A reliable serum C-peptide index for the selection of an insulin regimen to achieve good glycemic control in obese patients with type 2 diabetes: an analysis from a short-term study with intensive insulin therapy

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Abstract Insulin regimens achieving favorable glycemic control in patients with type 2 diabetes are expected to closely relate to residual insulin secretory ability. We herein attempted to identify a reliable C-peptide immunoreactivity (CPR) index as an insulin secretory marker that would contribute to the selection of an appropriate insulin regimen for patients with type 2 diabetes. We near-normalized blood glucose in 246 obese patients with type 2 diabetes using our protocol (which included short-term intensive insulin therapy, IIT), and administered an oral hypoglycemic agent (OHA). Based on responsiveness to OHA, patients were classified into three therapy groups: non-insulin therapy ($n = 78$), basal-insulin supported oral therapy (BOT) ($n = 109$), and multiple daily insulin injection (MDI) therapy ($n = 59$). Glucagon-loading CPR increment (Δ CPR), fasting CPR (FCPR), CPR2h after breakfast (CPR2h), ratio of FCPR to fasting plasma glucose (CPI), CPI2h after breakfast (CPI2h), and secretory unit of islets in transplantation (SUIT) were assessed with receiver operating characteristic (ROC) and multiple logistic analyses to discriminate the MDI group from the other therapy groups. ROC analysis revealed that CPR2h had the greatest area under the curve and specificity. Multiple logistic analysis identified CPR2h and CPI2h as the most significant explanatory variables for

identifying patients assigned to the MDI group. A postprandial serum CPR marker such as CPR2h or CPI2h was shown to be the best index for predicting an appropriate insulin regimen to achieve good glycemic control in obese patients with type 2 diabetes.

Keywords C-peptide · Insulin regimen · Obesity · Type 2 diabetes

Introduction

Glucose-lowering therapies with oral hypoglycemic agents (OHA) and insulin for type 2 diabetes have become increasingly advanced, although these therapies have also become more complex [1, 2]. Type 2 diabetes is a progressive disease characterized by an annual decline in insulin secretion with the duration of diabetes [3]; therefore, insulin regimens that achieve good glycemic control in patients with type 2 diabetes are expected to be closely related to residual insulin secretory ability. However, the relationship between insulin secretion and insulin regimen has not yet been investigated. We recently described [4] the relationship between insulin regimen and insulin secretory ability, and showed that postprandial serum C-peptide immunoreactivity (CPR) indices were the most useful indices for selecting insulin regimens to achieve favorable blood glucose levels in non-obese patients with type 2 diabetes. The pathophysiological characteristics of obese patients differ from those of non-obese patients [3], and the CPR index that best aids the selection of insulin regimens in obese patients with type 2 diabetes has not yet been identified, so further studies are warranted. In order to determine a reliable CPR index that can be used as an insulin secretory marker for the selection of an appropriate

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insulin regimen in obese patients with type 2 diabetes, we performed a retrospective analysis and evaluated serum CPR values in patients with body mass index (BMI) values of ≥ 25 using a treatment protocol that included short-term intensive insulin therapy (IIT).

Materials and methods

Patients

We initially extracted 1039 patients with type 2 diabetes who had been hospitalized and treated with insulin for poor glycemic control over a 36-month period between October 2007 and September 2010 from our departmental diabetes database. A total of 470 of these patients were excluded because of inappropriate medical conditions, which left 569 patients. Of these, 246 obese (BMI ≥ 25) patients with type 2 diabetes were enrolled in the present study (the exclusion criteria are presented in Fig. S1 of the Electronic supplementary material, ESM). Table 1 shows the baseline clinical characteristics and types of diabetes treatments of the patients.

