

Screening of Previously Undiagnosed Paediatric Cases of Mental Retardation and Autism for Specific Metabolic Disorders

CA Datar*, BN Apte**

Abstract

The results of a pilot study organized to examine and investigate paediatric cases of mental retardation and autism are presented. There are a large number of paediatric cases of mental retardation and autism where the exact cause cannot be ascertained after routine cytogenetic and molecular genetic investigations. We investigated such undiagnosed cases.

Cases : A total of 37 cases (0-12 yrs) from special schools were examined. Cases were diagnosed as having idiopathic mental retardation, autism or showed delay/regression of developmental milestones.

Methods : Blood and urine samples were obtained from 30 patients. Biochemical (metabolic) investigations including some enzyme estimations were done.

Results : We reached a conclusive diagnosis in 16 out of the 30 patients (54%). Of these 37% were diagnosed as having one of the mucopolysaccharoidosis, 25% had homocystinuria, another 25% had biotinidase deficiency. 12.5% cases had both homocystinuria and biotinidase deficiency.

Conclusion : From the above data it can be appreciated that, in 33% cases (cases of biotinidase deficiency and homocystinuria), the disorder can be managed by metabolic intervention so as to bring about some improvement in the severity of the symptoms. In all the above diagnosed cases, prenatal diagnosis can be offered for the next issue of the couple. These results point to an important finding that metabolic derangements could be responsible for some, if not all symptoms in mentally retarded and autistic children. It would indeed be helpful to perform biochemical investigations in all cases of mental retardation and autism, to determine the manageable element of these disorders and thus improve the quality of life of these patients. The results of this pilot study are encouraging and demand further study.

Introduction

A camp was conducted at Spandan Holistic Institute, Mumbai, in 2007. Children from special schools for autism and mental retardation participated in the camp along with their parents. These children were being trained in these special schools. Their training included occupational therapy, physiotherapy, speech therapy, social development etc. The

aim of conducting the camp was to examine and investigate undiagnosed paediatric cases of mental retardation and autism, for various inborn errors of metabolism (IEM).

In India, we do not have proper data on the prevalence of IEM in the society. The perception that a particular symptom/sign is due to a metabolic cause is often underestimated or overlooked. This commonly occurs in cases of mental retardation.

Mental retardation is defined as a

*Vidyasagar Institute of Genetic Studies and Research, Dadar, Mumbai. **Head, Department of Human Genetics, Bombay Hospital, Mumbai.

significantly sub average intellectual functioning (IQ < 70) with deficits or impairment in the adaptive functioning and an onset before 18 years of age. Mental retardation is considered to be mild if the measured IQ levels lie between 50 and 70, moderate if they are between 25 and 50 and severe if IQ is less than 25. The incidence is estimated to be 1.5 - 3% of the population (USA). It has been postulated that a specific cause is more likely to be found in cases of severe mental retardation. On the other hand, in most cases of mild-moderate mental retardation, a specific cause cannot be ascertained. 40-50% cases of mental retardation cannot be attributed to a specific cause, and are labelled as idiopathic. In roughly 5-10% cases of mental retardation known genetic causes of hereditary factors play a role. These include structural chromosomal abnormalities like deletions and translocations, chromosomal disorders like Down Syndrome, Edward Syndrome; single gene disorders like Fragile X Syndrome, Cri-du chat syndrome, Prader Willi syndrome, Angelman syndrome, and inborn errors of metabolism like phenylketonuria, mucopolysaccharoidosis, homocystinuria etc.¹

In our screening, we had excluded patients who were diagnosed as having a known genetic cause for mental retardation especially Down Syndrome, which is the commonest genetic cause of mental retardation. This left us with a majority of children in whom the symptoms were due to an idiopathic cause.

Autism, despite having its definitive diagnostic criteria, has become a loosely used term for any psycho-behavioural disorder whose cause cannot be ascertained after routine investigations. Once labelled as autistic, the children are deprived of a specific diagnosis throughout their lives and are often given only supportive and symptomatic

treatment. Therefore, in our study we also included autistic children.²

Cases

We examined and investigated 37 cases. All children were in the age group 0-12 yrs, and of Asian ethnicity. The children were randomly selected. As mentioned earlier, children with diagnosed causes of MR especially Down syndrome, Fragile X syndrome etc. were not considered for screening. Out of 37 children, 29 were male and 7 were females.

