

The Impact of Insulin Resistance on Proinsulin Secretion in Pregnancy

Hyperproinsulinemia is not a feature of gestational diabetes

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OBJECTIVE— Excessive secretion of the insulin precursor proinsulin, as manifested by an increased serum proinsulin-to-insulin ratio, has been associated with β -cell dysfunction. In women with gestational diabetes mellitus (GDM), previous studies of the proinsulin-to-insulin ratio have yielded conflicting results, despite the presence of β -cell dysfunction. The interpretation of the proinsulin-to-insulin ratio, however, may be confounded by the variable effects of hepatic insulin extraction. Thus, we sought to determine whether GDM is characterized by relative hyperproinsulinemia as measured by the proinsulin-to-C-peptide ratio, an alternate measure of proinsulin secretion that is not affected by hepatic insulin extraction.

RESEARCH DESIGN AND METHODS— Serum proinsulin, C-peptide, and insulin were measured in a cross-sectional study of 180 women undergoing oral glucose tolerance tests (OGTTs) in the late second or early third trimester. Based on the OGTT, participants were stratified into three groups: 1) normal glucose tolerance (NGT; $n = 93$), 2) impaired glucose tolerance (IGT; $n = 39$), and 3) GDM ($n = 48$). Insulin sensitivity (IS) was measured using the IS_{OGTT} index of Matsuda and DeFronzo, which has been previously validated in pregnant women.

RESULTS— There were no significant differences in mean fasting proinsulin-to-C-peptide ratio between the three glucose tolerance groups (NGT, 0.024; IGT, 0.022; GDM, 0.019; $P = 0.4$). Furthermore, adjustment for age, weeks' gestation, prepregnancy BMI, ethnicity, previous GDM, and family history of diabetes did not reveal any association between the proinsulin-to-C-peptide ratio and glucose tolerance status. Using Spearman univariate correlation analysis, fasting proinsulin-to-C-peptide ratio was significantly correlated with IS_{OGTT} ($r = 0.29$, $P < 0.0001$) and inversely related to the homeostasis model assessment of insulin resistance ($r = -0.36$, $P < 0.0001$) and prepregnancy BMI ($r = -0.23$, $P < 0.005$). On multiple linear regression analysis, IS_{OGTT} emerged as the strongest independent correlate of the dependent variable proinsulin-to-C-peptide ratio. Furthermore, after adjustment for potential covariates, a stepwise decrease in proinsulin-to-C-peptide ratio was observed per decreasing tertile of IS_{OGTT} (trend $P = 0.0019$), consistent with enhanced efficiency of proinsulin processing (i.e., reduced proinsulin-to-C-peptide ratio) as insulin resistance increases.

CONCLUSIONS— GDM is not independently associated with hyperproinsulinemia as measured by the proinsulin-to-C-peptide ratio. Instead, in pregnant women, increased insulin resistance is associated with decreased proinsulin-to-C-peptide ratio, independently of glucose tolerance status. These data suggest that relative proinsulin secretion in late pregnancy is primarily related to insulin resistance and does not necessarily reflect β -cell function.

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Abbreviations: AUC, area under the curve; GDM, gestational diabetes mellitus; HOMA-B, homeostasis model assessment for β -cell function; HOMA-IR, HOMA of insulin resistance; IGT, impaired glucose tolerance; NGT, normal glucose tolerance; OGTT, oral glucose tolerance test.

A table elsewhere in this issue shows conventional and Système International (SI) units and conversion factors for many substances.

