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Supplemental Material

Effects of Cyanobacterial Harmful Algal Bloom Toxin Microcystin-LR on Gonadotropin-Dependent Ovarian Follicle Maturation and Ovulation in Mice

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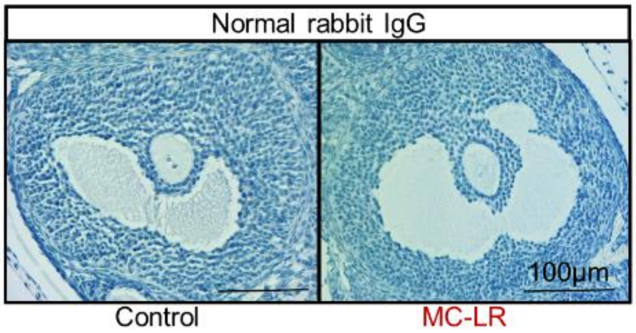

Figure S1. Detection of non-specific antibody binding in mouse ovary for the immunohistochemistry using normal rabbit IgG.

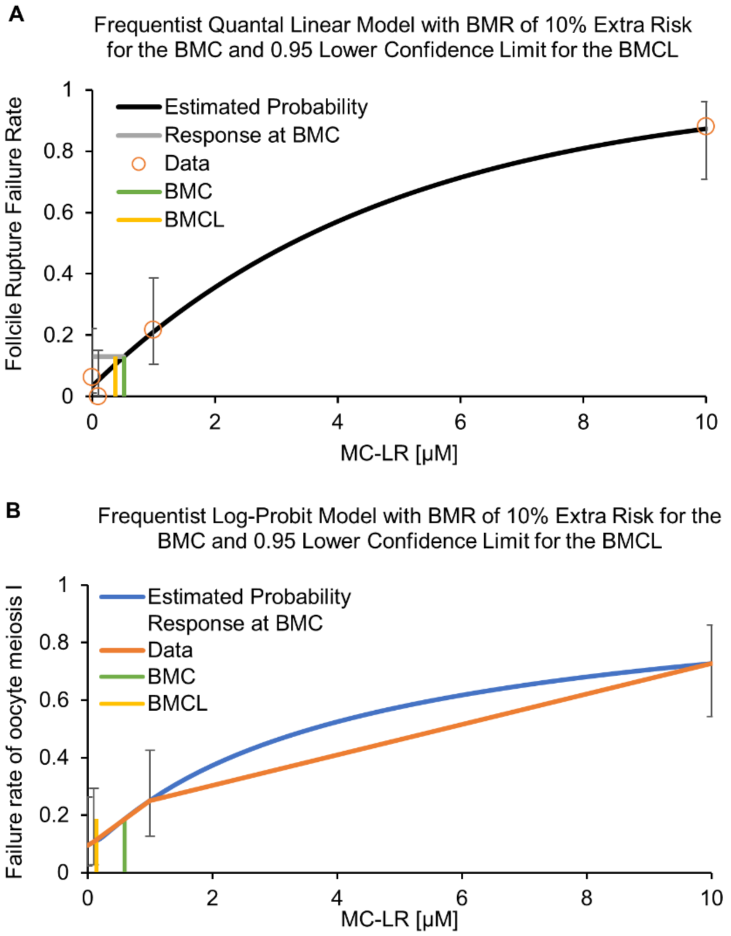

Figure S2. The *ex vivo* point of departure of MC-LR on failed follicle rupture and oocyte meiosis based on benchmark dose modeling. (A) Frequentist quantal linear model with BMR of 10% extra risk for the BMC and 95% lower confidence limit for the BMCL. (B) Frequentist Log-Probit model with BMR of 10% extra risk for the BMC and 95% lower confidence limit for the BMCL. BMR: benchmark response; BMC: benchmark concentration; BMCL: 95% lower confidence limit of BMC.

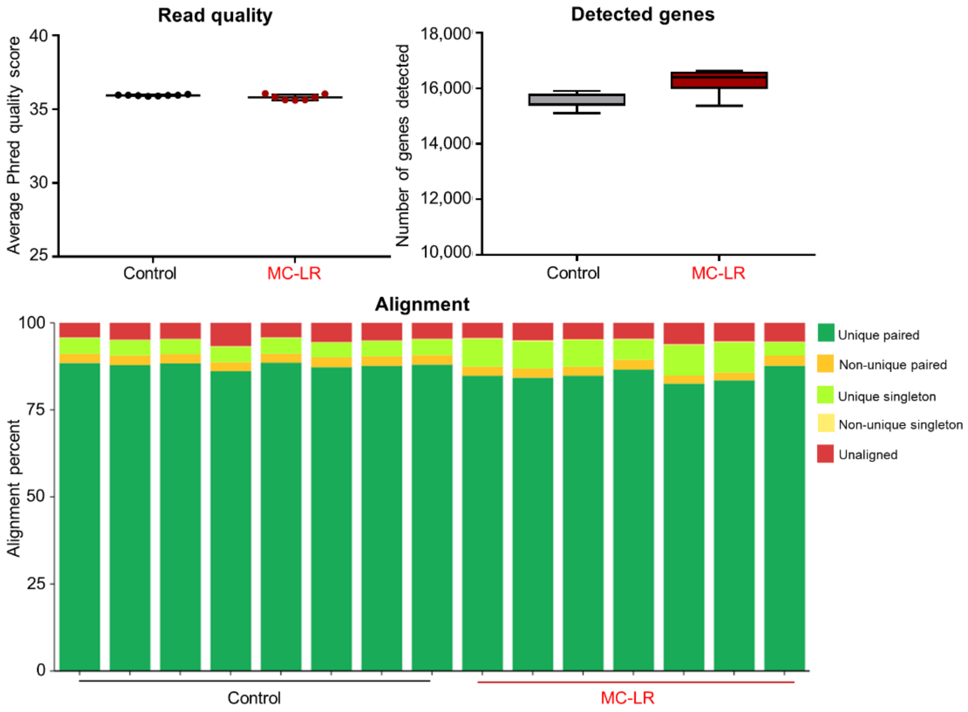

Figure S3. Quality control of single-follicle RNA sequencing reveals good sequencing quality, including sequencing read quality, number of detected genes, and read alignments. Data for Figure S3 can also be found in Table S26.

Table S1. Primer sequences used for RT-qPCR in this study.

Target gene	**Forward Primer (5'-3')**	**Reverse Primer (5'-3')**
Mouse		
Gapdh	CATCACTGCCACCCAGAAGACTG	ATGCCAGTGAGCTTCCCGTTCAG
Inha	CAGGCTATCCTTTTCCCAGCTAC	AAGTCACCTGGTGGCTGCGTAT
Inhba	GGAGATAGAGGACGACATTGGC	ACGCTCCACTACTGACAGGTCA
Inhbb	CTCCGAGATCATCAGCTTTGCAG	GGAGCAGTTTCAGGTACAGCCA
Comp	GTGCCCAACTTTGACCAGAGTG	ACAGGCATCACCCACAAAGTCG
Cyp19a1	CATGGTCCCGGAAACTGTGA	GTAGTAGTTGCAGGCACTTC
Fshr	GAGGCAGATGTGTTCTCCAACC	TCGGAGACTGGGAAGATTCTGG
Lhcgr	GACGCTAATCTCGCTGGAGT	GGCCTGCAATTTGGTGGAAG
Pappa	CAGAAAGCCAGCACCTGTAGCT	GGCAAAGGTCACATGCTGATCC
Human		
Gapdh	GTCTCCTCTGACTTCAACAGCG	ACCACCCTGTTGCTGTAGCCAA
Inha	TCCCAAGCCATCCTTTTCCCAG	TCACCTGGCGGCTGCGTGTAT
Inhba	GGATGACATTGGAAGGAGGGCA	ACTGACAGGTCACTGCCTTCCT
Inhbb	GAAATCATCAGCTTCGCCGAGAC	GGCAGGAGTTTCAGGTAAAGCC
Comp	GGAGATGCTTGTGACAGCGATC	TGAGTCCTCCTGGGCACTGTTA
Cyp19a1	GACGCAGGATTTCCACAGAAGAG	ATGGTGTCAGGAGCTGCGATCA
Fshr	GGTTTGTCCTCACCAAGCTTCG	GGTTGGAGAACACATCTGCCTC
Lhcgr	GGAGAAGATGCACAATGGAGCC	CGTGGCAATTAGCCTCTGAATGG
Pappa	GGAACTGAAGAGAGTGAGCCATC	CGTCGCATTGTTCACCTTGGTC

Table S2. Numeric data for Figure 1B: body weight (g).

