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FERTILITY PRESERVATION

Live birth rate after use of cryopreserved oocytes or embryos at the time of cancer diagnosis in female survivors: a retrospective study of ten years of experience

Anne Mayeur[1] · Vincent Puy[1,2] · Victoria Windal[1] · Laetitia Hesters[1] · Vanessa Gallot[3] · Alexandra Benoit[3] · Michael Grynberg[3] · Charlotte Sonigo[3] · Nelly Frydman[1,2]

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Abstract

Purpose The aim of this study was to evaluate the outcomes of frozen oocytes or embryos cryopreserved after controlled ovarian stimulation (COS) or in vitro maturation (IVM) for female cancer patients who underwent a fertility preservation (FP) prior to gonadotoxic therapy.

Methods A retrospective cohort study from 2009 to December 2017 was conducted. Among the 667 female cancer patients who underwent oocytes or embryos cryopreservation for FP, 40 (6%) have returned to the fertility clinic between 2011 and 2019 to use their frozen material after being cured. We compared these thaw cycles outcomes according to the techniques used at the time of cryopreservation.

Results Among the 40 women cancer survivors who used their cryopreserved material, thirty patients have benefited from at least one embryo transfer. Ten patients did not have an embryo transfer since the oocytes did not survive after the thawing process or because no embryo was obtained after fertilization. We related three live births following FP using IVM (two from frozen oocytes and one after embryo cryopreservation). Five live births were obtained when COS was performed at the time of FP (one from frozen oocytes and four after embryo cryopreservation).

Conclusions Our preliminary results, although they are obtained in a small sample, are encouraging and show that different FP techniques can be used in female cancer patients and lead to live births. IVM is one of the options available that does not delay the start of chemotherapy or if ovarian stimulation using gonadotropins is contraindicated.

Keywords Oncofertility preservation · Oocyte cryopreservation · Zygote cryopreservation · In vitro maturation · Live birth

Introduction

Thirteen percent of newly diagnosed cancers occur in women of reproductive age [1]. Breast cancer is the most common of them. According to cancer statistics in France, 2344 women under the age of 40 were diagnosed with breast cancer in 2012 [2]. Over the past few decades, diagnosis and treatment have both improved, leading to better long-term survival [3]. However, most cancer treatments jeopardize fertility due to chemotherapeutic agent exposure and pelvic radiotherapy [4]. The field of fertility preservation (FP) has grown during the last two decades, driven by the increasing recognition of potential fertility loss [5, 6]. Preserving female fertility before any gonadotoxic treatment is important to increase the chance of motherhood after curative therapies have ended, without having to resort to egg donation [7, 8].

Charlotte Sonigo and Nelly Frydman contributed equally to this work.

✉ Anne Mayeur
anne.mayeur@aphp.fr

[1] Reproductive Biology Unit CECOS, Antoine Béclère Hospital APHP, Paris-Saclay University, 157 rue de la porte de Trivaux, Clamart, 92140 Paris, France

[2] Laboratory of Development of the Gonads, UMRE008 Genetic Stability Stem Cells and Radiation, Paris University, Paris-Saclay University, CEA, F-92265 Fontenay-aux-Roses, Paris, France

[3] Department of Reproductive Medicine and Fertility Preservation, Antoine Béclère Hospital, APHP, Paris-Saclay University, Clamart, 92140 Paris, France

Current options proposed by oncologists and reproductive specialists include cryopreservation of oocytes, embryos, and/or ovarian tissue. Decisions about FP strategy are made according to patient's medical data and cancer characteristics. The patient's age, relationship status, personal wishes, and ovarian reserve parameters as determined by antral follicle count (AFC) and anti-Mullerian hormone (AMH) serum level are taken into account for FP decision. Moreover, the cancer type and its systemic impact (estrogen-sensitivity, hemorrhagic risk …), the proposed treatment (dose, duration, and potential gonadotoxicity), and the timeframe available before beginning cancer treatment are also evaluated to decide which of the FP strategy to use [9]. According to the American Society of Reproductive Medicine (ASRM), embryo or oocyte cryopreservation after controlled ovarian stimulation (COS) is the best established method to preserve female fertility [10]. For many year, embryo cryopreservation was the only established clinical method for FP of adult women. No more than 80 live births have been reported after an embryo thaw cycle for patients cured of cancer [11–26]. A paradigm shift occurred in 2013 when cryopreservation of mature oocytes through vitrification showed proof of its efficacy in egg banking programs. These facts resulted in an international consensus in 2013 to recognize oocyte cryopreservation after COS as a clinically established method for female FP [27, 28]. To date, only 38 live births have been reported after oocyte vitrified/warmed cycles in cancer patients after COS [11–13, 29–33].

