Isolation and characterization of extracellular vesicles secreted by pre-pubertal Sertoli cells

<u>Giovanni Luca</u>¹ - Giulia Falabella¹ - Mario Calvitti¹ - Stefano Giovagnoli² - Francesca Mancuso¹ - Iva Arato¹ - Cinzia Lilli¹ - Catia Bellucci¹ - Alessio Becchetti³ - Cinzia Antognelli¹ - Maria Bodo¹ - Tiziano Baroni¹ - Riccardo Calafiore⁴

¹ Università degli Studi di Perugia, Dipartimento di Medicina Sperimentale, Perugia, Italia - ² Università degli Studi di Perugia, Dipartimento di Scienze Farmaceutiche, Perugia, Italia - ³ Hospital Universitario de la Ribera de Alzira, Alzira, Valencia, Spagna – ⁴ Università degli Studi di Perugia, Dipartimento di Medicina, Perugia, Italia

Recent studies have shown that extracellular vesicles (Ev) are an important mechanism of intercellular communication. In fact, Ev may contain proteins, DNA and mRNA. In particular, the latter play an important role in various biological processes including regulation and cell differentiation [1]. Sertoli cells (SC), previously considered as a mere "sustentacular cell", were re-evalued in their functions and elevated to the rank of a "sentinel" in spermatogenesis due to production of trophic, differentiation and immunemodulators factors. Porcine pre-pubertal SC, isolated by our method [2], upon 48 hours culture, SC were stimulated with recombinant a-follitropin (rFSH) or FSH + testosterone (T) to evaluate both the presence in the medium of SC-derived Ev (SC-Ev) and SC-Ev content, in terms of mRNA for Anti-Müllerian hormone (AMH), inhibin B, Androgen Binding Protein (ABP) and FSH-receptor (FSH-r), by RT-PCR. SEM analysis highlighted the presence of SC-Nv in culture medium with mean diameters < 149 nm. We have also demonstrated, within the SC-Ev, significant increase in mRNA for AMH, ABP and FSH-r after both FSH and FSH+T stimulation, as compared to unstimulated SC-Ev. Differently from unstimulated SC-Ev, mRNA inhibin B levels were unchanged in FSH-stimulated SC-Ev, and increased after FSH+T stimulation. Interestingly, an opposite trend was shown in mRNA secretion, in control SC monolaver where, we demonstrated a decrease of AMH and FSH-r mRNA (after both stimulations with FSH or FSH + T) and an increase of inhibin B mRNA. On the contrary, mRNA ABP levels, in SC monolayer, decreased after stimulation with FSH but were unchanged in the presence of FSH+T. For the first time in the Literature, our work has shown the presence of SC-Nv containing AMH, inhibin B, ABP and FSH-r mRNA regulated by FSH with or without T. This result may suggest that other testicular cells could produce factors that, until now, were considered an exclusive SC secretory product.

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References

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Keywords

Sertoli cells; extracellular vesicles.