MedCrave
Step into the World of Research

Obstetrics & Gynecology International Journal

Review Article

Open Access

CrossMark

Freezing techniques as fertility preservation strategies: a narrative review

Abstract

Advances in the development of new chemotherapy and radiotherapy regimens have significantly improved the survival of cancer patients but many of these treatments have detrimental effects on gonadal function. These treatments may cause premature ovarian failure in females and azoospermia in males. Non-oncological conditions may also require therapies that put women's and men´s reproductive potential at risk. Moreover, an increasing number of women today decide to postpone maternity beyond the age of 35 due to social reasons with a possible affectation of future fertility due to the effect of age. The development and evolution of assisted reproductive techniques offer these patients new alternatives to preserve their fertility. The objective of this review is to describe the different options of fertility preservation. Oocyte vitrification for female and sperm banking for male are the first line for fertility preservation at the present time. Other techniques such as ovarian and testis tissue preservation, in vitro maturation of gametes, pharmacological protection (GnRH agonists and antiapoptotic agents) and surgical treatments (oophoropexy, selective radical trachelectomy) will be described. One of the most important steps in optimizing the results of these fertility preservation procedures in cancer patients is the need for prompt and timely referral to reproductive medicine specialists. Fertility preservation should be integrated as part of the oncological healthcare. Optimal counselling from healthcare professionals should always be present.

Keywords: fertility preservation, oocyte vitrification, sperm cryopreservation, oncological patients

Volume 13 Issue 6 - 2022

Ciarmatori Silvia, Rey Valzacchi Gaston
Italian Hospital of Buenos Aires, Buenos Aires, Argentina

Correspondence: Silvia Inés Ciarmatori, Italian Hospital of Buenos Aires, Buenos Aires, Argentina, Tel 4815 5818, Email silvia.ciarmator@hospitalitaliano.org.ar

Received: November 29, 2022 | **Published:** December 29, 2022

Introduction

The concept of fertility preservation refers to the application of medical, surgical and laboratory procedures to preserve the reproductive function of both women and men who are at risk of impaired reproductive capacity for health or social reasons. Fertility preservation strategies started developing over two decades ago, aiming to maintain reproductive capacity in cancer patients, since the chemotherapy and/or radiotherapy treatments that these patients receive have a high probability of causing infertility.[1] Advances in the development of new chemotherapy and radiotherapy regimens have significantly improved the survival of cancer patients, but many of the treatments have detrimental effects on gonadal function. As result, a large population of young individuals are cured of their disease but are faced with reproductive difficulties because of antitumor treatments.

In addition to infertility related to oncological treatments in male and female patients, there are other situations in which reproductive functions may be permanently affected, such as women who have had multiple ovarian surgeries, treatments for some collagen diseases or gender affirmation treatments. Moreover, an increasing number of women today decide to postpone maternity beyond the age of 35 due to social reasons such as professional development or a lack of a partner. This social change leads many women to be unable to get pregnant spontaneously, due to the impact of the years on the ovarian reserve. Due to these very diverse situations, there has been an enormous development in preexisting fertility preservation techniques, as well as the investigation of new strategies. Fertility preservation has now become a subspecialty within reproductive medicine, with many international and national scientific societies dedicated to the study and research of new techniques, diffusion of the subject into different areas, and recording of the results of all these procedures around the world.

Undoubtedly, one of the most significant progresses in this field has been the advances in cryopreservation techniques.[2] The freezing of embryos, oocytes and spermatozoa are now established techniques for fertility preservation. However, in some specific pathologies, surgical strategies are necessary. Such is the case of selective radical trachelectomy, offered to women with early-stage cervical cancer.[3] Hysterectomy is the conventional treatment for this patient. However, in very selected patients who wish to get pregnant, selective radical trachelectomy may be offered to preserve the uterus. Ophoropexy may be offered to patients who should receive pelvic radiotherapy. It consists of surgical fixation of the ovaries to the pelvic wall to remove them from the radiotherapy field. The ovaries are particularly radiosensitive, and even low radiotherapy doses may result in impaired or complete loss of ovarian function, causing hormonal disturbances and infertility.[4]

The following review presents options for fertility preservation focusing on cryopreservation techniques, which are the most frequently applied techniques, both in women and men.