When COS is not feasible (estrogen-sensitive tumors and/or if neoadjuvant chemotherapy is scheduled) or in urgent FP situations [34], the combination of immature-oocyte harvest from the ovaries followed by in vitro maturation (IVM) treatment in the laboratory [35, 36] can be performed. This FP solution does not require ovarian stimulation with gonadotropins prior to oocyte retrieval (OR) and can be carried out urgently if needed, whatever the phase of the menstrual cycle. To date, one live birth has been published after a frozen-thaw embryo transfer cryopreserved after IVM for a cancer patient [12]. We recently published the first live birth after cryopreservation of in vitro matured oocytes in a cancer patient [37]. However, this procedure is still regarded as an experimental FP procedure.

In this sense, it is important that patients and professionals be informed about the wider range of preservation options and their respective outcomes regarding the use of cryopreserved oocytes or embryos in cured cancer patients. The objective of this study is to report the reproductive outcomes of 40 female cancer survivors, who came back to use their oocytes or embryos cryopreserved after IVM or COS at the time of FP.

Materials and methods

Population studied

Our study population consisted of 40 patients cured from cancer who asked to use their oocytes or embryos cryopreserved at the time of FP. The cryopreservation techniques were performed between January 2009 and December 2017. The thawing cycles (n = 49) occurred between January 2011 and December 2019. This studied population represents a small part (6%) of cancer patients who underwent our FP program before a gonadotoxic treatment (667 patients, 275 COS cycles, and 420 IVM cycles). Hence, 627 patients were excluded from the study because they had not yet thawed their cryopreserved material at the time of the study.

Among these 40 patients, thirty-two returned after being cured from breast cancer, three from Hodgkin's lymphoma, one from acute lymphoblastic leukemia, one from MALT lymphoma, one from idiopathic medullary hypoplasia, and two from ovarian borderline tumors. All patients had the approval from their oncologist to start a pregnancy.

We divided this population into four groups according to FP technique performed: oocyte or zygote cryopreservation after COS (OO-COS and ZYG-COS) or after IVM (OO-IVM and ZYG-IVM) (Fig. 1).

Written informed consent was obtained from each patient or couple for the use of their medical data for publication. The database was approved by the appropriate French authority (Commission Nationale de l'Informatique et des Libertés, CNIL no. 1217921) on February 21st, 2007. According to the "Jardé Law" (decree no. 2016-1537, November 16, 2016), institutional review board approval was not required for this retrospective study.

Fertility preservation technique

When a patient was referred to our FP center, clinicians provided information regarding the impacts of cancer and their treatments on reproductive function and fertility. During the consultation, the clinician presented the FP options after assessment of serum AMH, progesterone levels, and antral follicle count (AFC) by ultrasound examination performed using a 3.7–9.3 MHz multi-frequency transvaginal probe (RIC5-9H, Voluson 730 Expert, General Electric Medical Systems, Paris, France). When oncologists contraindicated COS in estrogen-sensitive tumors or if neoadjuvant chemotherapy was required, an IVM cycle without ovarian stimulation was advised.

