

## FELLOWSHIP SUMMARY REPORTS

**Cover page :**

**Fellow:**

Valentina Lodde

Reproductive and Developmental Biology Laboratory (<http://www.redbiolab.unimi.it>)

Department of Health, Animal Science and Food Safety

University of Milan, Milan Italy

**Programme:**

Co-operative Research Programme: Biological Resource Management for Sustainable Agricultural Systems (Fellowship Awards Programme 2016)

**Theme number of the research fellowship:**

Theme III: The food chain

**Project title:**

Focus on zinc function in oocyte development to improve reproductive performance in dairy cattle

(Contract number: JA00091594)

**Your host institution:**

Département des sciences animales, Institut sur la nutrition et les aliments fonctionnels (INAF), Faculté des sciences de l'agriculture et de l'alimentation - Université Laval, Quebec Canada

**Host supervisor:**

Professor Marc-Andr e Sirard

**Dates of the fellowship:**

5 December 2016 - 27 March 2017

I consent to the publication of the present summary report on the Co-operative Research Programme's website,



## 1. Relevance

### To the Co-operative Research Programme objectives

The present research project fulfils the Co-operative Research Programme's main objective, which is to '*strengthen scientific knowledge and provide relevant scientific information and advice that will inform future policy decisions related to the sustainable use of natural resources, in the areas of food, agriculture, forests and fisheries*'. The main goal of the present project is to acquire new knowledge on the factors impairing cow oocyte quality and therefore female fertility, focusing on the role of zinc. Poor quality oocyte, with limited ability of undergoing fertilization, embryo development and subsequent pregnancy is one of the major limits to sustainable cattle productivity. Thus, understanding the factor and mechanism implicated in oocyte quality is the first step to develop and make available new tools and strategies to maximize the developmental potential of cattle oocytes, with obvious positive consequences on fertility and sustainable use of natural resources.

### To the theme

The present proposal is related to the Theme III of Co-operative Research Programme – The food chain - since the data generated will ultimately aid a more efficient use of genetic resources in livestock and improvement of reproductive techniques in dairy cattle. The dairy cattle industry profitability is threatened by reproductive disorders due mainly to the genetic selection for milk production traits, which reduces the reproductive performance of dairy cow. Moreover, the metabolic stress of milking in the most productive animals interacts with the energy balance and result in abnormal ovarian environment leading to compromised fertility. Thus, improving dairy cow fertility is a priority for sustainability of modern dairy farming in Europe and worldwide. Assisted reproductive technologies (ART) and in particular in vitro embryo production (IVP) are available but still not fully efficient since only 30-40% of the bovine oocytes fertilized in vitro develop into viable embryo that can be transferred in a recipient.

Importantly, the present research will ultimately lead to improvement of the overall efficiency of in vitro embryo production by proposing novel strategies to improve oocyte developmental competence acquisition during culture in vitro before fertilization. Ultimately, this will aid the development of a pathogen-free food chain, since it is well known that biosecurity is an added value to in vitro embryo production and embryo transfer [1-3]

Our research also fulfils the goal of providing new knowledge in the post-genomic area. In particular, transcriptomic (microarray) data that were already generated within the EmbryoGENE network (<http://emb-bioinfo.fsaa.ulaval.ca>), leaded by Professor Sirard, have been integrated with functional studies to identify the mechanism that ultimately determines oocyte quality. Finally, since several ovarian follicular compartments (oocyte, cumulus cells, granulosa cells) have been considered, the present research fosters the progress of system biology.

### To agricultural and food policy

Milk production depends on the cows becoming pregnant and subsequently delivering a calf. Ideally, cows will deliver a calf every 12-14 months in order to keep the milk production level at the optimum. Suboptimal reproductive performance leads to extended days non-milk producing days, increased culling due to reproductive failure, and decreased milk yield. Modern farming, is increasingly relying on in vitro embryo production. A recent analysis of worldwide bovine in vivo and in vitro embryo production between 1997 and 2013 revealed that IVP has increased significantly year after year and in 2013 up to 42% of the total embryos produced were of in vitro origin [4, 5]. These technologies are mostly used in North and South America,



while European Countries, and Italy in particular, still make large use of artificial insemination and lesser use of embryos fully produced in vitro. Remarkably, if we consider that on average only 30-40 % of in vitro cultured oocytes reaches the blastocyst stage of embryonic development, the overall IVP efficiency can be improved substantially, with obvious advantages for, but not limited to, dairy farming sustainability and profitability. The joined overall goal of the two research teams involved in the present project, at the University of Milan and Laval University, is to improve IVP efficiency thus making them more affordable for the stakeholders. This will be a major advantage for Europe and Italy in particular that still takes limited advantages of these technologies.

