A Model for Glucose Control of Insulin Secretion During 24 h of Free Living

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The aim of this work was to develop a mathematical model describing the functional dependence of insulin secretion on plasma glucose concentrations during 24 h of free living. We obtained hourly central venous blood samples from a group of healthy volunteers who spent 24 h in a calorimetric chamber, where they consumed standardized meals. Insulin secretory rates were reconstructed from plasma C-peptide concentrations by deconvolution. The relationship between insulin release and plasma glucose concentrations was modeled as the sum of three components: a static component (describing the dependence on plasma glucose concentration itself, with an embedded circadian oscillation), a dynamic component (modeling the dependence on glucose rate of change), and a residual component (including the fraction of insulin secretion not explained by glucose levels). The model fit of the individual 24-h secretion profiles was satisfactory (within the assigned experimental error of glucose and C-peptide concentrations). The static component yielded a dose-response function in which insulin release increased quasi-linearly (from 40 to 400 pmol/min on average) over the range of 4–9 mmol/l glucose. The dynamic component was significantly different from zero in coincidence with meal-related glucose excursions. The circadian oscillation and the residual component accounted for the day/night difference in the ability of glucose to stimulate insulin release. Over 24 h, total insulin release averaged 257 ± 58 nmol (or 43 ± 10 U). The static and dynamic component together accounted for ~80% of total insulin release. The model proposed here provides a detailed robust description of glucose-related insulin release during free-living conditions. In nondiabetic subjects, non–glucose-dependent insulin release is a small fraction of total insulin secretion. *Diabetes* 50 (Suppl. 1):S164–S168, 2001

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Quantitating insulin secretion is an important step in the study of disordered carbohydrate metabolism. A well-established approach is based on measurement of plasma C-peptide concentrations. C-peptide is secreted in equimolar amounts with insulin; both are transported to the liver, from which insulin is removed to a substantial (~50%) extent, whereas hepatic extraction of C-peptide is negligible. Thus, the systemic (venous) appearance of C-peptide equals insulin secretion. Because C-peptide kinetics are linear, systemic C-peptide appearance can be calculated by deconvolution from the measurement of plasma C-peptide concentrations and from a C-peptide kinetic model (1). The calculated systemic C-peptide appearance equals insulin secretion. Among other applications, this method has been successfully used to evaluate the 24-h profiles of insulin secretion in healthy subjects and obese patients (2).

The β-cell dose-response function, i.e., the relationship between glucose concentration and insulin secretion, has been derived exclusively from experiments using impulsive (3) or graded intravenous glucose infusions (4) over periods of a few hours. Under conditions of free living, the ingestion of mixed meals—with their contents of insulin secretagogues other than glucose (mostly amino acids [5]), the release of gastrointestinal peptides that potentiate glucose-induced insulin release (e.g., gastric inhibitory polypeptide [6]), and the accompanying neural activation (7)—give reason to expect that the β-cell glucose dose-response function should be different from that reconstructed from intravenous glucose administration. This, however, has not been determined. We therefore measured the 24-h profile of insulin secretion and developed a model to relate it to the concomitantly measured glucose concentrations. We report here the results obtained in a group of healthy subjects.

RESEARCH DESIGN AND METHODS

Experimental design. Seven healthy volunteers (four women, three men, age 35 ± 4 years [range 23–56], BMI 26 ± 0.4 kg/m^2 [25–28]) agreed to spend 24 h in a calorimetric chamber. During this period, four meals were administered for a total caloric intake of 30 kcal/kg of lean body mass (20% breakfast, 40% lunch, 10% afternoon snack, and 30% dinner). Diet composition was 17% protein, 35% fat, and 48% carbohydrate. In the afternoon, a 40-min session of bicycle exercise was performed at 40% of the individual maximal aerobic capacity. Hourly blood samples were drawn from a central venous catheter derived outside the chamber through long plastic tubing for the measurement of glucose, insulin, and C-peptide concentrations. The protocol was reviewed and approved by the Institutional Ethics Committee of the Catholic University of Rome.