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During insulin biosynthesis, the intracellular processing of proinsulin generates other peptides that are cosecreted by the pancreatic β -cell alongside insulin. Although their physiologic relevance is unclear, serum concentrations of these products, which include C-peptide, split products (des 31,32 and des 64,65), and the insulin precursor proinsulin may provide insight into β -cell function. Indeed, individuals with type 2 diabetes typically exhibit a disproportionate increase in serum proinsulin concentration in relation to insulin levels (as manifested by an elevated proinsulin-to-insulin ratio) (1,2). Importantly, this relative hyperproinsulinemia is significantly correlated with decreasing acute insulin response to glucose in subjects with type 2 diabetes (3). In addition, hyperproinsulinemia in pre-diabetic individuals has been prospectively associated with incident diabetes (4–6). Taken together, these observations have led to the hypothesis that relative hyperproinsulinemia is a marker of β -cell dysfunction, possibly early in the pathophysiology of type 2 diabetes.

The evaluation of proinsulin concentration in populations at risk of diabetes can be complicated by several factors, including insulin resistance, obesity, and familial predisposition to diabetes. For example, although established type 2 diabetes is associated with hyperproinsulinemia, it has been suggested that the normal β -cell response to increased secretory demand is enhanced efficiency of proinsulin processing, leading to a decreased proinsulin-to-insulin ratio (1). In support of this concept, both insulin resistance and its clinical correlate, obesity, have been shown to be associated with decreased proinsulin-to-insulin ratios in normoglycemic subjects (7–9). On the other hand, the interpretation of these relationships as a reflection of β -cell function may be confounded by the effects of hepatic insulin extraction. Specifically, in insulin-resistant states, the proinsulin-to-insulin ratio may be decreased because of reduced insulin clearance (reflecting hepatic insulin resistance) rather than en-

hanced proinsulin processing at the β -cell.

Since C-peptide is cosecreted in equimolar amounts with insulin but is not subject to hepatic clearance, the proinsulin-to-C-peptide ratio has been proposed as an alternate, more robust measure of relative proinsulin secretion (10,11). Indeed, support for this idea has been provided by Vaukhonen et al. (10) in their recent evaluation of the proinsulin-to-C-peptide ratio in normoglycemic offspring of patients with type 2 diabetes. After first showing that the proinsulin-to-insulin ratio in the offspring varied according to insulin resistance, they demonstrated that there is no difference in proinsulin-to-C-peptide ratio, regardless of insulin resistance, when comparing offspring with healthy control subjects. Thus, whereas earlier studies had yielded conflicting results regarding the relationship between family history of diabetes and proinsulin-to-insulin ratio, Vaukhonen et al. (10) suggested that hyperproinsulinemia is not a characteristic feature in the offspring, despite their increased risk of future diabetes.

Women with gestational diabetes mellitus (GDM) represent another population at high risk of developing type 2 diabetes. GDM arises in a subset of women in whom the considerable insulin resistance of late pregnancy unmasks a latent β -cell defect (12). Despite the presence of this β -cell defect, previous studies of proinsulin-to-insulin ratios in GDM have yielded conflicting results, ranging from a normal ratio to hyperproinsulinemia (13–15). Importantly, however, these studies have not evaluated the proinsulin-to-C-peptide ratio. Thus, in the current cross-sectional study in late pregnancy, we sought to determine whether GDM is characterized by relative hyperproinsulinemia as measured by the proinsulin-to-C-peptide ratio.

RESEARCH DESIGN AND METHODS

The study design and laboratory methods have been fully described previously (16,17). The protocol was approved by the research ethics board at Mount Sinai Hospital, and all subjects gave written informed consent. In brief, study participants consisted of 180 healthy pregnant women attending outpatient obstetrics clinics who had been referred for a 100-g oral glucose tolerance test (OGTT) following an abnormal result on a screening 50-g glucose challenge test (plasma glucose ≥ 7.8

mmol/l at 1 h p.c.). Demographic and historical information was collected by interviewer-administered questionnaire at the time of the OGTT, as previously described (16,18). The OGTT stratified participants into three glucose tolerance groups: 1) GDM, as defined by National Diabetes Data Group criteria (19) (which requires at least two of the following: fasting glucose >5.8 ; 1-h p.c. >10.6 ; 2-h p.c. >9.2 ; 3-h p.c. >8.1); 2) impaired glucose tolerance (IGT), defined by meeting only one of the above criteria; and 3) normal glucose tolerance (NGT), defined as not meeting any of the above criteria. It should be noted that the National Diabetes Data Group report does not define an IGT subclassification for pregnancy in this way (i.e., on the basis of a single abnormal OGTT value) (19). In the current analysis, this customized definition of IGT has been used to identify subjects with an intermediate degree of glucose intolerance (between NGT and GDM), as evidenced by their single abnormal glucose value.

