European Society of Human Reproduction and Embryology

Agonist triggering and freeze-all: oocytes or zygotes?

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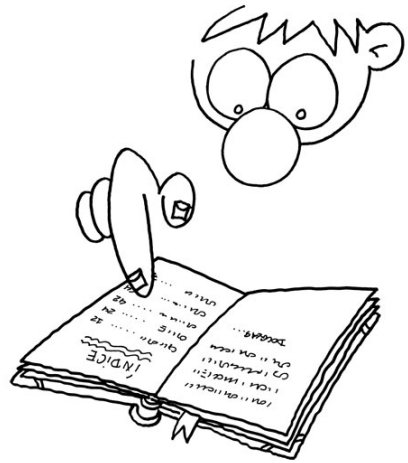
Summary



1. **OHSS** and how to prevent it.
2. **Cryopreservation**, definition and strategies
3. **Oocyte cryopreservation**
4. **Publication**s about zygote and embryo cryopreservation
5. A paper about **oocyte cryopreservation to prevent OHSS**
6. Conclusions.

Background

Ovarian hyperstimulation syndrome (OHSS):

Serious **complication derived of ovarian stimulation** in assisted reproduction patients

There are different **ways to prevent** OHSS:

- ✓ **cycle cancellation** before HCG administration *(Forman et al., 1990)*
- ✓ **coast** the patient *(García-Velasco et al., 2006)*
- ✓ **Embryo-zygote cryopreservation** *(Griesinger et al., 2007, 2010)*

A **new possibility** is to **cryopreserve the oocytes** obtained after triggering final oocyte maturation with GnRH agonists, **and transfer the embryos at a later stage**.

Prevention

- ✓ **Substitution of hCG** for a single **GnRH** agonist bolus is the **safest protocol** and avoids the frustration of cycle cancelling.

- ✓ Excellent **results obtained with egg or embryo vitrification** (*Kuwayama et al., 2005; Cobo et al., 2008)*

may allow a **combination** of these two approaches to avoid both early- and late-onset OHSS, while eliminating the need for adequate and specific luteal support.

Cryopreservation

"… Is a process where **cells or tissues are preserved by cooling** to low sub-zero temperatures, such as −196 °C (the boiling point of liquid nitrigen). At these low temperatures, **any biological activity**, including the biochemical reactions that would lead to cell deat, **is effectively stopped**"

Cryopreservation itself has always played
a **central role in ART**.

- ✓ Zygotes, cleavage stage embryos and blastocysts
- ✓ Increase **pregnancy chances** with frozen ET
- ✓ Increasing the **outcome**/stimulation cycle

Cryopreservation

Slow freezing	Vitrification
Conventional	Recently developed
Controlled cooling rate: 0.5ºC/min.	Cooling rate 15000-30000ºC/min.
Low CPA concentration	High CPA concentration
±0.5 ml volume	<1µl volume
Ice formation	NO Ice formation
Procedure duration: >90 min.	Procedure duration: 15min.
Easy work protocol	Training required

Oocyte vitrification

Oocyte cryopreservation **is an extremely useful technique in different clinical situations**

- ✓ **fertility preservation:** cancer patients or Postpone maternity
- ✓ **low responders**
- ✓ risk **of OHSS**
- ✓ **ethical reasons** as an alternative to embryo freezing
- ✓ **Donor** oocyte banking
- ✓ No **semen** available.

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Vitrification versus slow freezing of oocytes: effects on morphologic appearance, meiotic spindle configuration, and DNA damage

Publications…

Human Reproduction Vol.22, No.5 pp. 1348–1352, 2007
Advance Access publication February 15, 2007
doi:10.1093/humrep/dem006

Elective cryopreservation of all pronuclear oocytes after GnRH agonist triggering of final oocyte maturation in patients at risk of developing OHSS: a prospective, observational proof-of-concept study

G.Griesinger[1], S.von Otte, A.Schroer, A.K.Ludwig, K.Diedrich, S.Al-Hasani and A.Schultze-Mosgau

