Debate: Medical protection by GnRH agonists

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DISCLOSURE

- Unconditional grant from the Ipsen Pharmaceutical Group for the POF Intergroup Clinical trial (A prospective open randomized trial on the efficacy of gonadotropin-releasing hormone agonist depot –triptorelin- to prevent chemotherapy-induced premature ovarian failure for lymphoma)

>20 years later after …….

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Temporary Ovarian Suppression With Gonadotropin-Releasing Hormone Agonist During Chemotherapy for Fertility Preservation: Toward the End of the Debate?

LUCIA DEL MASTRO,[a] MATTEO LAMBERTINI[b]

The Oncologist 2015

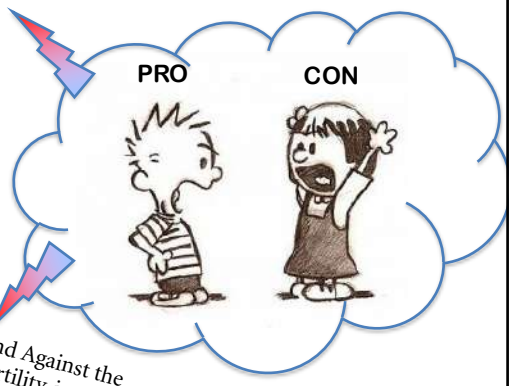
Judging the Fertility Protective Effect of GnRH Agonists in Chemotherapy—It Is a Matter of Perspective

Michael von Wolff and Petra Stute*

Frontiers in Endocrinology , 2017

Appraising the Biological Evidence for and Against the Utility of GnRHa for Preservation of Fertility in Patients With Cancer

Kutluk Oktay and Giuliano Bedoschi, New York Medical College and Innovation Institute for Fertility Preservation, New York, NY



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WHY?

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- What are the evidence regarding the mechanisms of ovarian protection of GnRHa?
- What really showed the clinical trials?

25/12/17

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WHY?

- What are the evidence regarding the mechanisms of ovarian protection of GnRHa?
- What really showed the clinical trials?

EXPERIMENTAL DATA

References	Species	Analogues	Main conclusion
Bokser 1990	Rat	Agonist	Protect mainly secondary follicles during Cy treatment
Montz 1991	Rat	Agonist (vs progesterone)	GnRHa as efficient as Prog to maintain fertility but not fecondity
Ataya 1995	Monkey	Agonist	Prevent Cy-induced follicular loss
Meirow, 2004	Mice	Antagonist	Prevent Cy-induced primordial follicular loss
Letterie, 2004	Rat	Agonist	No protection against Cy-induced follicular attrition
Yuce, 2004	Mice	Agonist	Partial protection from Cy inducing primordial follicular loss (Cy dose dependent)
Danforth, 2005	Mice	Agonist/antagonist	Agonist prevent Cy-induced primordial follicular loss but not antagonist (toxic effect)
Tan, 2010	Mice	Agonist	Dose-dependent protective effect of GnRHa on ovarian reserve against Cy
Lemos, 2010	Rat	Antagonist	No difference in total follicular density between CTL, Cy and Cy+Antago groups. Fertility protection
Zhao, 2010	Rat	Antagonist	Reduce Cy-induced apoptosis
Kishk, 2012	Mice	Agonist	Dose-dependent protective effect of GnRHa on ovarian reserve against Cy
Li, 2013	Rat	Agonist/antagonist	Prevent Cy-induced follicular loss
Parlakgumus, 2015	Rat	Agonist	No protection against Cy-induced follicular loss
Rossi, 2017	Mice	LH/FSH	LH and in a lesser extend FSH favored primordial follicles survival and DNA repair trough action on somatic cells when exposed to cisplatin

