Oral Glucose Tolerance Test Minimal Model Indexes of β-Cell Function and Insulin Sensitivity

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The simultaneous assessment of quantitative indexes of insulin secretion and action in a single individual is important when quantifying their relative role in the evolution of glucose tolerance in different physiopathological states. Available methods quantify these indexes in relatively nonphysiological conditions, e.g., during glucose clamps or intravenous glucose tolerance tests. Here, we present a method based on a physiological test applicable to large-scale genetic and epidemiologic studies—the oral glucose tolerance test (OGTT). Plasma C-peptide, insulin, and glucose data from a frequently sampled OGTT with 22 samples throughout 300 min ($FSOGTT_{300\text{-}22}$) were analyzed in 11 subjects with various degrees of glucose tolerance. In each individual, two indexes of pancreatic sensitivity to glucose (Φ_s [10^9 min^{-1}] and Φ_d [10^9]) and the insulin sensitivity index (S_I) (10^5 dl/kg per min per pmol/l) were estimated by using the minimal model of C-peptide secretion and kinetics originally proposed for intravenous graded glucose infusion and the minimal model approach recently proposed for meal/OGTTs. The indexes obtained from $FSOGTT_{300\text{-}22}$ were used as a reference for internal validation of OGTT protocols with reduced sampling schedules. Our results show that 11 samples in a 300-min period ($OGTT_{300\text{-}11}$) is the test of choice because the indexes it provides (Φ_s = 36 ± 3 [means ± SE]; Φ_d = 710 ± 111; S_I = 10.2 ± 2.4) show excellent correlation and are not statistically different from those of $FSOGTT_{300\text{–}22}$ (Φ_s = 33 ± 3; Φ_d = 715 ± 120; S_I = 10.1 ± 2.3). In conclusion, $OGTT_{300\text{-}11}$, interpreted with C-peptide and glucose minimal models, provides a quantitative description of β-cell function and insulin sensitivity in a single individual while preserving the important clinical classification of glucose tolerance provided by the standard 120-min OGTT. *Diabetes* 50:150–158, 2001

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CV, coefficient of variation; DI, disposition index; $FSOGTT_{300\text{-}22}$, frequently sampled OGTT with 22 samples throughout 300 min; IGT, impaired glucose tolerance; ISR, insulin secretion rate; IVGTT, intravenous glucose tolerance test; NGT, normal glucose tolerance; OGTT, oral glucose tolerance test; S_I, insulin sensitivity index.

The simultaneous assessment of β-cell function and insulin sensitivity in a single individual is of primary importance when quantifying their relative role in the evolution of glucose tolerance in different physiopathological states. Two methods are available for this purpose: the clamp technique, which uses a euglycemic-hyperinsulinemic and a hyperglycemic-euinsulinemic clamp in the same individual (1), and the intravenous glucose tolerance test (IVGTT) interpreted by the minimal models of glucose disposal (2) and C-peptide kinetics and secretion (3). Both these approaches give accurate and precise estimates of insulin sensitivity and β-cell function in a single individual, but plasma glucose, C-peptide, and insulin concentrations achieved during these studies are relatively nonphysiological. Recently, the need to quantify β-cell function and insulin sensitivity under more normal life conditions has encouraged many investigators to use more physiological protocols, including meal-like studies (4), graded up and down glucose infusions (5), meals (6,7), and oral glucose tolerance tests (OGTTs) (8,9). However, an approach to measure indexes of β-cell function and insulin action in a single individual based on a physiological test such as the OGTT is not available. The ability to derive in a single individual important information such as the clinical classification of oral glucose tolerance while simultaneously quantifying his or her β-cell function and insulin sensitivity could provide a unique tool potentially applicable to large-scale genetic and epidemiologic studies.

Therefore, the aim of the present study was to investigate whether indexes of β-cell function and insulin sensitivity could be simultaneously assessed in a single individual from OGTT data by extending to the OGTT the up and down C-peptide minimal model (5) and the insulin sensitivity formula recently derived for a meal (7). The database consisted of a frequently sampled 300-min OGTT performed on 11 subjects with various degrees of glucose tolerance. Indexes of insulin sensitivity and β-cell function based on OGTTs with reduced number of samples were also calculated and compared with those derived from the frequently sampled OGTT to arrive at a robust clinical protocol.

RESEARCH DESIGN AND METHODS

Subjects. Studies were performed on 11 nondiabetic subjects (4 men and 7 women); 7 had normal glucose tolerance (NGT), and 4 had impaired glucose tolerance (IGT). Mean age was 37 ± 3 years (means ± SE) (range 20–50), and BMI was 30.5 ± 2.1 kg/m^2 (range 21.3–46.2) (Table 1). Glucose tolerance was determined by using the American Diabetes Association Expert Committee criteria (10). All subjects had a normal screening blood count and chemistries and took no medications known to affect insulin secretion or action. All protocols were approved by the Institutional Review Board of The University of Chicago. Written informed consent was obtained from each subject.

Protocol. All studies were performed in the Clinical Research Center at the University of Chicago starting at 0800 in the morning after an overnight fast. Intravenous

TABLE 1
Clinical characteristics of the study subjects

	Glucose tolerance	Age (years)	Sex (M/F)	Weight (kg)	Height (cm)	BMI (kg/m^2)
Subject						
1	IGT	20	F	75.7	161.7	29.0
2	NGT	50	F	82.6	165.8	30.0
3	NGT	28	F	68.4	169.6	23.8
4	IGT	30	M	64.0	173.5	21.3
5	NGT	44	M	94.9	191.5	25.9
6	NGT	45	M	73.7	162.8	27.8
7	IGT	37	F	99.6	164.2	36.9
8	NGT	26	M	100.7	182.2	30.3
9	IGT	36	F	100.4	147.4	46.2
10	NGT	43	F	84.9	168.8	29.8
11	NGT	46	F	92.7	164.1	34.4
Mean		37	4/7	85.2	168.3	30.5
SE		3	—	4.0	3.5	2.1

catheters were placed into antecubital veins. The blood sampling arm was heated to obtain arterialized venous samples. At time 0, subjects ingested a 75-g glucose load. Blood samples were collected at –15, 0, 10, 20, 30, 45, 60, 75, 90, 105, 120, 135, 150, 165, 180, 195, 210, 225, 240, 255, 270, 285, and 300 min for measurement of glucose, insulin, and C-peptide concentrations. This complete protocol, consisting of 22 blood samples taken in a 300-min period after glucose ingestion, will be referred to as frequently sampled OGTT (FSOGTT$_{300\text{-}22}$).

