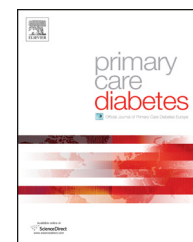




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Original research

HbA1c as a predictor of diabetes after gestational diabetes mellitus



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ABSTRACT

Aim: We wanted to investigate third-trimester HbA1c as a predictor of diabetes after gestational diabetes mellitus (GDM).

Methods: Women with GDM were followed up prospectively for five years from pregnancy to detect the development of diabetes. The ability of HbA1c to predict diabetes was evaluated with receiver-operating characteristic (ROC) curves and logistic regression analysis.

Results: By five years, 73 of 196 women had been diagnosed with diabetes. An optimal cut-off point for HbA1c of 36 mmol/mol (5.4%) could predict diabetes with 45% sensitivity and 92% specificity. For HbA1c ≥ 39 mmol/mol ($\geq 5.7\%$), sensitivity, specificity, and positive predictive value were 30%, 97%, and 91%, respectively. In logistic regression analysis, adjusting for the diagnostic glucose concentration during pregnancy, HbA1c levels in the upper quartile (≥ 36 mmol/mol) were associated with a 5.5-fold increased risk of diabetes.

Conclusion: Third-trimester HbA1c levels in the pre-diabetes range revealed women with post-partum diabetes with high specificity and high positive predictive value. HbA1c testing could be used as a strategy to select high-risk women for lifestyle interventions aimed at prevention of diabetes starting during pregnancy. The results should encourage further validation in other populations using new diagnostic criteria for GDM.

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Abbreviations: AUC, area under the curve; BMI, body mass index; CI, confidence interval; EASD, European Association for the Study of Diabetes; GDM, gestational diabetes mellitus; IFG, impaired fasting glucose; IGT, impaired glucose tolerance; NGT, normal glucose tolerance; NPV, negative predictive value; OGTT, oral glucose tolerance test; OR, odds ratio; PPV, positive predictive value; ROC, receiver-operating characteristic; SD, standard deviation; WHO, World Health Organization.

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1. Introduction

Post-partum follow-up of pregnancies with gestational diabetes mellitus (GDM) is important, as these women have a several-fold increased risk of progression to type-2 diabetes after delivery [1]. It has been shown that there is a beneficial effect of lifestyle intervention on the 10-year risk of diabetes in women with a history of GDM [2]. However, the uptake of post-partum screening after GDM is suboptimal, and women fail to attend the post-partum visit, even in a research setting [3–6]. An easy way of identifying those who are at highest risk of developing diabetes after pregnancy is needed, so that midwives and physicians can pay more attention to these women and start intervention already in pregnancy when the women are more likely to be highly motivated.

HbA1c analysis was recently endorsed as a screening test for unrecognized diabetes in early pregnancy [7–9], but it has not yet been advocated as a diagnostic test for GDM. There is some interest in finding an HbA1c threshold at other stages of pregnancy that could even be used for intervention during pregnancy. HbA1c as a diagnostic test has advantages for both patients and physicians. It can be performed without fasting, and is more reproducible and less cumbersome than an oral glucose tolerance test (OGTT) [10]. However, we and others have found a low sensitivity of HbA1c testing relative to an OGTT in diagnosing diabetes and pre-diabetes in women who have previously had GDM [5,11–16]. Very few studies have evaluated the clinical usefulness of third-trimester HbA1c levels as a way of predicting the development of post-partum diabetes [17–21]. In a previous study from our geographical area, we found that 30% of the women with GDM in the study cohort had already developed diabetes five years after delivery, and that HbA1c levels ≥ 38 mmol/mol ($\geq 5.6\%$) at the diagnostic OGTT during pregnancy, corresponding to the upper quartile, were associated with a four-fold increased risk of developing diabetes [17]. The aim of the present study was to investigate the HbA1c level measured close to the twenty-eighth week of pregnancy as a predictor of diabetes development up to five years after pregnancy.