Treatment protocol

The treatment involved a 2-week protocol, as described previously [4]. On day 1, a standard diabetes meal, 30 kcal/kg of standard body weight: $22 \times \text{body height (m)}^2$, consisting of 62 % carbohydrate, 17 % protein, and 21 % fat, was initiated. Patient self-measurements of capillary blood glucose using a portable blood glucose monitor four times daily (before each meal and at bedtime) were also started. On day 2, plasma glucose (PG, mg/dl) concentrations were measured at seven points (before and 2 h after three meals, and at bedtime) under the baseline treatment. A glucagon loading test was performed in the morning on day 3, and IIT was subsequently started as follows. Insulin injections were administered to all patients before each meal with 4 U of regular or ultra-short-acting analog insulin, and at 10 pm with 4 U of intermediate-acting insulin (NPH), insulin glargine, or insulin detemir. The insulin dose was adjusted daily to attain a target glucose value of 110 mg/dl before the three meals for 7 days. Insulin was discontinued on days 8–9, and OHA was started with 40–80 mg gliclazide, 1–2 mg glimepiride, 270 mg nateglinide, or 30 mg mitiglinide in combination with 150–300 mg acarbose, 0.9 mg voglibose, or 150 mg miglitrol (daily doses of each). These medicines were administered for 3 days and the OHA treatment was continued if glucose values were equal to the values obtained with IIT. If glucose was ≥ 140 mg/dl before breakfast, the original bedtime insulin (same type and dose as administered at 10 pm) was added. If glucose

Table 1 Baseline clinical characteristics of patients enrolled in the study ($n = 246$)

Male/female	130/116
Age (years old)	58.8 ± 13.5
Duration of diabetes (years)	10.2 ± 8.9
BMI (kg/m^2)	29.6 ± 5.1
FPG (mg/dl)	184 ± 56
HbA1c (% NGSP)	9.8 ± 1.7
ΔCPR (ng/ml)	2.44 ± 1.41
FCPR (ng/ml)	2.07 ± 1.31
CPR2h (ng/ml)	4.88 ± 2.84
CPI (ng/mg)	1.22 ± 0.93
CPI2h (ng/ml)	1.99 ± 1.51
SUIT (%)	32.2 ± 32.0
Diabetes treatment, n (%)	
Diet only	47 (19.1)
OHA alone	126 (51.2)
(Sulfonylurea)	84 (34.1)
(Metformin)	48 (19.5)
(Pioglitazone)	28 (11.4)
Insulin	73 (29.7)
(with OHA)	32 (13.0)
Complicated diseases, n (%)	
Peripheral neuropathy	130 (52.8)
Diabetic retinopathy	118 (48.0)
Diabetic renal disease	87 (35.4)
Ischemic heart disease	51 (20.7)
Cerebral vascular disease	39 (15.9)
Arteriosclerotic disease of the legs	21 (8.5)

Data shown are mean $\pm$ SD; number of patients and percentages in parentheses

ΔCPR C-peptide immunoreactivity increment during glucagon test, BMI body mass index, CPI C-peptide index, CPI2h C-peptide index 2 h after breakfast, CPR2h C-peptide immunoreactivity 2 h after breakfast, FCPR fasting C-peptide, FPG fasting plasma glucose, HbA1c glycated hemoglobin, NGSP National Glycohemoglobin Standardization Program, OHA oral hypoglycemic agent, SUIT secretory unit of islets in transplantation

was ≥ 140 mg/dl both before breakfast and before dinner, all OHAs were discontinued, and a biphasic analog-mixed insulin (at 80 % of the total daily insulin dose prior to switching, split in a 2:1 ratio) was started before breakfast and before dinner. The four-times-daily insulin regimen was resumed when the blood glucose levels that reached IIT before switching to OHA were not maintained in spite of twice-daily injections. According to this protocol, the final treatment type was divided into three groups: OHA alone, basal insulin plus OHA (basal-insulin supported oral therapy, BOT), and insulin two to four times daily (multiple daily insulin injection, MDI). Details of the oral agents and insulin used in each therapy group at the final stage are presented in Table S1 of the ESM.

PG and CPR sampling

PG excursion was measured on day 2, as were fasting CPR (FCPR) (ng/ml) before breakfast and CPR2h after starting the meal (CPR2h). Intravenous 1-mg glucagon loading was performed before breakfast on day 3, and CPR was measured at 0 and 6 min. PG excursion was measured again after IIT.

