Clinical and Genetic Risk Factors for Type 2 Diabetes at Early or Late Post Partum After Gestational Diabetes Mellitus

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Context: Women with a history of gestational diabetes mellitus (GDM) are at increased risk of type 2 diabetes (T2DM). However, the time to progression to diabetes differs individually.

Objective: We investigated the clinical and genetic risk factors that are associated with T2DM early or late post partum after GDM pregnancy.

Design and Setting: This was a hospital-based prospective cohort study that enrolled GDM women.

Patients and Outcome Measures: A total of 843 GDM subjects were followed for the development of T2DM. Clinical risk factors were investigated during pregnancy, 2 months post partum, and annually thereafter. GDM subjects were genotyped for 21 known T2DM-associated genetic variants, and their genotype frequencies were compared with elderly nondiabetic controls.

Results: At 2 months post partum, 105 (12.5%) subjects had T2DM (early converters). Among the 370 remaining subjects who underwent more than 1 year of follow-up, 88 (23.8%) had newly developed T2DM (late converters). Independent risk factors for early converters were higher prepregnancy body mass index, higher area under the curve of glucose during an antepartum oral glucose tolerance test, lower fasting insulin concentration, and decreased β-cell function. Independent risk factors for late converters were higher prepregnancy body mass index and higher glucose area under the curve. Variants in *CDKN2A/2B* and *HHEX* were associated with early conversion, whereas variants in *CDKAL1* were associated with late conversion.

Conclusions: Obesity was a risk factor for both early and late T2DM converters. However, early converters had more pronounced defects in β-cell function, which might be explained, in part, by differences in genetic predisposition. **(*J Clin Endocrinol Metab* 98: E744–E752, 2013)**

Women with a history of gestational diabetes mellitus (GDM) are at increased risk of type 2 diabetes mellitus (T2DM). In a recent meta-analysis, women with a previous history of GDM had as much as a 7.43-fold relative risk of T2DM compared with those who had a normoglycemic pregnancy (1). However, there are ethnic differences in the risk of developing T2DM after GDM pregnancy. In addition, there are large individual varia-

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Abbreviations: AUC, area under the curve; BMI, body mass index; CI, confidence interval; FSIVGTT, frequently sampled iv glucose tolerance test; GDM, gestational diabetes mellitus; GWA, genome-wide association; HbA1c, hemoglobin A1c; IGT, impaired glucose tolerance; IQR, interquartile range; IR, insulin resistance; IS, insulin secretion; NGT, normal glucose tolerance; OGTT, oral glucose tolerance test; T2DM, type 2 diabetes mellitus.

tions in the time to progression to T2DM. In Asians, it is estimated that 5% to 15% of GDM women have early postpartum T2DM (2, 3). The rest of the women develop T2DM at rates of 20% to 60% within the following 5 to 10 years (4, 5). Factors that increase the risk of T2DM after GDM pregnancy include age, obesity, increasing parity, requirement of insulin therapy during pregnancy, family history of diabetes, and the degrees of hyperglycemia and decreased pancreatic β-cell function during pregnancy (5–9). However, it is not well known whether these risk factors differ between those who have diabetes in the early postpartum period and those who progress to diabetes later.

Recently, more than 60 T2DM-associated genetic risk loci have been identified by large-scale genome-wide association (GWA) studies (10–13). Because GDM women are at increased risk of T2DM and have a high rate of positive family history of diabetes, it is assumed that GDM and T2DM share similar genetic backgrounds to some extent. In our previous GWA study regarding GDM, we have shown that genetic risk variants of T2DM are enriched in GDM women (14). These genetic factors might also be associated with early or late development of T2DM in GDM women.

The objective of this study was to investigate the clinical and genetic risk factors that are associated with development of T2DM early or late post partum after GDM pregnancy. For this purpose, we investigated the anthropometric and metabolic characteristics of women who had diabetes at 2 months post partum (early converters). Women who did not have diabetes early post partum but developed T2DM more than 1 year after parturition (late converters) were also studied. A total of 21 genetic variants in 10 known T2DM-associated genes were genotyped in a subset of GDM women and an independent cohort of elderly (≥60 years old) nondiabetic controls. The genotype frequencies of these variants in early converters or late converters were compared with those of the elderly nondiabetic controls.