Clinical Features

Majority of the children who participated in the camp were previously diagnosed as being mentally retarded (n=35). According to the DSM IV criteria for mental retardation, most of them had mild to moderate mental retardation with IQ in the range of 50-70.

Some other children (n=29) showed autistic features like non-social behaviour, no eye-to-eye contact, signs of self mutilation etc. The other important features seen were hyperactivity, non-development of speech, gross developmental delay in all domains, and/or regression of milestones.

Almost all children had some or the other facial and/or limb dysmorphic features.

Six children had a history of convulsions.

Other clinical features seen were- skeletal abnormalities (spine), hypotonia, distended abdomen (with palpable liver in a few cases), umbilical hernia, undescended testes, dermatitis and hyper/hypo pigmentation, skin rash, alopecia, ophthalmic abnormalities, etc.

Methods

The children were accompanied by their parents, who provided the history. The genetic history was taken down in a prescribed format. Chief complaints, birth, developmental, family and dietary history

were noted. Anthropometric measurements were done for all patients. Physical examination to detect phenotypic abnormalities was done meticulously and then systemic examination was carried out. Since the children were mentally retarded and autistic, their behavioural patterns were noted in detail. The investigations done in the past were also recorded.

We received samples from 30 out of the 37 patients. 10 ml of heparinized blood was collected in a sterilized vacutainer tube and 50 ml of urine sample (kept as cold as possible) was collected.

Urine colour, odour and pH were recorded. Microscopy was performed to detect urinary crystals, if any. Spot tests were carried out on the urine sample to detect urine sugars and any specific amino acids, if excreted.

Few drops of the urine sample were spotted onto a TLC plate, and then run in different solvent systems to detect specific sugars, amino acids and the presence of organic acids.

A simple slide gel electrophoresis was performed to detect the presence of mucopolysaccharides in the urine sample.³ If found positive, a confirmatory test was done on the blood leucocytes to detect the deficiency of the specific enzyme leading to MPS. Blood ammonia, pyruvate and lactate levels were determined to detect other derangements in the metabolism.

The quantitative estimation of the biotinidase enzyme was performed on the leucocytes using a chromogenic substrate.

After the results were obtained the children along with their parents were called for a counselling session.

Results

After the tests were done, the data was compiled. We could reach to a conclusive

diagnosis in 16 out of the 30 patients (54%). 37% of these (6 out of 16) were diagnosed as having one of the mucopolysaccharoidosis. They showed a positive result on slide gel electrophoresis, when run along with a known standard solution. Most of these children (5 out of 6) showed an increased excretion of organic acids in the urine and all had a positive Benedict's test.

In 25% of the diagnosed cases (4 out of 16) biotinidase deficiency was identified. In one patient the value of biotinidase was as low as 0.5 nmol/ml of protein/min (Normal range-4.3-7.54 nmol/ml/min), in two other children it was 1.5 and 2.3 respectively.

Homocystinuria was detected on urine spot tests and TLC in 25% of the diagnosed cases (4 out of 16). Urine spot tests showed a positive sodium nitroprusside and silver nitroprusside tests and TLC showed a band in the region of homocystine.

Two children were having a double dose with biotinidase deficiency and homocystinuria being present in them. They constituted about 12.5% of the diagnosed cases. Many children (n=16) were excreting amino acids and/or organic acids in large quantities. Fructosuria was present in 7 of the 30 children.

The results were tabulated, correlated with the phenotypical findings and then conclusions were drawn.

Discussion

This pilot study, done on a sample of 37 children with features of mental retardation and autism, has revealed some interesting findings. 54% of the children in whom the mental retardation or autism was labelled as being idiopathic in nature, showed some metabolic defect. In these children no biochemical investigations were ever performed. They were being treated for their

symptoms and were being given supportive therapy like physiotherapy, occupational therapy, speech therapy, etc. But the basic defect was never investigated for, and corrected.