OGTT protocols with reduced data sets were also considered (Fig. 1): *1*) sampling schedule similar to the one commonly adopted for a standard OGTT, with samples at 0, 30, 60, 90, 120, 150, 180, 240, and 300 min (9 samples throughout 300 min) referred to as OGTT$_{300\text{-}9}$; *2*) same as *1*) with two additional samples at 10 and 20 min (OGTT$_{300\text{-}11}$); *3*) same as *2*) but without the 300-min sample, thus shortening the duration of the test from 5 to 4 h (OGTT$_{240\text{-}10}$).

Assay. Plasma glucose was measured immediately using a glucose analyzer (YSI Model 2300 STAT; Yellow Springs Instruments, Yellow Springs, OH). The coefficient of variation (CV) of this method was <2%. Serum insulin was assayed by a double-antibody technique (11) with a lower limit of sensitivity of 20 pmol/l and an average intra-assay CV of 6%. The cross-reactivity of proinsulin in the radioimmunoassay for insulin was ~40%. Plasma C-peptide was measured as previously described (12). The lower limit of sensitivity of the assay was 0.02 nmol/l and the intra-assay CV averaged 6%.

β-Cell function. The minimal model of C-peptide secretion and kinetics originally applied to intravenous glucose graded infusion data (5) has been applied to assess β-cell secretion during an oral glucose perturbation.

C-peptide kinetics are described by using the well-known two-compartment model originally proposed by Eaton et al. (13):

$$\dot{CP}_1(t) = -[k_{01} + k_{21}]CP_1(t) + k_{12}CP_2(t) + SR(t) \qquad CP_1(0) = 0 \qquad (1)$$

$$\dot{CP}_2(t) = k_{21}CP_1(t) - k_{12}CP_2(t) \qquad CP_2(0) = 0 \qquad (2)$$

where the overdot indicates time derivative; CP_1 and CP_2 (nmol/l) are C-peptide concentrations above basal in the accessible and peripheral compartments, respectively; k_{ij} (min^{-1}) are C-peptide kinetic parameters; and SR (pmol · l^{-1} · min^{-1}) is the pancreatic secretion above basal, entering the accessible compartment, normalized by the volume of distribution of compartment 1.

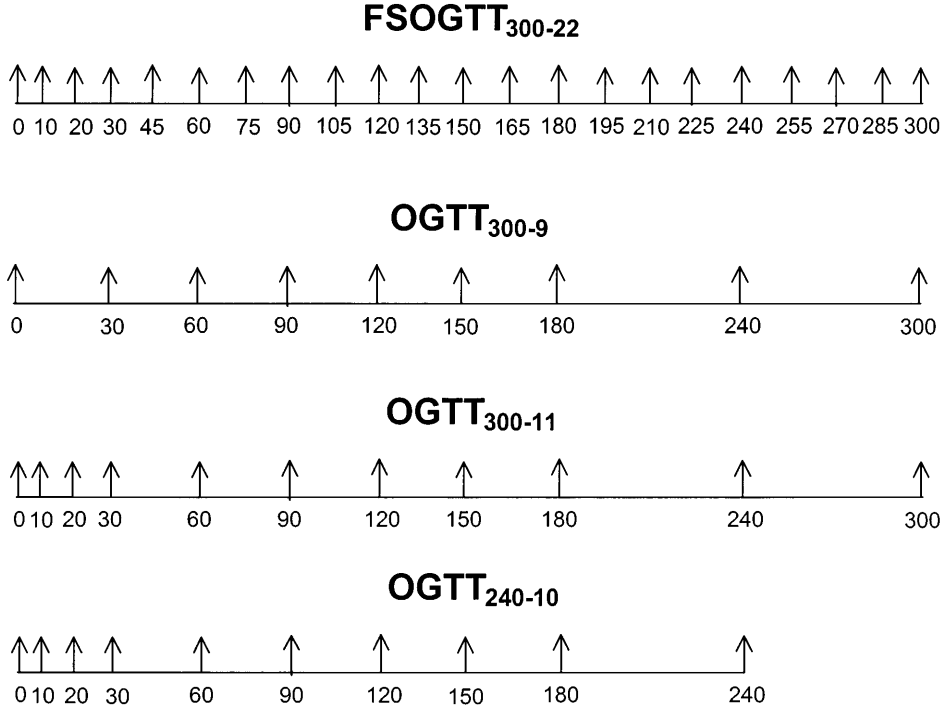

FIG. 1. The original FSOGTT$_{300\text{-}22}$ sampling schedule and its reduced versions.

Pancreatic secretion SR has been described as the sum of two components controlled respectively by glucose concentration (static glucose control [SR_s]) and by its rate of increase (dynamic glucose control [SR_d]):

$$SR(t) = SR_s(t) + SR_d(t) \tag{3}$$

SR_s is assumed to be equal to the provision of new insulin to the β-cells (Y) (pmol·l^{-1}·min^{-1}):

$$SR_s(t) = Y(t), \tag{4}$$

which is controlled by glucose according to the following:

$$\dot{Y}(t) = \alpha\{Y(t) - \beta[G(t) - h]\} \qquad Y(0) = 0 \tag{5}$$

It is worth noting that SR_s is not linearly related to glucose concentration but tends, with a time constant 1/α (min), toward a steady-state value linearly related through the parameter β to glucose concentration G above a threshold level h (mmol/l).

SR_d, on the other hand, represents the secretion of insulin stored in the β-cells in a promptly releasable form (labile insulin) and is proportional to the rate of increase of glucose:

$$SR_d(t) = \begin{cases} k(G) \cdot \dot{G}(t) & \text{if} \quad \dot{G}(t) > 0 \\ 0 & \text{if} \quad \dot{G}(t) \le 0 \end{cases} \tag{6}$$

where:

$$k(G) = \begin{cases} K_d \cdot \left(1 - \dfrac{G(t) - G_b}{G_t - G_b}\right) & \text{if} \quad G_b \le G(t) < G_t \\ 0 & \text{otherwise} \end{cases} \tag{7}$$

According to Eqs. 6 and 7, the dynamic control is at its maximum when glucose increases just above its basal value G_b, then it decreases linearly with glucose concentration and vanishes when glucose concentration exceeds the threshold level G_t, which is able to promote the secretion of all stored insulin. If parameter G_t assumes an elevated value, $k(G)$ approximates the constant K_d.

Indexes. The model allows the estimation of two indexes of β-cell function: the static and the dynamic sensitivity to glucose. In addition, a single global index of β-cell sensitivity to glucose, which suitably combines both the static and the dynamic control indexes, can be calculated.

