





degradation. A large number of miRNAs have been identified and/or predicted, and in humans evidence indicate to a key regulatory function for microRNAs (miRNAs) in gene expression regulation. Further investigation of microRNA target genes may manifest the molecular pathogenesis of these diseases as well as identify potential therapeutic targets and diagnostic biomarkers. About 30% of human genes are regulated by miRNAs (14). More than 1500 human microRNAs have been identified experimentally or by bioinformatics (15). As a member of nonprotein coding short RNAs family miRNAs through binding to the 3' untranslated region of specific target genes regulate their expression through translational regression. Important molecules involved in miRNA biogenesis, such as Droscha, Exportin 5, Dicer, Argonaute 2 (Ago 2) and DP103, have also been known in trophoblast cells (16) confirming that the miRNA biogenesis pathway is active in human placenta. MiRNA expression in the placenta is regulated by environment factors (17), signaling pathways and epigenetic modification (18).

MicroRNAs regulate mRNA, which encodes proteins that modulate cellular functions, therefore, miRNAs play key roles in physiological homeostasis in health and pathophysiological states in disease. MicroRNAs are known to have function in pathological process and prognosis of diseases such as diabetes (19), neurodegenerative diseases (20), preeclampsia (21), cancer and its resistance against chemotherapy (22). It has been also believed that the presence of single nucleotide polymorphism (SNP) in the processing machinery and target binding sites genes of miRNA affects cancer risk, treatment efficacy and prognosis of some diseases.

The first study that linked miRNA and PE was done by Pineles et al (23). The study was conducted to determine whether PE and small-for-gestational age (SGA) are associated with aberrations in placental miRNA expression. Thus they evaluated placental miRNAs' expression from patients with PE, SGA, PE + SGA along with a control group (Table 1).

They found that seven miRNAs (miR-210, miR-155, miR-181b, miR-182\*, miR-200b, miR-154\*, and miR-183) had higher expression between PE + SGA and the control group. The expression of

miR-182 and miR-210 was significantly higher in PE than in the control group.

Some studies have reported the expression of a number of miRNAs in placentas and fetal membranes with altered expression in these tissues affected by preeclampsia (10,11). The association between PE and altered miRNA expression indicates the possibility of a functional role for miRNA in this disease (13). MicroRNAs produced by human trophoblast cells could be secreted into maternal plasma or serum via an exosome-mediated pathway and have the potential to be used as biomarkers (24). Detection of miRNAs in the maternal circulation consider the possibility of using miRNAs as biomarkers to monitor the progression of normal pregnancy and gestational diseases such as preeclampsia. Aberrant expression of miRNAs in placenta from compromised pregnancies also discuss the potential of using miRNAs as therapeutic targets.

#### Placental Specific miRNAs

Many miRNAs are expressed in human placenta and some of them, such as the C19MC and C14MC clusters, are specifically or preferentially expressed in the placenta (25). The three most famous clusters are the chromosome 19 miRNA cluster (C19MC), C14MC and miR-371-3 cluster, which is also localized on chromosome 19. MiRNA members of these clusters are not only detected in the placenta, but also in other compartments, e.g. in serum where they have the potential to become novel biomarkers of pregnancy disorders. Antagonism of some of these miRNAs or their targets may lead to novel therapeutic points for the development of new drug classes in pregnancy disorders (26). The C19MC, located in chromosome 19q13.41, is the largest miRNA cluster identified to date and is encoded by paternally imprinted genes. This cluster 46 pre-miRNAs transcribed from a non protein coding host gene and expressed only in the placenta (27).

The C19MC is primate specific and expressed from the paternal harbors allele (28). On the other hand, the C14MC cluster, containing 46 miRNAs in 14q32, is also highly expressed in the human placenta but is encoded by maternally imprinted genes (29). MicroRNAs 518b belongs to C19MC.

MiR-154\* is located on 14q32.31 and has predominant expression pattern from placenta tissue. It is one of the pregnancy-associated

miRNAs and showed major decreased concentrations in maternal plasma after pregnancy (30).