Fertility preservation in women

Fertility preservation for oncological reasons

Approximately 10% of cancers occur in women under 45 years old.[5] Chemotherapy, radiation, surgical resections and bone marrow transplantation have greatly improved survival rates. However, many of these treatments can cause decreased ovarian reserve and premature ovarian failure.[6,7] Indeed, these therapies can affect the ovarian reserve through different mechanisms: inducing loss of primordial follicles, accelerating activation of primordial follicles, promoting follicular atresia, damage to the stroma and ovarian vasculature or inflammation of the gonads. Furthermore, the risk of ovarian failure depends on the ovarian reserve, the age of the patient, and the type and dose of drugs used (Table 1).[8,9] For this reason, it is essential to individualize

the strategy offered to each patient. The optimal approach must take multiple and diverse factors into consideration: patient treatment (radiation, surgery, chemotherapy, or a combination thereof), time available before treatment starts, the age of the patient, the pubertal state (pre- or post-pubertal), the patient´s disease, the presence of a partner, the consideration of future long-term problems such as storage and availability of frozen embryos, cells or tissues, among others.[10] Elderly women are more at risk of presenting with ovarian failure after gonadotoxic treatment than girls and younger women, due to the physiologic depletion of oocytes throughout life.

Table 1 Chemotherapeutic agents according to their gonadotoxic impact in women (amenorrhea) and men (azoospermia) (Adapted from Rodriguez-Wallberg KA et al. Fertility preservation for young adults, adolescents, and children with cancer[8])

High risk of chemotoxicity
Cyclophosphamide
Ifosfamide
Melphalan
Busulfan
Nitrogenous mustard
Procarbazine
Dacarbazine
Chlorambucil
Intermediate risk of chemotoxicity
Cisplatin (with low cumulative dose)
Carboplatin (with low cumulative dose)
Adriamycin
Low risk of chemotoxicity
Bleomycin
Actinomycin d
Vincristine
Methotrexate
5-fluorouracil
Radioactive iodine treatment for thyroid cancer
Undefined risk of chemotoxicity
Paclitaxel and docetaxel for the treatment of breast cancer
Irinotecan
Trastuzumab
Imatinib
Erlotinib
Bevacizumab

In women affected by oncological disease, cryopreservation techniques are the most used, especially oocyte freezing. Embryo cryopreservation can also be offered. Although this strategy has a high success rate, it has some limitations, such as the dilemma of discarding embryos in case the patient, for health or personal reasons, decides not to transfer cryopreserved embryos. In prepubertal girls or women who must start chemotherapy immediately and ovarian stimulation is not possible, cryopreservation of ovarian tissue is the only proven method currently available.[11]

Other alternatives in cases where there is no time to perform ovarian stimulation and oocyte vitrification include in vitro oocyte maturation,[12] or the administration of agents that "protect" the ovary. The latter consists of using GnRH agonist analogues, before and during chemotherapy treatment.[13] By suppressing ovarian function, have been theorized GnRH agonist would protect the ovary from a toxic insult such as chemotherapy. The results of the studies that evaluated this alternative were not conclusive.[14] It is difficult to assess the efficacy of GnRH therapy in preserving fertility because most published studies have used unreliable or inadequate surrogate outcomes (resumption of menses rather than pregnancy or live birth rate). Even so, GnRH therapy is usually offered, since it does not delay the start of cancer treatment, it is easy to implement, and it is not invasive. Several antiapoptotic drugs are currently being developed and evaluated with the aim of protecting the ovary.[15] These drugs would provide protection either by inhibiting one or more pathways in which the chemotherapy acts, causing damage to the ovary, or by reducing the delivery of the drug to the ovary.

Table 2 describes the different strategies used to preserve fertility for oncological reasons in women.

