SUPPLEMENTARY DATA

MATERIALS AND METHODS

OGTT Tests
For ethical reasons, the OGTT was not performed in patients presenting with FPG greater than 11.1 mmol/l. Hence, in the AER group 15 patients underwent the OGTT at baseline and 13 patients underwent the OGTT after physical training. Similarly, in the FOR group 13 patients underwent the OGTT at baseline and 15 patients underwent the OGTT after physical training.

During the entire test patients were sitting in a comfortable cardiac chair. One teflon (21 g) venous catheter was inserted into an antecubital vein for blood sampling and kept patent with heparinized normal saline solution. After a 30’ rest to establish baseline, at time = 0’ subjects ingested 75 g of glucose in 300 ml of water over 5 min. Blood samples to measure glucose, C-peptide and insulin concentrations were collected at times -10’, 0’, +15’, +30’, +45’, +60’, +90’, +120’, +150’, +180’, +210’, +240’, +270’ and +300’. Urines were collected to measure glycosuria.

All blood samples were collected in pre-chilled tubes and readily spun at 1,500 g. Plasma and serum specimens were stored at –80° C. Serum C-peptide and insulin concentrations were measured by chemiluminescence as previously described (1).

Mathematical modeling of beta-cell function
The analysis of the glucose and C-peptide curves during the OGTT follows the general strategy described in previous publications (2, 3) with some modifications (4,5) and builds upon previous works from other laboratories (6,7). The kinetics of C-peptide is described with a two-compartment model, in which the two pools (1 and 2) exchange with each other and the irreversible loss of the hormone is from pool 1, the same where C-peptide concentration is measured. C-peptide kinetic parameters are computed according to the equations by Van Cauter et al. (8).

Herein are the equations describing the model of glucose induced insulin secretion during an OGTT:

\[
dcp_1(t)/dt = ISR(t) + c_{p2} \cdot k_{12} - (k_{01} + k_{21}) \cdot c_{p1} \quad (Eq. 1)
\]

Where \( ISR = \) insulin secretion rate, \( c_{p1} = \) C-peptide mass in the sampling (accessible) compartment, \( c_{p2} = \) C-peptide mass in the remote compartment, \( k_{12} \) and \( k_{21} \) = rate constants of the exchange between the two C-peptide compartments, and \( k_{01} \) = rate constant of the irreversible loss of C-peptide from the accessible compartment. Note that the values of the volume of distribution of C-peptide pool 1 (accessible compartment), \( k_{12}, k_{21}, \) and \( k_{01} \) are computed according to the equations by Van Cauter et al.(8).

\[
ISR(t) = BSR + DSR(t) + PSR(t) \quad (Eq. 2)
\]

Where \( BSR = \) basal insulin secretion rate, \( DSR = \) insulin secretion rate due to the derivative (or dynamic) component, and \( PSR = \) insulin secretion rate due the proportional (or static) component.

\[
BSR = CP_{ss} \cdot V_1 \cdot k_{01} \quad (Eq. 3)
\]

Where \( CP_{ss} \) is basal C-peptide concentration and \( V_1 \) is the volume of the accessible compartment of C-peptide.
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From the modeling viewpoint, DSR(t) and PSR(t) are the components which in intravenous glucose tolerance tests or hyperglycemic clamps describe classical first phase insulin secretion and second phase insulin secretion, respectively. Furthermore, from a physiological viewpoint, the sum of BSR and PSR(t) describes the relationship linking glucose concentration and insulin secretion rate, in the absence of the derivative component (DSR).

DSR(t) and PSR(t) are mathematically defined as follows:

\[ DSR(t) = X1(t) \cdot \tau^{-1} \quad (Eq. 4) \]

\[ dX1(t) / dt = \sigma1 \cdot [dG(t)/dt]/[\log(1.1+t)] - X1(t) \cdot \tau^{-1} \quad \text{if } dG(t)/dt > 0 \quad (Eq. 5) \]

\[ dX1(t) / dt = - X1(t) \cdot \tau^{-1} \quad \text{if } dG(t)/dt \leq 0 \quad (Eq. 6) \]

Where \( \sigma1 \) = glucose sensitivity of derivative control of insulin secretion, \( G \) = plasma glucose concentration, \( X1 \) = C-peptide (insulin) mass made available for the derivative component of insulin secretion, \( \tau \) = time constant of the derivative component of insulin secretion, and the term \( \log(1.1+t) \) accommodates the time-associated decline of \( \sigma1 \) documented in humans during a hyperglycemic stimulus (9).

\[ PSR(t) = X2(t) \cdot \delta^{-1} \quad (Eq. 7) \]

\[ dX2(t) / dt = \sigma2 \cdot [G(t) - \theta] - X2(t) \cdot \delta^{-1} \quad (Eq. 8) \]

where \( \sigma2 \) = glucose sensitivity of the proportional component of insulin secretion, \( X2 \) = C-peptide (insulin) mass made available for the proportional component of insulin secretion, \( \delta \) = time constant of the proportional component of insulin secretion, \( \theta \) = glucose threshold above which the beta-cell responds with the proportional component of insulin secretion to plasma glucose concentration.