BACKGROUND: A bolus dose of GnRH agonist can substitute for hCG as a trigger for the resumption of meiosis in ovarian stimulation with GnRH antagonists, which has been suggested to reduce the risk of ovarian hyperstimulation syndrome (OHSS). As the efficacy of this measure in fresh embryo transfer (ET) cycles is unclear, we evaluated a new clinical concept of GnRH-agonist triggering. **METHODS:** In this prospective, observational proof-of-concept study, 20 patients considered at increased risk of developing OHSS (≥20 follicles ≥10 mm or estradiol ≥4000 pg/ml, or a history of cycle cancellation due to OHSS risk or the development of severe OHSS in a previous cycle) after ovarian stimulation and concomitant GnRH-antagonist administration had final oocyte maturation triggered with 0.2 mg triptorelin s.c. All two pronucleate (2 PN) oocytes were cryopreserved by vitrification, and frozen–thawed ETs (FT-ETs) were performed in an artificial cycle. Main outcome measures were the cumulative ongoing pregnancy rate per patient and the ongoing pregnancy rate per first ET. Secondary outcomes included the incidence of moderate-to-severe OHSS. **RESULTS:** Of the 20 patients triggered with GnRH agonist, 19 patients underwent 24 FT-ETs in the observational period. The cumulative ongoing pregnancy rate was 36.8% (95% confidence interval: 19.1–59.0%). The ongoing pregnancy rate per first FT-ET was 31.6% (15.4–54.0%). No cases of moderate or severe OHSS were observed. **CONCLUSIONS:** The present study is the proof of the concept that GnRH-agonist triggering of final oocyte maturation in combination with elective cryopreservation of 2 PN oocytes offers OHSS risk patients a good chance of pregnancy achievement, while reducing the risk of moderate and severe OHSS.

Publications…

Fertility and Sterility® Vol. 88, No. 3, September 2007

Triggering of final oocyte maturation with gonadotropin-releasing hormone agonist or human chorionic gonadotropin. Live birth after frozen-thawed embryo replacement cycles

Objective: To report the outcome of frozen-thawed embryo replacement cycles after GnRH-agonist triggering of final oocyte maturation in the collecting cycle with GnRH-antagonist.
Design: Prospective, observational, multicentric clinical study.
Setting: Tertiary university-affiliated IVF centers.
Patient(s): Patients under observation previously had been recruited into two concurrently performed, independent, randomized controlled trials (comparing hCG with GnRH-agonist for triggering final oocyte maturation in GnRH-antagonist multiple-dose protocols in normal responder patients) encompassing a total of 228 participants. Surplus embryos or oocytes at the pronuclear stage were cryopreserved in 53 patients after hCG administration and 32 patients after GnRH-agonist administration on the basis of patient choice, pronuclear/embryo availability, and local laws.
Intervention(s): Transfer of frozen-thawed embryos.
Main Outcome Measure(s): Live birth rate.
Result(s): Thirty-one and 23 patients after administration of hCG and GnRH-agonist, respectively, started a frozen-embryo replacement cycle by September 2005, with 25 and 16 patients eventually undergoing at least one frozen-thawed ET. Live birth rate per ET was 18.5% (95% confidence interval [CI], 8.2–36.7) and 30.0% (95% CI, 14.5–51.9) after hCG and GnRH-agonist triggering, respectively. Cumulative live birth rate per patient starting a frozen-embryo replacement cycle was 16.1% (95% CI, 7.1–32.6) and 26.1% (95% CI, 12.5–46.5) for hCG and GnRH-agonist, respectively.
Conclusion(s): The likelihood of live birth in frozen-embryo replacement cycles after GnRH-agonist triggering of final oocyte maturation does not appear to be impaired. (Fertil Steril® 2007;88:616–21. ©2007 by American Society for Reproductive Medicine.)