The CPR indices submitted for analysis included the glucagon loading CPR increment (Δ CPR), FCPR, CPR2h, the ratio of FCPR to fasting PG (FPG) and the C-peptide index (CPI, $\text{FCPR}/\text{FPG} \times 100$, ng/mg), CPI2h after breakfast (CPI2h), and the secretory unit of islets in transplantation (SUIT, %), which was calculated by the formula $1500 \times \text{FCPR}/(\text{FPG} - 61.7)$ [5].

CPR was measured by a radioimmunoassay (RIA) using the “Daiichi” III C-peptide kit (TFB Inc., Tokyo, Japan). PG values were measured in venous blood by the hexokinase method.

HbA1c values were converted from the Japan Diabetes Society to National Glycohemoglobin Standardization Program values using a conversion equation [6].

Statistical analysis

Clinical markers, the period from the diagnosis of diabetes (duration of diabetes), BMI, FPG, HbA1c, and individual CPR values were analyzed and compared among the OHA alone, BOT, and MDI groups using the Steel–Dwass test. Receiver operating characteristic (ROC) analysis of each CPR index and the duration of diabetes was performed to determine the requirement for MDI therapy. Comparisons of the AUC of the CPR index to that of CPR2h were performed by the method of Hanley [7]. Multiple logistic analysis was performed to select CPR parameters to use as significant explanatory variables for discriminating patients who required MDI from those who responded to the OHA therapy. The Mann–Whitney *U* test was carried out to identify significant differences between the OHA (the OHA-alone and BOT groups combined) and MDI groups. The statistical software used for analyses included Excel Statistics 2012 for Windows ver. 1.13 (Social Survey Research Information Co. Ltd., Tokyo, Japan) and IBM SPSS Statistics ver. 20 (IBM Japan, Tokyo, Japan).

Results

Daily plasma glucose excursions before and after IIT

Under baseline treatment conditions before starting IIT, the daily PG excursion values at the seven points were 184 ± 56 , 275 ± 86 , 240 ± 89 , 242 ± 95 , 183 ± 78 ,

240 ± 83 , and 224 ± 82 mg/dl (mean $\pm$ SD) in all subjects. Under the final treatment conditions after IIT, these values were 118 ± 22 , 164 ± 48 , 140 ± 41 , 159 ± 47 , 127 ± 32 , 169 ± 44 , and 163 ± 38 mg/dl (mean $\pm$ SD). Significant decreases were observed at all points ($p < 0.01$). The percentages of patients with PG that attained the target value before the 3 meals (110 mg/dl) were 39, 25, and 33 %, respectively.

Clinical characteristics and CPR levels in the three treatment groups

Of the 246 patients who participated, the number in each final therapy group was: OHA alone, 78; BOT, 109; and MDI, 59. Details of the oral agents or insulin used in the three groups are shown in Table S1 of the ESM.

Table 2 shows the baseline clinical characteristics of the three groups, including the following six CPR indices: Δ CPR, FCPR, CPR2h, CPI, CPI2h, and SUIT. The Steel–Dwass test revealed that the duration of diabetes was longer in the MDI group than in the OHA-alone and BOT groups ($p < 0.01$). An analysis of CPR values revealed no significant differences between the OHA-alone and BOT groups except for CPI, CPI2h, and SUIT, while all CPR values were significantly lower in the MDI group than in the OHA-alone and BOT groups ($p < 0.01$; $p < 0.05$, BOT vs MDI group in SUIT). Although significant differences were observed in three of the six CPR values between the OHA-alone and BOT groups, both groups were responsive to OHAs; therefore, they were combined into the “OHA group,” as we did in our previous study [4]. As shown in Table 3, a comparison of the clinical characteristics and CPR values between the OHA and MDI groups revealed that the duration of diabetes was longer and all of the CPR levels were lower in the MDI group than in the OHA group ($p < 0.01$). The average of the capillary blood glucose results obtained at the four diurnal time points based on patient self-measurements in each therapy group was 128 ± 20 (OHA alone), 132 ± 21 (BOT), and 137 ± 27 mg/dl (MDI) 1 week after starting IIT, respectively (mean $\pm$ SD; not significant among the 3 groups). The daily insulin dose in each group was 29.4 ± 16.4 , 48.6 ± 22.9 , and 48.4 ± 19.1 U/day 1 week after starting IIT, respectively (mean $\pm$ SD; $*p < 0.01$ vs OHA alone). The final insulin dose was 18.8 ± 13.2 in the BOT group and 37.1 ± 19.4 U/day in the MDI group (mean $\pm$ SD; $p < 0.01$) (Table S4 in the ESM).

