Red Blood Cells in The First Trimester and The Risk of Gestational Diabetes: A Prospective Cohort Study

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Research Article
Abstract

**Background:** This research aimed to assess the potential association of gestational diabetes (GDM) with early trimester hematological parameters including hemoglobin (Hb), red blood cell (RBC), white blood cell (WBC), and platelet count (PLT) through a prospective cohort study.

**Methods:** The prospective cohort included pregnant women subjected to prenatal examination at Shantou and Beijing hospitals in China from March 2014 to December 2015. Data were collected since the first perinatal visit in obstetrics clinics, and then participants were followed up at 24, 32, 36 gestational weeks and the time of delivery, respectively. Multivariable adjusted logistic regression models were conducted to estimate odds ratio (OR) and its corresponding 95% confidence interval (95% CI).

**Results:** A total of 1004 pregnant women with singletons, less than 12 gestational weeks, and without history of chronic disease were eligible for analysis. The incidence of GDM was 18.82%, and the mean age was 29.50 ± 3.84 years. Total of 187 (18.63%) women who had abnormal RBC level and 222 (22.11%) had abnormal Hb in the first trimester of pregnancy. After multivariable adjustment, each unit increment in numeric RBC or Hb was associated with 177% and 4% increased risk for GDM. The risk for GDM was significantly increased with higher RBC (OR: 2.00 for RBC>4.55×10^{12} /L) and Hb (OR: 2.14 for Hb>139 g/L) levels in the first trimester.

**Conclusions:** Elevated RBC and Hb in the first trimester are associated with increasing risk of GDM. Further evidence are warranted to confirm these possible causal relationships.

Introduction

Gestational diabetes (GDM) is a condition diagnosed for the first-time during pregnancy, and its prevalence ranges from 1.8–24.5% worldwide [1]. As a high-risk disorder during pregnancy, GDM increases the risk of adverse pregnancy outcomes in the perinatal period [2] and is associated with numerous future morbidities for both mothers and fetus in long term [3–5]. However, no global consensus on the optimal management of GDM and its complications exists, except for lifestyle intervention and insulin therapy, which have limited effectiveness on reducing health damages caused by hyperglycemia [6]. Additional primary or secondary prevention are required to identify pregnancy at risk as early as possible and then taking active measures to reduce GDM incidence and improve outcomes for the mother and baby [7, 8].

The first trimester of pregnancy is a critical period to identify pregnant women at high risk of GDM and reduce adverse intrauterine exposures throughout pregnancy. Many studies have demonstrated that elevated hemoglobin (Hb) and hemoglobin A1c (HbA1c) in the first trimester remarkably increased the morbidity of GDM and can be used as biomarkers either alone or in conjunction with other maternal risk factors to predict the risk of GDM [9–12]. Hb is the specific protein molecule in red blood cell (RBC) carrying almost all of oxygen molecules and is closely related to the number of RBC [13]. Previous studies have confirmed that RBC count is significantly associated with insulin resistance [14], β-cell dysfunction [15], and the incidence of type 2 diabetes [16], which suggest that peripheral RBC count may also play a role in GDM development. However, few studies have examined the relationship between RBC count in early pregnancy and the risk of GDM. In this study, we aimed to explore the potential association of early trimester maternal hematological parameters including Hb, RBC, white blood cell (WBC), and platelet count (PLT) with GDM through a prospective cohort study.

Methods

**Study design and population**

The hospital-based prospective cohort study was designed and conducted among pregnant women who were registered in the obstetric archives and completed their first perinatal visit in the obstetrics clinic of the First Affiliated Hospital of Shantou University Medical College and Beijing Friendship Hospital of Capital Medical University from March 2014 to December 2015. The purpose of this cohort was to assess the association between increased hematological parameters in early pregnancy and GDM incidence. All participants had provided written informed consent before participation. This study was designed and
conducted in accordance with the Declaration of Helsinki (2000) of the World Medical Association and approved by the Institutional Research Review Board of the National Research Institute for Family Planning, Beijing, China.

Pregnant women aged 20–49 years old, of Han nationality, with singletons, registered in the obstetric archives of the two hospitals, and living in local areas for more than half a year without tendency to move out during pregnancy were enrolled at the baseline of this cohort study. Participants were excluded if they were more than 12 gestational weeks, have missing values of hematological parameters, or had a history of chronic diseases (including diabetes, hypertension, or chronic nephritis) at the baseline. After the baseline interviews, all participants were followed up at 24, 32, 36 gestational weeks and the time of delivery, respectively. The flowchart of the recruited population is provided in Fig. 1.

