Sex-specific ncRNAs Expression in Placenta

Subjects: Developmental Biology

Contributor: CARMELA SANTANGELO

The placenta is a temporary and sex-specific endocrine organ that regulates maternal–fetal exchanges, with a central role in fetal growth and development. Sex differences in placenta become evident from the beginning of pregnancy. Specifically, a male fetus grows faster than a female fetus, and it invests more in fetal than placental development. Consequently, male placentas are smaller in size and more vulnerable to adverse events during pregnancy than female placentas. In this context, an increasing number of placental noncoding RNAs contributes to fetal development and pregnancy progression, by regulating gene expression in a sexual dimorphic manner.

ncRNA miRNA pregnancy complications placenta

1. Introduction

Pregnancy is a physiological state in which numerous and complex modifications occur in the mother in order to accommodate the growing fetus [1]. According to the theory of the fetal origin of adult diseases (FOAD), hypothesized by David Barker, the intrauterine environment plays a relevant role in fetal growth and development and markedly contributes to the risk of developing diseases later in life [2]. It is well known that fetal growth and development occur in a sexually dimorphic manner [3]. However, growing evidence indicates that fetal sex differently influences the course of pregnancy and associated complications, as well maternal health [4][5][6][7]. Although ethnic differences exist in the maternal response, several data indicate that women carrying a male show an increased risk of gestational diabetes (GDM), placental abruption [7][8], and preterm birth [9], whereas gestational hypertension/preeclampsia (PE) appear to be linked to carrying a female fetus [10]. Genetics, nutrition, and other known and not yet completely known environmental factors contribute to successful pregnancy [1][11]. Deregulation of the complex interactions between these factors is responsible for the onset of pregnancy-related complications with long-term impact on offspring health until adulthood, as well as on the mother's health [1][11]. Despite it being known that the biological differences between sexes become evident early in embryogenesis, the processes involved in these sex differences are not completely known [9]. Knowledge about the mechanisms through which female and male fetuses respond to external and internal stimuli or insults during pregnancy is rather fragmentary [12]. Embryo development requires proliferation and differentiation processes that are tightly regulated in time and duration [12][13]. Several mechanisms and numerous factors, including an increasing number of noncoding RNAs (ncRNAs), modulate gene transcription at every stage of embryo and fetus development [13] and pregnancy progression [14]. Non-coding RNAs play a significant role in epigenetic modifications and exhibit dynamic expression patterns in the regulation of biological processes [13]. They are expressed by almost all cells, allowing cell-to-cell interactions, and are detected in body fluids [15]. In recent years, ncRNAs have been studied as promising diagnostic and prognostic factors, as well as being studied as therapeutic targets in tumors [15][16] and

neurodegenerative and metabolic disorders [17], thus representing an area of intensive ongoing investigation in the context of human physiology and pathophysiology. The growing evidence of the importance of sexual dimorphism in health and disease imposes a wider knowledge of the biological processes underlying sex differences.

2. The Importance of Sexual Dimorphism in Placental ncRNAs Expression

The placenta is a temporary and sex-specific endocrine organ that regulates maternal–fetal exchanges, with a central role in fetal growth and development [18]. Sex differences in placenta become evident from the beginning of pregnancy. Specifically, a male fetus grows faster than a female fetus already in the pre-implantation phase, and it invests more in fetal than placental development [19]. Consequently, male placentas are smaller in size and more vulnerable to adverse events during pregnancy than female placentas [19]. These findings indicate that placental adaption to maternal environmental stimuli can influence fetus programming with long-term effects [20]. Of note, the plasticity of the placenta allows fetal signals to influence in a sex-specific manner maternal health as well [4]. However, the complex interaction between placental function and fetal development is not well known. To this end, in the last few years, several studies have tried to shed light on the mechanisms related to sex-specific fetus adaptation during pregnancy by using omics approaches as well [21]. Metabolomics of mouse placentas showed that sexual dimorphism contributes to a different placental adaptation to nutrition, stress, hormones, and chemicals to ensure normal fetal development [21]. A growing amount of data has shown that placental miRNAs and lncRNAs have a regulatory role in fetal developmental programming [22] and pregnancy progression [23][24], as well as in genomic imprinting and susceptibility to diseases later in life [25], by influencing many signaling pathways (**Table 1**).

Table 1. Sex-specific ncRNAs expression in placentas.

