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Setting up a fertility preservation programme

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A B S T R A C T

With improved survival rates from cancer, young people can expect to lead a normal life, including having their own children. However, cancer or other serious disease itself, and more often its treatment, often leads to a significant reduction in fertility or premature gonadal insufficiency. There is increasing acknowledgement for the importance of fertility preservation (FP) options to be discussed and offered to young people whose fertility is at risk, ideally before the gonadotoxic therapy begins. FP options currently include oocyte, embryo and ovarian tissue cryopreservation; ovarian protection during chemotherapy and semen, sperm and testicular tissue cryopreservation. A multidisciplinary team consisting of committed and enthusiastic doctors, scientists, nurses, counsellors, administrators and researchers is required to provide a holistic FP service with rapid response capacity for acute consultation and procedures and a robust system for long-term follow-up. This speciality is developing rapidly with exciting scientific advances that have relevance for the whole spectrum of reproductive medicine.

Introduction

What is fertility preservation?

Fertility preservation (FP) refers to the practice of optimising fertility for the future. FP can be considered because of the risk of damage to fertility from cancer or other serious disease, or more

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commonly, it is necessary and often a life-saving treatment. FP is also important for young patients with gender dysphoria conditions who may wish to transition and for young patients with genetic fertility compromise, such as Turner Syndrome.

The requirement for FP for the situations mentioned above is called medical FP, and there is an emerging medical speciality called oncofertility, which brings together oncologists, haematologists, physicians, fertility specialists, scientists, counsellors, nurses and researchers, all focused on maximising fertility opportunities for patients, at present and in the future [1]. The term oncofertility encompasses fertility, not only for patients with cancer but also for patients with non-malignant serious conditions wherein fertility is at risk.

FP can also be considered for women, who are not in a condition to have a baby currently and who wish, given the inexorable decline in fertility with age, to freeze their oocytes and thus to optimise the opportunity for future fertility.

In this chapter, we discuss the concept of medical FP; however, the scientific advances and the techniques are clearly referable to both the conditions mentioned above.

Why do we need fertility preservation programmes?

More than 80% of childhood and adolescent patients with cancer become long-term survivors [2]. Improvements in survival allow the expectation of a 'normal' life after cancer treatment, including having children. However, the compromise to fertility from many cancer treatments is profound [3,4]. Fertility is almost always compromised, at the very least temporarily, often with some resurrection of function. However, for young women receiving chemotherapy, given the reduction in the total follicular pool with cancer treatment, subsequent onset of premature ovarian insufficiency (POI) is to be anticipated, often at the time that young women, now cured of their cancer, would be wishing to conceive. Therefore, the risk to fertility needs to be addressed as early as possible after a diagnosis is made. There are considerable challenges associated with adding urgent fertility discussion and optimisation to the acute management of a cancer diagnosis in a young person. However, despite the trauma and distress during this time, patients and their families appreciate the opportunity for urgent fertility discussion and treatment, where possible, particularly as this represents optimism for their future. Therefore, it is essential for an operational FP programme to be able to provide discussion, information, treatment and organisation for follow-up, as a rapid response service.

Survivorship studies document the protracted and enduring psychological distress associated with not only post-cancer sterility but also perceived missed opportunity for pre-treatment FP, or at least discussion of the consequences [5].

International guidelines for fertility preservation

There are currently multiple international guidelines produced by craft groups including the American Society of Clinical Oncology (ASCO), European Society of Human Reproduction and Embryology (ESHRE) and the Clinical Oncology Society of Australia (COSA), all recognising the importance of, and mandating the requirement for, discussion and consideration for FP in patients with cancer whose fertility may be compromised [6–9].

Fertility preservation options

FP for girls and women

FP options for females include freezing oocytes (eggs), embryos and ovarian tissue as well as also administering medications before and during chemotherapy to try and protect the ovaries and the oocytes from the toxic effects [10].