This model was implemented in the SAAM 1.2 software (SAAM Institute, Seattle, WA) (10) to estimate its unknown parameters. Numerical values of the unknown parameters were estimated by using nonlinear least squares. Weights were chosen optimally, i.e., equal to the inverse of the variance of the measurement errors, which were assumed to be additive, uncorrelated, with zero mean, and a coefficient of variation (CV) of 6-8%. The unknown parameters of the model are: \( CP_{ss}, \sigma1, \tau, \sigma2, \delta, \) and \( \theta \). They were estimated with good precision, as shown by their CVs (ESM table 1). A good fit of the model to data was obtained as shown by the table of the weighted residuals (ESM table 2).

There are two main physiological outputs of the model:
1. Derivative control (units: \([\text{pmol.m}^{-2}.\text{BSA}] \cdot [\text{mmol.l}^{-1} \cdot \text{min}^{-1}]^{-1}\)): it is the amount of insulin secreted in response to a rate of glucose increase of 1 mmol/l per min which lasts for 1 minute;
2. Proportional control, i.e. the stimulus-response curve linking glucose concentration (x axis) to insulin secretion rate (y axis): as explained above, it is the sum of BSR and PSR. With the purpose of avoiding artifactual increases in the power of statistical analyses, we used the stimulus-response curve at the predetermined glucose concentrations of 5.5, 8.0, 11.0, 15.0 and 20.0 mmol/l.

RESULTS

Beta-cell function
At baseline both derivative control and proportional control of beta-cell function were almost superimposable in both groups (fig. 1a and 1b). After physical training, the derivative control showed a nonsignificant fall in both groups (p=0.65). After physical training, the proportional control of beta-cell function...
function showed a tendency to rise in the AER group and to fall in the FOR group (fig. 1), with the interaction term time-by-group hitting just the statistical significance (p=0.05). However, the direct comparison of proportional control of beta-cell function after physical training between the AER group and the FOR group was short of statistical significance (p=0.07).
SUPPLEMENTARY DATA

Supplementary Table 1. Coefficients of variation of the beta-cell model parameters. \( CP_{ss} \) = basal C-peptide concentration; \( \sigma_1 \) = parameter regulating glucose sensitivity of derivative control of insulin secretion, \( \tau \) = time constant of derivative control of insulin secretion, \( \sigma_2 \) = glucose sensitivity of proportional control of insulin secretion, \( \delta \) = time constant of proportional control of insulin secretion, \( \theta \): glycemic threshold of proportional control of insulin secretion.

<table>
<thead>
<tr>
<th>Model Parameter</th>
<th>Coefficients of Variation (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Median I.Q. Range</td>
</tr>
<tr>
<td>( CP_{ss} )</td>
<td>11.0 6.5 – 15.0</td>
</tr>
<tr>
<td>( \sigma_1 )</td>
<td>42.5 30.7 – 93.3</td>
</tr>
<tr>
<td>( \tau )</td>
<td>61.0 58.9 – 61.2</td>
</tr>
<tr>
<td>( \sigma_2 )</td>
<td>12.4 9.4 – 16.7</td>
</tr>
<tr>
<td>( \delta )</td>
<td>39.9 22.7 – 61.1</td>
</tr>
<tr>
<td>( \theta )</td>
<td>15.9 9.9 – 25.7</td>
</tr>
</tbody>
</table>

Supplementary Table 2. Weighted residuals of the model fit to the C-peptide data of the OGTT. Data are means±SD. The weighted residuals are a quantitative point-by-point assessment of the goodness-of-fit of the model to the data: a theoretically perfect fit should generate weighted residuals with mean 0 and SD of 1.

<table>
<thead>
<tr>
<th>Time</th>
<th>15’</th>
<th>30’</th>
<th>45’</th>
<th>60’</th>
<th>90’</th>
<th>120’</th>
<th>150’</th>
<th>180’</th>
<th>210’</th>
<th>240’</th>
<th>270’</th>
<th>300’</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean</td>
<td>-0.85</td>
<td>-0.27</td>
<td>+0.39</td>
<td>+0.57</td>
<td>+0.28</td>
<td>-0.004</td>
<td>-0.28</td>
<td>-0.45</td>
<td>+0.10</td>
<td>+0.58</td>
<td>+0.19</td>
<td>+0.26</td>
</tr>
<tr>
<td>SD</td>
<td>1.10</td>
<td>0.99</td>
<td>1.16</td>
<td>1.17</td>
<td>1.49</td>
<td>1.22</td>
<td>1.02</td>
<td>1.30</td>
<td>1.31</td>
<td>1.11</td>
<td>1.10</td>
<td>0.91</td>
</tr>
</tbody>
</table>

Supplementary Figure 1.  

a. Left panel: Derivative control of beta-cell function before and after physical training in the AER group and in the FOR group. No statistically significant differences were detected.

b. Right panel: Proportional control of beta-cell function before and after physical training in the AER group and in the FOR group. P=0.05 for the interaction term time-by-group. P=0.07 for the contrast post-intervention AER group vs post-intervention FOR group.
REFERENCES