Static. The static sensitivity (Φ_s [10^9 min^{-1}]) is a measure of the effect of glucose on β-cell secretion and is the ratio between SR and glucose concentrations (above the threshold level h) at steady state:

$$\Phi_s = \beta \tag{8}$$

Dynamic. The dynamic sensitivity (Φ_d [10^9]) is a measure of the stimulatory effect of the rate at which glucose increases upon the secretion of stored insulin. It is defined as the amount of insulin (per unit of C-peptide distribution volume) released in response to the maximum glucose concentration G_{max} achieved during the experiment, normalized by the glucose increase $G_{max} - G_b$:

$$\Phi_d = \frac{\int_{G_b}^{G_{max}} k(G)\,dG}{G_{max} - G_b} = \begin{cases} K_d \cdot \left[1 - \dfrac{G_{max} - G_b}{2 \cdot (G_t - G_b)}\right] & \text{if} \quad G_t > G_{max} \\ \dfrac{K_d \cdot (G_t - G_b)}{2 \cdot (G_{max} - G_b)} & \text{if} \quad G_t \le G_{max} \end{cases} \tag{9}$$

If parameter G_t assumes an elevated value, Φ_d approximates the constant K_d.

Global. In addition to Φ_s and Φ_d, which give a detailed portrait of β-cell function, it is also useful to define and derive a single global index of β-cell sensitivity to glucose (Φ [10^9 min^{-1}]), which suitably combines both the static (Φ_s) and the dynamic (Φ_d) control indexes. This is particularly advantageous in calculating the so-called disposition index, i.e., β-cell function × insulin sensitivity (see below).

The global index of β-cell sensitivity to glucose is defined as the average increase above basal of pancreatic secretion (Eq. 3) over the average glucose stimulus above the threshold level h:

$$\Phi = \frac{\int_0^\infty SR(t)\,dt}{\int_0^\infty [G(t) - h]\,dt} \tag{10}$$

Φ (see APPENDIX) can be calculated from model parameter h, model indexes Φ_s and Φ_d, and the area under the curve of G above the threshold level h:

$$\Phi = \Phi s + \frac{\Phi_d \cdot (G_{max} - G_b)}{\int_0^\infty [G(t) - h]\,dt} \tag{11}$$

Φ has been derived from the model, but it can also be calculated by a virtually model-independent formula (see APPENDIX):

$$\Phi = \frac{k_{01}\int_0^\infty CP_1(t)\,dt}{\int_0^\infty [G(t) - h]\,dt} \approx \frac{k_{01}\int_0^\infty CP_1(t)\,dt}{\int_0^\infty [G(t) - G_b]\,dt} \tag{12}$$

Insulin secretion rate. The model also provides the profile of the insulin secretion rate (ISR [pmol/min]) during the OGTT:

$$ISR\ (t) = [SR_b + SR(t)] \cdot V_1 \tag{13}$$

where SR_b (pmol·min^{-1}·l^{-1}) is insulin secretion in the basal state, and V_1 (liters) is the distribution volume of the accessible compartment.

Insulin sensitivity. To calculate insulin sensitivity, we have applied to the OGTT the formula for a meal recently proposed by Caumo et al. (7). As detailed in this study, insulin sensitivity index (S_I [10^5 dl/kg per min per pmol/l]) can be calculated with an area under the curve formula:

$$S_{I(OGTT)} = \frac{f \cdot D_{OGTT} \cdot \dfrac{AUC[\Delta G(t)/G(t)]}{AUC[\Delta G(t)]} - GE \cdot AUC[\Delta G(t)/G(t)]}{AUC[\Delta I(t)]} \tag{14}$$

where G is plasma glucose concentration; ΔG and ΔI are glucose and insulin concentrations above basal, respectively; AUC denotes the area under the curve calculated from time 0 to $t \to \infty$; GE is glucose effectiveness (dl · kg^{-1} · min^{-1}); D_{OGTT} is the dose of ingested glucose per unit of body weight (mg/kg); and f is the fraction of ingested glucose that actually appears in the systemic circulation. When glucose falls below basal, a slightly different formula needs to be used (we refer to Eq. 7 in Caumo et al. (7) for details). Calculation of S_I requires insertion of values for GE and f. Here we used the values proposed by Caumo et al.: GE = 0.024 dl · kg^{-1} · min^{-1} and f = 0.8.

Disposition index. The so-called disposition index (DI), i.e., β-cell function × insulin sensitivity (14,15), is a parsimonious and effective way to express β-cell function in relation to the degree of insulin resistance. To this end, it is convenient to use the single global β-cell function index Φ. The DI is thus defined as follows:

$$DI = \Phi \cdot S_I \tag{15}$$

Numerical identification. All C-peptide model parameters are a priori uniquely identifiable. However, numerical identification of the model requires the knowledge of C-peptide kinetics. Standard kinetic parameters were calculated by using the method proposed by Van Cauter et al. (16).

C-peptide model secretory parameters were estimated, together with a measure of their precision, by nonlinear least squares (17,18) using SAAM II software (19). When α was elevated and estimated with poor precision, the Bayesian approach implemented in SAAM II was used. Measurement error of C-peptide concentration has been assumed to be independent and gaussian, with zero mean with a constant but unknown variance. Glucose concentration, linearly interpolated between data, and its time derivative have been assumed as error-free model inputs. Area under the curve in the global index and insulin sensitivity formulas was calculated using the trapezoidal rule.

To evaluate the precision of S_I, Monte Carlo methods (20) have been applied to Eq. 14 (or, when appropriate, to Eq. 7 from Caumo et al. [7]), taking into account both measurement errors of glucose and insulin concentrations (assumed to be independent and gaussian, with zero mean and a constant CV of 2 and 6%, respectively) and population variability of f and glucose effectiveness (assumed to be gaussian with a CV of 10 and 25%, respectively [7]).

Statistical analyses. Results are given as means ± SE. The statistical significance of differences between the same parameters calculated from different sampling schedules has been calculated using the Wilcoxon's signed-rank test. Linear regression and Spearman rank correlation analyses were used to examine the relationship between parameters. Significance was declared at $P < 0.05$.

