IMPACT OF OVARIAN STIMULATION ON THE LUTEAL ENDOMETRIUM

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DISCLOSURE

Nothing to disclose

Learning Objectives

At the conclusion of this presentation, the participant should be able to:

- **Discuss changes in hormone secretion in stimulated cycles**
- **Describe the impact of high levels of ovarian steroids on endometrial maturation**
- **Summarize morphological and functional genomic changes in the endometrium**
- **Evaluate the impact of changes in endometrial receptivity on clinical outcome**

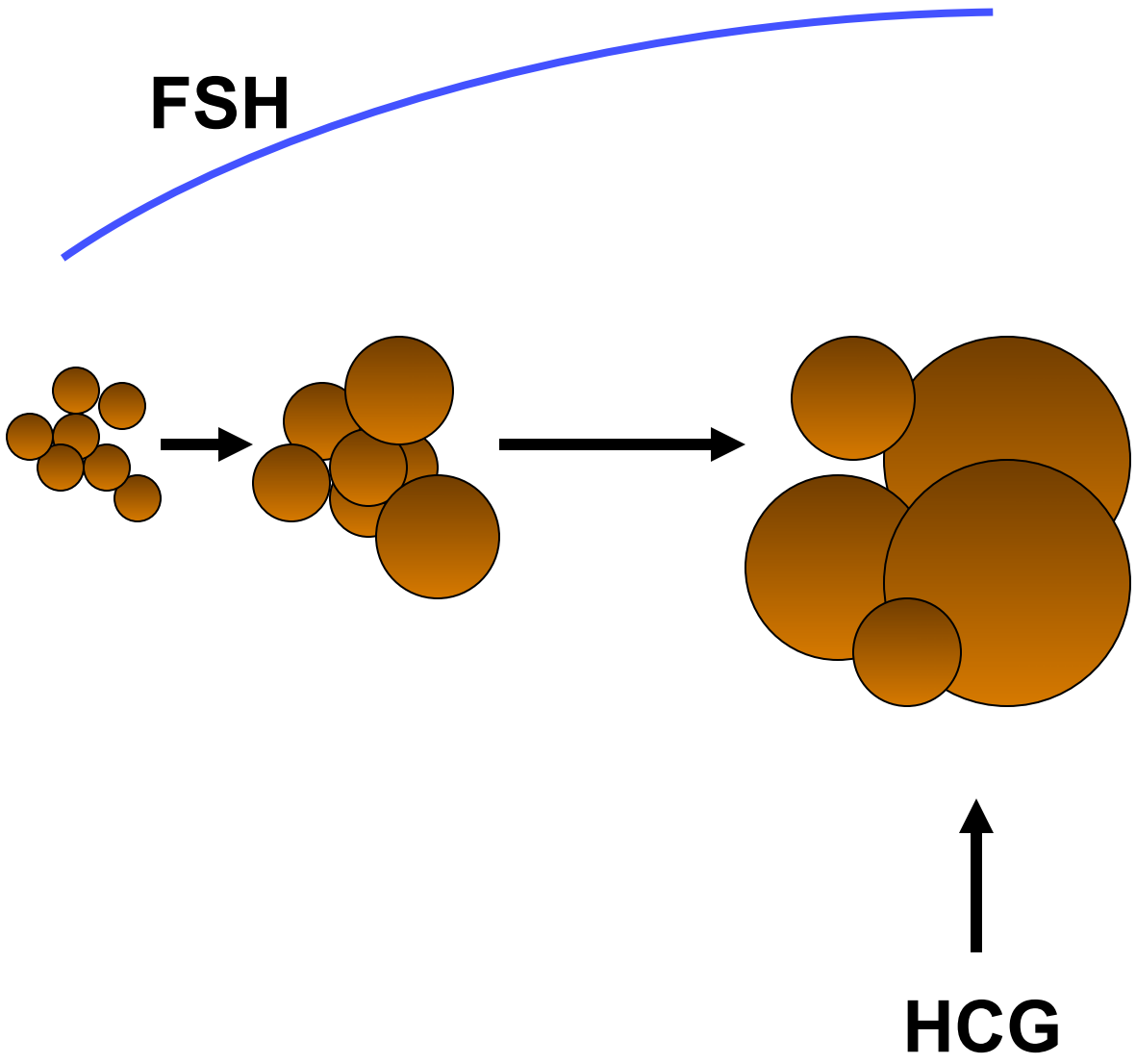
OVARIAN STIMULATION (Luteal phase)

- Endocrine changes
- Endometrial morphology changes
- Gene transcripts
- Clinical outcome
- Luteal support

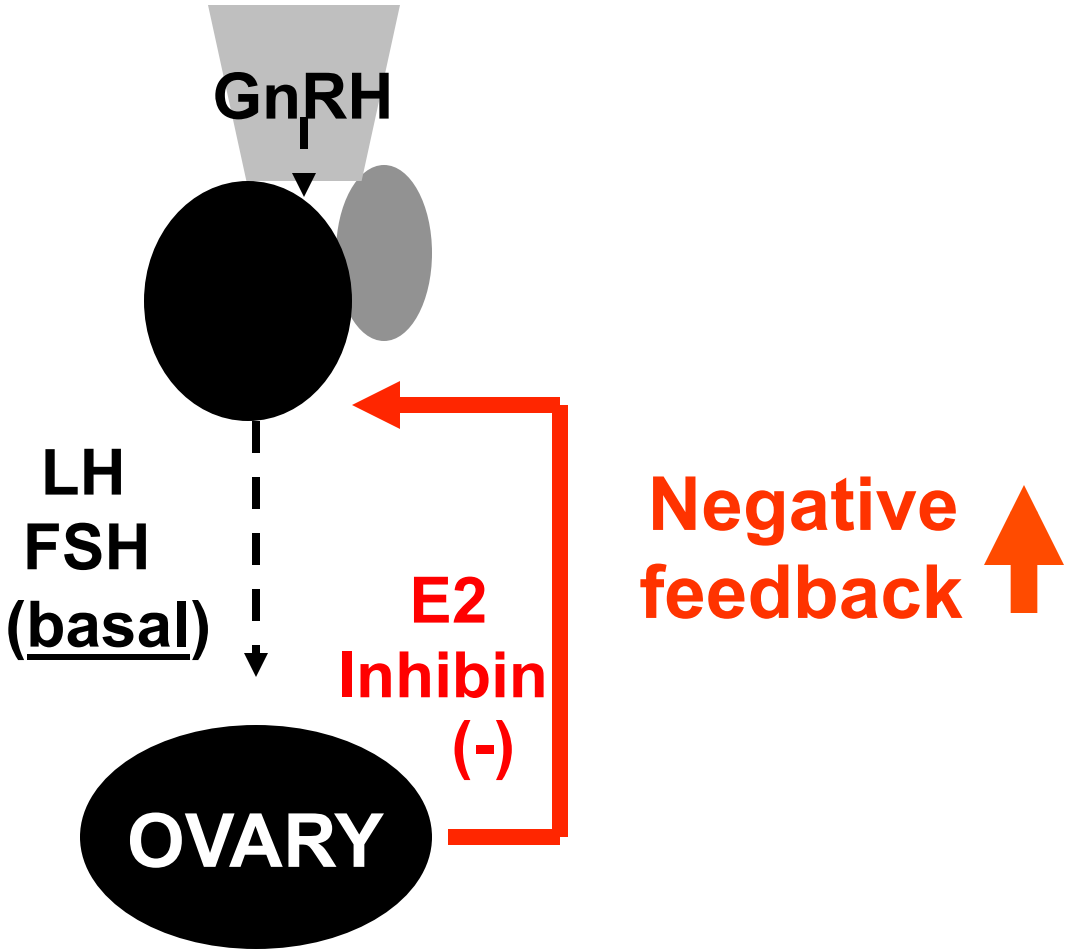
OVARIAN STIMULATION (Luteal phase)

- **Endocrine changes**
- Endometrial morphology changes
- Gene trnscripts
- Clinical outcome
- Luteal support

