**SUPPLEMENTAL FILE**

**Lactation vs formula feeding: Insulin, glucose and fatty acid metabolism during**

**the postpartum period**

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**Supplemental Table 1.** Characteristics of all postpartum subjects

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **Variable** | **Lactating** | | **Formula** | | | ***P*-Value** |
|  | NGT (n=6) | GDM (n=12) | NGT (n=6) | GDM (n=4) |  | |
| **Demographics and Anthropometrics** | | | | | | |
| Age (years) | 34 ± 6 | 34 ± 4 | 32 ± 2 | 37 ± 3 | 0.21 | |
| Parity | 2.7 ± 0.5 | 3.3 ± 0.9 | 3.0 ± 1.5 | 2.3 ± 1.0 | 0.35 | |
| Maternal weight (kg) |  |  |  |  |  | |
| ---Before pregnancy | 64 ± 5 \*, † | 79 ± 14 | 75 ± 10 | 70 ± 9 | 0.10 | |
| ---Term | 78 ± 10 | 86 ± 15 | 85 ± 11 | 81 ± 4 | 0.53 | |
| ---At 6 wk postpartum | 68 ± 10 | 78 ± 15 | 75 ± 8 | 71 ± 7 | 0.35 | |
| ---Retention at 6 wk postpartum | 3.9 ± 5.3 | -0.1 ± 5.0 | 0.8 ± 3.2 | 0.8 ± 2.2 | 0.36 | |
| BMI at 6 wk postpartum (kg/m2) | 27.2 ± 2.3 | 31.1 ± 4.8 | 30.4 ± 2.5 | 31.0 ± 0.7 | 0.19 | |
| WHR at 6 wk postpartum | 0.93 ± 0.02 ‡ | 0.96 ± 0.04 § | 0.94 ± 0.06 ‖ | 1.01 ± 0.04 | 0.06 | |
| Intrahepatic lipid (%) | 7.6 ± 4.0 | 11.0 ± 6.3 | 8.6 ± 5.5 | 13.4 ± 6.1 | 0.40 | |
| DEXA at 6 wk postpartum |  |  |  |  |  | |
| ---Body fat in kg | 25.7 ± 4.0 | 32.0 ± 10.7 | 29.6 ± 4.7 | 28.3 ± 3.0 | 0.45 | |
| ---Fat free mass in kg | 40.2 ± 5.9 | 44.3 ± 6.0 | 43.7 ± 6.0 | 40.8 ± 3.8 | 0.46 | |
| Infant birth weight (kg) | 3.54 ± 0.29 | 3.56 ± 0.40 | 3.24 ± 0.47 | 3.58 ± 0.31 | 0.39 | |
| Exclusive lactation (%) | 4 (67) †, ¶ | 11 (92) # | 0 \*\* | 0 | 0.0001 | |
| Amount of formula fed (oz) | 1.0 ± 1.8 †, ‡ | 0.4 ± 1.4 # | 24.0 ± 0.0 \*\* | 24.0 ± 0.0 | <0.0001 | |
| Gestational age at delivery (days) | 274 ± 6 | 276 ± 7 | 279 ± 12 | 275 ± 4 | 0.76 | |
| Family history of type 2 diabetes (%) | 3 (50) | 11 (92) | 3 (50) | 3 (75) | 0.11 | |

Data are mean ± SD. Abbreviations: WHR, waist-to-hip ratio; NGT, normal glucose tolerance during pregnancy; GDM, gestational diabetes mellitus. Comparisons were made by one-way ANOVA for normal distribution or Kruskal-Wallis test for non-normal distribution among four groups. For post-hoc comparisons, two-sided t tests were used for variables with a normal distribution or by Wilcoxon Rank Sum test for non-normal distribution. Categorical variables were analyzed by Fisher’s exact test.