Day	**Control (n=5)**		**MC-LR (n=5)**	
	Mean	SD	Mean	SD
1	23.3	1.6	22.9	1.6
2	23.9	1.5	23.8	1.7
3	24.1	1.6	23.7	1.7
4	24.3	1.4	24.6	1.6
5	24.5	1.6	24.7	1.6
6	24.6	1.4	25.2	1.8
7	24.4	1.7	25.2	1.9
8	24.6	1.8	25.4	1.9
9	24.4	1.6	25.3	2.2
10	24.5	1.8	24.9	2.1
11	24.3	1.8	25.1	2.1
12	24.6	1.6	25.4	1.7
13	24.9	1.7	25.9	1.8
14	25.1	1.6	24.8	1.8
15	25.4	1.8	25.3	2.2
16	25.2	1.6	25.5	2.4
17	25.0	1.7	25.7	2.5
18	25.4	2.0	25.9	2.4
19	25.4	2.0	25.7	2.4
20	26.1	2.0	26.2	2.4
21	25.7	1.9	26.0	2.4
22	25.8	1.9	25.8	2.5
23	26.4	2.0	26.1	2.1
24	26.1	2.0	25.9	2.0
25	26.1	2.1	26.0	2.2
26	26.5	1.9	26.7	2.4
27	26.8	1.5	26.9	2.2
28	26.8	1.7	26.9	2.1
29	27.4	2.1	26.9	2.0
30	26.9	2.2	27.2	2.1
31	27.1	2.2	27.2	2.1
32	26.8	2.1	27.1	2.2
33	27.1	1.9	27.2	2.0
34	27.9	2.3	27.7	1.9
35	27.6	2.6	27.4	1.8
36	27.4	2.7	27.3	1.9
37	27.6	2.7	27.3	2.1
38	28.0	2.8	27.5	1.8
39	28.1	2.7	27.6	1.8
40	27.7	2.5	27.9	2.0
41	27.5	2.1	28.1	2.2
42	27.9	2.5	28.4	2.2
43	28.2	2.9	29.1	2.3

Table S3. Numeric data for Figure 1C: estrous cycle days.

	Control (n=5)		**MC-LR (n=5)**	
	Mean	SD	Mean	SD
Proestrus	3.2	0.8	3.4	1.1
Estrus	3.2	0.4	3.8	1.9
Metestrus	3	0.7	3.4	0.5
Diestrus	5.6	0.5	4.4	1.1

Table S4. Numeric data for Figure 1E: total numbers of various stages of follicles per ovary in 1xPBS or MC-LR treated mice.

Follicle stage	**Control**					**MC-LR**				
	1	**2**	**3**	**4**	**5**	**1**	**2**	**3**	**4**	**5**
Primordial	704	615	654	834	631	918	587	650	623	559
Primary	348	254	334	204	358	383	343	233	376	452
Secondary	143	133	128	111	226	183	120	100	156	173
Antral	19	12	12	14	23	12	15	15	16	24
Atretic	97	52	62	84	99	94	99	78	94	93

Table S5. Numeric data for Figure 1F: total numbers of corpora lutea (CL) per ovary in 1xPBS or MC-LR treated mice.

Control					**MC-LR**				
1	**2**	**3**	**4**	**5**	**1**	**2**	**3**	**4**	**5**
8	17	13	8	13	9	4	4	9	7

Table S6. Numeric data for Figure 2B: total numbers of ovulated oocytes from mice treated with 1xPBS or MC-LR.

Group	**Mean**	**SD**	**n/group**
Control	38	8	9
MC-LR	25	10	10

Table S7. Numeric data for Figure 2D: percentages of ovulated MII oocytes (%) in mice treated with 1xPBS or MC-LR.

Group	**Mean**	**SD**	**n/group**
Control	97.1	2.8	9
MC-LR	98.5	2.4	10

Table S8. Numeric data for Figure 2F: total numbers of un-ovulated late-staged antral follicles per mouse in 1xPBS or MC-LR treatment groups.

Group	**Mean**	**SD**	**n/group**
Control	9	2	4
MC-LR	16	4	4

Table S9. Numeric data for Figure 3A: total numbers of ovulated oocytes and percentages of MII oocytes from mice treated with 1xPBS or MC-LR at 1 hour before hCG injection.

Group	**Number of ovulated oocytes**		**Percentage of MII oocytes (%)**		**n/group**
	Mean	SD	Mean	SD	
Control	40	10	94	3	12
MC-LR	42	12	95	3	14

Table S10. Numeric data for Figure 3B: total numbers of ovulated oocytes and percentages of MII oocytes from mice treated with 1xPBS or MC-LR during follicle maturation window.

Group	**Number of ovulated oocytes**		**Percentage of MII oocytes (%)**		**n/group**
	Mean	SD	Mean	SD	
Control	36	11	95	3	8
MC-LR	24	9	92	6	9

Table S11. Numeric data for Figure 4B: follicle diameter (μm).

Group	**Mean**	**SD**	**n/group**
Control	434.9	29.2	95
MC-LR	438.1	35.1	91

Table S12. Established follicle maturation-related genes and their functions.

Gene symbol	**Gene name**	**Functions and references**
Inha	Inhibin subunit alpha	Synthesis and secretion of ovarian peptide hormones of inhibin and activin and regulation of folliculogenesis [1]
Inhba	Inhibin subunit beta A	
Inhbb	Inhibin subunit beta B	
Comp	Cartilage oligomeric matrix protein	A biomarker of FSH-dependent follicle maturation [2]

Cyp19a1	Cytochrome P450 family 19 subfamily A member 1	Estrogen synthesis [3]
Fshr	Follicle stimulating hormone receptor	Membrane receptor of FSH and activating FSH target signaling and genes [4]
Lhcgr	Luteinizing hormone/choriogonadotropin receptor	Activation of LH mediated signaling pathways and target genes [5]
Pappa	Pregnancy-associated plasma protein A	IGFBP protease; regulation of folliculogenesis and steroidogenesis [6]

Table S13. Numeric data for Figure 4D: gene expression.

Gene	Group	Mean	SD	n/group
Inha	Control	74.4	35.8	24
	MC-LR	52.1	23.2	24
Inhba	Control	44.5	26.1	19
	MC-LR	35.8	17.5	23
Inhbb	Control	3.3	2.2	23
	MC-LR	2.4	1.1	24
Comp	Control	1.7	0.7	20
	MC-LR	1.3	0.5	25
Cyp19a1	Control	11.1	6.8	24
	MC-LR	7.8	5.2	23
Fshr	Control	2.1	1.5	23
	MC-LR	2.0	1.1	23
Lhcgr	Control	5.1	4.2	24
	MC-LR	3.0	2.2	22
Pappa	Control	0.3	0.3	23
	MC-LR	0.1	0.0	23

Table S14. Numeric data for Figure 4E: Immunohistochemistry quantification for LHCGR.

	Mean	SD	n/group
Control	37.8	6.5	5
MC-LR	8.0	2.2	5

Table S15. Numeric data for Figure 4F: Immunohistochemistry quantification for PAPPA.

	Mean	SD	n/group
Control	27.1	4.6	5
MC-LR	5.2	2.3	5

Table S16. Numeric data for Figure 5B: follicle survival rate (%) upon MC-LR treatment.

Day	Group	Mean	SD	n/group
0	0 µM	100.0	0.0	43
	0.1 µM	100.0	0.0	40
	1 µM	100.0	0.0	44
	10 µM	100.0	0.0	39
2	0 µM	100.0	0.0	43
	0.1 µM	100.0	0.0	40
	1 µM	100.0	0.0	44
	10 µM	100.0	0.0	39
4	0 µM	97.6	4.1	43
	0.1 µM	94.8	4.5	40
	1 µM	97.8	3.9	44
	10 µM	100.0	0.0	39
6	0 µM	95.2	4.1	43
	0.1 µM	92.1	8.4	40
	1 µM	95.6	3.9	44
	10 µM	92.3	7.7	39

Table S17. Numeric data for Figure 5C: follicle diameter (µm) upon MC-LR treatment.