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Publications…

Human Reproduction Vol.20, No.10 pp. 2887–2892, 2005
Advance Access publication June 24, 2005.
doi:10.1093/humrep/dei150

A lower ongoing pregnancy rate can be expected when GnRH agonist is used for triggering final oocyte maturation instead of HCG in patients undergoing IVF with GnRH antagonists

E.M.Kolibianakis[1,3], A.Schultze-Mosgau[2], A.Schroer[2], A.van Steirteghem[1], P.Devroey[1], K.Diedrich[2] and G.Griesinger[2]

Centre 2

Patients	≤39 y. FSH D3 ≤12. ≤3 IVF. BMI 18-29.	
Stimulation protocol	FSH+GnRH antagonist	
Trigger of final oocyte maturation	10.000 IU hCG	0.2 triptorelin
N Frozen/thawed cycles	7/14	7/11
N embryo transfers	4	4
Positive pregnancy test	1/4 25%	0/4
Live birth	1/4 25%	0/4
Cumulative live birth	1/7 14.3%	0/7

Publications…

Human Reproduction Vol.20, No.5 pp. 1213–1220, 2005
Advance Access publication March 10, 2005
doi:10.1093/humrep/deh765

GnRH agonist (buserelin) or hCG for ovulation induction in GnRH antagonist IVF/ICSI cycles: a prospective randomized study

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Centre 1

Patients	25-40 y. FSH&LH D3 ≤12. BMI 18-30.	
Stimulation protocol	FSH+GnRH antagonist	
Trigger of final oocyte maturation	10.000 IU hCG	0.5 m. buserelin
N Frozen/thawed cycles	13/26	9/12
N embryo transfers	11	6
Positive pregnancy test	5/11 45.5%	3/6 50%
Live birth	4/11 36.4%	1/6 16.7%
Cumulative live birth	4/13 30.7%	1/9 11.1%

Publications…

Fertility and Sterility® Vol. 88, No. 3, September 2007

Triggering of final oocyte maturation with gonadotropin-releasing hormone agonist or human chorionic gonadotropin. Live birth after frozen-thawed embryo replacement cycles

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Trigger of final oocyte maturation	hCG	GnRH-agonist
N Frozen/thawed cycles	31/53	23/32
N embryo transfers	27/31	20/23
Positive pregnancy test/ET	7/27 25.29%	9/20 45%
Live birth	5/27 18.5%	6/20 30%
Cumulative live birth	5/31 16.1%	6/23 26.1%

Publications…

European Journal of Obstetrics & Gynecology and Reproductive Biology 149 (2010) 190–194

Cumulative live birth rates after GnRH-agonist triggering of final oocyte maturation in patients at risk of OHSS: A prospective, clinical cohort study

Georg Griesinger*, Henriette Berndt, Laura Schultz, Marion Depenbusch, Askan Schultze-Mosgau

ABSTRACT

Objectives: To prospectively study the incidence of OHSS, live birth likelihood and neonatal outcome after GnRH-agonist triggering of final oocyte maturation and vitrification of all pronucleate (2PN) oocytes for later frozen–thawed embryo transfer (FRET) in an OHSS-risk population.

Study design: Prospective, clinical cohort study (12/2004–5/2009). Forty patients undergoing ovarian stimulation in a GnRH-antagonist protocol and at risk of developing severe OHSS underwent triggering with 0.2 mg triptorelin and elective vitrification of all 2PN-oocytes for later frozen–thawed embryo transfer.

Results: The incidence of OHSS was 0% (0/40; 95% confidence interval: 0.0–6.4%). Thirty-nine patients underwent 87 FRETs (mean number of FRETs per patient: 2.2 ± 1.6; range: 1–7). The cumulative live birth rate per patient was 35.0% (14/40; 95% confidence interval: 23.9–48.0%). Mean time-to-conception resulting in live birth after agonist triggering was 24.2 (±17.1; range: 9–67) weeks. Nine healthy singletons and five twins were born.

Conclusions: A treatment algorithm combining agonist trigger with vitrification of all 2PN-oocytes is feasible and safe, and provides patients with a good cumulative chance of live birth.