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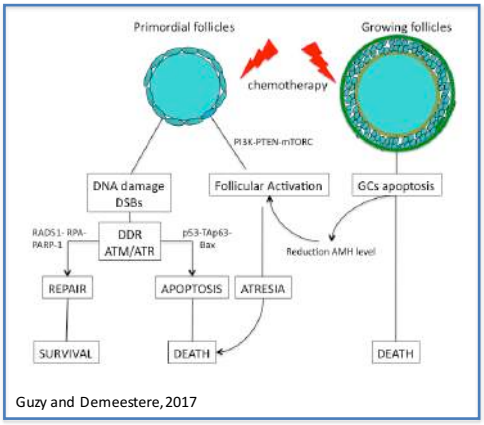
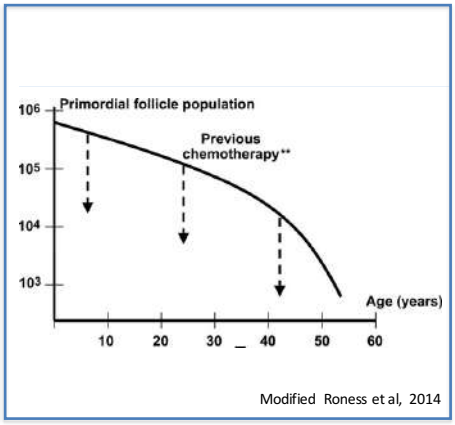
GONADOTOXICITY OF CHEMOTHERAPEUTIC AGENTS



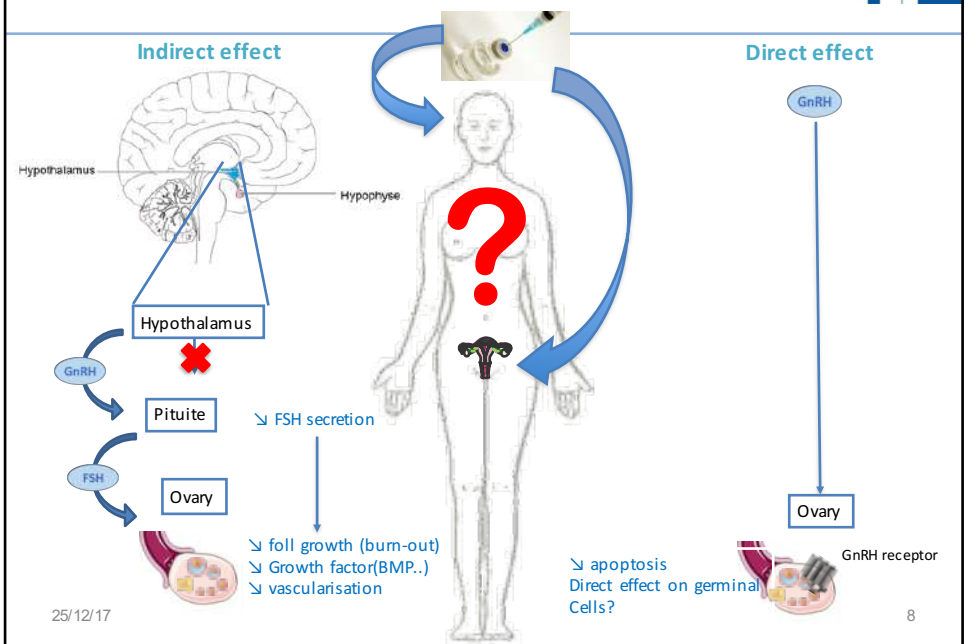
Guzy and Demeestere, 2017



Modified Roness et al, 2014

Infertility- POI
« Menopausal » Sd

Mechanisms of action of GnRH: Hypothesis



INVESTIGATION

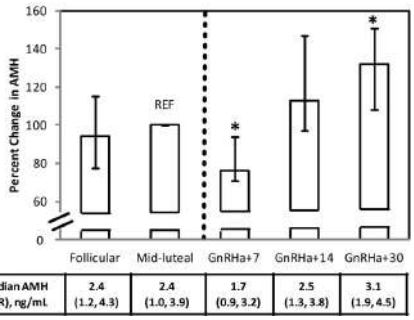
- **Question 1**: Does GnRHa prevent follicular recruitment?
- **Question 2**: Does inhibition of FSH indirectly protect the follicular pool?
- **Question 3**: Does GnRHa prevent follicular damage by directly acting on the ovary through GnRHa receptors?
- **Question 4**: Does GnRHa act through reduction of vascularisation?