ROC analysis of CPR indices to determine MDI

In the ROC analysis of the duration of diabetes and six CPR indices using baseline data, AUC and specificity were as follows: duration of diabetes, 0.772 and 82.9 % (cutoff,

Table 2 Baseline clinical profiles and C-peptide immunoreactivity levels in the three final therapy groups

Indices	OHA alone (<i>n</i> = 78)	BOT (<i>n</i> = 109)	MDI (<i>n</i> = 59)
Male/female	37/41	60/49	33/26
Age (years old)	57.5 ± 14.4	57.6 ± 13.2	62.7 ± 12.1*†
Duration of diabetes (years)	6.3 ± 5.6	9.1 ± 7.6	17.2 ± 10.5***††
BMI (kg/m ²)	29.8 ± 5.5	30.1 ± 5.7	28.2 ± 2.7
FPG (mg/dl)	158 ± 49	203 ± 55**	182 ± 53*†
HbA1c (% NGSP)	9.3 ± 1.8	10.1 ± 1.7**	9.8 ± 1.6
ΔCPR (ng/ml)	2.85 ± 1.50	2.52 ± 1.36	1.74 ± 1.08***††
FCPR (ng/ml)	2.29 ± 1.45	2.28 ± 1.28	1.38 ± 0.89***††
CPR2h (ng/ml)	5.88 ± 3.25	5.14 ± 2.43	3.08 ± 2.00***††
CPI (ng/mg)	1.61 ± 1.25	1.18 ± 0.71*	0.79 ± 0.51***††
CPI2h (ng/mg)	2.79 ± 2.00	1.83 ± 1.07**	1.24 ± 0.82***††
SUIT (%)	47.7 ± 45.7	27.9 ± 21.3**	19.7 ± 13.0***†
Prior diabetes therapy, <i>n</i> (%)			
Diet only	20 (25.6)	23 (21.1)	4 (6.8)
OHA alone	54 (69.2)	58 (53.2)	14 (23.7)
(Sulfonylurea)	31 (39.7)	41 (37.6)	12 (20.3)
(Metformin)	17 (21.8)	24 (22.0)	7 (11.9)
(Pioglitazone)	9 (11.5)	15 (13.8)	4 (6.8)
Insulin	4 (5.1)	28 (25.7)	41 (69.5)
(with OHA)	0 (0.0)	18 (16.5)	14 (23.7)

Baseline clinical profiles, C-peptide immunoreactivity (CPR) levels, and prior diabetes therapies of patients in each final therapy group are shown. Data are presented as mean ± SD; number of patients and the percentages in parentheses. Statistical analysis was carried out by the Steel–Dwass test

ΔCPR C-peptide immunoreactivity increment during glucagon test, BMI body mass index, CPI C-peptide index, CPI2h C-peptide index 2 h after breakfast, CPR2h C-peptide immunoreactivity 2 h after breakfast, FCPR fasting C-peptide, FPG fasting plasma glucose, HbA1c glycated hemoglobin, MDI multiple daily insulin injection, SUIT secretory unit of islets in transplantation

* $p < 0.05$, ** $p < 0.01$ vs oral hypoglycemic (OHA) alone

† $p < 0.05$, †† $p < 0.01$ vs basal insulin-supported oral therapy (BOT)

15 years); ΔCPR, 0.707 and 66.3 % (1.9 ng/ml); FCPR, 0.740 and 77.5 % (1.3 ng/ml); CPR2h, 0.782 and 80.7 % (3.2 ng/ml); CPI, 0.720 and 66.8 % (0.8 ng/mg); CPI2h, 0.741 and 65.2 % (1.4 ng/mg); SUIT, 0.691 and 65.2 % (20 %), respectively. AUC, sensitivity, and specificity were greater in CPR2h than in the other CPR indices (Table 4 and Fig. S2 in the ESM). A comparison of the AUC of each CPR index to the largest AUC of CPR2h revealed that the AUCs of ΔCPR, CPI, and SUIT were smaller than ($p < 0.05$ or $p < 0.01$) and the AUCs of FCPR and CPI2h were identical to that of CPR2h (Table 4).