### **To society**

The improvement of reproductive efficiency will have a direct impact on the environment since it will reduce the culling rate and the number of replacement cattle that are needed to maintain herd size. In Italy in particular, the development of more efficient IVP strategies will possibly foster a wider use of IVP technologies, also in small and medium sized heard, thus contributing to the development of new specialized IVP centres across the country and/or boost the ones that are already present in the territory. Clearly improved IVP efficiency will also speed genetic progress in cattle breeding industry.

Finally, since the cow is a good model for human infertility, there is a high probability that basic knowledge generated in the present research project will impact also human infertility field.

## **2. Objectives of the fellowship**

The proposed research considers the zinc as one of the essential players in the process of oocyte competence acquisition. Although a relationship between zinc deficiency and infertility in dairy cows has been known from many years the molecular basis of this relationship are still unknown, which is one of the main reason why zinc modulation during oocyte in vitro culture has not been yet optimized. The aim of the research project is firstly to define the molecular basis of oocyte zinc modulation in the establishment of the oocyte competence in bovine, during a critical step of the oocyte growth, by integrating 'omics' and functional data. In the last years Professor Sirard, together with some of the most renowned scientist in the field of reproductive biology, has developed the EmbryoGENE bovine transcriptomic platform within the EmbryoGENE network (<http://emb-bioinfo.fsaa.ulaval.ca/Home/index.htm>). Within this platform, the EmbryoGENE bovine microarrays, was made available to many researchers of the network, for the study of gene expression profiles of oocytes, cumulus and granulosa cells coming from a multiplicity of experimental conditions and models of developmental competence, with the aim of gathering information on the conditions leading to competent oocytes. So far, the platform has generated a huge amount of gene expression profiling data spanning small to ovulatory or degenerating follicles [6] ([7] (<http://emb-bioinfo.fsaa.ulaval.ca/granulosaIMAGE/>)). Clearly this offered a unique opportunity to fulfil the scope of defining the extent to which zinc controls oocyte function and development. The first objective of the fellowship was then Dr's Lodde being trained in the bioinformatic meta-analysis of the EmbryoGENE data to allow the identification of key players in the control of oocyte developmental competence acquisition. This analysis gave a list of target molecules and mechanism that can now be targeted during protocols of oocyte in vitro growth and/or oocyte in vitro maturation to improve the overall efficiency of in vitro embryo production, which is the second objective of the present project.

## **3. Major achievements (up to three)**

1 - As part of the training in bioinformatics analysis of data generated within the EmbryoGENE network, Dr Lodde was involved in a study aimed at conducting a meta-analysis of existing microarray datasets from bovine oocytes during the final stages of oocyte differentiation to analyze the expression profiles of histone and histone remodelling enzyme mRNAs, and correlated these with the major histone modifications known to occur at the same period, which are in turn pivotal to ensure a proper developmental programme in the



oocyte. This work generated a manuscript, with Dr Lodde as first Author, that has been already accepted for publication as a chapter in the book entitled 'Oocytes - Maternal information and functions' within the series 'Results and Problems in Cell Differentiation' from Springer/Nature. In addition, the result of this research has been recently presented as an oral communication at the 'XI National Congress of the Italian Association of Italian veterinary morphologist held in Rome May 25<sup>th</sup>-26<sup>th</sup> 2017 (title: Chromatin remodeling and histones modifying enzymes in bovine oocyte: interpretation of transcriptomic data meets morphology)

(<http://www.morfovet.it/XI%20Congresso%20AMV%20Roma%202017/Congresso%20AMV%20Roma%202017.htm> )

2- Generation of preliminary 'in silico' data for the study of the role of zinc in the acquisition of developmental competence in bovine oocytes. As expected the bioinformatics analysis revealed information on a multiplicity of target molecules thus generating hypothesis that are currently being analysed by Dr Lodde research team at the University of Milan. Importantly another research group, led by Prof. Claude Robert at the University of Laval, was involved in the study and is currently collaborating to the successful outcome of the research project, thus enlarging the research network. The results of the present joined research will result in a manuscript that will be submitted for publication end 2017-early 2018 in a high impact factor scientific journal in the field of reproductive biology.