As discussed earlier, mental retardation can be caused by many inborn errors of metabolism which include multiple carboxylase (biotinidase) deficiency, mucopolysaccharoidosis, neurolipidosis, phenylketonuria, Wilson's disease, Menke disease, Lesch-Nyhan syndrome, etc. But patients of mental retardation are rarely investigated for these disorders.

The deficiency of the enzyme biotinidase has been cited as an important differential diagnosis in a case of mental retardation. Seemingly this enzyme deficiency is much more prevalent than it was previously thought to be. This enzyme deficiency leads to reduced or absent conversion of biocytin to biotin. Free biotin is an essential cofactor in many enzyme reactions especially those involving carboxylases. Biotinidase is ubiquitously expressed in the body and thus the effects of its deficiency are widespread. Our patients with biotinidase deficiency had severe symptoms like epilepsy (6 patients), gross developmental delay, regression of milestones, non development of speech, and mild symptoms like alopecia, hair loss, erythematous rashes, hypotonia, lethargy, etc.

All these patients were mentally retarded. These neurological symptoms and defects are probably due to the accumulation of lactic acid in the brain due to the deficiency of the enzyme pyruvate carboxylase.

Marginal biotinidase deficiency commonly occurs during pregnancy, but severe deficiency can lead to spontaneous abortions as this deficiency is embryolethal. In our case, the obstetric history of the mother of one of

the child showed that she had 4 spontaneous abortions before the birth of this child. She was probably a homozygote for the diseased gene. This deficiency can also be iatrogenic, as seen after the administration of antiepileptic medications, prolonged antibiotic use, patients on total parenteral nutrition (TPN) and in infants fed on amino acid formulae. Biotinidase deficiency if detected at an early stage, can help to prevent permanent damage to the nervous system, hearing and vision. Treatment includes the administration of biotin and a diet restricted in carbohydrate. Past studies have shown that biotinidase deficient patients who were administered biotin showed rapid improvement in seizures, muscle tone, skin rashes and alopecia and moderate improvement in the IQ and developmental milestones, however, hearing and visual loss are not reversed.⁴

Increased excretion of mucopolysaccharides was found in 6 patients. These patients presented with coarse facial features, limb dysmorphic features, mental retardation of varying degree and delayed development or non development of milestones. One infant of 9 months had a global delay of milestones as the only presenting features. Of the remaining 5 children, all had moderate mental retardation and the two of them showed autistic features. Three children had umbilical hernia, out of which 2 also had hepatosplenomegaly. It was found that patients having a lesser degree of mental retardation without any serious visceromegaly suffered from a milder variety of MPS like Morquio B or Maroteaux-Lamy syndrome, confirmed by enzyme estimations. Others probably had a severe variety of MPS like Hurlers, Hunters or Sanfillipo syndrome. The prognosis is better for the first group while it is not always good for the second group. In these cases, the exact type of MPS

can be determined by measuring the deficient enzymes.

There is no way one can prevent the progress or fully cure MPS, atleast not until the enzyme replacement therapy becomes affordable and easily available. But MPS can certainly be prevented by offering prenatal diagnosis to mothers whose earlier child/children were affected by MPS. The parents can be assessed for carrier status and the exact MPS can be identified by enzyme estimation. This allows us to accurately determine the status of affection of the child in the offspring.

Another large set of patients (25%) were found to have homocystinuria. The children presented with mental retardation, inattentiveness, hyperactivity. Two children had Marfanoid features like tall stature, arachnodactyly, knock knees, scoliosis, an altered US/LS ratio, etc. The detection of homocystinuria by spot tests and TLC is very easy. The treatment of this disorder by administration of Vitamin B6 and batatine along with dietary management is also fairly straightforward and can bring about a dramatic improvement in the condition of the patient.⁵

For the lack of an early screening procedure, the cases of homocystinuria mostly come to notice after the child is 5-6 years of age when most of the irreversible neurological and structural damage is done. This again stresses the need for an early neonatal screening programme.

One more observation worth mentioning is that in there were 3 children among those affected, who were born of a consanguineous marriage. Among these two had a double dose of biotinidase deficiency and homocystinuria. All the disorders discussed here follow an autosomal recessive inheritance; therefore they can be expected to present singularly or

along with other recessive disorders in high frequency in cases involving consanguinity.