Table 2 Preservation of fertility in cancer patients: Therapeutic strategies

Cryopreservation
· Embryos
· Mature oocytes
· Immature oocytes for ivm
· Ovarian tissue
· Complete ovary
Pharmacological protection
· GnRH agonists
· Antiapoptotic Agents (Sphingosine 1 Phosphate, Ceramide 1 Phosphate, CHK2 Inhibitors, Tyrosine Kinase Inhibitors, mTorc Inhibitors, Melatonin, LH, Mirtazapine, Ghrelin, G-CSF -Granulocyte Colony Stimulating Factor, Imatinib, Bortezomib)
Surgical treatments
· Selective radical trachelectomy
· Oophoropexy(ovarian transposition)
· Ovarian / ovary tissue transplant
· Uterine transplant

Cryopreservation techniques

Cryopreservation consists of using extreme sub-zero temperatures to stop the vital functions of a cell, a tissue or an organism and allowing it to remain in conditions of "suspended life" for a long time. Cryopreservation techniques in reproductive medicine are associated with a higher probability of successfully generating offspring.[16,17]

These methods try to achieve low temperatures without ice forming during freezing, which would cause damage to the cell structure. Therefore, the cells must be mixed with different cryoprotective solutions, depending on the type of cell or tissue to be frozen. Cryopreservation techniques are used today for the preservation of embryos, gametes (sperm and eggs) or tissues (ovaries, testicles). Two freezing techniques exist: the classic or "slow freezing" - today almost abandoned - and the rapid freezing or "vitrification", developed over a decade ago.[18] The latter consists of converting water into a solid without the formation of ice crystals using very "fast freeze" and cryoprotectants. The cells instantaneously solidify into a glass-like structure. Vitrification was initially developed for oocytes but is now the preferred method for embryos and even sperm as it results in higher survival, implantation and pregnancy rates compared to the older slow freezing technique.[19]

Oocyte cryopreservation

Oocyte cryopreservation is the most widely used fertility preservation strategy in women. As previously explained. Egg freezing is not only indicated for cancer patients or women undergoing treatments that can potentially affect ovarian reserve, but is currently offered to healthy women who plan to become pregnant in the future.[20] This procedure is named fertility preservation for social reasons or social egg freezing, and it is the most frequent indication at

present. Social egg freezing allows women to preserve their fertility in anticipation of age-related fertility decline and ineffective fertility treatments at older ages.

With the traditional slow freezing technique, the pregnancy rate with cryopreserved oocytes was very low and the results were difficult to reproduce. It was considered an experimental technique. Unlike the cryopreservation of embryos and spermatozoa, the cryopreservation of oocytes is technically more complex, since oocytes are cells that contain more water and, therefore, are more sensitive to injuries due to the formation of ice crystals. The meiotic spindle, cytoskeleton, cortical granules, and zona pellucida are the structures most likely to be altered during freezing. However, with the fast freezing or "vitrification" technique, which prevents the formation of ice crystals, the oocyte survival rate can be as high as ninety percent.

To obtain the oocytes, the woman must previously undergo an ovarian stimulation. Stimulation treatment begins on the second day of the cycle (early follicular phase) and gonadotropins are applied. Human Menopausal Gonadotropin (hMG), recombinant follicle stimulating hormone (rFSH) and recombinant luteinizing hormone (rLH), for 9 to 12 days.[21] The objective is to achieve the maturation of a set of follicles and thus be able to obtain as many eggs as possible. By increasing the number of oocytes aspirated for fertilization, the number of available embryos increases, the best quality embryo can then be selected to transfer and thus improve the possibility of pregnancy and live birth. Follicle growth is monitored with a transvaginal ultrasound. Once the ovarian stimulation is finished, an transvaginal ovarian aspiration punction is performed and, through this process, oocytes are removed from the interior of the follicles. To perform this, a needle is inserted through the vagina, guided into the follicles by ultrasound visualization.