Oocyte (egg) cryopreservation

Oocyte cryopreservation can be performed in post-menarchal girls and young women, ideally before chemotherapy and can also be considered after treatment in those who have regained ovarian

function [11]. The process involves supervised stimulation of the ovaries with gonadotrophin hormones for 10–14 days to produce multiple follicles. Monitoring is performed throughout this time with ultrasound and sometimes endocrine assessment. The follicles are aspirated in a minor surgical procedure, usually performed with sedation anaesthesia, using a needle attached to a transvaginal ultrasound probe through the vagina (Fig. 1). This procedure takes approximately 10–15 min. The oocytes are then extracted from the follicular fluid by in vitro fertilisation (IVF) specialists, and mature oocytes are cryopreserved by the process of vitrification. On average, 10–14 mature oocytes are obtained, but this depends very much on the patient's age and pre-existing ovarian reserve, and in young women, many more oocytes can be cryopreserved. The vitrification process allows excellent survival, with approximately 85–90% of oocytes to be available once thawed. However, as with fresh oocytes, there is considerable attrition in the subsequent course from mature oocyte to usable embryo stage, with approximately one to four blastocysts expected for every 10 oocytes cryopreserved (Fig. 2).

Fortunately, pregnancy rates approximate those from fresh oocytes, with a 35–40% chance of a clinical pregnancy from every embryo transferred in women with oocytes removed before the age of 38 years. It has been estimated that 6–10 mature oocytes frozen for women less than 38 years of age will provide a 30–50% chance of having a live birth; however, in clinical practice, fertility specialists would feel that 10–20 oocytes would give a more reasonable expectation of successful live birth [11].

The risks of this process are very low; however, these risks must always be considered for each patient situation. Apart from the time delay, the risks relate mostly to the sedation anaesthetic, hormonal stimulation and risk of infection, especially in a vulnerable patient. In the past, ovarian hyperstimulation syndrome complicated approximately 0.5% of cycles, with occasionally serious clinical features requiring hospitalisation. The use of gonadotrophin-releasing hormone (GnRH) agonist triggers, which dramatically reduce the risk of hyperstimulation and can be used in most patients, has really transformed the safety of the practice of stimulation for cryostorage [12].

There is no evidence that the use of gonadotrophins for 10–14 days increases any tumour-propagation risk in non-high-risk hormone-sensitive breast cancers, but usually an aromatase inhibitor or an anti-oestrogen agent will be given concurrently during the stimulation phase to reduce the level of oestrogen or block its interaction with breast cells [13].

Various stimulation protocols have been developed to maximise the opportunity for young women to undergo one or two cycles of oocyte cryopreservation within a limited time period and thus avoid the delay of commencement of chemotherapy, including the conventional start cycle and the 'random-start' cycle with comparable results [14].

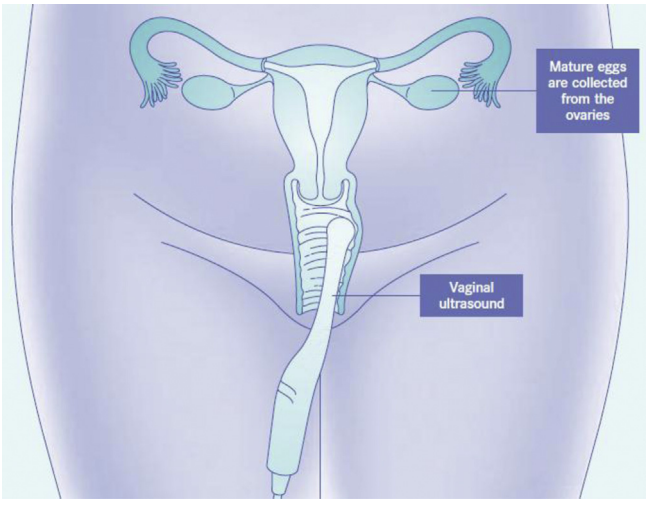

Fig. 1. Ultrasound-guided aspiration of mature oocytes.

Fig. 2. Oocyte attrition. The subsequent course from mature oocyte to usable embryo stage shows approximately one to four blastocysts ensue for every 10 oocytes cryopreserved.