Thus a pilot study conducted in a group of children from mentally retarded and autistic schools has revealed the presence of metabolic derangements in more than 50% of the cases examined. Despite the fact that mental retardation and autism can be caused by defective metabolic processes, investigations to detect these are rarely ordered. Early diagnosis of these disorders can lead to an effective management in some if not all cases.⁶

Seeing the burden of mental retardation and autism in the society, it will certainly be cost effective to perform metabolic screening tests in all neonates. Biochemical investigations can also be offered prenatally to mothers at high risk. Early management can be instituted in those who are found to be affected, thus reducing the severity of the disorder or preventing it completely as the case may be.

From our pilot study we conclude that a large proportion of cases with mental retardation and autism are likely to have some metabolic defect, which if detected early and corrected can improve the quality of life of these patients. A larger study needs to be conducted in this regard.

Acknowledgement

The authors acknowledge the contribution of Spandan Holistic Institute and its Director, Dr. Prafulbhai Barvalia and his team of doctors, for the arrangement and smooth functioning of the camp.

We also acknowledge the contribution of the Human Genetics Department, Bombay Hospital, Mumbai.

References

1. Apte BN, Sharma D, Dhar HL. Metabolic changes in children with behavioral disorders, abstracted in the Journal of Association of Physicians of India, 2001; 49 : 64.

2. Zaffanello M, Zamboni G, Fontana E, Zoccante L, Tato LA. Case of partial biotinidase deficiency associated with autism. *Child Neuropsychology* 2003 Sep; 9 (3) : 184-8.
3. Apte BN, Bhingarde SS. Prenatal diagnosis of Maroteaux – Lamy Syndrome, *Bombay Hospital Journal* 2001; 43 (1) : 214-6.
4. Apte BN, Tibrewala VN. Multiple Carboxylase Deficiency, *Bombay Hospital Journal* 2001 : 43(1).
5. Apte BN. 1) Homocysteinuria and 2) Biotinidase deficiency – A review of two cases, abstracted in *Bombay Hospital Journal* 1997; 39 (4) : 791 211-213.
6. Apte BN, Gogate SG. Population screening for genetic disorders- Jalna Camp Experience. *Bombay Hospital Journal* 2005; 47 (2) : 217.

EXHALED NITRIC OXIDE IN GUIDELINE-BASED ASTHMA MANAGEMENT

The result is high risk of too little or too much treatment, or both. Therefore the medical community has been hoping for a single, easy-to-measure, and reliable biomarker (such as haemoglobin A_{1c} in diabetes) that could facilitate the assessment of asthma control and help physicians to appropriately increase or decrease treatment. In this respect, exhaled nitric oxide (FE_{NO}) - an indirect marker of airway inflammation-has generated much enthusiasm FE_{NO} is easy to measure, correlates with eosinophilic airway inflammation, and is increased during periods of uncontrolled asthma and reduced during treatment.

The measurement of FE_{NO} is relatively easy and has shown promising results, but is not cheap.

The investigators conclude that adding measurements of FE_{NO} to guideline - recommended asthma management did not provide any important clinical benefit.

It seems unlikely that adding FE_{NO} measurement to guideline - recommended asthma management is going to improve asthma control or reduce under-treatment. This finding is highly relevant to daily practice.

Even if daily asthma control was not improved by the use of FE_{NO}, the measurement might still have improved individualised dosing, such that the same level of asthma control could be achieved with less treatment. By contrast, the use of FE_{NO} resulted in higher doses of inhaled corticosteroids and more frequent use of long-acting β₂ agonists. At first glance, this finding could lead to the conclusion that, in the population studied, use of FE_{NO} to help manage asthma could lead to over-treatment.

It takes more than one good study to completely rule out potential benefits of a new biomarker.

Until such data are available, a recommendation to use FE_{NO} measurements routinely in patients treated according to guidelines is *not* ready to be made yet.

Sren Pedersen, Paul M O'Byrne, The Lancet, 2008; 372 : 1015-16.