Data collection

Baseline information about demographic characteristics, medical history of diseases, and lifestyle behaviors of each participant were collected using a structured questionnaire by trained staff through face-to-face interviews. Demographic characteristics included age, educational level, occupation, pre-pregnancy weight, height, and monthly income. Medical history of disease regarded history of GDM, hypertension, adverse pregnancy outcomes, anemia, thyroid disease, and infection of hepatitis B virus. Family history of diseases included diabetes mellitus, hypertension, and GDM. Medication history covered the use of iron contained health products, oral contraceptives, antibiotics, and painkillers. Lifestyle behaviors included activities of smoking, drinking, passive smoking, psychological pressure, and transportation to work. All questionnaires were reviewed and entered independently by two persons under the settings for logical error correction. Physical examinations, previous obstetric history, laboratory parameters, and diagnoses were extracted from the medical records. Information of physical examinations contained height, weight, waist circumference, and blood pressures. Laboratory parameters recorded indicators of blood tests, liver function tests, renal function tests, fasting blood glucose tests, and Hepatitis B antigen tests. All these laboratory indicators were tested from fasting blood samples which collected and immediately stored at 4–8°C in 24 h.

Follow-up information in prenatal medical care tests about maternal and neonatal health status were all documented. Recorded information included GDM diagnosis, lifestyle behaviors during pregnancy (smoking, passive smoking, drinking, and diet), gestational weight, uterine height, abdominal circumference, and blood pressures.

Definitions

Age was classified as < 35 and ≥ 35 years old; education level was categorized as below college and equal or above college; occupation was sorted as farmers and workers; monthly income was grouped as RMB ≤ 5000 and RMB > 5000. Pre-pregnancy body mass index (BMI) was calculated as weight in kilograms divided by the square of height in meters. Participants with BMI < 18.5, 18.5–23.9, 24–27.9, and > 28.0 kg/m² at pre-pregnancy were regarded as underweight, normal, overweight, and obese, respectively, according to the guidelines for Chinese adults [17]. Gestational weeks were further categorized as ≤ 8 and 8–12 groups. Weight gain in the first trimester was calculated by weight before 12 gestational weeks minus weight in pre-pregnancy. Weight gain in the second trimester was calculated by weight in 24 gestational weeks minus weight in pre-pregnancy. The information about whether used iron contained health products, such as Elevit or Kingsley, were also collected. Early pregnancy fasting glucose was divided into < 6.1mmol/L and ≥ 6.1 mmol/L levels. Self-reported dietary taste was categorized as light, moderate, and salty. Work transportation was grouped as non-working, driving, taking subway, and riding bicycle.

As recommended by Abbassi [18], the normal range of RBC count, Hb, WBC, and PLT in the first trimester were defined as 3.42–4.55 (×10¹² /L), 116–139 (g/L), 5.7–13.6 (×10⁹ /L), and 174–391 (×10⁹ /L), respectively. Then, all categorized hematological parameters in the first trimester were classified into two levels (Class I), namely, normal RBC (3.42–4.55×10¹² /L) and abnormal RBC (< 3.42 or > 4.55×10¹² /L); normal Hb (116–139 g/L) and abnormal Hb (< 116 or > 139 g/L); normal WBC (5.7–13.6×10⁹ /L) and abnormal WBC (< 5.7 or > 13.6×10⁹ /L); normal PLT (174–391×10⁹ /L) and abnormal PLT (< 174 or > 391×10⁹ /L). Successively, all categorized hematological parameters in the first trimester were classified into three levels (Class II), RBC count was categorized as < 3.42 (×10¹² /L), 3.42–4.55 (×10¹² /L), and > 4.55 (×10¹² /L) groups; Hb concentration was stratified into < 116 (g/L), 116–139 (g/L), and > 139 (g/L) levels; WBC count was classified as < 5.7 (×10⁹ /L), 5.7–13.6 (×10⁹ /L), and ≥ 13.6 (×10⁹ /L) groups.
/L), and > 13.6 (×10⁹ /L) groups; and PLT count was grouped into < 174 (×10⁹ /L), 174–391 (×10⁹ /L), and > 391 (×10⁹ /L) levels.