Modeling. The model used is schematized in Fig. 1. The model consists of three subunits: a model for fitting the glucose concentration profile, a model describing the relationship between glucose concentration and insulin (or C-peptide) secretion, and a model of C-peptide kinetics.

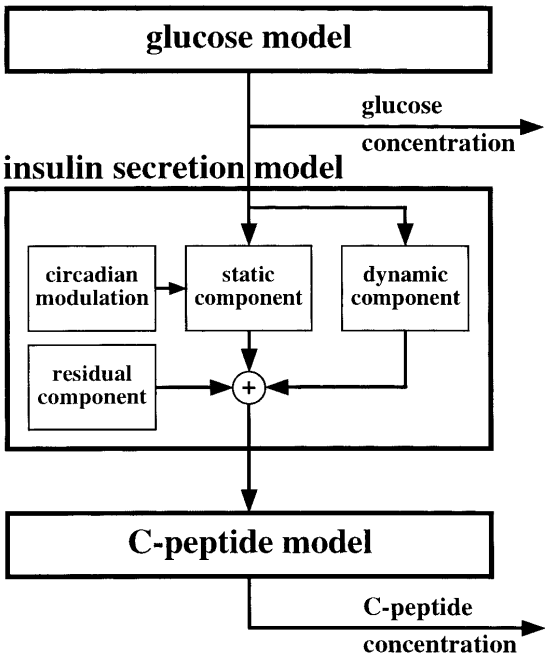

FIG. 1. Model used for the calculation of insulin secretion and β-cell glucose dose-response function.

The purpose of the glucose model is to smooth and interpolate glucose concentrations. It is described by the following differential equation:

$$\frac{dG(t)}{dt} = -kG(t) + R(t) \quad (1)$$

where $G(t)$ (expressed in millimoles per liter) is glucose concentration, k = 0.012 min^{-1} is an assigned constant, and $R(t)$ is a function of time, represented in discrete form as a piece-wise linear function over 20-min intervals. Equation 1 yields a glucose concentration profile continuous in time and its time derivative. Formally, Eq. 1 was a single-compartment model of glucose kinetics, although in this context, it is used only as a method for smoothing the glucose concentration.

In the insulin secretion model, insulin secretion [$S(t)$, in picomoles per minute] is represented as the sum of three components. The first component [$S_s(t)$] expresses a static relationship between insulin secretion and glucose concentration, i.e., it embodies a β-cell dose-response function, assumed to be modulated by a circadian rhythm, represented by means of a sinusoidal function with a 24-h period:

$$S_s(t) = p_1[\ln(1 + p_2 e^{G(t)}) - \ln(1 + p_2)] + p_3 + q(t) \quad (2a)$$

$$q(t) = p_4 \sin(t + p_5) \quad (2b)$$

where G (in millimoles per liter) is the glucose concentration, t is time (in hours), and p_1–p_5 are parameters. The term $q(t)$ represents the circadian modulation. When the modulating term $q(t)$ is zero, the dose-response function (Eq. 2a) is a curvilinear convex function. The parameter p_3 represents the intercept for $G = 0$, p_1 is the slope of the curve for high G values, and p_2 determines the curvature (i.e., for high [with respect to 1] values of p_2 the dose-response function is quasi-linear), whereas for low values of p_2, the dose-response curve exhibits a pronounced convexity. The parameters p_4 and p_5 are the amplitude and phase ($0 \le p_5 < 2\pi$) of the circadian oscillation, respectively.

The second insulin secretion component [$S_d(t)$] represents a dynamic dependence of insulin secretion on the rate of change of glucose concentration. $S_d(t)$ is proportional to the derivative of glucose concentration when the derivative is positive and is zero otherwise:

$$S_d(t) = \begin{cases} p_6 \dfrac{dG(t)}{dt}, & \dfrac{dG(t)}{dt} > 0 \\ 0, & \dfrac{dG(t)}{dt} \le 0 \end{cases} \quad (3)$$

The third insulin secretion component represents a residual secretion term [$S_r(t)$], which accounts for the possibility that the secretory controls represented in Eqs. 2a, 2b, and 3 are not exactly modeled or that other secretion components are present. $S_r(t)$ is modeled in discrete form as a generic piecewise linear function over 20-min intervals with zero mean. Because $S_r(t)$ may take on both positive and negative values, it represents an additive correction term rather than a real secretion component.

