FERTILITY PERSERVATION

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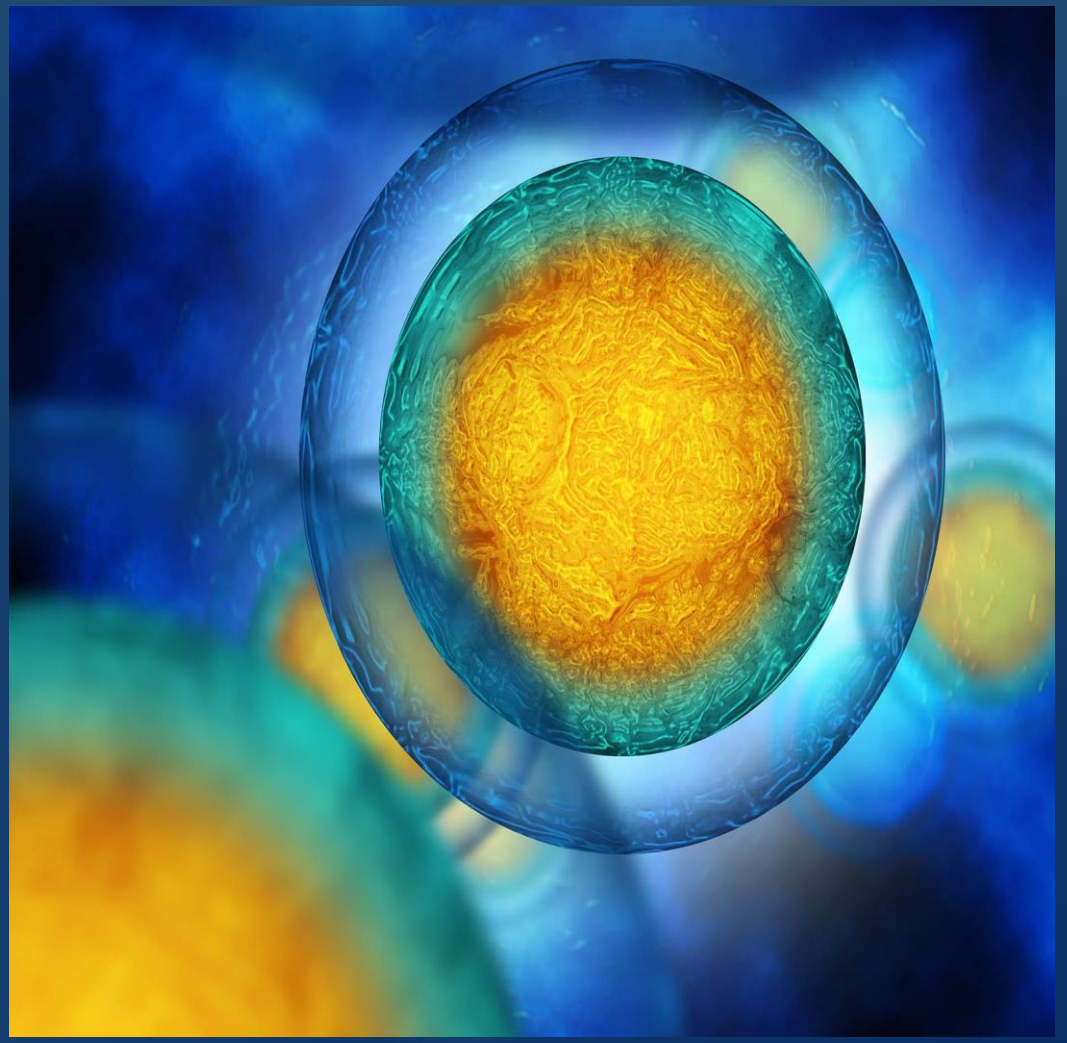
FETO-MATERNAL FELLOWSHIP

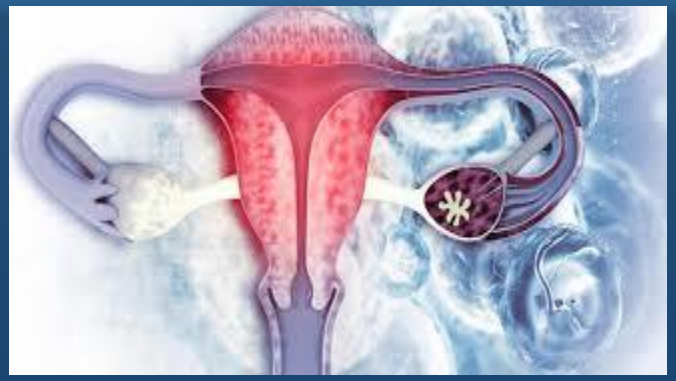
FERTILITY IN THE AGING FEMALE

- A woman's best reproductive years are in her 20s. Fertility gradually declines in the 30s, particularly after age 35.
- Each month that she tries, a healthy, fertile 30-year-old woman has a 20% chance of getting pregnant. By age 40, a woman's chance is less than 5% per cycle



EGG QUALITY

- Women become less likely to become pregnant and more likely to have *miscarriages* because egg quality decreases as the number of remaining eggs dwindle in number.
- These changes are most noted as she reaches her mid-to-late 30s.
- ***Therefore, a woman's age is the most accurate test of egg quality.***
- An important change in egg quality is the frequency of *genetic* abnormalities called aneuploidy



- As ovarian reserve declines, the follicles become less and less sensitive to FSH stimulation, so that they require more stimulation for an egg to mature and ovulate.
- At first, periods may come closer together resulting in short cycles, 21 to 25 days apart. Eventually, the follicles become unable to respond well enough to consistently ovulate, resulting in long, irregular cycles.
- Age does not appear to have a significant effect on morphological or histological responses of the uterus to steroid stimulation
- The prevalence of uterine pathology, such as fibroids and endometrial polyps, increases with age, yet there is little evidence that uterine factors have a significant impact on age-related infertility.