Multiple logistic analysis of CPR indices for determining MDI

Multiple logistic analysis of six CPR indices was performed to determine their contributions to the requirement for MDI therapy. To avoid multicollinearities among CPR indices, each CPR was separately analyzed together with common clinical markers: the duration of diabetes, BMI, and HbA1c (refer to Table S2 in the ESM). Of the six CPR

parameters, FCPR, CPR2h, CPI, and CPI2h were selected as significant explanatory variables; however, the standardized partial regression coefficient was advantageous for CPR2h and CPI2h (Table 5).

Discussion

In the present study, the CPR levels of patients in the MDI therapy group were significantly lower than those of the patients in the other therapy groups. Furthermore, post-prandial indices were found to be the most reliable of the 6 CPR indices tested to select the most appropriate insulin regimen (MDI or either of the other therapies) for obese patients with type 2 diabetes. These results were based on a short-term study including IIT and challenging patients with OHA after the near-normalization of hyperglycemia. Since SU and glinide are rapidly effective oral agents [1] including α-glucosidase inhibitors, these were adaptable to the short-term study design; therefore, the list of medicines that could be used in our protocol did not include

Table 3 Comparison of clinical markers and C-peptide immunoreactivity levels between the oral hypoglycemic agent group (OHA group; oral hypoglycemic agent alone + basal insulin-supported oral therapy) and multiple daily insulin injection group (MDI group)

Indices	OHA group (OHA alone + BOT) (<i>n</i> = 187)	MDI group (<i>n</i> = 59)	Mann–Whitney <i>U</i> test: <i>p</i> value
Male/female	97/90	33/26	n.s. ^a
Age (years old)	57.6 ± 13.7	62.7 ± 12.1	<0.01
Duration of diabetes (years)	7.9 ± 7.0	17.2 ± 10.5	<0.01
BMI (kg/m ²)	30.0 ± 5.6	28.2 ± 2.7	n.s.
FPG (mg/dl)	184 ± 57	182 ± 53	n.s.
HbA1c (%)	9.8 ± 1.7	9.8 ± 1.6	n.s.
CPR levels at baseline			
ΔCPR (ng/ml)	2.66 ± 1.43	1.74 ± 1.08	<0.01
FCPR (ng/ml)	2.29 ± 1.35	1.38 ± 0.89	<0.01
CPR2h (ng/ml)	5.45 ± 2.82	3.08 ± 2.00	<0.01
CPI (ng/mg)	1.36 ± 0.99	0.79 ± 0.51	<0.01
CPI2h (ng/ml)	2.23 ± 1.60	1.24 ± 0.82	<0.01
SUIT (%)	36.1 ± 35.0	19.7 ± 13.0	<0.01

C-peptide immunoreactivity (CPR) levels and other clinical markers at baseline in the oral hypoglycemic agent (OHA) alone + basal insulin-supported oral therapy (BOT) group (the “OHA group”) and the multiple daily insulin injection group (“MDI group”) are shown as mean ± SD, respectively

ΔCPR increment of C-peptide immunoreactivity during glucagon test, *BMI* body mass index, *CPI* C-peptide index, *CPI2h* C-peptide index 2 h after breakfast, *CPR2h* C-peptide immunoreactivity 2 h after breakfast, *FCPR* fasting C-peptide, *n.s.* not significant, *SUIT* secretory unit of islets in transplantation

^a Chi-square test

Table 4 Receiver operating characteristic analysis of duration of diabetes and C-peptide immunoreactivity indices for identifying patients who require multiple daily insulin injection therapy