Controlled ovarian stimulation protocols

The COS protocol was performed using a gonadotropin-releasing hormone (GnRH) antagonist protocol, with the

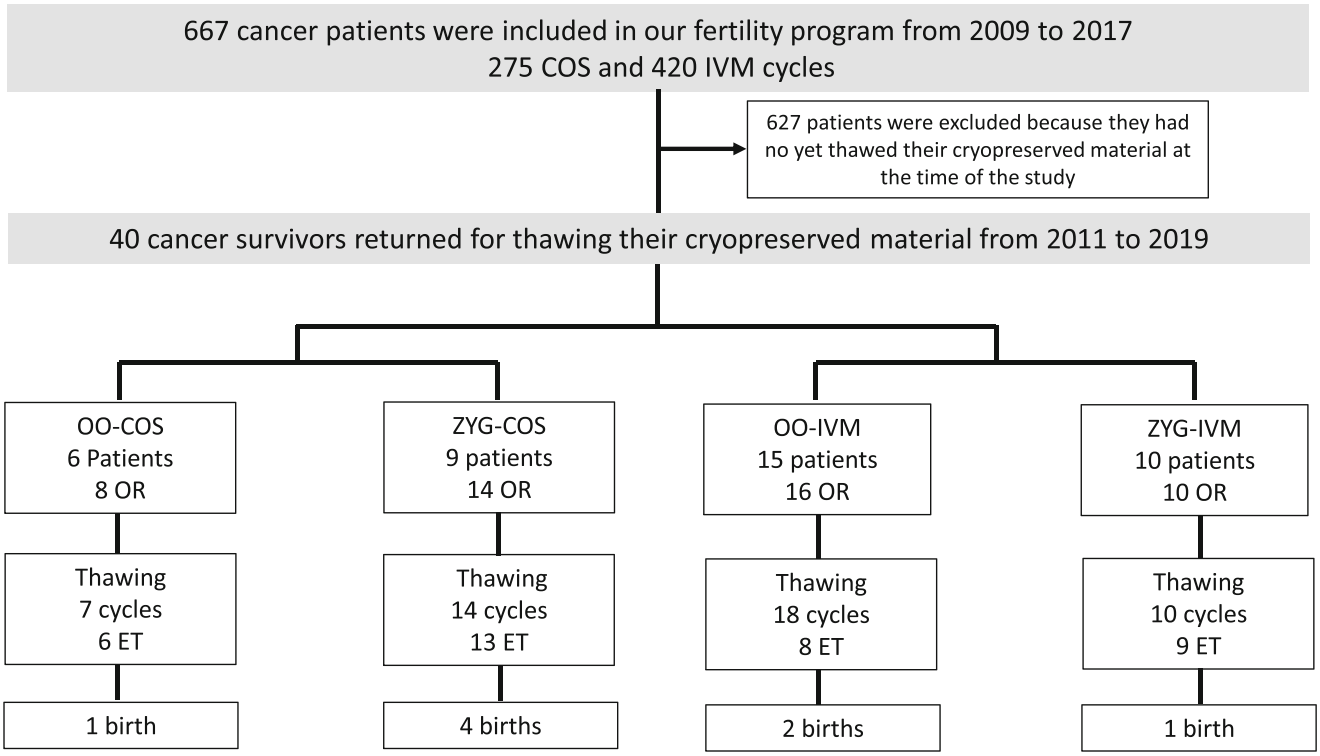

Fig. 1 Flowchart of the study; 667 female patients underwent our FP program before a gonadotoxic treatment from January 2009 to October 2017. Forty patients cured from cancer asked for the use of oocytes or embryos cryopreserved in this context. We divided this population into four groups according to FP technique performed: oocyte or zygote cryopreservation after COS (OO-COS and ZYG-COS) or after IVM (OO-IVM and ZYG-IVM). COS, controlled ovarian stimulation; IVM, in vitro maturation; OO-COS, oocytes cryopreserved after controlled ovarian stimulation; OO-IVM, oocytes cryopreserved after in vitro maturation; ZYG-COS, zygotes cryopreserved after controlled ovarian stimulation; ZYG-IVM, zygotes cryopreserved after in vitro maturation

