

Effects of Somatotropin and Human Vascular Endothelial Growth Factor on the *In Vitro* Development of Porcine Oocytes using Chemically Defined Medium

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Porcine follicular fluid (PFF) has frequently been used in porcine IVM to achieve enhancement of nuclear and cytoplasmic maturation of porcine oocytes *in vitro*. However, because PFF contains various unknown factors, many researchers have been trying to develop a chemically defined IVM medium excluding PFF. For this reason, this research was conducted to determine the effect of two growth factors, somatotropin (STH) and human vascular endothelial growth factor (hVEGF), on the development of porcine oocytes using chemically defined medium. Ovaries were collected from a local slaughter house then oocytes aspirated by 10mL syringes with 18 gauge needles from the follicles ranging from 3mm to 6mm in diameter. Oocytes with more than three layers of cumulus cells were selected and *in vitro* matured in TCM 199 medium supplemented with 0.1% PVA, PMSG (100 IU/mL), hCG (100 IU/mL), and 10ng/mL of EGF. IVM was performed in two step culture. In the first 22h, hormones (PMSG and hCG) were supplemented then removed for next 22h of maturation. For Experimental purpose, STH (1 μ g/mL) or hVEGF (10 ng/mL) were added to maturation medium. COCs were denuded by 0.1% hyaluronidase and polar body extrusion rate was evaluated by Hoechst staining. *in vitro* development to Blastocyst after parthenogenic activation evaluated using NCSU-23 as embryo culture media. Blastocyst rate was checked at day 7 of IVC. Parthenogenic embryos were obtained by single electrical pulse (2 kV/Cm, 30 μ sec). In Experiment 1, polar body extrusion rate was evaluated. STH

treated group ($80.80 \pm 3.19\%$, $n=237$) and VEGF treated group ($73.84 \pm 5.43\%$, $n=214$) show significantly higher efficiency of nuclear maturation compared to non-treated control group ($72.36 \pm 2.61\%$, $n=227$). In Experiment 2, developmental competence of blastocyst formation was examined. In STH and VEGF supplemented group, BL-formation rate was also improved ($11.95 \pm 3.61\%$) compared to non-treated group. ($6.92 \pm 2.79\%$). Result of Exp.1 show that supplementation of STH and VEGF during IVM will be helpful for complete maturation of porcine oocytes. These growth factors also beneficial to further development after activation as shown in Exp.2. In conclusion, both STH and VEGF are useful for optimizing chemically defined *in vitro* maturation medium. But still more studies are required to assess exact role and mechanisms of both hormones on *in vitro* maturation of porcine oocytes.

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