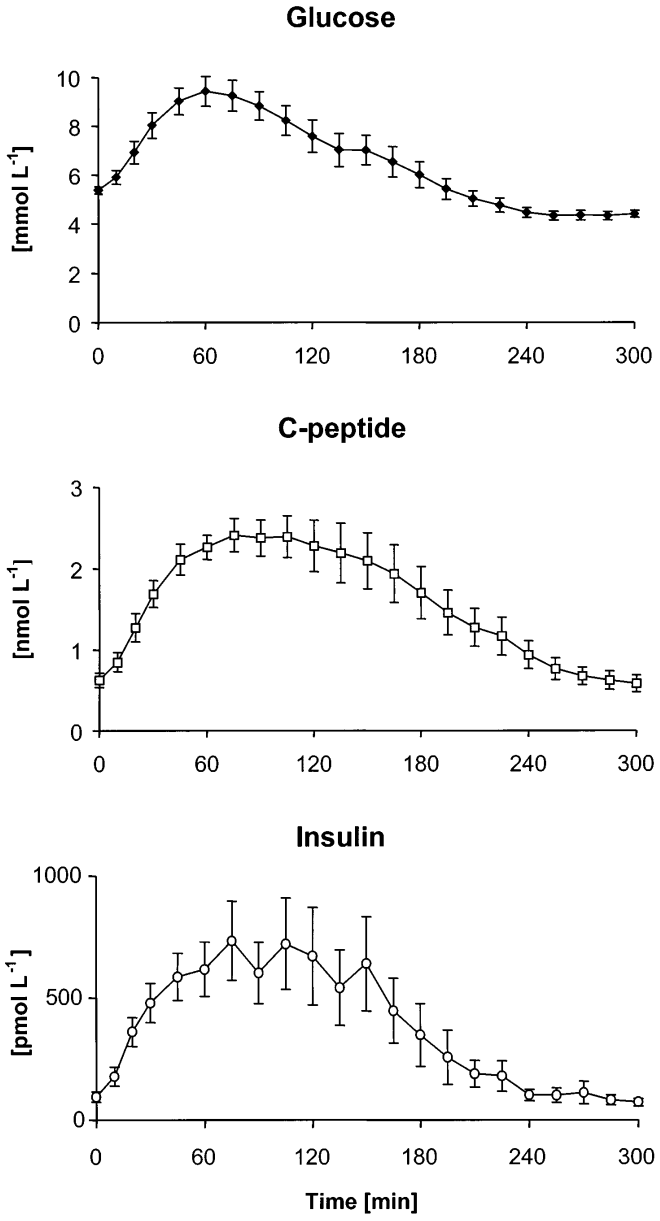

FIG. 2. Average (mean ± SE, n = 11) concentration of plasma glucose, C-peptide, and insulin obtained during the 75-g FSOGTT$_{300\text{-}22}$.

RESULTS

FSOGTT$_{300\text{-}22}$. Mean plasma glucose, C-peptide, and insulin concentrations during the FSOGTT$_{300\text{-}22}$ are shown in Fig. 2. The C-peptide minimal model well describes experimental data, as shown by the weighted residual plot (Fig. 3). Average β-cell sensitivity indexes were as follows: Φ_d = 715 ± 120 and Φ_s = 33 ± 3 (means ± SE). They were estimated with good precision for all subjects with an average CV of 25 ± 4 and 9 ± 1%, respectively. The ISR profile was also reconstructed and is shown in Fig. 4. Average S_I was 10.1 ± 2.3, and its precision averaged 12 ± 1%.

OGTT$_{300\text{-}9}$. The C-peptide minimal model is obviously able to fit the reduced data set (Fig. 3), but Φ_d values (1,114 ± 224, CV 44 ± 9%) were statistically different from those estimated by using the FSOGTT$_{300\text{-}22}$ (Fig. 5) and were not correlated with them (Fig. 6). The early portion of the ISR profiles (not shown) was not superimposable to that calculated using the FSOGTT$_{300\text{-}22}$. These results indicate that OGTT$_{300\text{-}9}$ does not accurately describe the early portion of the data, where the dynamic control of glucose is active. The values of Φ_s (35 ± 4, CV 15 ± 4%) and S_I (10.1 ± 2.4, CV 13 ± 1%), on the other hand, did not significantly change (Figs. 5 and 6).

OGTT$_{300\text{-}11}$. All the indexes, including Φ_d, were not different from and were well correlated with those obtained from the FSOGTT$_{300\text{-}22}$ (Φ_d = 710 ± 111, CV 39 ± 5%; Φ_s = 36 ± 3, CV 13 ± 3%; S_I = 10.2 ± 2.4, CV 13 ± 1%) (Figs. 5 and 6). Individual values of Φ_d, Φ_s, and S_I are summarized in Table 2, together with their precision. The average ISR profiles estimated using the two protocols were also very similar (Fig. 4), thus indicating that the more accurate description of the early portion of the experiment by OGTT$_{300\text{-}11}$ with respect to OGTT$_{300\text{-}9}$ is essential to obtain results similar to those of the FSOGTT$_{300\text{-}22}$. The weighted residual plot of the C-peptide minimal model is shown in Fig. 3.

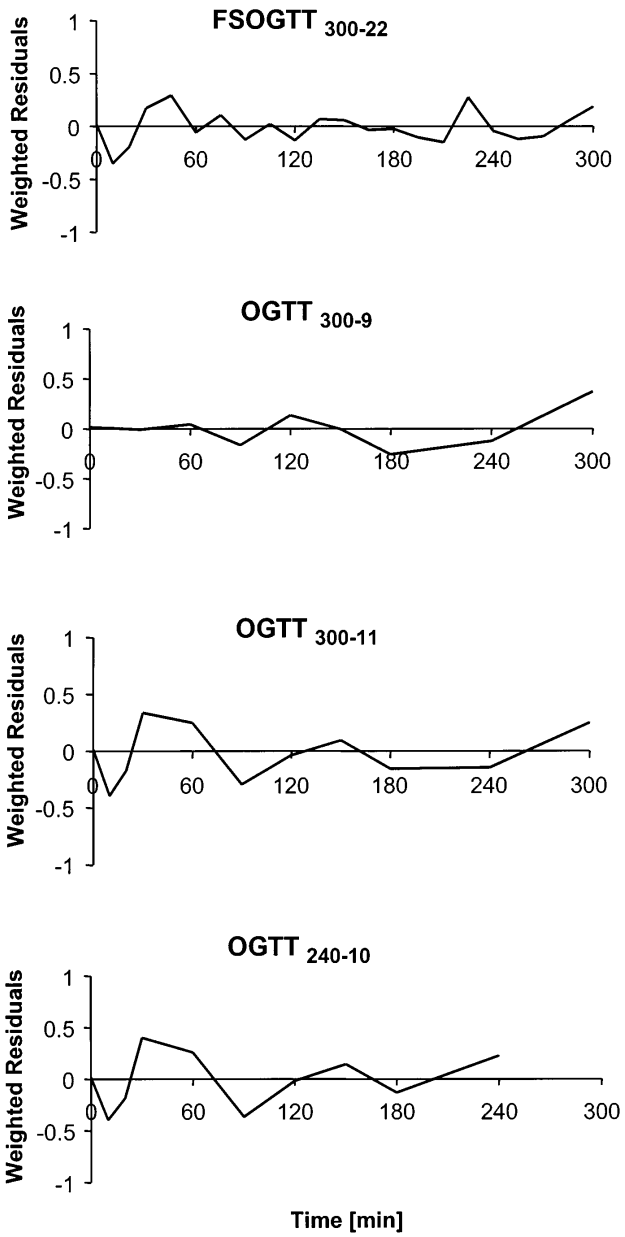

FIG. 3. Average weighted residuals of the C-peptide minimal model.

