

**MASTER  
"MEDICINA DELLA RIPRODUZIONE"  
25/27-03-2009**

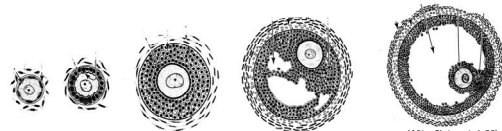
Valutazione (morfo)-funzionale degli ovociti nelle donne  
affette da alterazioni endocrino-metaboliche

*Giovanni Cotichio*



**Follicle Development**

Primordial follicle formation      Follicle recruitment and growth      Antral cavity formation      Growth and maturation of Graafian follicle



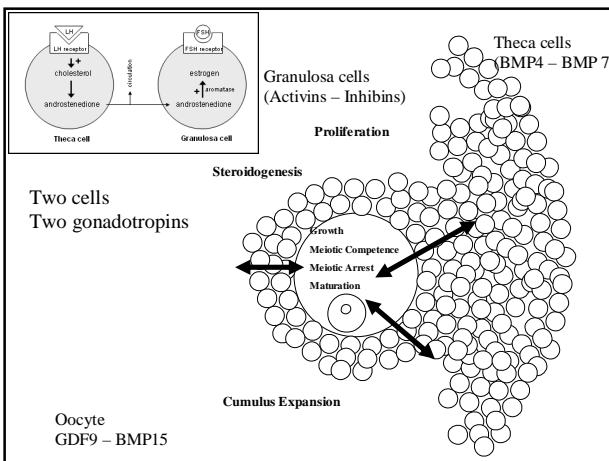
(Adapted from Picton et al. 198)

**Acquiring Competencies for:**  
MPF activation  
Genomic imprinting

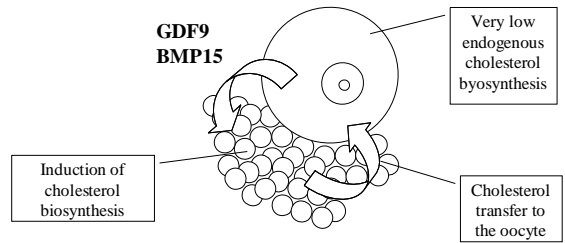
**Mature GV Oocyte**  
(meiotically competent)

**Oocyte Development**

Ca<sup>2+</sup> oscillations



**Example of metabolic loop  
between oocyte and  
cumulus cells**



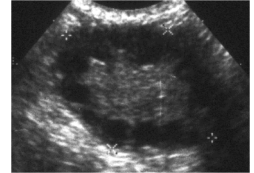
Su et al., 2008

## Topics

Oocyte / Follicle alterations in

- PCOS
- Diabetes (animal models)
- Obesity

## PCOS



- Ovarian hyperandrogenism
- LH hypersecretion
- Hyperinsulinemia (insulin resistance)
- Reduced fecundity

## PCOS

- Ovarian hyperandrogenism
  - Stimulation of primary, preantral, small antral follicle growth
  - Reduced TGF $\beta$ -related genes in oocytes
  - Inhibition of E2 synthesis as an effect of increased levels of 5 $\alpha$ -reduced androgens
  - Inhibition by P4 of the effects of E2 on oocyte cytoplasmic maturation (development of [Ca]<sup>2+</sup> oscillations)
  - Abnormal expression of genes involved in signal transduction, RNA processing, cell cycle regulation (many sharing promoter sequences containing putative transcription factor binding sites with sequence homology for androgen receptor)

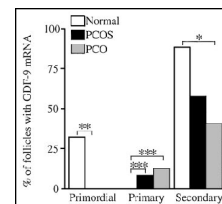
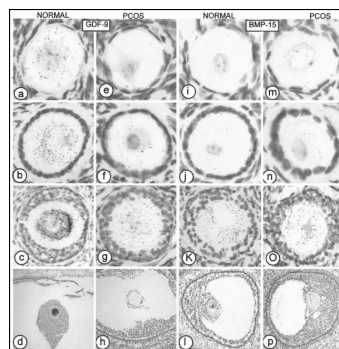
## PCOS

- Hyperinsulinemia
  - intrinsically impaired from abnormal post-receptor signal transduction
  - insulin resistance independent and additive with that of obesity
  - Increased androgen production, steroidogenic shift (E2 to P4 production), increased LH receptor expression, premature luteinization
  - Arrest of cell proliferation and follicle growth
  - Impairment of oocyte-somatic cell coordination

## PCOS

- TGFβ-related proteins
  - Reduced GDF9 expression in oocytes (effects on oocyte competence?)
  - Reduced AMH in primordial and primary follicles (increased primordial follicle recruitment?)