2. Material and methods

2.1. Participants

The prospective Mamma Study followed women in southern Sweden who gave birth during the years 2003–2005, for up to 5 years from delivery, to detect the development of post-partum diabetes. A detailed description of the study design has already been reported [6]. Briefly, pregnant women, representing different glucose categories according to an OGTT, were invited to take part in the study. A 75-g OGTT was offered to all women in the twenty-eighth week of gestation, excluding those who were diagnosed with diabetes before pregnancy. The diagnostic criteria for GDM were a slight modification of the European Association for the Study of Diabetes (EASD) criteria, defining GDM as a 2-h capillary blood glucose concentration of ≥ 9.0 mmol/l [22], corresponding to a plasma glucose concentration of ≥ 10.0 mmol/l [23]. Based on this definition,

391 women were recruited. HbA1c was measured within two weeks of the diagnosis of GDM. Participants were followed for the development of diabetes by means of an OGTT at 1–2 years and at 5 years after pregnancy—or until the diagnosis of diabetes. Based on the stated country of origin of at least three grandparents, women were grouped according to whether they were of European or non-European origin. Diagnostic criteria during follow-up were those proposed by the World Health Organization (WHO) 1999 [24]. According to the results of the OGTT, women were classified as having normal glucose tolerance (NGT), impaired fasting glucose (IFG), impaired glucose tolerance (IGT), or diabetes. Participants gave written informed consent and the Ethics Committee of Lund University approved the study (LU 259-00), which was performed according to the Declaration of Helsinki.

2.2. Metabolic measurements

The HemoCue Glucose 201+ system (HemoCue AB, Ängelholm, Sweden) was used for immediate measurement of plasma glucose concentrations (mmol/l). HbA1c was measured with ion-exchange chromatography procedures (Variant II from BioRad; Tosoh G7 from Tosoh Bioscience; and in-house Mono S) with results that were traceable to the Mono S procedure at the Swedish Reference Laboratory. Values given in % (Mono S) were converted to NGSP units (%) and IFCC units (mmol/mol) using the regression equations developed by the IFCC Working Group [25].

2.3. Statistical analysis

HbA1c values are given as mmol/mol with % NGSP units in parentheses or brackets. Continuous variables are summarized by means and standard deviations (SDs) or 95% confidence intervals (CIs). Differences between group means were compared with analysis of variance (ANOVA). Logistic regression analysis was used to calculate the odds ratios (ORs) and 95% CI for 5-year diabetes risk in different quartiles of HbA1c levels. A receiver-operating characteristic (ROC) curve was plotted to evaluate the diagnostic performance of HbA1c in diabetes prediction. Sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV), and the area under the curve (AUC) were calculated. Threshold for discrimination was calculated with the Youden index [26]. IBM SPSS Statistics 22.0 for Windows (IBM Corporation, Armonk, NY, USA) was used for analysis. Two-sided *p*-values of less than 0.05 were considered to be statistically significant.

3. Results

Of the 391 women who agreed to participate prospectively, 5-year data were available for 196 of them. Among these, 73% were of European origin (mostly Swedish) and 27% were of non-European origin (with Arab and Asian origin being the largest groups).

Mean values for maternal age, diagnostic 2-h plasma glucose concentration, and HbA1c level during pregnancy in participants were 33.3 (SD 4.9) years, 11.1 (1.7) mmol/l, and 33.1 (7.1) mmol/mol [5.2% (1.1%)], respectively. The correspond-

Table 1 – Diagnostic indices of various HbA1c thresholds to predict diabetes five years after pregnancy using normal glucose tolerance at 5-year follow-up as a reference.

HbA1c cut-off	n ^a	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)
≥48 mmol/mol (≥6.5%)	10	13.7	100.0	100.0	48.8
≥45 mmol/mol (≥6.3%)	12	16.4	100.0	100.0	49.6
≥42 mmol/mol (≥6.0%)	15	19.2	98.3	93.3	50.0
≥39 mmol/mol (≥5.7%)	24	30.1	96.7	91.2	53.2
≥36 mmol/mol (≥5.4%)	38	45.2	91.7	86.8	57.8
≥32 mmol/mol (≥5.1%)	75	71.2	61.7	69.3	63.8

PPV, positive predictive value; NPV, negative predictive value.

^a Number of women who reached the threshold value.