QUESTION 1: DOES GnRHa PREVENT FOLLICULAR RECRUITMENT?

Administration of GnRHa alone did not reduce AMH level

Parameters	Baseline	4 weeks after Zoladex	8 weeks after Zoladex
FSH (IU/L)	7.1± 1.8	3.7± 2	3.5± 1.7
LH (IU/L)	4.5± 1.3	0.4± 0.3	0.4± 0.3
AMH (ng/ml)	-	4.5± 2.7	3.8± 2.3

Patients with stage II-IV endometriosis (n=21; 32 ± 5.8y)
Modified from Mohamed et al, Fertil Steril 2006



Median AMH (IQR), ng/mL	2.4 (1.2, 4.3)	2.4 (1.0, 3.9)	1.7 (0.9, 3.2)	2.5 (1.3, 3.8)	3.1 (1.9, 4.5)

Healthy women (n=33; 30.3 ± 8.5y)
Su et al, JCEM 2013

➤ Maintain the pool of growing follicles and follicular recruitment process

Inhibition of Ovarian function after GnRH treatment: Mice model

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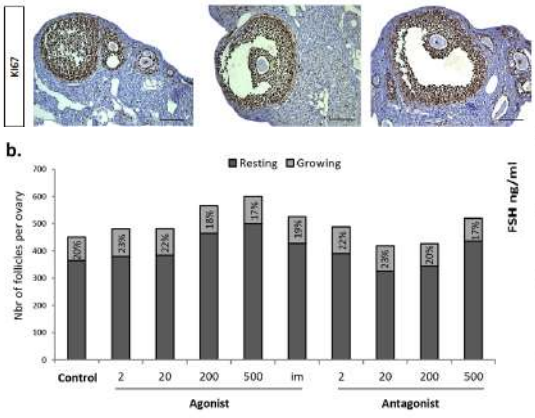
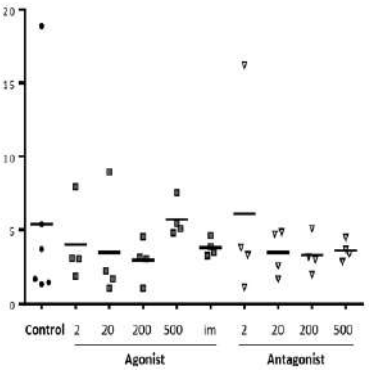
GnRHa Inhibited oestrus cycle but not follicular growth

Follicular growth after 2 weeks of GnRha treatment:



FSH levels after 2 weeks of GnRHa treatment:



Horicks et al, PlosOne 2015

Follicular depletion after treatment with cyclophosphamide

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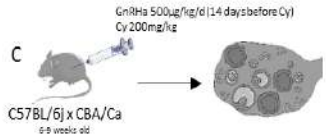


Table 2. Total follicular depletion, and proportion of resting and growing follicles population per ovary according to the treatment in mice.

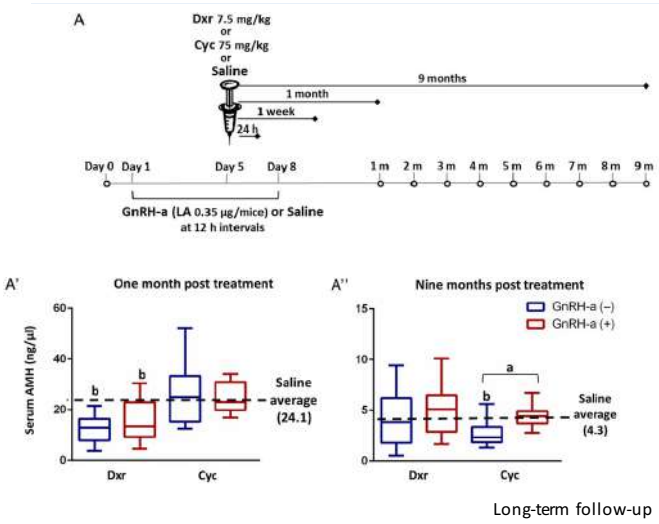
Treatment	N	Total follicular loss (%)	Distribution of follicles	
			Number of resting follicles (%)	Number of growing follicles (%)
Control	3	–	426.0±135.0 (81.05)	94.7±5.1 (18.95)
Cy	3	58,26*	133.7±41.1 (60.83) →	83.7±20.6 (39.17)
Ago	3	–	500.7±142.6 (83.50)	99.3±34.3 (16.50)
Ago + Cy	3	54,78*	173.7±48.0 (63.64) →	97.7±20.4 (36.36)
Ant	3	–	435.7±104.2 (83.30)	84.0±13.0 (16.70)
Ant + Cy	4	55,68*	139.0±23.43 (60.28) →	91.3±12.9 (39.72)

