Aminoacidopathies and organic acidurias in Tunisia: a retrospective survey over 23 years

Sameh Hadj-Taieb*, Fehmi Nasrallah, Mohamed B Hammami, Monia Elasmi, Haifa Sanhaji, Feki Moncef, Naziha Kaabachi

Laboratory of biochemistry, Rabta Hospital. Tunis-Tunisia.
Tunis Medical School - Tunis El Manar University

Sa. Hadj-Taieb, F. Nasrallah, M. B Hammami, M. Elasmi, H. Sanhaji, F. Moncef, N. Kaabachi

Aminoacidopathies et aciduries organiques en Tunisie: étude rétrospective sur 23 ans

LA TUNISIE MEDICALE - 2012 ; Vol 90 (n°03) : 259 - 262

Sa. Hadj-Taieb, F. Nasrallah, M. B Hammami, M. Elasmi, H. Sanhaji, F. Moncef, N. Kaabachi

Aminoacidopathies and organic acidurias in Tunisia: a retrospective survey over 23 years

LA TUNISIE MEDICALE - 2012 ; Vol 90 (n°03) : 259 - 262

RÉSUMÉ

Prérequis : Dans les pays en développement, les erreurs innées du métabolisme ne sont pas aussi communes que les maladies infectieuses et nutritionnelles. En Tunisie, aucune information n'est disponible sur l'incidence et les caractéristiques épidémiologiques de ces maladies métaboliques héréditaires.

But: Etudier le profil des aminoacidopathies autres que la phénylcétonurie et celui des aciduries organiques et estimer leur incidence.

Méthodes: Durant la période entre 1987 à 2009, notre laboratoire a reçu 13171 demandes d'analyses pour les patients présentant des symptômes évocateurs d'erreurs innées du métabolisme. La chromatographie échangeuse d'ions des acides aminés a été effectuée sur un analyseur d'acides aminés. Les profils des acides organiques urinaires ont été déterminés par chromatographie en phase gazeuse couplée à la spectrométrie de masse.

Résultats: Nous avons trouvé 370 cas pathologiques (2,8%), répartis en 212 cas d'aminoacidopathies (57,3%) et 158 cas d'aciduries organiques (42,7%). La leucinose (32,5%), la tyrosinémie type I (28,8%) et l'hyperglycinémie sans cétose (16%) étaient les plus fréquentes parmi les aminoacidopathies. Les aciduries méthylmalonique (33,5%), propionique (18,4%) et 2-hyrdoxy glutarique (10,8%) étaient les plus fréquentes des aciduries organiques. Les incidences ont été estimées à 1/13716 pour la leucinose, 1/14804 pour la tyrosinémie de type I, 1/16144 pour l'acidurie méthylmalonique et de 1/23176 pour l'acidurie propionique.

Conclusion: les aminoacidopathies et les aciduries organiques semblent être fréquentes en Tunisie, en raison d'un taux élevé de consanguinité. Nous pensons que leur incidence est sous estimée. Afin d'améliorer leur diagnostic, il est nécessaire de disposer d'équipements lourds ce qui permettrait une prise en charge précoce des patients.

Mots-clés

Acides aminés, pathologie; aciduries, organiques; erreurs innés du métabolisme; épidémiologie/Tunisie; incidence; étude rétrospective.

SUMMARY

Background: Inborn errors of metabolism are neglected in developing countries because they are not as common as infectious and nutritional disorders. In Tunisia, no information is available on the incidence and epidemiological features of these inherited metabolic diseases.

Aims: To precise the profile of aminoacidopathies other than phenylketonuria and organic acidurias and to estimate their incidences in Tunisia.

Methods: Between 1987 and 2009, our laboratory received 13171 requests for analysis of patients with symptoms suggestive of inborn errors of metabolism. For these cases, ion exchange chromatography of free amino acids was performed on amino acids analyser. Urinary organic acids profiles were determined by gas chromatography-mass spectrometry.