GDM was diagnosed if at least one value of plasma glucose concentration was equal to or exceeded the thresholds of 5.1, 10.0, and 8.5 mmol/L for fasting, 1 h, and 2 h post-glucose load values, respectively, after performing a 75 g oral glucose tolerance test (OGTT) at gestational 24–28 weeks according to the Guidelines for Diagnosis and Treatment of Diabetes in Pregnancy (2014) in China [19].

Statistical analysis

Kolmogorov–Smirnov test was used to determine the normality of hematological parameters, and baseline characteristics were described by median and interquartile range. Mann–Whitney U test or Kruskal-Wallis test was conducted to evaluate the differences in hematological parameters among participants with categorized basic characteristics. An independent sample t-test or χ² test was also performed to evaluate the difference in baseline characteristics between GDM and non-GDM cases. The changes of RBC and Hb in the first and the second trimester were also described and the differences of GDM incidence within RBC or Hb levels were examined by χ² test. Afterwards, the association (odds ratio [OR] and corresponding 95% confidence interval [95%CI]) between continuous hematological parameters at the first or the second trimester and the risk of GDM were assessed using age-adjusted and stepwise multivariable-adjusted logistic regression models, respectively. To further evaluate the association between categorized hematology parameters in the first trimester and the incidence of GDM, numeric hematological parameters were grouped into two classes, and each class used two multivariable-adjusted logistic regression models. Covariates in the multivariable-adjusted model I included age, gestational weeks, BMI before pregnancy, weight gain in the first trimester, dietary taste, iron contained health product consumption, passive smoking status, transportation to work, fasting glucose, history of GDM, and family history of GDM at the baseline. In model II, weight gain in the first trimester was replaced with weight gain in the second trimester based on model I. All analyses were used with R software 3.5.0, and two-sided p < 0.05 values were considered statistically significant.

Results

Overall, 1326 pregnant women were enrolled at the baseline and completed delivery from March 2014 to December 2015 in the two research sites. Finally, a total of 1004 women were eligible for our analysis. The flow chart of recruited participants is presented in Figure 1. The mean age of participants was 29.50 ± 3.84 years, and the median of gestational weeks was 10. Specifically, there were 187 (18.63%) women who had abnormal RBC level, 222 (22.11%) had abnormal Hb, 70 (7.00%) had abnormal WBC, and 72 (7.17%) had abnormal PLT at the first trimester of pregnancy. Participants with higher hematological parameters, including higher RBC, Hb, WBC and PLT, were more likely to be urban workers, higher monthly income, and higher BMI; they also made their first prenatal-care visit at very early gestational weeks (P<0.05; Table 1).

A total of 871 women continued to test hematological parameters at the 24 gestational weeks in the follow-up study. Among them, 812 and 803 women whose RBC and Hb in 24 gestational weeks were decreased, and only 59 and 68 women whose RBC and Hb in 24 gestational weeks were equal to or increased than that of the first trimester. There was no statistical difference between the proportion of GDM cases among women with equal or increased RBC or Hb and that of women with decreased RBC or Hb at the second trimester (P= 0.591 and P= 0.152) (data not shown).

From baseline until delivery, total of 189 (18.82%) women were diagnosed as GDM. Compared with non-GDM women, GDM patients had higher RBC, Hb, and WBC levels at the first trimester (P<0.001, P<0.001, and P=0.001, respectively), and higher RBC and Hb levels at the second trimester (P<0.001 and P<0.001) (Table 2). Results from multivariable adjusted logistic regressions revealed that each unit increment of RBC mass or Hb at the first trimester was associated with 177% and 4% increased risk for GDM, and that each unit increment of RBC mass or Hb at the second trimester was associated with 187% and 4% increased risk for GDM, respectively (Table 2).
Furthermore, categorized variables showed that women who had abnormal RBC count or Hb concentration in the first trimester have higher risks for GDM after multivariable adjustment compared with reference group (Table 3). Specifically, the risks for GDM were significantly increased with elevated RBC (OR: 2.00, 95% CI: 1.33–3.03) and Hb levels (OR: 2.14, 95% CI: 1.36–3.36) (Table 2). However, no association was found between WBC and PLT count in early pregnancy and the risk of GDM. After replacing weight gain in the first trimester with weight gain in the second trimester in Model II, the results remained stable (Table 3).

Discussion

To our knowledge, our research is the first prospective cohort study to demonstrate the association between RBC in the first trimester and the risk of GDM. We found that increased RBC and Hb levels in the first trimester significantly increased the risk of GDM, providing clinical evidence that elevated RBC count in early pregnancy could be a novel risk factor and biomarker of GDM in addition to Hb concentration.