Total insulin secretion is the sum of the three components described above:

$$S(t) = S_s(t) + S_d(t) + S_r(t) \quad (4)$$

Total insulin secretion is calculated every 20 min for the whole 24-h period.

The model for C-peptide kinetics is the two-exponential model proposed by Van Cauter et al. (8), in which model parameters are determined in each individual on the basis of the subject's sex, weight, height, and age. C-peptide concentration [$C(t)$] is the convolution between the individualized two-exponential C-peptide impulse response $h(t)$ and C-peptide secretion given in Eq. 4:

$$C(t) = h(t) \otimes S(t) \quad (5)$$

where $\otimes$ denotes convolution.

The model resulting from the combination of Eqs. 1–5 embodies three differential equations (Eq. 1 for glucose and two differential equations for Eq. 5). The model predicts glucose and C-peptide concentration once the parameters p_1–p_6 and $R(t)$ (Eq. 1) and $S_r(t)$ (Eq. 4) are known. Conversely, p_1–p_6, $R(t)$, and $S_r(t)$ can be estimated using least-squares techniques from the glucose and C-peptide data. For this purpose, it is necessary to introduce regularization constraints on $R(t)$ and $S_r(t)$, as done in deconvolution schemes. The regularization method used adds penalty terms for $R(t)$ and $S_r(t)$ to the standard sum of squares term, which eliminate the spurious oscillations that $R(t)$ and $S_r(t)$ would otherwise exhibit. The function that the least-squares algorithm actually minimizes is as follows:

$$\sum w_G(t)^2[G(t) - \hat{G}(t)]^2 + \sum w_c(t)^2[C(t) - \hat{C}(t)]^2 + w_R^2 \sum [\hat{R}''(t)]^2 + w_S^2 \sum [S_r''(t)]^2 + w_0^2 \sum [S_r(t)]^2 \quad (6)$$

where the sums for t are over all the 24 time points, the hat denotes the model prediction, the quote sign represents the second derivative with respect to time, and the w represent weight. In Eq. 6, the first two terms are the standard weighted sums of squares for glucose and C-peptide, the third and fourth terms are the regularization terms based on the second derivatives of $R(t)$ and $S_r(t)$ (which are normally used in deconvolution algorithms), and the last term is the sum of $S_r(t)$ squared.

The weights of the first two standard least-squares terms, $w_G(t)$ and $w_C(t)$, were set to the inverse of the expected SD of the measurement error for glucose and C-peptide concentration. For glucose, the SD was assumed to be constant and equal to 2% of the mean glucose concentration for each individual experiment. For C-peptide, the measurement error was experimentally found to be concentration dependent and was estimated for each point with the formula: SD = 2.14 × C + 1,008, where SD and C are in picomoles per liter. The weights w_R and w_S determine the degree of smoothness of $R(t)$ and $S_r(t)$: high weights give smoother $R(t)$ and $S_r(t)$. The weight w_0 constrains $S_r(t)$ to be small in a least-square sense. These weights were iteratively selected so that the SD of glucose and C-peptide concentration calculated from the difference between the observed and the model-predicted values are close to the values expected from the measurement error. Model simulation and parameter estimation by minimization of Eq. 6 has been performed using the language of technical computing Matlab.

From the estimated model parameters, other parameters describing the β-cell dose-response characteristics are calculated. From Eq. 2a, by letting G = 5 and $q(t) = 0$, the insulin secretion value corresponding to a glucose concentration of 5 mmol/l (S_5, in picomoles per minute) is calculated. This parameter quantifies insulin secretion at or around normal fasting glucose levels. The slope of the dose-response function at 5 mmol/l glucose (ΔS_5, in picomoles per minute per millimole per liter) is also obtained from Eq. 2a. This parameter quantifies the sensitivity of β-cells to glucose concentration changes in the vicinity of 5 mmol/l. The time of the day (T_{max}) for maximal amplitude of the circadian rhythm is calculated from Eq. 2b. A measure of the average amplitude of the residual insulin secretion component (SD_r, in picomoles per minute) is obtained from its SD.