Indice	AUC (95 % CI)	<i>p</i> value	Cutoff	Sensitivity (%)	Specificity (%)	<i>p</i> value vs CPR2h
Duration of diabetes	0.772 (0.696–0.848)	<0.001	15 (years)	57.1	82.9	n.d.
ΔCPR	0.707 (0.630–0.784)	<0.001	1.9 (ng/ml)	59.3	66.3	<0.05
FCPR	0.740 (0.664–0.816)	<0.001	1.3 (ng/ml)	66.1	77.5	n.s.
CPR2h	0.782 (0.714–0.851)	<0.001	3.2 (ng/ml)	71.2	80.7	–
CPI	0.720 (0.645–0.795)	<0.001	0.8 (ng/mg)	66.1	66.8	<0.05
CPI2h	0.741 (0.671–0.811)	<0.001	1.4 (ng/mg)	66.1	65.2	n.s.
SUIT	0.691 (0.617–0.766)	<0.001	20 (%)	66.1	65.2	<0.01

Results of receiver operating characteristic (ROC) analysis using baseline data are shown (refer to Fig. S2 of the ESM). Comparisons of the AUC to that of CPR2h were performed by the method of Hanley [7]

ΔCPR C-peptide immunoreactivity increment during glucagon test, *AUC* area under the ROC curve, *CPI* C-peptide index, *CPI2h* C-peptide index 2 h after breakfast, *CPR2h* C-peptide immunoreactivity 2 h after breakfast, *FCPR* fasting C-peptide immunoreactivity, *SUIT* secretory unit of islets in transplantation, *n.d.* not determined, *n.s.* not significant

metformin or thiazolidine in spite of their usefulness in obese patients with type 2 diabetes (they are not fast-acting). Accordingly, the patients’ responsiveness to insulin

secretagogues mainly contributed to the results obtained in the present study. The averages of the final PG values before the 3 meals were 118, 140, and 127 mg/dl, which

Table 5 Summary of multiple logistic analysis of C-peptide immunoreactivity indices for identifying patients who require multiple daily insulin injection therapy

Parameter	Partial regression coefficient (95 % CI)	Standardized partial regression coefficient	<i>p</i> value	Odds ratio (95 % CI)
ΔCPR	−0.3216 (−0.6670 to 0.0239)	−0.4574	n.s.	0.7250 (0.5132–1.0242)
FCPR	−0.4887 (−0.9090 to −0.0684)	−0.6346	<0.05	0.6134 (0.4029–0.9339)
CPR2h	−0.3716 (−0.5778 to −0.1654)	−1.0684	<0.01	0.6896 (0.5611–0.8476)
CPI	−0.7339 (−1.4371 to −0.0306)	−0.6847	<0.05	0.4801 (0.2376–0.9698)
CPI2h	−0.7911 (−1.3136 to −0.2686)	−1.1903	<0.01	0.4534 (0.2688–0.7645)
SUIT	−0.0247 (−0.0507 to −0.0013)	−0.7900	n.s.	0.9756 (0.9506–1.0013)

A summary of the multiple logistic analysis of C-peptide immunoreactivity parameters (see Table S2 in the ESM) is shown

ΔCPR C-peptide immunoreactivity increment during glucagon test, *CI* confidence interval, *CPI* C-peptide index, *CPI2h* CPI2h after breakfast, *CPR2h* C-peptide immunoreactivity 2 h after breakfast, *FCPR* fasting CPR, *n.s.* not significant, *SUIT* secretory unit of islets in transplantation

were slightly higher than the preset target (110 mg/dl), and the percentages of patients with PG that attained the target value before the 3 meals were 39, 25, and 33 %, respectively. These results suggest that the glucose toxicity caused by baseline hyperglycemia was not suitably cleared, despite IIT. However, when considering inpatient PG goals in the American Diabetes Association's standards [8] (premeal target <140 mg/dl), and in the studies reported by Funakoshi et al. [9] (premeal PG was approximately 130 mg/dl on discharge) and Chen et al. [10] (mean FPG was approximately 130 mg/dl on discharge), the final PG levels of our patients before meals were within a reasonable range.