Model	ncRNAs	Sex Regulation Female ♀ vs. Male ♂ Fetus	Targeted Genes/Processes	Study
Overweight and obese pregnant women	miRNAs	♀ (↑) miR-210	♀ (↑) TNFα and NFκB1; mitochondrial dysfunction	[26]
Increasing maternal pre-pregnancy BMI	miRNAs	♀ (↓) miR-20a, miR-34a, miR-146a, miR-210, and miR-222	♀♂ Cell proliferation, cell growth and invasion, inflammation, angiogenesis, and oxidative stress	[27]
Healthy human pregnancies	miRNAs	♀ (↑) miR-21, miR-34a, miR-146a, miR-210, and miR-222	♀♂ Inflammation, oxidative stress, cell cycle, cellular senescence, and apoptosis	[28]

			♀ longer placental relative telomere length	
Human low gestational age newborns (<28 weeks gestational age)	miRNAs	♀ (↑) 27 miRNAs, including miR-323b, miR-15a, and miR-223 ♂ (↑) 32 miRNAs, including miR-23a, miR-543, miR-495, miR-323a, miR-30a, miR-155, and miR-222	♀♂ Embryo implantation and development ♀ X chromosome inactivation, epigenetic control (HDAC1, HDAC8), and cell cycle progression (CDKN1B)	[29]
Human low gestational age newborns (<28 weeks gestational age)	miRNAs	♀ (↑) miR-4747-5p, miR-3674, miR-4252, miR-4461, miR-1285-5p, miR-4421, miR-4728-3p, miR-6081, miR-6890-3p, iR-564, miR-1225-3p, miR-541-3,p miR-3187-5p, miR-1976, miR-548w, miR-5589-5p, miR-889-3p, miR-4734 ♂ (↑) miR-4057, (↓) miR-128	♀♂ Underweight status ♀ No target genes associated with underweight status ♂ regulate 43 genes related to nutrition, growth, and angiogenesis	[30]
Healthy human pregnancies	miRNAs	♀ 76 (↑), and 66 (↓) miRNAs, most significant miR-361-5p ♀ (↑) miR-361-5p, miR-139-5p, miR-4769-3p♀ (↑) miR-520e, miR-518d-3p, miR-331-3p, miR-423-3p, and miR-330-3p ♀ (↑) miR-4287 ♀ (↓) miR-1207-5p, miR-4530, and miR-4687-3p ♀ (↓) miR-4299	♀♂ Metabolic, immunological, apoptotic, and neurogenesis processes underlying sex differences (↓) *PCDH11X* *(↓) CD99 gene* *(↓) DDX3Y* *(↑) KDM6A* *(↑) CDK16*	[31]
Healthy pregnancy from women with/without daily supplement with n-3 LCPUFA	miRNAs	♂ (↑) basal miR-99a ♀ (↑) miR-99a following supplementation	♀♂ Maternal-fetal amino acids homeostasis ♂ ↑ basal mTOR and SLC7A5 mRNA ♀ ↑ mTOR and SLC7A5 mRNA following supplementation	[32]
Healthy pregnancies from major ethnic groups: African Americans, European Americans, South Asians, and East Asians	miRNAs	♂ (↑) 14 miRNAs 371a-5p,372-3p,181b-2-3p, let-7g-3p, 185-3p, 3615, and 3158-3p ♀ (↑) 18 miRNAs	♂ glutamate receptor signaling and endocrine processes ♀ steroid hormones, estradiol, and glucocorticoid response; differentiation and metabolic processes	[33]

↑ Increases; ↓ decreases; ♀ female; ♂ male; BMI, body mass index; CD99, cluster of differentiation 99; CDK16, cyclin-dependent kinase 16; CDKN1B, cyclin-dependent kinase inhibitor 1B; DDX3Y, DEAD-box helicase 3 Y-

linked; HDAC, histone deacetylase; KDM6A, lysine demethylase 6A; miRNA, microRNA; mTOR, mammalian target of rapamycin; ncRNA, non-coding RNA; NFκB, nuclear transcription factor kappa B; PCDH11X, protocadherin 11 X-linked; n-3 LCPUFA, n-3 long-chain polyunsaturated fatty acids; SLC7A5, solute carrier family 7 member 5; TNFα, tumor necrosis factor α.