\* Lactating NGT versus Lactating GDM, *P* < 0.05

† Lactating NGT versus Formula NGT, *P* < 0.05

‡ Lactating NGT versus Formula GDM, *P* < 0.05

§ Lactating GDM versus Formula GDM, *P* > 0.05 and ≤ 0.10

‖ Formula NGT versus Formula GDM, *P* > 0.05 and ≤ 0.10

¶ Lactating NGT versus Formula GDM, *P* > 0.05 and ≤ 0.10

# Lactating GDM versus Formula GDM, *P* < 0.05

\*\* Lactating GDM versus Formula NGT, *P* < 0.05

**Supplemental Table 2.** Biochemistries, metabolic indicators, insulin sensitivity, and plasma hormones by both infant-feeding method at six weeks postpartum and glucose tolerance during pregnancy

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Variable** | **Lactating** | | **Formula** | | ***P*-Value** |
|  | NGT (n=6) | GDM (n=12) | NGT (n=6) | GDM (n=4) |  |
| **Biochemistries and metabolic indicators** | | | | | |
| HbA1c in percent (mmol/mol) | 5.8 ± 0.3 (40) | 5.7 ± 0.3 (39) | 5.5 ± 0.4 (37) | 5.7 ± 0.1 (39) | 0.25 |
| Fasting glucose (mg/dL) | 82 ± 9 | 85 ± 8 | 84 ± 7 | 87 ± 2 | 0.80 |
| Fasting insulin (μU/mL) | 2.1 ± 2.1 | 2.9 ± 2.2\* | 3.6 ± 2.2 | 5.9 ± 2.1 | 0.06 |
| Fasting FFA (mmol/L) | 0.56 ± 0.14 | 0.62 ± 0.15 | 0.56 ± 0.13 | 0.62 ± 0.13 | 0.73 |
| Total cholesterol (mg/dL) | 197 ± 13 | 199 ± 26 | 198 ± 38 | 186 ± 51 | 0.90 |
| HDL cholesterol (mg/dL) | 48 ± 7† | 52 ± 12‡ | 39 ± 7 | 45 ± 6 | 0.08 |
| Triglycerides (mg/dL) | 143 ± 61 | 110 ± 48 | 194 ± 103 | 157 ± 95 | 0.16 |
| LDL cholesterol (mg/dL) | 121 ± 11 | 125 ± 23 | 121 ± 33 | 111 ± 32 | 0.80 |
| Matsuda Index | 16.2 ± 10.8§ | 9.7 ± 8.9 | 8.3 ± 4.8 | 4.8 ± 3.0‖ | 0.06 |
| HOMA-IR | 0.5 ± 0.4 | 1.2 ± 1.2 | 1.1 ± 0.5 | 2.0 ± 1.2‖ | 0.05 |
| Adipo IR | 1.2 ± 0.7† | 3.4 ± 3.8\* | 3.2 ± 1.9 | 6.0 ± 2.7‖ | 0.02 |
| Insulin Secretion-Sensitivity Index-2 | 3.4 ± 2.3 | 1.8 ± 0.5‡ | 2.8 ± 0.9 | 1.7 ± 1.0 | 0.03 |
| First phase Stumvoll (μU/mL) | 93 ± 12† | 99 ± 75 | 159 ± 54 | 186 ± 122¶ | 0.10 |
| Second phase Stumvoll (μU/mL) | 27 ± 3 | 29 ± 18 | 43 ± 14 | 50 ± 30 | 0.11 |
| Insulin infusion rate, 10/20 mU/m2/min | 3 | 9 | 3 | 3 | 0.57 |
| Insulin infusion rate, 10/40 mU/m2/min | 3 | 3 | 3 | 1 | 0.57 |
| Fasting respiratory quotient | 0.83 ± 0.02 | 0.81 ± 0.10 | 0.86 ± 0.05 | 0.85 ± 0.01 | 0.68 |
| Clamp step 1 respiratory quotient | 0.87 ± 0.05 | 0.84 ± 0.07 | 0.90 ± 0.06 | 0.88 ± 0.03 | 0.29 |
| Clamp step 2 respiratory quotient at IIR=20 | 0.92 ± 0.13 | 0.87 ± 0.09 | 0.99 ± 0.09 | 0.87 ± 0.04 | 0.28 |
| Clamp step 2 respiratory quotient at IIR=40 | 0.94 ± 0.06 | 0.91 ± 0.04 | 0.93 ± 0.05 | 0.94 | 0.84 |
| **Oral glucose tolerance test (OGTT)** | | | | | |
| Plasma glucose AUC (g/dL) • min | 15.4 ± 1.2# | 19.1 ± 1.9‡ | 14.1 ± 3.6 | 18.6 ± 4.2 | 0.003 |
| Plasma insulin AUC (mU/mL) • min | 3.2 ± 0.4 | 5.1 ± 3.1 | 6.1 ± 3.7 | 8.5 ± 6.9 | 0.36 |
| Plasma FFA AUC (mmol/L) • min | 28.9 ± 5.3 | 37.5 ± 9.7 | 35.6 ± 12.4 | 38.5 ± 9.5 | 0.31 |
| NGT/IFG/IGT/Combined IFG and IGT | 6/0/0/0# | 2/1/9/0‡ | 5/0/1/0 | 2/0/1/1 | 0.002 |
| **Hormone concentrations** | | | | | |
| Prolactin (ng/mL) | 240 ± 119† | 243 ± 152\*\* | 14 ± 10‡ | 14 ± 8‖ | 0.0003 |
| Total adiponectin (μg/mL) | 3.92 ± 1.18 | 4.26 ± 1.30 | 4.01 ± 1.20 | 3.67 ± 0.83 | 0.84 |
| HMW adiponectin (μg/mL) | 1.98 ± 0.89 | 2.14 ± 1.08 | 1.86 ± 0.69 | 1.71 ± 0.50 | 0.85 |
| HMW / total adiponectin | 0.48 ± 0.10 | 0.48 ± 0.11 | 0.45 ± 0.06 | 0.46 ± 0.05 | 0.93 |
| Leptin (ng/mL) | 4.63 ± 3.63 | 5.29 ± 4.31 | 5.85 ± 2.11 | 8.10 ± 2.65 | 0.14 |
| Total adiponectin / leptin (μg/mL per ng/mL) | 1.94 ± 2.63 | 1.27 ± 0.94 | 0.74 ± 0.27 | 0.47 ± 0.06 | 0.12 |
| Estradiol (pg/mL) | 35 ± 15† | 27 ± 18\*\* | 73 ± 27‡ | 79 ± 35‖ | 0.003 |

