

Role of oocyte-secreted factors in prevention of cumulus cell apoptosis and enhancement of oocyte developmental competence

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A thesis submitted to the University of Adelaide in total fulfillment of the requirements for the degree of Doctor of Philosophy.

July 2006

Table of Contents

Abstract.....	VI
Declaration.....	VIII
Acknowledgements.....	IX
Glossary/Abbreviations.....	XI
Publications.....	XIII
Conference Proceeding.....	XIV
Provisional Patent.....	XV
Visits to Overseas Laboratories & Seminars.....	XV
Awards, Scholarship & Prizes.....	XVI

CHAPTER 1: LITERATURE REVIEW..... 1

1.1	Introduction.....	2
1.2	Follicle and Oocyte Development.....	4
1.2.1	<i>Follicular development.....</i>	<i>4</i>
1.2.2	<i>Oocyte-follicular cell interactions.....</i>	<i>8</i>
1.2.2.1	<i>Gap-junctional communication.....</i>	<i>9</i>
1.2.2.2	<i>Paracrine soluble factors.....</i>	<i>11</i>
1.2.3	<i>Granulosa cell regulation of oocyte growth.....</i>	<i>15</i>
1.3	Transforming Growth Factor –β Superfamily.....	15
1.3.1	<i>TGF-β superfamily signalling pathways.....</i>	<i>16</i>
1.3.2	<i>Transforming growth factor-β.....</i>	<i>20</i>
1.3.3	<i>Activins and inhibins.....</i>	<i>20</i>
1.3.4	<i>Growth differentiation factor 9.....</i>	<i>22</i>
1.3.5	<i>Bone morphogenetic protein 15.....</i>	<i>24</i>
1.3.6	<i>GDF-9 & BMP-15 deficient animal models.....</i>	<i>26</i>
1.3.7	<i>OSFs regulation of COC function.....</i>	<i>28</i>
1.4	Follicular Atresia, Apoptosis and Oocyte Quality.....	28
1.4.1	<i>Follicular atresia.....</i>	<i>28</i>
1.4.2	<i>The cellular mechanism of apoptosis.....</i>	<i>29</i>
1.4.3	<i>Cumulus cell apoptosis and oocyte quality.....</i>	<i>31</i>
1.5	Oocyte Maturation.....	33
1.5.1	<i>Oocyte nuclear and cytoplasmic maturation.....</i>	<i>33</i>
1.5.2	<i>The effect of cumulus cells on oocyte developmental competence.....</i>	<i>35</i>
1.5.3	<i>In vitro & in vivo oocyte maturation outcomes.....</i>	<i>37</i>
1.6	Summary.....	38
1.7	Hypothesis and Aims for PhD Project.....	40
1.7.1	<i>Hypothesis.....</i>	<i>40</i>
1.7.2	<i>Aims.....</i>	<i>40</i>

CHAPTER 2: OOCYTES PREVENT CUMULUS CELL APOPTOSIS BY MAINTAINING A MORPHOGENIC PARACRINE GRADIENT OF BONE MORPHOGENETIC PROTEINS..... 41