Sex-dependent miRNA and mRNA expression has been observed in placentas from the extremely low gestational age newborn cohort (ELGAN study; < 28 weeks gestational age). Genome-wide RNA-sequencing analysis showed that 59 miRNAs were differentially expressed between sexes. Of these, miR-23a, miR-543, miR-495, miR-323a, miR-30a, miR-155, and miR-222 were upregulated in male, and miR-323b, miR-15a, and miR-223 were increased in female placentas. All of them are differently involved in placental functions. Specifically, miR-23a, miR-495, miR-222, and miR-223 target genes were associated with PE, and miR-323a with ectopic pregnancy. Interactome analysis revealed that the mRNAs upregulated in female placentas included genes involved in epigenetic control (HDAC1, HDAC8) and cell cycle progression (CDKN1B). A large number of these miRNAs are located on the X-chromosome, suggesting for some of them the escape from XCI (X chromosome inactivation). It is worth noting that miR-543, miR-495, miR-323a, and miR-323b are imprinted miRNA genes belonging to the maternally expressed C14CM miRNA cluster involved in prenatal growth and placentation [29]. Recently, the same groups examined sexually dimorphic mRNA–miRNA expression in relation to pre-pregnancy BMI in placentas from ELGAN study subjects. Data showed that 572 mRNAs associated with pre-pregnancy underweight status in male placentas. Almost all of them (96%) were overexpressed and were involved in known regulatory networks related to nutrition, growth, and angiogenesis, including eukaryotic initiation factor 2 (EIF2) signaling, mammalian target of rapamycin (mTOR) signaling, insulin-like growth factor 1 (IGF-1) signaling, and vascular endothelial growth factor (VEGF) signaling. Of note, only 43 of 572 mRNAs were targets of the two miRNA (miR-4057 and miR-128) that were associated with underweight status in male placentas. In female placentas 18 overexpressed miRNA (miR-4747-5p, miR-3674, miR-4252, miR-4461, miR-1285-5p, miR-4421, miR-4728-3p, miR-6081, miR-6890-3p, iR-564, miR-1225-3p, miR-541-3,p miR-3187-5p, miR-1976, miR-548w, miR-5589-5p, miR-889-3p, miR-4734) were associated with underweight status. The differentially expressed genes in male placentas were also expressed in female placentas. However, none of the mRNAs were associated with underweight status, nor were they a target of the 18 overexpressed miRNAs in female placentas. The authors hypothesized that the altered gene expression in male placentas can represent a protection against intrauterine growth restriction. None of the genes were differentially expressed in overweight and obese status. Of note, the authors confirmed the association between miR-210 and overweight status [30].

Sedlmeier and colleagues revealed a sex-specific response to maternal supplementation with n-3 long-chain polyunsaturated fatty acid (LCPUFA) during pregnancy (INFAT study). They observed that n-3 LCPUFA supplementation had a more evident impact on gene expression in female placentas than in male [34]. The same authors analyzed the effect of n-3 LCPUFA supplementation on placental miRNA expression in a sex-specific manner. They highlighted a differential expression of miR-99a between sexes. MiR-99a can target the expression of mTOR , SLC7A5 , and SLC6A6 genes, encoding the nutrient sensor mTOR and the amino acid transporters LAT1 and TauT, respectively. The authors observed that miR-99a, mTOR , and SLC7A 5 mRNA levels were higher in male placentas compared with female ones, and n-3 LCPUFA supplementation stimulated elevated levels of

miR-99a, mTOR, and SLC7A5 mRNA only in female placentas. Amino acids levels were analyzed in maternal plasma (at 15 and 32 weeks of gestation), placenta, and cord plasma. Increased tryptophan (Trp) levels were found in maternal plasma (32 weeks), with significantly higher levels in mothers with female offspring, whereas lower levels of taurine (Tau) were detected in cord plasma of male offspring after supplementation. Significant associations were found for Trp and Tau and for miR-99a mTOR, SLC7A5 mRNA mTOR, and miRNA-99a with offspring body composition. No amino acids changes were detected in placenta after supplementation. The authors concluded that miRNA-99a could be a novel regulator of a complex network in placenta and in maternal and fetal compartments, regulating amino acids homeostasis and offspring's body composition [32].

Sex-specific ncRNAs expression in placentas.