Initially, stimulation to cryopreserve oocytes for oncological reasons had some drawbacks. Firstly, the time required until the beginning of the next cycle, plus the time required for the ovarian stimulation itself, could excessively delay the start of cancer treatment (radio or chemotherapy). Secondly, ovarian stimulation is associated with an increase in oestradiol to supraphysiological levels, which could generate greater tumor growth in hormone-dependent tumors. However, greater knowledge of the ovarian cycle, female gametogenesis, and the pharmacodynamics of certain drugs has made it possible to overcome these obstacles. In patients who cryopreserve ovules for oncological reasons, stimulation can be started at any time of the cycle, and not necessarily at the beginning of the follicular phase; this is known as random start. In fact, women can be stimulated twice in the same cycle, in order to increase the number of oocytes aspirated. This is known as duo stimulation (duo stim). In patients with estrogen-dependent malignancies (for example, breast cancer), the drug of choice for ovarian stimulation is letrozole (an aromatase enzyme inhibitor), which prevents the increase in estrogen levels, that occurs with traditional stimulation schemes.[22] Although many oncologists are concerned about the use of traditional assisted reproductive technology (ART) in women with hormone-sensitive malignancies, the results of studies using letrozole show reassuring results. In healthy women, whose objective is to preserve oocytes for deferred maternity, conventional ovarian stimulation treatments for in vitro fertilization procedures are used.

In addition to being a technique used in women who want to preserve fertility for social reasons, it is important to offer these procedures to women who are at risk of having a decreased ovarian reserve due to medical histories or pathologies that affect reproductive capacity. Such is the case of women who have received multiple ovarian surgeries or who have endometriosis.

Once the woman decides to seek pregnancy (in the case of cancer patients, when she is discharged from cancer treatment), the oocytes are devitrified and fertilized with sperm in a plate with special culture medium. This procedure is named in vitro fertilization (IVF). In cases where the number and/or quality of the spermatozoa are not adequate, intracytoplasmic sperm injection or ICSI (Intracytoplasmic sperm injection) is performed.

In randomized clinical trials, the pregnancy rate with cryopreserved/thawed mature oocytes was generally similar to that of fresh oocytes, although these trials were largely limited to populations of donor oocytes.[23,24] In contrast, two large observational studies in European population reported acceptable success rates, but lower than with fresh oocytes.[25,26] Importantly, the probability of obtaining embryos and ongoing pregnancy from cryopreserved eggs is related to the age of the woman at the time the eggs were retrieved. The younger the woman is at the time of oocytes retrieval, the greater the chance of achieving a viable pregnancy and live birth.

In addition to allowing reproductive potential to be maintained over time, egg cryopreservation reduces genetic risk, since the probability of the embryo presenting a chromosomal abnormality is related to the woman's age at the time of egg retrieval and not to the age of the woman when receives the embryo from the devitrified and fertilized oocytes

Ovarian tissue cryopreservation. In vitro maturation of oocytes

Occasionally, cancer patients need to start gonadotoxic oncological treatment urgently. Since the time required to perform ovarian stimulation followed by transvaginal oocyte retrieval (almost 2 weeks) is an unacceptable delay, the only available option in these cases is ovarian tissue cryopreservation.[27–29] The frozen ovarian tissue is thawed and autotransplanted once the woman is discharged and allowed to get pregnant. Ovarian tissue is cut into extremely thin strips, squares or fragments and is frozen by slow freezing or vitrification.[30] When the woman is allowed to become pregnancy, the ovarian cortex bands are transplanted back into the patient. Ovarian tissue can be transplanted in the remaining ovarian tissue, in the ovarian fossa, or in the pelvic peritoneum (orthotopic transplantation)[31] or in the subcutaneous tissue of abdominal wall, forearm, or chest wall (heterotopic transplantation).[32]

Once transplanted, the ovarian tissue can restore hormonal function and produce mature eggs. This alternative is valid only in oncological pathologies in which there is certainty that there are no primary or secondary tumors in the ovary. In some malignant pathologies, such as leukemias or lymphomas, there is a potential risk of reintroducing malignant cells when the autologous tissue transplantation is performed. Therefore, these techniques cannot be applied to all tumors. Ovarian tissue transplantation is also indicated in prepubertal girls who must receive gonadotoxic treatment[33] or in patients with hormone-sensitive tumors.

