In comparison with the non-pregnant state, the postprandial glucose concentration is elevated in pregnant subjects, especially during late pregnancy [1]. Postprandial maternal hyperglycemia is thought to accelerate glucose transfer from mother to fetus, which guarantees normal fetal growth during pregnancy. An increase in maternal insulin resistance during pregnancy manifests in early gestation [2], and causes physiological maternal postprandial hyperglycemia. Accordingly, the degree of maternal insulin resistance manifested during pregnancy is theoretically associated with the degree of glucose flux from mother to fetus. Gestational diabetes mellitus (GDM) is a typical example. Accompanied with beta cell dysfunction, excessive manifestation of insulin resistance during pregnancy is associated with the development of GDM [3]. In women with GDM, it is thought that maternal hyperglycemia leads to fetal hyperglycemia and hyperinsulinemia, which cause fetal macrosomia. Excessive insulin resistance during pregnancy is also observed in obese subjects without abnormal glucose tolerance [4], and fetal macrosomia is also common in these patients.

However, to the best of our knowledge, there have been few studies performed to address the association between the degree of physiological increase in maternal insulin resistance during pregnancy and neonatal birthweight in non-diabetic pregnancy. The aim of this study was to determine whether elevated maternal insulin resistance, as measured by the homeostasis model assessment-insulin resistance (HOMA-IR) in the second and third trimesters, is associated with increased neonatal birthweight, and therefore a risk of...
macrosomic infants after controlling for maternal obesity and plasma glucose (PG) levels in uncomplicated healthy pregnancies.

**Material and Methods**

This was a retrospective study using our GDM screening database over a six-year period from 2004 to 2009 at the Nagasaki Medical Center. This study was approved by the Institutional Review Board of Nagasaki Medical Center with written informed consent obtained from all subjects. All pregnant subjects were screened for GDM between the second and third trimesters using a 50 g glucose challenge test (GCT). In subjects with a positive GCT (≥ 135 mg/dL), we performed a 75 g oral glucose tolerance test (OGTT) after an overnight fast. Among the subjects who underwent OGTT during the trimesters, we included only those with healthy singleton pregnancies with normal OGTT results as determined by Japan Society of Obstetrics and Gynecology (JSOG) criteria [5]. We defined a normal OGTT test result as all three normal values of <100 mg/dL at fasting, <180 mg/dL at 1-hour, and <150 mg/dL at 2-hour after a 75 g oral glucose loading. We did not apply the new diagnostic criteria, i.e. the International Association of Diabetes and Pregnancy Study Groups (IADPSG) criteria [6] during the study period, because it was not adopted by the JSOG until June, 2010. At the time of OGTT, we also measured serum immunoreactive insulin (IRI) concentrations at fasting. We used HOMA-IR as a surrogate index of maternal insulin resistance, which was calculated by the equation \([\text{fasting PG level} \times \text{fasting IRI level}] / 405\). We defined large-for-gestational age (LGA) infants by using the 90th percentile of the standard Japanese infantile growth curve [7].

We excluded patients with GDM in order to avoid treatment bias, as we treated the patients with the aim of preventing perinatal complications associated with GDM, including neonatal macrosomia. We also excluded women with hypertensive disorders, other maternal medical complications, including systemic lupus erythematosus and thyroid disease, and those with fetal malformation. In the remaining patients, we examined the association between the neonatal birthweight and predictive variables, including maternal PG and HOMA-IR at OGTT, parity, pre-pregnancy body mass index (BMI), and weight gain during pregnancy by using univariate regression analysis after adjusting for gestational age (GA) at delivery. Then we tested the association between the maternal HOMA-IR and neonatal birthweight by multivariate regression analysis to adjust for confounding variables, including parity, pre-pregnancy BMI, weight gain during pregnancy, glucose values at OGTT, and GA at delivery. We also examined the association between maternal HOMA-IR and risk for LGA infants by using multiple logistic regression models after adjusting for these confounders. A \(p\)-value < 0.05 was considered to be significant.

**Results**

The maternal characteristics, results of 75 g OGTT, and neonatal outcomes of the 655 Japanese women involved in this study are summarized in Table 1. Mean GA at the time of OGTT was 28.3±6.3 weeks of gestation, and 224 (34%) and 431 (66%) subjects had the 75 g OGTT in their second and third trimester, respectively. Mean HOMA-IR among all subjects was 1.4±0.8. Since the mean HOMA-IR values were 1.34±0.92 and 1.46±0.79 in the second and third trimesters, respectively, and these were not significantly different \((p=0.38)\), we combined the data together for the analysis.