RESULTS

Figure 2 shows the observed daily profile of plasma glucose and C-peptide concentrations. It can be appreciated that the model fit is satisfactory. The model residuals, i.e., the differences between the measured and the model-predicted glucose and C-peptide concentrations, were not systematically different from zero at most of the time points. Small deviations

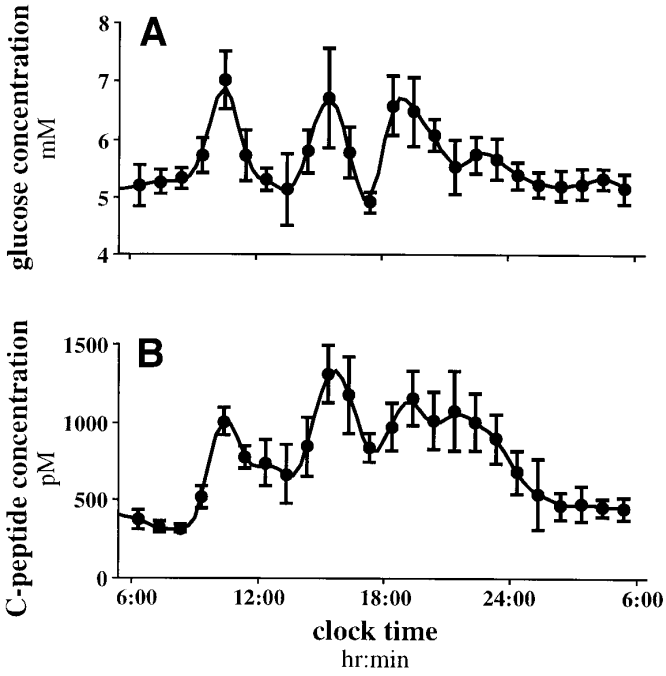

FIG. 2. Mean (± SE) plasma glucose (*A*) and C-peptide (*B*) concentrations over 24 h. The symbols represent the data, and the solid line represents the model fit.

from zero (0.1 mmol/l) were observed only for the largest glucose concentration swings.

Table 1 reports the parameters characterizing β-cell function in each study subject. The slope of the dose-response function (ΔS_5) varied less than threefold and was correlated with the parameter of the derivative component (p_6) (r = 0.86, $P < 0.02$). The amplitude of the circadian component (p_4) was correlated with the amplitude of the residual insulin secretion component (SD_r) ($r = 0.81$, $P < 0.05$).

Figure 3 shows the mean values of insulin secretion and its components throughout 24 h. The static component (Eq. 2), which also includes the circadian modulation, is the most important component. In fact, in an integral sense, the absolute value of the difference between total insulin secretion and the static component (the two lines shown in the top panel of Fig. 3) is <30% of total insulin secretion. The dynamic component accounts for ~9% of total insulin secretion. The values of the 24-h integral of total insulin secretion are given in Table 1: the group average was 257 nmol (43 U) with a less than twofold range.

Figure 4 shows the β-cell dose-response function in a typical subject (subject number 3). The dose-response function is plotted for a zero value of the circadian modulation. The circles depict the residual insulin secretion component $S_r(t)$, which is the vertical distance between the circles and dose-response line. The figure shows that the dose-response function is reasonably well identified despite uncertainty due to the residual insulin secretion component. The mean β-cell dose-response function is shown in Fig. 5.

DISCUSSION

The C-peptide deconvolution method was developed more than a decade ago to measure 24-h profiles of insulin secretion (2). Separate experiments have been necessary to explore the dependence of insulin secretion on plasma glucose concentrations (4). In this article, we have combined the C-peptide deconvolution methodology with a formal description of glucose control of insulin secretion. The obvious advantage of this approach is that the 24-h secretion profile and its functional relation to glucose concentration are determined in the same experiment.