### In Oocytes of Women with Polycystic Ovary Syndrome Altered Expression of Growth Differentiation Factor-9



Teixeira Filho, et al., 2002.. J Clin Endocrinol Metab. 87,1337-44

### Oocyte quality and treatment outcome in intracytoplasmic sperm injection cycles of polycystic ovarian syndrome patients

	PCOS patients	Age-matched controls	P value
Treatment cycles	51	105	—
Stimulation days (mean ±SD)	15.98 ± 3.95	15.22 ± 2.51	NS
Number of ampoules (mean ±SD)	34.88 ± 10.88	42.37 ± 13.83	< 0.01
Oestradiol/HCG day (mean ±SD) pg/ml	3843.59 ± 2578.73	2367.40 ± 1580.41	< 0.01
Severe OHSS (%)	12	3	< 0.01
Oocytes (mean ±SD)	15.18 ± 9.59	10.07 ± 6.09	< 0.01
Metaphase II oocytes (mean ±SD)	7.98 ± 4.76	6.31 ± 4.09	0.05
Metaphase II oocytes/total oocytes (%)	53.48	62.66	NS
Fertilized oocytes/metaphase II oocytes (%)	62.66	56.42	NS
Germinal vesicle oocytes (mean ±SD)	0.69 ± 1.79	0.75 ± 1.32	< 0.01
Rate of germinal vesicle oocytes (%)	4.63	7.45	NS
2 pronuclei oocytes (mean ±SD)	5.00 ± 3.89	3.56 ± 3.01	< 0.01
Polyploid oocytes (mean ±SD)	0.61 ± 1.44	0.29 ± 0.76	NS
Embryo quality (mean ±SD)	27.35 ± 16.99	20.3 ± 13.75	< 0.05
Embryos/transfer (mean ±SD)	2.69 ± 0.84	2.17 ± 1.12	< 0.01
Fertilization failure (%)	7.84	14.29	NS
Transfer rate/oocyte retrieval (%)	94.12	85.71	NS
Pregnancy rate/transfer (%)	25	22	NS
Implantation rate (%)	10.21	11.84	NS
Multiple pregnancy rate (%)	16.67	30	NS
Clinical abortion rate (%)	41.67	21	< 0.05

Ludwig et al, Hum Reprod 1999, 14: 354–358

### Comparison of oocyte quality and intracytoplasmic sperm injection outcome in women with isolated polycystic ovaries or polycystic ovarian syndrome

Variable	PCOS	PCO-only	Control	P value
No. of cumulus-oocyte complexes	14.1 ± 5.7	16.1 ± 5.0	12.4 ± 4.9	<0.05
No. of metaphase II oocytes	12.1 ± 5.2	14.3 ± 4.5	10.5 ± 4.4	<0.05
Metaphase II oocytes/total oocytes (%)	86.4	88.9	86.0	NS
Normal oocytes	45.9	42.8	42.8	NS
Central granular cytoplasm (%)	16.2	17.5	23.1	NS
Evenly granular cytoplasm (%)	15.6	16.1	15.2	NS
Refractile body (%)	2.2	4.0	2.9	NS
Aggregation sER (%)	9.2	8.1	6.2	NS
Vacuolization (%)	2.6	2.2	3.7	NS
Fragmented polar body (%)	39.2	34.8	40.3	NS
Abnormal zona pellucida (%)	22.6	21.1	28.9	NS

Values are expressed as mean ± SD  
NS not significant, sER smooth endoplasmic reticulum

Sahu et al. Arch Gynecol Obstet (2008) 277:239–244

### Comparison of oocyte quality and intracytoplasmic sperm injection outcome in women with isolated polycystic ovaries or polycystic ovarian syndrome

**Table 4** The embryological data and pregnancy outcome of the PCOS, PCO-only, and control groups

Variable	PCOS	PCO-only	Control	P value
No. of 2 pronucleated oocytes	7.6 ± 4.3	8.9 ± 4.0	7.3 ± 3.8	NS
Two pronucleated/metaphase II oocytes	63.2 ± 22.9	60.9 ± 17.1	72.5 ± 40.7	NS
No. of top grade embryos on day 3	3.9 ± 3.5	4.3 ± 3.0	4.0 ± 2.8	NS
Day of embryo transfer	3.3 ± 0.9	3.2 ± 0.7	3.1 ± 0.8	NS
No. of embryos transferred	2.0 ± 0.2	2.0 ± 0.2	1.9 ± 0.09	NS
No of cycles with blastocyst transfer (%)	16.2	12	13.7	NS
No. of cycles with embryo freezing (%)	41.3	56.3	44.9	NS
No of moderate/severe OHSS (%)	2	0	1.9	NS
Clinical pregnancy/embryo transfer (%)	59	52	44	NS
Implantation rate (%)	33	30	27	NS
Multiple pregnancy rate (%)	13	19	21	NS
Miscarriage rate (%)	31	15.4	14	<0.05