Venous blood samples for laboratory measurement of insulin, C-peptide, and proinsulin were drawn at fasting and hourly during the OGTT. Specific insulin was measured using the Roche Elecsys 1010 immunoassay analyzer and the electrochemiluminescence immunoassay kit. This assay shows 0.05% cross-reactivity to intact human proinsulin and the primary circulating split form (des 31,32). C-peptide was measured by chemiluminescent enzyme immunoassay (Immulate 2000; Diagnostic Products, Los Angeles, CA). Cross-reactivity of this assay is 0% for insulin and 17% for proinsulin (at 10 ng/ml). Proinsulin was measured by radioimmunoassay (catalog no. HPI-15K; Linco Research, St. Charles, MO). This assay shows $<0.1\%$ cross-reactivity to both human insulin and C-peptide, respectively.

Measures of proinsulin/insulin secretion and insulin sensitivity/resistance

The fasting proinsulin-to-insulin and proinsulin-to-C-peptide ratios were calculated as measures of relative proinsulin secretion. The fasting C-peptide-to-insulin ratio was calculated as a measure of hepatic insulin extraction (C-peptide-to-insulin ratio decreases as hepatic insulin extraction decreases) (20). Stimulated indexes for glucose, insulin, C-peptide, proinsulin, C-peptide-to-insulin, proinsulin-to-insulin and proinsulin-to-C-

peptide ratios were determined as the area under the curve (AUC) for the respective measures during the OGTT by applying the trapezoidal rule to hourly measurements of these analytes.

The Stumvoll first-phase measure of insulin secretion is defined by the following formula: $1,194 + 4.724 \times \text{Ins}_0 - 117.0 \times \text{gluc}_{60} + 1.414 \times \text{Ins}_{60}$ (21). The homeostasis model of assessment for β -cell function (HOMA-B) and insulin resistance (HOMA-IR) were calculated as described by Matthews et al. (22). The IS_{OGTT} index of insulin sensitivity was calculated as described by Matsuda and DeFronzo (23). In a validation study in pregnant patients, the IS_{OGTT} index showed better correlation with insulin sensitivity derived using the euglycemic-hyperinsulinemic clamp technique than did the HOMA-IR model (24).

Statistical analysis

All analyses were conducted using the Statistical Analysis System (SAS version 8.02; SAS Institute, Cary, NC). A P value <0.05 was considered statistically significant. Means and SDs (or medians and interquartile ranges for skewed variables) were presented by glucose tolerance group, and ANOVA was used to assess univariate differences among continuous variables (Table 1). The distributions of fasting insulin, AUC insulin, fasting proinsulin, AUC proinsulin, AUC C-peptide, fasting C-peptide-to-insulin ratio, AUC C-peptide-to-insulin ratio, fasting proinsulin-to-insulin ratio, AUC proinsulin-to-insulin ratio, fasting proinsulin-to-C-peptide ratio and AUC proinsulin-to-C-peptide ratio, IS_{OGTT} , HOMA-IR, HOMA-B, and Stumvoll first phase were skewed, and thus, medians and interquartile ranges were presented for these variables (Table 1). The natural logarithmic transformations of these skewed variables were used in univariate and multivariate analyses, with back-transformed results from multivariate analyses presented in Fig. 1. Univariate associations of fasting proinsulin-to-C-peptide ratio with continuous measures of age, adiposity, glucose, insulin resistance, and insulin secretion were assessed with Spearman's correlation analysis. ANCOVA was used to test differences in fasting proinsulin-to-C-peptide ratios across categories of glucose tolerance after adjustment for covariates including age, weeks' gestation, prepregnancy BMI, weight gain in pregnancy, ethnicity, personal history of previous GDM or delivery

