Plasma microRNA Profiling of Obese Pregnant Women with/without Gestational Diabetes Mellitus

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Introduction: Increasing Maternal Obesity (MO) worldwide is contributing to Gestational Diabetes Mellitus (GDM) occurrence, both leading to adverse short/long term maternal and offspring outcomes.
miRNAs, as reversible epigenetic actors, are appealing targets for early therapy and intervention strategies. Limited studies explored their association in pregnancy, being also affected by a huge variability in methods, sample quality, subjects' selection criteria and timing. We recently found an altered epigenetic profile in saliva of obese pregnancies.
Here we perform plasma miRNA profiling of normoglycemic and diabetic obese *vs* lean pregnant women.

Methods: Caucasian women with single term pregnancies (normal-weight controls, NW=7; obese without comorbidities, OB/GDM-=6; obese with GDM, OB/GDM+=6) were enrolled at elective cesarean section.
Plasma expression of 179 miRNAs was analyzed by miRCURY LNA SybrGreen PCR.
Bioinformatic analyses were performed with GeneGlobe software and predictive tools miTALOS v.2/miRPath v.3; statistics with SPSS v.27.

Results: Except for maternal BMI and glycemia, the 3 groups did not differ in maternal, placental and neonatal characteristics.
Plasma profiling revealed a number of differentially expressed miRNAs: 4 miRNAs (**A**) in OB/GDM- *vs* NW, 1 miRNA (**B**) in OB/GDM+ *vs* NW, and 14 miRNAs (**C**) OB/GDM+ *vs* OB/GDM- (**Figure**).
The bioinformatic enrichment analysis found associations between these miRNAs and **A)** 35, **B)** 15, **C)** 38 different pathways, mostly involved in fatty acid and vitamin B6 metabolism, steroid homeostasis, amino acid (lysine, valine, leucine, isoleucine) biosynthesis/degradation, ECM-receptor interaction, thyroid hormones signalling, FoxO and insulin regulation, *AMPK*, *mTOR*, *TGF-β* and *HIF-1* signalling.

Conclusion: In this preliminary study, we profiled specific patterns of circulating miRNAs in the presence of MO and GDM. miRNAs with altered expression are involved in adipocyte differentiation and impaired glucose/lipid metabolism, and few of them have been proposed as metabolic regulators in GDM.
Moreover, among the identified miRNAs-associated pathways, macro/micro nutrients metabolism and signaling, energy production, inflammation, oxidative status and insulin resistance may represent interlinked driving processes in MO and GDM pathogenesis.
A larger sample size will help highlighting altered miRNAs and their role in the obesogenic and diabetic pregnancy context.

Keywords: *maternal obesity; gestational diabetes mellitus; miRNA Profiling*

(**Figure**)

Figure. YELLOW- upregulated miRNAs; BLU- downregulated miRNAs, all listed by increasing p-value. Data analyzed with *GeneGlobe* (*Qiagen*). Statistical significance: fold regulation |2| and p≤0.05 in OB ±GDM *vs* NW.

OB/GDM- *vs* NW		
miRNA ID	**Fold Regulation**	**p-value**
hsa-miR-27a-3p	2.13	0.0016
hsa-miR-324-5p	2.29	0.0018
hsa-miR-33a-5p	3.38	0.0042
hsa-miR-186-5p	-2.26	0.0155

OB/GDM+ *vs* NW		
miRNA ID	**Fold Regulation**	**p-value**
hsa-miR-454-3p	-2.32	0.0216

OB/GDM+ *vs* OB/GDM-		
miRNA ID	**Fold Regulation**	**p-value**
hsa-miR-186-5p	2.13	0,0065
hsa-miR-320d	2.18	0.0148
hsa-miR-2110	2.04	0.0196
hsa-let-7b-5p	2.09	0.0260
hsa-miR-574-3p	2.45	0.0266
hsa-miR-320c	2.26	0.0377
hsa-miR-324-5p	-2.75	0.0023
hsa-miR-142-3p	-3.03	0.0115
hsa-miR-33a-5p	-4.09	0.0144
hsa-miR-21-5p	-12.16	0.0162
hsa-miR-27a-3p	-2.02	0.0186
hsa-let-7f-5p	-3.45	0.0294
hsa-miR-30e-3p	-4.57	0.0403
hsa-miR-339-5p	-2.46	0.0467