The present study revealed that four out of the six CPR indices examined using multiple logistic analysis were significant explanatory variables. Of these four parameters, the postprandial CPR indices CPR2h and CPI2h were found to have advantageous standard partial regression coefficients for identifying patients who required MDI. The ROC analysis supported these results. The postprandial serum CPR marker is reported to be a valuable index for predicting insulin therapy [11]. Our results were consistent with this finding and were identical to those obtained in a previous study [4] of non-obese patients with type 2 diabetes. Two studies [9, 12] showed that the fasting CPR indices CPI and SUIT were useful predictors of the requirement for insulin in patients with type 2 diabetes. We also identified FCPR and CPI as significant indices of the need for insulin therapy; however, postprandial indices were more reliable than these fasting ones. Accordingly, aside from the BMI, the postprandial CPR marker was a common and essential index for selecting which of the

MDI or other insulin regimens was most appropriate for patients with type 2 diabetes. Nevertheless, the cutoff values of the CPR indices were slightly higher, by 0.2–0.4, in obese patients than in non-obese ones (Table S3 in the ESM). These higher cutoff values may reflect higher baseline serum CPR levels in obese patients than in non-obese patients, in spite of the presence of identical baseline FPG levels among the patients enrolled in the present and previous [4] studies (184 ± 56 and 184 ± 59 mg/dl, respectively, mean $\pm$ SD); this showed that the patients in the present study had greater insulin resistance. These findings are important when selecting the mode of insulin therapy, because the cutoff levels for CPR indices depend on the patient's BMI. Cutoff values were determined under a fasting hyperglycemia of approximately 180 mg/dl, a level commonly observed in clinical practice, and had sufficient specificity (80.7 %, CPR2h), suggesting that these values will be practically useful for selecting an appropriate insulin regimen.

Being obese or overweight may influence the selection of insulin regimens because of insulin resistance in obese subjects [3, 13]. Whether the grade of insulin resistance contributes to the need for insulin treatment was not examined in the present study, which may limit the application of the results of this study for clinical use. Nevertheless, defects in the actions and secretion of insulin, which are the two main pathophysiological features of type 2 diabetes, have been shown to independently contribute to increases in plasma glucose levels [14]. Although insulin resistance is known to be the predominant driver of diabetes in obese patients, a similar degree of islet cell dysfunction has been observed upon comparing obese and leaner

patients [1, 14], thereby supporting the usefulness of our indices. The insulin dosage required per day has been shown to be dependent on insulin resistance in type 2 diabetes [15], while the need for treatments with insulin was mainly indicated by an annual decline in insulin secretion, as shown in our study. The daily insulin dosage of obese patients receiving the final treatment in the present study was significantly larger than that of non-obese subjects in both the BOT and MDI groups (Table S4 in the ESM).

Using insulin-sensitizing medicines such as metformin or pioglitazone, which were not an option in the present protocol, improves fasting glucose levels in obese patients by suppressing hepatic gluconeogenesis. Additionally, pioglitazone was reported to delay insulin initiation in a subanalysis of the PROactive study [16]. Therefore, the basal insulin dose in the BOT group could be tapered or terminated when any of the insulin sensitizers was challenged. Nevertheless, although the efficacies of these agents for type 2 diabetes with decreased insulin secretory ability should be validated, insulin sensitizers are still a supplementary option for advanced-stage patients requiring MDI therapy [17]. Because there is no direct enhancing effect of insulin sensitizers on insulin secretion [18], these agents would not change the requirement for MDI therapy. A study comparing the efficacies of insulin sensitizers for glucose metabolism in obese type 2 diabetes patients with sustained insulin secretion to those with decreased insulin secretion is needed.

Riddle [19] and Shank et al. [20] described combined therapy with basal insulin and oral agents in which bedtime NPH insulin was administered with daytime SU therapy. A long-acting insulin analog (glargine or detemir) is currently used as basal insulin, and a combination therapy of oral agents with basal insulin is termed BOT. However, the clinical characteristics and insulin secretory abilities of patients with type 2 diabetes successfully treated with BOT have not been thoroughly investigated [21]. Although patients assigned to the BOT group responded to insulin secretagogues, their blood glucose levels before breakfast did not attain the target level even with the maximum oral doses determined with the protocol, so a basal insulin dose was added to the regimen. Serum CPR levels of the patients in the BOT group were sustained more than those in the MDI group, but some CPR values in the BOT group were significantly lower than those in the OHA-alone group. As seen in Table 2, the CPR values in the OHA-alone group were higher than those in the BOT group, which in turn were higher than those in the MDI group. This shows that BOT represents the midpoint between non-insulin and intensive insulin therapy on the clinical road map for type 2 diabetes.

