Differential regulation of vascular mediators by GnRHa triggering

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Why do we use hCG

60´s – LH was measured to time OPU

70´s – agonist triggering described

Nakano, 1973

80´s – hCG binds and activates LH receptor

Kessler, 1979

- good for timing OPU
- OHSS developed

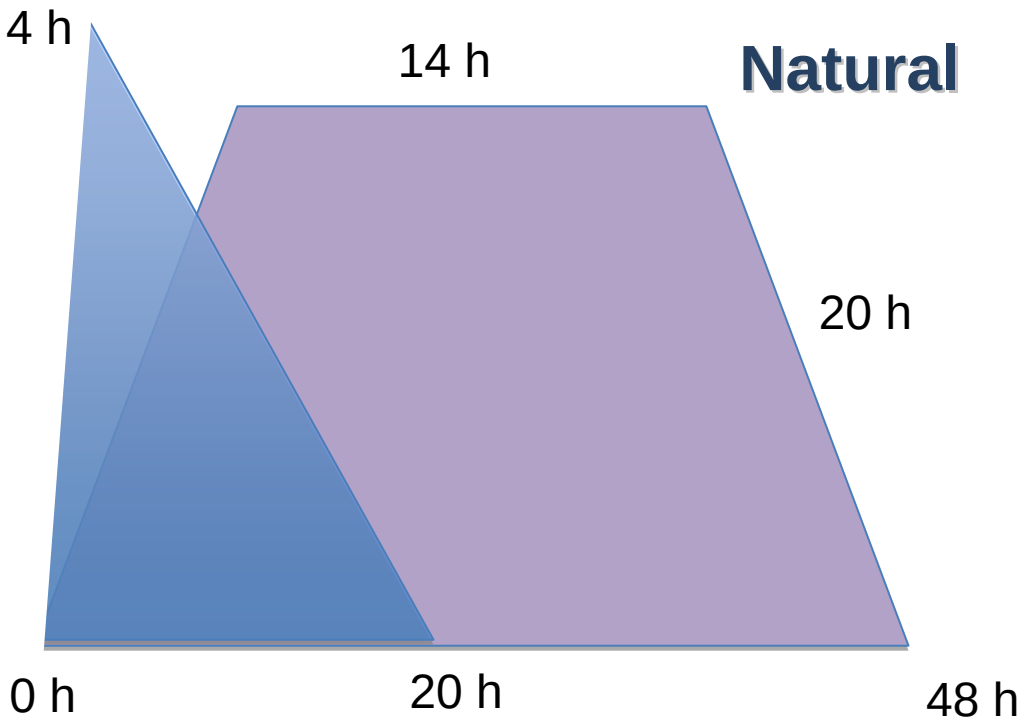
00´s – interest renewed in the antagonists era

- **OHSS still remains a complication of ART**
- **-ovarian stimulation**
- **- HCG for triggering**

Prevalence: 0.6 – 14% in IVF
Severe OHSS: 0.2-0.5%

Mozes, Lancet 1965
García Velasco & Pellicer, 2002

LH surge: GnRHa vs natural



Additional FSH surge

-induce LH receptor in GC
-promote oocyte nuclear maturation

Hoffer, 1983; Gonen, 1990; Itskovitz, 1991

GnRHa triggering

- Triggering final oocyte maturation with a bolus of GnRHa **is possible** under the antagonist protocol

Itskovitz et al 1988; Lanzone et al 1989; Gonen et al 1990

- GnRHa triggering has shown to **prevent OHSS** but **poor reproductive outcome** was reported (LPD)

Fauser et al 2002; Griesinger et al 2006

GnRH agonist: poorer outcome

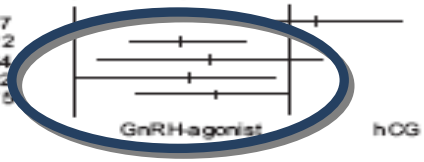
GnRH agonist for triggering final oocyte maturation in the GnRH antagonist ovarian hyperstimulation protocol: a systematic review and meta-analysis

G.Griesinger[1,3], K.Diedrich[1], P.Devroey[2] and E.M.Kolibianakis[2]

[1]Department of Obstetrics and Gynecology, University Clinic of Schleswig-Holstein, Campus Luebeck, Luebeck, Germany and [2]Centre of Reproductive Medicine, Dutch Speaking, Brussels Free University, Brussels, Belgium

Pregnancy rate per randomised patient

Citation	EffectName	GnRH-agonist	hCG	NTotal	Effect	Lower	Upper	PValue
Fauser, 2002	PR	6 / 32	2 / 15	47	1,500	,265	8,489	,645
Humaidan, 2005	PR	3 / 55	24 / 67	122	,103	,029	,367	,000
Kolibianakis, 2005	PR (centre 2)	1 / 34	5 / 30	64	,152	,017	1,380	,060
Kolibianakis, 2005	PR (centre 1)	1 / 18	10 / 24	42	,082	,009	,724	,008
Combined (4)		11 / 139	41 / 136	275	,214	,055	,840	,027



Pregnancy loss

Citation	EffectName	GnRH-agonist	hCG	NTotal	Effect	Lower	Upper	PValue
Fauser, 2002	Preg. loss	5 / 11	3 / 5	16	,556	,065	4,755	,590
Humaidan, 2005	Preg. loss	11 / 14	1 / 25	39	58,000	8,201	944,219	,000
Kolibianakis, 2005	Preg. loss (centre 2)	5 / 6	1 / 6	12	5,000	1,200	520,734	,021
Kolibianakis, 2005	Preg. loss (centre 1)	2 / 3	1 / 11	14	,000	,848	471,576	,031
Combined (4)		23 / 34	6 / 47	81	,515	,954	138,988	,054

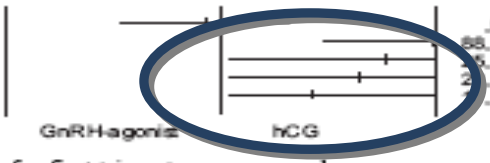
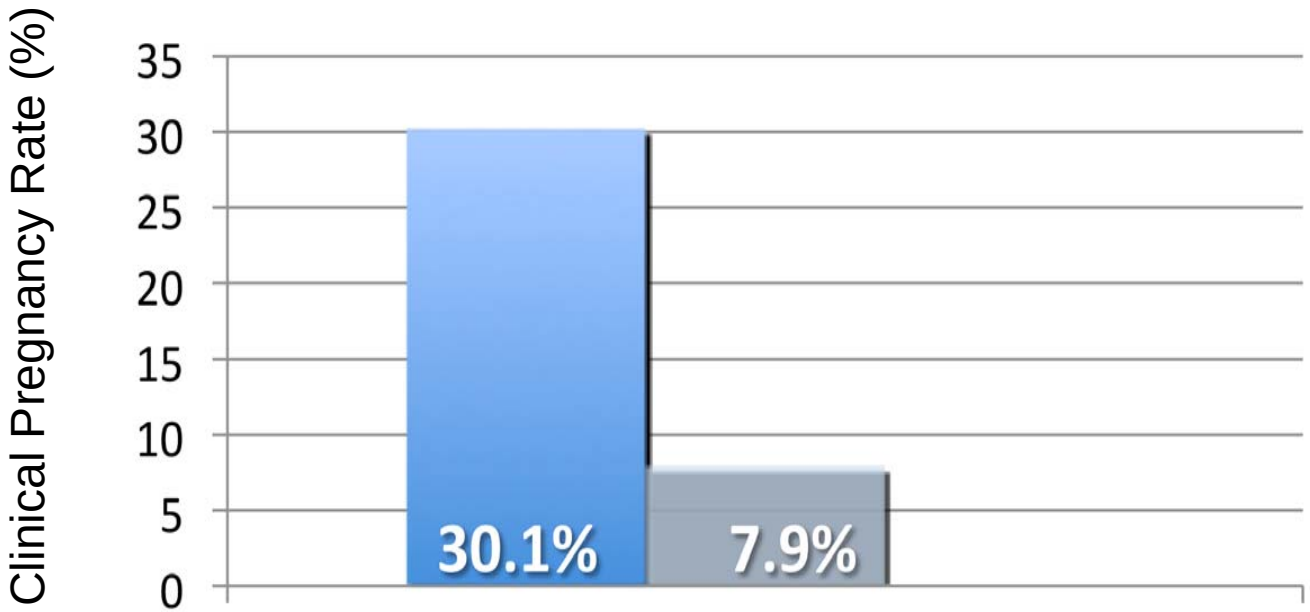


Figure 1. Forest plots of odds ratios. Outcomes were heterogenous at $P = 0.01$ for first trimester pregnancy loss.

