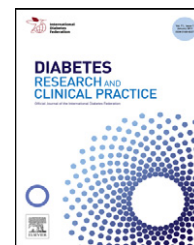


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Brief report

Clinical and molecular basis of transient neonatal diabetes mellitus in Brazilian children

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ABSTRACT

We report a series of patients with transient neonatal diabetes mellitus (TNDM). Paternal uniparental isodisomy of chromosome 6 and heterozygous *KCNJ11* and *ABCC8* mutation were the mutations found. This first reported series of Brazilian patients expands the geographical data on TNDM contributing to better understanding of its pathophysiology.

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1. Introduction

Transient neonatal diabetes mellitus (TNDM) is now referred to as a unique genetic entity classified as: TNDM1 (OMIM 601410) caused by genetic and epigenetic mutations at chromosome 6q24, TNDM2 (OMIM 610374) caused by *ABCC8* mutation(s); and TNDM3 (OMIM 610582) caused by a *KCNJ11* mutation (OMIM 600937) [1]. TNDM1 is the most frequent cause of transient neonatal diabetes mellitus, while mutations in the *ABCC8* and *KCNJ11* genes which encode the SUR1 and KIR6.2 subunits of the pancreatic ATP-sensitive potassium (K-ATP) channel are the second most common cause of the disorder [2].

Due to the rarity of this condition, the present study is aimed to describe, for the first time, a series of Brazilian

children with TNDM, with emphasis on its clinical and molecular aspects.

2. Materials and methods

The medical records of four patients with TNDM treated at a Brazilian university hospital were reviewed.

Laboratory investigation included: glycated hemoglobin (HbA1c), insulin, C peptide, anti-insulin antibody, anti-IA2 antibody, anti-islet cell antibody and anti-GAD antibodies. These tests were performed at the time of diagnosis, at the time of disease remission and at the time of the last assessment.

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Table 1 – Clinical data in Brazilian patients with transient neonatal diabetes mellitus.

Case	Gender	GA	Pregnancy	BW (centiles)	Physical exam	
					Macroglossia	Umbilical hernia
#1	M	33 weeks	Arterial hypertension	1190 g (<3th)	Absent	Absent
#2	M	33 weeks	Premature rupture of membranes	1385 g (3–10th)	Absent	Absent
#3	F	38 weeks	Urinary tract infection	3250 g (50–90th)	Absent	Absent
#4	M	38 weeks	Normal	2870 g (10–50th)	Absent	Absent

M: male; F: female; g: grams; GA: gestational age; BW: birth weight.

Table 2 – Laboratory data upon diagnosis, remission and follow-up in Brazilian patients with transient neonatal diabetes mellitus.

Exams	Patients							
	Case # 1		Case #2		Case #3		Case #4	
	Diag (1 day)	Rem (4 months)	Diag (1 day)	Rem (2 months)	Diag (15 days)	Rem (7 months)	Diag (50 days)	Rem (3 months)
Glycemia (70–99 mg/dL)	274	81	463	86	360	78	514	
Insulin (92–25 uUI/mL)	1.3	NP	<2.0	NP	0.7	NP	NP	<2.0
C Peptide (1.1–1.5 ng/mL)	0.2	NP	0.75	NP	0.1	1.1	NP	0.78
HbA1c (4.5–7.0%)	NP	5.6	NP	5.1	6.5	6.5	NP	6.0
Anti-GAD (<1.0 U/mL)	0.6	NP	0.5	NP	0.2	NP	NP	0.2
Anti-IA2 (<0.5 U/mL)	0.2	NP	0.3	NP	0.1	NP	0.2	NP
Genetic cause for the transient neonatal diabetes	UPD6		UPD6		p.R1183Q (c.3548G > A) ABCC8 mutation		p.R50Q (c.149G > A) KCNJ11 mutation	

Diag: age at diagnosis; Rem: age at remission; HbA1c: glycated hemoglobin; anti-GAD: antibody anti-decarboxylase of glutamic acid; anti-IA2: antibody anti-tyrosine phosphatase; NP: not performed; UPD6: paternal uniparental isodisomy of chromosome 6.

Studies to investigate methylation status at chromosome 6q24 were performed at Wessex Regional Genetics Laboratory, Salisbury, UK, as previously described [3]. Studies to investigate the ABCC8 and KCNJ11 mutations were performed at Peninsula Medical School, University of Exeter, UK, as previously reported [2].

This work was approved by the Ethics Research Committee of the participant institution.