Results: Abnormal cases were 370 (2.8%), divided into 212 cases of aminoacidopathies (57.3%) and 158 cases of organic acidurias (42.7%). The most frequent aminoacidopathies, were maple syrup disease (32.5%), tyrosinemia type I (28.8%) and nonketotic hyperglycinemia (16%). Methylmalonic aciduria (33.5%), propionic aciduria (18.4%) and 2-hyrdoxy glutaric aciduria (10.8%) were the most frequent organic acidurias. The incidences were calculated using the Hardy-Weinberg formula and were estimated at 1/13716 for maple syrup disease, 1/14804 for tyrosinemia type I, 1/16144 for methylmalonic aciduria and 1/23176 for propionic aciduria.

Conclusion: Aminoacidopathies and organic acidurias turned out to be highly frequent in Tunisia, mainly because of a high rate of consanguinity. We believe that they are underestimated. To improve their diagnosis, it is necessary to have available sophisticated equipment which would allow early treatment of patients.

Key-words

Amino acids, pathology; organic, acidurias; inborn errors of metabolism; incidence; epidemiology/Tunisia; retrospective study

Inborn errors of metabolism (IEM) seem to be common in the Middle East and North Africa, where high consanguinity rates still prevail due to frequents marriages between close relatives [1-3]. Diagnosis of these diseases on the basis of clinical symptoms followed by biological confirmation constitutes a reliable approach for both providing early diagnostic and therapeutic services. Among these IEM, worldwide, aminoacidopathies (AAP) and organic acidurias (OAP) have a relatively high incidence [4-6]. IEM are not given great interest in developing countries because they are not as common as infectious and nutritional disorders, and consequently are generally under-diagnosed. However, they may contribute significantly to morbidity and mortality. Hence a reliable and cost effective screening for early diagnosis is vitally important [7]. In Tunisia, no information is available on the incidence and epidemiological features of these diseases.

This retrospective study reports, the different AAP other than Phenylketonuria (PKU) and OAP, diagnosed in the metabolic division of the laboratory of biochemistry of the Rabta Hospital during a period of 23 years (1987-2009 inclusive). We estimate also the incidences of the most prevalent disorders of them in Tunisia. PKU is the most common AAP in Tunisia and has been explored in other studies [8- 9].

MATERIAL AND METHODS

Patients

In Biochemistry Laboratory of Rabta Hospital, 13171 patients with symptoms suggestive of IEM were investigated between 1987 and 2009. The cases were from different regions of Tunisia and therefore constitute a random representative sample from the entire country population. Data can thus be considered reliable estimate of the actual disease incidence. Main original hospital departments of these patients were paediatrics (75.4%), neonatology (10%) and neurology (4.6%).

Methods

Samples

The samples included random urine, fasting venous blood (collected into heparin-containing tube) and cerebrospinal fluid whenever the diagnosis of nonketotic hyperglycinemia (NKH) was suspected. These samples were complemented with information sheets where the following variables were recorded: age, gender, geographical origin, consanguinity, previous family history of IEM, the main clinical symptoms and current therapy.

Initial testing

Screening included blood and urine one-dimensional amino acid thin layer chromatography in n-butanol: acetone: acetic acid: water (35:35:10:20) [10]. Cephalic spinal fluid amino acids were also studied in patients with suspected NKH.

Specialized testing

In the case of abnormal result of the amino acid thin layer chromatography, ion exchange chromatography on amino acids analyser (Beckman 6300) was performed for the quantification of aminoacids. Urinary organic acids profiles were determined by gas chromatography-mass spectrometry (Hewlett Packard 5890/HP 5972).

Statistical analysis

Collected data on confirmed patients (AAP and OAP cases) were analyzed using Epi-Info software.