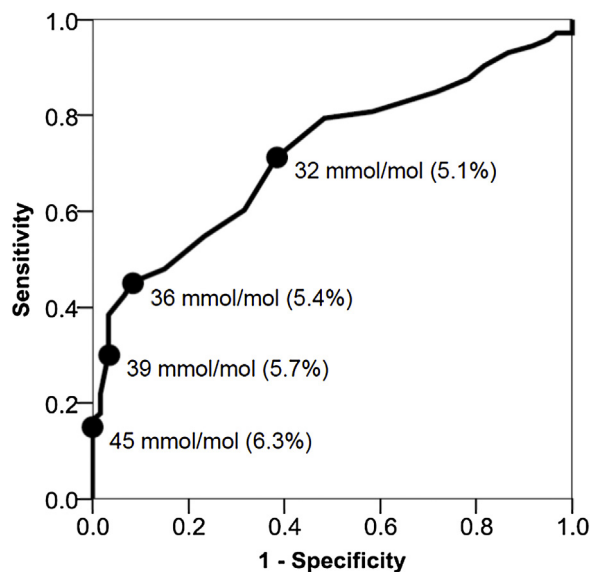


Fig. 1 – Predictive accuracy of HbA1c in detecting diabetes five years after gestational diabetes, using women with normal glucose tolerance as a reference. Various cut-off points are shown.

ing figures for non-participants were 32.4 (5.8) years, 11.0 (1.1) mmol/l, and 32.7 (5.8) mmol/mol [5.1% (0.9%)], and the differences compared to participants were not significant. After five years, 73 women had been diagnosed with diabetes: 14 before the first follow-up, 25 at the first (1- to 2-year) follow-up, 13 between the first follow-up and the final (5-year) follow-up, and 21 at the final follow-up. Of the remaining 123 women who participated in the 5-year follow-up (out of a total of 144), 60 were classified as having NGT and 63 were classified as having IFG/IGT (pre-diabetes).

The mean HbA1c level during pregnancy in women who had developed diabetes after 5 years was 36.7 (95% CI: 34.5–38.8) mmol/mol [5.5% (5.3–5.7%)], as compared to 31.4 (30.4–32.4) mmol/mol [5.0% (4.9–5.1%)] in women with pre-diabetes and 30.6 (29.5–31.7) mmol/mol [4.9% (4.8–5.1%)] in women with NGT at 5 years ($p < 0.0001$).

Using NGT at 5-year follow-up as a reference, an ROC curve was constructed to evaluate HbA1c as a predictor of diabetes up to five years after pregnancy (Fig. 1). The ability of the ROC curve to predict diabetes was fair (AUC=0.720, 95% CI: 0.634–0.806, $p < 0.0001$), with an optimal cut-off point of 36 mmol/mol (5.4%), resulting in a sensitivity of 45% and a

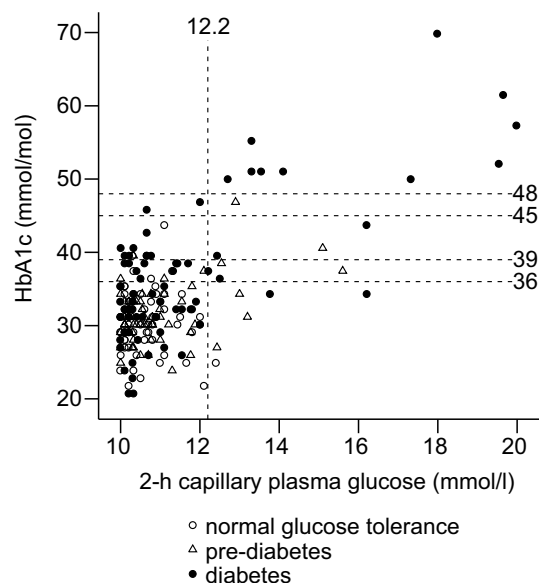


Fig. 2 – HbA1c levels plotted against the diagnostic 2-h glucose concentration during pregnancy for 196 women with gestational diabetes. Various diagnostic cut-off levels are shown, and the diagnoses at the 5-year follow-up are indicated by symbols.

specificity of 92%. Table 1 shows the sensitivity, specificity, PPV, and NPV for various cut-offs. Overall, HbA1c showed high specificity and PPV, but the sensitivity was low. The prediction did not improve by using both NGT and IFG/IGT at 5-year follow-up as a reference (AUC=0.710, 95% CI: 0.630–0.791, $p < 0.0001$). Similar results were obtained when we included women of Nordic origin only (diabetes, $n = 23$ vs. NGT, $n = 44$; AUC=0.734, 95% CI: 0.588–0.879, $p = 0.002$).