Day	Group	Mean	SD	n/group
0	0 µM	149.7	13.7	43
	0.1 µM	150.7	11.3	40
	1 µM	150.0	13.1	44
	10 µM	151.0	11.0	39
2	0 µM	194.9	25.4	43
	0.1 µM	197.0	19.2	40
	1 µM	194.9	27.0	44
	10 µM	195.8	22.2	39
4	0 µM	279.9	30.2	43
	0.1 µM	280.9	28.4	40
	1 µM	277.1	38.7	44
	10 µM	286.2	34.4	39
6	0 µM	337.5	34.6	43
	0.1 µM	335.9	43.0	40
	1 µM	333.4	41.1	44
	10 µM	330.6	36.8	39

Table S18. Numeric data for Figure 5D: estradiol concentration (ng/mL) upon MC-LR treatment.

Group	Mean	SD	n/group
0 µM	26.3	18.4	10
0.1 µM	28.9	18.4	11
1 µM	22.4	23.7	12
10 µM	21.1	10.5	10

Table S19. Numeric data for Figure 5E: testosterone concentration (pg/mL) upon MC-LR treatment.

Group	Mean	SD	n/group
0 µM	1142.9	714.3	10
0.1 µM	964.3	696.4	12
1 µM	1000.0	875.0	11
10 µM	1214.3	928.5	12

Table S20. Numeric data for Figure 5G: follicle rupture percentage (%) and MII oocyte percentage (%) upon MC-LR treatment.

	Group	Mean	SD	n/group
Follicle rupture percentage	0 µM	100.0	0.0	43
	0.1 µM	100.0	0.0	40
	1 µM	77.5	7.9	44
	10 µM	11.9	3.2	39
MII oocyte percentage	0 µM	91.1	8.4	43
	0.1 µM	89.2	1.4	40
	1 µM	74.6	4.0	44
	10 µM	27.2	7.9	39

Table S21. Numeric data for Figure 5H: progesterone concentration (ng/mL) upon MC-LR treatment.

Group	Mean	SD	n/group
0 µM	94.5	59.1	14
0.1 µM	103.6	62.7	14
1 µM	74.6	48.3	17
10 µM	23.6	10.9	16

Table S22. Numeric data for Figure 6B: expression of follicle maturation-related genes.

Gene	Day	Group	Mean (a.u.)	SD	n/group
Comp	0	Control	1.3	0.5	8
	2	Control	1.3	0.3	8
	4	Control	1.5	0.6	8
		MC-LR	2.6	1.4	7
	6	Control	16.7	5.3	8
		MC-LR	4.6	2.8	8
Inha	0	Control	143.5	60.9	8
	2	Control	252.2	50.0	8
	4	Control	265.2	41.3	8
		MC-LR	221.7	34.8	7
	6	Control	352.2	71.7	8
		MC-LR	178.3	52.2	8
Inhba	0	Control	0.9	0.4	8
	2	Control	17.6	25.4	8
	4	Control	80.5	22.7	8
		MC-LR	58.8	27.6	7

	6	Control	114.5	17.3	8
		MC-LR	79.4	12.8	8
Inhbb	0	Control	2.5	1.5	8
	2	Control	10.4	9.2	8
	4	Control	17.7	2.8	8
		MC-LR	9.6	4.1	7
	6	Control	17.6	4.6	8
		MC-LR	11.9	3.5	8
Cyp19a1	0	Control	0.5	0.2	8
	2	Control	1.0	0.9	8
	4	Control	2.8	0.6	8
		MC-LR	2.1	0.5	7
	6	Control	5.5	1.1	8
		MC-LR	2.9	1.6	8
Pappa	0	Control	0.1	0.1	8
	2	Control	0.1	0.1	8
	4	Control	0.1	0.1	8
		MC-LR	0.1	0.0	7
	6	Control	0.6	0.2	8
		MC-LR	0.1	0.1	8
Fshr	0	Control	1.6	0.8	8
	2	Control	4.5	1.0	8
	4	Control	4.7	1.0	8
		MC-LR	5.1	1.2	7
	6	Control	5.2	1.1	8
		MC-LR	3.3	1.2	8
Lhcgr	0	Control	1.7	0.8	8
	2	Control	3.1	0.5	8
	4	Control	2.0	0.5	8
		MC-LR	1.5	0.4	7
	6	Control	9.2	3.4	8
		MC-LR	1.7	1.2	8

Note: the mRNA expression levels of each gene were normalized by the expression of glyceraldehyde-3-phosphate dehydrogenase (*Gapdh*).

Table S23. Numeric data for Figure 6C: expression of follicle maturation-related genes in isolated mural granulosa cells.

Gene	**Group**	**Mean (a.u.)**	**SD**	**n/group**
Comp	Control	4.8	3.9	9
	MC-LR	1.8	0.8	8
Inha	Control	86.3	13.2	9
	MC-LR	48.2	23.9	8
Inhba	Control	55.8	13.9	9
	MC-LR	39.7	15.1	8
Inhbb	Control	6.6	1.7	9
	MC-LR	4.2	2.6	8

Cyp19a1	Control	1.5	0.2	9
	MC-LR	1.0	0.5	8
Pappa	Control	0.4	0.1	9
	MC-LR	0.1	0.1	8
Fshr	Control	1.9	0.6	9
	MC-LR	1.2	0.7	8
Lhcgr	Control	2.1	1.2	9
	MC-LR	0.4	0.3	8

Table S24. Established ovulatory genes and their identified functions in ovulation.

Gene symbol	**Gene name**	**Functions and references**
Pgr	Progesterone receptor	Follicle rupture [7]
Runx1	Runt-related transcription factor 1	Progesterone production [8]
Areg	Amphiregulin	Activation of EGF signaling to regulate COC expansion, oocyte maturation, follicle rupture, and luteinization [9]
Ereg	Epiregulin	
Btc	Betacellulin	
Plau	Plasminogen activator, urokinase	ECM remodeling and follicle rupture [10]
Ptgs2	Prostaglandin-endoperoxide synthase 2	Prostaglandin production, COC expansion, and oocyte maturation [11, 12]
Tnfaip6	Tumor necrosis factor-inducible gene 6	COC expansion [13]

Table S25. Numeric data for Figure 6D: expression of established ovulatory genes in vehicle or MC-LR treated follicles.

Gene	Group	Mean (a.u.)	SD	n/group
Pgr	Control	1.0	0.4	12
	MC-LR	0.1	0.1	10
Runx1	Control	1.0	0.1	12
	MC-LR	0.5	0.2	10
Areg	Control	1.0	0.6	12
	MC-LR	0.5	0.3	10
Ereg	Control	1.0	0.5	12
	MC-LR	0.5	0.3	10
Btc	Control	1.0	0.3	12
	MC-LR	0.4	0.2	10
Plau	Control	1.0	0.4	12
	MC-LR	0.3	0.1	10
Ptgs2	Control	1.0	0.4	12
	MC-LR	0.3	0.1	10
Tnfaip6	Control	1.0	0.3	12
	MC-LR	0.4	0.1	10

Table S26. Quality control analysis of single-follicle RNA sequencing, including sequencing read quality and number of detected genes in control and MC-LR treated follicles.

	Average Phred quality score		**Number of detected genes (x10^3)**					**n/group**
	Mean	SD	Max	Upper Quartile	Median	Lower Quartile	Min	
Control	35.92	0.04	15.94	15.79	15.46	15.43	15.14	8
MC-LR	35.78	0.19	16.67	16.60	16.44	16.06	15.40	7

Table S27. Original data for Figure 7C: Gene Ontology (GO) analysis of differentially expressed genes; Top 10 biological process (BP) enrichment results.

