Lab practical session: IVM

Session 1

Three step procedure

1 step Ovary selection :

M & M 1) Timing and transport of the ovaries from slaugtherhouse
2) Selection of follicles containing meiotically competent oocytes (medium antral follicles) that have not yet responded to the LH stimulus
3) exclude prebupertal ovaries with follicles bigger that 5 mm
4) exclude pubertal ovaries containing albicans CL

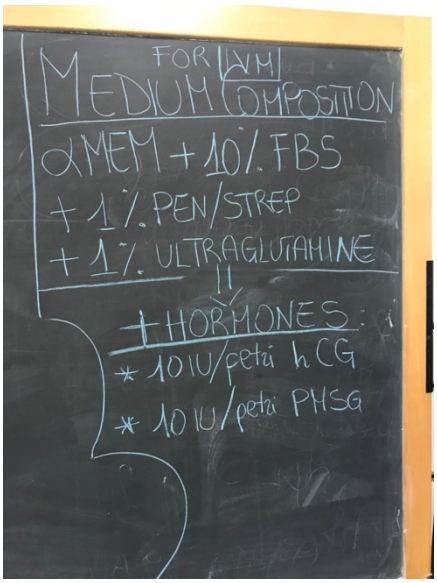
Quality assesment: number of Cumulus oocyte complexes (COCs) of good quality isolated from each ovary (in pig 6-10)

2 step COCs selection:

M &M to isolate COCs of good quality to perfome IVM you have to considered
Healthy indicators: compactness of cumulus cells (the zona pellucida can be easily appreciate through the cumulus)
Several layer of cumulus cells
Continuity of cumulus layers surrouding the oocyte
Grey vacuolate ooplasm
Degeneration markers: Oocyte darkness
Cumulus cell darkness,, loss of compatness and expansion

Functional indicators: Presence of abundant somatic component (cumulus but also mural granulosa cells attached to the cumulus) is the more suitable condition to transduce the LH surge

Quality assesment: % of MII oocyte after IVM good quality isolated (in pig ≥90%)



3 Step IVM

M&M Use adequate maturation medium (i.e Alpha MEM + supplements)
Hormonal stimulation of maturation (LH or hCG)
Controlled cultural conditions (temperature, CO2/O2 tension, and timing 40-44 h pig, 20-24h shepp etc
Quality assesment: % of fertilized oocyte with a monospermic assect (>60% in pig)