Data are mean ± SD. Abbreviations: FFA, free fatty acids; HOMA, homeostatic model assessment; Adipo-IR, adipose insulin resistance; HMW, high molecular weight. Comparisons were made by one-way ANOVA for normal distribution or Kruskal-Wallis test for non-normal distribution among four groups. For post-hoc comparisons, two-sided t tests were used for variables with a normal distribution or by Wilcoxon Rank Sum test for non-normal distribution. Categorical variables were analyzed by Fisher’s exact test.

\* Lactating GDM versus Formula GDM, *P* > 0.05 and ≤0.10

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# Lactating NGT versus Lactating GDM, *P* < 0.05

\*\* Lactating GDM versus Formula GDM, *P* < 0.05

**Supplemental figure 1.** Plots of glucose and insulin concentrations during 2-h oral glucose tolerance tests



A) Glucose and B) insulin data for the 12 lactating women (filled circles) and the 6 formula-feeding women (open circles) discussed in the main text. C) Glucose and D) insulin data for the 28 postpartum women included in the supplemental tables: Lactating women with normal glucose tolerance in pregnancy, filled square. Lactating women with recent GDM, filled triangle. Formula-feeding women with normal glucose tolerance in pregnancy, open square. Formula-feeding women with recent GDM, open triangle. Data are mean ± SD for each time point. The statistical analysis reported is for total AUC glucose and total AUC insulin.