2.1	Abstract	42
2.2	Introduction	43
2.3	Materials and Methods	46
2.3.1	<i>Collection of bovine oocytes and culture conditions</i>	46
2.3.2	<i>Treatment of cumulus cells</i>	47
2.3.2.1	<i>Generation of oocyctomized complexes</i>	47
2.3.2.2	<i>Generation of denuded oocytes</i>	47
2.3.2.3	<i>Growth factors and binding proteins</i>	48
2.3.3	<i>Determination of DNA damage by TUNEL (assessment of cumulus cell apoptosis)</i>	48
2.3.4	<i>Confocal microscopy and analysis</i>	49
2.3.5	<i>Western blot analysis</i>	50
2.3.6	<i>Experimental Design</i>	51
2.3.6.1	<i>Experiment 1: Effect of oocyctomy on cumulus cell apoptosis</i>	51
2.3.6.2	<i>Experiment 2: Effect of oocyte-secreted factors on cumulus cell apoptosis</i>	51
2.3.6.3	<i>Experiment 3: Pattern of apoptosis in relation to proximity to oocyte-secreted factor origin</i>	52
2.3.6.4	<i>Experiment 4: Dose response of GDF-9, BMP-6 & BMP-15 on cumulus cell apoptosis</i>	52
2.3.6.5	<i>Experiment 5: Effect of oocytes, GDF-9 and BMP-15 on CC expression of Bcl-2 and Bax proteins</i>	53
2.3.6.6	<i>Experiment 6: Effect of oocytes, BMP-6, and BMP-15 on cumulus cell apoptosis induced by staurosporine</i>	53
2.3.6.7	<i>Experiment 7: Effect of BMP antagonists on cumulus cell apoptosis</i>	54
2.3.6.8	<i>Experiment 8: Role of BMP-15 and BMP-6 in the anti-apoptotic actions of oocytes on cumulus cells</i>	54
2.3.6.9	<i>Experiment 9: Effect of BMP-7 and its antagonist, gremlin, on cumulus cell apoptosis</i>	55
2.3.7	<i>Statistical analysis</i>	55
2.4	Results	56
2.4.1	<i>Experiment 1: Effect of oocyctomy and FSH on cumulus cell apoptosis</i>	56
2.4.2	<i>Experiment 2: Effect of oocyte-secreted factors on cumulus cell apoptosis</i>	56
2.4.3	<i>Experiment 3: Pattern of apoptosis in relation to proximity to oocyte-secreted factor origin</i>	59
2.4.4	<i>Experiment 4: Dose response of GDF-9, BMP-6 and BMP-15 on cumulus cell apoptosis</i>	61
2.4.5	<i>Experiment 5: Effect of oocytes, GDF-9 and BMP-15 on CC expression of Bcl-2 and Bax proteins</i>	61
2.4.6	<i>Experiment 6: Protection of cumulus cells from staurosporine-induced apoptosis by oocytes, BMP-6 and BMP-15</i>	62
2.4.7	<i>Experiment 7: Effect of BMP antagonists on cumulus cell</i>	

	<i>apoptosis</i>	65
2.4.8	<i>Experiment 8: Role of BMP-15 and BMP-6 in the anti-apoptotic actions of oocytes on cumulus cells</i>	66
2.4.9	<i>Experiment 9: Effect of BMP-7 and its antagonist, gremlin, on cumulus cell apoptosis</i>	67
2.5	Discussion	71
2.6	Acknowledgements	80
2.7	References	81

CHAPTER 3: OOCYTE-SECRETED FACTORS ENHANCE OOCYTE DEVELOPMENTAL COMPETENCE.....87

3.1	Abstract	88
3.2	Introduction	89
3.3	Materials and Methods	92
3.3.1	<i>Collection of oocytes and culture conditions</i>	92
3.3.2	<i>Treatment of cumulus-oocyte complexes</i>	93
3.3.2.1	<i>Generation of denuded oocyte</i>	93
3.3.2.2	<i>Growth factors & antagonists</i>	93
3.3.3	<i>In vitro fertilization and embryo culture</i>	94
3.3.4	<i>Differen</i>	
3.3.5	<i>Experimental design</i>	96
3.3.5.1	<i>Experiment 1: Effect of co-culture of intact COCs with DOs during IVM on subsequent developmental competence</i>	96
3.3.5.2	<i>Experiment 2: Effect of BMP-15 and/or GDF-9 during IVM on oocyte developmental competence</i>	97
3.3.5.3	<i>Experiments 3 & 4: Effect of GDF-9 or BMP-15 antagonists on oocyte developmental competence</i>	97
3.3.6	<i>Statistical Analysis</i>	98
3.4	Results	99
3.4.1	<i>Experiment 1: Effect of co-culture of intact COCs with DOs during IVM on subsequent developmental competence</i>	99
3.4.2	<i>Experiment 2: Effect of BMP-15 and/or GDF-9 during IVM on oocyte developmental competence</i>	99
3.4.3	<i>Experiments 3 & 4: Effect of GDF-9 or BMP-15 antagonists on oocyte developmental competence</i>	101
3.5	Discussion	105
3.6	Acknowledgements	112
3.7	References	113