Previous studies have demonstrated that patients with GDM had higher RBC count than controls at the middle-term [20] and late period of pregnancy [21]. Only one study has explored the association between RBC count at the first prenatal-care visit and the risk of GDM, in which found that RBC was associated with GDM in univariable analysis [22]. Therefore, our study is the first to demonstrate that RBC count (numeric and categorized forms) in the early trimester was remarkably associated with the incidence of GDM after adjusting for multiple clinical risk factors including fasting glucose, history of GDM, weight gain in the first or the second trimester and whether taking iron contained health product. Weight gain was confirmed to be an independent risk factor for GDM [23], especially increasing in early pregnancy[24]. After adjusting the weight gain at the first or the second trimester, we still found an increased risk for GDM among women who have elevated RBC in the first trimester. In addition, we found that GDM risks were increased with elevated Hb level at early pregnancy, which were consistent with previous results [9]. The increased RBC and Hb in the first trimester could be a surrogate for the nutritional improvement. In our study, total of 567 (56.47%) women who were self-reported taking iron contained health product, including Elevit and Kingsley which contains iron or yellow iron oxide. Besides of weight gain, iron supplementation has been noted to be related with glucose tolerance in pregnancy [25,26]. Whereas, no difference of GDM incidence was existed between those people who were taking iron contained health product or not, except for higher Hb levels among people in the former group. Therefore, whether the positive association between risk of GDM and serum RBC or Hb levels is related to iron supplementation needs to be further explored.

Moreover, the changes of RBC and Hb from the early pregnancy to the second trimester were observed, in which the RBC and Hb concentrations in the second trimester were decreased. RBC and Hb concentrations in the second trimester were also found to be associated with the incidence of GDM, which was consistent with previous studies [20,27]. These findings emphasized that increased RBC and Hb concentration during pregnancy were probably related to the pathologic effects of GDM in addition to its normal changes. Further evidence based on large-scaled cohort studies and randomized clinical trials are required to evaluate whether these possible causal relationships are existed. Given that the discovery of symptoms or signs at the early stage of GDM are difficult, combining elevated RBC and Hb in the first trimester with clinical risk factors or laboratory indicators may be able to identify women at higher risk as early as possible.

In our study, the incidence of GDM was 18.82%, which was significantly higher than that of mainland China (14.80%) reported by a meta-analysis in 2019[28]. This difference could be explained from two aspects. On the one hand, all participants are residents of the first- and second-tier urban cities. It was speculated that the accelerated urbanization, lifestyle changes and population aging in China may induce higher GDM prevalence [29], especially in megacities with large population included in our study, given that the GDM incidence in our analysis was similar to the prevalence of hyperglycemia during pregnancy in large cities of North America [30]. On the other hand, the research sites in our study were ranked as grade A tertiary hospital, which means a high opportunity to admit high-risk pregnant women. Therefore, the GDM incidence in our study may have been overestimated.

It is worth noting that we found RBC and Hb in the first trimester was positive associated with the incidence of GDM. With increasing RBC (×10^{12}/L) and Hb (g/L) per unit, the risk of GDM increased 177% and 4%, respectively. This finding indicated
that RBC and Hb in the first trimester could be used as potential biomarkers for predicting GDM. However, low sensitivities and specificities for GDM prediction within single biomarkers were reported, including RBC, fasting plasma glucose, or HbA1c [20,31,32]. These studies suggested that GDM probably cannot be well predicted by a single biomarker or method in the first trimester [33,34]. As a routine prenatal-care test with standardized process and attractive cost benefit, tests of RBC count and Hb concentration in the first trimester combined with multiple information about clinical diagnoses may offer a valuable method for predicting GDM.

The association observed in our study between RBC count and the risk of GDM may be biologically plausible. Insulin receptor structure in RBC is identical to that in actual target cells of insulin [35] and can exhibit binding characteristics similar to those observed in classical target tissues for insulin action [36]. Other studies demonstrated that circulating RBC could be used to reflect short-term fluctuations in insulin-receptor affinity and long-term changes in receptor concentration [37]. Accumulating population-based evidence further support that high levels of RBC count and Hb are positively associated with underlying pathophysiological changes of hyperglycemia, including insulin resistance [14,15], β-cell dysfunction [15], and decreased insulin sensitivity [15]. HbA1c level is also affected by RBC lifespan and positively correlated with the RBC count since the lifespan of RBCs can directly affect the reaction time of glycosylation between Hb and blood glucose [38]. Lastly, the increase in blood viscosity leading by increased RBC count is associated with insulin resistance and may also increase the risk for GDM [39].

