

Oocyte banking: Does the period of storage affect ART outcomes?

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Introduction: Oocyte cryopreservation may be a valuable alternative to preserve fertility. According to our knowledge, there are no descriptions about the effect of the time of storage on oocyte performance after thawing. The aim of our report is to evaluate if the period of oocyte storage impact ART outcomes.

Materials: 124 eggs from 10 patients were thawed to perform an ICSI procedure. The oocytes were cryopreserved and thawed according to the protocol 1,2-propanediol-sucrose slow freeze-rapid thaw. Duration of storage, survival rate, fertilization rate, embryo cleavage rate, implantation rate and ongoing pregnancy rate were analyzed.

Results: Survival rate was 39%, Fertilization rate was 48%, cleavage rate was 83%, implantation rate 15,7% and pregnancy rate was 30%.

2 out of the 3 pregnancies (66%), were from the longest stored eggs (2 years storage). The third one had been stored for 3 months.

Conclusions: Even when oocyte banking is a novel experience and there are still a low number of thawed cases, duration of storage seems to have no relation with the pregnancy outcome.

This finding should encourage women who can not bear children immediately to apply for oocyte storage.

Keywords: oocyte ;cryopreservation ;storage; banking.

1. Introduction

The pace of modern day life has resulted in considerable changes. One of these has been the total insertion of the professional and non-professional woman in all areas of society. This situation has brought with it the postponement of events in women's personal lives, such as the forming of a couple and maternity. The average age of first-time mothers in the first world and the developing world has changed and this event now occurs around the fourth decade of their lives.

As available data shows that pregnancy rate decreases and genetic abnormalities in the conceptus increases after 35 years, can we offer these women who are over thirty and have neither a stable partner nor any desire for fertility at the present time, the possibility of putting their fertility into storage?

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The idea of oocyte cryopreservation and storage arose from the desire to reduce the number of frozen embryos and offer the freezing of one cell which brings with it less ethical and legal conflicts. With the passing of the years freezing and thawing techniques have progressed with regard to the survival rates because of changes in the freezing process: an increase in the sucrose concentration and the exposition times of the ovum during the process[1], the freezing with and without cumulus) [2], and changes in the sodium concentration in the freezing medium[3]. With the arrival of ICSI, the fertilization rates[4,5], improved and by-passed the problems caused by the hardening of the zona[6]and the premature releasing of cortical granule [7]..

According to our knowledge, there are no descriptions about the effect of the time of storage on oocyte performance after thawing. The aim of our report is to evaluate if the period of oocyte storage impact ART outcomes.

2. Materials and methods /Patients:

Twenty five patients who desired to preserve their future fertility for social and religious reasons and one patient who failed to obtain the sperm sample on the procedure's day cryopreserved 239 oocytes according to the protocol 1,2-propanediol –sucrose using a slow freeze- rapid thawing programme. Oocytes were transferred to human tubal fluid (HTF) supplemented with 0.5% human serum albumin (Irvine Scientific, Santa Ana, CA) at 37°C in an atmosphere of 5% CO₂ in air. Oocytes were frozen 5–6 hours after recovery. Following preequilibration of the oocytes for ten minutes in 1.5M 1,2-propanediol alone and transfer to 1.5M 1,2-propanediol containing 0.1M sucrose, the oocytes were immediately loaded into plastic straws and placed in a Planer Kryo 10 biological freezer (Planer Biomed, Sudbury, Middlesex, U.K.) and then cooled from 16°C to -8°C. Ice nucleation was induced manually by seeding each straw with precooled forceps. Gradual temperature reduction was continued to -30°C followed by rapid reduction to -150°C. Oocytes were stored in liquid nitrogen at -196°C prior to thawing.

One hundred and twenty four oocytes from ten patients (mean age 30.5 ± 6) were thaw. The straws were removed from liquid nitrogen, held at room temperature for 30 seconds, and transferred into a 30°C water bath for 40 seconds by which time complete thawing had occurred. The cryoprotectant was removed by transferring the oocytes through decreasing concentrations of propanediol solution (1.5– 0.5 M in each) containing 0.2M sucrose, followed by a final step dilution of 0.2M sucrose alone. Oocytes were then transferred into fresh culture medium, and after an incubation period of 1 hour ICSI procedure was done.

Different parameters were analyzed : duration of storage, survival rate, fertilization rate, embryo cleavage rate, implantation rate and ongoing pregnancy rate.

3. Results:

The period of time that the oocytes were stored varied from 3 months to 2 years.

124 oocytes from 10 patients were thawed ,58 were cryopreserved with cumulus corona complex and 66 without cumulus corona complex.

48 out of 124 thawed oocytes survived (39%). In all the cycles we had at least two oocytes which survived thawing with subsequent insemination.48 oocytes underwent ICSI procedure ,23 out of 48 had 2 pronuclei (fertilization rate 48%).The number of oocytes damaged was 6 (12.5 %). 19 out of 23 embryos cleaved (cleavage rate 83%)and were transferred (mean embryos transf.2.1±1.2). All cases had embryo transfer. Implantation rate was 15.7% and pregnancy rate 30%. There were no miscarriages so take home baby rate was 30%.

Three healthy babies, one female and two males, were born with a birth weight of 2,800 grams, 3,000 grams and 2700 grams respectively. All deliveries were cesarian sections and were performed at gestational ages of 36th, 38th and 37th weeks. There were no malformations in the newborn babies. They underwent until now a normal psychometric and neurological development.

2 out of the 3 pregnancies (66%), were from the longest stored eggs (2 years storage). The third one had been stored for 3 months.as shown inTable 1.

Table 1
Oocyte banking:Period of storage and pregnancies.

Cycle no.	Period of storage (months)	Pregnancy
1	5	No
2	21	Singleton
3	3	No
4	6	No
5	3	No
6	4	No
7	23	Singleton
8	6	No
9	5	No
10	3	Singleton

4.Discussion :

Even when oocyte banking is a novel experience and there are still a low number of thawed cases, duration of storage seems to have no relation with the pregnancy outcome as it was observed in embryo cryopreservation.

Our data showed cleavage, implantation and pregnancy rates acceptable for oocyte cryopreservation programs, with neither pregnancy loss nor malformations. 2 out of the 3 pregnancies (66%), were from the longest stored eggs (2 years storage). The third one had been stored for 3 month.

This findings should encourage women who can not bear children immediately for social, ethical and also for a variety of medical reasons such as cancer [8] to apply for oocyte storage, no matter how long it may take them to use their eggs.

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