

## Oviductal epithelial cells culture selects spermatozoa without DNA fragmentation

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*In vivo*, the interaction of oviductal epithelial cells (OEC) with the spermatozoa has a very important role in sperm selection, preventing that lower quality sperm reach the oocytes [1]. Several authors have characterized some aspects of the superior sperm quality associated with oviductal binding [2,3], concluding that sperm-oviduct binding plays a key role in the maintenance of sperm fertilizing ability. An important factor that affects fertility is DNA fragmentation. Although there is still controversy regarding this, it has been shown that DNA fragmentation in sperm, are correlated with fertility failure [4,5]. The aim was analyse the levels of DNA fragmentation of sperm bound to oviductal epithelial cells and compare them with not bound sperm. Spermatozoa were initially selected through a physical medium such as isotonic discontinuous Percoll gradient (mimicking the uterus) and subsequently were selected through a biological system such as OEC (mimicking the oviduct's action). After 30 minutes of co-culture we obtained two subpopulations: sperm bound to OEC (B group) and sperm from the supernatant not bound to OEC (NB group). As a control group, we used washed sperm through Percoll and incubated for 30 minutes without oviductal cells (W30 group). Apo-BrdU Tunel Assay Kit (Invitrogen S.A., Barcelona, Spain) was employed to determine sperm DNA fragmentation. In brief, the samples were fixed in 2% formaldehyde for 60 minutes to induce sperm membrane permeabilisation, washed twice with PBS and resuspended in 50 µl of DNA-labelling solution (10 µl reaction buffer, 0.75 µl Tdt enzyme, 8 µl of BrdUTP and 31.25 µl of dH<sub>2</sub>O) and incubated for 120 minutes at 37°C. Samples were evaluated using *in situ* detection. We found DNA fragmentation in W30 and NB groups without differences between them; however B group doesn't have DNA fragmentation. The results showed that, under *in vitro* conditions, spermatozoa with fragmented DNA are not able to bind to OEC, therefore sperm selection by OEC could in the future represent a solution to obtaining the best spermatozoa for fertilization in assisted reproduction technologies.

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### Referencias

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