Luo *et al.* 2008 found that miRNAs are exported from the human placental syncytiotrophoblast into maternal circulation, where they could target maternal tissues (24). Miura *et al.* (2010) showed that as the pregnancy progressed into the third trimester, the plasma concentrations of cell-free chromosome 19-derived miRNAs (has-miR-515-3p, has-miR-517a, has-miR-517c, has-miR-518b, and has-miR-526b) enhanced significantly, whereas that of cell-free has-miR-323-3p on chromosome 14q32.31 showed no change (31).

Hromadnikova *et al.* (2012) showed that both quantification approaches revealed significant up regulation in extracellular placenta-specific miRNA levels over time in women with normally progressing pregnancies; however, they did not have the capacity to differentiate between normally progressing and complicated pregnancies at the time of preeclampsia and/or IUGR onset. Also, significant elevation of extracellular miRNA levels was observed during early gestation (ie, within the

12th to 16th weeks) in women who later developed preeclampsia and/or IUGR (32). In Kotlabova *et al.* study (2011) seven microRNAs (miR-516-5p, miR-517\*, miR-518b, miR-520a\*, miR-520h, miR-525 and miR-526a) were identified as new pregnancy associated microRNAs with diagnostic potential but they mentioned only miR-518b had all criteria for selection as microRNA marker with diagnostic and or prognostic potential because of: 1) detection rate of 100% in full-term placentas, (2) detection rate of  $\geq 67\%$  in maternal plasma throughout gestation (at least four positive wells out of six tested wells) and (3) not detectable in blood and plasma samples of nonpregnant women (33). A quantitative real-time RT-PCR analysis of the expression of eight placenta-specific miRNAs in trophoblast cells with different proliferative activities by WANG *et al.* (2012) indicated that the expression levels of two miRNAs (miR-517b and miR-1283) were increased and miR-519a was decreased in trophoblast cells with lower proliferative activities (34).

**Table 1. Expression of some microRNAs in placenta**

| miR name  | Sample             | Expression      | References   |
|---|--------------------|-----------------|--|
| 210, 155, 181b, 182, 200b, 154, 183, 493, 372, 34c, 195   | Placenta           | High Expression | Pineles <i>et al.</i> 2007<br>Enquobahrie <i>et al.</i> 2010<br>Mayor-Lynn <i>et al.</i> 2010<br>Hu <i>et al.</i> 2009 |
| 16, 29b, 195, 26b, 181a, 335, 222, 181a, 584, 30a-3p, 210, 152, 517, 518b, 519e, 638, 296, 362, 512-3p            | severe PE placenta | High Expression | Hu <i>et al.</i> 2009<br>Zhu <i>et al.</i> 2009<br>Mayor <i>et al.</i> 2011  |
| 328, 584, 139-5p, 500, 1247, 340-5p, 1, 15b, 181a, 210, 483-5p, 154*, 34c-5p                                      | Placenta           | Low Expression  | Enquobahrie <i>et al.</i> 2010<br>Mayor-Lynn <i>et al.</i> 2010<br>Zhu <i>et al.</i> 2009<br>Daniel <i>et al.</i> 2011 |
| 101, 10b, 218, 590, 204, 32, 126, 18a, 19a, 411, 377, 154, 625, 144, 195, 150, 1, 18b, 363, 342-3p, 450, 223, 374 | severe PE placenta | Low Expression  | Zhu <i>et al.</i> 2009   |

## Conclusion:

MicroRNAs (miRNAs) are expressed in the human placenta and could be detected in maternal plasma. Specific miRNA profiles have been explained for the placenta, maternal plasma and pregnancy disorders such as PE. It has been observed that numerous miRNAs, which are predominantly or exclusively expressed during pregnancy in placenta, are clustered in chromosomal regions, may be controlled by the same promoters, may have similar seed regions and targets, and work synergistically. MiRNA members of these clusters are not only detected in the placenta, but also in serum where they have the potential to become novel biomarkers for early detection of pregnancy disorders. Antagonism of selected miRNAs or their targets may lead to novel targets for the development of new drug classes in pregnancy disorders such as PE.

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