administration of recombinant follicle-stimulating hormone (recFSH; Gonal-F®, Merck-Serono Pharmaceuticals, France) and a GnRH antagonist (Orgalutran®, 0.25 mg, S.C., MSD Pharmaceuticals, France). COS protocols were adjusted according to the menstrual cycle phase [38]. For six patients diagnosed with an estrogen-sensitive breast cancer, an aromatase inhibitor was associated during COS (Letrozole® 5 mg/day; Teva, Paris, France). In all patients, final oocyte maturation was achieved using either a recombinant human chorionic gonadotropin (hCG; Ovitrelle®, Merck-Serono, 0.25 mg, S.C.) or a GnRH agonist (Decapeptyl®, Ipsen Pharma, Paris, France, 0.2 mg, S.C.). Oocyte retrieval under moderate sedation was performed 36 h after ovulation triggering, using a 19-gauge needle (K-OPS-7035; Cook Medical Products, France) guided by vaginal ultrasound. The aspiration vacuum was fixed at 150 mmHg. Follicular fluid was then analyzed in Nucleon™ culture dishes (Nunc A/S, Denmark), where cumulus–oocyte complexes (COCs) were isolated and washed with culture medium (Universal IVF Medium, Origio, Versailles, France). Oocytes were denuded with a hyaluronidase solution (SynVitro Hyadase, Origio, Versailles, France), and nuclear oocyte maturation was assessed. Depending on the patient's choice, metaphase II oocytes were either frozen or fertilized by intracytoplasmic sperm injection (ICSI).

In vitro maturation

When IVM was required, management was carried out as quickly as possible regardless of follicular or luteal phases. No FSH was administered for follicular growth support. A 10,000 UI of hCG priming (Organon Pharma, Saint-Denis, France) was performed 36 h before oocyte retrieval to promote in vitro oocyte maturation [39]. Transvaginal ultrasonographically guided oocyte collection was done using a specially designed 19-gauge single-lumen aspiration needle (K-OPS-7035-Wood; Cook, France). The aspiration pressure was set at 7.5 kPa. Follicular aspirates containing cumulus–oocyte complexes were collected in 15 ml Nucleone (Nunc A/S, Denmark) tubes containing 3 ml of prewarmed sodium heparinate 2 IU/ml (Sanofi-Synthelabo, France).

Cumulus–oocyte complexes (COCs) were washed and then placed in IVM media (Origio, Versailles, France) enriched with 20% inactivated maternal serum, FSH (0.75 UI/ml), and luteinizing hormone (LH, Menopur®, Ferring, Germany, 0.75 UI/ml) [40]. After 24 to 48 h, COCs were

denuded with hyaluronidase solution (SynVitro Hyadase, Origio, Versailles, France), and nuclear oocyte maturation was assessed. Depending on the patient's choice, metaphase II oocytes were either frozen or fertilized by ICSI.

Oocyte/zygote cryopreservation

Metaphase II oocytes (MII) after COS or IVM were either directly cryopreserved (oocyte cryopreservation) or fertilized by ICSI (embryo cryopreservation). Fertilization was checked 17 h after ICSI, and embryos were frozen at the two pronuclear stages (zygote stage).

The vitrification method was set up in our IVF department in 2013. Prior to this date, oocytes or zygotes were frozen using a slow-cooling method as described elsewhere [33]. Mature oocytes and/or zygotes were vitrified using a RapidVit™ Blast kit (Vitrolife, Vastra Frolunda, Sweden) according to the manufacturer's instructions. A maximum of 2 oocytes or zygotes were loaded in one Rapid-i device and placed in high security straws cooled in liquid nitrogen.

Cryopreserved oocyte or embryo utilization

When patients came back to our center to ask for utilization of their cryopreserved oocytes/embryos, the approval for pregnancy from their oncologists was systematically required.

Oocyte/zygote thawing

Warming of oocyte(s) and zygote(s) was performed using a RapidWarm™ Blast Kit (Vitrolife, Vastra Frolunda, Sweden) for both slow-cooling and vitrification techniques.

Embryo culture and transfer

After oocyte or zygote thawing, embryos were cultured in a controlled atmosphere. The day after embryo thawing or 48 h after oocyte injection (day 2 of embryo development), embryo quality was assessed according to the consensus on embryo assessment [41] with respect to cell number, percentage of fragmentation (< 10%; 10–25%; > 25%), multi-nucleation, and cell-specific stage size. Embryos with 4–5 mononucleated blastomeres, a stage-specific cell size, and a fragmentation < 10% were given an A score (good quality embryos). Day 2 embryos with the best scores were transferred under ultrasound guidance (Voluson 730 Expert; General Electric Medical Systems, Paris, France) using a classic Frydman catheter (CCD Laboratories, Paris, France).