For many years, ovarian tissue transplantation was considered an experimental technique because first results were limited to case reports and small series of patients. However, a meta-analysis published in 2017 that included pregnancy-related outcomes reported a cumulative clinical pregnancy rate of 57.5%, similar to or even higher than that obtained with oocyte cryopreservation.[34] In addition, 64% of the women recovered endocrine function and approximately two-thirds of the women conceived naturally, without the need for assisted reproduction.[35] Most pregnancies after ovarian tissue grafting were in women younger than 30 years of age at the time of the

cryopreservation procedure. A recent systematic review on ovarian tissue processing size for cryopreservation evaluated 92 participants who underwent ovarian tissue transplantation.[36] The review reported a pregnancy rate of 81.3%, 45.5%, 66.7% and a live birth rate of 56.3%, 18.2%, 66.7% in the strip, square and fragments, respectively. This data has shown that ovarian tissue transplantation is no longer considered an experimental technique.

Another technique described for emergency situations is the in vitro maturation of oocytes.[37,38] In this case, the ovaries are punctured without prior stimulation and the oocytes obtained are matured in vitro, before or after freezing. Currently, the results of in vitro maturation are limited.

Embryo cryopreservation

This procedure consists in freezing extra embryos that have not been transferred in the fresh cycle. Embryos frozen in liquid nitrogen at very low temperatures can be kept for many years. Since no more than one embryo should be transferred to avoid the risk of a multiple pregnancy, all additional embryos should be frozen. For this reason, today this practice is routinely performed in all IVF/ICSI cycles in which more than one embryo was obtained or in cases in which no embryo is transferred in the fresh cycle ("freeze all"). Due to the vast experience with embryo cryopreservation, the possibility of pregnancy after the transfer of frozen/thawed embryos is very high. Embryo cryopreservation presents some limitations and complex ethical issues. For example, if the woman has a partner, she must determine before obtaining the embryos what her reproductive plan is, or what to do in the event of the patient's death or dissolution of the couple. Some couples may be philosophically opposed to embryo cryopreservation. Ovarian stimulation for embryo freezing is not ethically acceptable in postmenarchal girls with cancer. Furthermore, these procedures should only be performed where there are strictly enforced embryo protection laws. In some countries, such as Germany, it is illegal.

Fertility preservation in men

As in female cancer patient, new chemotherapy and radiotherapy treatments have significantly improved the survival of men with cancer. Different tumors, such as leukemia and lymphomas in children and adolescents and testis cancer in adults can now be successfully cured. However, infertility can arise as a consequence of treatment of oncological conditions. The germinal cell division is extremely high through increased meiotic and mitotic activity and allowing for increase sensitivity to cytotoxic agents. Patients have to be informed about the expected risk of infertility, which can vary in magnitude on the basis of the specific treatment.

Sperm cryopreservation

Sperm freezing is the process of collecting, analyzing, freezing, and storing a man's sperm in liquid nitrogen indefinitely. It is the main strategy indicated for patients who must undergo a treatment that may potentially affect spermatogenesis, such as chemotherapy.[39,40] It is a long established and proven procedure. Both the slow freezing technique and the vitrification technique can be used. Some patients who cannot obtain the sample may be able to do so by penile vibration or via electroejaculation. When it's not possible to obtain spermatozoa in the ejaculation, testicular tissue can also be frozen, or spermatozoa can be obtained through punction of the testicle or epididymis.[41] The cryopreserved sperm is then thawed and used to fertilize in an in vitro reproductive procedure (IVF/ICSI). An ICSI must be performed with the post-thawing spermatozoa.

Spermatozoa may be damaged by freezing and thawing,[42] although post-thawing sperm recovery rate is usually high. Recovery of motile sperm after thawing is less likely in men with severe oligospermia and possibly in those with testicular cancer or a basal semen parameter below reference values. The possibility of storing semen should be systematically offered to all men who receive a treatment with a possible deleterious effect on the gonads.

Other fertility preservation techniques in men

In prepubertal boys, cryopreservation of testicular tissue is the only preservation alternative, since spermatozoa are not produced at this stage of life.[29,30] A potential risk when performing autotransplantation of cells or tissues is that of reintroducing malignant cells together with the cryopreserved cell suspension. For this reason, the alternative of transplanting these cells into another animal species (xenotransplantation) has been proposed.[43] These techniques are currently considered experimental.