Values are expressed as mean ± SD  
NS not significant

Sahu et al. Arch Gynecol Obstet (2008) 277:239–244

### Lack of association between polycystic ovary syndrome and embryonic aneuploidy

**TABLE 3** Results of PGD and cycle outcomes according to number of oocytes retrieved.

Variable	10-20 Oocytes retrieved			≥21 Oocytes retrieved		
	PCOS (n = 30)	Controls (n = 52)	P	PCOS (n = 43)	Controls (n = 29)	P
Female age (y)	36.2 ± 4.2	36.7 ± 4.1	NS	36.4 ± 4.1	35.8 ± 3.7	NS
No. euploid embryos	2.7 ± 1.7	2.3 ± 1.6	NS	3.7 ± 2.3	3.5 ± 2.5	NS
No. aneuploid embryos	1.5 ± 1.6	1.7 ± 1.9	NS	3.6 ± 3.8	2.4 ± 2.8	NS
No. complex abnormal embryos	2.1 ± 2.4	1.7 ± 1.6	NS	3.1 ± 2.8	2.3 ± 2.3	NS
Percentage of euploid embryos	54.7 ± 29.5	52.2 ± 31.4	NS	45.6 ± 27.0	51.5 ± 28.2	NS
Implantation percentage rate per ET	25.4 ± 26.4	25.3 ± 30.2	NS	24.2 ± 33.5	33.7 ± 31.8	NS
Clinical pregnancies per ET	17 (58.6)	24 (49.9)	NS	18 (42.9)	20 (69.0)	<.05
Miscarriage	4 (23.5)	3 (12.5)	NS	1 (5.6)	1 (5.0)	NS
Ongoing pregnancies per ET	13 (44.8)	21 (42.9)	NS	17 (40.5)	19 (65.5)	<.05

Note: Data are mean ± SD or are n (%). NS = not statistically significant; ET = embryo transfer.

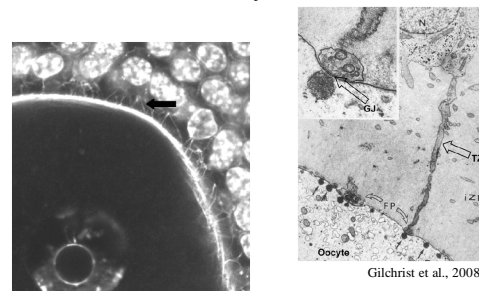
Weghofer et al. Fertility and Sterility Vol. 88, No. 4, Oct 2007

### A meta-analysis of outcomes of conventional IVF in women with polycystic ovary syndrome

- Significantly reduced chance of oocyte retrieval per started cycle in PCOS patients
- Significantly more oocytes per retrieval obtained in PCOS patients compared with controls
- The number of oocytes fertilized did not differ significantly between PCOS patients and controls
- No significant difference observed in the clinical pregnancy or live birth rates per started cycle

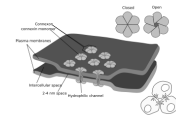
Heijnen et al Hum Reprod Update, Vol.12, No.1 pp. 13–21, 2006