GnRH agonist: poorer outcome

Meta-analysis

Pooled data
Reduced probability of clinical pregnancy



Griesinger et al 2006

Luteal phase

- All stimulated IVF/ICSI cycles have abnormal LP

vs 8.1% natural cycles

Edwards, 1980; Fatemi, 2007

- Main reason is the inhibition of LH secretion due to supraphysiological steroid levels
  - multifollicular development
  - long half life of hCG

Tavaniotou, 2001; Fauser and Devroey, 2003

- Shorter duration of LH secretion = luteolysis

How to rescue the luteal phase

hCG
or
rLH

Humaidan et al 2010
Castillo et al 2010
Papanikolau et al 2010

intensive
steroid
support

Engman et al 2008

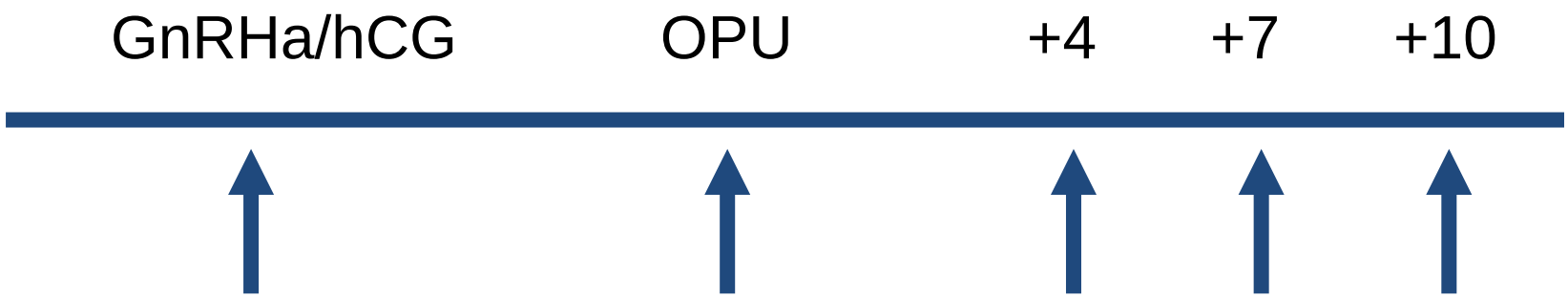
Trying to understand the LP

- Pilot study
- 20 egg donors
- 150 rFSH – antag day 5
- 0.2 mg triptorelin to trigger
- >6 and <20 folls >14mm

- **a) E2 patches /3 days + P4 200/12h**
- **b) same + 500 IU triggering day +4, +7 and +10**

Garcia-Velasco et al. Fertil Steril 2010

Trying to understand the LP



- US
- serum (FF)
- questionnaire
- duration LP

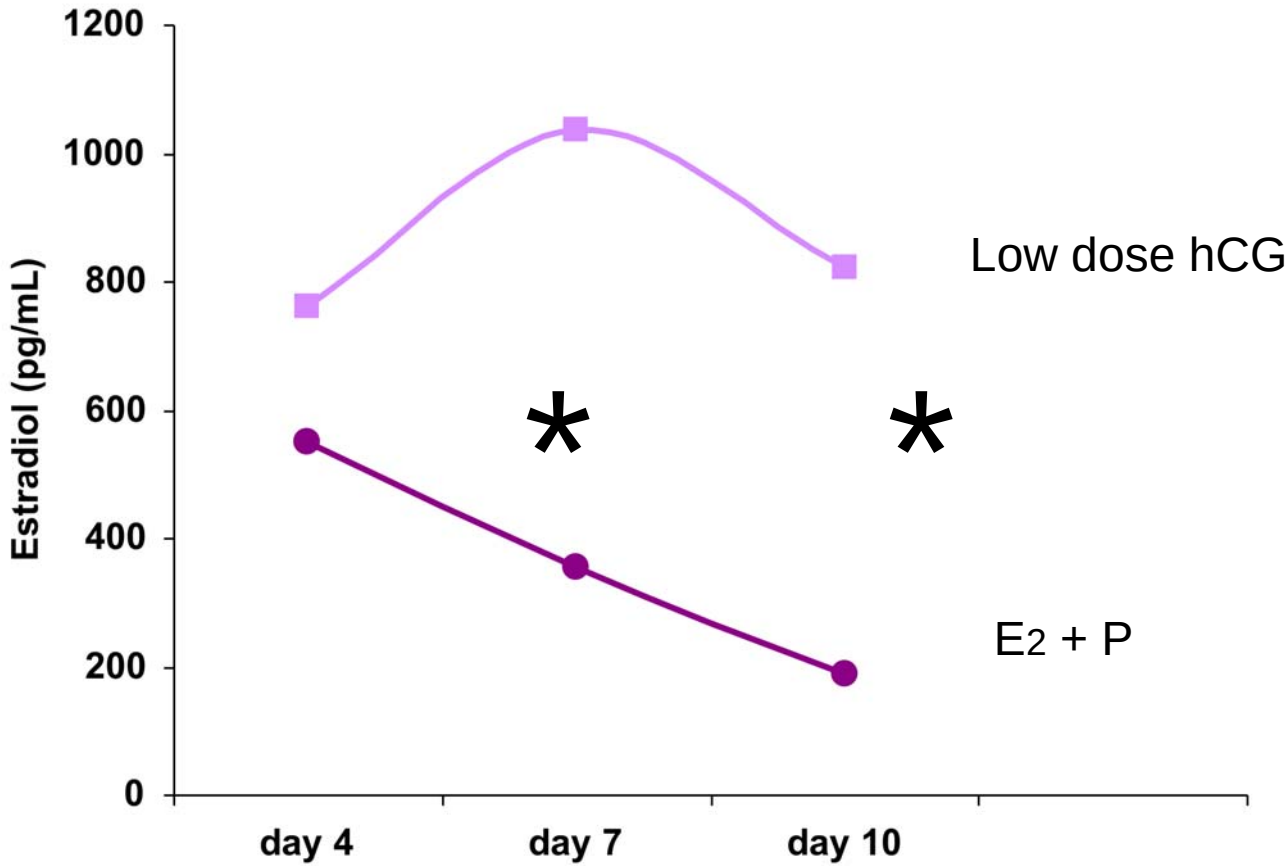
Characteristics

	(hCG) n=10	(E2+P4) n=10	p value
Age (y)	28.2	26.2	0.264
BMI (kg/m2)	22.1	22.0	0.935
total FSH (IU)	1980	1965	0.912
# oocytes	14.5	15.9	0.734

Luteal phase duration

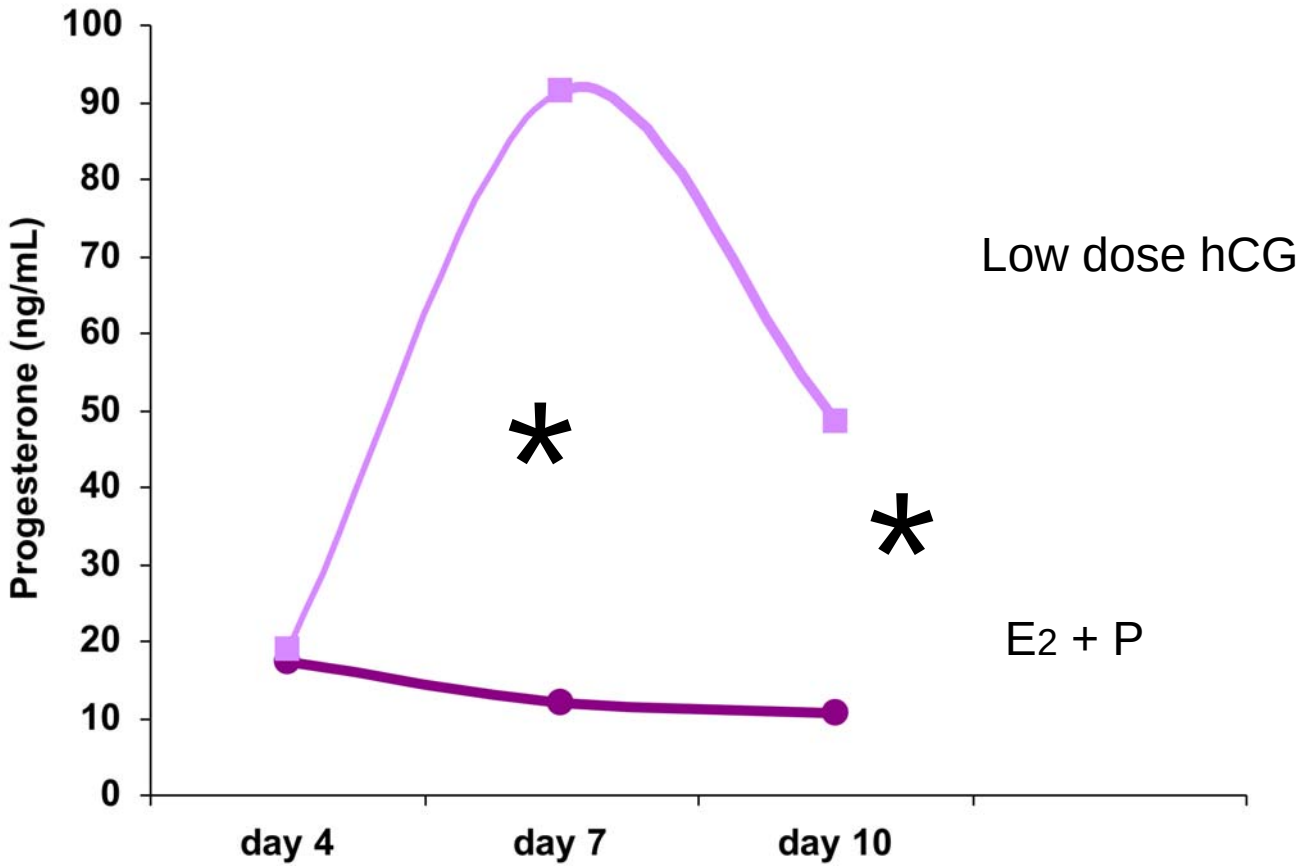
	(hCG) n=10	(E2+P4) n=10	
Duration LP (days)	15	11	p=0.003