Table 1—Insulin, proinsulin, and C-peptide secretion by glucose tolerance status

Characteristic	NGT	IGT	GDM	P*
n	93	39	48	
Age (years)	33.2 ± 4.5	32.8 ± 4.7	34.0 ± 4.3	0.3980
Weeks' gestation	29.2 ± 2.7	29.7 ± 1.9	29.1 ± 2.2	0.4268
Prepregnancy BMI (kg/m ²)	23.9 ± 4.5	24.1 ± 4.2	24.7 ± 5.1	0.5977
Insulin (pmol/l)				
Fasting	58 (40–77)	63 (47–81)	74 (54–104)	0.0036
AUC	1,320 (925–1,837)	1,561 (1,091–1,920)	1,709 (1,267–2,538)	0.0046
Proinsulin (pmol/l)				
Fasting	17.7 (9.3–29.4)	15.2 (7.9–22.9)	17.2 (9.1–27.3)	0.5286
AUC	70.5 (51.7–91.5)	66 (39.6–83.4)	57.7 (44.9–102.3)	0.6586
C-peptide (pmol/l)				
Fasting	742 ± 288	758 ± 258	863 ± 330	0.0654
AUC	7,076 (5,571–9,611)	7,928 (6,071–9,200)	8,717 (6,877–9,748)	0.1156
C-peptide-to-insulin ratio				
Fasting	12.3 (10.5–15.1)	12.3 (9.5–14.9)	11.2 (8.9–13.0)	0.0623
AUC	21.1 (17.1–24.9)	19.6 (16.4–24.9)	17.3 (14.8–21.2)	0.0211
Proinsulin-to-insulin ratio				
Fasting	0.277 (0.16–0.60)	0.197 (0.13–0.43)	0.221 (0.11–0.50)	0.1317
AUC	0.292 (0.18–0.48)	0.230 (0.13–0.36)	0.224 (0.11–0.36)	0.0155
Proinsulin-to-C-peptide ratio				
Fasting	0.024 (0.01–0.04)	0.022 (0.01–0.04)	0.019 (0.01–0.03)	0.4414
AUC	0.039 (0.02–0.05)	0.035 (0.02–0.05)	0.028 (0.02–0.05)	0.3465

Data are means ± SD for age, weeks' gestation, prepregnancy BMI, and fasting C-peptide. All other values are medians (interquartile range). *P values refer to overall differences across groups as derived from ANOVA. P values in bold are <0.05.

of an infant ≥10 lbs, and family history of type 2 diabetes or GDM (Fig. 1A). A similar analysis was performed across tertiles of prepregnancy BMI using the same covariates, except for the inclusion of variables representing glucose intolerance (IGT, GDM), in place of prepregnancy BMI (Fig. 1B). Finally, ANCOVA was also used to test differences in mean adjusted fasting proinsulin-to-C-peptide ratios across tertiles of IS_{OGTT} using the same covariates as in Fig. 1B in addition to prepregnancy BMI (Fig. 1C). Multiple linear regression analysis was used to determine factors that were significantly and independently associated with variations in the fasting proinsulin-to-C-peptide ratio.

RESULTS— As described in an earlier report, there were no significant differences between the glycemic tolerance groups with respect to age, weeks' gestation, prepregnancy BMI, weight gain during pregnancy, ethnicity, parity, smoking exposure, and family history of type 2 diabetes or GDM (Table 1) (16). In the GDM group, 18.8% of subjects had a history of GDM or delivery of a macrosomic infant in a previous pregnancy compared with 10.3% of IGT subjects and 4.3% of NGT subjects (overall $P = 0.0206$). As

expected, fasting insulin and AUC insulin were highest in GDM, followed in turn by IGT and NGT, respectively (fasting insulin $P = 0.0036$; AUC insulin $P = 0.0046$) (Table 1). On the other hand, both fasting and stimulated C-peptide-to-insulin ratios exhibited declining trends from NGT to IGT to GDM (fasting C-peptide-to-insulin ratio $P = 0.0623$; AUC C-peptide-to-insulin ratio $P = 0.0211$), suggestive of reduced hepatic insulin extraction with worsening glucose tolerance.

