10.1071/RD13007_AC
© CSIRO 2014
Supplementary Material: *Reproduction, Fertility and Development,* 2014, 26(2), 337-345.

Supplementary Material

Table S1. Details of primers used for quantitative reverse transcription-polymerase chain reaction of granulosa and theca cells

Gene abbreviation	Description[1]	Primer sequence forward and reverse	Product size (bp)	GeneBank accession no.
AMH	Anti-Müllerian hormone	5' GGACTTCCTGGCCTTGGT	161	NM_173890
		3' GGGTCTTTCGTGTGAAGCAG		
CYP19A	BT Cytochrome P450, family XIX, aromatase	5' CAAGCTCTCCTTCTCAAACCAGAC	142	NM_174305
		3' GCTTCTCTGCTGTTGAAATCCTG		
ESR1	Estrogen receptor 1 or alpha	5' TCCAGGGAGAAGAGTTTGTGTG	111	NM_001001443
		3' GGTGGATGTGGTCCTTCTCTTC		
ESR2	Estrogen receptor 2 or beta	5' AAGAGTCCTTGGCGTGAAAC	129	NM_174051
		3' GGCATCCCTCTTTGAACTTG		
FSHR	BT follicle stimulating hormone receptor	5' GGATGCCATCATCGACTCTG	133	NM_174061

		3’ TGACTCGAAGCTTGGTGAGAAC		
LHCGR	BT Luteinizing hormone/chorionic gonadotropin receptor	5’ GCACGCCTGGAGAAGATG 3’ CTGAATGGACTCTAGCCCGTAG	117	NM_174381
HSD3B1	HS Hydroxy-delta-5-steroid dehydrogenase, 3 beta- and sterid delta-isomerase 1	5’ GCTGGGCCCAACTCCTAC 3’ TTTCAGAGCCCACCCATTAG	141	NM_174343
VCAN	Chondroitin sulface proteoglycan, versican	5’ TCATAGCCACCCCAGAGC 3’TTCCTTCCCCATCATGTCTC	143	NM_181035
PTGS2	Prostaglandin-endoperoxide synthase 2, COX2	5’ TATCCCGCCCTTCTGGTAG 3’CACCGAAAGTGCTAGGCTTC	145	NM_174445
PTX3	Pentraxin-related gene 3	5’ TGGGTCAAAGCCACAGAAG 3’GCGACCAGTCTGTTTTCCTC	140	NM_001076259

[1] BT, *Bos Taurus*; HS, *Homo sapiens*.

Table S2. Details of primers used for quantitative reverse transcription-polymerase chain reaction of cumulus cells

Gene abbreviation	Description[1]	Primer sequence (5'-3') forward and reverse	Product size (bp)	GeneBank accession no.
ESR1	Estrogen receptor 1 or alpha	5' TCCAGGGAGAAGAGTTTGTGTG 3' GGTGGATGTGGTCCTTCTCTTC	111	NM_001001443
VCAN	Chondroitin sulface proteoglycan, versican	5' TCATAGCCACCCCAGAGC 3' TTCCTTCCCCATCATGTCTC	143	NM_181035
PTGS2	Prostaglandin-endoperoxide synthase 2, COX2	5' TATCCCGCCCTTCTGGTAG 3' CACCGAAAGTGCTAGGCTTC	145	NM_174445
PTX3	Pentraxin-related gene 3	5' TGGGTCAAAGCCACAGAAG 3' GCGACCAGTCTGTTTTCCTC	140	NM_001076259
AVEN	HS Apotosis, caspase activation inhibitor	5' CGAGGAGAAAGAGTGGGATG 3' CTGAACCAATTCAGCAGCAA	139	XM_590221

