P181 QUANTIFICATION OF TRANSFORMING GROWTH FACTOR b1 (TGF b1) mRNA EXPRESSION IN MOUSE PREIMPLANTATION EMBRYOS AND DETERMINATION OF TGFb RECEPTORS (TYPE I AND TYPE II) EXPRESSION IN MOUSE EMBRYOS AND REPRODUCTIVE TRACT

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We reported the quantification of TGFb1 mRNA in mouse embryos by competitive reverse transcription–polymerase chain reaction using RNA mimic. TGFb1 was first detected in the unfertilized oocyte, disappear after fertilization and expressed again at 2-cell stage (7.3±2.2 x 10-3 attomoles/embryo). It increases gradually and peaked at 8-cell stage (97.4±28.6 x 10-3 attomoles/embryo). The expression declines rapidly after morula stage and the concentration of TGFb1 mRNA at the blastocyst stage was 2.5±0.9 x 10-3 attomoles/embryo. The mRNA levels of TGFb1 at 8-cell and morula stages were significantly higher than that in other cell stages (p <0.05). TGFb receptors [Type I (ALK-5) and Type II] were detected in 1-cell, 2-cell, morula and blastocyst stage embryos by immunocytochemistry. Northern hybridization and immunohistochemistry showed a constant expression of both TGFb receptors in the oviduct from Days 1-4 of pregnancy, whilst there was a marked increase in the expression of TGFb Type I receptor in Day 3 uterus. Expression of TGFb Type II receptor in the uterus remained unaltered throughout the studied period. These observations indicate that preimplantation mouse embryos produce TGFb1, and that the embryos as well as the reproductive tract are responsive to TGFb1 in the preimplantation period. This study strengthens the hypothesis that there is a close interaction between the preimplantation embryo and the reproductive tract via TGFb1.

P182 IMMORTALIZED AND PRIMARY HUMAN OVIDUCTAL EPITHELIAL CELLS PRODUCE ETF-3 THAT STIMULATE EMBRYO DEVELOPMENT

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Objective: To study the glycoprotein fractions 3 (ETF-3) secreted from immortalized and primary human oviductal epithelial cells and their embryotrophic activities on mouse embryo development. **Methods:** The primary human oviductal epithelial cells (OE) were immortalized (OE-E6/E7). By using Liquid Chromatography system, ETF-3 was derived from OE-E6/E7 (F3) and OE (P3). F3 and P3 were separated by electrophoresis under native and denatured conditions. They were then reconstituted with Chatot-Ziomek-Bavister (CZB) medium and used to culture Day 1 MF1xBALC/c embryos for 4 days. CZB medium alone culture was included as control. Embryo development was assessed at 2-cell, 4-cell, morula and blastocyst stages. **Results:** After electrophoesis, similar protein patterns were found in F3 and P3 under native and denatured conditions. Embryo culture showed that there are no significant difference in the cleavage rate among control, F3 and P3. However, the sizes of F3 and P3 treated blastocysts were significantly larger than control group (101±1.27mm and 105±1.17mm Vs 93±2.14mm, respectively). **Conclusion:** These results showed that F3 has similar embryotrophic avtivity as P3. F3 can be used for further characterization of the embryotrophic factors secreted from the human oviductal epithelial cells.

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