Whereas fasting proinsulin-to-insulin ratios showed no significant differences between glucose tolerance groups, the AUC proinsulin-to-insulin ratio was highest in NGT, followed in turn by IGT and GDM, respectively ($P = 0.0155$). These differences in stimulated proinsulin-to-insulin, however, were abolished upon adjustment for variation in hepatic insulin extraction using AUC C-peptide-to-insulin ratio (adjusted mean AUC proinsulin-to-insulin ratio: NGT, 0.271; IGT, 0.243; GDM, 0.225; overall $P = 0.2391$). Both fasting and stimulated proinsulin-to-C-peptide ratio, on the other hand, showed no significant differences between the glucose tolerance groups. These data illustrate the advantage of evaluating relative proinsulin secretion in

relation to C-peptide, rather than insulin levels, since the potential confounding effects of variable hepatic insulin extraction are avoided with the proinsulin-to-C-peptide ratio.

Using Spearman univariate correlation analysis, fasting proinsulin-to-C-peptide ratio was inversely related to prepregnancy BMI ($r = -0.23$, $P < 0.005$). Similarly, proinsulin-to-C-peptide ratio was inversely related to HOMA-IR ($r = -0.36$, $P < 0.0001$) and positively correlated with the IS_{OGTT} index of insulin sensitivity ($r = 0.29$, $P < 0.0001$), suggesting that relative proinsulin secretion decreases with worsening insulin resistance in this population. Fasting proinsulin-to-C-peptide ratio was also inversely related to HOMA-B ($r = -0.27$, $P < 0.0005$) and to the Stumvoll first-phase index of insulin secretion ($r = -0.20$, $P < 0.01$), consistent with the concept of relative hyperproinsulinemia as insulin secretion declines. Interestingly, a weak inverse correlation between proinsulin-to-C-peptide ratio and age ($r = -0.17$, $P < 0.05$) was noted, consistent with an earlier observation in pregnant women (15). Finally, there was no significant association between proinsulin-to-C-peptide ratio and any glucose measurement during the OGTT, includ-