In the univariate analysis after adjusting for GA at delivery, parity (multipara vs. primipara, \(r^2=0.35, p=0.001\)), pre-pregnancy BMI \((r^2=0.36, p<0.001)\), weight gain during pregnancy \((r^2=0.37, p<0.001)\), fasting PG

**Table 1** Maternal characteristics, results of the 75 g OGTT, and neonatal outcomes (n=655)

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Mean±SD or N (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (y/o)</td>
<td>31.9±4.9</td>
</tr>
<tr>
<td>Primipara (%)</td>
<td>307 (46.9)</td>
</tr>
<tr>
<td>Pre-pregnancy BMI (kg/m²)</td>
<td>22.0±4.0</td>
</tr>
<tr>
<td>Weight gain during pregnancy (kg)</td>
<td>9.7±4.1</td>
</tr>
<tr>
<td>GA at 75 g OGTT (wk)</td>
<td>28.3±6.3</td>
</tr>
<tr>
<td>Fasting PG (mg/dL)</td>
<td>79.5±6.5</td>
</tr>
<tr>
<td>1h-PG (mg/dL)</td>
<td>144.1±25.0</td>
</tr>
<tr>
<td>2h-PG (mg/dL)</td>
<td>122.8±22.7</td>
</tr>
<tr>
<td>Fasting IRI (μU/mL)</td>
<td>7.1±3.8</td>
</tr>
<tr>
<td>HbA1c (%) (mmol/mol)</td>
<td>5.3±0.5 (34.0±3.1)</td>
</tr>
<tr>
<td>HOMA-IR</td>
<td>1.4±0.8</td>
</tr>
<tr>
<td>GA at delivery (wk)</td>
<td>39.1±1.7</td>
</tr>
<tr>
<td>Birth weight (g)</td>
<td>3,039±466</td>
</tr>
<tr>
<td>LGA infants (%)</td>
<td>91 (13.9%)</td>
</tr>
</tbody>
</table>

BMI, body mass index; GA, gestational age; PG, plasma glucose; IRI, immunoreactive insulin; HOMA-IR, homeostasis model assessment-insulin resistance; LGA, large-for-gestational age.
In healthy non-diabetic singleton pregnancies, we found that the maternal HOMA-IR in the second and third trimesters was significantly and positively associated with the neonatal birthweight after adjusting for the parity, pre-pregnancy BMI, weight gain during pregnancy, and PG levels. Elevated maternal HOMA-IR was an independent risk factor of giving birth to an LGA infant after controlling for these confounding variables. To the best of our knowledge, this is the first report to demonstrate a significant association between maternal HOMA-IR and fetal growth independent of maternal obesity and PG levels in normal pregnancy.

In previous studies, investigators did not find any independent association between maternal insulin resistance during pregnancy and neonatal birthweight in subjects with or without GDM. Voldner et al. [8] investigated the relationship between fetal macrosomia and maternal metabolic measures, including their fasting PG, fasting insulin, and HOMA-IR at 30-32 weeks of gestation in 553 non-GDM Caucasian females. They found that only the fasting PG, and neither the fasting insulin nor HOMA-IR, was associated with macrosomia after adjusting for covariates including maternal BMI. Bomba-Opon et al. [9] examined the association between maternal HOMA-IR in the third trimester and neonatal birthweight in 121 patients with GDM, and could not find any association. Das et al. [10] found a significant association between maternal HOMA-IR and ultrasonographically-determined fetal growth at 24-28 weeks of gestation in 86 women with normal glucose tolerance. Although they adjusted for maternal PG levels, they did not control for maternal obesity and did not address neonatal birthweight. The Japanese pop-
ulation is the least obese among developed countries [11], and in this study the subjects had a mean standard pre-pregnancy BMI of 22.0 kg/m². It is possible that the difference in basic obesity between Japanese and other ethnic populations may contribute to a difference between our study and other studies. Maternal BMI is strongly associated with neonatal birthweight independent of maternal glucose levels [12], and obesity is also significantly associated with insulin resistance. Therefore, to determine whether maternal insulin resistance is associated with fetal growth independent of maternal obesity in the obese population rather than less obese population like Japanese subjects, much more sample size would be necessary.