3. Results

Table 1 summarizes the main clinical information of the 4 patients (3 males, 1 female). The diagnosis of diabetes mellitus was made between 12 h and 55 days of life. Hyperglycemia was detected in three patients and diabetic ketoacidosis in one patient (Case #1). All patients required transient insulin replacement.

Table 2 summarizes the main laboratory information obtained at the time of diagnosis and TNDM remission.

4. Discussion

4.1. Molecular aspects

The genetic origin for more than 90% of the cases of TNDM is established. Disruption of the imprinted TNDM locus on chromosome 6 is the most common etiology [4,5].

Three types of abnormalities involving 6q24 cause TNDM1: (1) paternal uniparental disomy of chromosome 6 (UPD6), either complete or segmental [6]; (2) duplication of the imprinted TNDM1 region on chromosome 6q24 [7]; and (3) epimutations (maternal hypomethylation of the TNDM differentially methylated region in the absence of any detectable chromosome anomaly) [4]. The common underlying effect of these abnormalities is overexpression of genes in the TNDM locus [8]. The TNDM locus is genomically imprinted, with restriction of gene expression from the maternally inherited allele. However, since the ZAC/PLAGL1 has biallelic expression in white blood cells, this makes it difficult to demonstrate the relaxation of imprinting as a mechanism that causes TNDM through analysis of the blood of patients with this phenotype, since there is scarce availability of samples of other tissues, for obvious ethical reasons [9]. In a subset of TNDM patients, imprinting is disrupted at diverse chromosome locations throughout the genome, in a widespread DNA methylation defect of genomic imprinting [3,10].

The paternal isodisomy of chromosome 6 seen in patients 1 and 2, with homologous chromosomes 6 in all the loci, is consistent with paternal chromosomal duplication after fertilisation of maternal gamete nullisomic for chromosome 6; this is a sporadic event, with low recurrence risk [5]. In the present study, 2 of the 4 patients had paternal isodisomy of chromosome 6, in accordance with the published prevalence of UPD6 [11].

Flanagan et al. [2] identified K-ATP channel genes mutations in 25/28 patients (12 KCNJ11 and 13 ABCC8) with TNDM in

whom methylation defects at chromosome 6q24 had been excluded. In their study the phenotype associated with mutations in the two genes was similar, contrasting with the group of 6q24 patients, who had a lower birth weight, earlier diagnosis and disease remission.

Besides the defects previously described, TNDM may also be caused by mutations in the *INS* gene and, more rarely, in the *HNF1B* gene [1].

4.2. Clinical aspects

The clinical manifestations of TNDM begin in the first weeks of life. Polyuria, dehydration and low weight gain are the most frequent symptoms [8]. The patients are generally born at term, being small for gestational age and looking malnourished [12].

Apparently, there is no major phenotypic difference between patients with UPD6, duplication of 6q24, methylation defect, and no identifiable anomaly of chromosome 6, to support the hypothesis that most patients share the same genetic mechanism [13]. Nevertheless, among patients with TNDM, those with 6q24 abnormalities showed more marked intrauterine growth retardation. Consistently with such data, in the present report, the two patients with UPD6 were premature, and one was small for gestational age. In this report, remission of hypoglycemia occurred earlier in the patients with isodisomy when compared to the patients with *KCNJ11* and *ABCC8* mutations. Since our patients are still young, they will need long-term follow up, until puberty, for assessment of diabetes recurrence.

In general, diagnostic investigation shows hyperglycemia, glycosuria, moderate or absent ketonuria, insulinopenia, reduction in the serum C-peptide levels, slight increase in glycated hemoglobin and negative results for anti-insulin, anti-islet and anti-GAD [13,14]. All these laboratory findings were present in our patients.

Although insulin replacement is the first choice for neonatal diabetes, the identification of an activating mutation(s) in the *KCNJ11* or *ABCC8* genes is clinically important, because of a favorable response sulfonyleureas therapy (oral hypoglycemics that inhibit the potassium ATP channel, increasing insulin release), that not only improves glucose control but also reduces the risk of hypoglycemia [15,16].

5. Conclusions

The genetic basis of TNDM in Brazilian patients, first reported in this paper, is similar to molecular defects previously reported in the literature [17,18]. This information is important to assist in genetic counseling on recurrence of the disease and is essential to understand the role of genetics and epigenetics in the pathophysiology of TNDM.