CHAPTER 4: TEMPORAL EFFECTS OF OOCYTE-SECRETED FACTOR(S) DURING IN VITRO MATURATION ON BOVINE OOCYTE DEVELOPMENTAL COMPETENCE.....120

4.1	Abstract	121
4.2	Introduction	122
4.3	Materials and Methods	125
4.3.1	<i>Collection of oocytes and culture conditions</i>	125
4.3.2	<i>Treatment of cumulus-oocyte complexes</i>	126
4.3.2.1	<i>Generation of denuded oocytes</i>	126
4.3.2.2	<i>Growth factors</i>	126
4.3.3	<i>In vitro fertilization and embryo culture</i>	126
4.3.4	<i>Differential</i>	
4.3.5	<i>Experimental design</i>	128
4.3.5.1	<i>Experiment 1: Temporal effects of OSFs on oocyte developmental competence following co-culture of intact COCs with DOs at either 0 or 9 hour of IVM</i>	128
4.3.5.2	<i>Experiment 2: Assessment of oocyte developmental competence following treatment of COCs with GDF-9 or BMP-15 at either 0 or 9 hour of IVM</i>	130
4.3.6	<i>Statistical analyses</i>	130
4.4	Results	132
4.4.1	<i>Experiment 1: Temporal effects of OSFs on oocyte developmental competence following co-culture of intact COCs with DOs at either 0 or 9 hour of IVM</i>	132
4.4.2	<i>Experiment 2: Assessment of oocyte developmental competence following treatment of COCs with GDF-9 or BMP-15 at either 0 or 9 hour of IVM</i>	133
4.5	Discussion	136
4.6	Acknowledgements	140
4.7	References	141
CHAPTER 5: FINAL DISCUSSION		147
Final Discussion		148
<i>Future directions</i>		153
REFERENCES		156
APPENDICES		181
Appendix 1: Additional Experiments		182
Appendix 2: TUNEL Assay		187
Appendix 3: Culture Media		191
Appendix 4: Reagents		195
Appendix 5: Blastocyst Scoring System		197
Appendix 6: Published Version of Chapter 2		199
Appendix 7: Published Version of Chapter 3		200

Abstract

Paracrine factors secreted by the oocyte (oocyte-secreted factors, OSFs) regulate a broad range of cumulus cell functions. The capacity of oocytes to regulate their own microenvironment by OSFs may in turn contribute to oocyte developmental competence. The aim of this thesis was to examine whether cumulus cells exhibit a low incidence of apoptosis due to their close association with oocytes and their exposure to OSFs, and to investigate if OSFs have a direct influence on bovine oocyte developmental competence during in vitro maturation (IVM).

This thesis includes a series of studies designed to examine by various means the nature of the paracrine network of bone morphogenetic proteins (BMPs) and their binding proteins involved in the regulation of cumulus cell apoptosis. OSFs, in particular BMP-15 and BMP-6, but not growth differentiation factor 9 (GDF-9), reduced apoptosis of cumulus cells by following a gradient from the site of the oocytes. Moreover, follistatin and a BMP6 neutralizing antibody, which antagonized the anti-apoptotic effects of BMP15 and BMP6, respectively, whether alone or combined, blocked ~50% of the anti-apoptotic actions of oocytes. These results demonstrated that OSFs, particularly BMP-15 and BMP-6, maintain the low incidence of apoptosis by establishing a localized gradient of bone morphogenetic proteins.

Results from the present thesis also demonstrated that OSFs enhance oocyte developmental competence during IVM, whether in their native form as an uncharacterized mix of growth factors secreted by the oocyte, throughout the oocyte

maturation period, or as exogenous BMP-15 and GDF-9, during the first 9 hour of IVM. Also, OSFs improved embryo quality as evident by increased blastocyst total and trophoctoderm cell numbers. These results were further verified in neutralization experiments of the exogenous growth factors and of the native OSFs. Follistatin and the kinase inhibitor SB-431542, which antagonize BMP-15 and GDF-9, respectively, neutralized the stimulatory effects of the exogenous growth factors, and impaired the developmental competence of control cumulus-oocyte complexes (COCs).

The work presented in this thesis has provided multiple lines of evidence that OSF-regulation of the COC microenvironment is an important determinant of cumulus cell apoptosis and oocyte developmental programming.