Quick and timely referral to the specialist

One of the most important steps in optimizing the results of these fertility preservation procedures in cancer patients is the need for prompt and timely referral to reproductive medicine specialists. Medical oncologists should discuss the risk of treatment-induced infertility as well as potential fertility-preserving interventions before initiating gonadotoxic therapy or excisional surgery. This discussion should be raised as early as possible during treatment planning because some interventions are time consuming and could delay the initiation of cancer therapy. A delay in referral may cause the fertility preservation procedure no longer to be possible and therefore a unique opportunity is lost. Some institutions provide expedited consultations within 48 hours of diagnosis. Even though numerous scientific societies have strongly recommended that patients receive adequate advice on fertility preservation treatments, numerous studies show that there is still low awareness in many health teams about the need to refer these patients.[44,45]

Conclusion

At present, oncological treatments have managed to substantially increase the survival rate of adults, young people and children; however, in many cases, these treatments are associated with significant loss or decrease in reproductive function. Similar results are obtained for treatments applied to certain medical or gender affirming disorders. Patients requiring such treatments should be referred immediately for advice on fertility preservation options. Embryo or gamete cryopreservation is the established method of fertility preservation for adults, adolescents and post-pubertal children. In some women, the use of gonadotropin-releasing hormone (GnRH) agonists to suppress ovarian function during chemotherapy has been associated with post-treatment return of ovarian hormone production and some live births. However, the live birth rate after GnRH agonist treatment is lower than that with proven assisted reproductive techniques. In pre-pubertal girls or when there is not time to perform ovarian stimulation and transvaginal punction, cryopreservation of ovarian tissue can be offered. This technique is no longer experimental. In men, the technique of choice is semen freezing. For prepubertal boys, investigative techniques include possible cryopreservation of testicular tissue. Likewise, the oocyte cryopreservation technique should be offered to healthy women who plan to postpone the search for pregnancy. In all cases, the preservation of fertility requires that the strategy be individualized.

Citation: Silvia C, Gaston RV. Freezing techniques as fertility preservation strategies: a narrative review. *Obstet Gynecol Int J.* 2022;13(6):395–400.
DOI: 10.15406/ogij.2022.13.00683

Acknowledgments

None.

Funding

None.

Conflicts of interest

There are no conflicts of interest.