Contributors

CA participated in the conception and design of the study and the data acquisition, wrote the initial the draft and revised the final version; SF and SE tested the *ABCC8* and *KCNJ11*

mutations and contributed to the final version of the paper; DM tested the UPDS and contributed for the final version of the paper. All authors acknowledge that they have participated sufficiently in the work to take responsibility for its content approving its final version.

Conflicts of interest

There are no conflicts of interest.

REFERENCES

- [1] Mackay DJ, Temple IK. Transient neonatal diabetes mellitus type 1. *Am J Med Genet C Semin Med Genet* 2010;154C:335–42.
- [2] Flanagan SE, Patch AM, McKay DJ, Edghill EL, Gloyn AL, Robinson D, et al. Mutations in ATP-sensitive K channels genes cause transient neonatal diabetes and permanent diabetes in childhood or adulthood. *Diabetes* 2007;56:1930–7.
- [3] Mackay DJ, Hahnemann JM, Boonen SE, Poerksen S, Bunyan DJ, White HE, et al. Epimutation of the TNDM locus and the Beckwith-Wiedemann syndrome centromeric locus in individuals with transient neonatal diabetes mellitus. *Hum Genet* 2006;119:179–84.
- [4] Temple IK, Shield JP. Transient neonatal diabetes, a disorder of imprinting. *J Med Genet* 2002;39:872–5.
- [5] Temple IK, Shield JP. 6q24 transient neonatal diabetes. *Rev Endocr Metab Disord* 2010;11:199–204.
- [6] Temple IK, James RS, Crolla JA, Sitch FL, Jacobs PA, Howell WM, et al. An imprinted gene(s) for diabetes? *Nat Genet* 1995;9:110–2.
- [7] Docherty LE, Poole RL, Mattocks CJ, Lehmann A, Temple IK, Mackay DJ. Further refinement of the critical minimal genetic region for the imprinting disorder 6q24 transient neonatal diabetes. *Diabetologia* 2010;53:2347–51.
- [8] Polak M, Cavé H. Neonatal diabetes mellitus: a disease linked to multiple mechanisms. *Orphanet J Rare Dis* 2007;2:12.
- [9] Mackay DJ, Coupe AM, Shield JP, Storr JN, Temple IK, Robinson DO. Relaxation of imprinted expression of *ZAC* and *HYMAI* in a patient with transient neonatal diabetes mellitus. *Hum Genet* 2002;110:139–44.
- [10] Mackay DJ, Callaway JL, Marks SM, White HE, Acerini CL, Boonen SE, et al. Hypomethylation of multiple imprinted loci in individuals with transient neonatal diabetes is associated with mutations in *ZFP57*. *Nat Genet* 2008;40:949–51.
- [11] Suzuki S, Makita Y, Mukai T, Matsuo K, Ueda O, Fujieda K. Molecular basis of neonatal diabetes in Japanese patients. *J Clin Endocrinol Metab* 2007;92:3979–85.
- [12] Luzuriaga C, Cantero P, Llorca J, Martinez-Chamorro, Perez G. Diabetes neonatal. *An Esp Pediatr* 2001;54(Suppl. 1):1–8.
- [13] Alcaina M, Garcia-Algar O, Pérez S, Vidal R, Jurado L, Sierra A. Diabetes mellitus neonatal transitória asociada a isodisomía uniparental del cromosoma 6. *An Esp Pediatr* 2002;56:567–70.
- [14] Shield J, Temple K, Sabin M, Mackay D, Robinson O, Betts R, et al. An assessment of pancreatic endocrine function insulin sensitivity in patients with transient neonatal diabetes in remission. *Arch Dis Child Fetal Neonatal* 2004;89:F341–3.

- [15] Aguilar-Bryan L, Bryan J. Neonatal diabetes mellitus. *Endocr Rev* 2008;29:265–91.
- [16] Pearson ER, Fletchner I, Njosltad PR, Malecki MT, Flanagan SE, Larkin B, et al. Neonatal Diabetes International Collaborative Group. Switching from insulin to oral sulfonylureas in patients with diabetes due to Kir6.2 mutations. *N Engl J Med* 2006;355:467–77.
- [17] Hattersley AT, Ashcroft F. Activating mutation in Kir6.2 and neonatal diabetes. New clinical syndromes, new scientific insights, and new therapy. *Diabetes* 2005;54:2503–13.
- [18] Mackay DJ, Boonen SE, Clayton-Smith J, Goodship J, Hahnemann JM, Kant SG, et al. A maternal hypomethylation syndrome presenting as transient neonatal diabetes mellitus. *Hum Genet* 2006;120:262–9.