This study is the first prospective cohort study showing that RBC count in early pregnancy could be a new risk factor and biomarker of GDM. Effective measures have been taken to ensure the standardization of data collection, including training professional staff to interview and follow up, as well as recording standard information from medical records and laboratory examinations. Additionally, conventional risk factors of GDM including age, BMI before pregnancy, weight gain in the first or the second trimester, history of GDM, family history of GDM, passive smoking, and others were well controlled.

However, several limitations should be mentioned. This prospective cohort study is an observational study, which cannot provide a biological cause-effect relationship between RBC or Hb in early trimester and the risk of GDM. The small sample size with a deficient number of participants in higher WBC or PLT levels may have led to insufficient statistical power to assess the association between WBC or PLT and the risk of GDM. Similarly, small sample size was unable to evaluate the robustness and interaction of the observed association between RBC or Hb levels and the risk of GDM by using subgroup and stratified analyses. Moreover, the GDM incidence in our study may have been overestimated because the two research sites are top hospitals from megacities and have higher opportunities to admit high-risk pregnant women. In addition, the recruited participants were only from two cities in China, which may have introduced selection bias and limited the generalization of our findings. Finally, the serum iron or ferritin levels of the participants were not tested, which were unable to assess whether the observed association between RBC or Hb levels and the risk of GDM is related to iron or ferritin levels.

In conclusion, our study found that elevated RBC count and Hb concentration in the first trimester are associated with an increased risk of GDM. Further evidence are required to confirm the possible causal relationship between early pregnancy RBC and GDM. As a common and inexpensive prenatal-care test, combining the first-trimester RBC count and Hb concentration with multiple information about clinical diagnoses may offer a valuable, attractive and low-cost way to predict the risk of GDM before early signs or symptoms appear.

Declarations

Ethics approval and consent to participate

This study was designed and conducted in accordance with the Declaration of Helsinki (2000) of the World Medical Association and approved by the Institutional Research Review Board of the National Research Institute for Family Planning, Beijing, China. All participants provided written informed consent before participation.

Consent for publication

Not applicable.
Availability of data and materials

The ownership of datasets which supporting the conclusions of this cohort study are belonging to Ying Yang who employed at National Research Institute for Health and Family Planning, Beijing, China. These data are not open access to the public individuals or organizations, readers who want to access the data please contact Ying Yang, prof, E-mail: angela-yy65@hotmail.com

Competing interests

The authors declare no conflict of interest.

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Authors' contributions

All authors have contributed significantly and in keeping with the latest guidelines of the International Committee of Medical Journal Editors. The corresponding authors have full access to data in the study and takes responsibility for data integrity and the accuracy of data analysis. Jiajing Jia searched the literature, analyzed the data, interpreted the results, and drafted the manuscript. Ying Yang and Xu Ma designed and supervised the study. Ying Yang and Minjin Zhang revised the manuscript. Li Lin and Yequn Chen provided supervision in the two cohort study sites and trained staffs for investigation and sample collection. Tonglei Guo, Qin Xu, Long Wang, Zuoqi Peng, and Changlong Guo collected the data. Xingyu Wang and Yixin Chen finally revised and corrected the grammatical errors. Ying Yang and Xu Ma conceived of the study and provided overall guidance and revised the manuscript.