Frozen-thawed embryo transfer outcomes

A pregnancy test was systematically performed 12 days after embryo transfer. Clinical pregnancy was defined as the presence of a gestational sac with a fetal heartbeat by ultrasound examination at seven gestational weeks. The implantation rate was calculated by dividing the number of gestational sacs with a fetal heartbeat by the number of embryos transferred.

Endometrium preparation

Endometrium was prepared with an artificial cycle regimen using estrogen priming at a dose of 4 mg daily (Provames® 2 mg, France) on days 1–3 of the menstrual cycle. Progesterone treatment (micronized progesterone vaginally Progestan® 800 mg, Besins, France) was added when endometrial thickness reached 7 mm or more. This treatment was continued until the pregnancy test and until 11 weeks of gestation in case of pregnancy.

Statistical analysis

Data collected were analyzed using GraphPad Prism® software (version 6.0; GraphPad Software Inc., San Diego, CA) and were expressed as medians interquartile range (25th percentile–75th percentile). To compare unpaired values between two groups, we used either a Student's *t*-test when distributions passed the normality test or the Mann–Whitney test if distributions were not normal. To compare proportions between groups, according to sample size, either χ^2 or Fisher's exact ($n < 5$) tests were performed. The threshold for statistical significance was set to $p < 0.05$.

Results

Fertility preservation cycles in female cancer survivors

Patient characteristics and fertility preservation cycle outcomes are presented in Table 1.

No significant difference in patient age, ovarian reserve, and AFC ranges was observed between the four groups divided according to the technique used at the time of FP (OO-COS, ZYG-COS, OO-IVM, ZYG-IVM).

When analyzing data between patients who underwent COS on the one hand (OO-COS and ZYG-COS) versus IVM on the other hand (OO-IVM and ZYG-IVM), no difference in the total number of retrieved oocytes (8.0 [5.7–10.7] and 6.5 [4.0–9.0], $p = 0.34$) and mature oocytes (6.0 [4.7–8.5] and 5.0 [3.0–7.0], $p = 0.14$) was observed.

For IVM cycles, oocyte retrieval was performed during the follicular phase for half of the patients (13 cycles) and the other half (13 cycles) during the luteal phase. The number of COCs recovered was comparable between follicular and luteal phases (5.0 [4.0–8.0] vs 9.0 [4.5–12.0], $p = 0.21$). Maturation rate after oocyte retrieval during follicular phase was 74.7%

Table 1 Patient characteristics and fertility preservation cycle outcomes according to type of cryopreservation method used

	COS-FP			IVM-FP		
	OO-COS	ZYG-COS	All COS-FP	OO-IVM	ZYG-IVM	All IVM-FP
Female patients	6	9	15	15	10	25
Oocyte retrievals	8	14	22	16	10	26
Age (years)	35.5 [28.7–38.25]	36.0 [31.0–39.5]	36.0 [30.0–38.0]	33.0 [31.0–36.0]	37.0 [33.5–38.0]	34.0 [32.0–37.0]
AMH (ng/ml)	1.5 [0.6–2.2]	3.9 [2.1–5.1]	2.2 [1.3–4.7]	3.5 [2.8–6.6]	2.3 [1.5–5.9]	3.2 [1.6–6.2]
AFC	15.5 [10.8–17.5]	19.0 [11.5–24.5]	16.0 [11.0–23.0]	17.0 [12.5–26.5]	14.0 [12.0–18.0]	15.0 [12.0–22.0]
Stimulation duration (days)	12.5 [11.2–14.0]	11.5 [10.8–13.0]	12.0 [11.0–13.0]	-	-	-
Total FSH dose (IU)	3300 [3000–3900]	2550 [1772–3600]	3000 [2081–3600]	-	-	-
E2 level on HCG injection day (pg/ml)	336.0 [188.0–1001]	1710 [920.0–3005]	1034 [342.5–2071]	-	-	-
Retrieved oocytes (total no.)	6.0 [4.3–8.5] (49.0)	9.5 [6.7–13.2] (145.0)	8.0 [5.7–10.7] (194)	8.0 [5.0–12.5] (145.0)	4.5 [3.5–7.5] (51.0)	6.5 [4.0–9.0] (196)
Mature oocytes (total no.)	5.5 [4.2–7.7] (44.0)	6.0 [4.7–11.0] (104.0)	6.0 [4.7–8.5] (148)	6.0 [4.0–7.7] (98.0)	4.0 [2.7–5.2] (40.0)	5.0 [3.0–7.0] (138.0)
Fertilization rate*	-	72.2%	-	-	65.0%	-
Oocytes or zygotes cryopreserved/patient	7.5 [5.7–8.5]	6.0 [5.0–12.0]	7.0 [5.0–10.0]	6.0 [4.0–8.0]	3.0 [1.0–3.2]	4.0 [3.0–7.0]