Oocyte cryopreservation is currently considered as an established and generally safe method for FP, with the expectation of success almost equivalent to that from fresh oocytes. Reassuringly, oocyte yield and pregnancy rates for patients with cancer approximate those for people without cancer. However, the limited number of opportunities afforded by a small cohort of oocytes obtained must be considered when planning FP.

Embryo cryopreservation

For couples in established relationships, embryo cryopreservation may be considered [11]. Although there is no significantly higher chance of pregnancy associated with embryo cryopreservation than with oocyte cryopreservation, frozen embryos allow a more concrete estimation of success in the future, given that the stages of attrition from oocyte to embryo have already been completed.

However, embryos created with partner's sperm require partner (or ex-partner in the situation of a separation) involvement in plans for use, i.e. consent, with different jurisdictional implications. Therefore, it is possible that a woman may subsequently be prevented from using the cryopreserved embryos for fertility treatment, if consent for use is withdrawn by the ex-partner, even if she has no other opportunity to conceive with her own gametes.

Ovarian tissue cryopreservation and grafting

Storage of ovarian tissue, with hundreds or thousands or primordial follicles in the unstimulated ovary, provides the potential for many opportunities for fertility as well as restoration of ovarian function after grafting [15]. With more than 130 pregnancies and more than 100 births already variously reported, this form of FP is no longer regarded as experimental [16]. Improved data collection and registries currently allow a more realistic estimation of success, with reports of approximately 30% pregnancy rates after grafting [16]. Pregnancy can be achieved either spontaneously or with IVF treatment. However, it must be acknowledged that cycle dynamics in grafted tissue are different from standard treatment, with a low yield of good oocytes. Ovarian grafts may function for several years depending on the follicle density, age of the patient and exposure to toxic agents before cryopreservation.

The ovarian tissue cryopreservation process involves removal of ovarian tissue (either part of an ovary or the whole ovary), usually by a laparoscopic procedure; this is subsequently sliced into very small pieces and are cryopreserved, usually by slow freezing, but sometimes by vitrification, for grafting back later in the case of persistent ovarian failure [11].

This is the only FP option available for young girls, patients who require immediate cancer treatment and for those who would be at risk from a cycle of ovarian stimulation [17]. Ovarian tissue

harvesting may also be considered in addition to mature oocyte cryopreservation to maximise the opportunities for future fertility.

Grafting sites include the ovarian bed, pelvic side wall, pouch of Douglas and anterior abdominal wall (Fig. 3). On average, the tissue becomes functional approximately 3–5 months after grafting, with reduction in serum follicle-stimulating hormone (FSH), increase in oestradiol and restoration of menses in women with a functional uterus [15].

Apart from the surgical risks associated with the procedure, the main safety concern relates to the risk of tumour cell transmission in the tissue, particularly with leukaemia. Various strategies are being developed to allow subsequent safe grafting for women with leukaemia, including the diagnostic tools of immunohistochemistry, molecular biological techniques and xenografting [18]. Future therapeutic strategies may include the 'artificial ovary', purging of tumour cells from the tissue and in vitro follicle and even oocyte maturation [19].

Ovarian protection

It is possible to protect ovarian function, at least partially, during chemotherapy, by co-administration of a GnRH analogue [10,20]. Although the exact mechanism of protection is not yet conclusively understood, a reduction in the accelerated recruitment and destruction cycle during exposure to chemotherapy may be implicated [20].

Meta-analyses in patients with breast cancer have demonstrated a reduction in POI in patients receiving a GnRH agonist, with no increased risk of tumour progression or recurrence [10].

Other novel mechanisms including interruption of the apoptotic pathway are also currently being investigated with a view of developing additional therapeutic molecules [21].

FP for boys and men

Surgery, chemotherapy and particularly radiation can cause permanent damage to sperm production and sometimes impair testosterone production as well.