GO_BP_Term	Count	Gene ratio	FDR
transmembrane receptor protein tyrosine kinase signaling pathway	43	0.43	4.4351E-08
positive regulation of apoptotic process	98	0.29253731	2.9282E-08
positive regulation of angiogenesis	49	0.40495868	2.8712E-08
positive regulation of cell proliferation	141	0.2601476	2.2946E-08
inflammatory response	104	0.30232558	1.1675E-09
axon guidance	59	0.39597315	1.1133E-09
multicellular organism development	251	0.24392614	1.0842E-12
positive regulation of cell migration	83	0.408867	1.9365E-15
angiogenesis	93	0.38912134	1.044E-15
cell adhesion	170	0.35051546	1.8929E-24

Table S28. Original data for Figure 7D: Gene Ontology (GO) analysis of differentially expressed genes; Top 10 molecular function (MF) enrichment results.

GO_MF_Term	Count	Gene ratio	FDR
protein binding	785	0.19183773	3.47E-13
heparin binding	61	0.40397351	1.11E-10
integrin binding	47	0.47	1.11E-10
protein homodimerization activity	187	0.23433584	1.14E-07
calcium ion binding	165	0.2360515	6.56E-07
collagen binding	29	0.49152542	1.19E-06
extracellular matrix binding	19	0.67857143	1.19E-06
fibronectin binding	19	0.65517241	2.35E-06
transmembrane receptor protein tyrosine kinase activity	26	0.48148148	1.04E-05
receptor binding	104	0.25242718	1.41E-05

Table S29. Original data for Figure 7E: Gene Ontology (GO) analysis of differentially expressed genes; Top 10 cellular component (CC) enrichment results.

GO_CC_Term	Count	Gene ratio	FDR
membrane	1372	0.19605602	1.313E-40
proteinaceous extracellular matrix	138	0.43670886	2.5886E-32
extracellular region	445	0.25385054	1.1849E-31
cell surface	191	0.3036566	1.8126E-21
extracellular matrix	106	0.36054422	3.6617E-17
extracellular exosome	553	0.20680628	6.3114E-17
extracellular space	347	0.23071809	6.3114E-17
cell junction	189	0.2632312	4.9111E-14
basement membrane	48	0.49484536	3.7405E-13
cytoplasm	1160	0.17493591	8.4855E-12

Table S30. Original data for Figure 7F: Top 10 KEGG pathway enrichment results of differentially expressed genes. KEGG: Kyoto Encyclopedia of Genes and Genomes.

KEGG_Term	Count	Gene ratio	FDR
ECM-receptor interaction	51	0.57954545	2.8067E-16
Focal adhesion	81	0.39130435	1.3495E-13
PI3K-Akt signaling pathway	113	0.32193732	1.6404E-12
Proteoglycans in cancer	69	0.33990148	1.9592E-08
Pathways in cancer	109	0.27594937	9.9682E-08
Transcriptional misregulation in cancer	58	0.35151515	9.9682E-08
Axon guidance	48	0.37209302	3.1819E-07
MAPK signaling pathway	74	0.29482072	2.1515E-06
Amoebiasis	43	0.36752137	2.4771E-06
Hypertrophic cardiomyopathy (HCM)	33	0.41772152	3.1271E-06

Table S31. A summary of all overlapped 315 genes between MC-LR induced DEGs and previously identified FOXO1 target genes in the granulosa cells.

FOXO1 upregulated genes	**FDR**	**Fold change**	**FOXO1 downregulated genes**	**FDR**	**Fold change**
Spp1	2.11E-34	719.78	*Ache*	2.07E-05	-14.00
Serpinb2	4.19E-19	124.90	*Pappa*	1.28E-23	-8.42
Cemip	2.41E-14	76.18	*Gja5*	0.010856	-8.13
Ankrd1	8.14E-07	73.48	*Lhcgr*	2.79E-16	-6.81
Gbp5	7.23E-06	58.35	*Dnah2*	2.97E-32	-6.18
Eef1a2	9.42E-05	49.45	*Cacna1d*	9.89E-19	-5.10
Actc1	6.55E-07	48.60	*Pla2g1b*	0.000983	-4.18
Cldn2	1.26E-06	40.38	*Flvcr2*	2.4E-17	-3.79
Tnc	6.27E-26	33.03	*Fgf11*	3.28E-10	-3.44
Robo3	0.000251	21.22	*Hao2*	1.28E-15	-3.42
Edn1	7.53E-26	19.55	*Srcin1*	3.86E-25	-3.39
Ngf	0.03522	19.46	*Nxnl1*	8.73E-06	-3.34
Dusp4	4.89E-30	16.86	*Tmem25*	1.18E-13	-3.23
Hsd11b1	5.09E-05	16.84	*Nos3*	1.47E-10	-3.21
Muc6	0.000732	16.43	*Srgap3*	1.6E-06	-3.20
Tmod1	4.93E-17	16.27	*Cyp19a1*	6.27E-06	-3.18
Spink8	0.027359	15.60	*Cox4i2*	4.76E-08	-3.14
Prune2	2.99E-51	15.55	*Pcdh8*	0.006074	-3.08
Wnt16	0.009025	15.25	*Adgrg1*	1.01E-11	-3.03
Arid5a	2.79E-25	12.83	*Spdya*	0.000174	-2.94
Slfn2	1.29E-07	12.02	*Rapsn*	0.000418	-2.89
Creb5	8.14E-71	9.93	*Naglu*	6.03E-39	-2.89
Tnfrsf12a	1.26E-60	9.46	*Flt4*	1.52E-18	-2.87
Sfrp2	5.49E-08	8.50	*Ccrl2*	2.83E-05	-2.86
Pth2r	0.012956	8.23	*Fam180a*	0.00013	-2.86
Gpnmb	3.05E-06	8.09	*Pdxp*	1.43E-25	-2.83
Serpine1	7E-06	7.39	*Gldn*	3.46E-11	-2.79
B3gnt5	6.37E-06	7.24	*Nppc*	7.61E-05	-2.77
Tnnc1	0.000877	7.18	*Smpd3*	8.93E-05	-2.75
Zfp365	3.3E-25	7.09	*Slc35g2*	3.14E-09	-2.68
Megf10	1.74E-10	6.93	*Proser2*	1.98E-20	-2.54
Stmn4	2.18E-06	6.37	*Kcnh2*	4.54E-14	-2.49
Cdkn2b	5.14E-12	6.33	*Hsd17b1*	8.63E-19	-2.48
Xdh	1.38E-11	6.20	*Lmo2*	0.001455	-2.43
Mgp	1.73E-12	6.15	*Tmem107*	8.1E-14	-2.42
Fn1	1.15E-18	5.93	*Dock2*	0.000714	-2.41
Pamr1	1.53E-08	5.82	*Dhh*	5.6E-15	-2.38
Etv4	1.57E-08	5.55	*Sh3d21*	1.54E-07	-2.35
Thbd	1.6E-23	5.44	*Spata18*	4.57E-06	-2.34
Snai1	1.97E-14	5.31	*Sox8*	1.35E-05	-2.25