MULTIPLE FOLLICLES



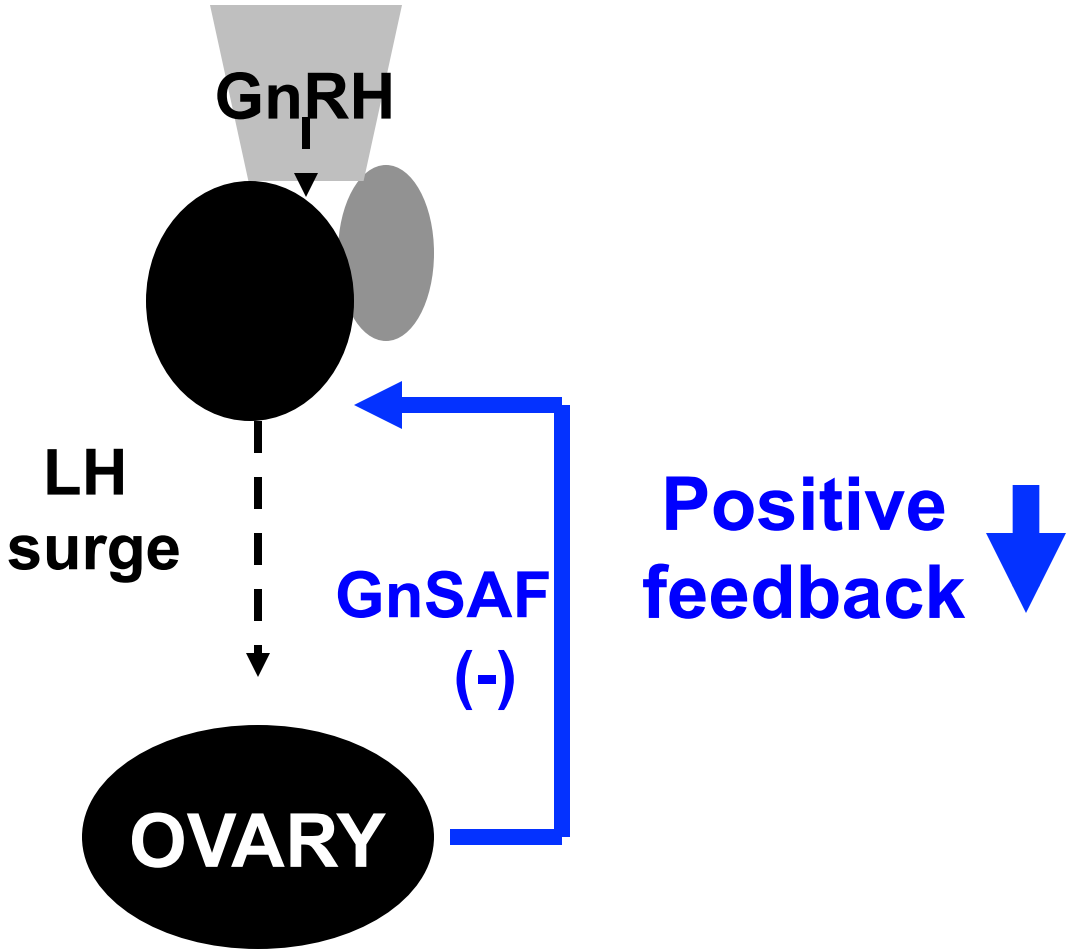
POTENTIATION OF THE NEGATIVE FEEDBACK



Multiple follicular development

Messinis, 2006
Hum. Reprod. Update 12, 557-571

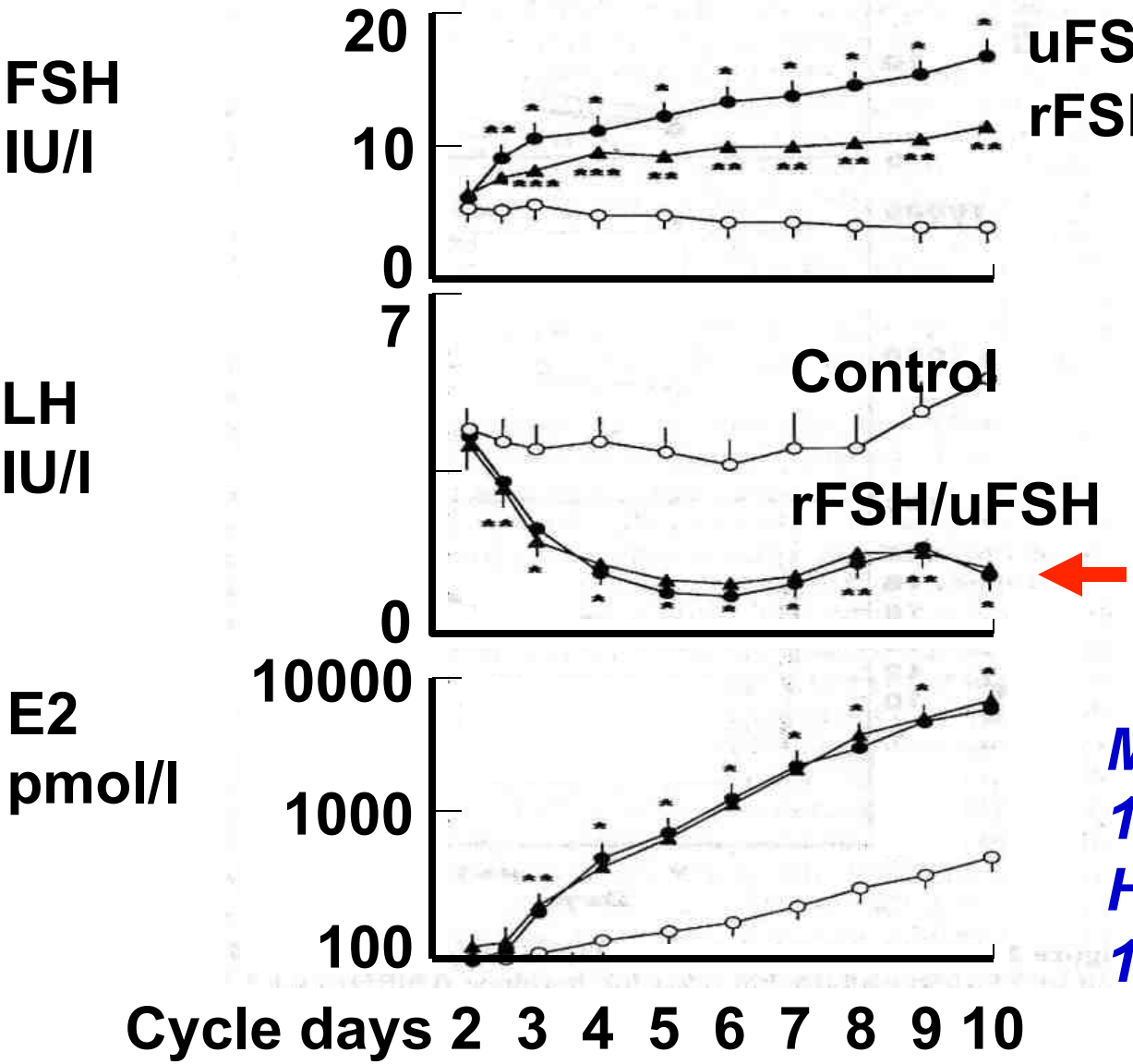
ATTENUATION OF THE POSITIVE FEEDBACK



Multiple follicular development

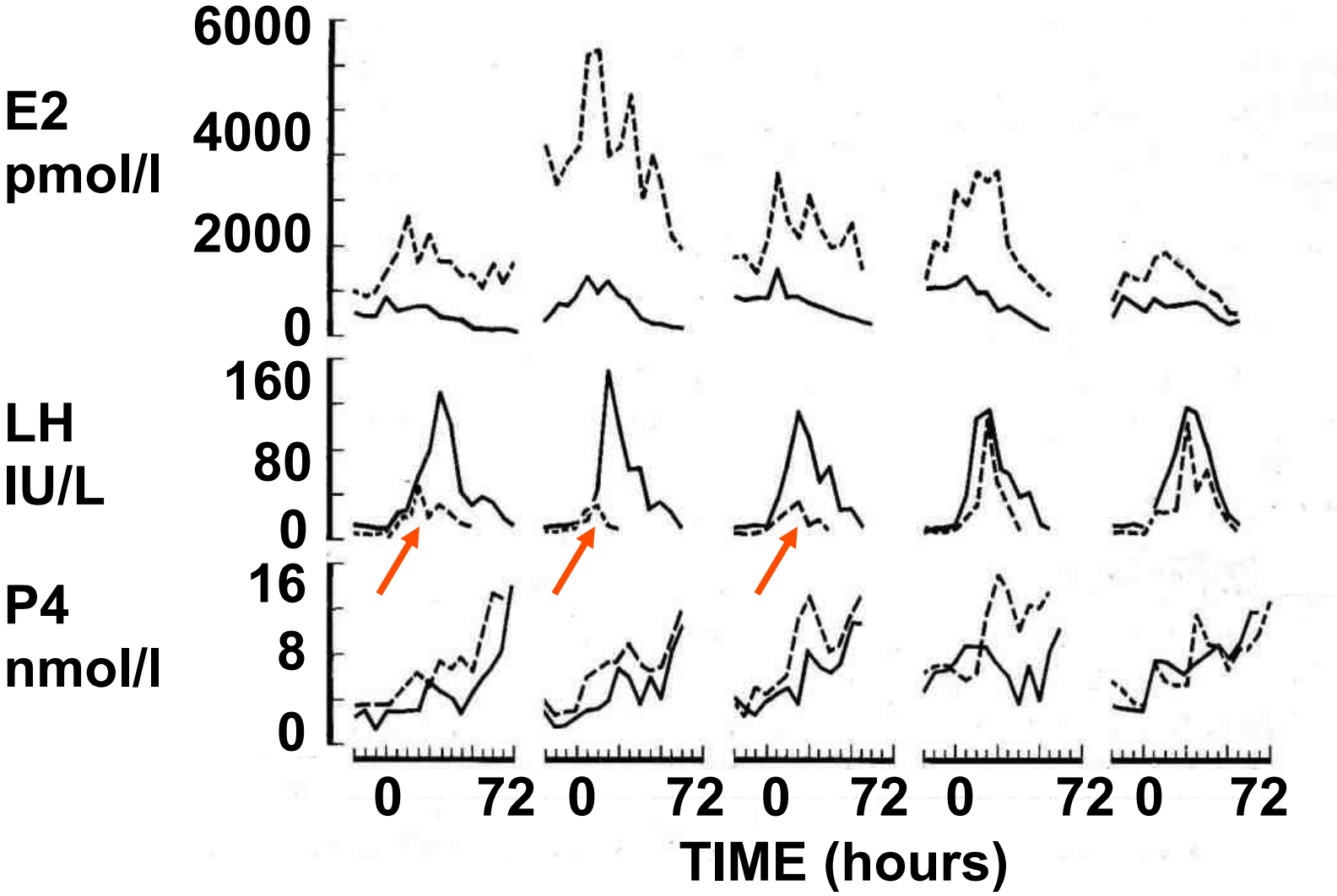
Messinis, 2006
Hum. Reprod. Update 12, 557-571

LH IS SUPPRESSED



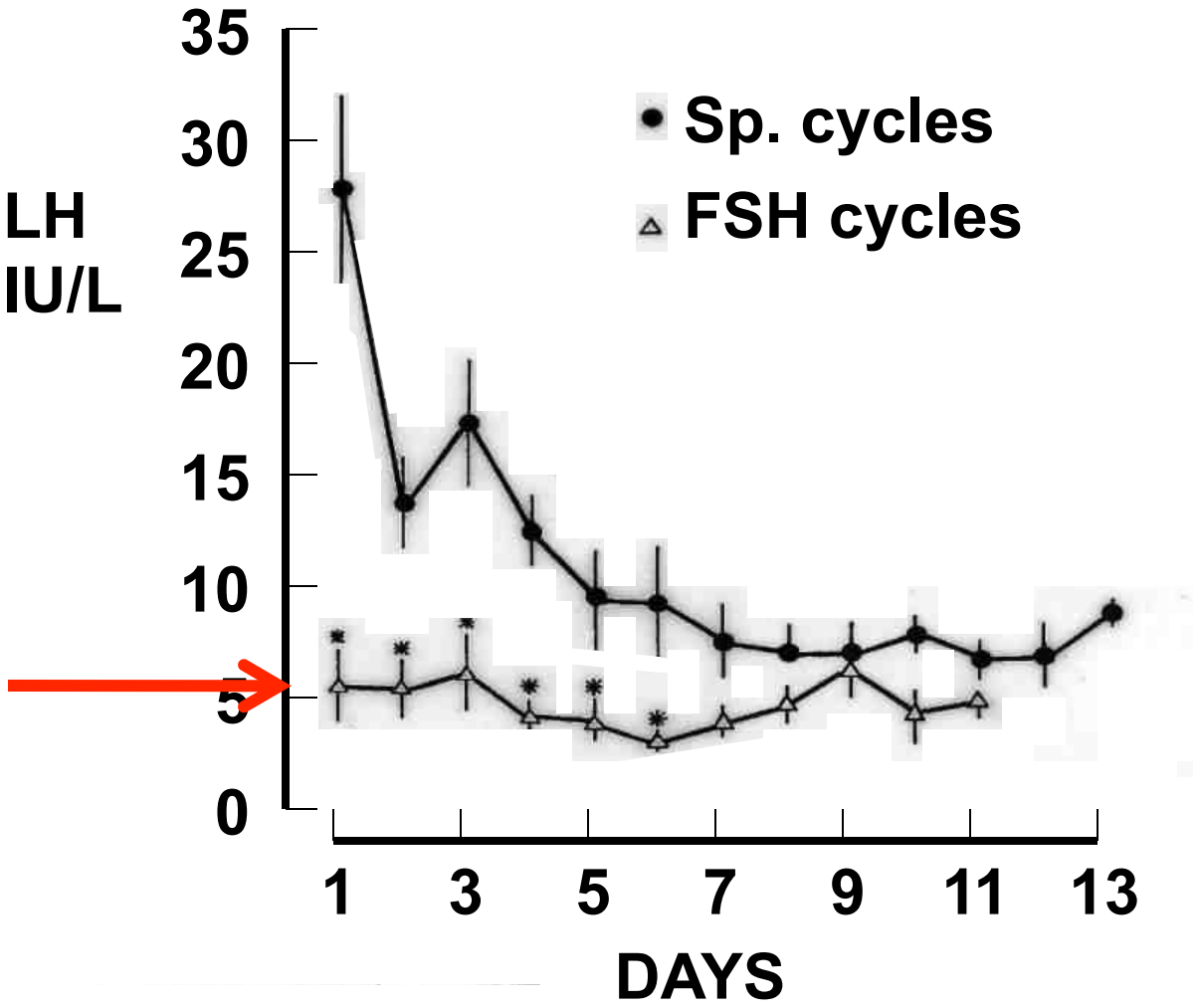
Messinis et al., 1998
Hum. Reprod. 13, 2415-20

SUPEROVULATION INDUCTION WITH PULSATILE FSH (Attenuated LH surges)