Subjects and Methods

Study Subjects

This was a prospective cohort study that enrolled GDM subjects between January 1996 and February 2003. The enrollment was done in Cheil General Hospital, Seoul, Korea, and the subjects were followed either at Cheil General Hospital or Seoul National University Bundang Hospital, Seongnam, Korea, until December 2010. The protocols for diagnosis and follow-up of GDM women were described in our previous publication (15). In brief, in a first screening step, all pregnant women performed a 50-g 1-hour glucose challenge test with a positive cutoff value of ≥7.2 mmol/L. Screen-positive women underwent a 100-g oral glucose tolerance test (OGTT). The diagnosis of GDM followed the criteria of the Third International Workshop-Conference on GDM (16). Women who had diabetes before pregnancy or positive results for GAD antibodies were excluded from the study. After parturition, all GDM women were scheduled for a 75-g OGTT at 2 months post partum and annually thereafter. Subjects were categorized into normal glucose tolerance (NGT), impaired glucose tolerance (IGT), and T2DM groups according to the American Diabetes Association criteria (17). A total of 843 women who underwent the 75-g OGTT at 2 months post partum were enrolled. Those who had T2DM at 2 months post partum were designated as early converters. Subjects who did not have diabetes at 2 months post partum but were subsequently diagnosed with T2DM more than 1 year after parturition were designated as late converters. A DNA sample for genotyping was available for a subgroup (n = 634) of subjects.

A total of 632 (345 women and 287 men) carefully selected, elderly, nondiabetic control subjects were used for comparing genotype frequencies with early converters and late converters. The inclusion criteria and the clinical characteristics of the elderly nondiabetic controls are described in our previous publication (18). In brief, the inclusion criteria were age ≥60 years, no previous history of diabetes, no first-degree relatives with diabetes, and fasting plasma glucose <6.1 mmol/L and hemoglobin A1c (HbA1c) <5.8%. The elderly nondiabetic control subjects were not used for comparison of metabolic phenotypes.

The Institutional Review Board of the Seoul National University Hospital and the Ethics Committee of the Cheil General Hospital approved the study protocol. Informed consent for genetic analysis was obtained from each subject. All clinical investigations were conducted according to the principles expressed in the Declaration of Helsinki.

Biochemical measurements

Plasma glucose was measured by the YSI 2300 STAT (YSI, Yellow Springs, Ohio) using the glucose oxidase method. Insulin was measured using a human-specific RIA kit (Linco Research, St Charles, Missouri). The areas under the curves (AUCs) of glucose and insulin were calculated using the trapezoidal rule. The 1-hour ΔI/ΔG (change in insulin concentration/change in glucose concentration) was calculated as $(\text{insulin}_{1\text{-h}} - \text{insulin}_0)/(\text{glucose}_{1\text{-h}} - \text{glucose}_0)$. Insulin sensitivity was evaluated by the Matsuda index as $10\,000/\sqrt{[(\text{fasting glucose}) \times (\text{fasting insulin}) \times (\text{mean glucose}) \times (\text{mean insulin})]}$ (19). Pancreatic β-cell function was assessed with the insulin secretion (IS)/insulin resistance (IR) disposition index, which is defined as [(AUC of insulin)/(AUC of glucose)] × (Matsuda index) (20, 21).

Genotyping

A total of 21 genetic variants in 10 genes (*PPARG*, *IGF2BP2*, *CDKAL1*, *SLC30A8*, *CDKN2A/2B*, *HHEX*, *TCF7L2*, *KCNQ1*, *KCNJ11*, and *FTO*) were genotyped in a subgroup (n = 634) of GDM subjects, which included 69 early converters and 70 late converters and an independent elderly cohort of nondiabetic controls (n = 632). The selection of genetic variants and the genotyping were performed as in our previous studies (22, 23). DNA was extracted from peripheral blood, and allelic discrimination was performed using the TaqMan assay (Applied Biosystems, Carlsbad, California). PCR amplification was performed by a PE 9700 thermal cycler (Applied Biosystems). The Sequence Detection System version 2.1 (Applied Biosystems)

was used for fluorescence detection. The overall genotype success rate was 99.4%, and the concordance rate based on duplicate comparisons was 99.6%.

Statistical analysis

Data are expressed as the mean ± SD in case of normal distribution or otherwise as median (interquartile range [IQR]). Student's *t* test was used to analyze the differences of quantitative traits between the NGT/IGT group and the T2DM group after pregnancy. Multivariate logistic regression analysis was used to identify independent risk factors for early and late converters.

Hardy-Weinberg equilibrium for each single-nucleotide polymorphism was analyzed using the χ^2 test. There was no significant deviation from Hardy-Weinberg equilibrium in the control subjects or in the overall population (data not shown). For the genetic association study, we compared the genotype frequencies between T2DM converters and elderly nondiabetic controls. The associations between genotypes and T2DM risk were assessed by logistic regression with an additive genotype model.

$P < .05$ was considered statistically significant in most of the analyses, except for the genetic association analyses, where Bonferroni correction was applied to adjust for multiple comparisons. In detail, because 10 genes and 2 comparisons (early converters and late converters) were used for the genetic association study, we divided the *P* value threshold (0.05) by the number of independent tests (0.05/10/2 = 0.0025) and considered $P < .0025$ to be significant. Statistical analyses were performed using SPSS version 19.0 for Windows software (SPSS, Chicago, Illinois) or PLINK version 1.07 (http://pngu.mgh.harvard.edu/purcell/plink/) (24).