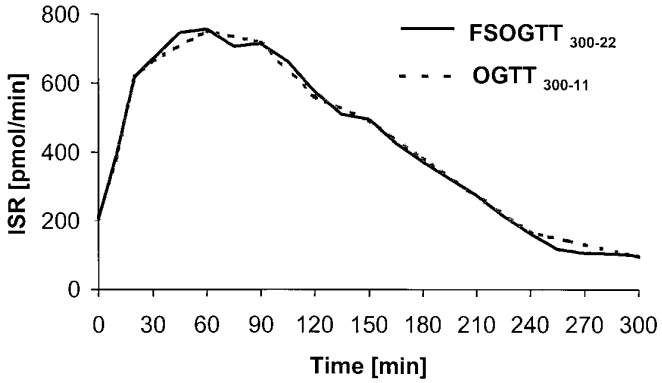

FIG. 4. Mean β-cell secretion during $FSOGTT_{300\text{-}22}$ and $OGTT_{300\text{-}11}$.

$OGTT_{240\text{-}10}$. β-Cell sensitivity indexes Φ_d (710 ± 126, CV 40 ± 7%), Φ_s (35 ± 3, CV 12 ± 1%), and S_I (8.9 ± 2.1, CV 15 ± 1%) were well correlated with those obtained during the $FSOGTT_{300\text{-}22}$ (Fig. 6). However, Φ_d and Φ_s did not significantly change, whereas S_I was statistically different from values calculated during the $FSOGTT_{300\text{-}22}$ (Fig. 5), thus indicating the importance of the 300-min sample for an accurate estimation of insulin sensitivity. The weighted residual plot of the C-peptide minimal model is shown in Fig. 3.

DISCUSSION

In this study, minimal model indexes of β-cell function and insulin sensitivity have been proposed and successfully assessed during an 11-sample 300-min OGTT in individuals with various degrees of glucose tolerance. While maintaining the possibility of estimating indexes in the single individual, the $OGTT_{300\text{-}11}$ offers a number of additional advantages with respect to available approaches based on the clamp or IVGTT techniques: the oral perturbation reproduces a physiological condition, and the test is easy to perform, with potential application to large scale genetic and epidemiologic studies. These features make it an appealing tool because it may exhibit a larger statistical power than population-oriented indexes such as those proposed by Matthews et al. (21) and Matsuda and DeFronzo (22).

Minimal models, through a parsimonious description of glucose/C-peptide and glucose/insulin relationships, provide reliable indexes of β-cell function and insulin sensitivity. More precisely, β-cell sensitivity indexes have been calculated by extending to the OGTT data the C-peptide minimal model recently developed for intravenous glucose graded up and down infusion (5). The model assumptions are particularly favorable to an OGTT protocol, i.e., a situation where C-peptide and glucose concentrations show slow dynamics compared with the IVGTT, where excursions are rapid and large. The model has already been successfully used to assess β-cell function during physiological tests in normal subjects (5) and in individuals with various degrees of glucose tolerance (23). It incorporates the assumption that glucose stimulates pancreatic insulin secretion by exerting both a static (dependent on glucose concentration) and a dynamic (dependent on glucose rate of change) control. The model provides the insulin secretion profile and indexes of β-cell function: the static Φ_s, the dynamic Φ_d, and the global Φ sensitivity to glucose.

S_I has been calculated by using a formula recently proposed for a meal glucose tolerance test (7). The approach associates a parsimonious parametric representation of splanchnic glucose absorption with the minimal model description of glucose disposal. This method has been validated by comparing its S_I estimates to those provided by an insulin-modified IVGTT performed on the same group of 10 normal subjects. The significant correlation between the two S_I indexes ($\rho_s = 0.89$, $P < 0.01$) indicates that a reliable measure of insulin sensitivity can be derived from either an oral test or an IVGTT.

To derive reliable estimates of β-cell function and insulin sensitivity during an OGTT, a rich database was initially used, with 22 samples taken in a 300-min OGTT ($FSOGTT_{300\text{-}22}$). Then, because our objective was to propose a protocol sufficiently simple but still robust, we examined reduced sampling schedule OGTTs by using the indexes obtained from the $FSOGTT_{300\text{-}22}$ as a reference for internal validation. The results indicate that 11 samples in a 300-min OGTT ($OGTT_{300\text{-}11}$) are sufficient to obtain results similar to those from the $FSOGTT_{300\text{-}22}$, since the two sets of indexes show high correlation (Fig. 6) and are not statistically different (Fig. 5). Also, the ISR profiles are virtually superimposable (Fig. 4). A