Semen cryopreservation

Conventional semen freezing, particularly when there is opportunity for collecting multiple samples, provides an excellent opportunity for future fertility, even if the semen is of poor quality, with subsequent use of IVF-intracytoplasmic sperm injection (ICSI). If semen quality is excellent and there are many straws in storage, then intrauterine insemination may be considered in the first instance, when patients are ready to attempt pregnancy [22].

Testicular biopsy for sperm freezing

For young men who are unable to produce a masturbated sample of semen, fine-needle testicular biopsy under local or general anaesthesia can be considered. Once removed, the seminiferous tubules are examined, and individual mature sperm are extracted and cryopreserved for later IVF-ICSI with excellent success rates [23].

Testicular biopsy for tissue freezing (for prepubertal boys)

FP for young boys provides a challenge, given the immaturity of sperm in the seminiferous tubules. Maturation of immature spermatogonia and spermatocytes to mature spermatozoa, capable of fertilisation of oocytes, is still highly experimental, with success only in animal models. Other hopeful strategies for the future include testicular tissue grafting [24].

Testicular tissue biopsy and cryopreservation is practised within a 'research' setting in some large paediatric institutional centres working with reproductive medicine units. Careful counselling is required for boys and their families given that this is still regarded as experimental FP.

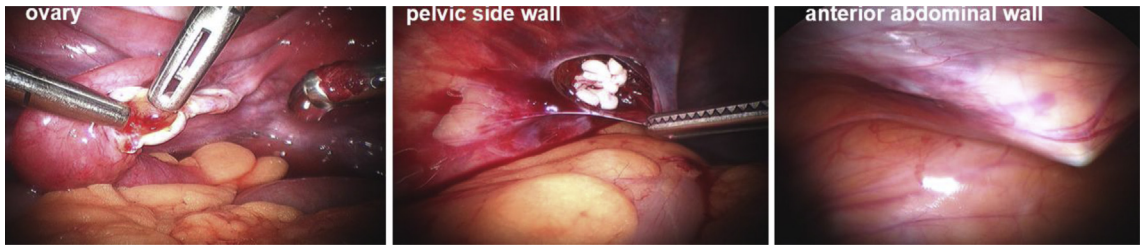

Fig. 3. Graft sites. Graft sites used are mostly determined at the time of operation depending on pelvic anatomy (such as the presence of adhesions) and past treatment such as radiotherapy.

Functions of a fertility preservation programme

Key components of a functioning FP programme

To provide appropriate care for patients, a FP service should aim to provide the following:

a) prompt referral for consultation and assessment of risk and urgent access to treatment options
b) multidisciplinary team
c) comprehensive range of services or strong referral pathways for additional services as required
d) short-, medium- and long-term follow-up

Additionally, a FP programme should aim to include the following:

a) rigorous data collection
b) public and professional education
c) strong collaborations with associated specialities including oncologists, haematologists and physicians
d) scientific and clinical research
e) procurement of funding

Logistics of a functional FP programme

Engagement of staff (multidisciplinary team)

A team approach is essential for a functional FP service. In addition to fertility specialists with a thorough working knowledge of fertility risks and the therapeutic modalities available, a nurse coordinator or administrator can facilitate communication between the different specialist groups and can optimally coordinate acute and follow-up care. Counselling for patients and their families is essential, not only to assist with decision making for the FP options but also to provide additional support during this time of acute distress and anxiety. The scientific team, with both clinical and research engagement, is also integral to the programme.

Close relationships between the oncology/haematology/physician/surgery teams and the FP team will facilitate timely and well-coordinated referrals (Fig. 4).

Accessibility to programme for patients and specialists

The key to a successful programme is prompt accessibility, and although this is less challenging in major urban centres, additional modalities may be required such as initial consultation by phone or Skype supported by online patient and practitioner resources. Some treatments may be possible at rural or regional sites, with transport of tissue or gametes for centralised processing and storage.

Rapid response for referrals and treatments

A robust acute referral pathway is essential, with Fast Fax referrals and accessibility to an on-call team ideal. To avoid delay for commencement of chemotherapy and other cancer treatments, it is essential to have comprehensive counselling and preparation for treatment facilitated through a 'rapid access' process. Where more than one modality is required, such as both ovarian tissue and oocyte cryopreservation, coordination is required between the different clinical and scientific teams.