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### Tables

**Table 1. Baseline demographic characteristics of eligible participants**
| Variables                      | N    | GDM, n(%) | RBC (x10^{12}/L) | Hb(g/L) | WBC (x10^{9}/L) | PLT(x10^{9}/L) |
|-------------------------------|------|-----------|-------------------|--------|-----------------|---------------|
|                               |      |           | Median            | IQR    | Median          | IQR           |
|                               |      |           | Median            | IQR    | Median          | IQR           |
|                               |      |           | Median            | IQR    | Median          | IQR           |
| Age (y)                       |      |           |                   |        |                 |               |
| <35                           | 901  | 160(17.76) | 4.20              | 0.50   | 129.00          | 14.00         |
| ≥35                           | 102  | 29(28.43)  | 4.20              | 0.40   | 130.00          | 11.70         |
| Education                     |      |           |                   |        |                 |               |
| Below college                 | 259  | 43(16.60)  | 4.20              | 0.45   | 128.00          | 14.50         |
| College and above             | 744  | 146(19.62) | 4.20              | 0.41   | 130.00          | 13.00         |
| Job                           |      |           |                   |        |                 |               |
| Farmers                       | 331  | 58(17.52)  | 4.20              | 0.40*  | 128.00          | 14.00*        |
| Workers                       | 673  | 131(19.47) | 4.30              | 0.50   | 130.00          | 13.00         |
| Monthly income (¥)            |      |           |                   |        |                 |               |
| ≤5000                         | 527  | 101(19.17) | 4.20              | 0.50   | 127.00          | 13.00***      |
| >5000                         | 477  | 88(18.45)  | 4.20              | 0.40   | 131.00          | 13.00         |
| Pre-pregnancy BMI (kg/m^2)    |      |           |                   |        |                 |               |
| <18.4                         | 191  | 20(10.47)  | 4.20              | 0.50***| 126.00          | 15.00***      |
| 18.5~23.9                     | 626  | 112(17.89) | 4.20              | 0.40   | 129.00          | 13.00         |
| 24~27.9                       | 121  | 36(29.75)  | 4.37              | 0.30   | 133.00          | 11.00         |
| 28~                           | 56   | 17(30.36)  | 4.40              | 0.40   | 136.00          | 14.00         |
| Gestational weeks             |      |           |                   |        |                 |               |
| ≤8                            | 254  | 49(19.29)  | 4.30              | 0.40*  | 131.00          | 13.00**       |
| 8~12                          | 750  | 140(18.67) | 4.20              | 0.40   | 129.00          | 14.00         |
| Intaking iron contained health product |      |           |                   |        |                 |               |
| No                            | 394  | 64(16.24)  | 4.20              | 0.40   | 128.00          | 15***         |
| Yes                           | 567  | 117(20.63) | 4.20              | 0.50   | 131.00          | 12            |
| Fasting glucose (mmol/L)      |      |           |                   |        |                 |               |
| <6.1                          | 990  | 185(18.69) | 4.20              | 0.40   | 129.00          | 14.00         |
| ≥6.1                          | 10   | 4(40.00)   | 4.20              | 0.98   | 123.50          | 20.75         |
| History of GDM               |      |           |                   |        |                 |               |
| NO                            | 996  | 186(18.67) | 4.20              | 0.40   | 129.00          | 14.00         |
|                           | YES  | 8   | 3(37.50) | 4.35 | 0.35 | 127.50 | 4.75 | 7.45 | 1.11 | 247.00 | 30.25 |
|--------------------------|------|-----|----------|------|------|--------|------|------|------|--------|-------|
| Family history of GDM   |      |     |          |      |      |        |      |      |      |        |       |
| NO                       | 976  | 179 | 18.34†   | 4.20 | 0.50 | 129.00 | 14.00| 8.41 | 2.71 | 232.00 | 63.00 |
| YES                      | 14   | 6   | 42.86    | 4.25 | 0.18 | 133.50 | 8.75 | 8.77 | 1.76 | 223.00 | 36.50 |
| Elevated BPs            |      |     |          |      |      |        |      |      |      |        |       |
| NO                       | 982  | 179 | 18.23††  | 4.20 | 0.40 | **129.00** | 14.00| 8.40 | 2.68** | 232.00 | 62.70 |
| YES                      | 22   | 10  | (45.45)  | 4.50 | 0.38 | 133.00 | 15.00| 9.84 | 3.01 | 236.50 | 74.75 |
| Passive smoking during pregnancy |      |     |          |      |      |        |      |      |      |        |       |
| NO                       | 628  | 132 | 20.02†   | 4.20 | 0.50 | **130.00** | 13.00| 8.35 | 2.87 | 232.00 | 64.00 |
| YES                      | 365  | 55  | (15.07)  | 4.20 | 0.40 | **128.00** | 13.00| 8.57 | 2.37 | 233.00 | 61.00 |
| Dietary taste            |      |     |          |      |      |        |      |      |      |        |       |
| Light                    | 248  | 45  | (18.15)  | 4.20 | 0.40 | **128.00** | 13.00| 8.43 | 2.36 | 227.00 | 60.00 |
| Moderate                 | 572  | 118 | (20.63)  | 4.30 | 0.50 | **130.00** | 13.00| 8.41 | 2.76 | 234.00 | 60.25 |
| Salty                    | 180  | 26  | (14.44)  | 4.20 | 0.40 | **130.00** | 12.00| 8.44 | 3.01 | 228.50 | 71.50 |
| Work transportation      |      |     |          |      |      |        |      |      |      |        |       |
| Non-work                 | 214  | 30  | (14.02)† | 4.20 | 0.50 | **126.50** | 14.00| 8.58 | 2.88 | 228.53 | 66.50 |
| Drive                    | 299  | 54  | (18.06)  | 4.20 | 0.40 | **129.00** | 14.00| 8.40 | 2.82 | 234.00 | 59.00 |
| Public transport         | 369  | 86  | (23.31)  | 4.22 | 0.50 | **131.00** | 12.00| 8.45 | 2.55 | 233.00 | 59.00 |
| Walk or bicycle          | 120  | 19  | (15.83)  | 4.20 | 0.40 | **129.00** | 15.00| 8.29 | 2.82 | 226.00 | 63.25 |