3. Sex-Based ncRNAs Expression in Fetal Development

Embryo and fetus growth and development are tightly regulated processes involving embryonic genome and transcriptional sexual dimorphism as well as epigenetic and maternal factors that control the sequence of stage-specific changes leading to a fully developed organism [13]. In this complex context, an increasing number of ncRNAs appear to regulate gene transcription at every stage of embryo and fetus development [35]. Several studies have shown the importance of miRNAs and piRNAs in human reproduction [13]. Specifically, they are involved in the regulation of pre- and post-fertilization stages, such as sex-specification, commitment, and maintenance of mammalian gonads; embryonic implantation and early development; silencing of X-linked gene expression; and maternal-embryo cross talk [36][35]. The increasing knowledge about stage-specific ncRNA profiles during fetal development is expected to take a step forward in improving assisted reproductive technology (ART) programs as well [35]. Various studies have shown that a differential expression of certain miRNAs was associated with implantation failure [37]. Moreover, an aberrant expression of miRNAs has been observed in reproductive cells/tissues in the context of infertility in humans, both in females [38][39] and males [38], indicating a fundamental role of these ncRNAs in the de-regulation of the reproductive system and early embryogenesis (**Table 2**).

Table 2. Sex-specific ncRNAs expression in fetal development.

Model/Tissue	ncRNAs	Sex Regulation Female ♀ vs. Male ♂ Fetus	Targeted Genes/Processes	Study
Cryopreserved human embryos (day 5 of development)	miRNAs	♀ (↑) miR-182, miR-206, miR-500, miR-601, miR-604, miR-875-5p ♂ (↑) miR-140-5p, miR-149, miR-151-5p, miR-26b, miR-31, miR-362-3p, miR-512-3p, miR-512-5p, miR-518d-5p, miR-518e, miR-525-3p, miR-886-3p, miR-886-5p, miR-92a	♀♂ Embryo development, cell cycle, and apoptosis	[40]

Mouse embryonic stem cells	miRNAs	♂ (↑) miR-302a, miR-302b, miR-302c, miR-302d, at differentiation day 5 ♂ (↓) miR-302, at differentiation day 10	♀♂ Chromatin remodeling; proliferation, differentiation, and cell fate determination	[41]
Human feto-placental endothelial cells from healthy pregnancies	miRNAs	♀ (↑) miR-23a-3p, miR-222-5p, miR-181a-3p, miR-151a-3p, miR-4286	♀♂ Endothelial barrier function: adherent junction, ECM receptor interaction, and focal adhesion;	[42]
		♂ (↑) miR-29b-3p, miR-15b-5p, miR-431-5p, miR345-5p	♂ cells have increased barrier resistance compared with female cells	

↑ Increases; ↓ decreases; ♀ female; ♂ male; ECM, extracellular matrix; miRNA, microRNA; ncRNA, non-coding RNA.

Indeed, different miRNA profiles seem to reflect embryonic ploidy status (i.e., euploid vs. aneuploid), as well as sexual dimorphism, in human blastocysts (**Table 2**). It has been shown that miR-140-5p, miR-149, miR-151-5p, miR-26b, miR-31, miR-362-3p, miR-512-3p, miR-512-5p, miR-518d-5p, miR-518e, miR-525-3p, miR-886-3p, miR-886-5p, miR-92a, and RNU48 were highly expressed in cryopreserved human male embryos (day 5 of development) from in vitro fertilization (IVF), and miR-182, miR-206, miR-500, miR-601, miR-604, and miR-875-5p were highly expressed in the female ones. Genes targeted by these miRNAs belong to several pathways that are essential for embryo development, cell cycle, and apoptosis, indicating the importance of ncRNAs in these processes and their possible role as diagnostic biomarkers of infertility and prognostic tools of embryo development [40].

In another complex study, sex-specificity was identified in miRNA expression in mouse embryonic stem cells (ES) at different day (D) of differentiation (i.e., D0, D2, D5). Specifically, the authors highlighted that the miR-302 genomic cluster (i.e., miR-302a, miR-302b, miR-302c, miR-302d) was more expressed in male ES at D5, compared with the female. The authors identified as putative miRNA targets the chromatin remodeling and the E2F-dependent transcription repressors Ari4a and Arid4b, involved in diverse cellular processes including proliferation, differentiation, and cell fate determination. Decreased miR-302 level at D10 in male ES suggested a specific role of this miRNA during a narrow window of male development [41]. By using next-generation miRNA sequencing, sex-dependent miRNA expression patterns were identified in feto-placental endothelial cells (fpEC) obtained from healthy pregnancies. Nine miRNAs were differentially expressed in male and female fpEC; specifically, miR-29b-3p, miR-15b-5p, miR-431-5p, miR345-5p, were overexpressed in male, and miR-23a-3p, miR-222-5p, miR-181a-3p, miR-151a-3p, miR-4286 were overexpressed in female. The analysis of the potential function of these miRNAs indicated their involvement in endothelial barrier function regulation. Specifically, they modulated adherent junctions, extracellular matrix (ECM) receptor interactions, and focal adhesion, eventually suggesting that male cells have increased barrier resistance compared with female ones. The authors hypothesized that the distinct features of endothelial cells already in utero can have a role in sexual dimorphism of