RESULTS

During 23 years of selective screening, 370 AAP and OAP cases were diagnosed corresponding to 22 IEM, giving 2.8 % of total of 13171 patients screened. These IEM were divided into respectively, 212 AAP cases (57.3 %) grouping 9 different enzyme deficiencies and 158 OAP cases (42.7 %) with 13 different types of abnormalities. The three most common disorders of amino acids metabolism were maple syrup urine disease (MSUD), tyrosinemia type I and NKH. Methylmalonic aciduria, propionic aciduria and 2-hyrdoxy glutaric aciduria were the most frequent OAP disorders (tables 1 and 2). Sex ratio was 1.36 for male/female. Consanguinity accounted for 86.7 % and the presence of previous similar disorders in the family was identified in 67.5 % of cases. When data is considered according to geographical origin, no hot spot location for these disorders could be singled out within the country, but for MSUD 97% of cases originate from west central Tunisia and for NKH 71% of patients were from central Tunisia.

Table 1 : Number and respective frequencies of different types of aminoacidopathies and age at diagnosis

Disorder type	Number [(%)]	Age at diagnosis Mean (min-max)/ months
Maple syrup urine disease	69 (32.5)	6 (1*-108)
Tyrosinemia type I	61 (28.8)	15 (1*-192)
Nonketotic hyperglycinemia	34 (16)	1 (1*-30)
Homocystinuria	21 (9.9)	104 (3-348)
Cystinuria	8 (3.8)	168 (18-360)
Tyrosinemia type 2	7 (3.3)	64 (9-180)
Hyperhomocystinemia with methyl tetrahydrofolate reductase deficiency	5 (2.4)	108 (20*-252)
Sulfite oxidase and xanthine oxidase deficiency	5 (2.4)	8 (10*-36)
Hypophosphatasia	2 (0.9)	25 (19 -31)

*Age on days; Min, minimum; Max, maximum.

Table 2 : Number and respective frequencies of different types of organic acidurias and age at diagnosis

Disorder type	Number [percent (%)]	Age at diagnosis Mean (min- max)/ months
Methylmalonic aciduria	53 (33.5)	12 (2*-144)
Propionic aciduria	29 (18.4)	7 (3*-58)
2-Hydroxyglutaric aciduria	17 (10.8)	80 (1*-456)
3-ß-Ketothiolase deficiency	13 (8.3)	18 (6-36)
Isovaleric aciduria	10 (6.3)	24 (1*-84)
Glutaric aciduria type I	10 (6.3)	22 (18*-78)
Alkaptonuria	9 (5.7)	200 (6*-588)
Pyroglutamic aciduria	5 (3.2)	20 (9*-89)
N-Acetylaspartic aciduria	5 (3.2)	41 (15-76)
Primary carnitine deficiency	3 (1.9)	20 (2-36)
3-Methylglutaconic aciduria	2 (1.2)	36 (4*-72)
3-Methyl crotonyl glycinuria	1 (0.6)	24
Mevalonic aciduria	1 (0.6)	7

Age at diagnosis varied between 1 day and 49 years with a mean age of 1 year and 9 months. NKH is the AAP whose clinical symptoms were the earliest followed by MSUD, sulfite oxidase and xanthine oxidase deficiency and tyrosinemia type I. For other AAP, onset of clinical symptoms was later in our patients. Concerning OAP, onset of clinical symptoms was very heterogeneous. Despite this variability, we remark early onset of clinical signs during propionic, methylmalonic and pyroglutamic aciduria (tables 1 and 2).

The main clinical symptoms likely related to clinical disease state in new born and infants patients were, hypotonia (73.6%), metabolic acidosis (65.2%), seizures (61%), failure to feed (59.5%), respiratory difficulties (58.4%), ketosis (57.3%), coma (54.8%), persistent vomiting (53.8%), hepatomegaly (47.5%), mental retardation (47%), delay in motor development (43%) and abnormal movements (36.5%).

Using Hardy-Weinberg formula [E= M/aN + f √M/aN; E, estimated incidence; M: pathological cases; a: period of study (23 years: 1987-2009 inclusive); N: total of births per year during the period of study; f: coefficient of consanguinity (0.0129)] [11], the diseases incidences were estimated for AAP and OAP as one per 4514 births (1/4514). Table 3 shows the calculated incidences for the most frequent AAP and OAP.

Table 3 : Calculated incidences of the frequent aminoacidopathies other than phenylketonuria and organic acidurias.