GDM: gestational diabetes; RBC: red blood cell count; Hb: hemoglobin concentration; WBC: white blood cell count; PLT: platelet count; IQR: interquartile range; BMI: body mass index; BP: blood pressure; 

n(%), n: number of participants; %: incidence of GDM; 

Mann-Whitney U test or Kruskal-Wallis test were used to examine differences of hematological parameters among groups with categorized baseline characteristics, *: \( P \text{ value} <0.05 \); **: \( P \text{ value} <0.01 \); ***: \( P \text{ value} <0.001 \); 

c\(^2\) tests were used to examine statistical differences of GDM incidence among baseline subgroups, †: \( P \text{ value} <0.05 \); ††: \( P \text{ value} <0.01 \); †††: \( P \text{ value} <0.001 \); 

**Table 2. Description of numeric hematological parameters in the first and the second trimester of pregnancy**
| Variables          | Non-GDM M(IQR) | GDM M(IQR) | Age-adjusted OR (95% CI) | Multivariable-adjusted OR (95% CI) |
|-------------------|----------------|------------|--------------------------|-----------------------------------|
| RBC (×10^{12}/L)  |                |            |                          |                                   |
| Before 12 gestational weeks | 4.20(0.40) *** | 4.40(0.50) *** | 2.98(1.87,4.74) | 2.77(1.68,4.57) † |
| 24 gestational weeks  | 3.70(0.40) *** | 3.80(0.33) *** | 2.50(1.49,4.20) | 2.87(1.61,5.11) ‡ |
| Hb (g/L)          |                |            |                          |                                   |
| Before 12 gestational weeks | 128(14.00) ^*   | 133(13.00) ***   | 1.05(1.03,1.07) | 1.04(1.02,1.06) † |
| 24 gestational weeks  | 116(12.00) ^*   | 117(11.00) **   | 1.04(1.01,1.06) | 1.04(1.02,1.07) ‡ |
| WBC (×10^{9}/L)   |                |            |                          |                                   |
| Before 12 gestational weeks | 8.31(2.62) ^*   | 8.92(2.79) **   | 1.12(1.04,1.21) | 1.11(1.02,1.21) † |
| 24 gestational weeks  | 9.81(2.85) **   | 10.03(2.76)     | 1.02(0.94,1.10) | 1.00(0.92,1.09) ‡ |
| PLT (×10^{9}/L)   |                |            |                          |                                   |
| Before 12 gestational weeks | 231(62.00) ^*   | 235(68.00)     | 1.00(1.00,1.01) | 1.00(1.00,1.00) † |
| 24 gestational weeks  | 225(64.00) ^*   | 217(58.50)     | 1.00(1.00,1.00) | 1.00(1.00,1.00) ‡ |

GDM: gestational diabetes; RBC: red blood cell count; Hb: hemoglobin concentration; WBC: white blood cell count; PLT: platelet count;

Continuous variables were described by median and interquartile range. M(IQR): median(interquartile range);

Mann-Whitney U test was used to examine differences of hematological parameters between GDM and non-GDM groups, *: P value <0.05; **: P value <0.01; ***: P value <0.001;

†: Adjusted for age, BMI before pregnancy, weight gain at the first trimester, pregnancy weeks, history of gestational diabetes, family history of gestational diabetes, hypertensive status, fasting glucose concentration, iron contained health product consumption, passive smoking during pregnancy, dietary taste, and work transportation;

