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ORIGINAL ARTICLE

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Three-dimensional in vitro maturation of rabbit oocytes enriched with sheep decellularized greater omentum

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Abstract

Background: To prevent ovarian hyperstimulation syndrome, in vitro maturation (IVM) allows the oocytes for infertility treatment without hormone therapy. Although many oocytes matured during IVM, some deficiencies in the culture conditions lead to inhibition of the growth and development of the cumulus cells and the oocyte nuclear and cytoplasmic maturation.

Objectives: The challenge of improving the oocyte culture conditions prompted us to use greater omentum (GOM), full of growth factors and proteins, as a rich supplement to the base culture medium.

Methods: Cumulus-oocyte complexes were recovered from rabbits and divided into 3D and 2D conditions cultured for 12 and 24 h. In 3D cultures, the oocytes embedded in alginate containing FBS decellularized GOM. Corresponding supplements were also added in 2D conditions—maturation of the oocytes evaluated by Aceto-Orcein, TEM, and RT-PCR for MAP2K1 and Cdk2.

Results: DNA quantification, Hoechst, and H&E staining confirmed cell depletion from GOM, and SEM showed the preservation of ultra-architecture after decellularization. Histochemical staining methods showed appropriate extracellular matrix preservation. ELISA assessment showed retention of VEGF content. MTT assessment indicated decellularized GOM was non-toxic. Both Aceto-Orcein assessment and ultra-structure study of the oocytes showed that supplementation of 2D or 3D cultures with decellularized omentum promoted oocyte maturation. Expression of MAP2K1 and Cdk2 also increased in the presence of GOM.

Conclusions: GOM supplementation has a beneficial impact on oocyte maturation, probably due to the presence of growth factors and proteins.

KEYWORDS
decellularization, greater omentum, in vitro maturation, oocyte, rabbit

1 | INTRODUCTION

In recent years, much progress has been made in reproductive technology. The main stage depends on the success rate of the oocyte in vitro maturation. Promoting nuclear and cytoplasmic maturation of the cumulus-oocyte complex (COCs) is isolated from the immature follicles in the culture medium to get prepared for fertilization is called in vitro maturation (IVM) (Heraud et al., 2017). Before IVM innovation,

oocytes showed that the 3D GOM culture condition provided a better microenvironment for the maturation of the oocyte. It may be due to the richness of GOM in the nutrients, growth factors, and proteins, and this solution can be suggested as beneficial supplementation to accelerate IVM (12 h). In addition, we successfully obtained the rabbit COCs without hormonal drugs in the present study. This is important in reducing the widespread side effects of using hormonal drugs in ART and the threatening risks to infertile women.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

ETHICS STATEMENT

This project was approved by the ethics committee of Shiraz University of Medical Sciences (IR.SUMS.REC. 1396.S1013).

AUTHOR CONTRIBUTIONS

KFD: involved in data collection, data analyses, and drafting of the manuscript; TTK: involved in experimental design, data analyses, revising the manuscript, and supervision; SFMA, involved in idea development, experimental design, data analyses, fund acquisition, and supervision.

DATA AVAILABILITY STATEMENT

The data of manuscript are available.

PEER REVIEW

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