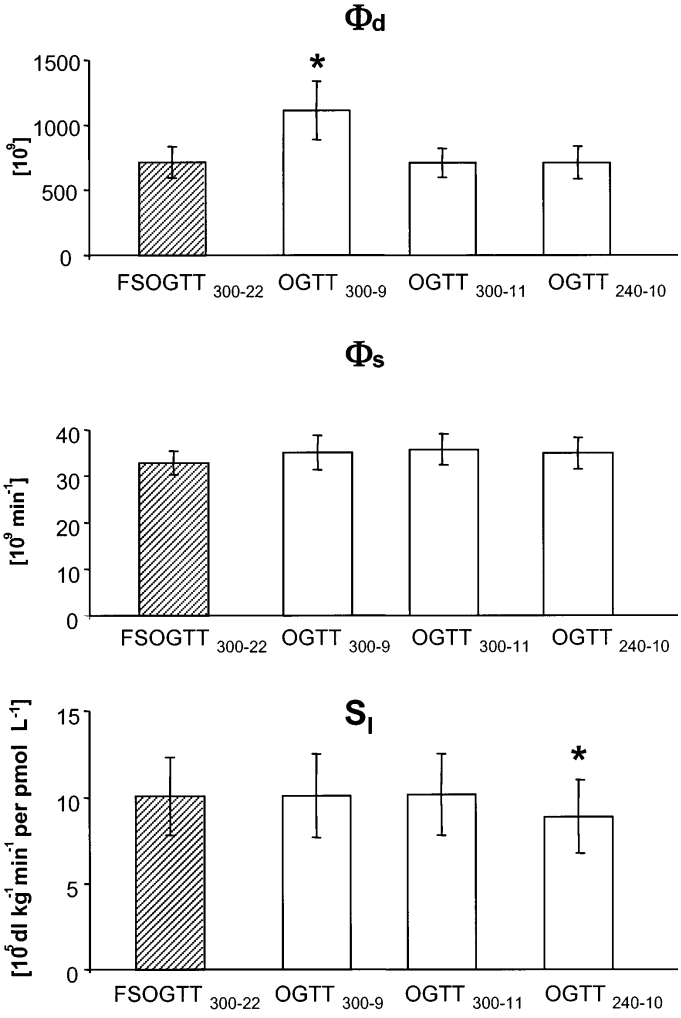

FIG. 5. Effect of reduced sampling schedules on indexes Φ_d, Φ_s, and S_I compared with the same indexes obtained from the $FSOGTT_{300\text{-}22}$. *Comparison to $FSOGTT_{300\text{-}22}$: $P < 0.05$.

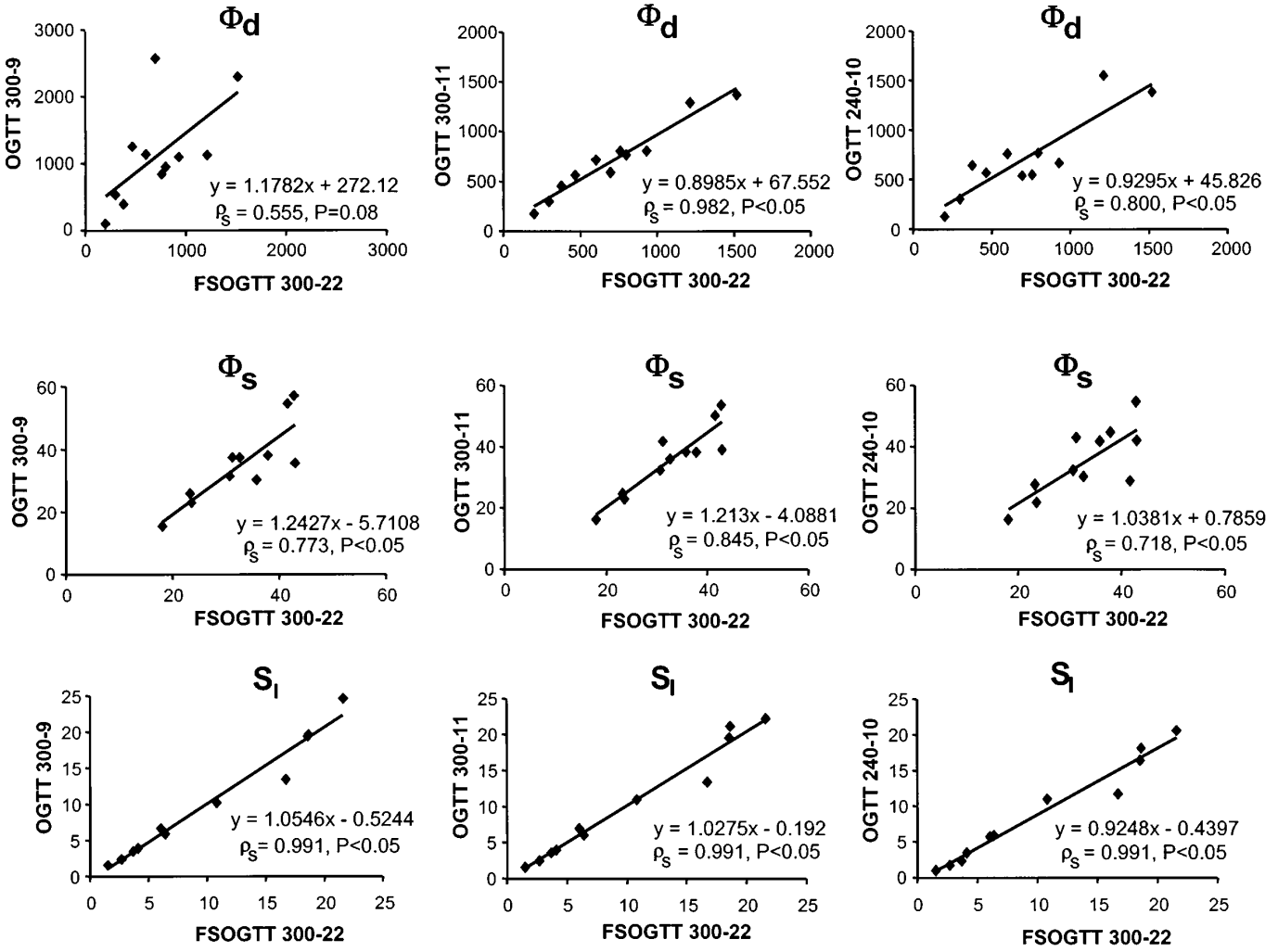

FIG. 6. Regression plots and Spearman rank coefficient of correlation (ρ_s) between the indexes Φ_d, Φ_s, and S_I obtained from the $FSOGTT_{300-22}$ and the same indexes obtained from the $OGTT_{300-9}$, $OGTT_{300-11}$, and $OGTT_{240-10}$, respectively.

300-min experiment is necessary to obtain such results because the values estimated for S_I during a shorter experiment ($OGTT_{240-10}$) are slightly lower than those estimated during a 300-min experiment ($FSOGTT_{300-22}$, $OGTT_{300-9}$, and $OGTT_{300-11}$). The accurate description of the early portion of the C-peptide and glucose curves, provided by the 10, 20, 30 min samples after the oral glucose ingestion, is also necessary to obtain reliable estimates of the index Φ_d.