Our model of insulin secretion is based on well-established concepts of β-cell function as well as experimental observations. Because of the complexity of the mechanisms governing β-cell function and the limitations imposed by experimental data, the model contains some simplifications. One assumption is that insulin secretion depends on plasma glucose concentration alone. Insulin secretion is known to respond also to secretagogues other than glucose, such as amino acids and free fatty acids (5). However, in the setting of free-living conditions, these secretagogues are expected to change in quasi-synchrony with the changes in glucose concentration, making differentiation of the effects difficult. Thus, the influence of other substrates is likely to be embed-

TABLE 1
Parameters of the insulin secretion model

	S_5 (pmol/min)	ΔS_5 (pmol · min^{-1} · mmol^{-1} · l^{-1})	p_2	p_4 (pmol/min)	T_{max} (clock time)	p_6 (pmol · mmol^{-1} · l^{-1})	SD_r (pmol/min)	TIS [nmol (U)]
Subject								
1	118	355	0.087	25	16:25	1,036	33	196 (33)
2	120	197	0.002	59	21:54	3	75	294 (49)
3	68	455	0.015	26	22:47	4,740	60	292 (49)
4	90	580	0.086	155	19:11	3,468	107	245 (41)
5	97	570	0.045	26	16:54	5,538	46	221 (37)
6	133	353	0.215	112	20:08	774	61	352 (59)
7	119	306	0.052	43	12:28	1,811	32	202 (34)
Mean	106	402	0.072	64	18:32	2,481	59	257 (43)
SD	23	140	0.071	51	3.6	2,122	26	58 (10)

The SD for T_{max} is in hours. ΔS_5, slope of the dose-response function at 5 mmol/l glucose concentration; p_2, parameter quantifying the curvature of the dose-response function (higher curvature = lower p_2 value); p_4, amplitude of the circadian modulation; p_6, parameter of the dynamic component; S_5, insulin secretion at 5 mmol/l glucose concentration; SD_r, standard deviation of the residual insulin secretion component; T_{max}, time of the day for maximum amplitude of the circadian modulation; TIS, 24-h integral of total insulin secretion.

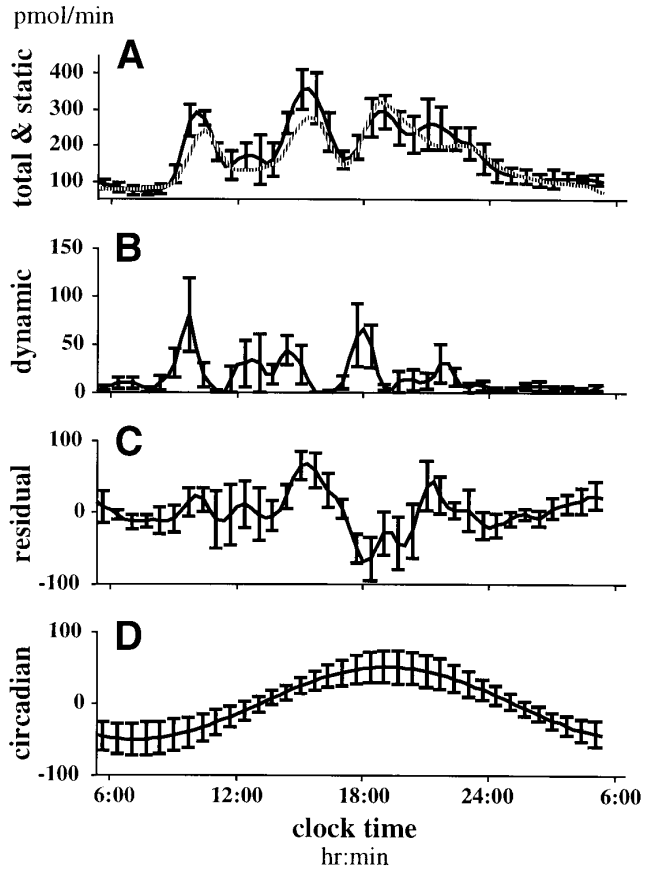

FIG. 3. Mean (± SE) insulin secretion and its components. All values are in picomoles per minute. *A*: Total insulin secretion (——) and its static component (·····; Eq. 2). *B*: Derivative component (Eq. 3). *C*: Residual component. *D*: Circadian modulation (Eq. 2b).