Slc9a9	8.26E-09	5.19	*Fam229b*	0.024098	-2.22
Tagln	1.36E-05	5.02	*Kctd14*	8.83E-06	-2.22
Pxdc1	2.88E-09	4.98	*Adamtsl2*	0.045963	-2.21
Bdnf	2.5E-09	4.81	*Pm20d2*	5.26E-10	-2.20
Fbxo32	3.57E-14	4.78	*Tbata*	0.004254	-2.19
Mreg	2.65E-09	4.57	*Pyroxd2*	0.00118	-2.16
Osmr	2.91E-05	4.54	*Prlr*	1.16E-11	-2.16
Prdm1	2.18E-07	4.44	*Lrrc56*	8E-05	-2.15
Ntn4	3.03E-08	4.39	*Cacna1a*	1.99E-08	-2.12
Adora2b	9.98E-05	4.25	*Acsbg1*	2.56E-11	-2.12
Rragd	5.38E-29	4.24	*Cbs*	2.9E-08	-2.10
Mmp28	0.000217	4.20	*E2f7*	1.58E-06	-2.08
Gadd45a	2.19E-35	4.03	*Rnf208*	0.004213	-2.03
Pdk4	1.16E-14	3.98	*Lyrm9*	6.22E-10	-2.03
Myc	8.35E-32	3.97	*Zfp879*	0.040633	-1.96
Hgf	6.83E-11	3.89	*Psmb9*	1.24E-10	-1.96
Lox	0.000106	3.85	*Cebpa*	1.03E-35	-1.95
Anxa1	5.93E-24	3.76	*Gsta2*	8.49E-05	-1.94
Synj2	4.21E-17	3.75	*Miip*	3.04E-05	-1.93
Gpr176	0.031448	3.71	*Plekhd1*	2.56E-17	-1.92
Hmgcll1	1.86E-05	3.53	*Mgarp*	4.19E-13	-1.92
Rnd3	3.86E-29	3.38	*Gstt3*	0.005121	-1.92
Dkk3	8.55E-18	3.37	*Plxnc1*	5.41E-40	-1.91
Gadd45b	4.85E-09	3.36	*Cbfa2t3*	0.005867	-1.91
Hspb1	3.01E-18	3.32	*Aldh3b1*	2.47E-09	-1.90
Cald1	3.78E-24	3.31	*Folr1*	4.24E-10	-1.90
Gypc	6.96E-12	3.28	*Rhbdl1*	6.58E-05	-1.89
Dock10	8.05E-05	3.25	*Fanca*	3.34E-06	-1.88
Cilp	0.010349	3.24	*Prkar2b*	3.65E-12	-1.87
Srgap1	9.4E-12	3.23	*Efnb3*	2.23E-05	-1.86
Fcgr2b	0.012425	3.21	*Slc16a3*	2.86E-08	-1.86
Scrn1	0.002486	3.12	*Pomgnt2*	2.14E-17	-1.86
Cyp7b1	0.030847	3.10	*Fbxo16*	5.88E-11	-1.85
Bmf	9.66E-16	3.07	*Fmc1*	5.38E-06	-1.85
Hmga1	2.12E-12	3.07	*Slc27a1*	1.68E-15	-1.84
Larp6	7.76E-06	3.06	*Scara5*	0.004585	-1.84
Ppfia4	0.001278	3.05	*Ccpg1os*	0.001801	-1.84
Zfp36	0.001061	3.05	*Grem2*	2.47E-11	-1.84
Rhoj	9.37E-16	2.99	*Nphp4*	1.98E-07	-1.81
Cd38	1.92E-06	2.95	*Sybu*	0.004977	-1.81
Serpinb8	0.001607	2.90	*Dhrs1*	4.98E-12	-1.80
Tlr4	0.000101	2.88	*Pm20d1*	0.000226	-1.80
Maff	3.5E-11	2.87	*Cyp17a1*	0.018829	-1.79
Cav1	3.13E-07	2.85	*Fbxo44*	0.025153	-1.78

Pmepa1	6.48E-21	2.85	*Rasd1*	2.58E-06	-1.77
Cd36	1.5E-17	2.80	*Ednrb*	0.034061	-1.76
Tgfb2	6.61E-07	2.79	*Tmem177*	1.84E-05	-1.75
F3	4.7E-09	2.76	*Map2k6*	7.89E-05	-1.74
Bmper	2.58E-06	2.75	*Ccdc142*	0.000466	-1.73
Tmeff1	6.74E-15	2.73	*Fbxl8*	0.007521	-1.72
Pear1	8.66E-11	2.72	*Angptl1*	2.57E-07	-1.72
Sptbn2	0.002188	2.68	*Sema7a*	2.73E-09	-1.72
Sh3bgrl2	7.39E-07	2.65	*Lrrc24*	6.01E-05	-1.71
Rab3b	0.027216	2.62	*Tssk6*	0.037664	-1.71
Rgcc	1.14E-10	2.54	*Rac3*	0.000443	-1.70
Sox9	6.79E-08	2.54	*Amigo2*	8.87E-11	-1.70
Pdgfrb	6.65E-12	2.48	*Dusp28*	0.001984	-1.67
Klf12	6.06E-10	2.48	*Tmem17*	0.011355	-1.67
Tpm1	2.23E-29	2.47	*Dnph1*	0.002209	-1.67
Spata13	6.76E-26	2.46	*Socs1*	0.022895	-1.67
Klhl29	0.008301	2.45	*Kcnt1*	0.000347	-1.66
Pkp2	2.16E-12	2.44	*Ankrd9*	0.000868	-1.66
Actn1	1.5E-28	2.42	*Mif4gd*	6.48E-11	-1.65
Anpep	9.22E-07	2.42	*Efna1*	1.25E-07	-1.65
Nfil3	0.003855	2.37	*Ptprz1*	0.018945	-1.64
Dusp1	0.002726	2.37	*Dnajb13*	0.005903	-1.63
Csrp1	3.63E-12	2.30	*Depdc1b*	0.025249	-1.62
Nid1	4.58E-06	2.27	*Crispld2*	1.28E-08	-1.62
Trpv2	0.017402	2.27	*Tst*	5.25E-06	-1.61
Orai2	5.64E-07	2.26	*Nr5a1*	8.82E-29	-1.59
Junb	0.049394	2.22	*Ppt2*	1.2E-11	-1.58
Adamts12	1.9E-12	2.22	*Kit*	0.000773	-1.57
Tjp2	1.41E-15	2.22	*Car14*	4.02E-05	-1.57
Shroom3	9.74E-06	2.20	*Dhrs11*	3.43E-05	-1.57
Vasn	2.91E-19	2.20	*Wdr90*	4.55E-05	-1.56
Nin	1.47E-46	2.19	*Trpv4*	0.040445	-1.56
Wipf1	4.67E-09	2.19	*Cdc25b*	5.64E-06	-1.56
Tes	3.99E-16	2.18	*Enkd1*	2.36E-05	-1.55
Fam129a	3.83E-09	2.17	*Cyb5d2*	0.029259	-1.54
Fam20a	0.00099	2.16	*Wnt6*	0.006427	-1.53
Map3k6	1.56E-06	2.16	*Csdc2*	9.05E-06	-1.53
Sorbs1	2.28E-09	2.15	*Clybl*	0.002102	-1.53
Wt1	0.031616	2.14	*Pex10*	0.016309	-1.52
Stk32c	0.00044	2.14	*Atl1*	0.000636	-1.52
Ccdc30	0.000885	2.14	*Naga*	5.6E-11	-1.52
Zyx	3.05E-16	2.09	*Sap18*	4E-07	-1.50
Cldn1	0.00837	2.06			
Scn7a	0.003023	2.06			

Lpar1	4.65E-06	2.06
Glis3	0.000225	2.05
Fjx1	1.92E-06	2.04
Shb	5.84E-13	2.03
Eng	0.000515	2.00
Ets1	0.000114	1.97
Tmem144	1.78E-06	1.97
Tpm2	0.020702	1.96
Rab30	0.00018	1.96
Lsp1	0.01133	1.96
Slc43a2	3.15E-05	1.94
Syt14	5.55E-09	1.94
Apba1	0.003343	1.94
Fam124a	0.034319	1.94
Arid5b	4.08E-10	1.93
Hectd2	2.58E-05	1.92
Dpysl3	8.85E-05	1.91
Cnn2	8.43E-06	1.91
Arhgap24	0.041478	1.89
Ston1	1.43E-07	1.88
Tpm4	3.35E-19	1.88
Klf6	0.025102	1.87
Satb2	0.003297	1.86
Lmcd1	0.007788	1.86
Col5a2	1.21E-08	1.85
S1pr1	0.002887	1.85
Clic4	4.27E-27	1.85
Aatk	0.006773	1.85
Efnb2	5.45E-05	1.84
Pcbp3	0.000977	1.82
Dact1	4.43E-05	1.82
Zfp423	0.003314	1.81
Lrrfip1	6.31E-09	1.80
Col11a2	0.033131	1.79
Shank3	0.002663	1.79
Cttnbp2nl	2.45E-08	1.78
Pde4a	0.002441	1.77
Enox1	0.001328	1.74
Stx3	1.07E-07	1.74
Kif5c	5.4E-06	1.74
Frmd4b	0.003996	1.74
Vgll3	0.022052	1.72
Dtx4	0.015132	1.72
Rassf5	0.002608	1.69