Results

Incidence of T2DM after GDM pregnancy

Among the 843 GDM women who were available at 2 months post partum, 105 (12.5%) subjects had T2DM (Figure 1). After this period, the number of subjects with T2DM steadily increased at a rate of 6.8% per year until 10 years of follow-up. The median time to diagnosis of T2DM was 8.0 (95% confidence interval [CI] 7.2–8.9) years. When women who were diagnosed with T2DM at 2 months post partum were excluded, the median time to diagnosis of T2DM was 8.5 (95% CI 8.0–9.0) years. Among the 738 subjects who did not have T2DM at 2 months post partum, 370 (50.1%) women attended follow-up visits for more than 1 year. Those who participated in the long-term follow-up had higher antepartum glucose and lower insulin concentrations in antepartum 100-g OGTT and were more likely to undergo insulin treatment during GDM pregnancy (Table 1). Among the 370 follow-up subjects, 88 (23.8%) had T2DM during a median follow-up of 49 (IQR 30–82) months.

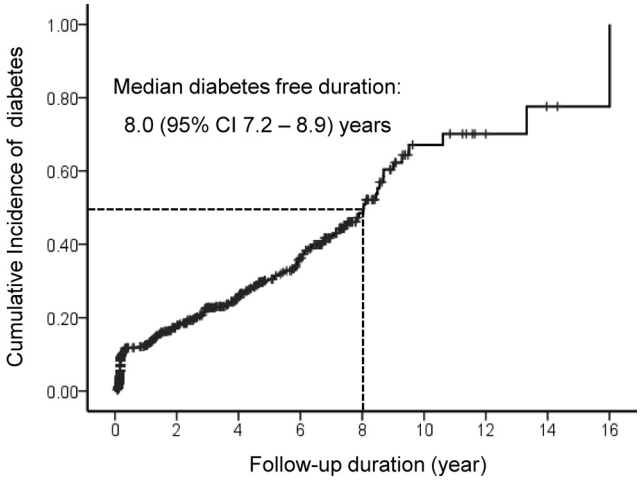

Figure 1. Cumulative incidence of T2DM after GDM pregnancy.

Clinical risk factors for early conversion to T2DM

We compared antepartum clinical and metabolic characteristics between the NGT/IGT group and T2DM group at 2 months post partum (Table 2). Prepregnancy body mass index (BMI) and pregnancy BMI at OGTT were significantly different between the 2 groups. Early T2DM converters had an earlier diagnosis of GDM during pregnancy compared with the NGT/IGT group at 2 months post partum (25.2 ± 5.3 vs 26.4 ± 3.0 weeks, $P = .030$). They were also more likely to receive insulin treatment during GDM pregnancy compared with the NGT/IGT group. Fasting, 1-, 2-, and 3-hour glucose and the AUC of glucose in the antepartum 100-g OGTT were significantly different between the 2 groups. The T2DM group had higher glucose values compared with the NGT/IGT group for all glucose measurements. The 1-hour $\Delta I/\Delta G$ of the early converters was about half of that of the NGT/IGT group (35 [IQR 16–55] vs 69 [IQR 46–107] pmol/mmol, $P < .05$). In contrast, no significant differences were noted regarding insulin sensitivity as assessed by the Matsuda index. Finally, the pancreatic β-cell function estimated by the IS/IR disposition index in early converters was only 59.0% of that in the NGT/IGT group at 2 months post partum (98 ± 53 vs 166 ± 60, $P < .001$).

Clinical factors for late conversion to T2DM

To identify risk factors associated with late conversion to T2DM, we investigated women who were NGT/IGT at 2 months post partum and attended follow-up for more than 1 year (Table 2). Late T2DM converters had higher prepregnancy BMI and pregnancy BMI compared with those who had NGT/IGT at more than 1 year of follow-up. Late converters were more likely to undergo insulin treatment during GDM pregnancy compared with the NGT/IGT group. When glucose values of the antepartum 100-g OGTT were compared, late converters had significantly higher glucose at all 4 time points and higher AUC of glucose compared with the NGT/IGT group. The insulin concentrations between the 2 groups

Table 1. Comparison of Antepartum Clinical Characteristics and Measures of Diagnostic 100-Gram OGTT Between Subjects Who Had Follow-up for More Than 1 Year and Those Who Did Not (Only 2-Month Postpartum NGT/IGT Subjects Are Included)[a]

