ADROPIN in polycystic ovarian syndrome: expression and impact on proliferation, apoptosis, steroidogenesis and signaling pathways in human granulosa cells.

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ADROPIN is a new adipokine, which regulates energy homeostasis and food intake. The serum and follicular fluid levels of adropin are decreased in women suffering from polycystic ovarian syndrome (PCOS), however its role in ovarian function is still unknow. The aims of the study were to determine the expression of adropin and its receptor G protein-coupled receptor 19 (GPR19) in human granulosa cells (Gc), its immunolocalization as well as direct *in vitro* effect on cell proliferation, apoptosis, steroidogenesis and signaling pathways.

The Gc were obtained from healthy women (normal weight (NW) and obese (OB)) and women diagnosed with PCOS (NW and OB) to determine *adropin/GPR19* mRNA expression. Paraffin ovarian slides from healthy NW women were used for its immunolocalization. Next, human Gc and KGN cells *in vitro* culture were performed to investigate the effects of recombinant adropin on cell proliferation/apoptosis (by alamarBlue and *PCNA, cyclin D, BAX/BCL2, caspase3, p53* mRNA and protein level) and steroidogenesis (by E2 secretion and mRNA and protein expression of *StAR* protein and enzymes: *CYP11A1, HSD3B, HSD17B, CYP17A1, CYP19A1)*. Additionally, adropin effect on MAP3/1, AKT, PKA and STAT3 phosphorylation was analyzed by Western blot. Statistical analyses were performed by Graph Pad Prism 5.

The mRNA expression of adropin in Gc between groups was stable, while level of *GPR19* mRNA was decreased in Gc from obese and PCOS women; adropin/GPR19 were localized in Gc, theca cells and oocyte. Also, adropin reduced cell proliferation and related gene expression (*PCNA* and *cyclin D*) in both human Gc and KGN, while increased cell apoptosis related genes level (*caspase3* and *BAX/BCL2* ratio). Additionally, adropin had a negative effect on steroidogenesis by inhibiting *STAR* and *CYP19A1* mRNA expression. The negative effect of adropin on ovarian function could be associated with a decrease in phosphorylation of AKT, PKA and STAT3. This effect of adropin was independent of the patient's condition ($P<0.05$, $n=6$).

The obtained data clearly indicate that adropin/GPR19 are present in human ovaries and its negative effect on ovarian physiology may be a medical target to contribute to restore fertility in PCOS suffering women.

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