Our paper…

The aim of our study is to present

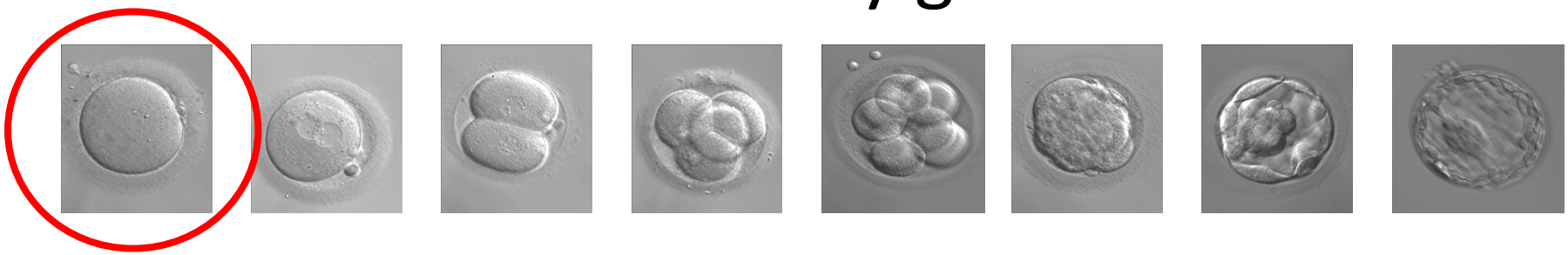
this **combined approach** of

GnRH agonist bolus to induce final oocyte maturation

and **egg vitrification** as

a new option for these patients at risk

of OHSS with very good results.



Materials and Methods

January 2009 to December 2009. IVI Madrid.

248 patients undergoing controlled ovarian stimulation for IVF show a **high response**:

- ✓Serum oestradiol ≥4000 pg/ml
- ✓≥20 mature follicles

Oocyte vitrification 1 hour after oocyte pick up

(in order to carry out microinjection and embryo transfer in a natural environment afterwards)

KITAZATO®

Fertility and Sterility® Vol. ■, No. ■, ■ 2010

Avoiding the use of human chorionic gonadotropin combined with oocyte vitrification and GnRH agonist triggering versus coasting: a new strategy to avoid ovarian hyperstimulation syndrome

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	Vitrified oocytes group	Coasted patients group	*p* value
N	96	152	
Transfer cancellation rate (N)	8.3% (8)	19.7% (30)	0.0151
Eggs retrieved (N)	20.4±3.2 (1958)	20.1±3. (3058)	0.5055
MII (rewarmed or obtained) (N)	10.7±4 (1026)	17.9±2.8 (2730)	<0.0001
Fertilization rate (N)	75.1 (570/857)	70% (1911/2730)	0.5628
Embryos transferred (N)	1.9±0.6 (168)	1.8±0.6 (224)	0.2023
Clinical pregnancy rate (N)	50% (44/88)	29.5% (36/122)	0.0012
Implantation rate (sacs/ET)	32.1% (54/168)	19.2% (47/224)	0.0209
Miscarriage rate (N)	9.1% (4/44)	11.1% (4/36)	0.6311

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Avoiding the use of human chorionic gonadotropin combined with oocyte vitrification and GnRH agonist triggering versus coasting: a new strategy to avoid ovarian hyperstimulation syndrome

	FET cycles belonging to cancelled transfers	**Vitrified oocytes group**	*p* value
N	28	96	
Transfer cancellation rate (N)	14.3% (4/28)	8.3% (8/96)	0.3442
Embryos transferred	1.8±0.6 (44)	1.9±0.6 (168)	0.4393
Clinical pregnancy rate (N)	41.6% (10/24)	50% (44/88)	0.4338
Implantation rate (sacs/TE)	25% (11/44)	32.1% (54/168)	0.4727
Twin pregnancy rate (N)	10% (1/10)	25% (11/44)	0.0897
Miscarriage rate (N)	10% (1/10)	9.1% (4/44)	0.8853

Conclusions

Oocyte vitrification after triggering with GnRH agonists **is a highly attractive, safe, and efficient alternative** to postpone embryo transfer **in patients at high risk of OHSS**, avoiding the risk for the patient.

Thanks
for your attention

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