endothelial dysfunction and cardiovascular disease in male versus female [42]. However, the advance in high-throughput sequencing technology, together with sexually dimorphic gene expression analysis, represent valuable tools for explaining the basis of sex differences in fetal programming and development and in the susceptibility to diseases.

References

1. Hsu, C.N.; Tain, Y.L. The Good, the Bad, and the Ugly of Pregnancy Nutrients and Developmental Programming of Adult Disease. Nutrients 2019, 11, 894.
2. Barker, D.J. Developmental origins of adult health and disease. J. Epidemiol. Community Health 2004, 58, 114–115.
3. Clifton, V.L. Review: Sex and the human placenta: Mediating differential strategies of fetal growth and survival. Placenta 2010, 31, S33–S39.
4. Vari, R.; Scazzocchio, B.; Silenzi, A.; Giovannini, C.; Masella, R. Obesity-Associated Inflammation: Does Curcumin Exert a Beneficial Role? Nutrients 2021, 13, 21.
5. Cowell, W.; Colicino, E.; Askowitz, T.; Nentin, F.; Wright, R.J. Fetal sex and maternal postpartum depressive symptoms: Findings from two prospective pregnancy cohorts. Biol. Sex Differ. 2021, 12, 6.
6. Enninga, E.A.; Nevala, W.K.; Creedon, D.J.; Markovic, S.N.; Holtan, S.G. Fetal sex-based differences in maternal hormones, angiogenic factors, and immune mediators during pregnancy and the postpartum period. Am. J. Reprod. Immunol. 2015, 73, 251–262.
7. Funaki, S.; Ogawa, K.; Ozawa, N.; Okamoto, A.; Morisaki, N.; Sago, H. Differences in pregnancy complications and outcomes by fetal gender among Japanese women: A multicenter cross-sectional study. Sci. Rep. 2020, 10, 18810.
8. Broere-Brown, Z.A.; Adank, M.C.; Benschop, L.; Tielemans, M.; Muka, T.; Goncalves, R.; Bramer, W.M.; Schoufour, J.D.; Voortman, T.; Steegers, E.A.P.; et al. Fetal sex and maternal pregnancy outcomes: A systematic review and meta-analysis. Biol. Sex Differ. 2020, 11, 26.
9. Alur, P. Sex Differences in Nutrition, Growth, and Metabolism in Preterm Infants. Front. Pediatrics 2019, 7, 22.
10. Yu, T.; Chen, T.S.; Liang, F.W.; Kuo, P.L. Does sex matter? Association of fetal sex and parental age with pregnancy outcomes in Taiwan: A cohort study. BMC Pregnancy Childbirth 2020, 20, 348.
11. Franzago, M.; Santurbano, D.; Vitacolonna, E.; Stuppia, L. Genes and Diet in the Prevention of Chronic Diseases in Future Generations. Int. J. Mol. Sci. 2020, 21, 2633.