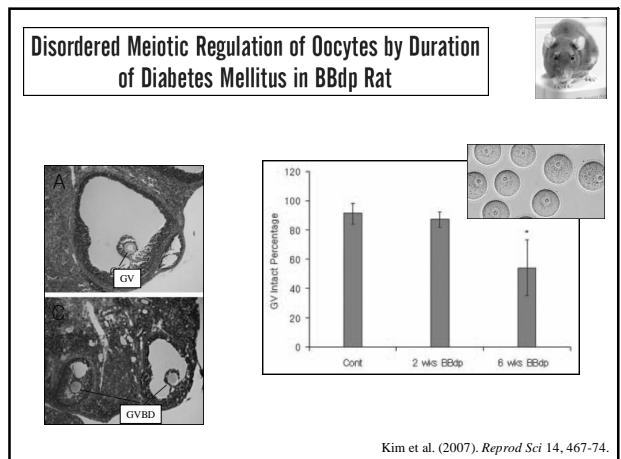
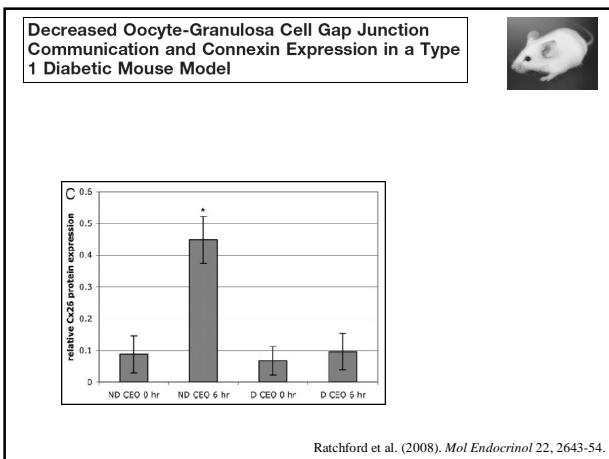
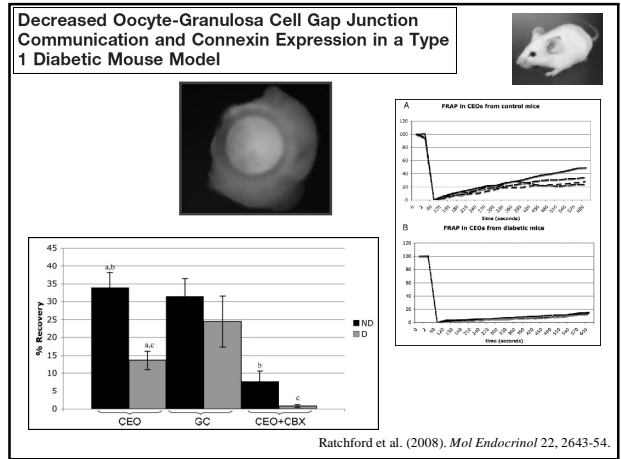
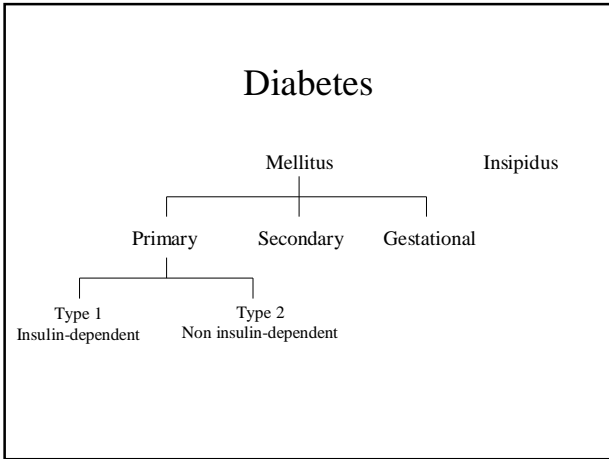
### Cellular bases of oocyte-cumulus cells association

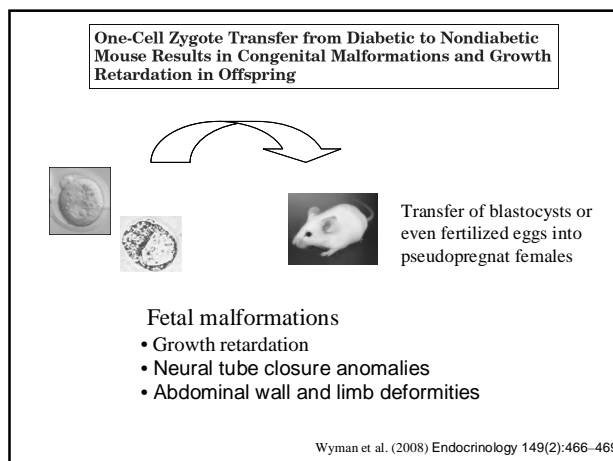
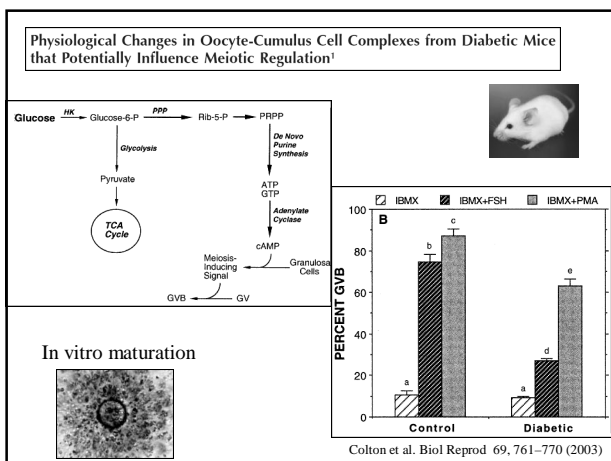
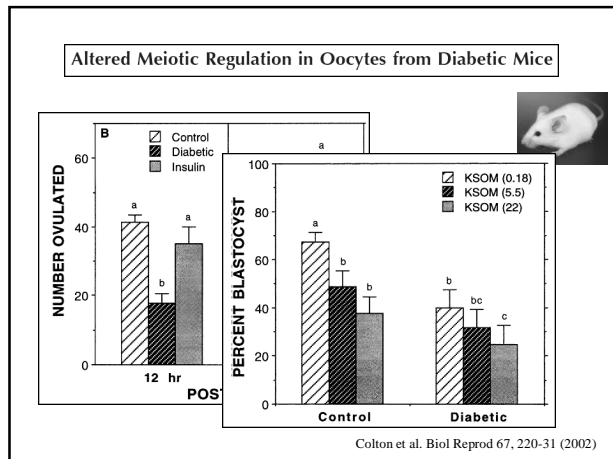
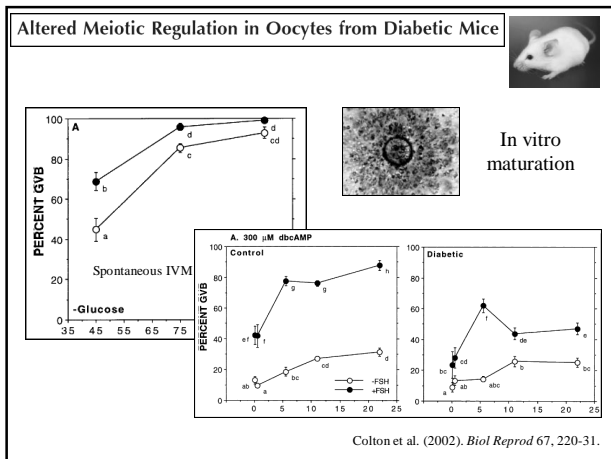