	No Long-term Follow-up	Follow-up for More Than 1 Year	*P*
n (%)	368 (49.9)	370 (50.1)	
Age at pregnancy, y	31.2 ± 3.7	31.5 ± 4.0	.315
Pre-pregnancy BMI, kg/m^2	22.9 ± 3.7	22.5 ± 3.1	.103
Pregnancy BMI at OGTT, kg/m^2	27.3 ± 3.4	27.0 ± 3.2	.204
Weight gain during pregnancy, kg	10.9 ± 4.4	10.1 ± 4.3	.610
Gestational week at diagnosis, wk	26.4 ± 2.8	26.3 ± 3.1	.695
Parity, n	0.47 ± 0.64	0.48 ± 0.64	.816
Family history of DM, %	38.6	40.8	.538
Insulin treatment, %	10.6	24.6	<.001
Glucose, mmol/liter			
0-h	5.0 ± 0.8	5.1 ± 0.9	.004
1-h	10.8 ± 1.4	10.8 ± 1.6	.870
2-h	9.7 ± 1.5	10.0 ± 1.7	.010
3-h	7.8 ± 1.6	8.2 ± 1.8	.001
AUC of glucose	26.8 ± 3.1	27.5 ± 3.7	.011
Insulin (pmol/liter)[b]			
0-h	83 (63–104)	76 (53–97)	.007
1-h	535 (368–736)	417 (278–632)	<.001
2-h	670 (458–941)	563 (403–806)	<.001
3-h	517 (368–743)	458 (313–688)	.039
AUC of insulin	1527 (1111–2121)	1306 (917–1799)	<.001
1-h ΔI/ΔG, pmol/mmol[b]	79 (51–113)	60 (39–102)	<.001
Matsuda index[b]	3.28 (2.41–4.35)	3.62 (2.61–5.13)	<.001
IS/IR disposition index	173 ± 60	160 ± 60	.004

[a] Data are shown as the mean ± SD in case of normal distribution or otherwise as median (IQR).

[b] These variables were log transformed before statistical analysis.

did not show significant differences, except for a small difference at 2 hours. However, the late converters had significantly decreased AUC of insulin, 1-hour ΔI/ΔG, and IS/IR disposition index, which reflect β-cell dysfunction. Among the 155 subjects with 2-month postpartum IGT, 40 (25.8%) reverted to NGT, 56 (36.1%) maintained IGT, and 59 (38.1%) progressed to T2DM at greater than 1 year post partum (Supplemental Table 1, published on The Endocrine Society's Journals Online web site at http://jcem.endojournals.org).

When early converters and late converters were directly compared (Table 2), there were no differences in antepartum clinical risk factors between the 2 groups, except for an earlier diagnosis of GDM and higher insulin treatment rate in early converters. When the glucose and insulin concentrations of antepartum 100-g OGTT were compared, early converters had significantly higher glucose and lower insulin concentrations and decreased β-cell function compared with late converters.

Genetic risk factors for early or late conversion to T2DM

We investigated the genetic risk factors for early or late conversion to T2DM by comparing the genotypes of 21 known T2DM-associated genetic variants in early converters (n = 69), late converters (n = 70), and the independent cohort of elderly nondiabetic controls (n = 632, 345 women and 287 men) (Table 3). Compared with elderly nondiabetic controls, early converters had significantly increased risk allele frequencies of rs10811661 ($P <$ .00043) in *CDKN2A/2B* and rs1111875 ($P <$ .0010) and rs7923837 ($P <$ .0011) in *HHEX*. Late converters had an increased risk allele frequency of rs7754840 ($P <$ .000045) in *CDKAL1*. Although several other variants were associated with risk of early or late conversion of T2DM, they were not significant after Bonferroni correction ($P >$ .05/10/2 = .0025). When risk allele frequencies of early converters and 2-month postpartum NGT/IGT subjects were compared, there were nominally significant differences in variants of *HHEX* (Supplemental Table 2). We were not able to find any differences in risk allele frequencies between late converters and NGT/IGT subjects at more than 1 year follow-up because there was a substantial decrease in the statistical power (Supplemental Table 3).

Independent risk factors for early or late T2DM

Multivariate logistic regression analysis was used to identify independent risk factors for early or late T2DM conversion (Table 4). Clinical and metabolic risk factors

Table 2. Clinical Characteristics and Measures of Diagnostic 100-Gram OGTT During Pregnancy According to the Glucose Tolerance State at 2 Months Post Partum and at More Than 1 Year (Excluding Early T2DM Converters)[a]

