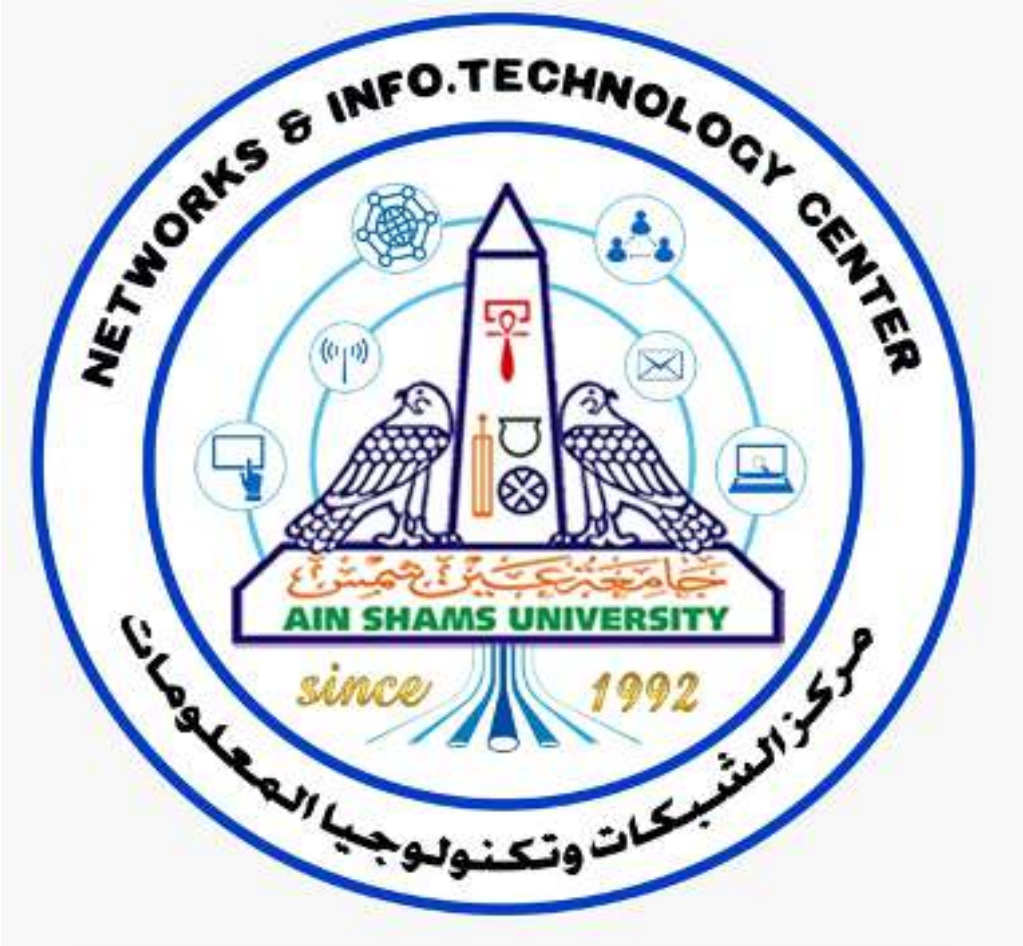
Mona Maghraby

بسم اللّه الرحمن الرحيم

مركز الشبكات وتكنولوجيا المعلومات

قسم التوثيق الإلكتروني



Mona Maghraby

جامعة عين شمس

التوثيق الإلكتروني والميكروفيلم

قسم

نقسم باللّه العظيم أن المادة التي تم توثيقها وتسجيلها

علي هذه الأقراص المدمجة قد أعدت دون أية تغيرات

MOLECULAR STUDIES ON OOCYTE MATURATION IN EGYPTIAN BUFFALO USING DIFFERENT MEDIA

By

Said EL- Hussein Said Mahmoud Abu Hamed
B.Sc. Agric. Sci. (Biotechnology), Fac. Agric., Alex Univ., 2009

THESIS

Submitted in Partial Fulfillment of the Requirements for the Degree of

MASTER OF SCIENCE

In

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Department of Genetics
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Format Reviewer **Vice Dean of Graduate Studies**

APPROVAL SHEET

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Professor of Genetics, Faculty of Agriculture, Cairo University