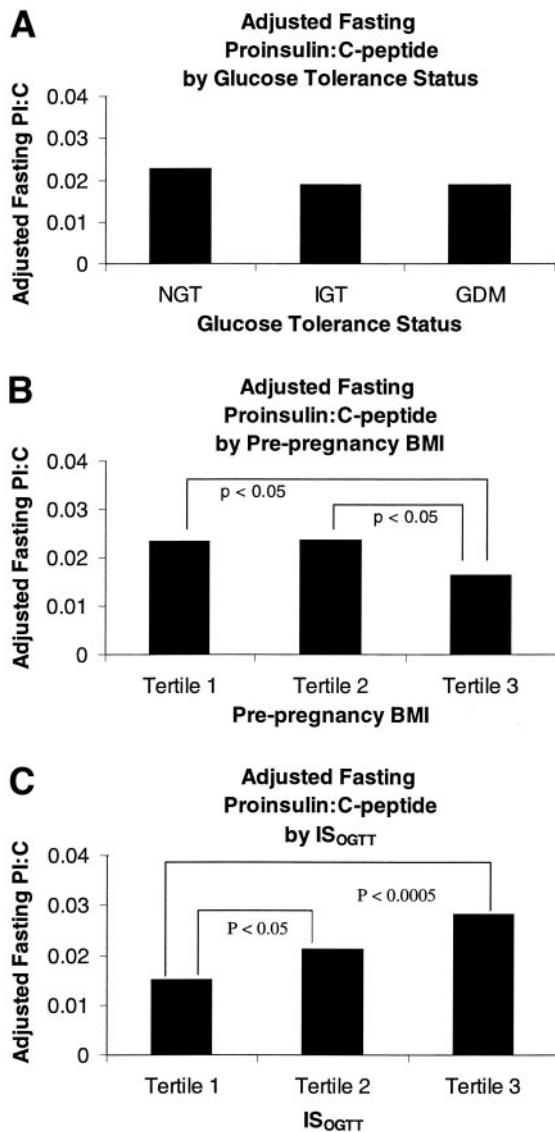


Figure 1— Mean adjusted fasting proinsulin-to-C-peptide (proinsulin-to-C-peptide ratio) by glucose tolerance status (A), tertile of prepregnancy BMI (B), and tertile of IS_{OGTT} (C). A: Fasting proinsulin-to-C-peptide ratio by glucose tolerance status adjusted for age, weeks' gestation, prepregnancy BMI, ethnicity, personal history of previous GDM, and family history of diabetes. *Trend $P = 0.4083$, and all pairwise P values are not significant. B: Fasting proinsulin-to-C-peptide ratio by tertile of prepregnancy BMI, adjusted for age, weeks' gestation, ethnicity, history of previous GDM, family history of diabetes, and current glucose intolerance (IGT, GDM). *Trend $P = 0.0509$; **Tertile 1: 16.5–21.4 kg/m²; tertile 2: 21.4–25.4 kg/m²; tertile 3: 25.6–41.0 kg/m². C: Fasting proinsulin-to-C-peptide ratio by tertile of IS_{OGTT}, adjusted for age, weeks' gestation, ethnicity, prepregnancy BMI, personal history of previous GDM, family history of diabetes, and current glucose intolerance (IGT, GDM). *Trend $P = 0.0019$; **tertile 1: 0.65–3.59; tertile 2: 3.61–5.25; tertile 3: 5.25–13.84.

ing AUC glucose. Indeed, even after adjustment for potential covariates (including age, weeks' gestation, prepregnancy BMI, ethnicity, personal history of previous GDM, and family history of type 2 diabetes/GDM), there were no significant differences in mean adjusted fasting proinsulin-to-C-peptide ratio across the glucose tolerance groups (Fig. 1A).

To evaluate independent correlates of relative proinsulin secretion, multiple linear regression analysis was performed with fasting proinsulin-to-C-peptide ratio as the dependent variable. In a model fully adjusted for age, weeks' gestation, prepregnancy BMI, ethnicity, current glucose intolerance (IGT, GDM), personal history of previous GDM, and family history of type 2 diabetes/GDM, the sole independent and negative correlates of fasting proinsulin-to-C-peptide ratio

were prepregnancy BMI ($t = -2.32$, $P = 0.0214$) and age ($t = -1.98$, $P = 0.0494$). Repeating this multiple linear regression analysis with inclusion of IS_{OGTT} as an additional covariate increased the explained variance in proinsulin-to-C-peptide ratio considerably from 10.0 to 17.9%. In this model, IS_{OGTT} emerged as an independent correlate of fasting proinsulin-to-C-peptide ratio ($t = 3.98$, $P = 0.0001$), and age remained a negative covariate ($t = -2.56$, $P = 0.0112$) (pregnancy BMI was no longer an independent correlate).

To further study the inverse independent relationship between maternal obesity and relative proinsulin secretion, study participants were stratified into tertiles based on prepregnancy BMI. After adjustment for potential covariates (including age, weeks' gestation, ethnicity,

current glucose intolerance, personal history of previous GDM, and family history of type 2 diabetes/GDM), mean fasting proinsulin-to-C-peptide ratio remained significantly lower among women in the highest tertile of prepregnancy BMI compared with their leaner counterparts in either of the other two tertiles (both comparisons, $P < 0.05$) (Fig. 1B). Further adjustment for IS_{OGTT}, however, abolished these relationships (data not shown), suggesting that differences in insulin sensitivity likely mediate the relationship between BMI and fasting proinsulin-to-C-peptide ratio.