HSP90B1	HS Heat shock protein 90, beta (Grp94), member 1, (Endoplasmin precursor)	5' CCCATATGGCAGAGACCATC 3' GGATATATGCCATGGGGTCA	100	NM_174700
WASL	Wiskott-Aldrich syndrome like Regulation of the cortical actin cytoskelton	5' CCTTTGCTGGAGATACCTGTC 3' GGGGTCTCGTCTTTTCTCAG	128	NM_174219
BAG3	HS BCL2-associsted athanogene 3	5' CCAGCCAGATAAACCGAGTG 3' AGGACGAGGAGCAGTCTGAG	116	NM_001082471
BUB3	Budding uninhibited by benzimidazoles 3	5' GCTGGGATCAGACGGTTAAA 3' AAGTCCCACACCAGCACTCT	148	NM_001076177
ATRX	Alpha thalassenmia/mental retardation syndrome X-linked / Homo sapiens similar to transcriptional regulator ATRX (LOC652455)???	5' GCTTGTGGACAACAGGTCAA 3' CATCTACACTGTTCATCCATTCC	149	XM_592333
EGR1	Early growth response (protein) 1	5' GGGAGAAAGTTTGCCAGGAG	124	NM_001045875

		3’ ACGGGTACGAAGGGAGAGAG		
TNFAIP6	HS Tumour necrosis factor, alpha-induced protein 6	5’ TGTCCTGCTATGGGAAGAGG 3’ TGCTTGTAGGTGGCAAGATG	186	NM_001007813

[1] BT, *Bos Taurus*; HS, *Homo sapiens*.

Table S3. Effect of cumulus cell biopsy before or after maturation on development embryos (Experiment 2a, 6 or 7 replicates)

Culture method			No. of	Cleaved	Blastocyst yield (%)		
IVM	IVF	IVC	oocytes[1]	(%)	Day 7	Day 7–9	Hatching[2]
Biopsy/ Group	Group	Group	119	79.7[2] 3.7	18.4[2] 2.3	22.2[2] 1.4	42.2[2] 11.8
Group	Group	Group	116	77.7[2] 2.7	26.0[2] 2.6	30.4[2] 3.4	53.1[2] 9.1
	P value			*P* = 0.7	*P* = 0.05	*P* = 0.07	*P* = 0.2
Group	Biopsy/ Group	Group	135	85.1[2] 3.7	27.9[2] 4.6	36.9[2] 3.4	56.2[2] 7.6
Group	Group	Group	141	84.1[2] 3.2	27.9[2] 3.2	35.0[2] 3.5	52.5[2] 5.5
	P value			*P* = 1.0	*P* = 0.8	*P* = 0.8	*P* = 0.7

[1] Each droplet contained 50 oocytes in IVM or 20 oocytes or zygotes in IVF and IVC, irrespective of treatment.

[2] Hatching blastocysts at Day 7-9 as a percentage of total blastocysts.

10.1071/RD13007_AC
© CSIRO 2013
Supplementary Material: *Reproduction, Fertility and Development*

Table S4. Effect of cumulus cell biopsy before or after maturation and the following IVM, IVF and IVC in the polyester mesh individual culture on development embryos (Experiment 2b, 8 replicates)

Culture method			No. of	Cleaved	Blastocyst yield (%)		
IVM	IVF	IVC	oocytes[1]	(%)	Day 7	Day 7–9	Hatching[2]
Biopsy/ L mesh	L mesh	S mesh	147	75.7[a] 3.5	10.1[a] 2.8	12.9[a] 3.2	30.3[a] 13.0
Group	Group	Group	116	72.2[a] 2.7	24.5[a] 2.8	26.4[a] 3.5	58.8[a] 8.9
	P value			*P* = 0.4	*P* = 0.003	*P* = 0.01	*P* = 0.09
Group	Biopsy/ L mesh	S mesh	151	76.0[a] 6.0	29.2[a] 2.2	32.0[a] 2.7	53.5[a] 9.7
Group	Group	Group	162	80.8[a] 1.5	36.5[a] 3.2	37.1[a] 3.3	67.9[a] 9.9
	P value			*P* = 0.3	*P* = 0.08	*P* = 0.2	*P* = 0.3

[1] Each droplet contained 20 oocytes, irrespective of treatment

[2] Hatching blastocysts at Day 7-9 as a percentage of total blastocysts.