Date: / /

SUPERVISION SHEET

MOLECULAR STUDIES ON OOCYTE MATURATION IN EGYPTIAN BUFFALO USING DIFFERENT MEDIA

M.Sc. Thesis
In
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Title of Thesis: Molecular Studies on Oocyte Maturation in Egyptian Buffalo Using Different Media
Supervisors: Dr. Salah EL-Din Sayed Mohamed EL-Assal
Dr. Ahmed Yehia Gad
Dr. Swiefy Abd El-Rahim Swiefy (late)
Department: Genetics **Approval:** / /

ABSTRACT

Retinoic acid, vitamin A metabolite, plays a role in oocyte development and maturation in different ways including gene expression alteration and/or prohibiting oxidative stress. The objective of this study was to examine the effect of 9-cis-retinoic acid (9-cisRA) on the quality and maturation rate of buffalo oocytes. Cumulus oocyte complexes (COCs, n = 460) were collected from ovaries of slaughtered buffalos. Varying concentrations of 9-cisRA (0, 5, 50, and 200 nM) were added to the maturation medium, and the following parameters were analyzed: (i) maturation and cleavage rates, (ii) mitochondrial activity and reactive oxygen species (ROS) levels, (iii) expression level of antioxidant-related genes (PRDX1, SOD1, CAT, HOMX1, and GPX4) using RT-qPCR. (iv) expression level of genes related to oocyte quality (GDF9,BMP15) Maturation rate was significantly improved in 5 nM 9-cisRA oocyte group (95.8%, $P < .05$) compared to control and other treatment groups (86.7% in control group). The same oocyte group exhibited significantly higher mitochondrial membrane potential activity and lower ROS accumulation level compared to other treatment groups. Antioxidant-related genes were up-regulated in oocytes matured with 5 or 50 nM 9-cisRA compared to control and 200 nM 9-cisRA groups. In contrast, 200 nM of 9-cisRA showed a clear down-regulation for antioxidant-related genes except for PRDX1. In conclusion, supplementation of 9-cisRA with a lower concentration (5 nM) to the buffalo oocytes maturation media promotes maturation rate through a protection mechanism that maintains adequate levels of antioxidant-related transcripts and improves mitochondrial activity. However, 9- cisRA has no significant effect on the cleavage rate of the treated oocytes.

Key words: Antioxidant genes, Cleavage rate, Follicular fluid, Mitochondrial activity Reactive oxygen species.

LIST OF ABBREVIATIONS

ART	Assistant Reproductive Technology
AtRNA	All trans Retinoic Acid
CEOs	Cumulus Enclosed Oocytes
Cocs	Compact Complex Oocyte Cumulus Cells
CRABP	Cellular Retinoic Acid Binding Protein
CT	Threshold Cycle
ET	Embryo Transfer
ICM	Inner Cell Mass
IVC	*In vitro* Culture
IVEP	*In vitro* Embryo Production
IVF	*In vitro* Fertilization
IVM	*In vitro* Maturation
IVP	*In vitro* Production
MOET	Multiple ovulation Embryo Transfer
mtDNA	Mitochondrial DNA
OS	Oxidative Stress
OSFs	Oocyte Secreted Factors
PI	Propidium Iodide
RA	Retinoic Acid
RAR	Retinoic Acid Receptors
ROS	Reactive Oxygen Species
RXR	Retinoid X Receptors
SO	Super Ovulation
TCN	Total Cell Number
TNF	Tumer Necrosis Factor
ZP	Zona Pellucida

ACKNOWLEDGMENTS

I would like to express my deepest sincere gratitude and appreciation to ***Dr. SALAH EL-DIN SAYED MOHAMED EL-ASSAL****, Professor of Genetics, Fac. Agric., Cairo University, for his keen supervision, valuable suggestions, and continuous encouragement and generously given the experience and guidance.*

Also I would like to express my deep thanks to ***Dr. AHMED YEHIA GAD,*** *Associate Professor of Animal Physiology, Faculty of Agriculture, Cairo University, for his collaboration and showing me the practical importance of work study, suggesting the point, enthusiastic help and generously given the experience and guidance.*

I am extremely grateful to ***Dr. SWIEFY ABD EL-RAHIM SWIEFY,*** *Professor, Biotechnology department, Animal Production Research Institute, Agriculture Research Center, Giza, Egypt, for his kind supervision, valuable help, instructive criticism and supplying the facilities throughout this work. And call upon God to bestow His mercy TO* ***DR SWIEFY ABD EL-RAHIM SWIEFY,*** *Who died before the completion of this study.*

I am sincerely thankful to the professional help of all the staff of Biotechnology and Embryology Labs, Cairo University Research Park (CURP), Faculty of Agriculture, Cairo University.