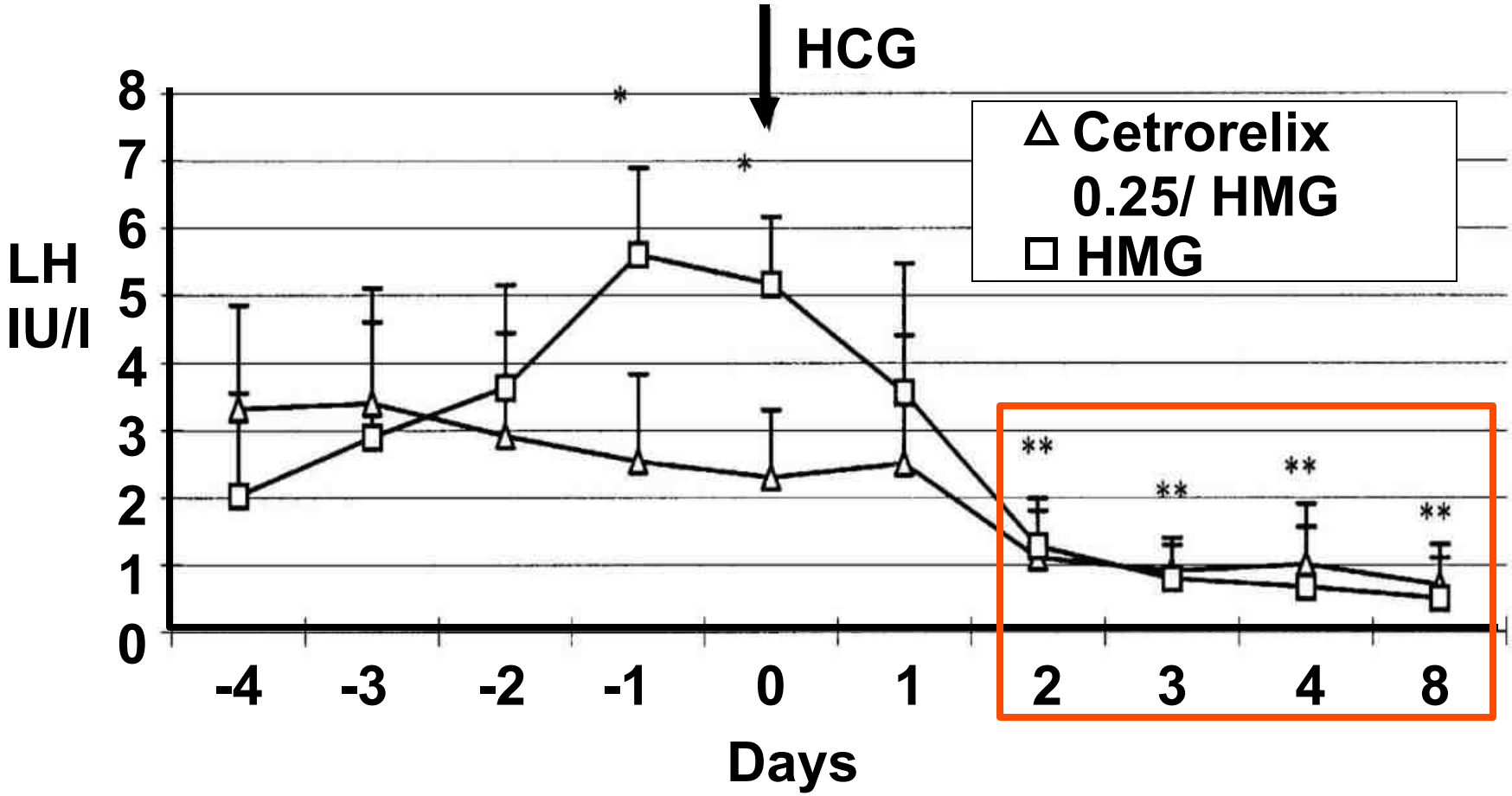
Messinis & Templeton, 1986; Clin. Endocrinol. 25, 633-640

LUTEAL PHASE LH VALUES FOLLOWING AN ATTENUATED LH SURGE



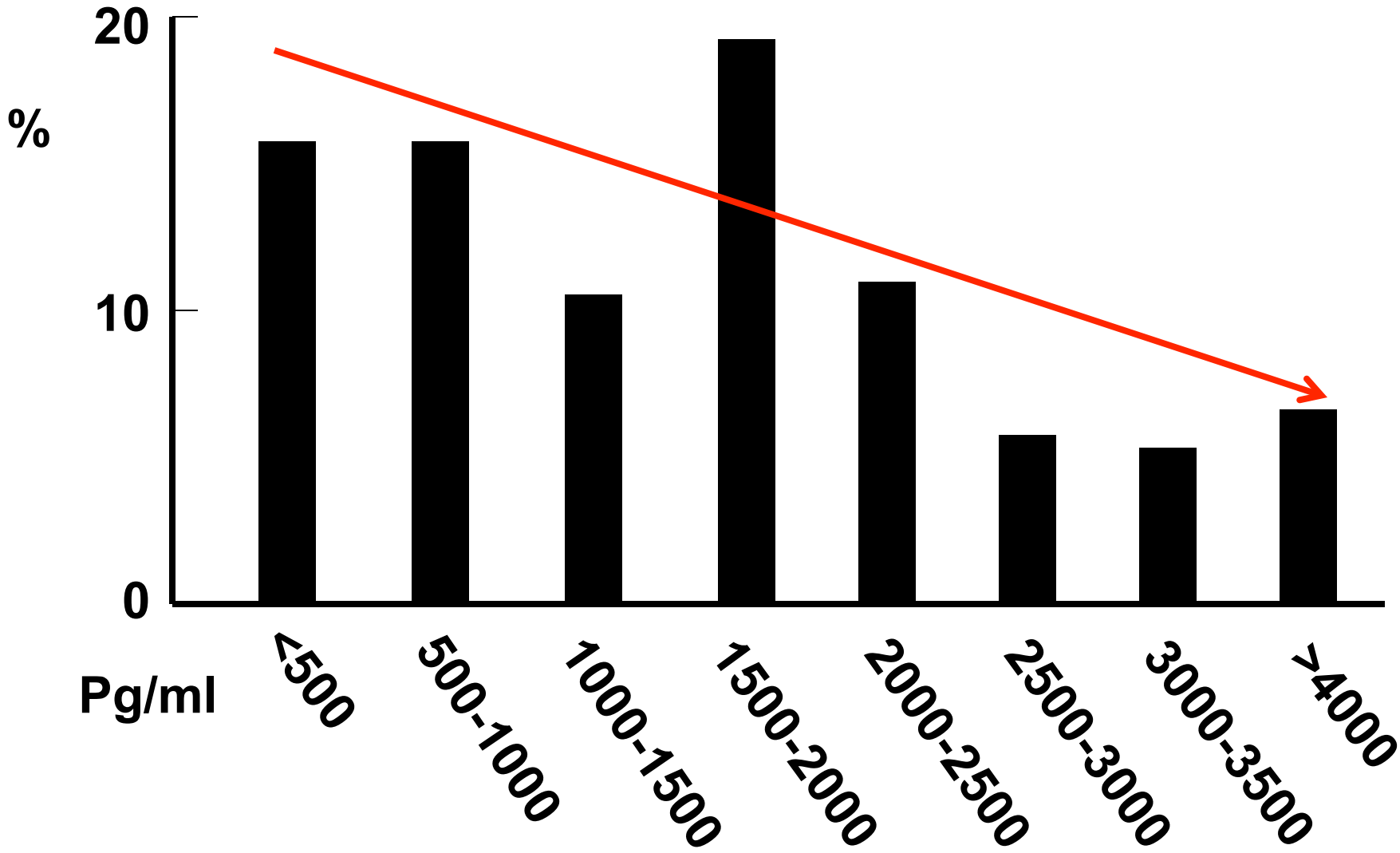
Messinis & Templeton, 1987
J. Reprod. Fert. 79, 549-554

SERUM LH VALUES (Luteal)



Tavaniotou et al., 2001
Hum. Reprod. 16, 663-667

IMPLANTATION RATE AND ESTRADIOL LEVELS ON DAY OF HCG



Simon et al., 1995; Hum. Reprod. 10, 2432-7

SERUM PROGESTERONE

CYCLES	DAY OF HCG INJECTION	DAY OF FOLLICLE ASPIRATION
Unstimulated	0.5±0.2	0.5±0.1
Hyperstimulated	1.1±0.6*	8.5±2.2**

* P<0.01, ** P<0.001

Kolb & Paulson, 1997
Am. J. Obstet. Gynecol. 176, 1262-7

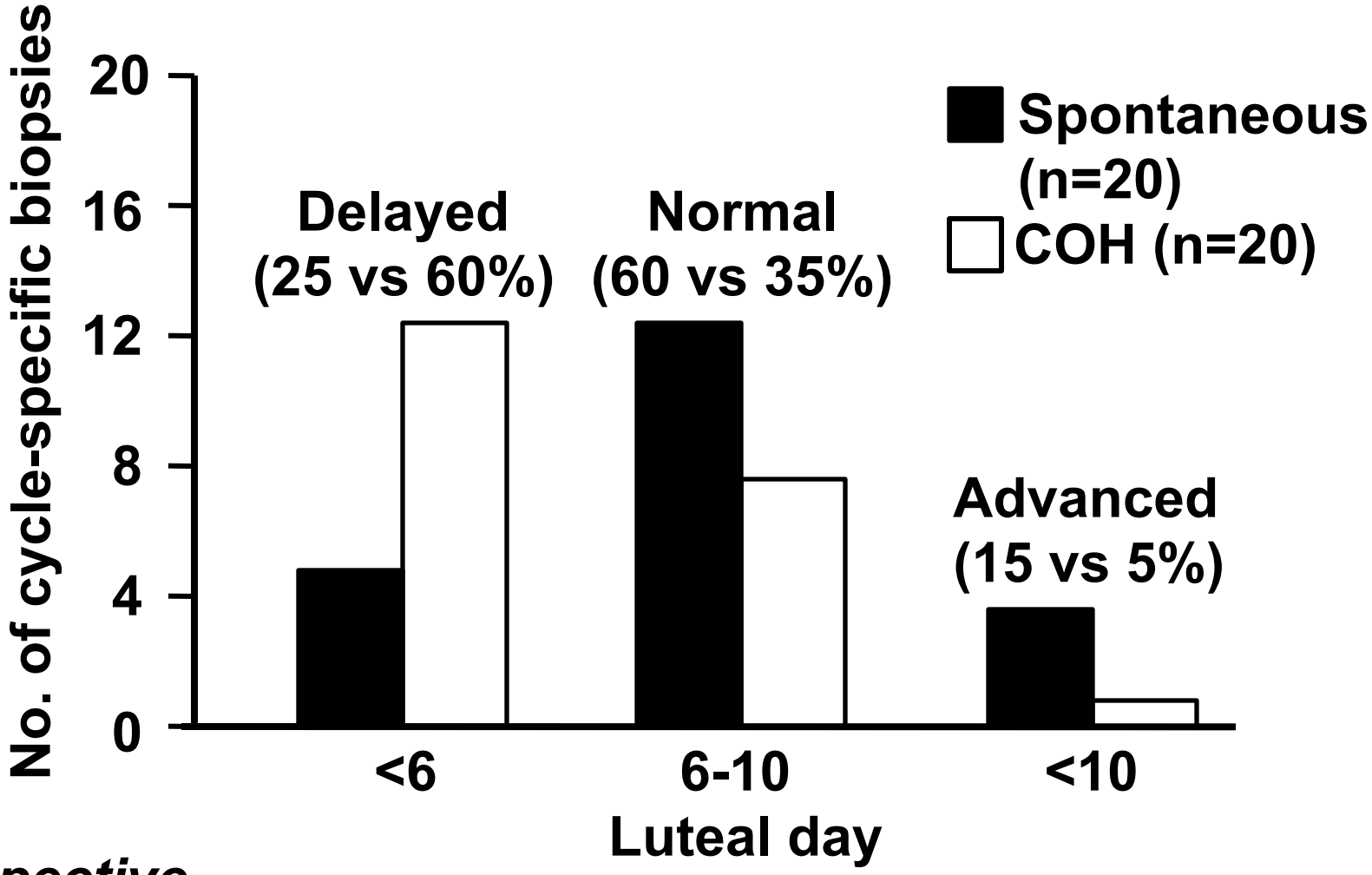
OVARIAN STIMULATION (Luteal phase)

- Endocrine changes
- **Endometrial morphology changes**
- Gene transcripts
- Clinical outcome
- Luteal support