‡: Adjusted for age, BMI before pregnancy, weight gain at the second trimester, pregnancy weeks, history of gestational diabetes, family history of gestational diabetes, hypertensive status, fasting glucose concentration, iron contained health product consumption, passive smoking during pregnancy, dietary taste, and work transportation;

Table 3. Association of categorized hematological parameters in the first trimester with gestational diabetes
| Variables          | N  | GDM    | n (%) | Age-adjusted OR (95% CI) | Multivariable-adjusted OR (95% CI) |
|--------------------|----|--------|-------|--------------------------|-----------------------------------|
|                    |    |        |       | Model I†                  | Model II‡                          |
| Class I            |    |        |       |                          |                                   |
| RBC (×10^{12}/L)   |    |        |       |                          |                                   |
| 3.42~4.55          | 817| 138    | (16.89)| 1                        | 1                                 |
| ~3.41 or 4.56‡     | 187| 51     | (27.27)| 1.98(1.36,2.90)          | 1.94(1.29,2.92)                   |
| Hb (g/L)           |    |        |       |                          |                                   |
| 116~139            | 782| 139    | (17.80)| 1                        | 1                                 |
| ~115 or 140~       | 222| 50     | (22.52)| 1.33(0.92,1.93)          | 1.31(0.88,1.95)                   |
| WBC (×10^{9}/L)    |    |        |       |                          |                                   |
| 5.7~13.6           | 934| 175    | (18.76)| 1                        | 1                                 |
| ~5.69 or 13.61~    | 70 | 14     | (20.00)| 1.11(0.60,2.05)          | 1.04(0.45,1.85)                   |
| PLT (×10^{9}/L)    |    |        |       |                          |                                   |
| 174~391            | 932| 181    | (19.44)| 1                        | 1                                 |
| ~173 or 392~       | 72 | 8      | (11.11)| 0.57(0.27,1.22)          | 0.66(0.31,1.44)                   |
| Class II           |    |        |       |                          |                                   |
| RBC (×10^{12}/L)   |    |        |       |                          |                                   |
| ~3.41              | 10 | 1      | (10.00)| 0.58(0.07,4.67)          | 0.91(0.11,7.60)                   |
| 3.42~4.55          | 817| 138    | (16.89)| 1                        | 1                                 |
| 4.56‡              | 177| 50     | (28.25)| 2.08(1.42,3.06)          | 2.00(1.33,3.03)                   |
| P for trend        |    |        |       | <0.001                   | 0.002                             |
| Hb (g/L)           |    |        |       |                          |                                   |
| ~115               | 97 | 8      | (8.25 )| 0.39(0.19,0.84)          | 0.40(0.17,0.93)                   |
| 116~139            | 782| 139    | (17.80)| 1                        | 1                                 |
| 140~               | 125| 42     | (33.60)| 2.40(1.57,3.66)          | 2.14(1.36,3.36)                   |
| P for trend        |    |        |       | <0.001                   | <0.001                             |
| WBC (×10^{9}/L)    |    |        |       |                          |                                   |
| ~5.69              | 49 | 8      | (16.33)| 1.05(0.51,2.13)          | 1.12(0.52,2.42)                   |
| 5.70~13.6          | 934| 175    | (18.76)| 1                        | 1                                 |
| 13.61~             | 21 | 6      | (28.57)| 1.28(0.57,2.90)          | 1.00(0.40,2.46)                   |
| P for trend        |    |        |       | 0.459                    | 0.694                             |

GDM: gestational diabetes; RBC: red blood cell count; Hb: hemoglobin concentration; WBC: white blood cell count; PLT: platelet count;

Categorical variables were described by number and row percentage.
†: Adjusted for age, BMI before pregnancy, weight gain at the first trimester, pregnancy weeks, history of gestational diabetes, family history of gestational diabetes, hypertensive status, fasting glucose concentration, iron contained health product consumption, passive smoking during pregnancy, dietary taste, and work transportation;

‡: Adjusted for age, BMI before pregnancy, weight gain at the second trimester, pregnancy weeks, history of gestational diabetes, family history of gestational diabetes, hypertensive status, fasting glucose concentration, iron contained health product consumption, passive smoking during pregnancy, dietary taste, and work transportation;

§: PLT count can only be classified into two layers (normal range and abnormal range) due to no one was diagnosed as GDM in group of PLT count >392 (×109/L).