Cy single ip injection of 200 mg/kg; GnRH agonist (Ago) daily subcutaneous (sc) injection of 500 µg/kg; GnRH antagonist (Ant) daily sc injection of 500 µg/kg. Results are expressed as mean ± SD. N = number of ovaries analysed.
*p < 0.05 compared with controls.

➤Proportion of growing follicles increases during chemotherapy whatever the mice recieved GnRha or not

Horicks et al, PlosOne 2015

AMH levels after chemotherapy with or without GnRHa



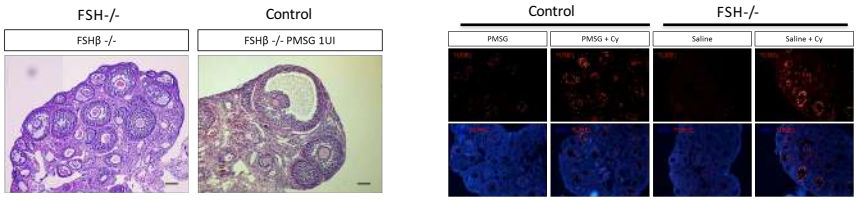
Long-term follow-up

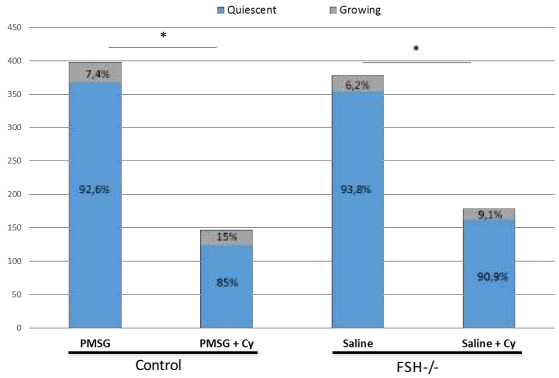
Hasky et al, Human Reprod, 2015

QUESTION 2:
DOES INHIBITION OF FSH INDIRECTLY PROTECT THE OVARIAN POOL?

- Maintain low FSH levels during chemotherapy
- Reduce secretion of growing factors by FSH-dependent large follicles
- Reduce the growth of secondary follicles more sensitive to chemotherapy.

FSH deficient mice model





Follicular loss: 62,9% 52,7%

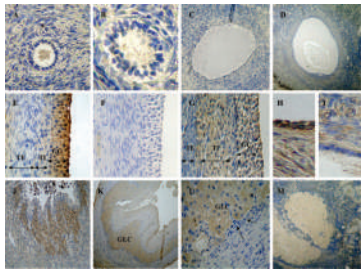
Horicks, et al submitted

QUESTION 3: DOES GnRHa PREVENT FOLLICULAR DAMAGE BY DIRECTLY ACTING ON THE OVARY THROUGH GnRHa RECEPTORS?

Role of GnRHR in the ovaries?