In Fig. 2, HbA1c levels are plotted against the diagnostic 2-h capillary plasma glucose concentrations during pregnancy for the whole study group. After five years, all ten women with HbA1c levels ≥ 48 mmol/mol ($\geq 6.5\%$) had been diagnosed with diabetes, six women before the first follow-up (HbA1c 51–70 mmol/mol [6.8–8.6%]), one woman at the first follow-up (HbA1c 57 mmol/mol [7.4%]), and three women at the five-year follow-up (HbA1c 50–55 mmol/mol [6.7–7.2%]). Similarly, in 13 women with HbA1c levels ≥ 45 mmol/mol ($\geq 6.3\%$) all but 1 woman (IGT) had been diagnosed with diabetes after five years. Altogether, five out of 27 women with HbA1c levels ≥ 39 mmol/mol ($\geq 5.7\%$) had not been diagnosed with diabetes

after five years (2 NGT, 1 IFG, and 2 IGT). The corresponding figure for women with 2-h capillary plasma glucose levels ≥ 12.2 mmol/l (the diagnostic limit for diabetes outside of pregnancy) was eight out of 24 (1 NGT, 3 IFG, and 4 IGT).

HbA1c levels for the total study group were grouped into quartiles. Median levels for HbA1c in mmol/mol [%] in the respective quartiles were: 27 (range: 21–29) [4.6% (range: 4.1–4.8%)] ($n = 56$), 31 (30–31) [5.0% (4.9–5.0%)] ($n = 43$), 33 (32–35) [5.2% (5.1–5.4%)] ($n = 51$), and 40 (36–70) [5.8% (5.4–8.6%)] ($n = 46$). A logistic regression analysis, testing the predictive value of HbA1c quartiles for the 5-year diabetes risk, showed that women with HbA1c levels in quartile four had a seven-fold increased risk of post-partum diabetes compared to women with HbA1c levels in quartiles 1–3 (OR = 7.0, 95% CI: 3.3–14.6, $p < 0.0001$). This association remained significant after adjustment for maternal age and the 2-h glucose level during pregnancy (OR = 5.5, 95% CI: 2.5–12.1, $p < 0.0001$).

4. Discussion

The results of the present study confirm our previous findings that HbA1c levels in the upper quartile, measured close to the diagnostic OGTT during pregnancy, predict diabetes development during the five years after delivery [17]. To the best of our knowledge, only four other studies have investigated an association between HbA1c levels during pregnancy and the risk of post-partum diabetes [18–21].

Using the WHO (1999) criteria for the diagnosis of GDM, Liu et al. evaluated HbA1c, measured at 26–30 gestational weeks, as a predictor of diabetes 1–5 years after delivery in 1263 Chinese women [18]. Adjusting for various risk factors in a Cox proportional hazards model, the hazard ratio for post-partum diabetes was 2.11 (95% CI: 1.50–2.97) for every unit (%) increase in HbA1c. Furthermore, when fasting glucose, 2-h glucose, and HbA1c level during pregnancy were entered into the model simultaneously, 2-h glucose and HbA1c level, but not fasting glucose, remained significant and positive predictors of post-partum diabetes. In our previous study, both HbA1c and the fasting glucose level during pregnancy were found to be independent predictors of the 5-year diabetes risk [17]. A number of risk factors for diabetes development after GDM have been identified, which may in part differ from one population to another [27]. The fact that the fasting glucose levels did not predict diabetes in the study by Hsu et al. may be specific to the Chinese population, as fasting plasma glucose has been reported to be less sensitive for diagnosis of diabetes than the 2-h glucose level in the Asian population [28].