Maintaining follow-up

There is always a risk of loss to follow-up, especially after completion of chemotherapy, when patients attend their oncology team for some years and then graduate to reduced follow-up, possibly without linkage to a 'late effects' service.

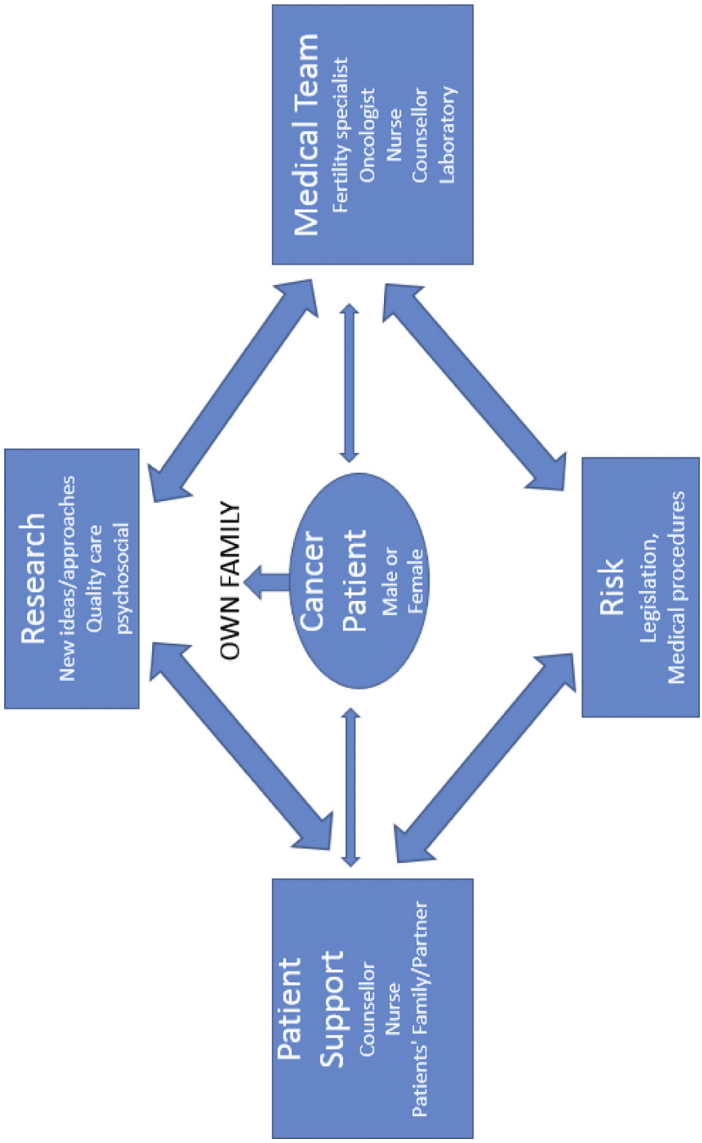

Fig. 4. The multidisciplinary approach to FP.

Long-term follow-up is essential for assessment of reproductive endocrine and sexual function, hormone replacement, if necessary, and opportunity for future FP if required. Given the possibility of temporary resurrection of reproductive function, patients may need advice for either/both contraception and fertility and pregnancy timing.

Storage of gametes, tissues and embryos is an ongoing responsibility for FP units, with associated legislative requirements. Patients may also find decision-making about storage distressing and require additional support to navigate the best path.

Data collection

Data collection is essential to assist with our understanding of the following aspects of FP:

1) magnitude of the problem
2) risk of POI
3) take-up rates of various options
4) pregnancy outcomes for patients who require FP
5) resourcing requirements
6) epidemiological, clinical and laboratory research aspects

Publication bias hampers our true understanding of treatment options for patients who require FP, particularly the less routine procedures such as ovarian tissue cryopreservation; therefore, there is a need for robust international registries, and this requires engagement from all who work in the area.