Finally, to further study this relationship, participants were stratified into tertiles based on IS_{OGTT} (Fig. 1C). After adjustment for potential covariates, a stepwise increase in fasting proinsulin-to-C-peptide ratio was observed with increasing tertiles of IS_{OGTT} (trend $P = 0.0019$). Accordingly, women with the greatest insulin resistance (i.e., those women in the lowest tertile of IS_{OGTT}) exhibited significantly lower mean adjusted fasting proinsulin-to-C-peptide ratio compared with their more insulin-sensitive counterparts in the middle tertile (pairwise $P < 0.05$) and highest tertile (pairwise $P < 0.0005$) of IS_{OGTT}, respectively.

CONCLUSIONS— In this report, we demonstrate that GDM is not independently associated with hyperproinsulinemia when using the proinsulin-to-C-peptide ratio to evaluate relative proinsulin secretion. Instead, rather than glucose intolerance, insulin resistance emerged as the primary determinant of relative proinsulin secretion in late pregnancy. Furthermore, increased insulin resistance was significantly associated with decreased proinsulin-to-C-peptide ratio, suggesting that the efficiency of proinsulin processing is enhanced in response to insulin resistance in pregnancy, independently of glucose tolerance status. Accordingly, relative proinsulin secretion in late pregnancy primarily reflects insulin resistance rather than β -cell dysfunction.

Previous studies of relative proinsulin levels in GDM have yielded conflicting results. In a small study ($n = 20$), Dornhorst et al. (13) found that the proinsulin-to-insulin ratio was elevated in women with GDM when compared with control subjects. In contrast, in a slightly larger sample ($n = 40$), Kautzky-Willer et al. (14) reported no differences in fasting proinsulin-to-insulin, although postprandial

proinsulin-to-insulin was elevated in GDM. Finally, Festa et al. (15) found no differences in fasting proinsulin-to-insulin ratios when comparing mild GDM ($n = 52$) with NGT ($n = 157$). Several factors may have contributed to the lack of consistent findings in these studies. These factors include differences among the study populations with respect to severity of glucose intolerance, body weight, and hepatic insulin extraction (i.e., using the proinsulin-to-insulin ratio). The current study thus reconciles these issues by evaluating the proinsulin-to-C-peptide ratio, rather than the proinsulin-to-insulin ratio, in a large study population across a broad range of both glucose tolerance and maternal BMI.

The present study highlights the advantage of using the proinsulin-to-C-peptide ratio rather than the proinsulin-to-insulin ratio in the evaluation of relative proinsulin secretion. Whereas stimulated proinsulin-to-insulin ratios exhibited an association with glucose tolerance status that was likely mediated by hepatic insulin extraction, the proinsulin-to-C-peptide ratio provided an unbiased measure of relative proinsulin secretion, unaffected by insulin clearance at the liver. The importance of this issue is underscored by the central role of the liver in regulating peripheral insulin concentration. For example, the gate-keeping role of the liver in the regulation of the systemic insulin response to glucose has been previously demonstrated in studies of obese patients undergoing weight-reduction surgery (25). Indeed, following massive weight loss, the initial mechanism underlying the observed reduction in insulin levels (i.e., reflecting improved insulin sensitivity) is enhanced hepatic clearance of insulin, rather than a decline in insulin production (which occurs subsequently) (25).

The current findings regarding differences between the proinsulin-to-C-peptide ratio and the proinsulin-to-insulin ratio in GDM mirror the experience of Vauhkonen et al. (10) described earlier, in their study of these measures in offspring of patients with type 2 diabetes. Like GDM, family history of diabetes had previously exhibited various associations with the proinsulin-to-insulin ratio (10). Thus, the proinsulin-to-C-peptide ratio emerges as a measure that may reconcile conflicting observations noted in the proinsulin literature to date.