Hey1	0.011576	1.68
Stard13	0.000256	1.67
Mtr	1.64E-06	1.63
Tnfaip8	0.007956	1.63
Tns2	0.000129	1.62
Cebpd	0.027523	1.62
2900026A02Rik	0.001507	1.59
Fnip2	1.86E-08	1.58
Ldlrad4	0.001038	1.57
Tmbim1	1.68E-06	1.56
Peak1	1.58E-07	1.54
Aatf	2.85E-13	1.54
Fam117a	0.000727	1.53
Pid1	0.003746	1.52
Zfp608	0.000131	1.52
Zfp583	0.034876	1.51
Specc1	5.92E-05	1.50

Note: Herndon *et al.* transduced rat granulosa cells with empty adenovirus or constitutively active FOXO1 and performed RNA deep sequencing post 24 hours of FSH treatment [14]. By using this strategy, they found that FOXO1 regulated the majority of FSH target genes in granulosa cells. To investigate the role of FOXO1 upon MC-LR exposure, we compared the differentially expressed genes in MC-LR treated follicles with the previously identified FOXO1 target genes. We found that there were 315 overlapped genes between two data sets. The fold change and FDR of these 315 overlapped genes were shown in this table. FDR: false discovery rate.

Table S32. Original data for Figure 7G: representative overlapped genes between differentially expressed genes identified in MC-LR treated murine follicles and FOXO1 target genes identified in rat granulosa cells.

Gene Symbol	**Control**							
	1	**2**	**3**	**4**	**5**	**6**	**7**	**8**
Cyp17a1	283.6	375.7	516.1	122.1	302.3	290.6	171.7	163.6
Cdc25b	38.6	35.5	45.7	47.0	35.8	45.4	38.2	37.6
Cebpa	852.7	749.3	695.5	719.4	851.9	841.9	785.6	709.9
Nr5a1	256.4	257.8	277.0	229.6	281.2	259.4	254.2	237.6
Inhbb	14915.7	14562.6	15015.5	17930.4	15518.8	16375.3	16407.8	14194.1
Dhh	129.7	144.3	132.1	146.1	143.1	150.5	152.8	146.5
Inha	48139.5	50601.7	47552.4	50314.7	47624.0	49898.1	48650.6	47555.2
Hsd17b1	6372.2	6259.9	5705.8	6142.5	6585.0	5780.6	6482.0	6583.7
Pappa	2.8	6.2	3.2	4.6	3.8	4.8	3.7	2.6
Nppc	599.4	639.0	437.7	472.1	527.6	577.3	548.8	466.4
Lhcgr	77.9	113.9	86.0	40.7	119.0	50.9	74.2	66.4
Prlr	27.0	33.6	29.8	19.8	35.0	30.0	32.2	28.2
Csdc2	15.6	14.5	19.2	16.1	19.4	14.6	20.6	14.2

Inhba	4767.4	5329.2	5413.9	5269.3	6554.5	5663.1	6798.0	5682.3
Cyp19a1	28.2	38.2	43.7	36.0	39.8	32.6	55.1	26.6
Vgll3	2.9	3.4	2.3	1.6	2.9	0.8	2.1	3.6
Gadd45a	56.0	52.0	40.4	39.9	37.7	37.6	56.9	81.3
Rnd3	11.7	8.9	7.3	8.9	8.1	7.5	8.1	12.1
Shb	1.3	1.0	0.8	1.2	1.0	0.9	1.0	0.9
Cald1	5.0	4.3	3.7	2.2	3.6	3.4	3.8	2.7
Fn1	35.5	48.0	15.4	28.8	26.9	32.2	41.9	23.3
Adamts12	0.9	0.9	1.0	0.6	0.7	0.8	0.7	0.7
Ets1	2.4	2.9	0.9	1.5	1.0	2.3	2.5	1.3
Wt1	2.6	1.3	1.1	1.1	0.2	0.7	0.6	0.3
Tgfb2	2.0	0.6	0.6	0.6	0.8	0.5	0.8	0.6
Dusp1	69.6	82.7	72.4	54.7	72.7	54.2	47.1	270.2
Klf6	98.0	107.1	164.1	114.3	171.1	104.9	91.7	375.1
Junb	294.7	436.2	878.7	212.4	738.2	298.6	218.0	2011.5

Gene Symbol				MC-LR			
	1	2	3	4	5	6	7
Cyp17a1	166.1	84.3	123.2	257.8	143.3	191.6	128.2
Cdc25b	27.4	33.9	28.9	30.9	17.4	22.8	21.4
Cebpa	410.5	394.0	351.5	466.5	357.6	441.8	377.1
Nr5a1	184.0	163.0	147.8	164.8	155.3	176.4	149.5
Inhbb	11416.0	9303.5	8714.5	8519.0	5255.1	7204.0	4519.5
Dhh	72.7	75.9	81.0	71.3	37.1	44.8	41.3
Inha	22819.6	21151.2	20043.4	26609.5	16320.0	10276.7	16361.9
Hsd17b1	3541.6	3219.2	2543.5	3161.8	1794.7	1845.7	1640.5
Pappa	0.5	0.4	0.2	0.8	0.2	0.4	0.7
Nppc	277.7	248.9	148.3	416.2	65.9	76.5	128.3
Lhcgr	11.6	13.7	6.5	24.9	6.2	7.2	11.2
Prlr	14.1	9.8	8.1	18.9	14.0	13.1	18.2
Csdc2	13.6	12.6	11.0	9.1	8.4	10.1	12.3
Inhba	4104.0	2791.8	2162.9	3807.9	2476.5	2408.1	2453.7
Cyp19a1	26.2	7.8	7.5	21.8	7.7	6.8	5.6
Vgll3	4.4	2.6	2.7	2.6	6.7	7.4	3.2
Gadd45a	220.2	215.9	168.4	198.0	227.5	223.8	174.9
Rnd3	24.3	26.6	36.1	23.1	31.9	39.1	35.3
Shb	2.2	1.7	2.0	1.5	2.5	2.4	2.1
Cald1	12.8	9.5	12.3	7.8	13.1	17.5	11.1
Fn1	188.2	104.3	167.9	111.1	245.6	332.9	166.1
Adamts12	2.4	1.5	2.3	1.2	1.5	1.8	1.5
Ets1	4.9	2.8	3.5	3.3	3.6	4.7	2.8
Wt1	0.7	0.9	2.4	5.4	1.6	1.7	2.5
Tgfb2	2.0	1.4	1.7	2.4	2.6	2.2	3.7
Dusp1	197.7	172.9	276.1	178.9	216.4	170.1	309.0

Klf6	203.9	170.5	313.3	251.4	298.5	245.2	546.2
Junb	1024.9	786.7	1749.1	1202.9	1651.6	1008.5	2587.7

Note: The raw read counts were normalized based on the Transcripts Per Million (TPM) method. The normalized results were shown in this table.

Table S33. Numeric data for Figure 8A: PP1 phosphatase activity (picomoles) in vehicle or 10 µM MC-LR treated follicles.

Time	Group	Mean	SD	n/group
20 min	Control	665.6	88.4	3
	MC-LR	528.5	60.9	3
60 min	Control	1615.1	201.7	3
	MC-LR	1246.7	249.1	3
100 min	Control	2214.7	236.4	3
	MC-LR	1843.8	219.3	3

Table S34. Numeric data for Figure 8B: PP2A phosphatase activity (picomoles) in vehicle or 10 µM MC-LR treated follicles.

Time	Group	Mean	SD	n/group
20 min	Control	408.7	66.1	3
	MC-LR	349.2	71.6	3
60 min	Control	1016.3	245.6	3
	MC-LR	827.5	134.8	3
100 min	Control	1645.2	214.1	3
	MC-LR	1456.9	221.8	3

Table S35. Numeric data for Figure 8D: quantification of examined proteins by western blotting.