In addition, patient contributions to the FP literature will allow resource allocation and refinements of infrastructure to support the patient before, during and after their treatment.

Challenges

There are considerable challenges associated with medical FP for patients and their treatment teams. The acute distress and psychological trauma associated with the diagnosis of a potentially life-threatening condition, as well as the serious acute and long-term sequelae of therapy, require careful patient discussion and ongoing counselling. Accurate, comprehensive and also accessible and understandable information for patients assists them with feeling some control over management and looking forward to the future.

Even with optimal counselling and documentation at the time of diagnosis, the risk of damage from cancer treatment, and the finite (and sometimes quite small) additional future fertility benefit from cryopreservation, may be incompletely understood by patients and their families. Therefore, follow-up counselling is essential with a particular focus on reiteration of the fertility situation, firm plans for ongoing assessment and reassurance regarding options.

The logistic difficulties for patients from regional, rural and remote areas can be somewhat overcome with ready access to communication modalities with specialists and a coordinated approach to regionalised or even centralised FP services.

Narrow windows of opportunity are expected in FP, and the key to timely treatment is prompt referral and access to services and optimal communication between the oncology and the FP teams. Clear and accessible patient resources and family support resources are essential, and decision aids may also be useful [25,26].

Coordination of care within a range of specialities for patients with complex and often multiple medical issues can be challenging. The risks of anaesthesia with mediastinal disease and the haematological compromise associated with acute leukaemia require multidisciplinary discussion and evaluation of risk and benefit.

The affordability of FP, including consultations, treatment and storage, varies between centres, and more widely between countries, depending on support from the government and resourcing of FP, as well as the commitment of FP team members to provide their services for minimal cost to patients. There is no doubt that FP and reduction of risk of gonadal failure will allow future pregnancy at a lower cost than when reliant on donor gametes and hence will reduce the risk of childlessness.

Finally, a genuine understanding of the different personal circumstances, expectations and plans of each patient is required to facilitate an optimal and, importantly, an individualised approach to management.

Summary

The emerging speciality of oncofertility, or medical fertility preservation, brings together a committed team of clinicians from a range of specialties including scientists, nurses, counsellors, researchers and support staff. The team is enthusiastically focused on addressing the urgent issue of fertility compromise for young people, often in the midst of a life-threatening diagnosis and all the accompanying distress. The provision of accurate information and support, as well as realistic options for protection and preservation of fertility, to those young patients and their families, is absolutely achievable. It is also a fundamentally important part of reproductive medicine, in terms of both clinical processes and scientific advancement.

Research agenda

There are many exciting directions in fertility preservation currently being explored in a research context and that will hopefully result soon in an expansion of opportunities for young patients whose fertility is compromised by treatment for cancer or other serious diseases.

Successful births after uterine transplants have now been recorded; however, the technical difficulties mean that this is still considered to be highly experimental [27].

The technical processes for developing ovarian follicles into mature form in vitro (in vitro maturation or IVM), although demonstrated in animal models, are not yet successful in humans. This technology will, once clinically reliable, be an enormous step forward, given the large cohort of primordial follicles found in ovarian tissue samples and the current risks of grafting of tissue in patients with haematological malignancy [28].

Creation of an artificial ovary', in which follicles are grown into a mature form, will also expand the opportunities for patients whose tissue is at risk of tumour cell contamination [19].

Activation of primordial follicles and the ability to switch off follicle destruction are exciting areas of research with enormous potential, not only for patients with cancer but also for all women who have compromised ovarian function and are wishing to conceive [21].

Development of strategies for IVM of sperm will similarly expand opportunity for prepubescent boys to achieve fertility in the future [29].

Practice points

The key to a successful FP programme is to ensure that the following considerations are addressed:

- A strong connection between the oncology team and the fertility specialist is required.
- Identification of key medical contacts and a multidisciplinary team facilitates the steering of patients across specialties within the tight timelines necessary for FP in patients with cancer, such that consultations can occur within 24–48 h.
- FP consultation with a fertility specialist is a patient's main resource regarding fertility and FP options; other useful resources include decision aids, brochures and websites as well as contact with other key personnel within the FP team (nurses or counsellors) where follow-up by phone or email can assist with the imposed time constraints.
- The establishment of registries on the short- and long-term outcomes of FP techniques assists with addressing gaps in knowledge, identifying new areas of research and establishing referral pathways.