	Follow-up at 2 Months Post Partum			Follow-up at More Than 1 Year (Excluding Early T2DM Converters)			
	NGT/IGT	T2DM (Early Converters)	P_1	NGT/IGT	T2DM (Late Converters)	P_2	Early vs Late Converters, P_3
n (%)	738 (87.5)	105 (12.5)		282 (76.2)	88 (23.8)		
Age at pregnancy, y	31.3 ± 3.8	32.1 ± 4.0	.065	31.5 ± 4.0	31.4 ± 3.9	.768	.216
Prepregnancy BMI, kg/m^2	22.7 ± 3.5	24.2 ± 3.8	<.001	22.2 ± 2.9	23.7 ± 3.6	<.001	.313
Pregnancy BMI at OGTT, kg/m^2	27.1 ± 3.3	28.3 ± 3.6	<.001	26.7 ± 2.9	27.6 ± 3.7	.038	.233
Weight gain during pregnancy, kg	11.0 ± 4.4	9.9 ± 4.8	.023	11.4 ± 4.3	10.1 ± 4.3	.011	.852
Gestational age at diagnosis, wk	26.4 ± 3.0	25.2 ± 5.3	.030	26.2 ± 2.8	26.7 ± 4.1	.308	.033
Parity, n	0.48 ± 0.64	0.49 ± 0.68	.913	0.49 ± 0.65	0.47 ± 0.62	.765	.835
Family history of DM, %	39.7	47.6	.132	37.9	50.0	.050	.743
Insulin treatment, %	17.6	59.0	<.001	19.5	40.9	<.001	.012
Glucose, mmol/liter							
0-h	5.0 ± 0.9	6.4 ± 1.7	<.001	5.0 ± 0.7	5.6 ± 1.3	<.001	<.001
1-h	10.8 ± 1.5	12.9 ± 2.7	<.001	10.6 ± 1.5	11.4 ± 1.9	<.001	<.001
2-h	9.8 ± 1.6	12.6 ± 3.2	<.001	9.7 ± 1.4	10.8 ± 2.3	<.001	<.001
3-h	8.0 ± 1.7	10.6 ± 3.3	<.001	8.0 ± 1.5	9.0 ± 2.2	<.001	<.001
AUC of glucose	27.1 ± 3.4	34.0 ± 7.8	<.001	26.8 ± 2.7	29.5 ± 5.3	<.001	<.001
Insulin, pmol/liter[b]							
0-h	76 (56–97)	69 (49–90)	.022	76 (51–97)	76 (56–104)	.119	.042
1-h	472 (306–681)	264 (174–417)	<.001	431 (292–667)	375 (243–514)	.053	<.001
2-h	625 (424–882)	382 (250–500)	<.001	563 (410–854)	542 (326–708)	.042	.002
3-h	486 (347–715)	333 (194–528)	<.001	458 (326–695)	444 (299–681)	.340	.005
AUC of insulin	1431 (1024–1934)	896 (558–1257)	<.001	1340 (953–1861)	1221 (767–1629)	.036	<.001
1-h ΔI/ΔG, pmol/mmol[b]	69 (46–107)	35 (16–56)	<.001	67 (42–107)	50 (34–74)	.011	<.001
Matsuda Index[b]	3.42 (2.51–4.65)	3.48 (2.65–5.63)	.120	3.70 (2.59–5.18)	3.46 (2.63–4.98)	.250	.204
IS/IR disposition index	166 ± 60	98 ± 53	<.001	169 ± 58	131 ± 57	<.001	<.001

[a] Subjects were categorized as NGT/IGT or T2DM group according to the glucose tolerance state at 2 months post partum and at more than 1 year of follow-up using a 75-g OGTT. Antepartum clinical characteristics and measures of glucose and insulin at a 100-g diagnostic OGTT during pregnancy were compared. Data are shown as the mean ± SD in case of normal distribution or otherwise as median (IQR). P_1 indicates comparison between early postpartum NGT/IGT group vs T2DM group; P_2, comparison between more than 1 year postpartum follow-up NGT/IGT group vs T2DM group; and P_3, comparison between early converters and late converters.

[b] These variables were log transformed before statistical analysis.

that showed significant differences between the NGT/IGT group and the T2DM group early or late post partum were included in model 1. Independent risk factors for early T2DM included higher prepregnancy BMI, higher AUC of glucose during antepartum OGTT, lower fasting insulin, and lower IS/IR disposition index. When 2 genetic variants (rs10811661 and rs1111875) that showed significant association for early converters in Table 3 were included, both of the variants did not have significant association (model 2 and model 3). The introduction of these variants reduced the association for all the variables except for earlier diagnosis of GDM and insulin treatment. Regarding late T2DM conversion, independent clinical risk factors were prepregnancy BMI and a higher antepartum AUC of glucose (model 1). When rs7754840 was included (model 2), all the associations were attenuated except for the IS/IR disposition index and insulin treatment.