ENDOMETRIAL DATING

- **Endometrial histology is advanced**
  - **at oocyte pick-up in GnRH agonist or antagonist cycles *(Kolibianakis et al., 2003; Saadat et al., 2004)*.**
  - **at mid-luteal phase in 60% of patients *(Kolibianakis et al., 2003)*, especially with the agonists *(Simon et al., 2005)*.**

HISTOLOGIC GLANDULAR MATURATION



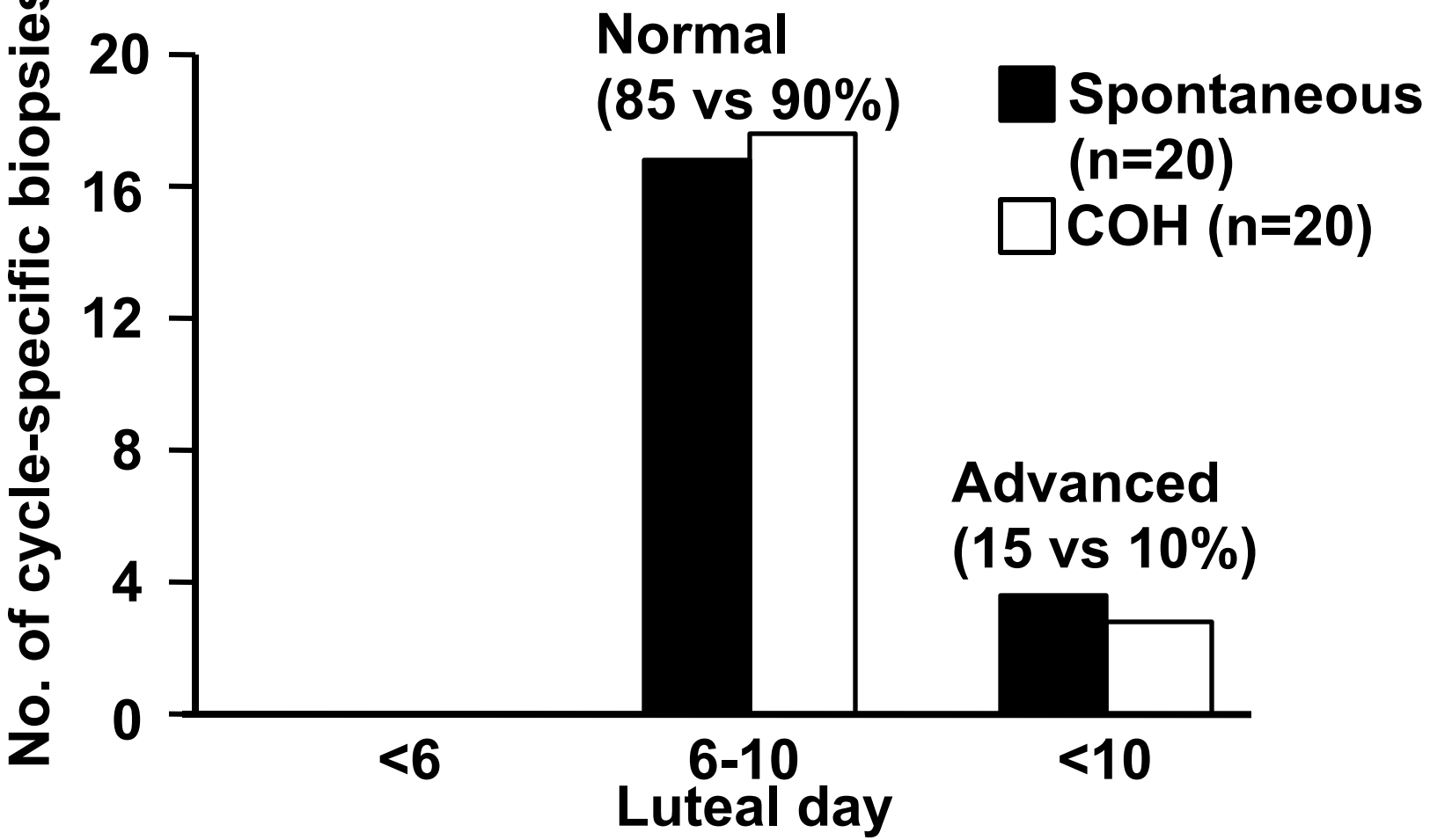
Prospective

LH/HCG+8 (n=19 oocyte donors) NC

Data not affected by P4 administration

Meyer et al., 1999
Fertil. Steril. 71, 109-14

HISTOLOGIC STROMAL MATURATION



Glandular-stromal dyssynchrony

In 80% of COH

In 30% of spontaneous

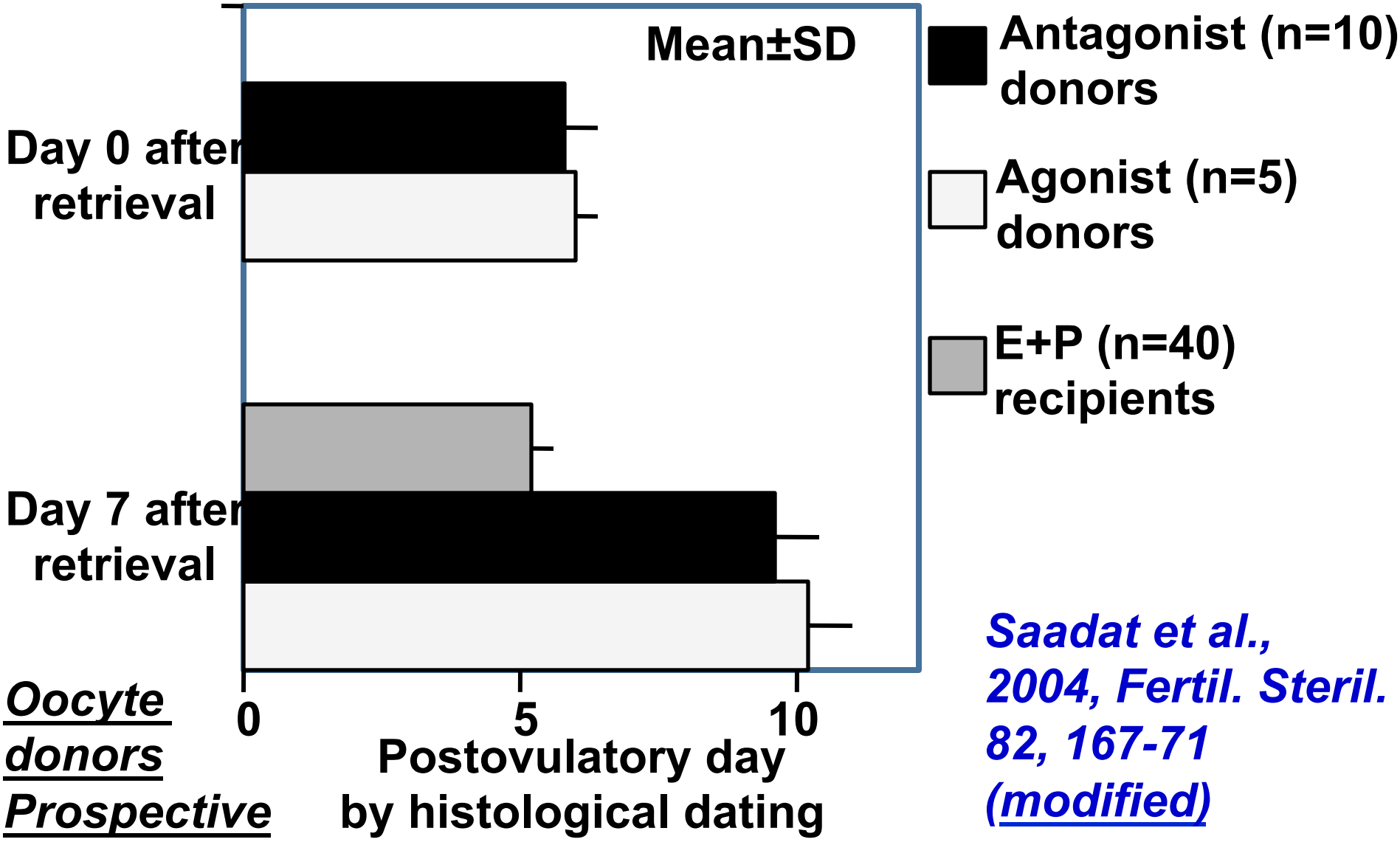
Meyer et al., 1999
Fertil. Steril. 71, 109-14

THE ENDOMETRIUM IN OVARIAN STIMULATION FOR IVF

- In peri- and post-ovulatory period ⟶ advanced endometrium
- In the early to mid-luteal phase ⟶ 'normal' aspect of the endometrium
- In the mid- to late luteal phase ⟶ glandular-stromal dyssynchrony

Bourgain and Devroey, 2003
Hum. Reprod. Update, 9, 515-22

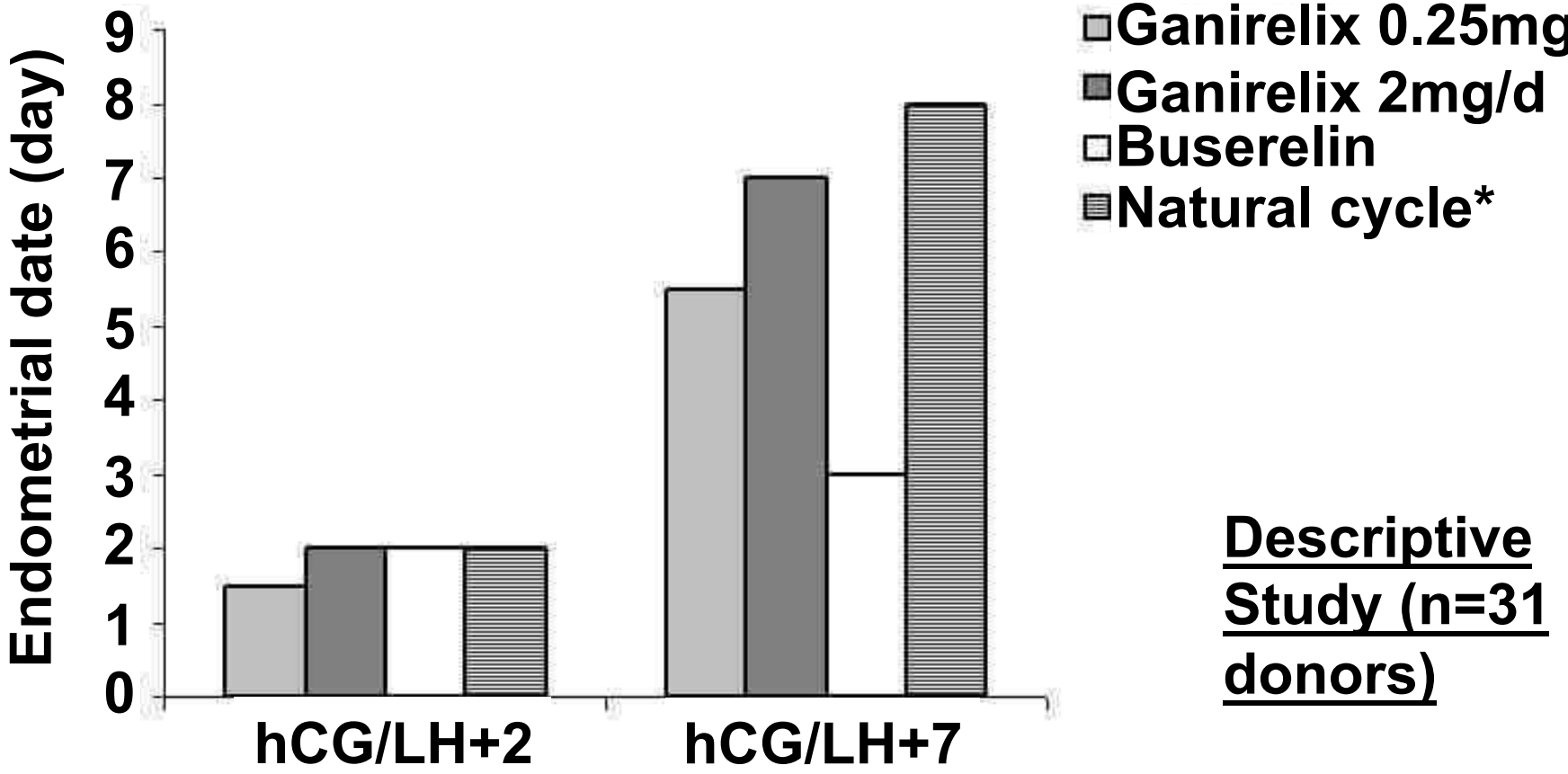
ACCELERATED ENDOMETRIAL MATURATION



Oocyte donors Prospective

Saadat et al., 2004, Fertil. Steril. 82, 167-71 (modified)