Conflicts of interest

Non-directional research funding was received from Merck-Serono and Ferring Pharmaceuticals. This funding was used towards our ovarian tissue research programme and to support the establishment of our national fertility preservation registry.

References

[1] Woodruff TK, Karrie AS. Oncofertility: fertility preservation for cancer survivors (cancer treatment and research). Chicago: Springer; 2006.
[2] Phillips SM, Padgett LS, Leisenring WM, Stratton KK, Bishop K, Krull KR, et al. Survivors of childhood cancer in the United States: prevalence and burden of morbidity. Cancer Epidemiol Biomark Prev 2015;24:653–63.
*[3] Anderson RA, Brewster DH, Wood R, Nowell S, Fischbacher C, Kelsey TW, et al. The impact of cancer on subsequent chance of pregnancy: a population based analysis. Hum Reprod 2018;33:1281–90.
*[4] Peccatori FA, Azim Jr HA, Orecchia R, Hoekstra HJ, Pavlidis N, Kesic V, et al., On behalf of the ESMO Guidelines Working Group. Cancer, pregnancy and fertility: ESMO Clinical Practice Guidelines for diagnosis, treatment and follow-up. Ann Oncol 2013;24(6):160–70.
*[5] Silva C, Almeida-Santos AT, Melo C, Ribeiro Rama AC. Decision on fertility preservation in cancer patients: development of information materials for healthcare professionals. J Adolesc Young Adult Oncol 2017;6(2):353–7.
*[6] Martinez F, International Society for Fertility Preservation–ESHRE–ASRM Expert Working Group. Update on fertility preservation from the Barcelona International Society for Fertility Preservation-ESHRE-ASRM 2015 expert meeting: indications, results and future perspectives. Fertil Steril 2017;108(3):407–15.
*[7] Lambertini M, Del Mastro L, Pescio MC, Andersen CY, Azim Jr HA, Peccatori FA, et al. Cancer and fertility preservation: international recommendations from an expert meeting. BMC Med 2016;14:1–16.
*[8] Oktay K, Harvey BE, Partridge AH, Quinn GP, Reinecke J, Taylor HS, et al. Fertility preservation in patients with cancer: ASCO clinical practice guideline update. J Clin Oncol 2018 Jul 1;36(19):1994–2001.
*[9] AYA Cancer Fertility Preservation Guidance Working Group. Fertility preservation for AYAs diagnosed with cancer: Guidance for health professionals. Sydney: Cancer Council Australia. [Version URL: https://wiki.cancer.org.au/australiawiki/index.php?oldid=170274, cited 2018 Jun 30]. Available from: https://wiki.cancer.org.au/australia/COSA:AYA_cancer_fertility_preservation.
[10] Lambertini M, Moore HCF, Leonard RCF, Loibl S, Munster P, Bruzzone M, et al. Gonadotropin-releasing hormone agonists during chemotherapy for preservation of ovarian function and fertility in premenopausal patients with early breast cancer: a systematic review and meta-analysis of individual patient-level data. J Clin Oncol 2018;36(19):1981–90.
*[11] Donnez J, Dolmans MM. Fertility preservation in women. N Engl J Med 2018;378(4):1657–65.
[12] Casper RF. Reducing the risk of OHSS by GnRH agonist triggering. J Clin Endocrinol Metab 2015;100(12):4396–8.
[13] Rodgers RJ, Reid GD, Koch J, Deans R, Ledger WL, Friedlander M, et al. The safety and efficacy of controlled ovarian hyperstimulation for fertility preservation in women with early breast cancer: a systematic review. Hum Reprod 2017;32(5):1033–45.
[14] Danis RB, Pereira N, Elias RT. Random start ovarian stimulation for oocyte or embryo cryopreservation in women desiring fertility preservation prior to gonadotoxic cancer therapy. Curr Pharmaceut Biotechnol 2017;18(8):609–13.