Disorder	Cases diagnosed	Incidence (number/live births)
Aminoacidopathies other than phenylketonuria	212	1/4464
Maple syrup urine disease	69	1/13716
Tyrosinemia type I	61	1/14804
Nonketotic hyperglycinemia	34	1/21088
Organic acidurias	158	1/5570
Methylmalonic aciduria	53	1/16144
Propionic aciduria	29	1/23176
2-Hydroxyglutaric aciduria	17	1/31573

DISSCUSION

A retrospective study of inherited metabolic diseases during 23 years in Tunisia showed that AAP were more frequent (1/4464) than AOP (1/5570). This study has been conducted on patients with a high rate of consanguinity (86%) and high rate of first-cousin marriages (56%). In addition, we notice a high frequency of similar disorders or unexplained deaths among the families of patients (67.5%). High consanguinity is common in the Middle East and North Africa, which increased the occurrence of IEM [7]. Indeed, a study of IEM in an Omani population of the Arabian Peninsula [1] found that the parental consanguinity was twice as frequent in the study patients as in the general population. Other studies from Riyadh, Saudi Arabia and Kuwait indicate that some diseases are confined to specific families and Tribal groups [12-16]. In contrast, in other studies in Italy, British Columbia, Brazil and West Midlands, patients with IEM had a low rate of consanguinity [4, 17-19].

In this study, we did not find geographic areas at higher risk of IEM, with the notable exception of west central Tunisia where MSUD (97% of cases) and NKH (72% of patients) showed a clear regional clustering. These results are similar to those in the Mexican study in which MSUD has been found more frequent in Hispanic population [20].

Age at diagnosis was very variable between different metabolic diseases. Acute diseases (such as NKH, MSUD, methylmalonic and propionic aciduria) were characterized by a sudden onset of deterioration of vital functions (like respiratory and neurological distress). motivating parents regardless of their level of consciousness to consult early. However, for other chronic pathologies (such as homocystinuria, cystinuria and 3-,-Ketothiolase deficiency) characterized by a gradual onset and progressive installation of clinical symptoms (such as mental retardation and delay in motor development), age at diagnosis was late because the consultation depends on the degree of awareness of parents and their socio economic conditions.

More than half of all diagnosed patients corresponded to amino acid disorders other than PKU.

It is possible that AAP were often diagnosed due to the simplicity and low-cost of the detection techniques required, such as chemical screening tests and amino acid chromatography. Following PKU, MSUD was the most frequent AAP; it is interesting that an even higher frequency has been found in Saudi Arabia [13] and in Mexico [20]. Because Spain was an Islamic country for 800 years, it is tempting to speculate whether some of the MSUD mutations in the Spanish-speaking populations are of Near Eastern origin [20].

Regarding OAP, methylmalonic aciduria was the most prevalent disorder in this study and also in other studies performed in Saudi Arabia [13], Mexico [20], Italy [17] and China [21].

Some factors need to be considered when comparing these results with those of other studies. The number of cases in Tunisia is under diagnosed because, on the one hand, the patients not identified in the acute phase die quickly and on the

other hand, our Laboratory is the only one specialised in the diagnosis of these diseases which creates difficulties for biological exploration of patients living far; in addition, the screening is selective and no systematic national screening programmes were done. The diagnosis is delayed for the majority of patients, because of poor socio economic conditions of some patients in isolated areas which prevent the parents to consult early.

CONCLUSION

In total, AAP and OAP seem to be highly frequent in Tunisia, because of a high rate of consanguinity. It is necessary to make available sophisticated equipment, to improve the diagnosis of AAP and AOP as well as other disorders; this is essential to early diagnosis and effective management and emergency treatments of patients with suspected IEM.

Aknowledgments

The authors would like to thank all clinicians of hospital departments in Tunisia for their collaboration, in particular Paediatrics, Neonatology, Neurology, Neuropaediatrics, Genetics, Dermatology and Ophthalmology. They would like to thank also technicians of the biochemistry laboratory: Dalinda Bouhadra, Salma Ben Ayed, Saloua Skhiri Mustpha Mzoughi and Aida Abidli, for their technical assistance.

