Brazil, Giugliani et al. (1985)[2] and Giugliani (1988)[1] have proposed special routines for the investigation of patients with suspected IEM. In developing countries, the detection of IEM in high-risk groups has been the strategy more frequently used for the diagnosis of these disorders, as studies in these groups have shown a frequency of IEM among high-risk patients about 200 times higher than the detected in unselected samples[12].

Diagnosis of IEM allows the introduction of a specific therapy for many patients. Due to the genetic nature of these diseases, their families could benefit from measures such genetic counselling, carrier detection, prenatal diagnosis and early identification of new cases. In this report we present the results of a pilot study performed in Salvador, Bahia for the detection of IEM in high-risk patients, the first publication about detecction of IEM in Northeast Brazil.

## MATERIAL AND METHODS

We investigated 65 patients referred to the Medical Genetics Laboratory of the Hospital Univeristario Prof. Edgard Santos (Salvador, Bahia). These patients were considered as having high-risk for an IEM, 58 of them due to the occurrence of mental retardation, neuropsychomotor delay or other neurologic symptom. Other signs as visceromegaly, bone dysplasia and lens dislocation were the main reason for referral in the remaning 7 cases.

An occasional urine sample and a blood sample were obtained from each subject for the screening protocol (set of screening tests in urine and paper chromatography of amino acids in blood and urine), as described by Wannmacher et al.[12]. Positive and doubtful results, and patients with suspicion of a metabolic disorder not detected by the screening protocol, were submitted to selected diagnostic tests, which included: a) thin-layer chromatography of urinary oligosaccharides, syalyloligosaccharides, sugars and glycosaminoglycans; b) quantitative assay of blood lactate, urinary glycosaminoglycans and urinary sialic acid; c) assay of the enzyme activity of plasma hexosaminidases, beta-glucuronidase and alfa-iduronidase, of white blood cell beta-galactosidase and arylsulphatase A and of liver glucose-6-phosphatase[6].

## RESULTS

Blood and urine samples were collected from all 65 patients. However, urine loss occurred in one case, thus permitting only blood amino acid chromatography, which proved to be normal. At least one altered screening test was observed in 8 patients. Of the 56 patients with normal screening tests, 22 continued the investigation for IEM due to a picture suggestive of an IEM not detectable by the screening protocol. In summary, 30 patients (8 with altered and 22 with normal screening tests) were submitted to selected diagnostic tests. The results allowed the detection of 5 cases of IEM among the 65 patients investigated (7,7%) (Table).

## DISCUSSION

The frequency of IEM in high-risk patients in Salvador (7.7 %) seems to be similar to that found in similar studies performed in Brazil[12] and abroad[5]. Mental retardation and/or other

*Table 1. Inborn errors of metabolism diagnosed in 65 high-risk patients in Salvador, Bahia, Brazil.*

| Metabolic disorder | Number of cases | Confirmatory results |
|---|---|---|
| Metachromatic leukodystrophy* | 1 | Deficient activity of leukocyte arylsulphatase A |
| Glycogen storage disease, type I* | 1 | Deficient activity of liver glucose-6-phosphatase |
| Mucopolysaccharidosis, type I | 1 | Deficient activity of plasma alpha-iduronidase |
| Mucopolysaccharidosis, type IV B | 1 | Deficient activity of leukocyte beta-galactosidase |
| Mucopolysaccharidosis, type not identified | 1 | Abnormal pattern of urinary glycosaminoglycans** |

* diseases not identified by the initial screening tests employed in the protocol;
** collection of a blood sample for the measurement of enzyme activities for identification of the type of mucopolysaccharidosis was not possible.

neurological signs were the main reason for investigation, in agreement to the observations of Villarcal et al.[11] in Mexico and Giugliani et al.[2] in Brazil. The fact that all 5 patients with IEM had storage diseases may reflect a referral bias, as these cases may be more frequently identified by pediatricians as potential metabolic diseases more often referred to specialized diagnostic services. Lysosomal storage disorders were also the largest group of IEM diagnosed by Giugliani et al.[3] in high-risk patients in South Brazil.

The frequency of IEM diagnosed in high-risk patients in this study, when compared to the frequency of metabolic disorders identified by newborn screening programs in unselected samples demonstrates that previous selection of patients for investigation increases more than 200-fold the chance of detecting a metabolic disorder[12]. In addition, the application of diagnostic tests in patients with normal screening tests but with a clinical picture suggestive of an IEM not detectable by the screening protocol allowed the diagnosis of 2 out of 5 cases diagnosed. This finding, as already pointed out by Giugliani et al.[4], stress the need of a careful clinical selection also after the results of screening protocol in order to plan and individually designed diagnostic investigation.

The results presented in this pilot study, the first insight on IEM in Northeast Brazil, indicate that the designed screening protocol has been successful, allowing the detection of affected patients in a frequency comparable to that observed in larger studies performed elsewhere. The continuation of the study and the enlargement of the sample will allow the identification of a significative number of patients and families with IEM, which could benefit from the available therapeutic and preventive measures.

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## REFERENCES

1. Giugliani R. Erros inatos do metabolismo: uma visão panorâmica. Pediatria Moderna 1988;23:29-40.
2. Giugliani R, Dutra JC, Pereira MLS. A flowsheet for the diagnosis of storage diseases. Rev Bras Genet 1985;8:771-774.
3. Giugliani R, Dutra JC, Barth ML, Dutra-Filho CS, Goldenfum SL, Wajner M. Seven-year experience of a reference laboratory for detection of inborn errors in Brazil. J Inher Metab Dis 1991;14:400-402.

4. Giugliani R, Dutra-Filho CS, Barth ML, Dutra JC, Wajner M, Wannmacher CMD, Montagner L. Inborn errors of metabolism: sensitivity of screening tests in high-risk patients. Clin Pediat 1989;28:494-499.
5. Hoffmann GF. Selective screening for inborn errors of metabolism - past, present and future. Eur J Pediatr 1994;153:S2-S8.
6. Hommes FA. Techniques in diagnostic human biochemical genetics: a laboratory manual. New York: Wiley-Liss, 1991.
7. Lubs H A. Frequency of genetic disease. In: Lubs HA, de La Cruz F (eds). Genetic counseling. New York: Raven Press, 1977:15-22.
8. McKusick V A. Heritable disorders of connective Tissue, Ed 5. St. Louis: C.V. Mosby, 1992.
9. Sinclair L. A new look of the inborn errors of metabolism. Ann Clin Biochem 1982;19:314-321.
10. Stanbury JB, Wyngaarden JB, Fredrickson DS, Goldstein JL, Brown MS. Inborn errors of metabolism in the 1980s. In: Stanbury JB, Wingaarden JB, Fredrickson DS, Goldestein JL, Brown MS (eds). The Metabolic Basis of Inherited Disease. Ed 5. New York: McGraw Hill, 1983:3-60.
11. Villareal ML, Velazquez A, Carnevale A, Castillo V, Solis EM. Descubrimiento de errores innatos del metabolismo de aminoacidos, acidos organicos e hidratos de carbono em un hospital infantil de La Ciudad de Mexico. Bol Med Hosp Infantil (Mexico) 1978;35:205-215.
12. Wannmacher CMD, Wajner M, Giugliani R, Giugliani ERJ, Costa MG, Giugliani MCK. Detection of metabolic disorders among high risk patients. Rev Bras Genet 1982;1:187-194.