Defective insulin secretion is a key feature in the pathogenesis of type 2 diabetes [1–3]. Both genetic and acquired factors contribute to the loss of beta-cell functional mass [1–3]. The candidate-gene approach and the genome wide-scanning technique have led to the identification of several type 2 diabetes susceptibility genes, most of which are involved in pancreatic beta-cell function and/or turnover [4,5]. Single nucleotide polymorphisms (SNP) in SLC2A2, the gene encoding glucose transporter 2 (GLUT2), for example, have been associated with type 2 diabetes in humans and with the conversion of impaired glucose tolerance to type 2 diabetes [6,7].

For this reason, we investigated whether the presence of two common SLC2A2 SNP (rs5393 and rs5394) can directly affect the function of human pancreatic islets. Human islets were prepared by collagenase digestion and density gradient purification [8] from the pancreas of 20 non-diabetic (ND; age: 63.5 ± 15.8 years; body mass index (BMI): 25.1 ± 2.9 kg/m²) and 20 type 2 diabetic (T2D; age: 66.3 ± 8.0 years; BMI: 27.0 ± 3.5 kg/m²) organ donors, and insulin secretion from the isolated islets was measured in response to acute (45 min) exposures to 3.3 and 16.7 mmol/L glucose [7,8]. SNP were identified by the TaqMan allelic discrimination assay (Applied Biosystems, Foster City, CA) [6]. GLUT2 mRNA expression was assessed by quantitative RT–PCR [9].

Subjects carrying the rs5393 and rs5394 polymorphisms were 50% and 20%, respectively, in ND, and 40% and 45%, respectively, in T2D. Both polymorphisms were present in 5% and 30% of the ND and T2D, respectively (P = 0.05 by the Chi² test). As expected [10], glucose (16.7 mmol/L) stimulated insulin release (GSIR) that was lower in T2D than in ND islets (0.129 ± 0.113 U/islet/min vs. 0.060 ± 0.071* U/islet/min, respectively; P < 0.01), and accompanied by a lower stimulation index (ratio of insulin release at 16.7 mmol/L glucose: 3.23 ± 2.06 vs. 1.54 ± 0.62, respectively; P < 0.01). Although the presence of rs5393 was not associated with significant changes in insulin release (not shown), the presence of rs5394 was associated with reductions in both insulin secretion and the stimulation index (Table 1). Accordingly, a statistically significantly (P < 0.05) higher percentage of patients with a stimulation index below the median value (2.6) was seen in the presence of rs5394 (76.9%) than in its absence (22.7%). The presence of the rs5394 polymorphism was, in addition, associated with an approximately 30% reduction of GLUT2 mRNA expression, which was 0.050 ± 0.030 in the variant islets and 0.028 ± 0.030 in control cells.

In conclusion, the combined presence of rs5393 and rs5394 polymorphisms of GLUT2 was more frequent in T2D than in ND in the present series. Furthermore, rs5394 appeared to be associated with significantly decreased GSIR and a tendency to a reduced GLUT2 gene expression.

Acknowledgements

Supported in part by the Italian Ministry of University and Research (PRIN 2006-2008).

References


Table 1

<table>
<thead>
<tr>
<th></th>
<th>Wild-type islets</th>
<th>rs5394 islets</th>
</tr>
</thead>
<tbody>
<tr>
<td>Insulin release (µU/islet/min)</td>
<td>0.129 ± 0.113</td>
<td>0.060 ± 0.071*</td>
</tr>
<tr>
<td>Stimulation index</td>
<td>3.23 ± 2.06</td>
<td>1.52 ± 0.89**</td>
</tr>
</tbody>
</table>

*P = 0.02 and **P < 0.01 vs. wild-type by Student’s t test.

1262-3636/$ – see front matter © 2009 Published by Elsevier Masson SAS. doi:10.1016/j.diabet.2009.01.002


S. Del Guerra
R. Lupi
V. D’Aleo
F. Filipponi
U. Boggi
P. Marchetti *

Department of Endocrinology and Metabolism, Cisanello Hospital, University of Pisa, via Paradisa 2, 56124 Pisa, Italy

* Corresponding author.
E-mail address: marchant@immr.med.unipi.it (P. Marchetti).

19 January 2009
Available online 9 March 2009