Discussion

There have been several reports on the incidence of T2DM in women with history of GDM (1, 5, 7, 8, 25, 26). This study is one of the largest prospective cohort studies in Asians to investigate the incidence and risk factors of T2DM in GDM women. The incidence of T2DM after GDM pregnancy varies from 2.6% to as much as 70%, depending on the ethnicity, protocols used for diagnosis of

Table 3. Genetic Risk Factors for Early Converters (n = 69), and Late Converters (n = 70) Compared With Elderly Nondiabetic Controls (n = 632, 345 Women and 287 Men)[a]

				Risk Allele Frequency			Odds Ratio (95% CI)		*P*	
CHR	SNP	Nearby Gene	Risk Allele	Early Converters	Late Converters	Elderly Nondiabetic Controls	Early Converters	Late Converters	Early Converters	Late Converters
3	rs1801282	*PPARG*	C	0.97	0.96	0.95	1.87 (0.67–5.21)	1.51 (0.60–3.81)	.231	.383
3	rs3856806	*PPARG*	C	0.85	0.86	0.82	1.19 (0.73–1.93)	1.28 (0.78–2.10)	.484	.326
3	rs4402960	*IGF2BP2*	T	0.36	0.41	0.30	1.30 (0.90–1.86)	1.64 (1.15–2.35)	.161	.0069
6	rs7754840	*CDKAL1*	C	0.58	0.65	0.46	1.61 (1.12–2.31)	2.17 (1.49–3.14)	.010	**.000045**
6	rs7756992	*CDKAL1*	G	0.62	0.65	0.53	1.47 (1.02–2.13)	1.67 (1.15–2.43)	.040	.0072
8	rs13266634	*SLC30A8*	C	0.65	0.70	0.59	1.33 (0.92–1.93)	1.63 (1.11–2.38)	.129	.013
9	rs564398	*CDKN2A/2B*	T	0.90	0.88	0.87	1.28 (0.73–2.25)	1.11 (0.66–1.89)	.385	.693
9	rs1333040	*CDKN2A/2B*	C	0.34	0.31	0.32	1.12 (0.77–1.63)	0.98 (0.67–1.43)	.548	.922
9	rs10757278	*CDKN2A/2B*	G	0.51	0.49	0.45	1.25 (0.87–1.80)	1.14 (0.80–1.62)	.223	.457
9	rs10811661	*CDKN2A/2B*	T	0.67	0.63	0.51	1.96 (1.35–2.84)	1.60 (1.12–2.29)	**.00043**	.010
10	rs1111875	*HHEX*	C	0.44	0.36	0.30	1.87 (1.29–2.70)	1.31 (0.90–1.91)	**.0010**	.157
10	rs5015480	*HHEX*	C	0.28	0.22	0.19	1.77 (1.18–2.67)	1.25 (0.82–1.93)	.0063	.303
10	rs7923837	*HHEX*	G	0.33	0.25	0.21	1.95 (1.30–2.91)	1.24 (0.82–1.88)	**.0011**	.313
10	rs7903146	*TCF7L2*	T	0.04	0.06	0.02	1.50 (0.56–4.00)	2.66 (1.24–5.69)	.416	.012
10	rs12255372	*TCF7L2*	T	0.01	0.01	0.00	4.62 (0.41–51.59)	4.55 (0.41–50.83)	.214	.219
11	rs2074196	*KCNQ1*	G	0.61	0.62	0.58	1.14 (0.79–1.64)	1.18 (0.82–1.69)	.499	.366
11	rs2237892	*KCNQ1*	C	0.67	0.66	0.61	1.32 (0.90–1.92)	1.25 (0.86–1.81)	.151	.235
11	rs2237895	*KCNQ1*	A	0.71	0.64	0.70	1.06 (0.71–1.56)	0.74 (0.51–1.08)	.784	.119
11	rs5215	*KCNJ11*	G	0.42	0.35	0.38	1.18 (0.84–1.67)	0.90 (0.63–1.27)	.344	.543
11	rs5219	*KCNJ11*	A	0.43	0.35	0.38	1.23 (0.86–1.75)	0.90 (0.63–1.29)	.251	.563
16	rs8050136	*FTO*	A	0.12	0.10	0.12	1.01 (0.59–1.72)	0.80 (0.45–1.42)	.979	.446

Abbreviation: SNP, single-nucleotide polymorphism; CHR, chromosome.

[a] Genotype frequencies of early converters and late converters were compared with those of elderly nondiabetic control subjects using logistic regression analysis assuming additive genotype model. *P* values < the significance threshold after Bonferroni correction (0.05/10/2 = .0025) are in bold.