Serum estradiol



Garcia-Velasco et al. Fertil Steril 2010

Serum Progesterone



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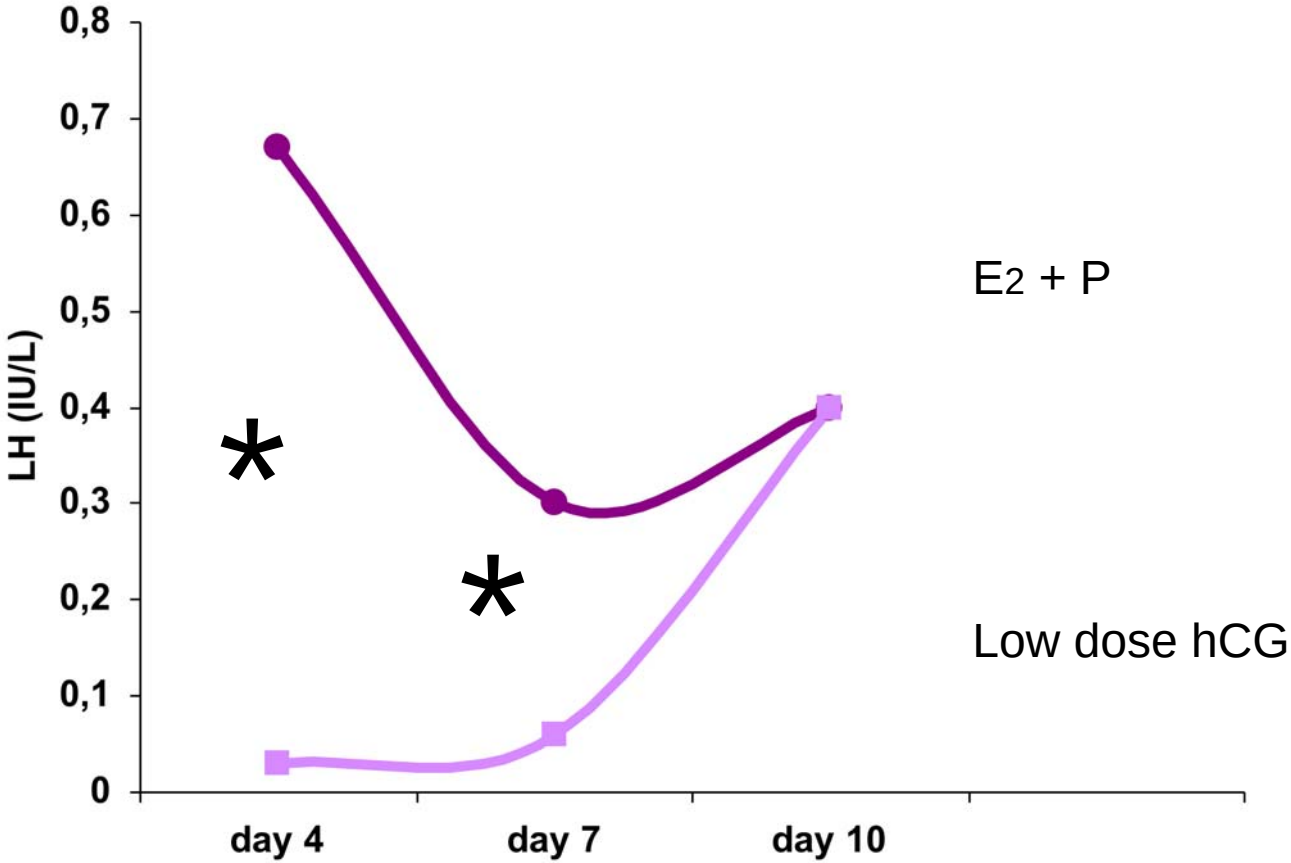
Serum LH



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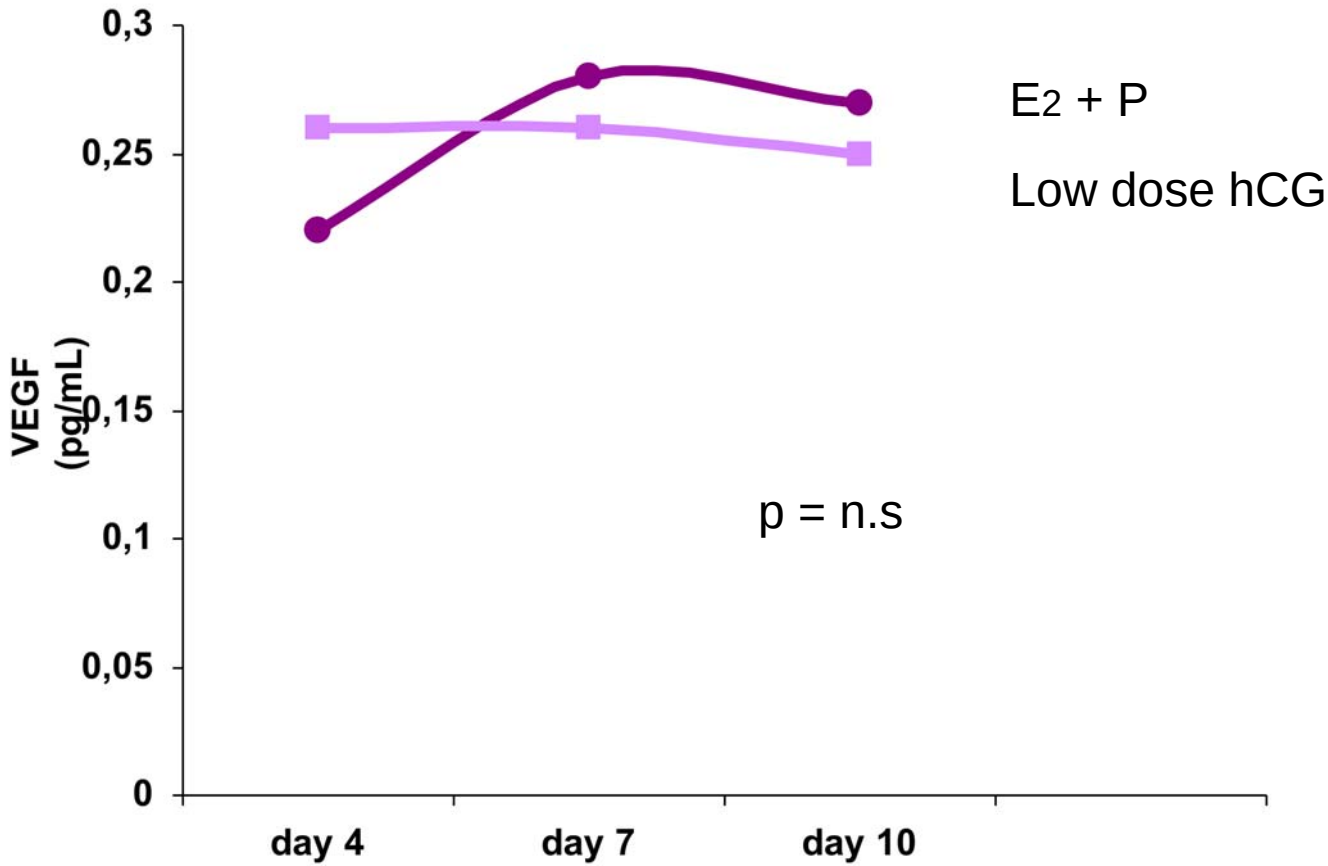
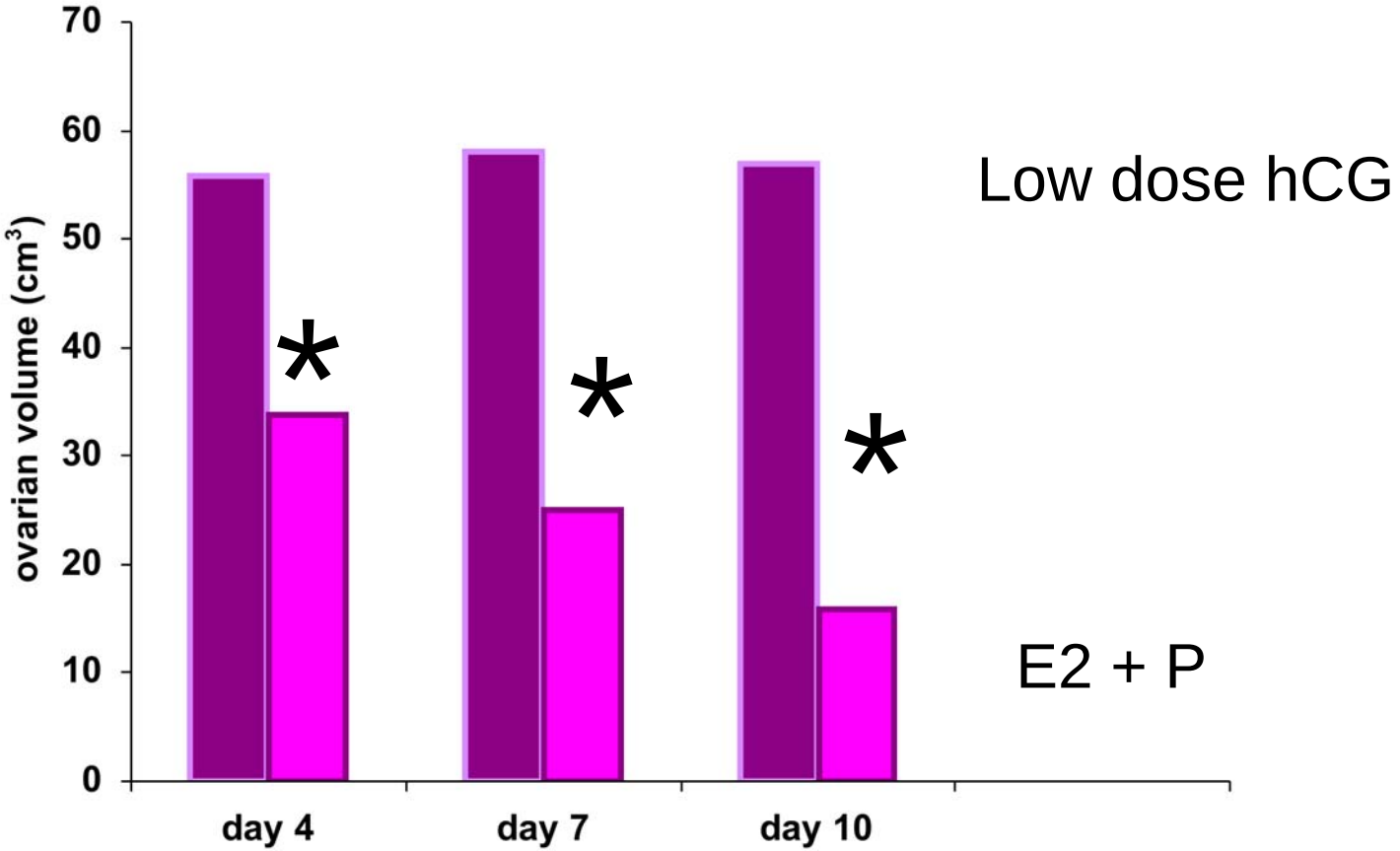
Serum VEGF