3- Dr. Lodde had the opportunity to present the results of her previous research project focused on the role of Progesterone Receptor Membrane Component 1 (PGRMC1) in mammalian fertility during an invited lecture entitled 'Insights into Progesterone Receptor Membrane Component 1's role in regulating bovine oocyte meiosis and granulosa cell mitosis' within the conference series of the 'Centre de recherche en reproduction, développement et santé intergénérationnelle (CRDSI, <http://www.crdsi.ulaval.ca>)' held in Quebec city on March 15<sup>th</sup> 2017. This will further enlarge Dr Lodde's research network and promoting possible future scientific collaboration with renowned scientist in the field of biology of reproduction, whose common interest is to improve reproductive efficiency and female fertility

#### 4. Follow-up

##### **Is a publication envisaged? Will this be in a journal or a publication? When will it appear?**

Yes, one full scientific publication has been already accepted and it will be published around August-September 2017. Another full paper will be prepared and submitted end 2017-early 2018. Conference proceedings of the oral communication at the 'XI National Congress of the Italian Association of Italian veterinary morphologist' held in Rome May 25<sup>th</sup>-26<sup>th</sup> 2017 will be published in the 'Annals of Anatomy', Elsevier.

##### *List of publication:*

1. **Lodde V**, Luciano AM, Franciosi F, Labrecque R, **Sirard MA**. Accumulation of chromatin remodelling enzyme and histone transcripts in bovine oocytes. In: Kloc M (ed.) *Oocytes - Maternal information and functions*, vol. in press: Springer; 2017.
2. **Lodde V**, Franciosi F, Dieci C, Terzaghi L, Modina SC, **Sirard MA**, Luciano AM. Chromatin remodeling and histones modifying enzymes in bovine oocyte: interpretation of transcriptomic data meets morphology. In: 11° Congresso, Associazione Morfologi Veterinari, vol. Special Issue Animal Anatomy Roma, Italy: *Annals of Anatomy*; 2017: in press

##### **Is your fellowship likely to be the start of collaboration between your home institution and your host?**

Yes, very much

##### **Is your research likely to result in protected intellectual property, novel products or processes?**

Possibly



## 5. Satisfaction

### **Did your fellowship conform to your expectations?**

Yes, I had great expectations from the collaboration with Prof. Sirard and his team, which were conformed with no exception

### **Will the OECD Co-operative Research Programme fellowship increase directly or indirectly your career opportunities? Please specify.**

Yes, I enlarged my collaborative network, which is pivotal for a scientist in the path to independency

### **Did you encounter any practical problems?**

Minor practical problem was the need to postpone my departure of a few days due to paperwork's delay in obtaining the Canadian work permit. This did not impact the overall outcome of the fellowship.

### **Please suggest any improvements in the Fellowship Programme.**

Inclusion of funding that could cover bench fees or publication fees could be helpful

## 6. Advertising the Co-operative Research Programme

### **How did you learn about the Co-operative Research Programme?**

From my former supervisor at the University of Milan, Professor Alberto M. Luciano

### **What would you suggest to make it more "visible"?**

Advertise in social media.

### **Are there any issues you would like to record?**

no

### **References:**

1. Thibier M. Embryo transfer: a comparative biosecurity advantage in international movements of germplasm. *Rev Sci Tech* 2011; 30:177-188.
2. Wrathall AE, Simmons HA, Bowles DJ, Jones S. Biosecurity strategies for conserving valuable livestock genetic resources. *Reprod Fertil Dev* 2004; 16:103-112.
3. Givens MD, Marley SD. Approaches to biosecurity in bovine embryo transfer programs. *Theriogenology* 2008; 69:129-136.
4. Perry G. 2013 Statistics of embryo collection and transfer in domestic farm animals. *Embryo Transfer Newslett* 2015; 33:14-26.
5. Blondin P. Status of embryo production in the world. *Anim Reprod* 2015; 12:356-358.
6. Sirard MA. Toward building the cow folliculome. *Anim Reprod Sci* 2014; 149:90-97.
7. Khan DR, Fournier E, Dufort I, Richard FJ, Singh J, Sirard MA. Meta-analysis of gene expression profiles in granulosa cells during folliculogenesis. *Reproduction* 2016; 151:R103-110.

Applying for fellowships and grants is an integral part of academic life in the sciences. External funding from government agencies, private foundations, corporations, and other entities outside of the university is essential to carrying out sustained research projects of significant scope. A fellowship application presents a comprehensive picture of an applicant as a graduate student, a research scientist, a future colleague, and a member of a profession. A consensus report, the so-called Evaluation Summary Report (ESR), establishes the proposal's final grade. Proposals are then ranked according to their grade. Funding will be provided to eligible projects in descending ranking order according to the available budget for each panel. For MSCA fellowships, inter- and multidisciplinary are part and parcel of "excellence" and should be addressed in this section. This part includes an introduction, a description of the state of the art, the research objectives and an overview of the action. Fellowships will also need to print out all of the forms from their GLACIER summary report, sign them, and attach copies of their I-20/DS-2019, I-94, and Visa. This report will be brought to GPO to be reviewed by the international tax consultant. Student Health Insurance. Fellows receive a medical insurance stipend supplement twice a year (August & December) in their paycheck.