In a retrospective study from Korea, evaluating HbA1c at 26–30 gestational weeks as a diagnostic test for GDM, follow-up data for at least 3 months after pregnancy were available for 54 of 321 women [19]. Based on ROC-curve analysis, an optimal cut-off value for HbA1c of 37 mmol/mol (5.5%) could predict diabetes with 79% sensitivity and 73% specificity. However, the restricted number of women included in the analysis made the results less reliable. Furthermore, long-term follow-up data were not available. In another study from Warsaw, Poland, Malinowska-Polubiec et al. evaluated various risk factors for diabetes 0.5–10 years after pregnancy in 150 women with a history of GDM [20]. In that population both second-trimester and

third-trimester HbA1c were associated with increased relative risks of post-partum diabetes. Finally, in a retrospective study of 305 women in the Czech Republic, Bartakova et al. found an optimal cut-off value from ROC-curve analysis (based on Youden statistics) for mid-trimester HbA1c of 36 mmol/mol (5.4%) for any degree of post-partum glucose abnormality during the first year after pregnancy [21].

In addition to the 2-h plasma glucose concentration during pregnancy, we have recently reported that (1) BMI at the first follow-up after pregnancy and (2) a non-European background were the most important risk factors for development of diabetes five years after pregnancy in the total Mamma Study cohort (defining GDM by the WHO (1999) criteria) [29]. However, HbA1c was not included in the prediction model since it was only measured in women diagnosed with GDM according to clinical routine (EASD criteria). For this reason, we performed a separate study restricted to these women.

At the time of the design of the Mamma Study, HbA1c was not recommended as a diagnostic test for diabetes, nor as a test early in pregnancy to rule out pre-existing diabetes. In our material, an HbA1c level of ≥ 48 mmol/mol ($\geq 6.5\%$) during the third trimester of pregnancy identified all women with a diabetes diagnosis five years after pregnancy, some of whom had been diagnosed with diabetes before the first follow-up and may have had pre-gestational diabetes. Furthermore, an HbA1c level of ≥ 45 mmol/mol ($\geq 6.3\%$) identified all but 1 woman with diabetes after five years, and an HbA1c level ≥ 39 mmol/mol ($\geq 5.7\%$) identified all but 5 women with a diabetes diagnosis during follow-up. On the other hand, for the various thresholds, HbA1c had low sensitivity in diagnosing diabetes using either NGT or NGT/IFT/IGT as a reference. These data provide evidence to suggest there may be a useful HbA1c threshold above which all women should be closely monitored, starting already during pregnancy, to prevent diabetes development after delivery. Furthermore, after adjustment for the 2-h glucose level, HbA1c levels equal to and above the optimal cut-off level of the ROC curve were associated with more than a 5-fold increased risk of post-partum diabetes. This indicates that HbA1c analysis could be an adjunct to the OGTT in identifying women who are at high risk of developing post-partum diabetes.

A major limitation of the study was that HbA1c measurements were only available for women with a clinical diagnosis of GDM. Moreover, as previously reported [6], the overall participation rate in the Mamma Study was less than 50%, which may have introduced selection bias. As the diagnostic criteria for GDM have changed worldwide since the time when this study was initiated, the findings should be repeated and validated in future studies using updated diagnostic criteria [7–9]. Generalization to populations with different ethnic backgrounds should also be done. The strengths of the study were the uniform screening procedure in the region, based on a universally applied OGTT, and the prospective study design with long-term follow-up after pregnancy.

5. Conclusions

An HbA1c level of ≥ 36 mmol/mol ($\geq 5.4\%$), obtained close to the twenty-eighth week of pregnancy, was associated with a

more than 5-fold increased risk of diabetes five years after pregnancy. A cut-off level for HbA1c of ≥ 39 mmol/mol ($\geq 5.7\%$), corresponding to the pre-diabetes range outside of pregnancy, could reveal women with post-partum diabetes with high specificity (97%) and high PPV (91%). Due to the low sensitivity, HbA1c does not appear suitable as a screening test to predict diabetes after GDM in all women, but it could be used as a strategy for selecting high-risk women for lifestyle interventions to prevent diabetes, starting already in pregnancy. The results should encourage further validation in large-scale studies using new diagnostic criteria for GDM.

Conflict of interest

The authors state that they have no conflict of interest.

Authors' contributions

KB conceived the study. All the authors contributed to the study design and interpretation of data. RC wrote the initial draft of the manuscript, and KB contributed to the next draft. CI and NS critically reviewed and edited the manuscript. All the authors approved the final version.

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