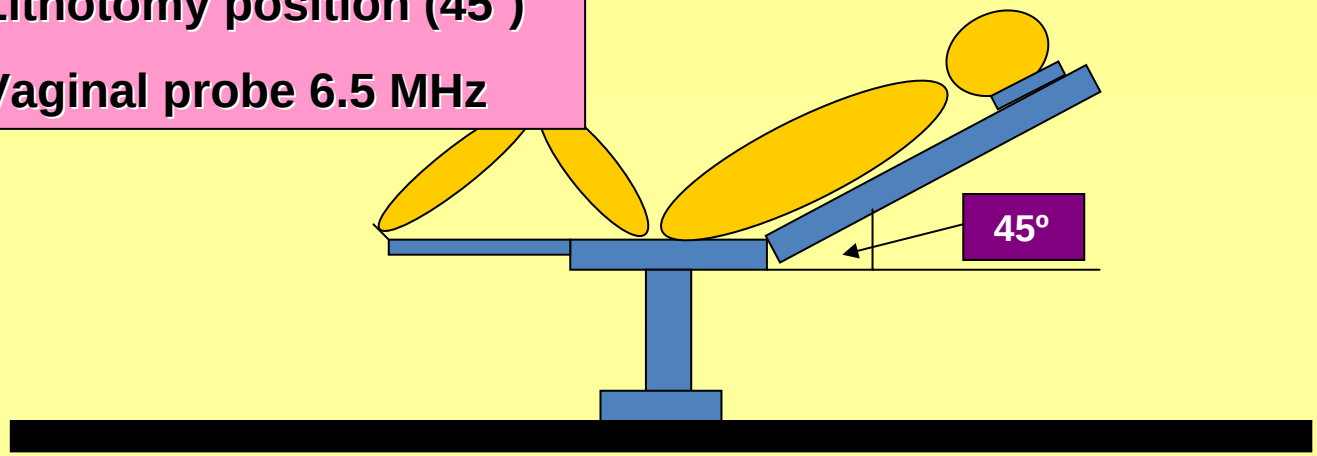
Garcia-Velasco et al. Fertil Steril 2010

Ovarian volume



Free fluid (cm^2)

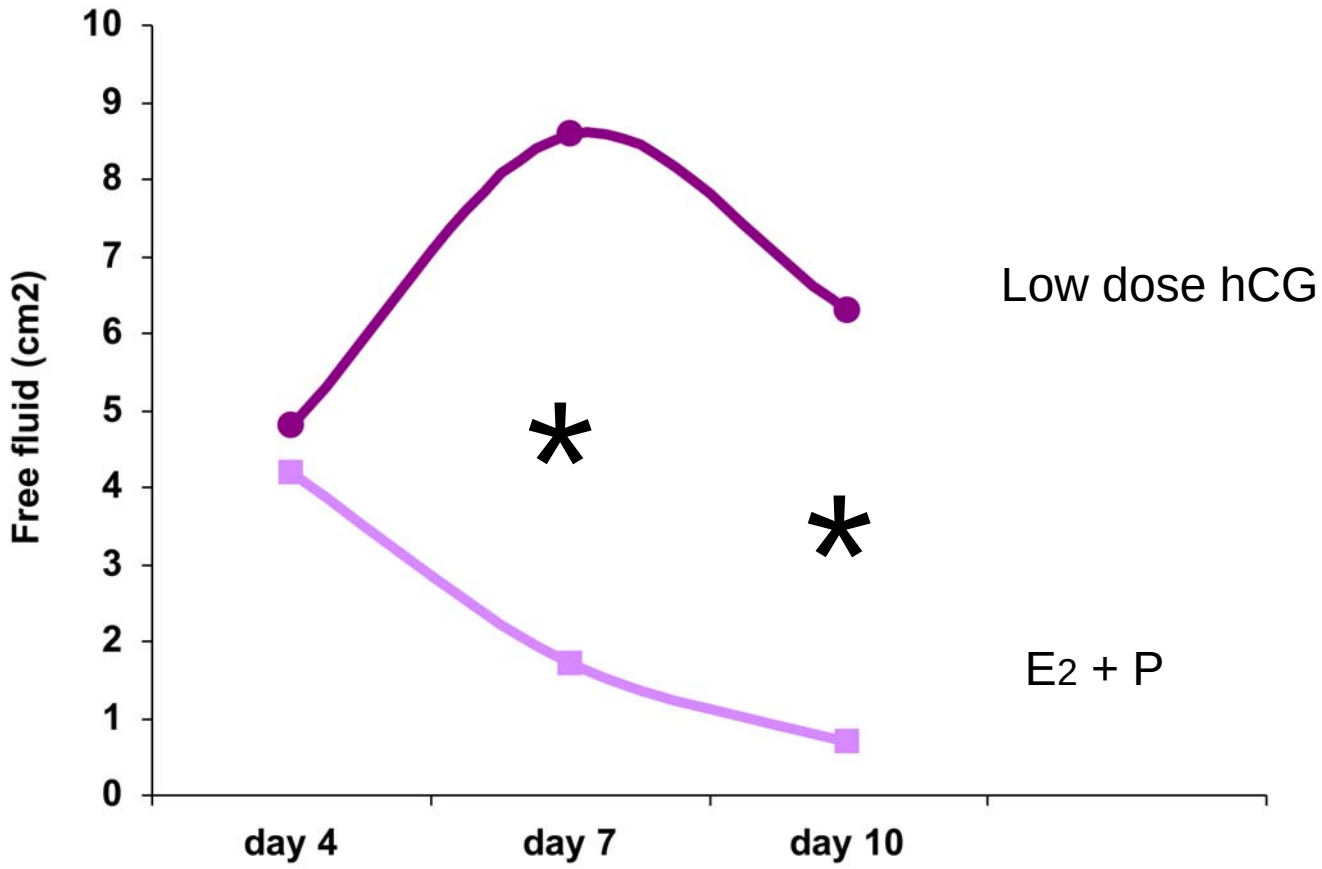
- Vaginal ultrasound
- Lithotomy position (45˚)
- Vaginal probe 6.5 MHz



Same explorer

Alvarez et al. J Clin Endocrinol Metab 2007

Free fluid (cm^2)



GnRH agonist triggering
OHSS prevention

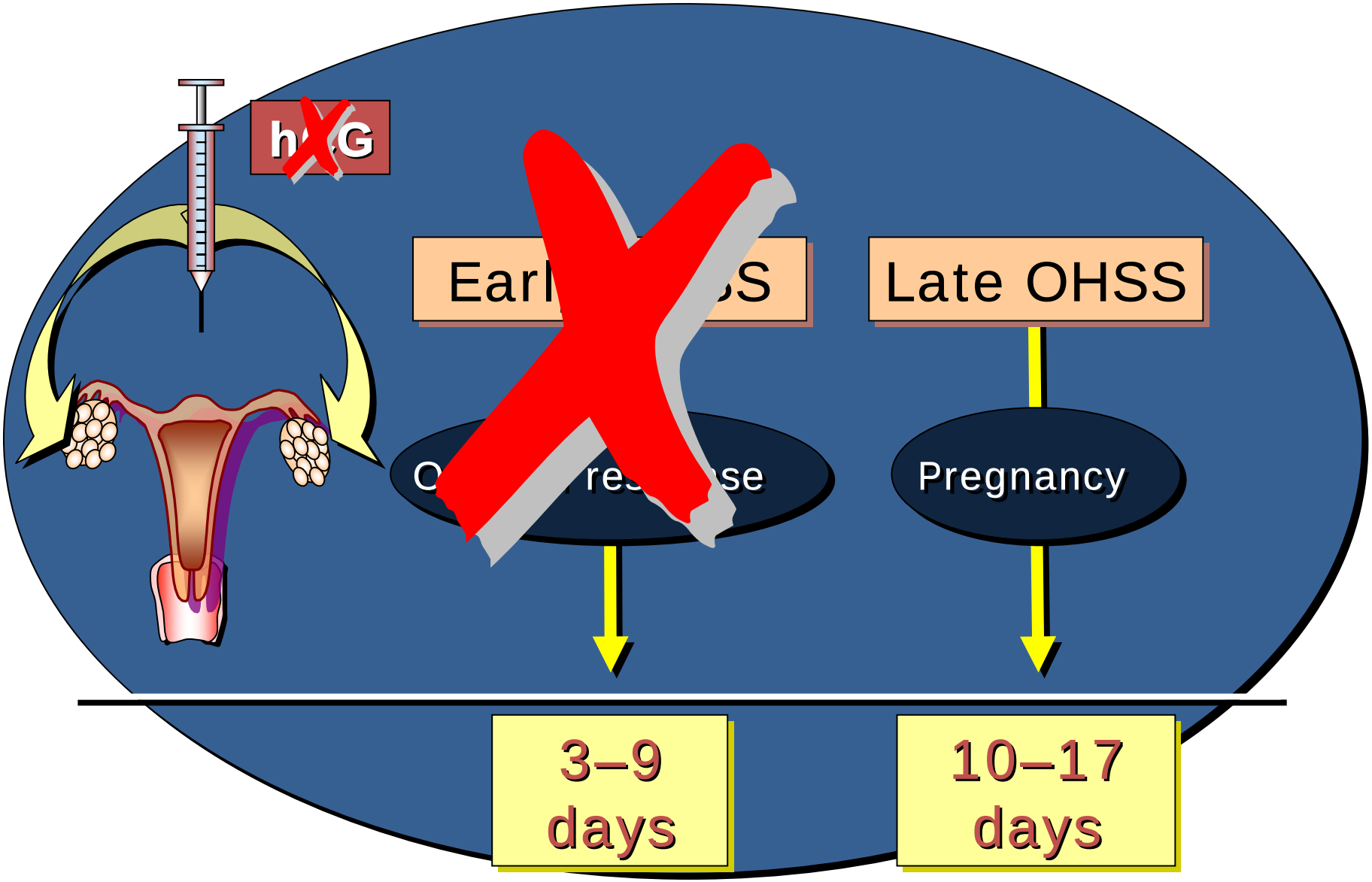
J Assist Reprod Genet (2008) 25:63–66
DOI 10.1007/s10815-008-9198-1