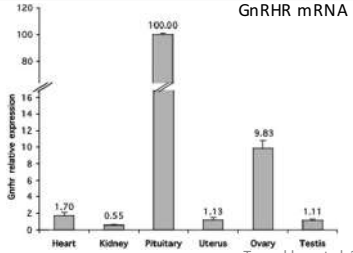
- Expressed mainly in antral follicles and CL→ Luteinization and apoptosis
- Protection from chemotherapy-induced granulosa cells damage *in vitro*

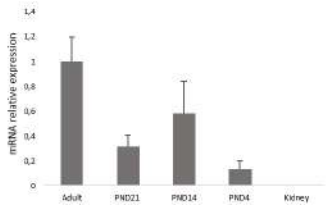
GnRHR expression in human ovaries



Choi et al, 2006

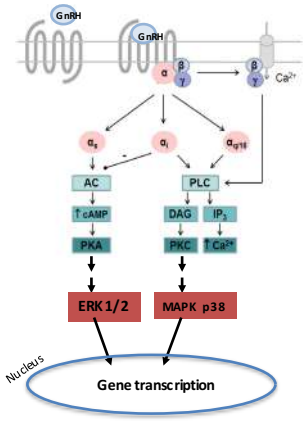
GnRHR mRNA expression in mouse ovaries





Torrealday et al, 2013

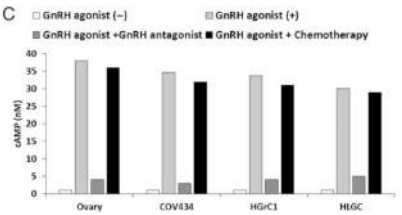
Signaling pathways ?



Extra-pituite tissue:

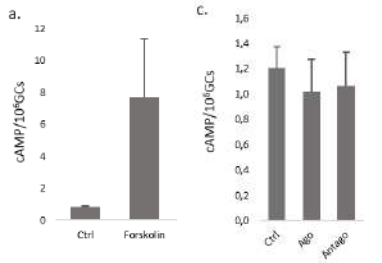
- lower affinity of the ligand
- Selective signaling cascade

cAMP levels in human ovarian tissue after GnRHa exposure



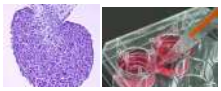
Bildik et al, 2015

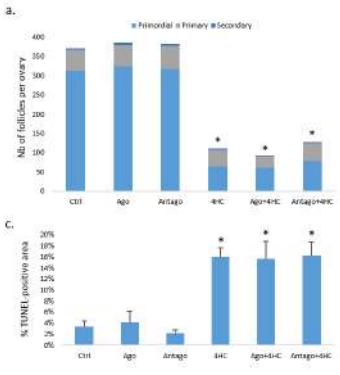
cAMP level inmouse GCs after GnRHa exposure



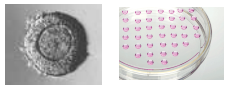
Direct effect of GnRHa on the ovary: Culture Model

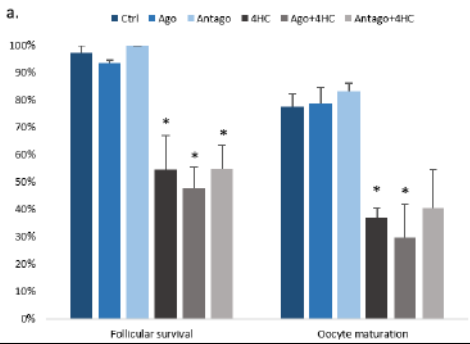
Quiescent follicles





Growing follicles





DIRECT EFFECT ON HUMAN OVARIAN TISSUE

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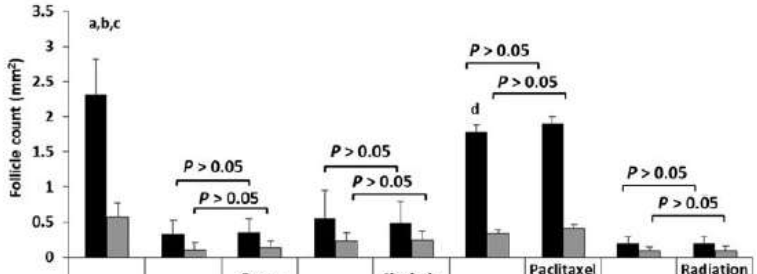
FOLLICLE RESERVE



	Control	Cyc	Cyc + GnRHa	Cisplatin	Cisplatin + GnRHa	Paclitaxel	Paclitaxel + GnRHa	Radiation	Radiation + GnRHa
Primordial	2.32 ± 0.5	0.33 ± 0.2	0.35 ± 0.2	0.55 ± 0.4	0.49 ± 0.2	1.79 ± 0.1	1.9 ± 0.1	0.2 ± 0.1	0.2 ± 0.1
Preantral-Antral	0.57 ± 0.2	0.11 ± 0.1	0.13 ± 0.1	0.23 ± 0.1	0.24 ± 0.1	0.34 ± 0.05	0.41 ± 0.05	0.1 ± 0.04	0.1 ± 0.05

Bildik et al, 2015

Question 4:
Does GnRHa act through reduction of vascularisation?