Data are expressed as Medians interquatile range (25th percentile–75th percentile)

AFC antral follicles count, *COS* controlled ovarian stimulation, *FP* fertility preservation, *IVM* in vitro maturation, *OO-COS* oocytes cryopreserved after COS, *OO-IVM* oocytes cryopreserved after IVM, *ZYG-COS* zygotes cryopreserved after COS, *ZYG-IVM* zygotes cryopreserved after IVM, *no*. number of *Fertilization rate: number of oocytes successfully fertilized divided by the number of mature oocytes

and is not different from maturation rate (67.2%) in luteal phase (p = 0.25). All COS cycles were started during the follicular phase.

The number of zygotes cryopreserved per patient was significantly decreased after IVM compared to COS (3.0 [1.0–3.2] vs 6.0 [5.0–12.0], p = 0.003) since 5 female patients benefited from two consecutive FP cycles in the ZYG-COS group. Per OR, the number of zygotes cryopreserved after COS or IVM was not significantly different (4.5 [2.0–8.7] vs 3.0 [1.0–3.2], p = 0.051). The number of oocytes cryopreserved per patient after COS or IVM was not significantly different (7.5 [5.7–8.5] vs 6.0 [4.0–8.0]).

Thawing cycle outcomes

Thawing cycle characteristics and embryo transfer outcomes are summarized in Table 2. Forty female cancer survivors asked for the utilization of cryopreserved oocytes/zygotes, representing 49 thaw cycles started. They waited at least 3 years before being able to benefit from a thawing cycle. Thirty patients underwent at least one embryo transfer, two patients had two embryo transfers, and two patients had three embryo transfers. Ten patients did not undergo an embryo transfer; eight of them were enrolled in the IVM protocol at the time of FP. The reason for embryo transfer failure was either the lack of oocyte survival after thawing (three patients), a fertilization failure (three patients), or embryo cleavage failure (four patients).

When compared to slow freezing, the survival rate after vitrification for both oocytes (54.5% vs 76.2%, p = 0.08) and zygotes (54.3% vs 69.2%, p = 0.29) was increased, but this difference was not significant. A decrease was observed in fertilization rates for oocytes cryopreserved after IVM as compared to after COS (49.1% vs 68.8%, respectively), but this difference was not significant (p = 0.11). A comparable fertilization rate was observed for oocytes injected before freezing as compared to after freezing for COS FP group (72.2% vs 68.8%, p = 0.71), and while in IVM FP group, a decrease was observed (65.0% vs 49.1%, p = 0.12).

When considering all FP strategies, among the 89 embryos obtained, only 11 (12.3%) were of good quality as described in the sect. “Materials and methods”. No good quality embryos were obtained from oocytes thawed after IVM. Thirty-six embryo transfers were completed, yielding live births in each group.