Incretin-related agents such as dipeptidyl peptidase-4 (DPP-4) inhibitors and glucagon-like peptide-1 (GLP-1)

receptor agonists are currently being administered to patients with type 2 diabetes, but these agents were not available when our treatment protocol was created. Although GLP-1 receptor agonists were previously shown to be effective for obese patients with type 2 diabetes [13, 22], the relationship between the efficacy of these agonists and the insulin secretory ability of the patient has not yet been fully elucidated. Two studies recently demonstrated that liraglutide was less responsive in Japanese patients with decreased beta-cell function than in those with sustained insulin secretion [23, 24]. The results of the present study suggest that many obese patients with type 2 diabetes whose serum CPR2h levels are below 3.2 ng/ml (cutoff) would not respond to the administration of GLP-1 receptor agonists because these patients require MDI therapy to achieve favorable blood glucose levels (Table 4). However, this hypothesis should be investigated further using a protocol that is designed to challenge patients with incretin-related agents, especially GLP-1 receptor agonists, after improving blood glucose levels with IIT [25].

The durabilities of the treatment efficacies of diabetic agents, except for insulin, must be linked to residual insulin secretory ability [26]. However, although the Δ CPR value obtained from a glucagon loading test is an index that can be used to estimate the yearly decline in insulin secretion in Japanese patients with type 2 diabetes [27, 28], the utility of this value as an indicator for some insulin regimens was found to be inferior to the utility of postprandial indices in the current analysis. The postprandial indices CPR2h and CPI2h, obtained from the physiological meal load, may reflect two insulin secretory mechanisms: postprandial glucose-stimulated insulin secretion and glucose-dependent insulin secretion by incretin [29]. This may have been why the postprandial indices proved advantageous as indices for determining the need for insulin therapy as compared to fasting or non-physiological CPR indices. The latter indices could not reflect the impaired regulation of incretin effects in type 2 diabetes [30].

Renal dysfunction delays the excretion of CPR into the urine, which elevates serum CPR levels. Accordingly, the presence of renal dysfunction may cause the usefulness of the CPR index to be underestimated because the sensitivity of the index is lowered in ROC analysis. This could be a limitation of our study. Nevertheless, the results of our present study are useful for practical applications beyond renal conditions in which the serum creatinine concentration ranges up to 1.5 mg/dl (Table S5 in the ESM).

The current results show that the likelihood of the treatment proceeding from non-insulin to BOT to MDI is closely related to the patient's residual insulin secretory ability. An appropriate insulin regimen can be identified with approximately 80 % specificity if any of the patient's postprandial serum CPR index levels are

known. Therapy individualization for type 2 diabetes should be based on more patient characteristics [17, 31], but the present study revealed that residual beta-cell function is one of the most significant determinants of whether insulin treatment is needed and, if so, the appropriate regimen to implement.

In conclusion, the postprandial serum CPR concentration or CPI value measured 2 h after breakfast was identified as the best index for predicting an appropriate insulin regimen to achieve good glycemic control in obese patients with type 2 diabetes. We hope to confirm our results in follow-up evaluations of the selected insulin regimen.

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Compliance with ethical standards

Conflict of interest The authors report no potential conflicts of interest, and no sponsors were involved in this retrospective study.

Human rights statement and informed consent The clinical study plan, treatment protocol, and informed consent form were submitted to the Clinical Research Approval Committee and approved by the Medical Ethics Committee of Kurashiki Central Hospital in accordance with the Helsinki Declaration of 1964 and later revisions of it. The attending physician provided a written explanation of the study protocol, and informed consent was obtained from all patients.

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