12. DiPietro, J.A.; Voegtline, K.M. The gestational foundation of sex differences in development and vulnerability. Neuroscience 2017, 342, 4–20.
13. Kamalidehghan, B.; Habibi, M.; Afjeh, S.S.; Shoai, M.; Alidoost, S.; Almasi Ghale, R.; Eshghifar, N.; Pouresmaeili, F. The Importance of Small Non-Coding RNAs in Human Reproduction: A Review Article. Appl. Clin. Genet. 2020, 13, 1–11.
14. Ali, A.; Hadlich, F.; Abbas, M.W.; Iqbal, M.A.; Tesfaye, D.; Bouma, G.J.; Winger, Q.A.; Ponsuksili, S. MicroRNA-mRNA Networks in Pregnancy Complications: A Comprehensive Downstream Analysis of Potential Biomarkers. Int. J. Mol. Sci. 2021, 22, 2313.
15. Szilagyi, M.; Pos, O.; Marton, E.; Buglyo, G.; Soltesz, B.; Keseru, J.; Penyige, A.; Szemes, T.; Nagy, B. Circulating Cell-Free Nucleic Acids: Main Characteristics and Clinical Application. Int. J. Mol. Sci. 2020, 21, 6827.
16. Reda El Sayed, S.; Cristante, J.; Guyon, L.; Denis, J.; Chabre, O.; Cherradi, N. MicroRNA Therapeutics in Cancer: Current Advances and Challenges. Cancers 2021, 13, 2680.
17. Lopez-Jimenez, E.; Andres-Leon, E. The Implications of ncRNAs in the Development of Human Diseases. Non-Coding RNA 2021, 7, 17.
18. Rosenfeld, C.S. Sex-Specific Placental Responses in Fetal Development. Endocrinology 2015, 156, 3422–3434.
19. Dearden, L.; Bouret, S.G.; Ozanne, S.E. Sex and gender differences in developmental programming of metabolism. Mol. Metab. 2018, 15, 8–19.
20. Cowell, W.; Deyssenroth, M.; Chen, J.; Wright, R.J. Maternal stress in relation to sex-specific expression of placental genes involved in nutrient transport, oxygen tension, immune response, and the glucocorticoid barrier. Placenta 2020, 96, 19–26.
21. Saoi, M.; Kennedy, K.M.; Gohir, W.; Sloboda, D.M.; Britz-McKibbin, P. Placental Metabolomics for Assessment of Sex- specific Differences in Fetal Development During Normal Gestation. Sci. Rep. 2020, 10, 9399.
22. Mouillet, J.F.; Chu, T.; Sadovsky, Y. Expression patterns of placental microRNAs. Birth Defects Res. A Clin. Mol. Teratol. 2011, 91, 737–743.
23. Medina-Bastidas, D.; Guzman-Huerta, M.; Borboa-Olivares, H.; Ruiz-Cruz, C.; Parra-Hernandez, S.; Flores-Pliego, A.; Salido-Guadarrama, I.; Camargo-Marin, L.; Arambula-Meraz, E.; Estrada-Gutierrez, G. Placental Microarray Profiling Reveals Common mRNA and lncRNA Expression Patterns in Preeclampsia and Intrauterine Growth Restriction. Int. J. Mol. Sci. 2020, 21, 3597.
24. Ali, A.; Bouma, G.J.; Anthony, R.V.; Winger, Q.A. The Role of LIN28-let-7-ARID3B Pathway in Placental Development. Int. J. Mol. Sci. 2020, 21, 3637.