Pregnancies were obtained in each cryopreservation procedure group (OO-COS, OO-IVM, ZYG-COS, ZYG-IVM). Eight healthy babies were born at term. Five of them were part of the zygote FP group (four were part of the ZYG-COS group, and one was part of the ZYG-IVM group), and

Table 2 Thawing cycle characteristics and embryo transfer outcomes

	COS-FP			IVM-FP		
	OO-COS	ZYG-COS	All COS-FP	OO-IVM	ZYG-IVM	All IVM-FP
Female-patients	6	9	15	15	10	25
Thawing cycles	7	14	21	18	10	28
Age (years)	40.0 [33.0–42.0]	37.5 [34.8–42.0]	38.0 [34.5–42.0]	39.0 [37.0–41.0]	40.5 [39.8–42.0]	39.0 [37.5–41.5]
Time from FP cycle to thawing (years)	3.0 [2.7–4.0]	3.0 [1.7–5.0]	3.0 [2.5–4.0]	5.0 [5.0–6.0]	5.0 [3.0–5.0]	5.0 [4.2–5.7]
Oocytes or zygotes warmed/patient (total no.)	7.5 [5.7–8.5] (44)	6.0 [4.0–9.0] (61)	7.0 [5.0–10.0] (105)	6.0 [4.0–8.0] (98)	3.0 [1.0–3.2] (26)	4.0 [3.0–7.0]
No. oocytes or zygotes still frozen	0	14	14	0	0	0
Survival rate slow freezing vitrification	-	54.3%	54.3%	54.5%	73.1%	59.2%
	72.7%	69.2%	71.4%	76.2%	-	76.2%
Fertilization rate*	68.8%	-	-	49.1%	-	-
No. embryos obtained	20	33	53	20	16	36
Percentage of good quality embryos (total no.)	25% (5)	12% (4)	17% (9)	0	13% (2)	5.5% (2)
No. embryo transfers	6	13	19	8	9	17
No. embryos transferred	9	22	31	11	12	23
Implantation rate**	11.1% (1/9)	18.2% (4/22)	16.1% (5/31)	18.2% (2/11)	8.3% (1/12)	13% (3/23)
No. live births	1	4	5	2	1	3
Live birth rate/patient	16.6%	44.4%	33.3%	13.3%	10.0%	12.0%

Data are expressed as medians interquartile range (25th percentile–75th percentile)

COS controlled ovarian stimulation, *IVM* in vitro maturation, *FP* fertility preservation, *OO-COS* oocytes cryopreserved after COS, *OO-IVM* oocytes cryopreserved after IVM, *ZYG-COS* zygotes cryopreserved after COS, *ZYG-IVM* zygotes cryopreserved after IVM, *no*. number of

*Fertilization rate: number of oocytes successfully fertilized divided by the number of mature oocytes

**Implantation rate: number of gestational sacs with a fetal heartbeat divided by the number of embryos transferred

three were from cryopreserved oocytes (one from the OO-COS group, and two from the OO-IVM group). It has to be noted that the two female-patients who gave live birth in the OO-IVM group were punctured at the luteal phase of their menstrual cycle at the time of fertility preservation. Table 3 shows the outcomes and patient characteristics in pregnancies after IVF-FP cycles due to cancer.

Discussion

The present study aimed to investigate the outcomes of oocyte or zygote thawing cycles according the procedure used for oocyte maturation at the time of FP in female cancer survivors. Eight healthy babies were born, three of which were born after the utilization of cryopreserved oocytes or zygotes following IVM.

In 10 years of clinical practice in our center, 667 women have undergone oocyte or embryo cryopreservation before cancer treatment. Once cured, 6% of them (40 patients) asked for frozen-thawed oocyte or embryo transfer cycles since attempts to obtain a natural pregnancy were unsuccessful. This low return rate after oncological FP is similar to that described in the literature for oocytes [29] and for banked sperm [42, 43]. Female patients must be psychologically able to attempt pregnancy after a long history of treatment. Moreover, in the case of breast cancer, tamoxifen is often prescribed for several years, leading to a delay in patients using their own oocytes or embryos.