In the current study, the absence of increased proinsulin-to-C-peptide ratio

in patients with GDM suggests that the β -cell defect in this condition is not associated with relative hyperproinsulinemia. β -Cell dysfunction in GDM is characterized by a quantitative defect in insulin secretion, previously illustrated using the disposition index, such that the compensatory increase in insulin secretion required to counter the severe insulin resistance of late pregnancy is insufficient to achieve normoglycemia (12). In light of the current findings, it may be surmised that, unlike in established type 2 diabetes (1,2), abnormal proinsulin processing is not a feature of this relative insulin insufficiency in GDM. Interestingly, this idea is consistent with a recent hypothesis that the quantitative insulin secretory defect in GDM may be related to reduced β -cell mass, secondary to progesterone-mediated inhibition of β -cell proliferation (26,27). Although this mechanism remains speculative at this time, reduced β -cell mass could nevertheless reconcile the maintenance of normal relative proinsulin secretion with a deficiency in total insulin output.

Given the risk of future type 2 diabetes in women with GDM, it is possible that relative hyperproinsulinemia may develop over time in tandem with progressive β -cell dysfunction and progression to type 2 diabetes. In support of this idea, in the Women's Health Study, an increased proinsulin-to-insulin ratio in healthy women was most strongly associated with an imminent diagnosis of type 2 diabetes (i.e., within 2 years, as opposed to 3 or 4 years) (28). Accordingly, when first presenting during pregnancy, β -cell dysfunction in women with GDM may not yet have advanced to the point of abnormal proinsulin processing. Longitudinal evaluation of proinsulin-to-C-peptide ratio in women with previous GDM is thus needed to address the possibility that hyperproinsulinemia may develop over time, as β -cell function worsens. In this regard, it is of interest to note that increased proinsulin-to-insulin ratios have been reported in women with a history of previous GDM (29).

The inverse relationship between maternal obesity/insulin resistance and the proinsulin-to-C-peptide ratio in pregnancy reported in this study is consistent with previous observations linking obesity and insulin resistance with decreased proinsulin-to-insulin ratios in normoglycemic individuals (7–9). Importantly, the use of the proinsulin-to-C-peptide ratio in the current context supports the idea

that enhanced processing of proinsulin (rather than decreased hepatic insulin extraction) underlies this phenomenon. In addition, the demonstration of this relationship in the setting of GDM suggests that, despite the β -cell defect, the efficiency of proinsulin processing can be increased in GDM, just as in women with normal β -cell function. It is possible that this ability to compensate for insulin resistance by enhancing the efficiency of proinsulin processing may diminish over time in these patients. Again, longitudinal evaluation is indicated.

We recognize that the cross-sectional nature of this study limits the ability to draw causal inferences. Nevertheless, this report represents the first analysis to evaluate the interrelationships between proinsulin-to-C-peptide ratio, maternal obesity, and glucose intolerance in pregnancy. It is also recognized that the inability to detect a relationship between GDM and relative hyperproinsulinemia could reflect a type II error. Arguing against this possibility, however, is the fact that the results reported herein are in agreement with those of the single larger study of relative proinsulin secretion in GDM (15). Finally, it should be noted that all study participants, including those comprising the NGT group, had a positive glucose challenge test before recruitment. Thus, findings with this NGT group may not reflect a truly normal patient population (i.e., with normal screening glucose challenge test and normal results on diagnostic OGTT). Further study is required.

In summary, when evaluated using proinsulin-to-C-peptide ratio, relative hyperproinsulinemia is not a feature of GDM. Instead, insulin resistance is the primary determinant of relative proinsulin secretion in late pregnancy. In pregnant women of all degrees of glucose tolerance including GDM, increased insulin resistance is associated with decreased proinsulin-to-C-peptide ratio, presumably due to enhanced proinsulin processing. In women with GDM, longitudinal study will be required to determine whether patterns of proinsulin secretion change over time as β -cell dysfunction progresses.

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