25. Majewska, M.; Lipka, A.; Paukszto, L.; Jastrzebski, J.P.; Gowkielewicz, M.; Jozwik, M.; Majewski, M.K. Preliminary RNA-Seq Analysis of Long Non-Coding RNAs Expressed in Human Term Placenta. Int. J. Mol. Sci. 2018, 19, 1894.
26. Muralimanoharan, S.; Guo, C.; Myatt, L.; Maloyan, A. Sexual dimorphism in miR-210 expression and mitochondrial dysfunction in the placenta with maternal obesity. Int. J. Obes. 2015, 39, 1274–1281.
27. Tsamou, M.; Martens, D.S.; Winckelmans, E.; Madhloum, N.; Cox, B.; Gyselaers, W.; Nawrot, T.S.; Vrijens, K. Mother's Pre-pregnancy BMI and Placental Candidate miRNAs: Findings from the ENVIRONAGE Birth Cohort. Sci. Rep. 2017, 7, 5548.
28. Tsamou, M.; Martens, D.S.; Cox, B.; Madhloum, N.; Vrijens, K.; Nawrot, T.S. Sex-specific associations between telomere length and candidate miRNA expression in placenta. J. Transl. Med. 2018, 16, 254.
29. Eaves, L.A.; Phookphan, P.; Rager, J.E.; Bangma, J.; Santos, H.P., Jr.; Smeester, L.; O'Shea, T.M.; Fry, R.C. A role for microRNAs in the epigenetic control of sexually dimorphic gene expression in the human placenta. Epigenomics 2020, 12, 1543–1558.
30. Clark, J.; Eaves, L.A.; Gaona, A.R.; Santos, H.P.; Smeester, L.; Bangma, J.T.; Rager, J.E.; O'Shea, T.M.; Fry, R.C. Pre-pregnancy BMI-associated miRNA and mRNA expression signatures in the placenta highlight a sexually-dimorphic response to maternal underweight status. Sci. Rep. 2021, 11, 15743.
31. Tsamou, M.; Vrijens, K.; Wang, C.; Winckelmans, E.; Neven, K.Y.; Madhloum, N.; de Kok, T.M.; Nawrot, T.S. Genome-wide microRNA expression analysis in human placenta reveals sex-specific patterns: An ENVIRONAGE birth cohort study. Epigenetics 2021, 16, 373–388.
32. Sedlmeier, E.M.; Meyer, D.M.; Stecher, L.; Sailer, M.; Daniel, H.; Hauner, H.; Bader, B.L. Fetal sex modulates placental microRNA expression, potential microRNA-mRNA interactions, and levels of amino acid transporter expression and substrates: INFAT study subpopulation analysis of n-3 LCPUFA intervention during pregnancy and associations with offspring body composition. BMC Mol. Cell Biol. 2021, 22, 15.
33. Guo, S.; Huang, S.; Jiang, X.; Hu, H.; Han, D.; Moreno, C.S.; Fairbrother, G.L.; Hughes, D.A.; Stoneking, M.; Khaitovich, P. Variation of microRNA expression in the human placenta driven by population identity and sex of the newborn. BMC Genom. 2021, 22, 286.
34. Sedlmeier, E.M.; Brunner, S.; Much, D.; Pagel, P.; Ulbrich, S.E.; Meyer, H.H.; Amann-Gassner, U.; Hauner, H.; Bader, B.L. Human placental transcriptome shows sexually dimorphic gene expression and responsiveness to maternal dietary n-3 long-chain polyunsaturated fatty acid intervention during pregnancy. BMC Genom. 2014, 15, 941.

35. Timofeeva, A.; Drapkina, Y.; Fedorov, I.; Chagovets, V.; Makarova, N.; Shamina, M.; Kalinina, E.; Sukhikh, G. Small Noncoding RNA Signatures for Determining the Developmental Potential of an Embryo at the Morula Stage. Int. J. Mol. Sci. 2020, 21, 9399.
36. Reza, A.; Choi, Y.J.; Han, S.G.; Song, H.; Park, C.; Hong, K.; Kim, J.H. Roles of microRNAs in mammalian reproduction: From the commitment of germ cells to peri-implantation embryos. Biol. Rev. Camb. Philos. Soc. 2019, 94, 415–438.
37. Paul, A.B.M.; Sadek, S.T.; Mahesan, A.M. The role of microRNAs in human embryo implantation: A review. J. Assist. Reprod. Genet. 2019, 36, 179–187.
38. Salas-Huetos, A.; James, E.R.; Aston, K.I.; Jenkins, T.G.; Carrell, D.T.; Yeste, M. The Expression of miRNAs in Human Ovaries, Oocytes, Extracellular Vesicles, and Early Embryos: A Systematic Review. Cells 2019, 8, 1564.
39. Gebremedhn, S.; Ali, A.; Hossain, M.; Hoelker, M.; Salilew-Wondim, D.; Anthony, R.V.; Tesfaye, D. MicroRNA-Mediated Gene Regulatory Mechanisms in Mammalian Female Reproductive Health. Int. J. Mol. Sci. 2021, 22, 938.
40. Rosenbluth, E.M.; Shelton, D.N.; Sparks, A.E.; Devor, E.; Christenson, L.; Van Voorhis, B.J. MicroRNA expression in the human blastocyst. Fertil. Steril. 2013, 99, 855–861.
41. Ciaudo, C.; Jay, F.; Okamoto, I.; Chen, C.J.; Sarazin, A.; Servant, N.; Barillot, E.; Heard, E.; Voinnet, O. Correction: RNAi-Dependent and Independent Control of LINE1 Accumulation and Mobility in Mouse Embryonic Stem Cells. PLoS Genet. 2015, 11, e1005247.
42. Cvitic, S.; Strutz, J.; Appel, H.M.; Weiss, E.; Brandl, W.T.; Thuringer, A.; Bernhart, E.M.; Lassance, L.; Wadsack, C.; Schliefsteiner, C.; et al. Sexual dimorphism of miRNA signatures in feto-placental endothelial cells is associated with altered barrier function and actin organization. Clin. Sci. 2020, 134, 39–51.