ARRESTED ENDOMETRIUM WITH THE USE OF BUSERELIN

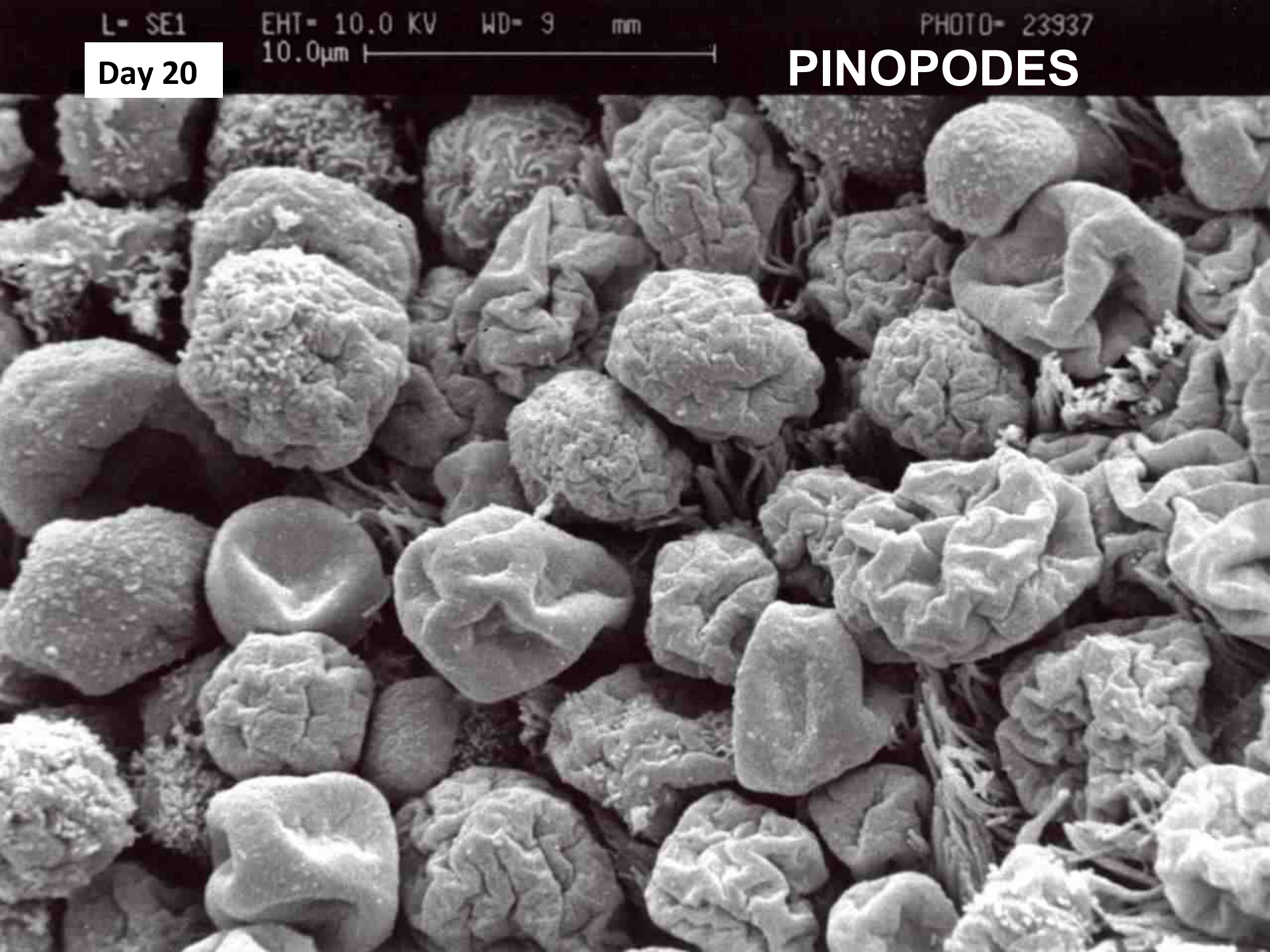


Descriptive Study (n=31 donors)

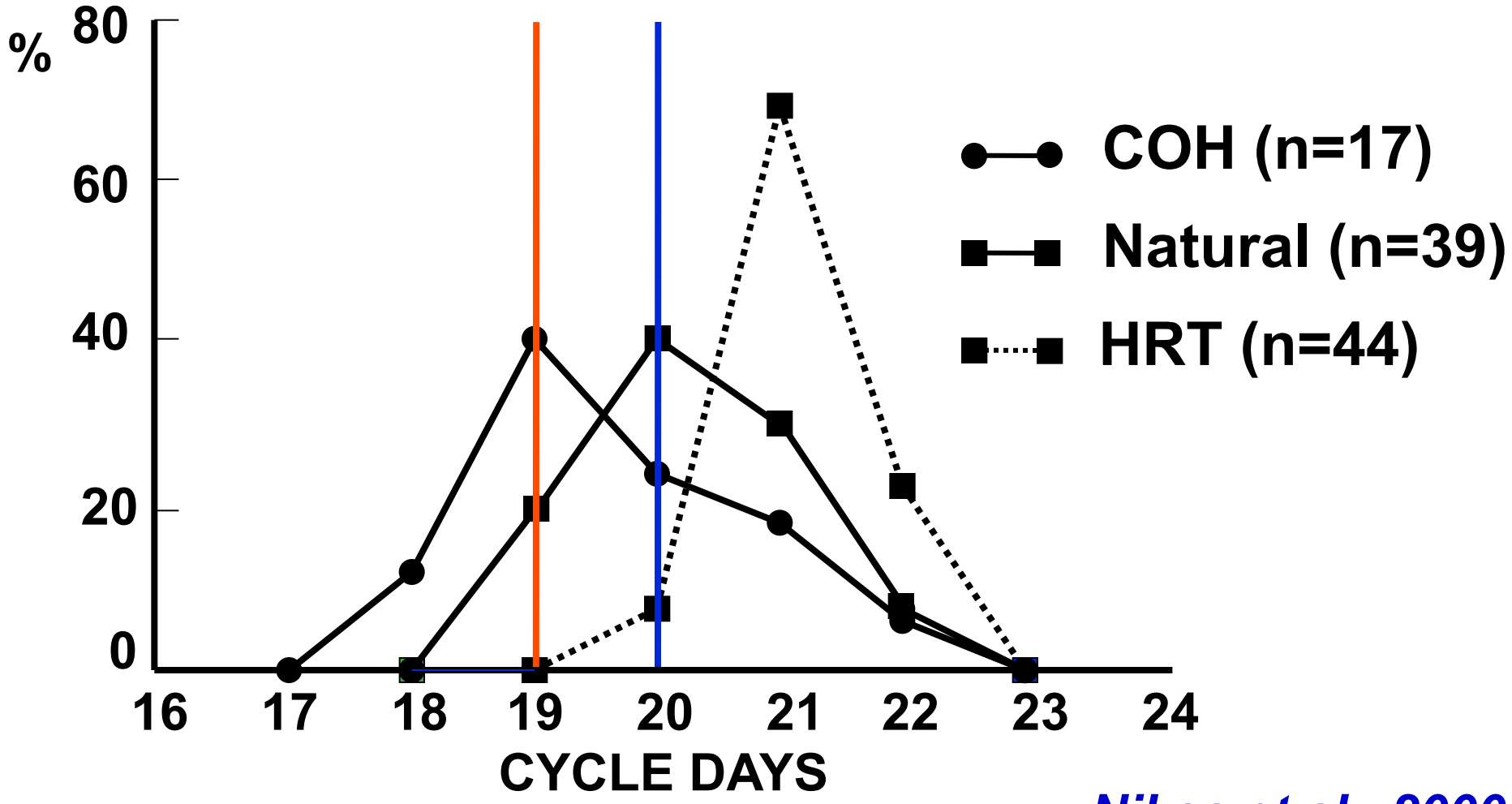
*** LH peak**

Noyes criteria

Simón et al. 2005
Hum. Reprod. 20, 3318-27


L= SE1
EHT= 10.0 KV
WD= 9 mm
PHOTO= 23937
10.0µm
Day 20
PINOPODES

DIFFERENTIAL EXPRESSION OF PINOPODES



Nikas et al., 2000

COH CYCLES

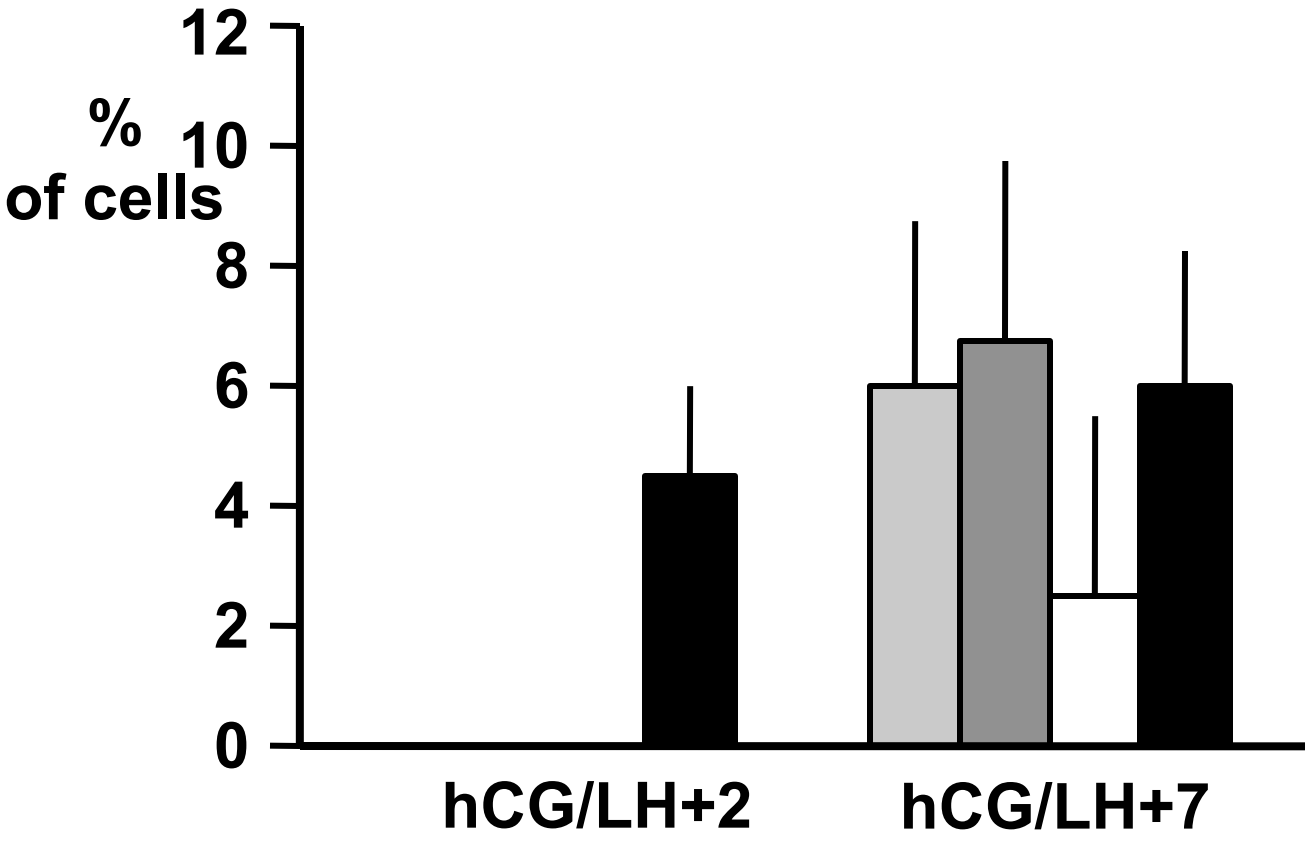
(Pinopodes and progesterone receptors)