References

1. Anderson RA, Brewster DH, Wood R, et al. The impact of cancer on subsequent chance of pregnancy: a population-based analysis. *Hum Reprod*. 2018;33:1281–1290.
2. Argyle CE, Harper JC, Davies MC. Oocyte cryopreservation: where are we now? *Hum Reprod Update*. 2016;22:440–449.
3. Abu-Rustum NR, Sonoda Y, Black D, et al. Fertility-sparing radical abdominal trachelectomy for cervical carcinoma: technique and review of the literature. *Gynecol Oncol*. 2006;103(3):807–813.
4. Moawad NS, Santamaria E, Rhoton-Vlasak A, et al. Laparoscopic ovarian transposition before pelvic cancer treatment: ovarian function and fertility preservation. *J Minim Invasive Gynecol*. 2017;24(1):28–35.
5. National Cancer institute Age and Cancer Risk. 2021.
6. van Dorp W, Haupt R, Anderson RA, et al. Reproductive function and outcomes in female survivors of childhood, adolescent, and young adult cancer: a review. *J Clin Oncol*. 2018;36:2169–2180.
7. Jayasinghe YL, Wallace WHB, Anderson RA. Ovarian function, fertility and reproductive lifespan in cancer patients. *Expert Rev Endocrinol Metab*. 2018;13:125–136.
8. Rodriguez-Wallberg KA, Anastacio A, Vonheim E, et al. Fertility preservation for young adults, adolescents, and children with cancer. *Whoops J Med Sci*. 2020;125(2):112–120.
9. Winkler-Crepaz K, Ayuandari S, Ziehr SC, et al. Fertility preservation in cancer survivors. *Minerva Endocrinol*. 2015;40:105–118.
10. Shliakhtsitsava K, Suresh D, Hadnott T, et al. Best practices in counseling young female cancer survivors on reproductive health. *Semin Reprod Med*. 2017;35(4):378–389.
11. Silber SJ, DeRosa M, Goldsmith S, et al. Cryopreservation and transplantation of ovarian tissue: results from one center in the USA. *J Assist Reprod Genet*. 2018;5(12):2205–2213.
12. Abir R, Ben-Aharon I, Garor R, et al Cryopreservation of in vitro matured oocytes in addition to ovarian tissue freezing for fertility preservation in pediatric female cancer patients before and after cancer therapy. *Hum Play*. 2016;31:750–758.
13. Blumenfeld Z. Fertility Preservation using GnRH agonists: rationale, possible mechanisms, and explanation of controversy. *Clin Med Insights Reprod Health*. 2019;13:1–13.
14. Elgindy E, Sibai H, Abdelghani A, et al. Protecting ovaries during chemotherapy through gonad suppression: a systematic review and meta-analysis. *Obstet Gynecol*. 2015;126(1):187.
15. Spears N, Lopes F, Stefansdottir A, et al. Ovarian damage from chemotherapy and current approaches to its protection. *Human Reprod Update*. 2019:25(6):673–693.
16. Oktay K, Harvey BE, Partridge AH, et al. Fertility preservation in patients with cancer: ASCO clinical practice guideline update. *J Clin Oncol*. 2018;36(19):1994–2001.
17. Practice Committee of the American Society for Reproductive Medicine. Fertility preservation in patients undergoing gonadotoxic therapy or gonadectomy: a committee opinion. *Fertile Sterile*. 2019;112(6):1022.
18. Levi-Setti PE, Patrizio P, Scaravelli G. Evolution of human oocyte cryopreservation: slow freezing versus vitrification. *Curr Opin Endocrinol Diabetes Obes*. 2016;23(6):445–450.
19. Rienzi L, Gracia C, Maggiulli R, et al. Oocyte, embryo and blastocyst cryopreservation in ART: systematic review and meta-analysis comparing slow-freezing versus vitrification to produce evidence for the development of global guidance. *Hum Reprod Update*. 2017;23(2):139–155.
20. Varlas VN, Bors RG, Albu D, et al. Social freezing: pressing pause on fertility. *Int J Environ Res Public Health*. 2021;18(15):8088.
21. Racca A, Drakopoulos P, Neves AR, et al. Current therapeutic options for controlled ovarian stimulation in assisted reproductive. *Technology Drugs*. 2020;80:973–994.
22. Bonardi B, Massarotti C, Bruzzone M, et al. Efficacy and safety of controlled ovarian stimulation with or without letrozole co-administration for fertility preservation: a systematic review and meta-analysis. *Front Oncol*. 2020;10:574669.
23. Cobo A, Kuwayama M, Pérez S, et al. Comparison of concomitant outcome achieved with fresh and cryopreserved donor oocytes vitrified by the Cryotop method. *Fertile Sterile*. 2008;89(6):1657–1664.
24. Parmegiani L, Cognigni GE, Bernardi S, et al. Efficiency of aseptic open vitrification and hermetical cryostorage of human oocytes. *Play Biomed Online*. 2011;23(4):505–512.
25. Scaravelli G, Vigiliano V, Mayorga JM, et al. Analysis of oocyte cryopreservation in assisted reproduction: the Italian National Register data from 2005 to 2007. *Reprod Biomed Online*. 2010;21(4):496–500.
26. Borini A, Levi Setti PE, Anserini P, et al. Multicenter observational study on slow-cooling oocyte cryopreservation: clinical outcome. *Fertile Sterile*. 2010;94(5):1662–1668.
27. Yasmin E, Balachandren N, Davies MC, et al. Fertility preservation for medical reasons in girls and women: British fertility society policy and practice guideline. *Hum Fertil (Camb)*. 2018;21:3–26.
28. Rodriguez-Wallberg KA, Oktay K. Fertility preservation during cancer treatment: clinical guidelines. *Cancer Manag Res*. 2014;6:105–117.
29. Practice Committee of the American Society of Reproductive Medicine. Fertility preservation in patients undergoing gonadotoxic therapy or gonadectomy: a committee opinion. *Fertil Steril*. 2019;112:1022–1031.
30. Gellert SE, Pors SE, Kristensen SG, et al. Transplantation of frozen-thawed ovarian tissue: an update on worldwide activity published in peer-reviewed papers and on the Danish cohort. *J Assist Reprod Genet*. 2018;35:561–567.
31. Practice Committee of the American Society for Reproductive Medicine. Evidence-based outcomes after oocyte cryopreservation for donor oocyte in vitro fertilization and planned oocyte cryopreservation: a guideline. *Fertile Sterile*. 2021;116(1):36–47.
32. Oktay K, Türkçüoğlu I, Rodriguez-Wallberg KA. Four spontaneous pregnancies and three live births following subcutaneous transplantation of frozen banked ovarian tissue: what is the explanation? *Fertil Steril*. 2011;95(2):804e7–810.
33. Poirot C, Brugieres L, Yakouben K, et al. Ovarian tissue cryopreservation for fertility preservation in 418 girls and adolescents up to 15 years of age facing highly gonadotoxic treatment. Twenty years of experience at a single center. *Acta Obstet Gynecol Scand*. 2019;98:630–637.
34. Pacheco F, Oktay K. Current success and efficiency of autologous ovarian transplantation: a meta-analysis. *Reprod Sci*. 2017;24(8):1111–1120.
35. Society for assisted reproductive technology. IVF success rates. Clinic summary report 2009.