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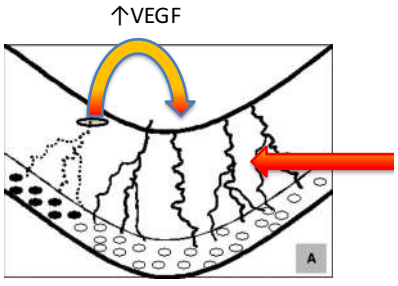
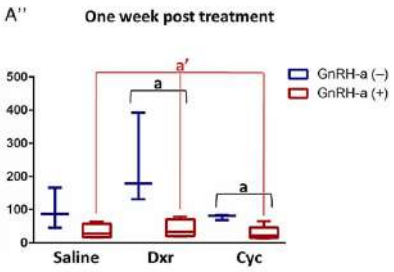
- Change in utero-ovarian flow principally results of the hypo-oestrogenism caused by GnRHa
- Direct effect on the ovary by decreasing VEGF secretion





Dada et al, 2001; Kitajima et al, 2006; Hasky et al, 2015; Meirow et al.2006

WHY?

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- What are the evidence regarding the mechanisms of ovarian protection of GnRHa?
- What really showed the clinical trials?

Ovarian protection in lymphoma patients

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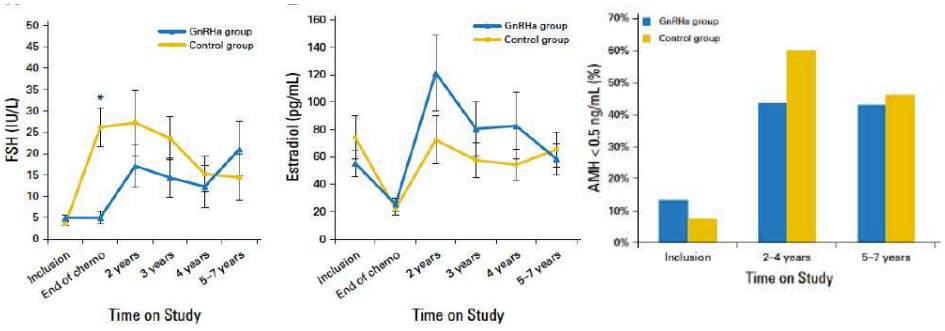
Prospective Randomized Controlled Trials

	Mean age (y)		N		FU		Outcomes and results
	Study	Control	Study	Control	Study	Control	
Waxman, 1987	28,5	25,9	8	10	2,3y	2y	No effect
Guisepe,2007	24,3	24,3	15	14	2,4y	5,9y	Protection (Menstruation)? No effect on ov. reserve
Behringer , 2010 (BEACOPP)	25,9	25,2	10	9 (OC)	≥1	≥1	No effect Amenorrhea Control 3/9 Treated 1/10 (1 unknown) Similar hormonal profile
Demeestere, 2012 Demeestere, 2016	25,6	27,2	45 32	39 35 (prog)	1 5	1 5	No effect POF rate 20% vs 19% AMH values in favor of GnRha after 1y (n=31) but not after 5y

