Effect of visfatin on human placental endocrinology. *In vitro* studies on BeWo cell line and villous explants from third-trimester human pregnancy

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Visfatin, an adipocytokine primarily known for its role in energy metabolism, has been recently implicated in pregnancy with evidence suggesting that its levels fluctuate across individual trimesters. Our previous data indicated visfatin expression in the placental cell lines and different compartments of the human placenta. In this study, we aimed to investigate the in vitro effect of visfatin on placental cell endocrinology with underlying molecular mechanisms.

Using placental BeWo cell line and villi obtained from both normal and pathological pregnancies, including intrauterine growth restriction (IUGR), preeclampsia (PE), and gestational diabetes mellitus (GDM), we examined the dose (1-100 ng/mL) and time (24-72 h) dependent impact of visfatin on the secretion of key steroids (progesterone and estradiol) and protein hormones (human chorionic gonadotropin, human placental lactogen, and placental growth hormone (ELISA). We also investigated the effect of visfatin on the protein expression of steroid enzymes, including 3-β-hydroxysteroid dehydrogenase and aromatase, as well as the aforementioned protein hormones (Western blot). Moreover, we studied the visfatin effect on phosphorylation of extracellular signal-regulated kinase 1/2 (ERK1/2) and protein kinase A (PKA). In contrast, pharmacological inhibition of the above kinases was used to further explain visfatin-mediated effects on selected hormones secretion. Experiments were carried out at least three times, and statistical analysis was performed in GraphPad Prism 8 using Student t-test as well as ANOVA ($p<0.05$).

Our results provide comprehensive insights into the modulatory role of visfatin in the human placenta and identify key endocrine molecular players involved in this process. The findings underscore the complex interplay between adipocytokines and placental endocrinology, shedding light on potential therapeutic targets for pregnancy-related complications. This research contributes to a deeper understanding of the intricate mechanisms governing maternal-fetal interactions and may pave the way for developing novel interventions to improve pregnancy outcomes.

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