GDM, follow-up duration, and diagnostic criteria used to define postpartum diabetes (5). In this study, the prevalence of T2DM in the early postpartum period was 12.5%, which is comparable to our previous observation of 15.1% (3). The median time to progression to T2DM was 8.0 years, and the annual incidence was estimated to be 6.8% per year after the early postpartum period. The incidence of T2DM was slightly higher than the finding of Metzger et al (8) who reported an annual incidence of T2DM of 5% to 6% per year and a 5-year cumulative incidence of approximately 50%. This difference could be attributed to the fact that Metzger et al (8) excluded subjects who had fasting hyperglycemia of more than 7.2 mmol/L during GDM pregnancy in their follow-up analysis. Because we did not have data on HbA1c during pregnancy, we were not able to preclude those who had undiagnosed pregestational diabetes or IGT, and this might have caused us to overestimate the incidence of T2DM. Still, it is evident from our study and others that GDM women are at particularly high risk of developing T2DM and that they require special consideration. Moreover, the identification of T2DM after GDM is important to those who want to have subsequent pregnancies, because undiagnosed and untreated hyperglycemia significantly increases risk of congenital anomalies (27).

Various potential risk factors for early postpartum T2DM have been reported (3, 6–8, 26). In this study, there was a significant difference in antepartum BMI between the NGT/IGT and T2DM groups. Obesity has consistently been associated with T2DM after GDM pregnancy (3, 5, 8). It is noteworthy that the mean prepregnancy BMI of early converters was only 24.2 kg/m^2, which reflects the fact that Asian women are more prone to metabolic derangement at a lower BMI threshold (28). Despite the higher degree of hyperglycemia during the antepartum 100-g OGTT, early converters had significantly lower insulin concentrations. The β-cell function estimated by the IS/IR disposition index was significantly decreased in early converters compared with the NGT/IGT group. In addition, this factor was one of the independent risk factors for early postpartum T2DM. Decreased β-cell function has been proposed to be a major determinant of T2DM after GDM pregnancy (4, 8, 29). Our results confirm that β-cell function and its compensation for the increased insulin resistance in pregnancy is a crucial factor determining the postpartum glucose tolerance status in GDM women (30, 31).

Late T2DM converters were also more obese compared with those who had NGT/IGT at more than 1 year follow-up. Similar to early converters, they also had significantly elevated glucose during the antepartum 100-g OGTT. On the other hand, antepartum insulin concentrations were mostly comparable between the NGT/IGT and T2DM groups. Although β-cell function estimated by the IS/IR disposition index was significantly different between the 2 groups, this was not an independent risk factor for late converters. Based on our findings, it could be speculated

Table 4. Independent Risk Factors for T2DM Early or Late Post Partum According to Multivariate Logistic Regression Analysis[a]

	Model 1		Model 2		Model 3	
	OR (95% CI)	*P*	OR (95% CI)	*P*	OR (95% CI)	*P*
T2DM at 2 mo post partum (early converters)						
Prepregnancy BMI	1.08 (1.01–1.16)	**.028**	1.08 (1.00–1.17)	.051	1.09 (1.00–1.18)	.045
Gestational week at diagnosis	0.95 (0.90–1.01)	.092	0.93 (0.86–1.00)	**.047**	0.93 (0.86–1.00)	**.048**
Insulin treatment	1.71 (0.81–3.60)	.159	1.97 (0.83–4.70)	.126	2.06 (0.87–4.87)	.101
Fasting glucose	0.98 (0.96–1.01)	.142	0.98 (0.96–1.01)	.308	0.99 (0.96–1.01)	.317
AUC of glucose	1.01 (1.00–1.01)	**.006**	1.01 (1.00–1.01)	.060	1.01 (1.00–1.01)	.076
Log fasting insulin	0.04 (0.00–0.38)	**.006**	0.05 (0.00–0.89)	**.041**	0.04 (0.00–0.71)	**.028**
Log AUC of insulin	1.40 (0.16–12.47)	.765	1.00 (0.07–13.91)	1.000	1.34 (0.10–18.57)	.825
IS/IR disposition index	0.10 (0.02–0.49)	**.005**	0.15 (0.02–0.99)	**.049**	0.13 (0.02–0.87)	**.036**
rs10811661 (*CDKN2A/2B*)			1.18 (0.76–1.84)	.462		
rs1111875 (*HHEX*)					1.44 (0.93–2.23)	.106
T2DM at more than 1 y of follow-up (late converters)						
Prepregnancy BMI	1.11 (1.02–1.20)	**.013**	1.08 (0.98–1.19)	.114		
Insulin treatment	0.92 (0.40–2.14)	.849	0.76 (0.30–1.92)	.558		
Fasting glucose	1.01 (0.98–1.04)	.622	1.01 (0.97–1.04)	.665		
AUC of glucose	1.01 (1.00–1.01)	**.013**	1.01 (1.00–1.01)	.082		
Log AUC of insulin	0.48 (0.14–1.71)	.260	0.73 (0.17–3.26)	.685		
IS/IR disposition index	0.61 (0.22–1.66)	.329	0.25 (0.06–0.99)	**.049**		
rs7754840 (*CDKAL1*)			1.39 (0.89–2.18)	.147		

Abbreviation: OR, odds ratio.