We combined the HOMA-IR data from the second and third trimesters, because there were no significant differences between them. Maternal insulin resistance is already increased in early gestation in comparison with the pre-pregnant state in healthy pregnant subjects [2]. Although the change in insulin resistance between the second and third trimesters has not been well documented in normal pregnancy, some authors have reported that no significant change was observed in the maternal HOMA-IR between the trimesters in either non-obese or obese women with normal glucose tolerance [13]. Cohen et al. reported that the HOMA-IR is appropriate for use during the second and third trimesters of pregnancy even in obese patients [14].

In terms of the association between maternal hyperglycemia and neonatal birthweight, Voldner et al. [8] reported that the maternal fasting PG was the only independent risk factor associated with neonatal macrosomia. A large multicenter prospective observational study called the HAPO study [15] confirmed that each maternal PG level during 75 g OGTT in mid-pregnancy was independently associated with giving birth to an LGA neonate, and that the fasting PG showed the strongest association. In the HAPO study, however, they did not address maternal insulin status. In the univariate analyses in our study, we also found that each PG during 75 g OGTT was significantly associated with neonatal birthweight, and that the fasting PG was the strongest. However, the association was no longer significant in the multivariate regression models including HOMA-IR and the pre-pregnancy BMI as covariates. While the association between maternal PG levels and neonatal birthweight cannot be independent from maternal HOMA-IR, since there is a link between maternal PG levels and insulin resistance in normal pregnancy, HOMA-IR in mid-pregnancy could well be a better predictive variable for neonatal birthweight and macrosomia than maternal PG levels because of its lack of reproducibility during OGTT in uncomplicated pregnancies. Again, the lack of statistical power may have affected the identification of such a modest association in non-diabetic healthy pregnant subjects in a previous study [8].

It is well-documented that maternal pre-pregnancy BMI and excessive gestational weight gain are independently associated with fetal macrosomia in non-diabetic pregnancy [16-18]. A subanalysis of the HAPO study showed that maternal obesity was significantly associated with macrosomia, independent of maternal PG levels [19]. In our study, we found that maternal pre-pregnancy BMI and gestational weight gain were individually associated with having an LGA infant, independent of not only maternal glycemic levels, but also insulin resistance status. Although maternal obesity, excessive gestational weight gain and insulin resistance during pregnancy are interrelated [20, 21], our results showed that these three factors may independently influence fetal overgrowth during normal pregnancy.

There are several limitations to this study. Firstly, we did not directly measure insulin resistance. Although the gold standard used to measure insulin resistance in vivo is the euglycemic glucose clamp method [22], because of the complexity of the clamp method, we were obliged to use surrogate indices of insulin resistance which are often used in the clinical setting. HOMA-IR is known to show a good linear correlation to insulin resistance directly measured by glucose clamp technique in non-pregnant adults [23-25]. Although HOMA-IR during pregnancy is less correlated to directly measured insulin resistance by clamp method in comparison with females in a non-pregnant state, it is still a significant predictor of total insulin sensitivity throughout pregnancy and may be a useful tool to assess maternal insulin status [14, 26]. Secondly, we did not measure neonatal adiposity. Neonatal adiposity is well recognized in infants born from diabetic and gestational diabetic mothers and is a very sensitive marker of abnormal fetal overgrowth [27, 28]. Walsh et al. [29] reported that the maternal fasting PG concentration at 24 weeks of gestation was significantly associated with both infant birthweight and adiposity in healthy non-diabetic mothers. Although further examinations are necessary, maternal HOMA-IR, a surrogate marker of insulin resistance, is expected to provide a
new predictor of neonatal adiposity in diabetic and/or non-diabetic healthy mothers.

In terms of clinical significance, we were able to use HOMA-IR levels to assess the risk of having a macrosomic infant in women without GDM. Although we did not include patients with gestational diabetes in this study due to therapeutic bias, HOMA-IR levels may also be useful to estimate the risk of macrosomia in such patients. It has been reported that the prevalence of GDM has increased since applying the new IADPSG diagnostic criteria [6], and it may be possible to make triage decisions based on HOMA-IR level in order to assess the risk of macrosomia.

In summary, maternal HOMA-IR in the second and third trimesters was significantly associated with neonatal birthweight and the risk of giving birth to an LGA infant after controlling for GA at birth, maternal parity, pre-pregnancy BMI, weight gain during pregnancy, and PG levels in uncomplicated pregnancies. Our findings suggest that the degree of insulin resistance in mid-pregnancy plays an important role in fetal growth in normal healthy pregnancies, independent of maternal obesity and glucose levels.

Disclosure

None of the authors have any potential conflict of interest to disclose associated with this research.

References