OGTT minimal model indexes: NGT versus IGT. The standard 120-min OGTT, with blood samples usually drawn at 0, 30, 60, 90, and 120 min, provides the important clinical classification of glucose tolerance (10). The $OGTT_{300-11}$ we propose here includes the standard samples, thus still allowing one to perform the standard classification of glucose tolerance but also enabling one to estimate indexes of β-cell function and insulin sensitivity, which help to better characterize glucose tolerance in a single individual. From our data, when $OGTT_{300-11}$ indexes in NGT subjects (n = 7) were compared with those obtained in IGT subjects (n = 4), no difference was found in the pancreatic sensitivity indexes (NGT: $\Phi_d = 728 \pm 124$, $\Phi_s = 37 \pm 4$, $\Phi = 48 \pm 6$; IGT: $\Phi_d = 678 \pm 241$, $\Phi_s = 34 \pm 6$, $\Phi = 42 \pm 9$), but a statistically significant difference was found in both the S_I (NGT 14.0 ± 2.7, IGT 3.4 ± 1.0) and the DI (NGT 678 ± 178, IGT 160 ± 80).

These results confirm that subjects with IGT are characterized by an inadequate insulin secretory response for the degree of insulin resistance (24) or, in other words, that IGT is characterized by a relative, rather than absolute, insulin deficiency.

OGTT versus intravenous glucose infusions. Pancreatic indexes Φ_s and Φ_d estimated from $OGTT_{300-11}$ were also compared with the same indexes estimated during either IVGTT or graded up and down glucose infusions. The IVGTT counterparts of Φ_d and Φ_s are respectively the first- and second-phase sensitivity indexes Φ_1 and Φ_2. The values obtained during an insulin-modified IVGTT (300 mg/kg) in normal subjects (n = 15) were $\Phi_2 = 10.9 \pm 1.4$ and $\Phi_1 = 191 \pm 29$ (25); β-cell indexes obtained during graded up and down glucose infusions (0, 4, 8, 16, 8, 4, and 0 $mg \cdot kg^{-1} \cdot min^{-1}$) in nondiabetic subjects (n = 8) were $\Phi_s = 18.8 \pm 1.8$ and $\Phi_d = 222 \pm 30$ (5). Both Φ_s and Φ_d are threefold higher during OGTT than during intravenous tests. This can probably be ascribed to the presence of the well-known insulin-stimulating gastrointestinal hormones known as the incretin effect, which are secreted in response to oral but not intravenous glucose administration (26). It will be of interest in future studies to compare β-cell indexes and S_I obtained from OGTTs with values obtained in

TABLE 2
Quantitative indexes of β-cell function and insulin sensitivity

	Φ_d (10^9)	Φ_s (10^9 min^{-1})	S_I (10^5 dl/kg per min per pmol/l)
Subject			
1	172 (10)	23 (5)	3.6 (12)
2	715 (61)	42 (19)	4.0 (15)
3	763 (60)	32 (13)	13.4 (12)
4	802 (45)	50 (40)	6.0 (19)
5	297 (16)	16 (10)	19.6 (10)
6	562 (31)	38 (10)	21.1 (11)
7	449 (25)	38 (9)	2.4 (10)
8	589 (33)	54 (9)	22.2 (10)
9	1,290 (62)	25 (10)	1.6 (16)
10	805 (45)	36 (10)	11.0 (16)
11	1,366 (46)	39 (11)	6.9 (11)
Mean	710 (39)	36 (13)	10.2 (13)
SE	111 (5)	3 (3)	2.4 (1)

Data in parentheses are precision of parameter estimates, expressed as percent coefficient of variation.

the same subjects during intravenous glucose infusions—possibly during a graded up and down glucose infusion, which better simulates an OGTT. Preliminary results are available on only six of the eleven subjects studied here (four with NGT and two with IGT) who underwent both an OGTT and a graded up and down glucose infusion. The results confirmed our expectations. Indexes of β-cell function (Φ_d and Φ_s) were markedly higher during the oral perturbation (OGTT: Φ_d = 677 ± 154, Φ_s = 39 ± 5; up and down: Φ_d = 105 ± 43, Φ_s = 17 ± 3, $P < 0.05$). The OGTT S_I was higher than the graded up and down S_I (OGTT S_I = 13.1 ± 3.6; up and down S_I = 8.8 ± 2.3, $P <$ 0.05) and showed a high correlation with it (ρ_s = 0.83)—a trend already observed during meal tolerance tests (7).

Importance of the dynamic control of glucose on insulin secretion. Indexes of β-cell function have been recently proposed in the literature based on modeling analyses of glucose and C-peptide data during a meal (6) and a 120-min OGTT (9). Both models assume a control of glucose, but not of its rate of change, on insulin secretion. This is a gross simplification because the importance of dynamic control of glucose on insulin secretion (active when glucose concentration increases) has been shown both in previous studies where graded up and down glucose infusion data were analyzed (5) and in the present study. If a model similar to the OGTT minimal model but not accounting for the dynamic glucose control is used to analyze the data, systematic deviations occur in the early portion of the OGTT (Fig. 7).

The approach proposed by Cretti et al. (9) is simplistic but particularly appealing because it only requires five samples throughout 120 min. To compare our approach with that proposed by Cretti et al., we applied the latter to our data by adopting the sampling schedule proposed in (9), i.e., 0, 30, 60, 90, and 120 min. Model fit and residual plots showed a systematic underestimation of the data between 15 and 75 min, thus indicating that the model proposed by Cretti et al. is too simplistic to describe OGTT data. The apparent glucose threshold θ (9) assumed values (often ~0) far from basal glucose, and β-cell sensitivity σ (9) was different when compared with the minimal model counterpart (Φ_s). This may reflect the fact that σ incorporates both the static and the dynamic glucose controls, whereas Φ_s describes the static glucose control only but is more likely the consequence of numerical compensations due to differences in the estimates of threshold θ in the Cretti et al. model (often ~0) and h in our model (always ~G_b). In conclusion, care must be exercised in adopting simplistic but appealing methods because structural errors can lead to compensations among parameters and consequently to inaccurate β-cell portraits. We think that the protocol and the methods proposed here, which enable one to obtain a precise description of both β-cell function and insulin action in a single individual, are a good compromise between model/protocol simplicity and accuracy.