- **Fully developed pinopodes**
  by *day 19* vs *day 21* in natural cycles

- **The decline in PR staining started**
  on *day 18* vs *day 20* in natural cycle

(Develioglou et al., 1999; Fertil. Steril. 71, 1040-7
Stavreus-evers et al., 2001; Fertil. Steril. 76, 782-91)

REDUCED EXPRESSION OF PINOPODES BY BUSERELIN



Descriptive Study (n=31 donors)

* LH peak

Simón et al. 2005
Hum. Reprod. 20, 3318-27

RU486 POSTPONED THE DEVELOPMENT OF PINOPODES

- **In rats:**
  - **On Pd6-8 vs day 5 in control**

    (Sarantis et al., 1988, Hum. Reprod. 3, 251-5)

- **In mice:**
  - **on Pd1, pinopodes were reduced**

    (Huang et al., 2005, Acta Pharmacol. Sin. 26, 212-9)

RU486 AND PINOPODES (Women)

	PINOPODES PRESENT (HCG+7)
• DONORS*	
- controls	1 of 4[a]
- study group (RU486)**	4 of 4
• RECIPIENTS	4 of 4

* COH with HMG/GnRH-a/HCG (endometrium was advanced)

[a]P<0.05

** 2.5 mg the day of OR and 2.5 mg the following day

Prosp. CT

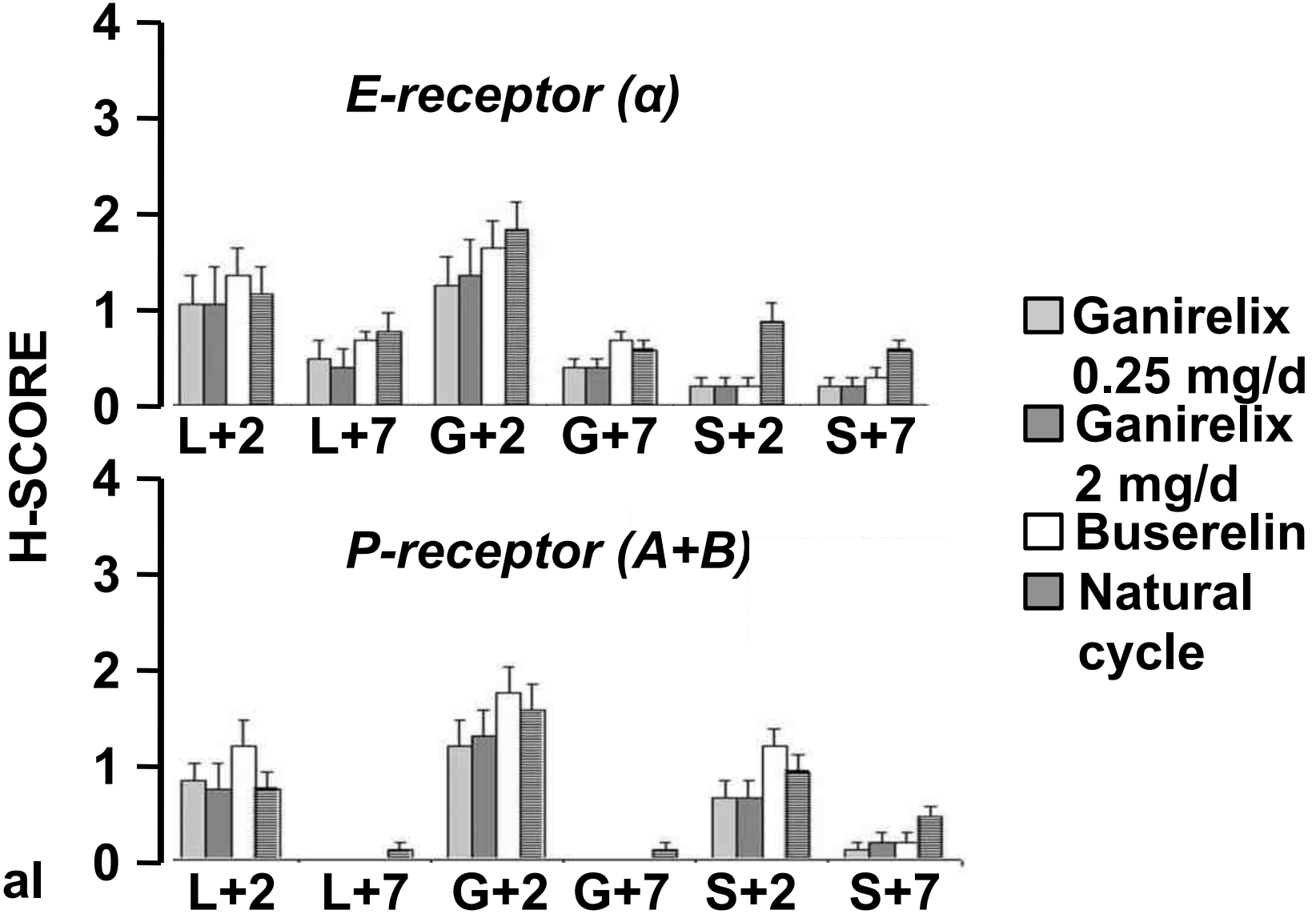
Paulson et al., 1997; Fertil. Steril. 67, 321-5

POTENTIAL OF RU486 IN IVF CYCLES

- **To block the endogenous LH surge**
  (Messinis et al., 1997; Clin. Endocrinol. 46, 309-14
  Escudero et al., 2005; JCEM 90, 2081-8)

- **To postpone the appearance of pinopodes**
  (Paulson et al., 1997; Fertil. Steril. 67, 321-5)

RECEPTORS* IN THE ENDOMETRIUM

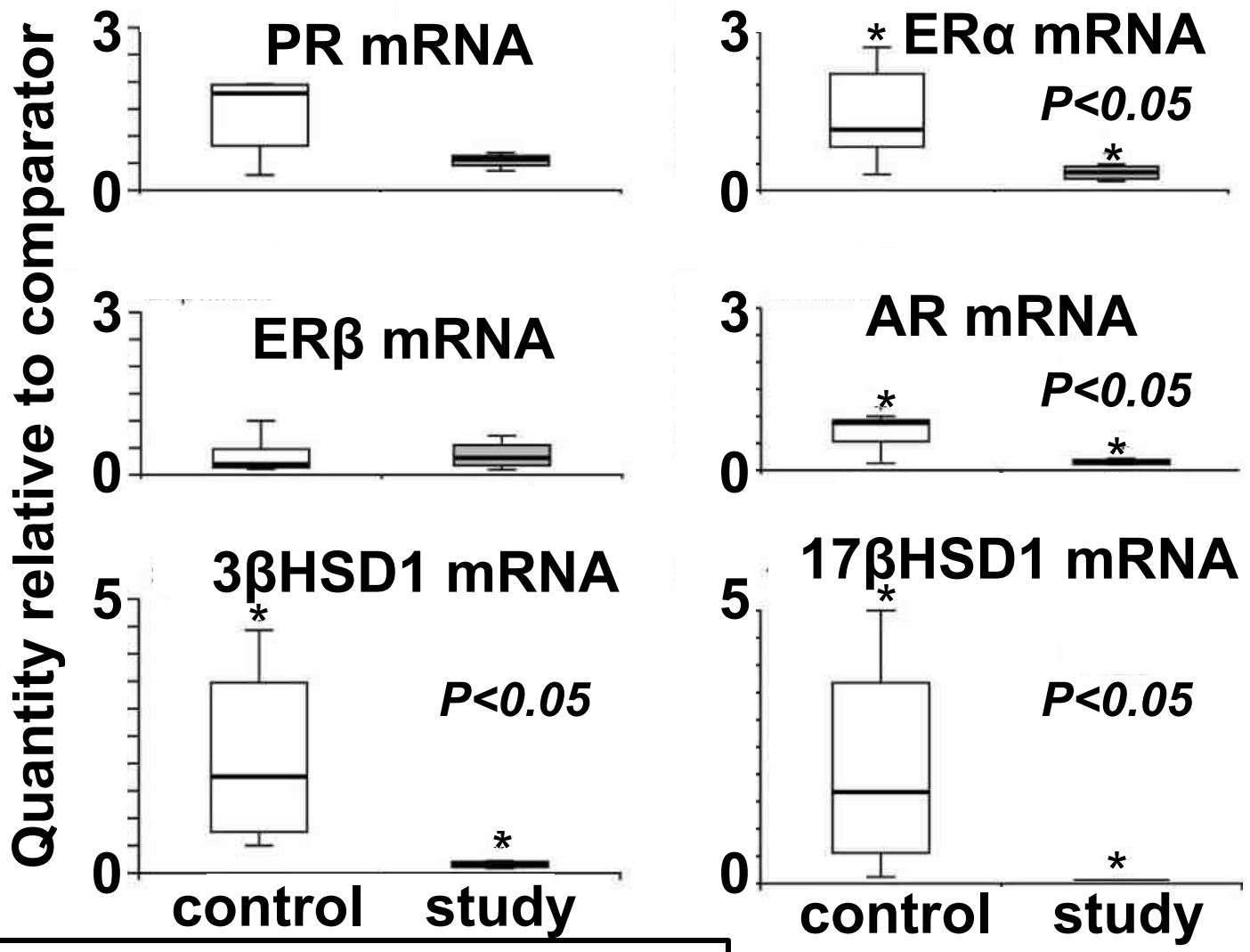


*protein

L=luminal
G=glandular
LH/hCG+2/7

Simón et al. 2005
Hum. Reprod. 20, 3318-27 (modified)

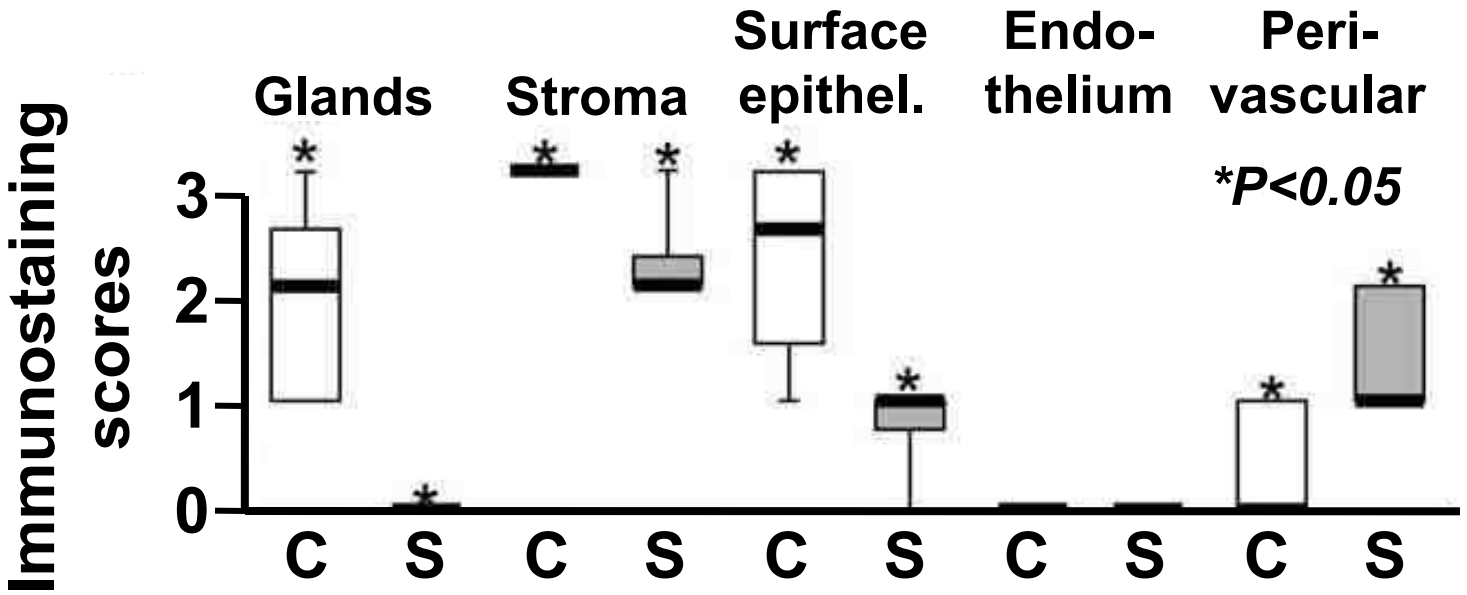
STEROID RECEPTORS AND METABOLIZING ENZYMES IN MID-LUTEAL ENDOMETRIUM



Donors (study)
COH (rFSH/GnRH Ant+P4 suppl.)