REVIEW

GnRH agonist for triggering final oocyte maturation in patients at risk of ovarian hyperstimulation syndrome: still a controversy?

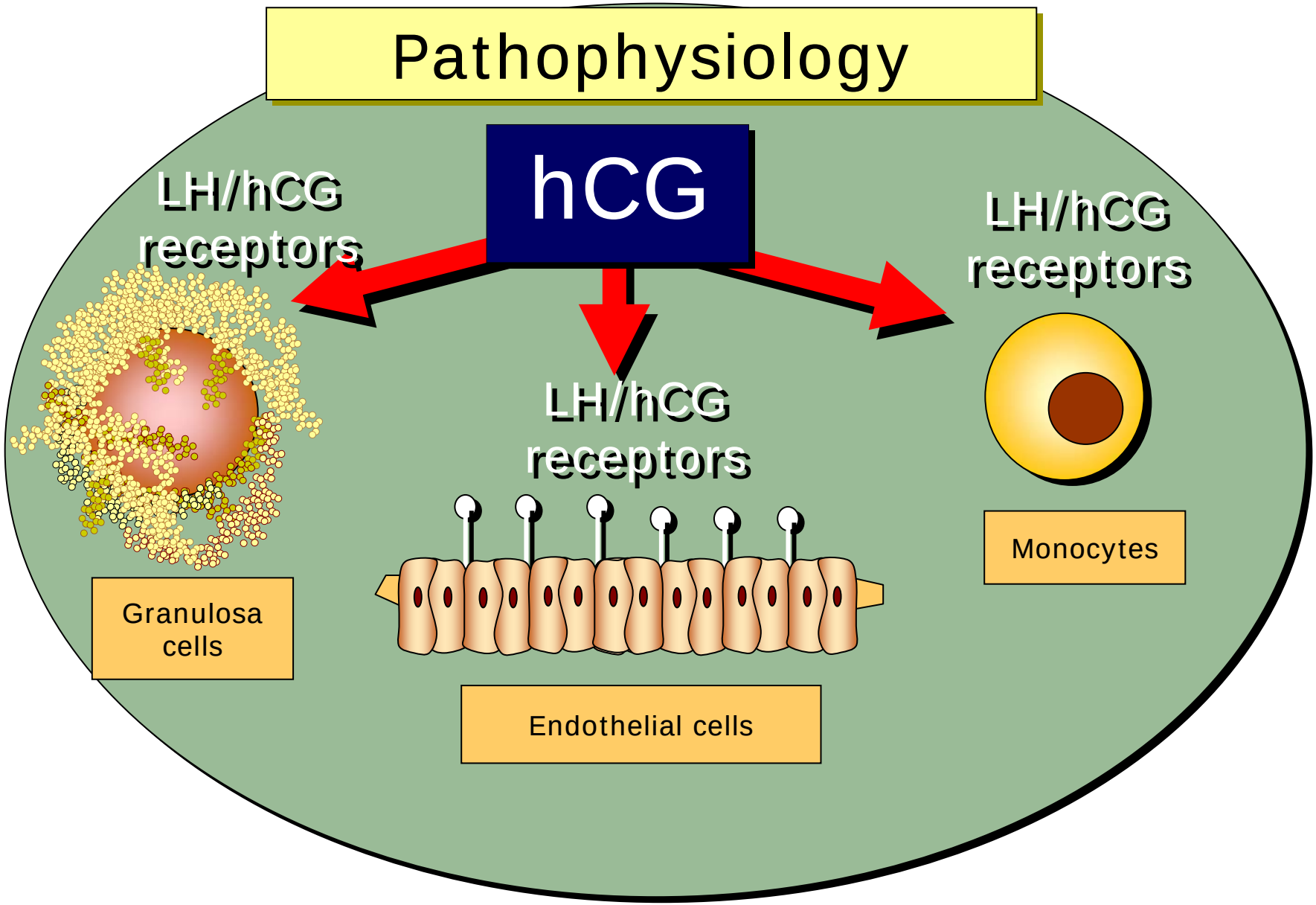
S. Kol · I. Solt

OHSS prevention



Lyons et al. 1994; Mathur et al. 2000

Pathophysiology



Gordon. 1996; Li 1995; Cohen 1996; Standaert et al., 1992; Toth 1994

GnRH agonist triggering
OHSS prevention

Donor studies

	Interventions	OHSS incidence	Luteal support in recipients	Nº eggs, embryo quality PR and IR
Acevedo et al 2006	Antagonist 1) GnRHa 2) hCG	0/30 5/30 (17%)	Oral E2 and natural P4 (600mg9	 NS
Melo et al 2007	Antagonist 1) GnRHa 2) HCG	0/35 6/35 (17%)	Oral E2 and natural P4	 NS
Bodri et al 2008	Antagonist 1) aGnRHa 2) HCG	0/1046 13/1031 (1.26%)	Oral E2 and natural P4	 NS
Shapiro et al 2007	Antagonist 1) GnRHa 2) HCG	0/36 1/42 (2.3%)	E2 oral and P4 im	NS