Courtesy of D. Albertini

Transzonal processes are regulated by FSH during follicle development and by LH at ovulation







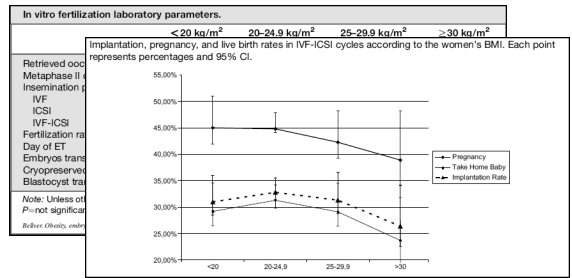
# Obesity

$$\text{BMI} = \text{Kg/m}^2$$

Adults	Women	Men
anorexia	< 17.5	
underweight	<19.1	<20.7
in normal range	19.1-25.8	20.7-26.4
marginally overweight	25.8-27.3	26.4-27.8
overweight	27.3-32.3	27.8-31.1
very overweight or obese	>32.3	>31.1
severely obese	35 - 40	
morbidly obese	40 - 50	
super obese	50 - 60	

## ARTICLE IN PRESS

### Female obesity impairs in vitro fertilization outcome without affecting embryo quality



### Obesity does not impact implantation rates or pregnancy outcome in women attempting conception through oocyte donation

Pregnancy outcome and BMI.	≤20	21-25	26-29	≥30	P value
N	101	284	74	77	
Mean age (y)	40.2 ± 5.9	41.2 ± 1.6	40.4 ± 8.4	41.5 ± 6.9	.17
Implantation rate (%)	55.2	53.8	54.7	54.8	.90
Pregnancy rate (%)	71.3	73.2	74.3	74	.91
Loss rate (%)	25.0	24.5	10.9	29.8	.096
Mean no. of embryos	2.2 ± 0.002	2.3 ± 0.005	2.4 ± 0.003	2.3 ± 0.002	.43
% Blastocyst transferred	55.4 <sup>a</sup>	60.6 <sup>a</sup>	51.4 <sup>a</sup>	59.7 <sup>a</sup>	
% Day 3 transferred	44.6 <sup>b</sup>	39.4 <sup>b</sup>	48.6 <sup>b</sup>	40.3 <sup>b</sup>	

<sup>a</sup> vs. <sup>b</sup>P=.46.

Styne-Gross et al. Fertil Steril 83:1629-34, June 2005

## Summary

- Various metabolic/endocrine conditions can affect oocyte and follicle physiology from the primordial to the preovulatory stage
- The oocyte-somatic cells cross-talk is often disrupted
- Impairment of oocyte function may be not apparent morphologically but involves more profound and partly unknown functional aspects
- Fetal malformations and abnormalities can occur independent of metabolic conditions during pregnancy