



# Exosomes as mediators of early embryo-maternal communication

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## Introduction

Successful pregnancy requires an appropriate communication between the mother and the embryo. Recent studies indicate that exosomes, small (30-200nm) membrane vesicles of endocytotic origin could act as intercellular vehicles in this unique communication system. Here, we proposed to study the dynamics of exosome production and secretion by the oviduct epithelium in response to female sexual hormones and the presence of the early developing embryo in an attempt to identify different dimensions of the embryo-maternal cross-talk.



## Research Questions

How does the female sexual hormones modulate the exosome production, their protein content and secretion by bovine oviductal epithelial cells (BOEC)?

How do exosomes regulate fertilization and early embryo development?

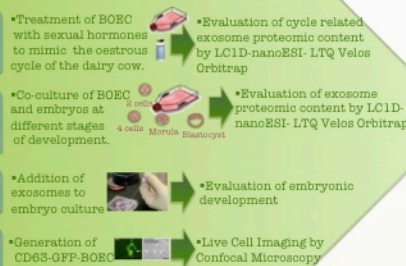
## Objectives and Methodology

**Objective 1** To determine the effect of female sexual hormonal stimulation on exosome production, protein content and secretion in vivo and in vitro by BOEC.

**Objective 2** To determine whether the embryo at different developmental stages modulates exosome production, protein content and secretion by BOEC.

**Objective 3** To determine the exosome content effect on embryonic development.

**Objective 4** To track exosome transfer between the BOEC and the embryo and spermatozoa



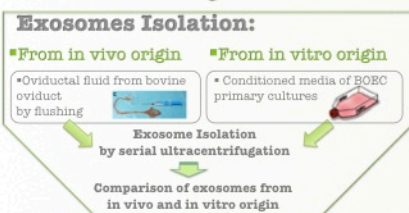
## Preliminary Experiments

BOEC have been thoroughly used to study embryo-maternal interactions and to improve embryo development in vitro.

**But, are there any differences between BOEC exosomes from in vivo and in vitro origin?**

Hence, our preliminary experiment aimed to provide a **morphologic and proteomic characterization of exosomes** secreted by BOEC in vivo in the oviductal fluid and in vitro in the conditioned media.

## Preliminary Results



### Characterization of exosome distribution and size

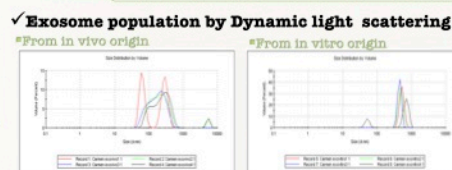


Figure 1. Size analysis confirms exosomes (30-200nm) and microvesicles (>200nm) in exosome preparations from in vivo and in vitro origin.

### Exosome observation by electron microscopy

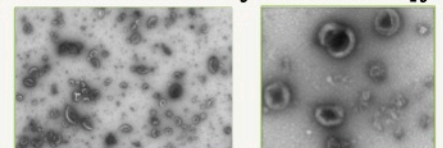
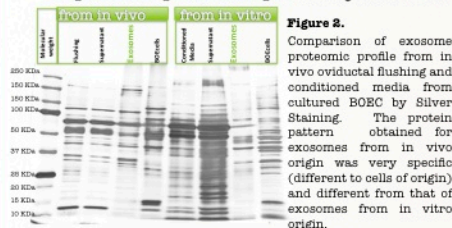


Figure 2. Demonstration of presence of exosomes and microvesicles in oviductal flushing by transmission electron microscopy.

### Comparison of Oviductal Exosome Proteomic Profile

#### Comparison of protein composition by SDS-PAGE



#### Comparison of proteomic profile by Mass Spectrometry

##### Analyses LC1D-nanoESI-LTQ Velos Orbitrap + MASCOT

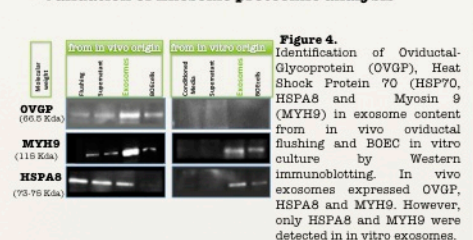
✓ A total of 319 proteins were identified in exosomes samples.

✓ From these proteins, 179 proteins were differentially expressed when in vivo exosomes were compared to in vitro exosomes (T-test, (p-value<0.01, ratio<0.5 ou ratio>2).

✓ Interestingly, 97 proteins were specific for in vivo exosomes, 47 proteins were specific for in vitro exosomes and 175 proteins were in common in both exosomes from in vivo and in vitro origin.



#### Validation of Exosome proteomic analysis



## Expected Impact

- To Increase our understanding of the role of exosomes as modulators of the embryo-maternal interactions
- To Improve reproductive biotechnologies such as in vitro fertilization, embryo cryopreservation and transfer by using exosomes
- To Improve livestock production, by using exosomes as novel molecular markers of infertility