



## Proteomic profile of mouse oocytes after vitrification: A quantitative analysis based on 4D label-free technique

Qingrui Zhuan <sup>a,1</sup>, Xingzhu Du <sup>a,1</sup>, Jiachen Bai <sup>b,1</sup>, Dan Zhou <sup>a</sup>, Yuwen Luo <sup>a</sup>, Hongyu Liu <sup>a</sup>, Wenquan Sun <sup>b</sup>, Pengcheng Wan <sup>c</sup>, Yunpeng Hou <sup>d</sup>, Jun Li <sup>e,\*</sup>, Xiangwei Fu <sup>a,c,\*\*</sup>

<sup>a</sup> Key Laboratory of Animal Genetics, Breeding and Reproduction of the Ministry of Agriculture and Rural Affairs, National Engineering Laboratory for Animal Breeding, Beijing Key Laboratory for Animal Genetic Improvement, College of Animal Science and Technology, China Agricultural University, Beijing, China

<sup>b</sup> Institute of Biothermal Science and Technology, School of Medical Instrument and Food Engineering, University of Shanghai for Science and Technology, Shanghai, China

<sup>c</sup> State Key Laboratory of Sheep Genetic Improvement and Healthy Breeding, Institute of Animal Husbandry and Veterinary Sciences, Xinjiang Academy of Agricultural and Reclamation Sciences, Shihhotze, China

<sup>d</sup> State Key Laboratories of Agrobiotechnology, College of Biological Sciences, China Agricultural University, Beijing, China

<sup>e</sup> Department of Reproductive Medicine, Reproductive Medical Center, The First Hospital of Hebei Medical University, Shijiazhuang, Hebei, China

### ARTICLE INFO

#### Article history:

Received 21 March 2022

Received in revised form

23 April 2022

Accepted 24 April 2022

Available online 27 April 2022

#### Keywords:

Mice  
Oocyte  
Vitrification  
Proteome  
4D label-free

### ABSTRACT

Mature oocyte cryopreservation represents an important trend for future fertility preservation, however, the relatively low efficiency has hampered its clinical application. Proteomic profiling is a method of choice for the exploration of the molecular mechanism underlying cryoinjuries. Here, a systematic comparison of protein expression between fresh and vitrified oocytes was performed based on the 4D label-free technique, an informative method with high sensitivity. Our results indicated that the oocyte survival rate was significantly reduced after vitrification. Proteomic results showed that 32 proteins were up-regulated, while 77 proteins were down-regulated in vitrified oocytes compared with the fresh counterparts. Gene Ontology (GO) functional analysis revealed that differentially expressed proteins (DEPs) were involved in metabolism, mitochondrial function, cytoskeleton and other cell functions. Moreover, proteins that participated in signaling transduction mechanisms were the largest category based on Clusters of Orthologous Groups of protein/Eukaryotic Orthologous Groups (COG/KOG) functional classification. In addition, over-expressed DEPs were enriched for “nucleus”, “protein binding”, “membrane”, “cytoplasm” as well as mitochondrial function. Furthermore, we discovered that the DEPs were clustered in pyruvate metabolism, citric acid (TCA) cycle and glucose metabolism by Protein-Protein Interaction (PPI) network evaluation. In conclusion, our data demonstrate that vitrification induces multi-level damages in oocytes, the dynamic proteomic profiling will provide systematic insights into uncovering the mechanism underlying cryoinjuries.

© 2022 Published by Elsevier Inc.

\* Corresponding author. The First Hospital of Hebei Medical University, Shijiazhuang, Hebei, China.

\*\* Corresponding author. Key Laboratory of Animal Genetics, Breeding and Reproduction of the Ministry of Agriculture and Rural Affairs, National Engineering Laboratory for Animal Breeding, Beijing Key Laboratory for Animal Genetic Improvement, College of Animal Science and Technology, China Agricultural University, Beijing, China.

E-mail addresses: [junliydy@126.com](mailto:junliydy@126.com) (J. Li), [xiangweif@126.com](mailto:xiangweif@126.com) (X. Fu).

<sup>1</sup> These authors contributed equally to this work.

## 1. Introduction

Assisted reproductive technologies (ARTs) such as oocyte *in vitro* fertilization (IVF) and gamete cryopreservation play an important role in basic research and the application of genetic preservation models as well as in clinical practice. Among them, oocyte cryopreservation is the most proven technique to maintain female fertility and genetic potential [1]. Challenges to oocyte cryopreservation include their relatively large size, high water content, unique chromosome arrangement and meiotic spindle [2], which make oocytes particularly vulnerable to damage from ice crystal formation during vitrification and thawing.