[a] Variables that showed statistically significant difference between the NGT/IGT and T2DM groups (Table 2) were selected for multivariate logistic regression analysis. If variables had significant colinearity, such as prepregnancy BMI and pregnancy BMI, those that had better clinical relevance were included in the model. *P* values less than .05 are in bold.

that late conversion to T2DM is more closely related to the degree of obesity than to β-cell function.

When early and late converters were directly compared, both groups had similar degrees of obesity but significantly different β-cell function. Early converters had more severe β-cell dysfunction and were more likely to receive insulin treatment during pregnancy. This difference in β-cell function might have resulted in conversion to T2DM at different times. Further studies are warranted to determine the relative importance of obesity and β-cell dysfunction in the development of T2DM after GDM pregnancy.

In this study, we found that several genetic variants known to be associated with T2DM at the genome-wide significance level were also associated with early or late conversion to T2DM after GDM pregnancy. The *HHEX* (hematopoietically expressed homeobox) gene encodes a transcription factor that is involved in ventral pancreas development (32, 33). The *CDKN2A* and *CDKN2B* genes encode $p16^{INK4a}$ and $p15^{INK4b}$, respectively, both of which regulate β-cell replication (34, 35). Variants in *HHEX* and *CDKN2A/2B* have been associated with decreased 30-minute insulin secretion after glucose challenge and decreased β-cell glucose sensitivity (36). In addition, risk alleles of *HHEX* have been most strongly associated with decreased AUC of insulin during antepartum 100-g OGTT in GDM women (22). These data imply that the decreased β-cell function in early converters might be due to genetic predisposition conferred by these variants in *HHEX*, at least in part. The variant that was significantly associated with late conversion to T2DM was located in *CDKAL1*. The *CDKAL1* gene encodes cyclin-dependent kinase 5 regulatory subunit-associated protein 1-like 1. This variant is also well known for its association with T2DM and decreased insulin secretion (37). Additionally, a recent large-scale GWA study showed that a variant in this gene is significantly associated with BMI in East Asians (38). Interestingly, that study associated the *CDKAL1* variant with lower BMI, lower insulin concentration, and increased risk of T2DM, suggesting opposite directions of association for obesity and T2DM. It might be possible that the risk of T2DM in late converters conferred by this variant is modulated through an interaction with obesity.

There are certain limitations to this study. First, approximately 50% of our subjects did not undergo long-term follow-up. In addition, those who underwent follow-up for more than 1 year were more likely to have higher glucose concentration and decreased β-cell function during pregnancy. Considering this, it could be possible that our estimation of the late T2DM conversion rate was overestimated. In comparing early converters and late

converters, the contrast between the 2 groups might have been obscured because more severe cases might have been preferentially skewed to the late converters. Second, the OGTT-derived 1-hour ΔI/ΔG index and IS/IR disposition index are not validated measures of pancreatic β-cell function. Among the GDM women included in this study, 56 subjects underwent a frequently sampled iv glucose tolerance test (FSIVGTT) at 1 year post partum (39). Therefore, we were able to compare the 1-hour ΔI/ΔG index with the FSIVGTT-derived acute insulin response. The 1-hour ΔI/ΔG index had a significant positive correlation with acute insulin response (Pearson's coefficient 0.463, $P < .001$). We also compared the IS/IR disposition index with the FSIVGTT-derived disposition index and found a significant positive correlation (Pearson's coefficient 0.422, $P < .001$). Therefore, we suggest that both the 1-hour ΔI/ΔG index and the IS/IR disposition index could be reasonable estimates of β-cell function when more complex measures are not available. Third, in comparing genetic risk factors, it could be argued that the control group should be NGT/IGT women at the corresponding time, not the elderly nondiabetic subjects. However, a large proportion of NGT/IGT subjects in the early postpartum period will develop T2DM later. Therefore, a significant ascertainment bias would have been present. This would also have been true for the NGT/IGT group at more than 1 year follow-up. The comparison made between early or late converters and elderly nondiabetic controls could be regarded as comparison of gene pools in subjects with high risk of T2DM and a very low risk of T2DM. Some of our elderly nondiabetic control subjects (13.3%) had fasting glucose (from 5.6–6.1 mmol/L) and HbA1c level (5.7%) in the range of IGT, which might have limited our ability to find positive genetic associations.

In conclusion, a significant portion of GDM subjects progress to T2DM in the early postpartum period and continue to have a high incidence of T2DM at later periods. This strongly supports the current recommendation that GDM women be tested for their glucose tolerance status at 2 months post partum and annually thereafter. Early T2DM converters were more obese and had significantly decreased β-cell function, which could be partly explained by genetic susceptibility. In late converters, antepartum obesity and hyperglycemia were the independent predictors of T2DM. Further investigations of genetic and environmental risk factors should develop useful prediction models of T2DM in previously GDM women.

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