

## **Streszczenie w języku angielskim**

Gestational diabetes mellitus (GDM) is one of the most common metabolic disorders during pregnancy. Undiagnosed and untreated GDM leads to serious complications in both the mother and the child. Currently, diagnostics are based on an oral glucose tolerance test performed between 24 and 28 weeks of pregnancy according to the criteria proposed by the IADPSG (International Association of Diabetes and Pregnancy Study Group).

Determination of changes in the miRNA expression profile and the concentration of individual ceramides in the serum in the first trimester of pregnancy may contribute not only to a better understanding of GDM pathogenesis, but also to the identification of clinically useful genetic and biochemical biomarkers for the early implementation of prophylaxis.

The aim of the study was to investigate the concentration of ceramides and their metabolites in the serum of pregnant women in the first and second trimesters of pregnancy compared to the control group, assessing whether they may constitute early biomarkers of gestational diabetes. Moreover, with the same intention, an examination of the miRNA expression profile in the serum of pregnant women in the first trimester of pregnancy was performed. An additional goal was to check the relationship between the measured parameters and the indicators of insulin resistance.

In the studies on sphingolipid concentrations, the study group (GDM) included women who had normal glucose tolerance in the first trimester (9-12 weeks) and developed gestational diabetes in the second trimester (24-26 weeks) ( $n = 53$ ). The reference group (NGT) was comprised by women with normal glucose tolerance during pregnancy ( $n = 82$ ). Additionally, non-pregnant women (control group) ( $n = 37$ ) were included in the study. The concentration of sphingolipids in pregnant women was measured in serum in the first and second trimesters and once in non-pregnant women. Expression of circulating miRNAs was determined in the serum of pregnant women in the first trimester of pregnancy. The study group (GDM) constituted of women who developed gestational diabetes in the second trimester ( $n = 24$ ), while in the control group (NGT) were women with normal glucose tolerance throughout pregnancy ( $n = 24$ ).

The concentration of ceramides and their metabolites in the serum was determined using ultra-high performance liquid chromatography combined with a triple quadrupole mass spectrometer (UHPLC / MS / MS). The miRNA profile in the serum of the patients was determined using NanoString technology, and validation of obtained results was performed using the RT-PCR method.

In the serum of pregnant women, significantly higher concentrations of C16:0-Cer, C18:1-Cer, C22:0-Cer, C24:1-Cer and C24:0-Cer were observed, as well as lower levels of SPA and S1P compared to the control group. The comparison of the sphingolipid concentration in the serum of the GDM group with the NGT group showed a higher concentration of C18:1-Cer in the serum of patients diagnosed with gestational diabetes in the second trimester. Across the study population, a negative correlation was observed between the concentration of SPA and the HOMA-IR and HOMA- $\beta$ . Moreover, the concentration of C22:0-Cer and C24:0-Cer positively correlated with the concentration of insulin, HOMA-IR and HOMA- $\beta$ . In addition, the concentration of C18:1-Cer and C24:1-Cer positively correlated with the concentration of insulin and HOMA- $\beta$ .

Analysis of the miRNA profile using the NanoString method allowed to select 4 miRNA molecules, the expression of which was significantly different in women diagnosed with GDM compared to NGT. Based on this analysis, the expression of miR-16-5p, miR-142-3p, miR-144-3p and miR-320e was assessed by RT-PCR. The validation showed significant differences between the groups in the expression of miR-16-5p, miR-142-3p and miR-144-3p. Further analysis showed a positive correlation between miR-16-5p and HOMA-IR and a negative correlation between ISIOGTT and miR-16-5p and miR-144-3p.

Based on obtained results, the following conclusions were drawn:

1. Pregnancy is associated with a significant increase in serum concentrations of C16:0-Cer, C18:1-Cer, C22:0-Cer, C24:1-Cer and C24:0-Cer.
2. The determination of C18:1-Cer concentration in the first trimester of pregnancy may be a new marker of the development of gestational diabetes.
3. Changes in the concentrations of C18:1-Cer, C22:0-Cer, C24:0-Cer and C24:1-Cer may be associated with increasing insulin resistance in pregnancy.
4. The miRNA profile during the first trimester of pregnancy is different in patients who developed gestational diabetes during pregnancy compared to healthy pregnant women.
5. miR-16-5p, miR-142-3p and miR-144-3p can potentially serve as markers for early diagnosis of GDM.