36. Diaz AA, Kubo H, Handa N, et al. A systematic review of ovarian tissue transplantation outcomes by ovarian tissue processing size for cryopreservation. *Front Endocrinol (Lausanne)*. 2022;13:918899.

37. Abir R, Ben-Aharon I, Garor R, et al Cryopreservation of in vitro matured oocytes in addition to ovarian tissue freezing for fertility preservation in pediatric female cancer patients before and after cancer therapy. *Hum Play*. 2016;31:750–758.

38. Ata B, Shalom-Paz E, Chian RC, et al. In vitro maturation of oocytes as a strategy for fertility preservation. *Clin Obstet Gynecol*. 2010;53:775–782.

39. Oktay K, Harvey BE, Partridge AH, et al. Fertility preservation in patients with cancer: ASCO clinical practice guideline update. *J Clin Oncol*. 2018;36(19):1994–2001.

40. Ethics Committee of the American Society for Reproductive Medicine. Fertility preservation and reproduction in patients facing gonadotoxic therapies: an Ethics Committee opinion. *Fertility and sterility*. 2018;110(3):380–386.

41. Flannigan R, Bach PV, Schlegel PN. Microdissection testicular sperm extraction. *Transl Androl Urol*. 2017;6(4):745–752.

42. Palomar Rios A, Gascón A, Martínez JV, et al. Sperm preparation after freezing improves motile sperm count, motility, and viability in frozen-thawed sperm compared with sperm preparation before freezing-thawing process. *J Assist Reprod Genet*. 2018;35(2):237–245.

43. Ntemou E, Kadam P, Van Saen D, et al. Complete spermatogenesis in intratesticular testis tissue xenotransplants from immature non-human primates. *Hum Play*. 2019;34(3):403–413.

44. McCray DK, Simpson AB, Flyckt R, et al. Fertility in women of reproductive age after breast cancer treatment: practice patterns and outcomes. *Ann Surg Oncol*. 2016;23(10):3175–3181.

45. Fabi A, Lanzetta G, Vizza E, et al. The unmet need for oncofertility preservation in women: Results of a survey by different oncological specialists in Lazio, Italy. *Curr Probl Cancer*. 2019;43(6):100479.

Citation: Silvia C, Gaston RV. Freezing techniques as fertility preservation strategies: a narrative review. *Obstet Gynecol Int J*. 2022;13(6):395–400.
DOI: 10.15406/ogij.2022.13.00683