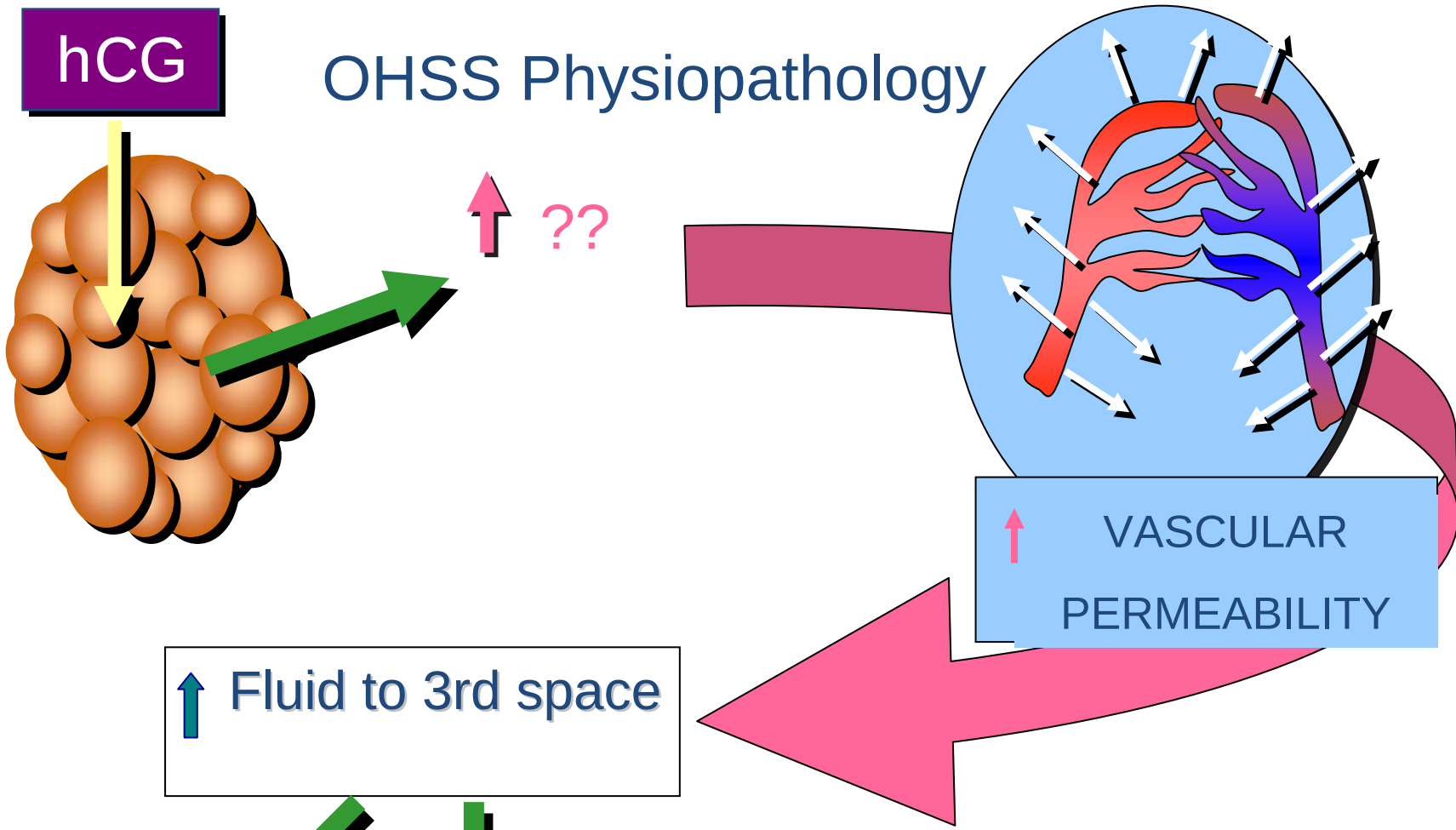
GnRH agonist prevents OHSS

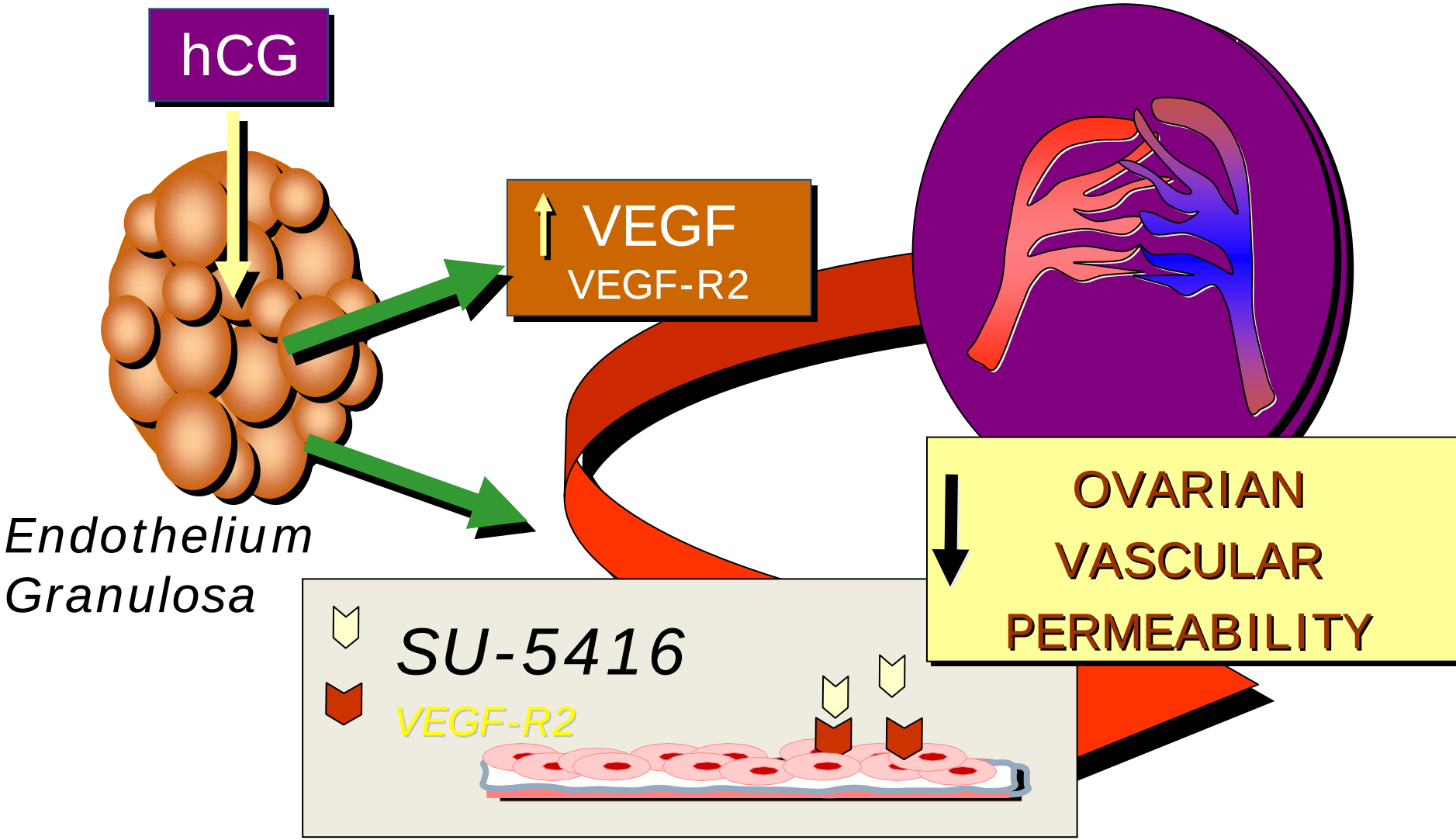
	GnRHa	hCG
n	760	84
age	25.6	26.4
BMI	22.2	22.1
cancelled	8.2	8.5
day [illegible] stim	[illegible]	10.1
FSH dose	1747	1726
# oocytes	13.8	12.5
LP duration	5.2	8.5
general cond	12.8	38.9

NO OHSS !!

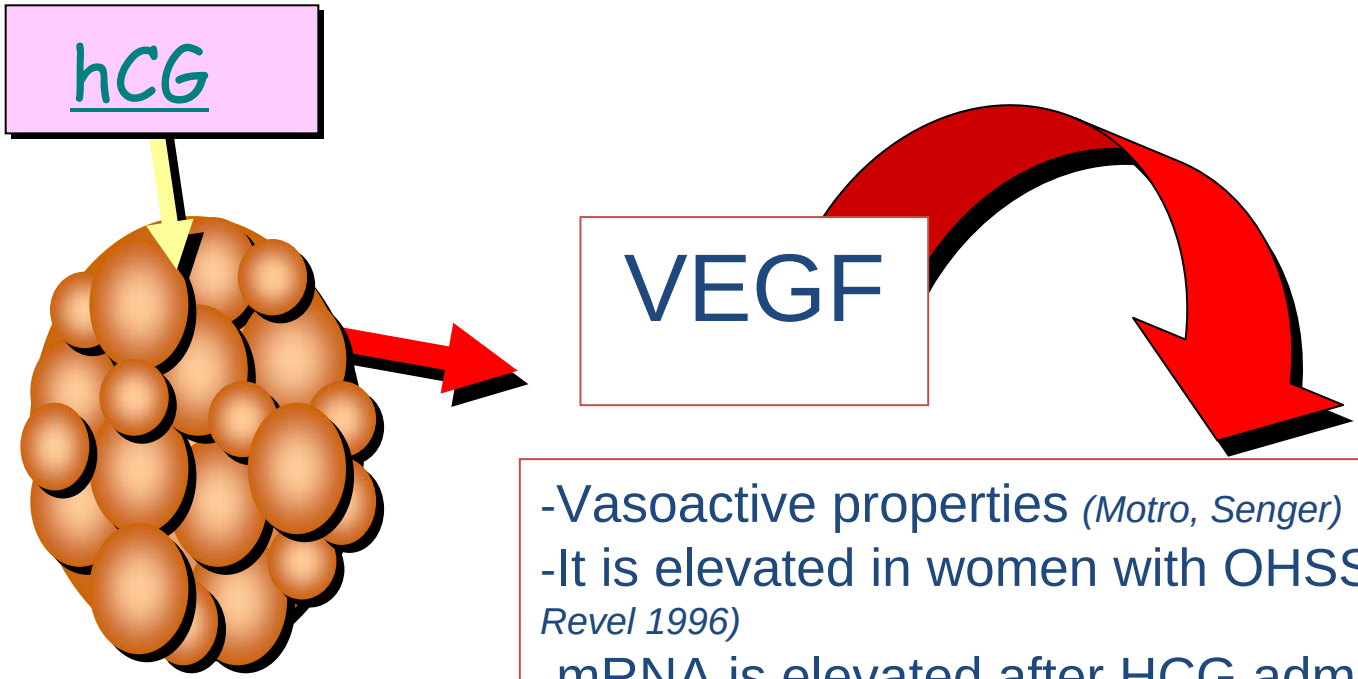
Alvarez A et al (ESHRE 2009)

OHSS Physiopathology





Gómez et al. Endocrinology 2002; 143:4339–4348
Gómez et al. Biol Reprod 2003; 68:2164–2171



-Vasoactive properties *(Motro, Senger)*
-It is elevated in women with OHSS *(Abramou 1997., Revel 1996)*
-mRNA is elevated after HCG administration *(Neulen 1995, Loret de Mola 1996)*
-HCG induce the expression of VEGF and VEGF-R2 in OHSS *(Wang, 2002, Gómez R 2002)*

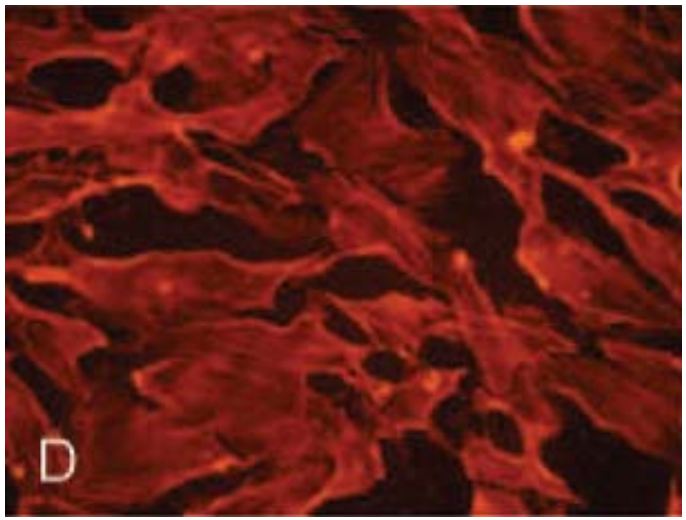
VEGF:

