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Fetoplacental-endothelial derived small extracellular vesicles (fp-sEVs) as modulators in the development of the fetal immune system

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Introduction: During pregnancy the placenta releases small extracellular vesicles (sEVs) into the maternal circuit. The main function of those placental sEVs is to maintain the immune tolerant environment towards the fetal allograft. The cell-to-cell communication between the placenta and the fetus is weakly understood, therefore, we aim to investigate the contribution of fetoplacental-endothelial derived sEVs (fp-sEVs) in the development of the fetal immune system. Methods: Primary fetoplacental-endothelial cells (fpECs) were isolated from placentae at term, preeclamptic (PE) and preterm (PT) pregnancies. fp-sEVs were enriched from media supernatants of fpECs by differential ultracentrifugation (100,000?g, 22 hours, 4°C pellet) and characterized by size (nanoparticle tracking analysis) and sEVs markers like ALIX, Syntenin, CD9, CD63 and CD81. In addition, a lectin microarray was used to profile glycosylation pattern of fp-sEVs and their parent fpECs cell membrane. Results: fpECs secrete fp-sEVs with a mean size of 125.3±3.9 nm (term, n=4), 162.4 nm (PT, n=1) and 153.4±20.44 nm (PE, n=3). Specific sEVs markers verified the presence of fp-sEVs. Moreover, on protein level we identified the placental specific marker Siglec-6, a sialic acid binding protein, and the endothelial marker CD31. The lectin microarray revealed that fpECs and fp-sEVs differ in their glycosylation profile in both term and PE. Compared to their parent fpECs, fp-sEVs are enriched with sialic acids, indicating a selective cargo loading in the sEVs biogenesis. Conclusion: These preliminary results suggest that the signature of fp-sEVs resembles their origin and the state of the parent fpECs. Based on the notion that glycans are important for intercellular communication; obtained sialic acid signatures on fp-sEVs may indicate an interplay of these vesicles and respective receptors on immune cells. Together, our findings could indicate an effective priming of the fetal innate immune system in utero.