References

1. Joshi SN, Hashim J, Venugopalan P. Pattern of inborn errors of metabolism in an Omani population of Arabian Peninsula. Ann Trop Paediatr 2002; 22:93-96.
2. Teebi A, El-Shanti H. Consanguinity: implications for practice, research and policy. Lancet 2006; 367:970-71.
3. Bittles AH. When cousins marry: a review of consanguinity in the Middle East. Prespect Hum Biol 1995; 1:71-83.
4. Sanderson S, Green A, Preece MA, Burton H. The incidence of inherited metabolic disorders in the West Mildlands, UK. Arch Dis Child 2006; 91:896-99.
5. Wasant P, Svasti J, Srisomsap C, Limmongkolkul S. Inherited metabolic disorders in Thailand. J Med Assoc Thai 2002; 85:S700-S9.
6. Widhalm K. 25 years Austrian screening program for inborn errors of metabolism at the Vienna University. Wien Klin Wochenschr 1992; 104:510-3.
7. Sadallah AA, Rached MS. Newborn screening: Experiences in the Middle East and North Africa. J Inherit Metab Dis 2007; 30:482-9.
8. Khemir S, Tebib N, Nasrallah F et al. Phenylketonuria in Tunisian institutions for the mentally handicapped. Arch Dis Child. 2009; 94:647-8.
9. Khemir S, Elasmi M, Sanhaji H et al. Phenylketonuria is still a major cause of mental retardation in Tunisia despite the possibility of treatment. Clin Neurol Neurosurg 2011; 113: 727-30.
10. Hans JB, Marinus D, Johannis PK et al. Disturbances of amino acid metabolism: clinical chemistry and diagnosis. USA: Urban & Schwarzenberg Baltimore-Munich; 1981:450
11. Monastiri K, Khairi H, Snoussi N, Ghannem H, Hajfredj A. Biométrie du nouveau né dans le Sahel Tunisien. Ann Pediatr 1994; 41:578-83.
12. Moammar H, Cheriyan G, Mathew R, Al-Sannaa N. Incidence and patterns of inborn errors of metabolism in the Eastern Province of Saudi Arabia, 1983-2008. Ann Saudi Med. 2010; 30:271-7.
13. Ozand PT, Devol EB, Gascon GG. Neurometabolic diseases at a national referral center: Five years experience at the King Faisal specialist hospital and research center. J Child Neurol 1992; S7:4-11.
14. Rashed M, Ozand PT, al Aqeel A, Gascon GG. Experience of King Faisal specialist hospital and research center with Saudi organic acid disorders. Brain Dev 1994; S16:1-6.
15. Ozand PT. Diagnosis of inborn errors of metabolism by tandem mass spectrometry. Ann Saudi Med 1998; 18:234-8.
16. Abdel-Hamid M, Tisocki K, Sharaf L, Ramadan D. Development, validation and application of tandem mass spectrometry for screening of inborn metabolic disorders in Kuwaiti Infants. Med Princ Pract 2007; 16:215-21.
17. Carlo Dionisi V, Risso C, Burlina Ab et al. Inborn errors of metabolism in the Italian pediatric population: a retrospective survey. J Pediatr 2002; 3:321-7.
18. Applegarth DA, Toone JR, Lowry RB. Incidence of inborn errors of metabolism in British Columbia, 1969-1996. Pediatrics 2000; 105:e10.
19. Simoni RE, Oliveira SPH, Grassiano DM, et al. Screening for inborn errors of metabolism in high-risk children from Rio de Janeiro, Brazil. Early Hum Dev 1998; 50:305-11.
20. Velázquez A, Vela-Amieva M, Cicerón-Arellano I et al. Diagnosis of inborn errors of metabolism. Arch Med Res 2000; 31:145-50.
21. Han LS, Ye J, Qiu WJ et al. Diagnosis of inborn errors of metabolism using tandem mass spectrometry and gas chromatography mass spectrometry. Zhonghua Yi Xue Za Zhi 2008; 88:2122-6.