Protein	Group	Replicate 1	Replicate 2	Replicate 3
pAKT-S473	Control	8881.4	10757.4	24792.6
	MC-LR	23474.2	31486.7	31072.0
pAKT-T308	Control	24217.8	32324.9	41896.4
	MC-LR	12513.5	17617.4	18052.4
AKT	Control	26437.0	24387.7	47112.8
	MC-LR	30678.0	31704.0	33373.4
pFOXO1	Control	20304.1	16704.1	28132.0
	MC-LR	11367.7	8061.5	15659.5
YH2AX	Control	23427.6	25712.3	33187.1
	MC-LR	21680.6	24481.9	28347.0
PCNA	Control	27097.9	26141.9	30253.6
	MC-LR	24379.7	23827.4	29692.5
β-Actin	Control	32006.7	35627.2	40385.8
	MC-LR	33581.8	36033.9	39726.2

Note: the western blotting bands were quantified using ImageJ software (v1.53; NIH). The original measured grey value of each protein was shown in this table.

Table S36. Numeric data for Figure 8E: diameters (µm) of follicles treated with vehicle or 10 µM MC-LR in the presence or absence of 25 µg/mL 740-YP.

Day	Group	Mean	SD	n/group
0	Control	144.5	13.5	40
	MC-LR	145.0	11.7	39
	740-YP-25 µg/mL	141.7	14.2	43
	740-YP-25 µg/mL & MC-LR	144.4	11.2	42
2	Control	176.6	20.7	40
	MC-LR	173.3	24.3	39
	740-YP-25 µg/mL	169.7	22.5	43
	740-YP-25 µg/mL & MC-LR	175.2	23.4	42
4	Control	250.3	39.3	40
	MC-LR	247.5	38.8	39
	740-YP-25 µg/mL	236.5	37.8	43
	740-YP-25 µg/mL & MC-LR	249.5	38.3	42
6	Control	321.5	55.1	40
	MC-LR	302.0	30.8	39
	740-YP-25 µg/mL	300.6	50.6	43
	740-YP-25 µg/mL & MC-LR	301.5	43.4	42

Table S37. Numeric data for Figure 8F: survival rates (%) of follicles treated with vehicle or 10 µM MC-LR in the presence or absence of 25 µg/mL 740-YP.

Day	Group	Mean	SD	n/group
0	Control	100.0	0.0	40
	MC-LR	100.0	0.0	39
	740-YP-25 µg/mL	100.0	0.0	43
	740-YP-25 µg/mL & MC-LR	100.0	0.0	42
2	Control	100.0	0.0	40
	MC-LR	100.0	0.0	39
	740-YP-25 µg/mL	100.0	0.0	43

	740-YP-25 µg/mL & MC-LR	100.0	0.0	42
4	Control	97.6	4.1	40
	MC-LR	97.8	3.8	39
	740-YP-25 µg/mL	100.0	0.0	43
	740-YP-25 µg/mL & MC-LR	95.2	4.1	42
6	Control	94.9	8.9	40
	MC-LR	91.1	10.2	39
	740-YP-25 µg/mL	93.3	11.5	43
	740-YP-25 µg/mL & MC-LR	90.1	8.6	42

Table S38. Numeric data for Figure 8G: expression of follicle maturation-related genes in follicles.

Gene	Group	Mean (a.u.)	SD	n/group
Comp	Control	5.6	4.0	10
	740-YP	6.8	2.5	11
	MC-LR	2.4	1.3	12
	740-YP & MC-LR	3.5	1.4	12
Inha	Control	182.1	27.1	10
	740-YP	186.9	47.1	11
	MC-LR	92.2	27.2	12
	740-YP & MC-LR	139.8	46.6	12
Pappa	Control	0.8	0.4	10
	740-YP	0.8	0.5	11
	MC-LR	0.2	0.1	12
	740-YP & MC-LR	0.4	0.1	12
Lhcgr	Control	3.4	1.9	10
	740-YP	3.4	1.9	11
	MC-LR	0.5	0.2	12
	740-YP & MC-LR	1.3	0.7	12

Table S39. Numeric data for Figure 8H: percentages of ruptured follicles at 16 hours post-hCG.

Group	Mean	SD	n/group
Control	93.9	5.4	36
740-YP	93.7	5.7	34
MC-LR	16.9	3.9	35
740-YP & MC-LR	48.2	7.4	34

Table S40. Numeric data for Figure 9A: PP1 phosphatase activities in vehicle or 10 µM MC-LR treated human primary granulosa cells.

Time	Group	Mean	SD	n/group
20 min	Control	57.2	16.2	3
	MC-LR	17.8	11.8	3
60 min	Control	54.4	16.1	3
	MC-LR	12.5	8.0	3
100 min	Control	68.2	15.5	3
	MC-LR	18.0	12.7	3

Table S41. Numeric data for Figure 9B: PP2A phosphatase activities in vehicle or 10 µM MC-LR treated human primary granulosa cells.

Time	Group	Mean	SD	n/group
20 min	Control	48.3	28.7	3
	MC-LR	32.2	23.3	3
60 min	Control	60.8	28.2	3
	MC-LR	38.2	25.1	3
100 min	Control	77.4	28.7	3
	MC-LR	51.6	25.8	3

Table S42. Numeric data for Figure 9C: quantification of western blotting for phosphorylated FOXO1 in human primary granulosa cells.

Protein	Group	Replicate 1	Replicate 2	Replicate 3
pFOXO1	Control	2977.8	12053.2	12212.3
	MC-LR	1436.9	6267.9	7953.8
β-Actin	Control	12888.9	10829.7	14628.7
	MC-LR	11432.2	11524.8	14032.2

Note: the western blotting bands were quantified using ImageJ software (v1.53; NIH). The original measured grey value of each protein was shown in this table.

Table S43. Numeric data for Figure 9D: expression of follicle maturation-related genes in vehicle or 10 µM MC-LR treated human primary granulosa cells.

Gene	**Group**	**Mean (a.u.)**	**SD**	**n/group**
Lhcgr	Control	3.3	0.6	3
	MC-LR	3.3	0.3	3
Fshr	Control	3.9	0.6	3
	MC-LR	1.7	0.6	3
Pappa	Control	3.9	0.3	3
	MC-LR	9.6	1.6	3
Cyp19a1	Control	19.2	3.0	3
	MC-LR	9.9	4.1	3
Comp	Control	1.1	0.4	3
	MC-LR	1.2	0.4	3
Inha	Control	6.9	0.6	3
	MC-LR	5.7	0.5	3
Inhba	Control	686594.4	53522.3	3
	MC-LR	639211.8	202804.7	3
Inhbb	Control	1.8	0.4	3
	MC-LR	1.1	0.3	3

Table S44. Differential gene expression of oxidative stress genes induced by MC-LR using single-follicle RNA-seq.

Gene Symbol	**FDR**	**Fold change**	**Gene Symbol**	**FDR**	**Fold change**
Glutathione peroxidase			*Aox2*	0.629712	-1.25
Gpx1	2.69E-09	1.49	***Aox3***	**0.047462**	**4.93**
Gpx3	0.234339	1.4	*Aox4*	0.012576	-1.44
Gpx4	0.017853	-1.14	*Ephx1*	0.000194	-1.41
Gpx6	0.762612	1.95	*Ephx2*	0.222292	1.10
Gpx7	0.088809	-1.17	*Ephx3*	0.523748	1.80
Gpx8	0.799543	1.02	*Ephx4*	0.891817	-1.39
Gstp1	9.72E-11	1.42	*Bnip1*	0.025701	-1.26
Gstp2	0.07082	2.73	*Bnip2*	0.225673	-1.07
Peroxiredoxins			***Bnip3***	**5.84E-20**	**-2.34**
Prdx1	0.484905	1.04	*Mpv17*	0.868155	-1.02
Prdx2	0.608307	1.03	Oxidative stress-responsive genes		
Prdx3	0.011774	-1.16	*Txn1*	0.000353	1.24
Prdx4	0.000122	-1.34	*Txn2*	0.4955	1.05
Prdx5	0.335076	-1.08	*Krt1*	0.887693	-1.06
Prdx6	0.725722	-1.03	*Gclm*	0.000764	1.29
Peroxidases and antioxidants related genes			*Msra*	0.388201	1.10
Cybb	0.133662	1.79	*Cat*	0.055657	1.18
Mgst1	3.98E-08	1.31	***Ccl5***	**0.005531**	**16.75**
Mgst2	**0.049618**	**2.47**	***Dusp1***	**0.002726**	**2.37**