POF TRIAL: RESULTS



Demeestere et al, JCO, 2016

Ovarian protection in Breast cancer patients

	PROMISE-GIM6 study [1]	POEMS-S0230 study [3]	OPTION Study [4]
Median age, years	39 (18-45y)	37.7 (18-49 y)	38.8 vs 37.9 (26 to 51y)
No. patients (ER pos/ER neg)	281 (226/51)	218 (0/218)	227 (95/126)
Primary end-point	no resumption of menses at 1y	Amenorrhea 6m and post-menopausal FSH levels (?) at 2y	Amenorrhea at 1-2 y
No. Patients eligible	269	135	202
Ovarian dysfunction (CT + LHRHa vs CT alone)	8.9 vs 25.9% OR = 0.28, P < .001 5-year cumulative incidence estimate of menstrual resumption was 72.6% in the LHRHa group and 64.0% in the control; age-adjusted HR, 1.48; *P* = .006. [2]	8% vs 22% stratified OR = 0.30, P = .04	22.1% vs 38.1% Amenorrhea 18.5% vs 34.8% POI (FSH>25IU/L)
Pregnancies (CT + LHRHa vs CT alone)	8 vs 3 age-adjusted HR = 2.40, P = .20	22 vs 12 adjusted OR = 2.45, P = .03	

[1]. Del Mastro L et al, JAMA 2011.
[2] Lambertini M et al, JAMA 2015.
[3] Moore HCF et al, N Engl J Med 2015
[4] Leonard et al, 2017

NO EFFECT ON THE OVARIAN RESERVE

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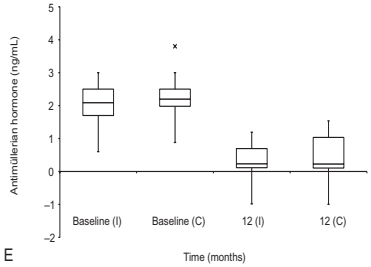
RCT breast cancer patients (18-40y)



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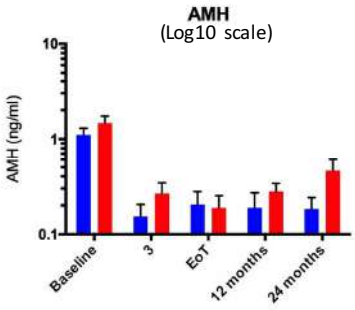
No effect on menstrual resumption and AMH levels

Elgindy et al, 2013

OPTION trial:
Reduction >95% in both groups at 2y



Leonard et al, 2017

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CONCLUSION

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- No evidence for the mechanism of action of GnRHa to prevent follicular depletion
- No evidence for a protective effect of GnRHa in young lymphoma patients.
- GnRHa analogues might be efficient and safe to improve ovarian function and fertility after chemotherapy in breast cancer patients but there is no evidence of a long-term benefit on the ovarian reserve
- Recent guidelines support GnRHa as a strategy to potentially preserve fertility in breast cancer patients but It should not replace gametes storage.

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CONCLUSION

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- Recent guidelines support GnRHa as a strategy to potentially preserve fertility in breast cancer patients but It should not replace gametes storage.

Belgian compromise?

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HYPOTHESIS

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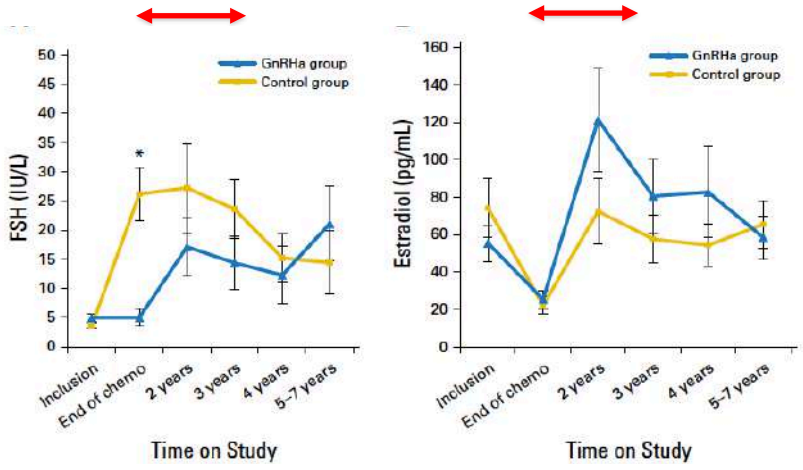
Favourable hormone environment after chemo to restore menstrual cycle more rapidly

« Window of opportunity » to get pregnant within 1-3 years after treatment



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ISFP

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