*[15] Gellert SE, Pors SE, Kristensen SG, Bay-Bjørn AM, Ernst E, Andersen C. 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(4):561–70.
[16] Andersen CY, Bollerup AC, Kristensen SG. Defining quality assurance and quality control measures in connection with ovarian tissue cryopreservation and transplantation: a call to action. Hum Reprod 2018;33(7):1201–4.
[17] Ladanyi C, Mor A, Christianson MS, Dhillon N, Segars JH. Recent advances in the field of ovarian tissue cryopreservation and opportunities for research. J Assist Reprod Genet 2017;34(6):709–22.
[18] Soares M, Saussoy P, Maskens M, Reul H, Amorim CA, Donnez J, et al. Eliminating malignant cells from cryopreserved ovarian tissue is possible in leukaemia patients. Br J Haematol 2017;178(2):231–9.
[19] Chiti MC, Dolmans MM, Orellana R, Soares M, Paulini F, Donnez J, et al. Influence of follicle stage on artificial ovary outcome using fibrin as a matrix. Hum Reprod 2016;31(2):427–35.
[20] Senra JC, Roque M, Talim MCT, Reis FM, Tavares RLC. Gonadotropin-releasing hormone agonists for ovarian protection during cancer chemotherapy: systematic review and meta-analysis. Ultrasound Obstet Gynecol 2018;51(1):77–86.
[21] Nguyen QN, Zerafa N, Liew SH, Morgan FH, Strasser A, Scott CL, et al. Loss of PUMA protects the ovarian reserve during DNA-damaging chemotherapy and preserves fertility. Cell Death Dis 2018;9(6):618.
[22] Abram McBride J, Lipshultz LI. Male fertility preservation. Curr Urol Rep 2018;19(7):49.
[23] Gies I, Oates R, De Schepper J, Tournaye H. Testicular biopsy and cryopreservation for fertility preservation of prepubertal boys with Klinefelter syndrome: a pro/con debate. Fertil Steril 2016;105(2):249–55.
[24] Radaelli MRM, Almodin CG, Minguetti-Câmara VC, Cerialli PMA, Nassif AE, Gonçalves AJ. A comparison between a new vitrification protocol and the slow freezing method in the cryopreservation of prepubertal testicular tissue. JBRA Assist Reprod 2017;21(3):188–95.
[25] Peate M, Smith SK, Pye V, Hucker A, Stern C, Stafford L, et al. Assessing the usefulness and acceptability of a low health literacy online decision aid about reproductive choices for younger women with breast cancer: the aLLIAnCE pilot study protocol. Pilot Feasibility Stud 2017;3:31.
*[26] Jones GL, Hughes J, Mahmoodi N, Greenfield D, Brauten-Smith G, Skull J, et al., On behalf of the Cancer, Fertility and Me research team. Observational study of the development and evaluation of a fertility preservation patient decision aid for teenage and adult women diagnosed with cancer: the Cancer, Fertility and Me research protocol. BMJ Open 2017;7(3), e013219.

[27] Wei L, Xue T, Tao KS, Zhang G, Zhao GY, Yu SQ, et al. Modified human uterus transplantation using ovarian veins for venous drainage: the first report of surgically successful robotic-assisted uterus procurement and follow-up for 12 months. Fertil Steril 2017;108(2):346–56.
[28] Lee J, Kim EJ, Kong HS, Youm HW, Kim SK, Lee JR, et al. Comparison of the oocyte quality derived from two-dimensional follicle culture methods and developmental competence of *in vitro* grown and matured oocytes. BioMed Res Int 2018; 2018:790–2.
[29] Oblette A, Rives N, Dumont L, Rives A, Verhaeghe F, Jumeau F, et al. Assessment of sperm nuclear quality after in vitro maturation of fresh or frozen/thawed mouse pre-pubertal testes. Mol Hum Reprod 2017 Oct 1;23(10):674–84.