In our clinic, IVM was set up in 2003 for women with polycystic ovary syndrome to avoid the risk of the ovarian hyperstimulation syndrome [44]. Although IVM requires specific expertise, it was relatively easy to adapt this procedure to FP. IVM can be applied in emergency situations, regardless of the menstrual cycle phase [12, 45]. This technique should be considered in the FP strategy when ovarian stimulation is unfeasible, in particular when markers of the follicular ovarian status are at a relatively high range to maximize the chance of obtaining a live birth [46]. Some authors encourage the combination ovarian tissue cryopreservation with IVM of oocytes retrieved from ovarian tissue ex vivo. Live births following this ex vivo IVM technique have been reported [14, 25]. Nevertheless, IVM is still regarded as experimental procedure in the field of FP [27] albeit the ESHRE guideline group on female FP

Table 3 Outcomes and patient characteristics in pregnancies after IVF-FP cycles due to cancer

Patient	Age at FP	Cancer diagnoses	Oocytes collected (*n*)	Type of oocyte maturation	Number of OO/ZYG cryopreserved	Time between FP and ET (months)	Embryos transferred (*n*)	Outcomes
1	29	Medullary hypoplasia	9	COS	8 OO	47	2	Live birth
2	29	Breast	7	IVM	6 OO	65	1	Live birth
3	31	Breast	9	IVM	8 OO	73	2	Live birth
4	36	Breast	9	IVM	4 ZYG	64	2	Live birth
5	36	Acute lymphoblastic leukemia						
First ET			27	COS	19 ZYG	33	2	Live birth
Second ET						77	1	Live birth
6	36	Borderline tumor	15	COS	6 ZYG	14	2	Live birth
7	26	Borderline tumor	16	COS	7 ZYG	12	2	Live birth

COS controlled ovarian stimulation, *ET* embryo transfer, *IVM* in vitro maturation, *OO* oocytes, *FP* fertility preservation, *ZYG* zygotes

has considered IVM as an innovative procedure because we lack hindsight on the outcomes that result from this technique.

Here, we report two healthy babies born from oocyte cryopreservation after IVM performed in the circumstance of breast cancer. Consequently, the live birth rate (LBR) per patient was 13.3% (2/15 patients). For zygote cryopreservation after IVM, the LBR per patient was 10.0% (1/10 patients). In the present study, slow cooling was used more often in IVM than in COS, and it is well known that vitrification provides better results compared to slow cooling in non-oncological contexts [47]. It is thus reasonable to speculate that vitrification may implement the LBR in the future in the field of IVM-FP. Nevertheless, it is difficult to really conclude on the efficiencies of IVM for FP since few data are available (Supplemental Table).

Since the first birth after transfer of cryopreserved embryos in the field of COS-FP [15], many studies reported the efficiency of this procedure in female cancer patients undergoing FP. As shown in Supplemental Table, the live birth rates per embryo transfer in the literature vary from 12.5% [19] to 50.0% [15] representing 67 healthy babies born. In our study, four healthy babies were born from ZYG-COS thawing cycles resulting in a LBR of 30.7% per embryo transfer and 44.4% per patient. Our results seem to be comparable with the mean results from several studies (Supplemental Table). Although embryo cryopreservation is an established option for FP, women in couples take the risk of losing their reproductive autonomy and facing eventual issues of ownership of stored embryos [48]. Regarding oocyte cryopreservation, we report one baby born among the 6 patients who used thawed oocytes cryopreserved in a context of oncological FP. As shown in Supplemental Table, the live birth rates per embryo transfer in the literature are a variable representing 38 healthy babies born with a mean of 25.6%.

It is difficult to compare our data with those collected in the Supplemental Table since only 6 female patients had COS followed by oocyte vitrification at the time of FP. The small size of our different groups is one of the main limitations of our study.

Conclusions

We report eight live births for female patients who used their cryopreserved oocytes or embryos after being cured of breast cancer. Given the relatively small numbers of patients in our cohort study, thawing cycles should be evaluated over time to draw definitive conclusions about the efficiency of each FP method. That being said, the results of this study may place IVM as an innovative procedure and some additional data on a larger sample must be obtained to confirm the effectiveness of this technique which led to 3 live births for female cancer patient in our FP clinic.

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Code availability Not applicable

Author contribution All authors have been involved in the conception and design, or acquisition of data, or analysis and interpretation of data. A.M., V.P.,V.W., L.H., V.G., and A.B. drafted the manuscript. M.G.,

C.S., and N.F. revised the article for important intellectual content. All authors approved the final version to be published.

Data availability Not applicable

Declarations

Conflict of interest The authors declare no competing interests.

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