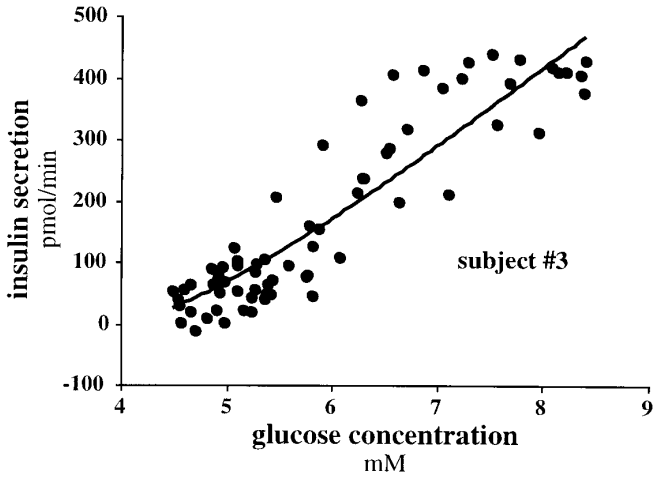

FIG. 4. β-Cell glucose dose-response function in subject number 3. The small circles depict the residual insulin secretion component $S_r(t)$. The vertical distance between the circles and the dose-response line is the value of $S_r(t)$.

ded in the relationship between glucose concentration and insulin secretion, although our results indicate that such influence is probably small (see below).

The static relationship between glucose concentration and insulin secretion we adopted (the dose-response function, Eq. 2a) is a mathematical function that can be linear or curvilinear depending on the parameters. The choice of a linear dose-response function is supported by the studies of Byrne et al. (4) in which graded glucose infusions were used to stimulate insulin secretion. Hovorka et al. (9) used a threshold-linear function in their meal test. In our study, a quasi-linear function was appropriate in most cases (Fig. 5), but in some subjects, a curvilinear function was necessary to fit the dose-response relationship, particularly at low glucose concentrations. The dose-response function obtained in our group of subjects (Fig. 5) was very similar to that obtained by Byrne et al. (4) in overnight fasted subjects receiving graded intravenous glucose infusions. Thus, insulin secretion increased tenfold quasi-linearly between 4 and 9 mmol/l glucose. This finding was somewhat unexpected considering the strong potentiation of insulin release provided by gastrointestinal factors. It is possible that oral glucose-induced enhancement of insulin release *1*) manifests itself as a derivative control (i.e., the dynamic component of insulin secretion formalized in our model), *2*) is hidden in an apparently circadian modulation, or *3*) is a transient phenomenon (more evident on a scale of minutes than hours). Only paired experiments, in which the oral glucose–induced glucose profile is mimicked by precise intravenous glucose infusions, can clarify whether and how gastrointestinal factors alter the glucose-insulin dose-response function over a 24-h period. Nevertheless, the current results do suggest that oral administration of glucose does not result in a major increase in β-cell response to glucose per se in comparison with intravenous glucose.

The dynamic dependence of insulin secretion on the rate of change of glucose concentration has been established in tests in which glucose concentration was abruptly increased (3,10). During an oral glucose or mixed meal test, this secretory component is definitely less evident because the changes in glucose concentration are slower. In the meal test model by Hovorka et al. (9), this component was not featured. Our model includes the dynamic component $S_d(t)$. Although its contribution to total insulin secretion is small (Fig. 3), we found that the coefficient expressing the dependence of insulin secretion on the derivative of glucose concentration (p_6) is correlated with the slope of the dose-response function (ΔS_5), suggesting that a dynamic component reflecting the sensitivity of the β-cell to glucose concentration changes is indeed present.