In conclusion, by extending to the OGTT the recently developed C-peptide minimal model during intravenous glucose graded up and down infusion (5) and by applying to the OGTT the minimal model formula recently proposed for a meal glucose tolerance test (7), we have shown that it is possible to simultaneously assess individual parameters of β-cell function and insulin sensitivity from an 11-sample 300-min OGTT. Of note is that $OGTT_{300\text{-}11}$ preserves the important clinical classification of glucose tolerance provided by a standard 120-min OGTT. The detailed description of β-cell function and insulin action thus available in a single individual, together with the ease of execution of the

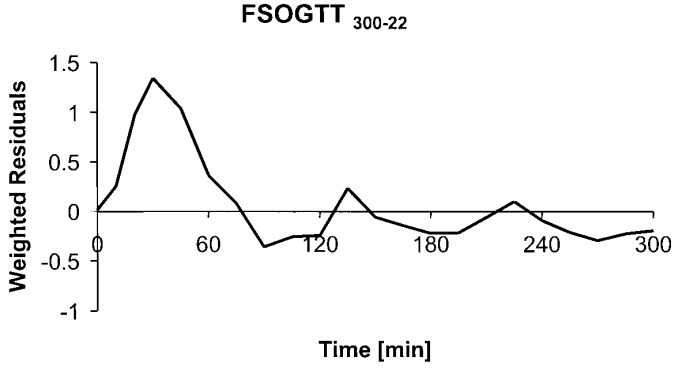

FIG. 7. Inadequacy of a C-peptide model that simply assumes a static glucose control on insulin secretion. Mean weighted residuals show a systematic deviation in the first 60 min.

protocol, should make this approach a powerful tool for measuring changes in insulin secretion and action that would also be applicable to large-scale genetic and epidemiologic studies. The present study is the first attempt to simultaneously assess insulin sensitivity and β-cell function in the single individual during an OGTT and shows encouraging results in subjects with various degrees of glucose tolerance. However, this study is definitely unfinished and further work needs to be performed to define the domain of validity of this approach throughout the whole range of glucose tolerance, including patients with diabetes.

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APPENDIX

The purpose of this section is to derive the global index of β-cell sensitivity to glucose, Φ.

Model dependent formula. Pancreatic secretion SR is the sum of a static (SR_s) and a dynamic (SR_d) component (Eq. 3).

SR_s is described in Eqs. 4 and 5. The integral from time 0 to ∞ of SR_s can thus be calculated by integrating Eq. 5:

$$\int_0^\infty \dot{Y}(t)\mathrm{d}t = \int_0^\infty \mathrm{d}Y = Y(\infty) - Y(0) = -\alpha\int_0^\infty Y(t)\mathrm{d}t + \alpha\beta\int_0^\infty [G(t) - h]\mathrm{d}t \quad \text{(A1)}$$

Because $Y(\infty) = Y(0)$, i.e., the system returns for $t \to \infty$ to the basal steady state, the expression for

$$\int_0^\infty Y(t)\mathrm{d}t$$

and thus for

$$\int_0^\infty SR_s(t)\mathrm{d}t$$

becomes the following:

$$\int_0^\infty SR_s(t)\mathrm{d}t = \beta\int_0^\infty [G(t) - h]\mathrm{d}t = \Phi_s\int_0^\infty [G(t) - h]\mathrm{d}t \quad \text{(A2)}$$

SR_d is described in Eqs. 6 and 7. The integral from time 0 to ∞ of SR_d can thus be calculated by integrating Eq. 6:

$$\int_0^\infty SR_d(t)\mathrm{d}t = \begin{cases} \int_0^\infty k(G)\cdot\dot{G}(t)\mathrm{d}t & if\ \dot{G}(t) > 0 \\ 0 & if\ \dot{G}(t) \le 0 \end{cases} \quad \text{(A3)}$$

then

$$\int_0^\infty SR_d(t)\mathrm{d}t = \int_{G_b}^{G_{max}} k(G)\mathrm{d}G \quad \text{(A4)}$$

By using the definition of Φ_d (Eq. 9), the integral from time 0 to ∞ of SR_d can be expressed as follows:

$$\int_0^\infty SR_d(t)\mathrm{d}t = \Phi_d\cdot(G_{max} - G_b) \quad \text{(A5)}$$

It is thus possible to calculate the global index of β-cell sensitivity to glucose as follows:

$$\Phi = \frac{\int_0^\infty SR(t)\mathrm{d}t}{\int_0^\infty [G(t) - h]\mathrm{d}t} = \frac{\int_0^\infty SR_s(t)\mathrm{d}t}{\int_0^\infty [G(t) - h]\mathrm{d}t} + \frac{\int_0^\infty SR_d(t)\mathrm{d}t}{\int_0^\infty [G(t) - h]\mathrm{d}t} = \Phi_s + \frac{\Phi_d\cdot(G_{max} - G_b)}{\int_0^\infty [G(t) - h]\mathrm{d}t} \quad \text{(A6)}$$

Model independent formula. By integrating Eqs. 1 and 2 from 0 to ∞:

$$\int_0^\infty \dot{CP}_1(t)\mathrm{d}t = \int_0^\infty \mathrm{d}CP_1 = CP_1(\infty) - CP_1(0) = -[k_{01} + k_{21}]\int_0^\infty CP_1(t)\mathrm{d}t + k_{12}\int_0^\infty CP_2(t)\mathrm{d}t + \int_0^\infty SR(t)\mathrm{d}t \quad \text{(A7)}$$

$$\int_0^\infty \dot{CP}_2(t)\mathrm{d}t = \int_0^\infty \mathrm{d}CP_2 = CP_2(\infty) - CP_2(0) = k_{21}\int_0^\infty CP_1(t)\mathrm{d}t - k_{12}\int_0^\infty CP_2(t)\mathrm{d}t \quad \text{(A8)}$$

Because $CP_1(\infty) = CP_1(0)$ and $CP_2(\infty) = CP_2(0)$, i.e., the system returns for $t \to \infty$ to the basal steady state, the following holds:

$$(k_{01} + k_{21})\int_0^\infty CP_1(t)\mathrm{d}t = k_{12}\int_0^\infty CP_2(t)\mathrm{d}t + \int_0^\infty SR(t)\mathrm{d}t \quad \text{(A9)}$$

$$k_{12}\int_0^\infty CP_2(t)\mathrm{d}t = k_{21}\int_0^\infty CP_1(t)\mathrm{d}t \quad \text{(A10)}$$

By substituting Eq. A10 in Eq. A9, Eq. A9 becomes:

$$\int_0^\infty SR(t)\mathrm{d}t = k_{01}\int_0^\infty CP_1(t)\mathrm{d}t \quad \text{(A11)}$$

Finally, by substituting Eq. A11 in Eq. 10, Φ is given by:

$$\Phi = \frac{k_{01}\int_0^\infty CP_1(t)\mathrm{d}t}{\int_0^\infty [G(t) - h]\mathrm{d}t} \approx \frac{k_{01}\int_0^\infty CP_1(t)\mathrm{d}t}{\int_0^\infty [G(t) - G_b]\mathrm{d}t} \quad \text{(A12)}$$

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