Mutations in the SLC29A3 Gene Are not a Common Cause of Isolated Autoantibody Negative Type 1 Diabetes

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Dear Sir:

Recessive mutations in the SLC29A3 gene have recently been shown to result in diabetes [1, 2]. In the last year eleven families with the H syndrome (OMIM#612391) and five families with pigmented hypertrichosis with insulin dependent diabetes (PHID) have been described, resulting from seven different recessive SLC29A3 mutations. The most common feature in all but two cases is pigmented hypertrichosis. Hyperglycaemia is an overlapping feature of the two syndromes although it is much rarer in the H syndrome where it is present in 1/15 subjects [1, 3] compared to 5/6 subjects with PHID [2, 4, 5]. The median age of diagnosis for diabetes was 12 years (range: 4-15 years), all patients were insulin treated with only 1/5 testing positive for GAD autoantibodies. It is not known if milder mutations in the SLC29A3 gene can cause autoantibody negative type 1 diabetes without associated syndromic features.

The SLC29A3 gene encodes ENT3, a member of the equilibrative nucleoside transporter family (SLC29), which mediates intracellular trafficking of nucleosides [6]. In man the SLC29A3 gene is most highly expressed in uterus [7]. In vivo studies of the Drosophila melanogatser ortholog of SLC29A3 (ENT1) have shown it interacts the insulin signalling pathway, although the molecular basis of the interaction has yet to be characterised [2]. In addition, it has been detected in total human pancreas but it is not known if it is expressed in the exocrine component or in the islets [7].

In order to determine whether the SLC29A3 gene is expressed in endocrine pancreas, we quantified SLC29A3 transcripts by real-time PCR in human islet, pancreas and uterine RNA, relative to that of the HNF4A gene, which has documented expression in the beta cell [8]. B2M was used as an endogenous control. Probes to SLC29A3 mRNA were targeted to the exon 5-6 junction of the SLC29A3 gene (NM_018344.4) and were validated by standard curve analysis over eight 1.10 serial dilutions (r² 0.99). The expression levels of B2M, HNF4A and SLC29A3 transcripts were calculated...
from average crossing points of triplicate samples, using the comparative Ct (ΔΔCt) method [9]. Compared to uterine SLC29A3 mRNA levels there were 17% and 2% expression in the pancreas and islets respectively (relative to B2M). Moreover SLC29A3 mRNA makes up only a small proportion of the beta cell transcriptome representing 0.4% of transcripts detected for HNF4A. Therefore SLC29A3 is detectable in both exocrine and endocrine pancreas, although the expression is lower in the latter.

Early-onset diabetes is a feature of both the H syndrome and PHID. We hypothesised that mutations in the SLC29A3 gene could cause isolated diabetes in children and screened 47 cases diagnosed at a median of 5 years (range: 1-16 years) with autoantibody negative type 1 diabetes (antibodies tested at or soon after diagnosis; glutamic acid decarboxylase and/or islet antigen 2), and without pigmented hypertrichrosis. Mutations in the HNF1A, HNF4A, KCNJ11 and INS genes were excluded by sequence analysis in all subjects.

We amplified the 6 exons of SLC29A3 (primer sequences available on request), including the exon/intron boundaries and non-coding exon 1. We did not identify any pathogenic SLC29A3 mutations, but the common non-synonymous polymorphisms rs2277257, rs780668, rs2252996, and rs2487068 were present at a minor allele frequency of 71%, 7%, 9% and 6%, respectively. We have shown that SLC29A3 is expressed in the human islet and recessive mutations are likely to result in beta cell failure, however mutations in this gene are not a common cause of isolated autoantibody negative diabetes diagnosed in children under 17 years.

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Conflict of interest None to declare

References