The addition of a circadian modulation of insulin secretion [$q(t)$, Eq. 2b] was essential to predict C-peptide concentrations adequately. Without $q(t)$, insulin secretion would have been significantly overestimated during the night compared with the diurnal period. The modulation was described as a vertical time-dependent shift of the dose-response function during the 24 h, following a sinusoidal pattern. Indeed, this variation is not necessarily a true circadian rhythm but can reflect other phenomena, e.g., daytime potentiation of insulin secretion due to exposure to meal-related hyperglycemia. Such a potentiation is in fact well documented in some experimental conditions (10). Circadian rhythms in insulin secretion have also been reported (11,12), but differences in the experimental protocols make direct comparison with the present findings difficult.

The functional mechanisms of insulin secretion discussed above were still not sufficient to explain the observed variations

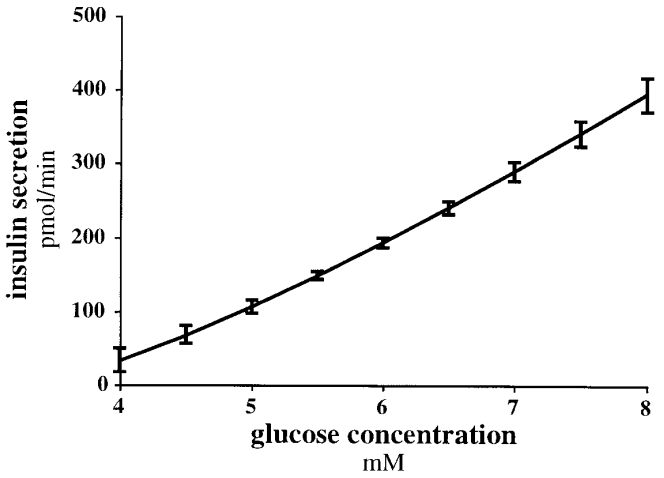

FIG. 5. Mean (± SE) β-cell glucose dose-response function of the whole group. Error bars are shown at selected plasma glucose concentrations.

in C-peptide concentration. For this reason, we included a residual secretion component [$S_r(t)$], with which C-peptide concentration could be predicted within the expected measurement error. $S_r(t)$ was not constrained to a specific functional form but was allowed to take on an arbitrary smooth zero-mean time course. The residual component was quite variable from subject to subject. On average, $S_r(t)$ was most significant between 1400 and 2200, whereas during the remaining period of time, its value was small. The peaks observed at 1530 and 2130, i.e., in concomitance with lunch and dinner, may reflect potentiation of the insulin response by previous exposure to hyperglycemia (10), whereas the negative values between these two peaks might be due to an inhibitory effect of the exercise bout that was performed by all subjects at 1730 (13). The existence of a potentiation phenomenon underlying both these variations in insulin secretion and the 24-h oscillation may explain the observed correlation between the amplitude of the oscillation and the SD of the residual component.

The residual secretion component adds some uncertainty to the determination of the dose-response function and the dynamic secretory component. Figure 4, which represents an average case and where the scatter around the dose-response function is due to the residual component, shows that despite $S_r(t)$, the dose-response function is identified with sufficient accuracy. As for the dynamic secretion component $S_d(t)$, it should be noted that the actual response to glucose concentration changes is different from that predicted by the dynamic component alone because total secretion is the sum of the static component and both $S_d(t)$ and $S_r(t)$. For instance, the peak of the dynamic response predicted at 1700 (Fig. 3) is virtually abolished by the negative peak of $S_r(t)$. However, even when $S_d(t)$ and $S_r(t)$ are considered together, an increase in secretion corresponding to the time periods during which the derivative of glucose concentration is positive is observed (except at 1700). This indicates that the incorporation of a dynamic secretion component in the model is appropriate. Furthermore, the correlation observed between the parameter of the dynamic component (p_6, Eq. 3) and the slope of the dose-response function (ΔS_5) suggests that both reflect an aspect of β-cell function. With regard to total insulin secretion, our approach is very similar to that used by Polonsky et al. (2), with the only differences being that our sampling schedule was less frequent and that C-peptide kinetics were not determined from individual bolus injections but from anthropometric data according to a validated approach (8,14). In this respect, our results essentially reproduce the previous findings (2).