- The key vascular mediator in OHSS
- It is released after HCG administration

Reis et al, Human Reprod Update, 2008

Other mediators

- **sVE-Cadherin**



↑ VASCULAR PERMEABILITY

- sVE-cadherin is expressed in endothelial cells

- Serum levels are higher in patients with OHSS

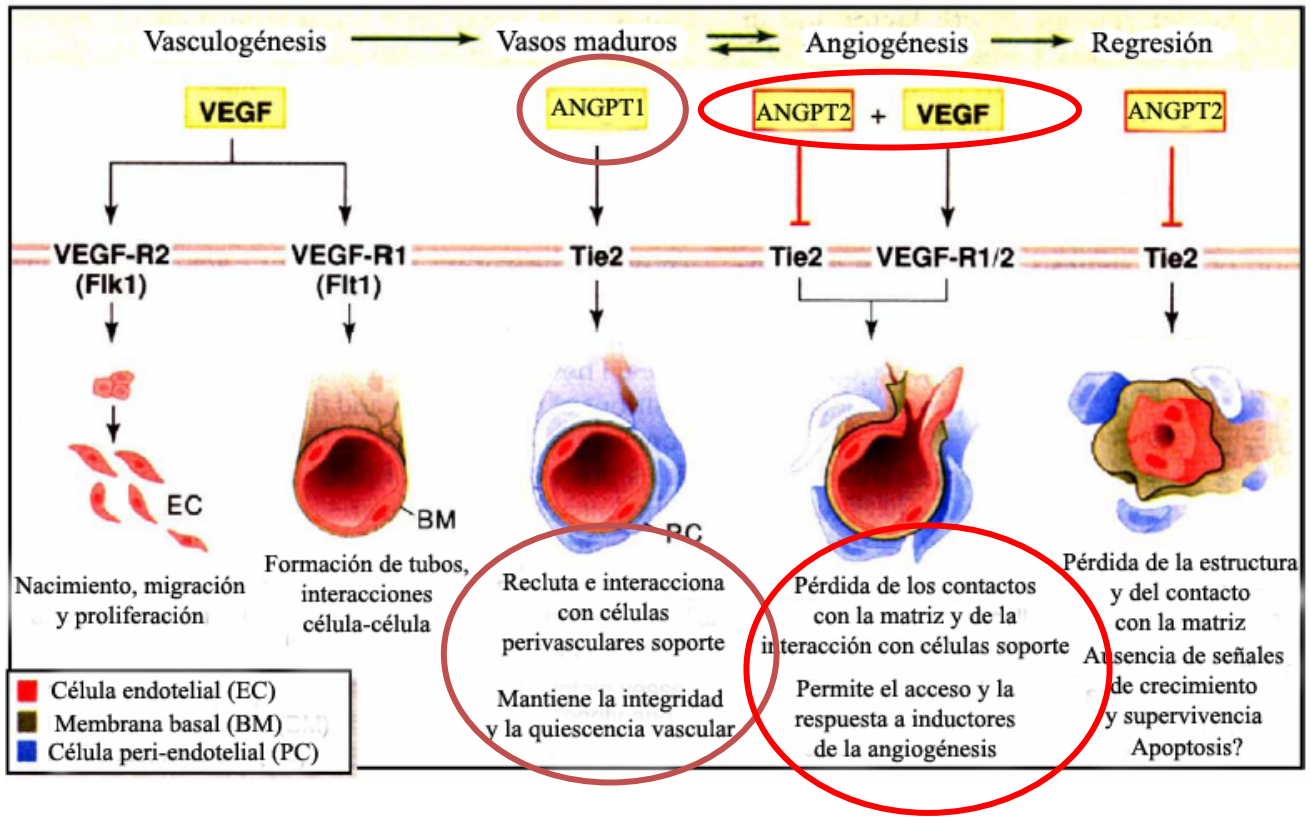
- sVE-cadherin concentration decreased with clinical improvement

Villasante et al Hum Reprod 2007; JCEM 2007

Angiopoietins 1 & 2

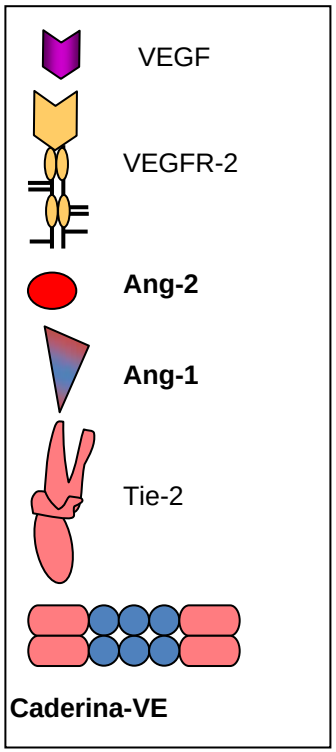
Growth factor family influencing endothelial cell regulation

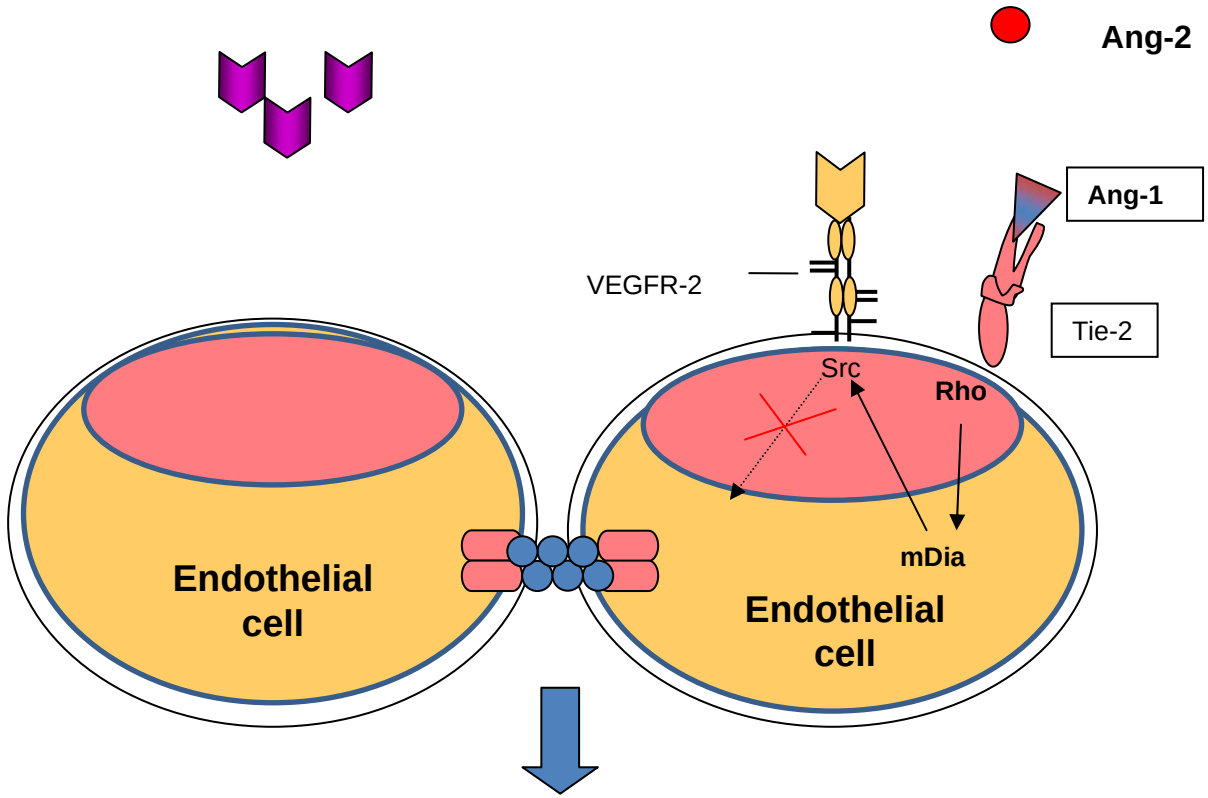
Both bind the same receptor (Tie-2)



Tesis María Cerrillo

Molskness et al. Fertil Steril 2006; Fraser H Reprod Biol Endocrinol 2006





Fluid leakage

Thomas et al, 2009, Hellmut et al 2009

Objectives

To investigate

- VEGF
- sVE-cadherin
- angiopoietin 2

modulation by hCG vs GnRHa in oocyte donors undergoing COH with antagonist protocols

Design

A. Pilot study

- 39 donors
- long agonist hCG vs antagonist GnRHa

B. Prospective, cohort study

- 64 donors
- antagonist hCG vs antagonist GnRHa

Differential regulation of VEGF after final oocyte maturation with GnRH agonist versus hCG: a rationale for OHSS reduction

- Prospective, proof-of-concept trial in egg donors
- Long + hCG vs antagonist-GnRHa

- No differences in # oocytes, IR or PR
- Shorter luteal phase and less subjective symptoms in donors that received GnRHa

TABLE 1

Plasma and follicular fluid VEGF concentration (pg/mL).

	hCG n = 19	GnRHa n = 20	*P* value
Plasma, day of hCG or GnRHa	158 ± 26 (44–326)	173 ± 23 (63–314)	NS
Plasma, day of egg retrieval	193 ± 28 (81–381)	219 ± 27 (81–411)	NS
Follicular fluid	1,666 ± 53 (1,267–1946)	1,207 ± 135 (436–2,117)	<.001

Note: Values are presented as mean ± SEM (95% confidence interval). GnRH = GnRH agonist.

Cerrillo. VEGF production after hCG or GnRH agonist. Fertil Steril 2008.