Vani et al., 2007
Hum. Reprod. 22, 2981-91

PROGESTERONE RECEPTOR* IN MID-LUTEAL ENDOMETRIUM



*protein

C: Control group (n=8)

S: Study group (n=5)

Vani et al., 2007

Hum. Reprod. 22, 2981-91

RECEPTORS IN HUMAN ENDOMETRIUM (Immunohistochemistry)

	Natural cycles (n = 12)	Stimulated cycles (n = 23)	P
ER			
Stroma	0.13±0.14	0.17±0.22	NS
Glands	0.38±0.35	0.81±0.53 ↑	.01
PR			
Stroma	1.29±0.23	1.61±0.45 ↑	.02
Glands	2.11±0.43	1.45±0.81 ↓	.02

LH/HCG+7 (Long agonist)
Infertile, no ET

Chai et al., 2011
Fertil. Steril. 96, 764-8

OVARIAN STIMULATION (Luteal phase)

- Endocrine changes
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- **Gene transcripts**
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FUNCTIONAL GENOMICS IN THE ENDOMETRIUM (COS cycles)

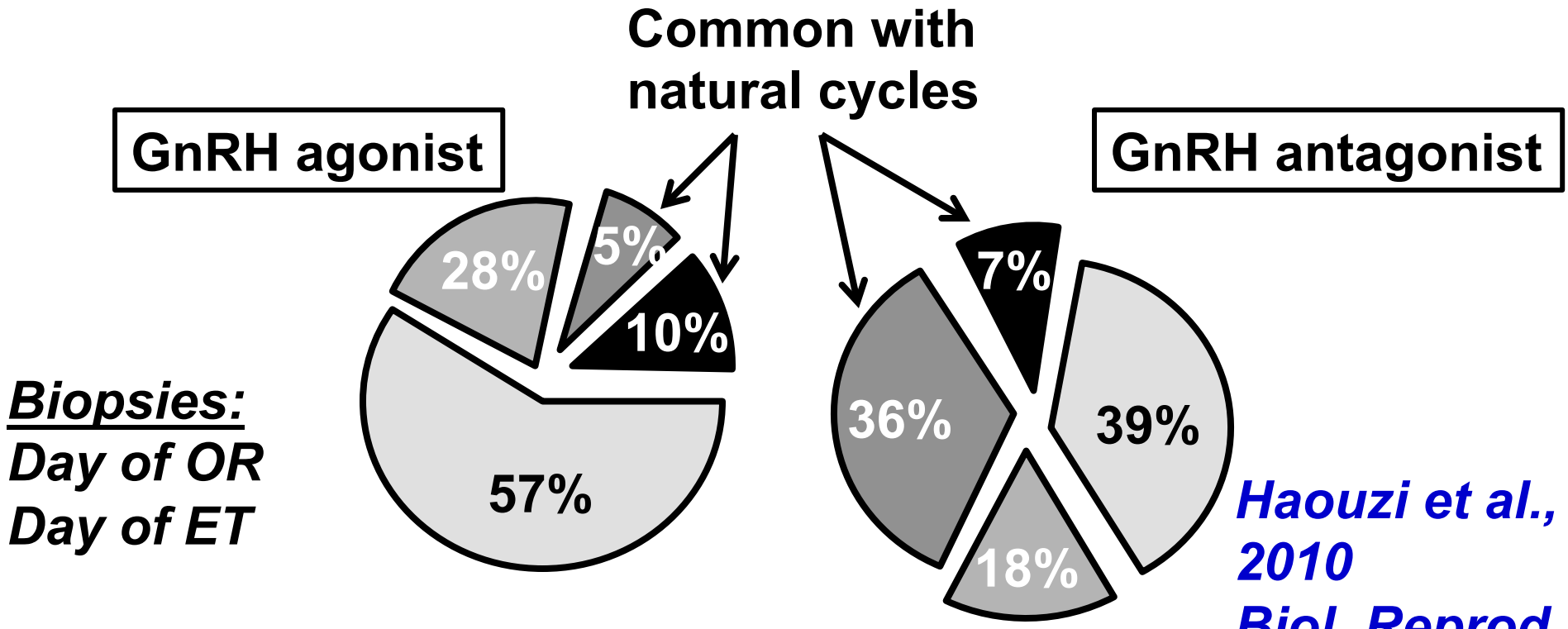
- **2-day delay in the:**
  - ***Activation*** **of 218 implantation genes**
  - ***Repression*** **of 133 implantation genes**

from the pre-receptive (days LH/HCG+1 until LH/HCG+5) to the receptive phase (day LH+7/HCG+7)

Microarray technology

Horcajadas et al., 2008
J Clin Endocrinol Metab 93: 4500–10

GENES IN COMMON DURING THE RECEPTIVE ENDOMETRIUM



FUNCTIONAL GENOMICS IN COS (Endometrial receptivity)

- **140 genes dys-regulated when serum P4 >1.5 on HCG day (n=6) vs P4<1.5 ng/ml (n=6):**
  - **64 up-regulated**
  - **76 down-regulated**

  } **Day rHCG+7 (biopsy)**

- **Genes related to cell adhesion, developmental processess, the immune system**

Microarray technology

Labarta et al., 2011
Hum. Reprod. 26, 1813-25

OVARIAN STIMULATION (Luteal phase)

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CLINICAL OUTCOME (Impaired endometrium)

	Fresh	Cryopreservation (artificial endometrial preparation)
Retrievals	67	70
Blastocyst transfers	53	50
Clin. Pregn. Rate (%)	54.7	84*
Ong. Pregn. Rate (%)	50.9	78*
Early preg, lossess (%)	19.4	13.3
Implantation rate (%)	38.9	70.8**

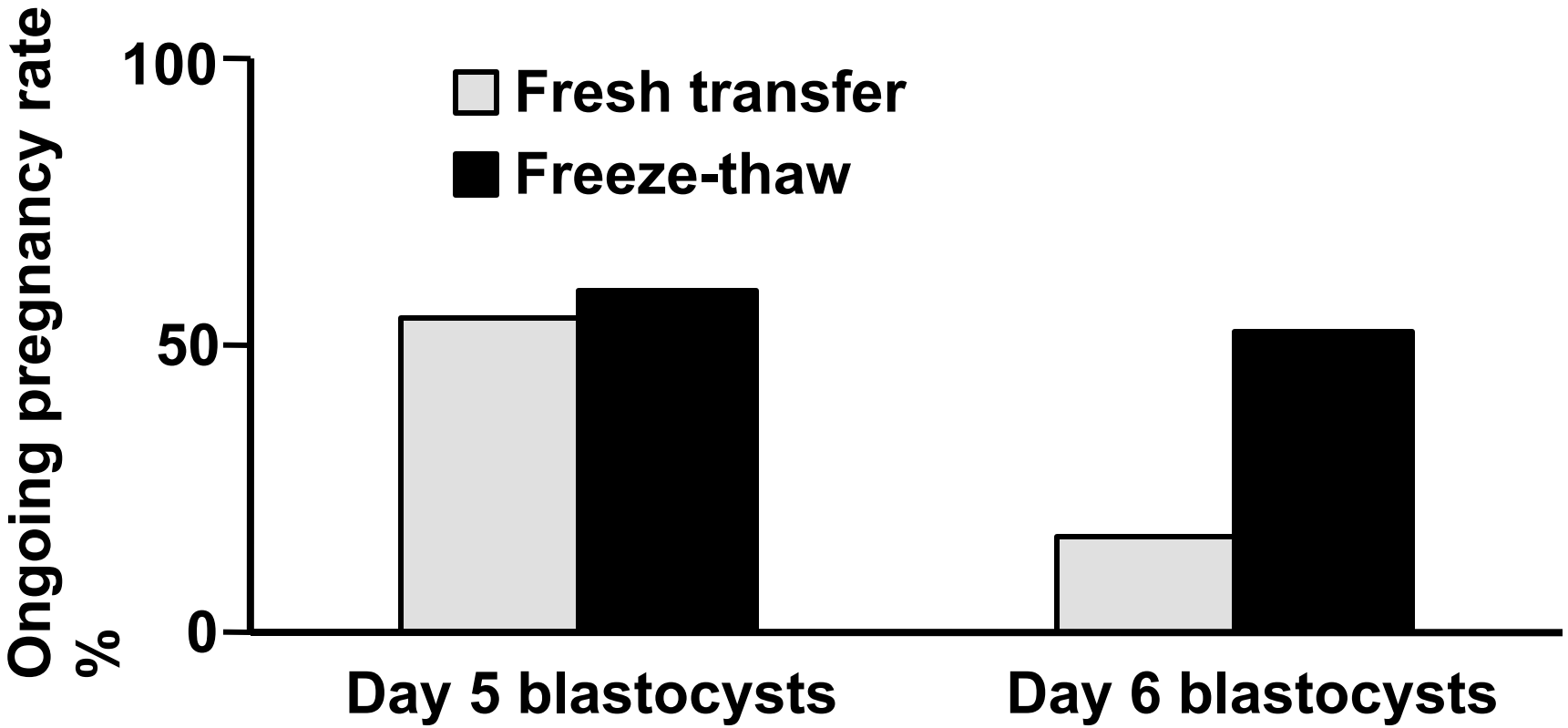
Prospective RCT

** P<0.05*

*** P<0.001*

Shapiro et al., 2011
Fertil. Steril. 96, 344-48

REDUCED PREGNANCY RATE ON DAY 6 (fresh vs thaw)



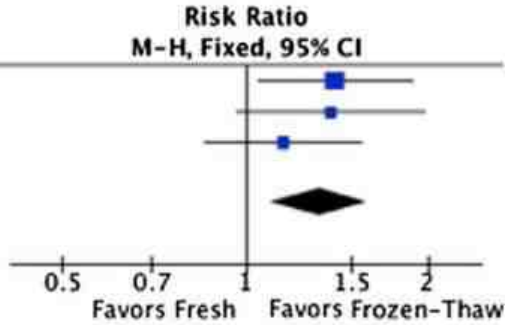
Shapiro et al., 2013
Fertil. Steril. 99, 389-92

FRESH vs FROZEN ET

Ong. Preg. A

Study or Subgroup	Frozen-Thawed Events	Total	Fresh Events	Total	Weight	Risk Ratio M-H, Fixed, 95% CI
Aflatoonian 2010	73	187	52	187	46.0%	1.40 [1.05, 1.88]
Shapiro 2011 - Normal	39	70	27	67	24.4%	1.38 [0.97, 1.98]
Shapiro 2011 -High	38	60	34	62	29.6%	1.15 [0.86, 1.55]
Total (95% CI)		**317**		**316**	**100.0%**	**1.32 [1.10, 1.59]**
Total events	150		113			