Mgst3	6.86E-12	-1.69	***Hmox1***	**2.23E-30**	**1.94**
Ptgs1	0.513398	1.59	***Txnrd1***	**3.27E-50**	**1.82**
Ptgs2	**3.22E-15**	**26.38**	*Txnrd2*	0.786981	1.04
Pxdn	0.236651	1.07	*Atox1*	0.000259	-1.32
Superoxide dismutases			***Hspa1a***	**8.59E-12**	**-2.48**
Sod1	0.454192	-1.05	*Dhcr24*	0.002132	-1.22
Sod2	3.25E-05	-1.26	***Nqo1***	**0.040914**	**1.56**
Sod3	0.057711	-1.47	*Nudt1*	0.465971	-1.13
Superoxide metabolism related genes			***Ttn***	**0.008176**	**2.30**
Ncf1	0.752746	1.28	*Gclc*	0.000177	1.29
Ncf2	**0.001975**	**2.24**	***Gss***	**8.83E-13**	**2.27**
Ncf4	**0.000201**	**10.59**	***Apoe***	**0.015151**	**1.60**
Nos1	0.360395	1.43	*Oxr1*	0.002446	-1.17
Nos2	0.427773	1.22	*Oxsr1*	0.000342	-1.19
Nos3	**1.47E-10**	**-3.21**	***Foxm1***	**7.55E-07**	**-1.63**
Nox1	0.358752	3.81	*Pdlim1*	0.008054	1.22
Nox4	0.895899	-1.05	*Pnkp*	0.546601	-1.05
Prex1	**0.001458**	**1.5**	*Prnp*	0.40789	-1.07
Prex2	**0.006074**	**-2.95**	*Fth1*	2.76E-20	1.30
Ucp2	0.593742	-1.08	*Rnf7*	1.28E-05	-1.29
Duox1	0.768706	-1.21	***Scara3***	**1.04E-11**	**3.84**
Duox2	**3.61E-08**	**2.79**	*Gsr*	0.00289	1.20
Mt1	0.003558	1.5	*Sirt2*	0.114793	-1.11
Mt2	**6.92E-10**	**2.64**	***Sqstm1***	**2.62E-18**	**1.52**
Mt3	0.411034	-1.46	***Srxn1***	**1.9E-106**	**6.53**
Ccs	0.013469	-1.27	*Stk25*	0.025903	-1.10
Alox5	**0.007233**	**7.01**	*Mbl2*	0.633729	1.24
Alox12	0.236651	1.84	Oxygen transporters		
ROS metabolism related genes			***Cygb***	**2.13E-10**	**3.17**
Aox1	**0.000763**	**1.62**			

Note: The differentially expressed genes (DEGs) were identified with absolute fold change > 1.5 and FDR adjusted *p*-value <0.05. The DEGs were highlighted in bold.

Table S45. Differential gene expression of autophagy related genes induced by MC-LR using single-follicle RNA-seq.

Gene Symbol	**FDR**	**Fold change**	**Gene Symbol**	**FDR**	**Fold change**
Initiation of autophagy			*Atg9b*	0.823436	1.11
Mtor	0.342134	-1.06	*Atg10*	0.400832	-1.20
Rptor	0.04807	1.11	*Atg12*	1.31E-09	-1.27
Ulk1	0.015702	1.15	*Atg14*	0.428791	-1.07
Atg13	0.381834	1.06	*Atg16l1*	0.046813	-1.16
Rb1cc1	0.001186	1.18	*Atg16l2*	0.04127	1.23
Atg101	0.795196	1.03	*Wipi1*	0.9456	1.01
Autophagosome formation			*Wipi2*	0.670857	-1.02

Atg2a	0.009368	1.13	*Pik3c3*	0.975624	-1.00
Atg2b	0.597959	1.03	*Pik3r4*	0.000121	-1.21
Atg3	0.227213	1.09	*Becn1*	0.126115	-1.08
Atg4a	0.110177	1.21	*Uvrag*	0.02529	-1.14
Atg4b	0.029092	1.13	*Ambra1*	0.843854	1.02
Atg4c	**0.000143**	**-1.54**	*Map1lc3a*	0.000251	1.20
Atg4d	0.264684	-1.10	*Map1lc3b*	0.011875	1.10
Atg5	0.792237	1.02	***Gabarapl1***	**3.99E-15**	**1.73**
Atg7	0.112621	1.17	*Gabarapl2*	0.281379	-1.11
Atg9a	0.449925	-1.06	*Gabarap*	0.014694	1.11

Note: The differentially expressed genes (DEGs) were identified with absolute fold change > 1.5 and FDR adjusted *p*-value <0.05. The DEGs were highlighted in bold.

Table S46. Differential gene expression of inflammation related genes induced by MC-LR using single-follicle RNA-seq.

Gene Symbol	FDR	Fold change	Gene Symbol	FDR	Fold change
Spp1	2.11E-34	719.78	*Mmp9*	4.63E-18	15.16
Mmp13	1.6E-10	162.18	*Cxcl1*	4.91E-08	14.81
Ccl6	7.67E-06	154.83	*Tnf*	2.93E-07	13.14
Serpinb2	4.19E-19	124.90	*Il11*	0.008537	12.74
Ccr1	1.97E-09	117.84	*Clcf1*	0.000598	12.38
Hp	2.52E-07	94.77	*Tnfaip6*	4.07E-55	11.83
Fmod	2.56E-09	86.07	*Ereg*	1.73E-10	11.41
Cemip	2.41E-14	76.18	*Il6ra*	1.71E-21	10.92
Ankrd1	8.14E-07	73.48	*Il6*	0.001003	9.70
Ifi202b	6.05E-30	72.22	*Mmp25*	0.008134	9.57
Ccl7	1.64E-07	69.88	*Cxcl5*	2.47E-06	8.40
Saa3	6.84E-16	64.61	*Mmp19*	8.63E-17	7.94
Gbp5	7.23E-06	58.35	*Cd44*	9.26E-45	7.13
Rgs1	1.07E-06	57.98	*Clec10a*	0.003004	7.10
Ptx3	1.91E-22	49.06	*Cxcl14*	1.14E-05	6.94
Mmp10	4.14E-05	48.08	*Il1rl2*	2.75E-33	5.70
Ccl9	7.65E-13	43.99	*Mmp24*	0.001205	5.46
Cxcl10	1.43E-11	39.11	*Lif*	5.69E-06	5.20
Tnr	9.63E-10	36.06	*Il7*	1.19E-24	5.12
Il33	6.09E-05	35.60	*Ccl3*	0.001668	5.01
Il7r	6.39E-07	35.42	*Ccn1*	8.54E-05	4.84
Tnc	6.27E-26	33.03	*Mmp28*	0.000217	4.20
Mmp12	7.99E-07	32.89	*Mmp2*	6.95E-09	3.82
Sema3e	3.85E-13	26.50	*Il16*	0.039833	3.54
Ptgs2	3.22E-15	26.38	*Il10ra*	0.000532	3.38
Pf4	0.037346	26.17	*Il13ra1*	1.31E-10	3.12
Ccr5	0.000111	25.14	*Mmp11*	1.01E-18	2.91
Fgf21	3.5E-07	24.57	*Mmp14*	4.42E-27	2.87

Il1rn	1.72E-09	24.36	*Il34*	0.019853	2.79
Ccl2	9.25E-07	24.18	*Tgfb2*	6.61E-07	2.79
Cxcl2	2.62E-08	21.61	*Il23a*	0.011036	2.71
Ptprn	1.31E-23	20.94	*Mt2*	6.92E-10	2.64
C3	6.83E-10	20.94	*Il2rb*	0.039101	2.56
Edn1	7.53E-26	19.55	*Il12rb1*	0.027228	2.55
Ngf	0.03522	19.46	*Il20rb*	0.014366	2.31
Ccl5	0.005531	16.75	*Cxcl12*	0.005769	2.06
Il1a	0.002639	16.14	*Il17ra*	5.33E-08	1.86
Ccl4	0.000695	15.58	*Mmp16*	0.000976	1.79

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