In summary, the model for glucose control of insulin secretion we present in this work yields an accurate profile of insulin secretion, the dose-response function for glucose-induced insulin secretion, and additional fine-tuning parameters of β-cell function. Phenomena such as non–glucose-dependent insulin release, potentiation, and memory, which have been predicted on the basis of short-term experiments using intravenous glucose, have proven difficult to model when dealing with 24-h free-living conditions; however, they are likely to be embedded in the features of the model. Whereas the main quantitative results in our healthy volunteers are fully consistent with the available literature, only the application of the model to different pathophysiological conditions can test its ability to provide new insight into the mechanisms of insulin release.

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REFERENCES

1. Eaton RP, Allen RC, Schade DS, Erickson KM, Standefer J: Prehepatic insulin production in man: kinetic analysis using peripheral connecting peptide behavior. *J Clin Endocrinol Metab* 51:520–528, 1980
2. Polonsky KS, Given BD, Van Cauter E: Twenty-four-hour profiles and pulsatile patterns of insulin secretion in normal and obese subjects. *J Clin Invest* 81:442–448, 1988
3. Ferrannini E, Pilo A: Pattern of insulin delivery after intravenous glucose injection, and its relation to plasma glucose disappearance. *J Clin Invest* 64:243–251, 1979
4. Byrne MM, Sturis J, Polonsky KS: Insulin secretion and clearance during low-dose graded glucose infusion. *Am J Physiol* 268:E21–E27, 1995
5. Floyd JC Jr, Fajans SS, Conn JW, Knopf RF, Rull J: Stimulation of insulin secretion by amino acids. *J Clin Invest* 45:1487–1502, 1966
6. Dupre J, Cutris JD, Unger RH, Waddell RW, Beck JC: Effect of secretin, pancreozymin, or gastrin on the response of the endocrine pancreas to administration of glucose or arginine in man. *J Clin Invest* 48:745–757, 1969
7. Robertson RP, Halter JB, Porte D Jr: A role for alpha-adrenergic receptors in abnormal insulin secretion in diabetes mellitus. *J Clin Invest* 57:791–795, 1976
8. Van Cauter E, Mestrez F, Sturis J, Polonsky KS: Estimation of insulin secretion rates from C-peptide levels: comparison of individual and standard kinetic parameters for C-peptide clearance. *Diabetes* 41:368–377, 1992
9. Hovorka R, Chassin L, Luzio SD, Playle R, Owens DR: Pancreatic β-cell responsiveness during meal tolerance test: model assessment in normal subjects and subjects with newly diagnosed noninsulin-dependent diabetes mellitus. *J Clin Endocrinol Metab* 83:744–750, 1998
10. Cerasi E: Differential actions of glucose on insulin release: re-evaluation of a mathematical model. In *Carbohydrate Metabolism: Quantitative Physiology and Mathematical Modelling*. Cobelli C, Bergman RN, Eds. Chichester, U.K., Wiley, 1981, p. 3–22
11. Van Cauter E, Blackman JD, Roland D, Spire JP, Refetoff S, Polonsky KS: Modulation of glucose regulation and insulin secretion by circadian rhythmicity and sleep. *J Clin Invest* 88:934–942, 1991
12. Van Cauter E, Shapiro ET, Tillil H, Polonsky KS: Circadian modulation of glucose and insulin responses to meals: relationship to cortisol rhythm. *Am J Physiol* 262:E467–E475, 1992
13. Levitt NS, Hirsch L, Rubenstein AH, Polonsky KS: Quantitative evaluation of the effect of low-intensity exercise on insulin secretion in man. *Metabolism* 42:829–833, 1993
14. Hovorka R, Koukkou E, Southerden D, Powrie JK, Young MA: Measuring pre-hepatic insulin secretion using a population model of C-peptide kinetics: accuracy and required sampling schedule. *Diabetologia* 41:548–554, 1998