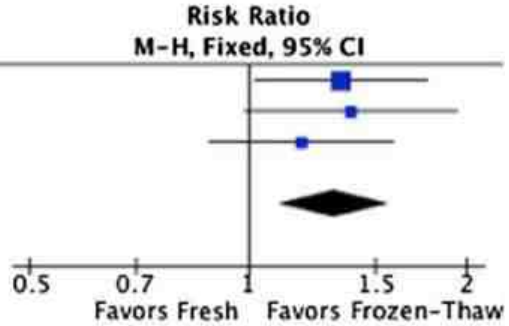
Heterogeneity: $Chi^2 = 1.03$, $df = 2$ ($P = 0.60$); $I^2 = 0\%$
Test for overall effect: $Z = 3.00$ ($P = 0.003$)



Clin. Preg. B

Study or Subgroup	Frozen-Thawed Events	Total	Fresh Events	Total	Weight	Risk Ratio M-H, Fixed, 95% CI
Aflatoonian 2010	78	187	58	187	47.9%	1.34 [1.02, 1.77]
Shapiro 2011 - Normal	42	70	29	67	24.5%	1.39 [0.99, 1.94]
Shapiro 2011 -High	39	60	34	62	27.6%	1.19 [0.88, 1.59]
Total (95% CI)		**317**		**316**	**100.0%**	**1.31 [1.10, 1.56]**
Total events	159		121			

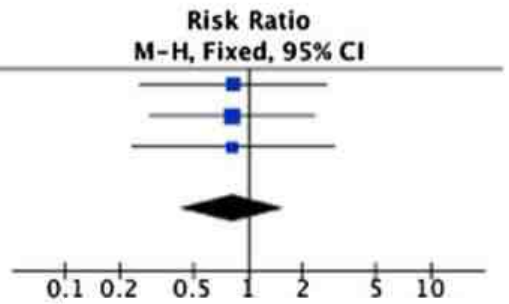
Heterogeneity: $Chi^2 = 0.60$, $df = 2$ ($P = 0.74$); $I^2 = 0\%$
Test for overall effect: $Z = 3.04$ ($P = 0.002$)



Misc. C

Study or Subgroup	Frozen-Thawed Events	Total	Fresh Events	Total	Weight	Risk Ratio M-H, Fixed, 95% CI
Aflatoonian 2010	5	187	6	187	33.2%	0.83 [0.26, 2.68]
Shapiro 2011 - Normal	6	70	7	67	39.6%	0.82 [0.29, 2.32]
Shapiro 2011 -High	4	60	5	62	27.2%	0.83 [0.23, 2.93]
Total (95% CI)		**317**		**316**	**100.0%**	**0.83 [0.43, 1.60]**
Total events	15		18			

Heterogeneity: $Chi^2 = 0.00$, $df = 2$ ($P = 1.00$); $I^2 = 0\%$
Test for overall effect: $Z = 0.56$ ($P = 0.57$)

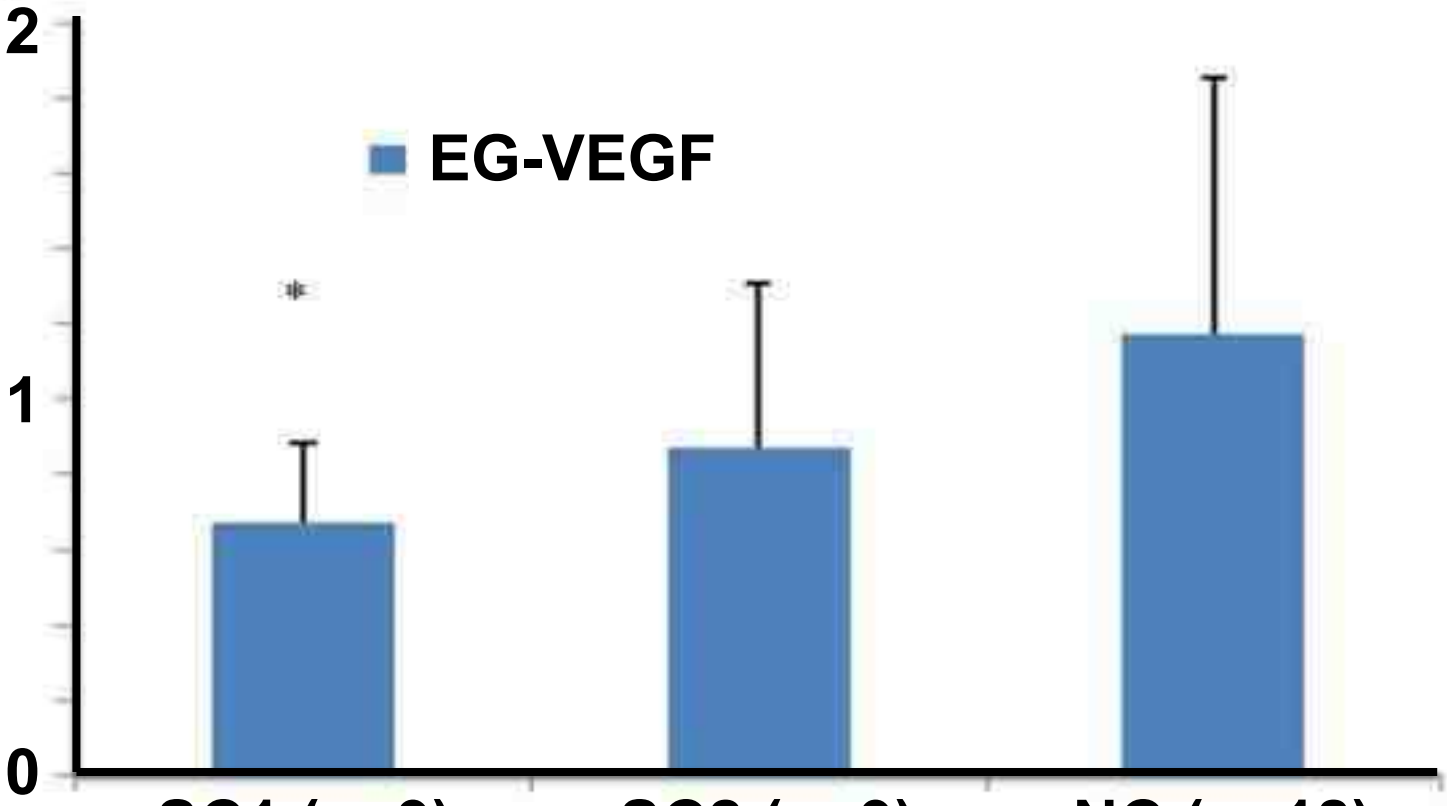


Favors fresh Favors frozen

IVF

Roque et al., 2013; Fertil. Steril. 99, 156-62

PERI-IMPLANTATION ENDOMETRIUM



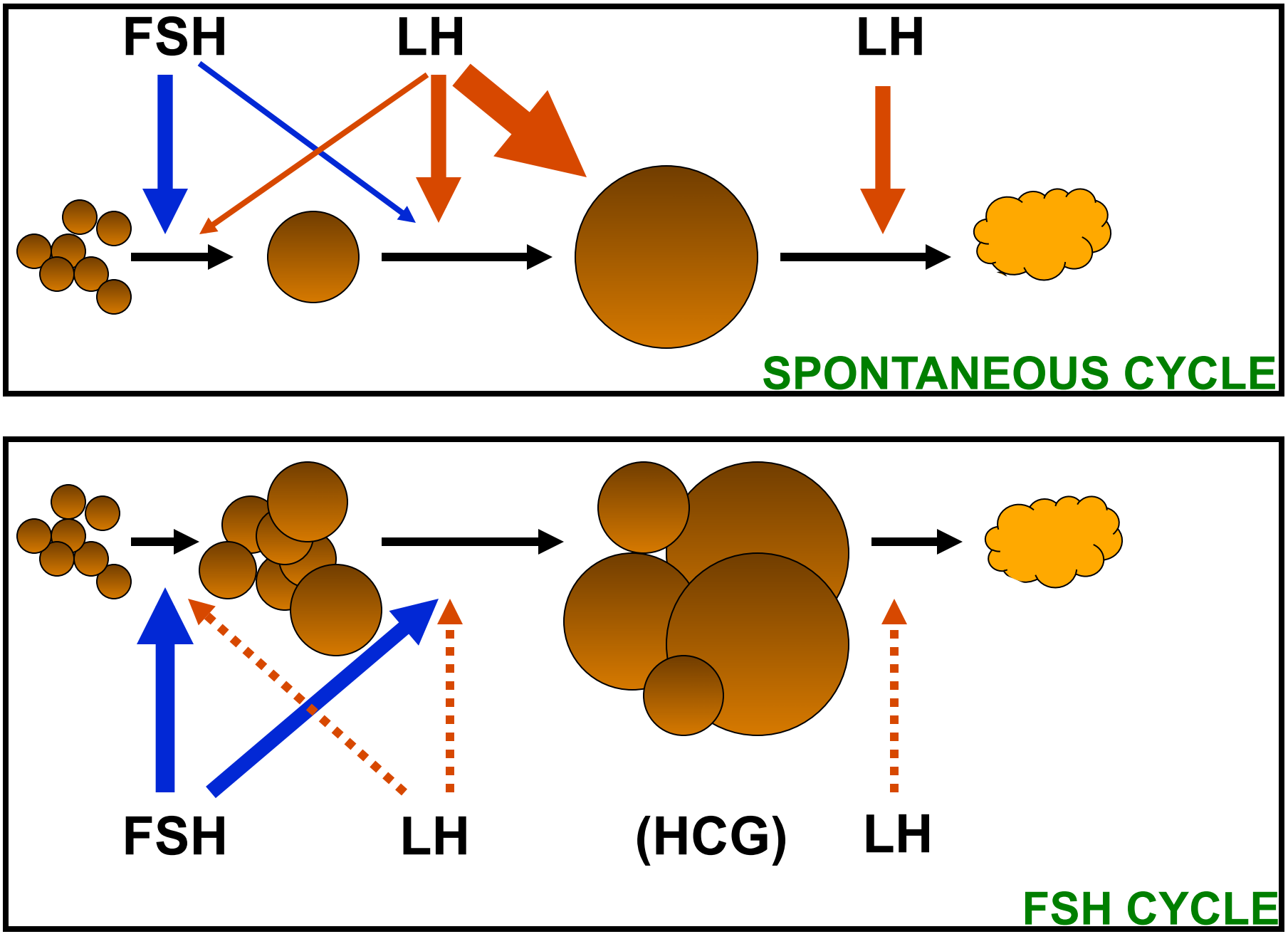
mRNA
RT PCR

Biopsy 6 days after ovul. or OR

Xu et al., 2015
Int. J. Clin. Exp. Pathol. 1, 8902-11

OVARIAN STIMULATION (Luteal phase)

- Endocrine changes
- Endometrial morphology changes
- Gene transcripts
- Clinical outcome
- **Luteal support**


FSH
LH
LH
SPONTANEOUS CYCLE
FSH
LH
(HCG)
LH
FSH CYCLE

LUTEAL PHASE SUPPORT

- **P4 or HCG higher Ong.PR and LBR than placebo**
- **GnRH-a added to P4 improves pregnancy outcomes**
- **HCG increases the OHSS rate**
- **Addition of E2 does not improve the clinical outcome**
- **The route of P4 administration does not affect the outcome**

Van der Linden et al., 2015
Cochrane Database Syst. Rev. Jul 7;7:CD009154

TAKE HOME MESSAGES

- **In stimulated cycles:**
  - **LH levels are markedly suppressed in the luteal phase**
  - **Morphological changes, mainly of advanced endometrial maturation (histology, pinopodes), take place**
  - **Changes in functional genomics occur**
  - **Transfer of cryopreserved embryos overcomes the severely compromised endometrial receptivity**