OHSS Prevention Antagonist Protocol

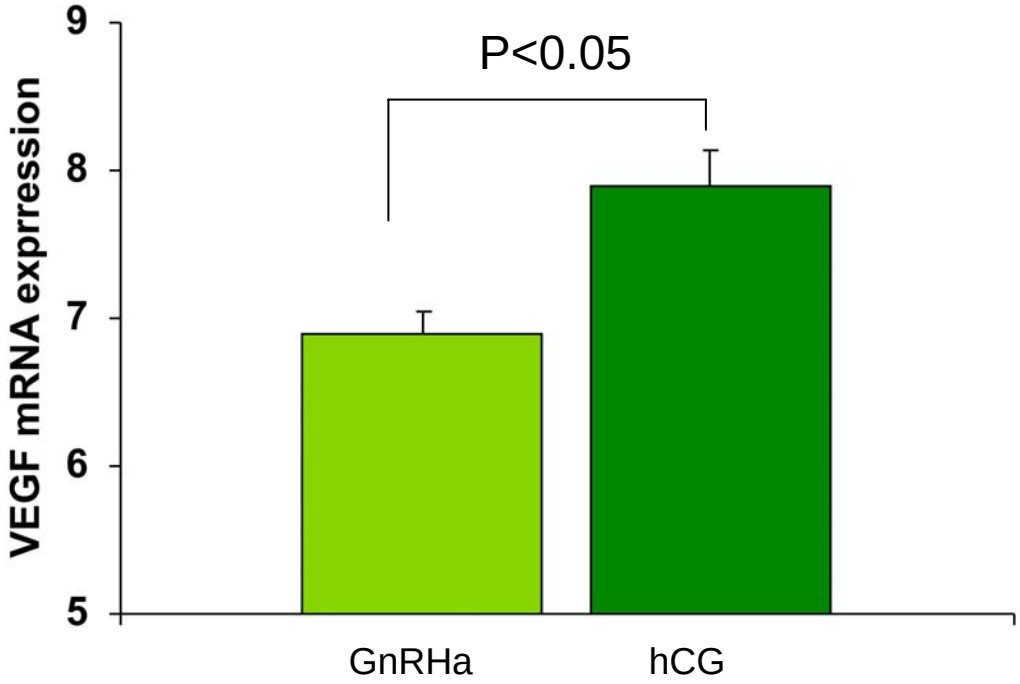
	hCG n:26	aGnRH n:30	P value
Age	25.4 ±4	23.8 ± 3.9	(0.22)
BMI (kg/m2)	21.5±2.9	22.04±2.34	(0.52)
Days stimulation	8.5±1.2	8.9±1.2	(0.29)
rFSH dose (IU)	1307±235	1377±234	(0.35)
Estradiol (pg/mL)	2537±1032	2213±979	(0.17)
Progesterone	0.78±0.5	0.82±0.33	(0.78)
# eggs	14±4.6	17±8	(0.12)
luteal phase (d)	10.2±1.1	5.2±1.65	**<0.001**
Symptoms	42% (8)	0	**<0.005**
Mod/sev OHSS (%)	0	0	NS

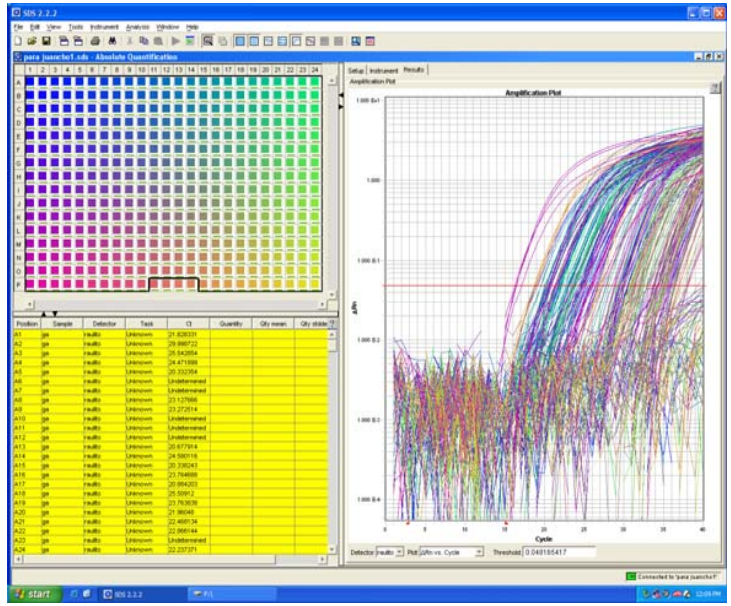
Cerrillo et al. Fertil Steril 2010

Vascular Mediators Regulation

	hCG n=26	GnRHa n=30	P value
VE Cadherin Serum (ng/mL)	0.32 ±0.13	0.34 ±0.10	NS
VE Cadherin FF	0.34 ±0.15	0.36 ±0.12	NS
Angiopoietin 2 Serum (pg/mL)	263 ± 85	460 ± 291	NS
Angiopoietin 2 FF	3341 ± 897	4713 ± 696	NS
VEGF Serum (pg/mL)	708 ± 332	594 ± 259	NS
VEGF FF	5094 ± 1280	3762 ± 1118	<0.001

VEGF mRNA expression





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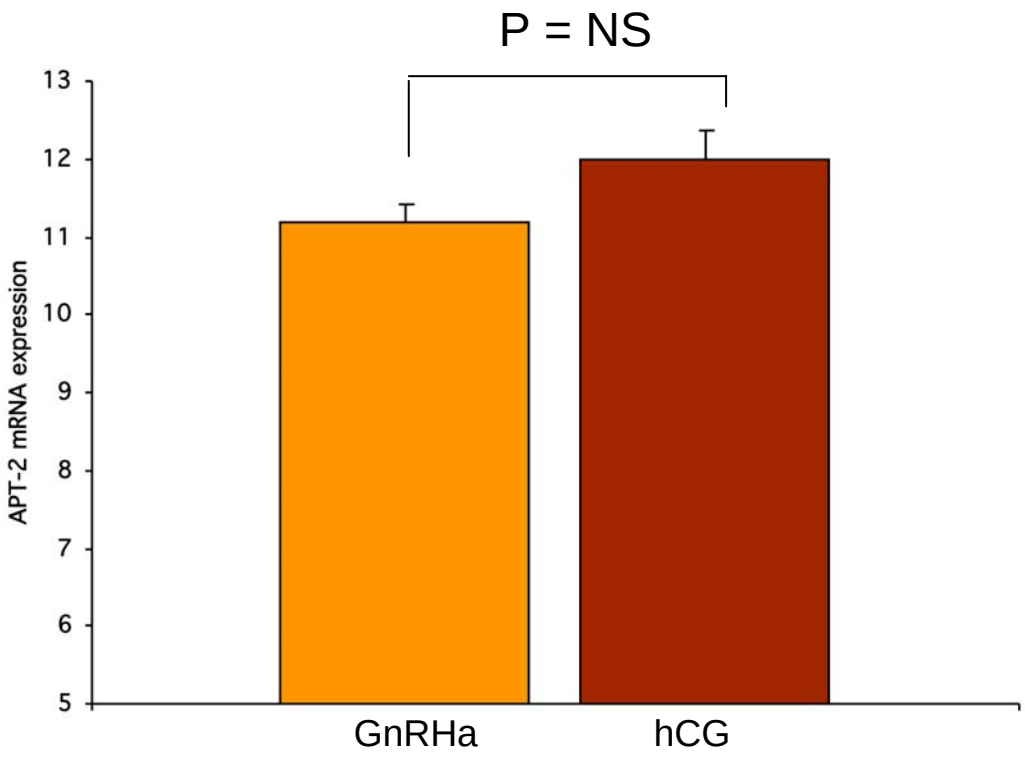
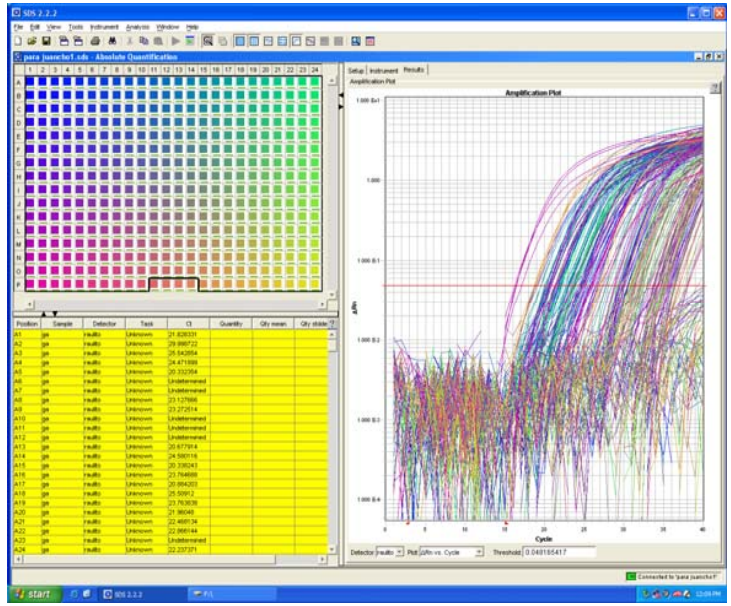
Angiopoietin-2 mRNA expression





Cerrillo et al. Fertil Steril 2010

Conclusions

- We have demonstrated a differential impact of steroids and low dose hCG on corpus luteum function
- Also, ovarian size, free-fluid and patient comfort are significantly different
- Triggering final oocyte maturation with GnRH agonists differentially regulates vascular mediators

Conclusions

- VEGF, rather VE-Cadherin and Angiopoietin 2, is significantly increased after HCG administration, contributing to increased vascular permeability

- The differential regulation of vascular proteins, such as VEGF, may explain the hypothetical benefits of protocols that avoid HCG to reduce OHSS incidence

Acknowledgments

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