Female age and fertility

- Studies of populations where virtually no birth control is practiced demonstrate:

fertility begins to decline from age 25 .The decline in oocyte quality becomes clinically relevant for women from their mid-30s .

The prevalence of infertility increases from 1% at age 25 to 55% at age 45 . While 75% of women attempting conception at age 30 conceive within 12 months, by age 40 this has declined to 44%.

Further, 20% of women who conceive at age 35 will have a spontaneous abortion. The monthly fecundity resulting in live-birth rate among women aged 30, 35 and 40 year is 17%, 12%, and 5% respectively.

Age and ART outcomes

- Even with IVF, the likelihood of successful ongoing pregnancy is compromised by age.
- After the age of 30 years, the probability for ongoing pregnancy decreases by about 1.5% per year.
- The rate of implantation decreases by more than two-thirds after the age of 40 years
- Miscarriage rates after IVF cycles are 15% in women less than 35 years old, 25% in women at age 40 years, and more than 70% in women more than 44 years of age.
- The likelihood of success with IVF decreases because of a diminished response to stimulation, a lower chance of proceeding to oocyte retrieval and embryo transfer, and both lower pregnancy rates and live birth rates per transfer.
- Poor embryo quality. Results of studies evaluating chromosomal screening before embryo transfer have shown that there is a decline in reproductive potential of embryos even after controlling for aneuploidy

FERTILITY PRESERVATION

- Women who wish to delay childbearing until their late 30s or early 40s may consider methods of fertility preservation.
- In 2012 the average age of women in Australia and New Zealand who underwent ART treatment using their own oocytes was 35.8 years .
- The live delivery rate per fresh cycle was 26% among women aged under30 and less than 1% in women aged 44 or older.
- Some studies report that women over the age of 40 can have a greater than 5% chance for success in ART but no pregnancies are reported for women aged 46 or more who are using their own oocytes.
- Women who conceive with ART after the age of 40 have a greater risk of

miscarriage, gestational diabetes, pregnancy-induced hypertensive disorders, instrumental deliveries and caesarean sections than younger women

- an evidence-based guideline for the provision of fertility preservation
- evaluate all aspects of this topic in relation to its application for adult women and specifically to include its relevance to transgender men.
- Its application for prepubertal girls is not included comprehensively, although the application of ovarian tissue cryopreservation for this patient group is alluded to in the relevant section.
- Additionally, this guideline seeks to be inclusive regarding indications for fertility preservation
- Women are increasingly opting to cryopreserve oocytes for age-related fertility loss, a process often called “social egg freezing”. The medical and ethical aspects of this indication are also included in this guideline.

PART A: Organization and availability of fertility preservation (FP) care

- This guideline aims to help providers meet a growing demand for FP options by diverse groups of patients, including those diagnosed with cancer undergoing gonadotoxic treatments, with benign diseases undergoing gonadotoxic treatments or those with a genetic condition predisposing to premature ovarian insufficiency, transgender men, and women requesting oocyte cryopreservation for age-related fertility loss.
- Despite differences between the needs of these groups, FP care should be organized in an optimal way to accommodate all of them, taking into consideration the appropriate local legal context. With regards to organization of care, the most important difference between the patient types lies in the urgency of FP treatment.

- In order to improve the quality of health care for patients undergoing FP, a multi-level approach is necessary, addressing issues specific to:

1. The patient (and his/her partner and/or parents),
2. Professionals,
3. Organization (clinic, hospital),
4. Policy makers and general population.

The FP team should consist of fertility specialists, and embryologists, but should also include a psychologist or counsellor.

A2: Legal aspects and availability

- Data on whether fertility preservation is allowed in European countries, for which indications and under which conditions, were collected in an online survey.
- In general, oocyte cryopreservation for FP is allowed in all countries for which data were collected.
- Embryo cryopreservation for FP is also allowed in all countries except for Italy and Portugal.
- Ovarian tissue cryopreservation for FP is allowed in 30 countries

A3: HOW LONG SHOULD REPRODUCTIVE MATERIAL BE STORED?

- With data for 30 countries, With regards to storage, a duration of 5 or 10 years is most often reported, and this is mostly extendable.
- The age limit for use of the oocytes is reported to be ranging from 42 to 55 years.
- Eight countries reported that there were no limits for duration of storage of oocytes, nor for the use of the stored gametes.
- Similar results were found for embryo cryopreservation

PART B: Patient information

Checklist 2 Checklist for clinicians to cover the information needs of patients undergoing fertility preservation counselling

Information needs	Cancer patients	Medical (non-cancer) patients	Trans-gender men	Women undergoing FP for age-related fertility loss
1) Impact of disease/treatment on reproductive function				
Menstrual changes/Amenorrhoea	√	√	√	-
Premature ovarian insufficiency	√	√	√	-
Information about contraception	√	√	√	-
2) Impact of disease/treatment on fertility				
Effects of disease on fertility	√	√	-	-
Effects of treatments on fertility / risk of infertility	√	√	√	-
Effects of hormonal therapy on fertility	√	√	√	-
3) Fertility preservation options				
Effects of hormonal stimulation for FP on disease recurrence	√	√	-	-
Impact of age at the time of FP on success rates	√	√	√	√
Fertility preservation options				
- Established and experimental FP techniques	√	√	√	√
- Time requirements of each FP option	√	√	√	√
- Success rates of each FP technique	√	√	√	√
- Pregnancy rates after each FP option	√	√	√	√
- Risks of each FP technique	√	√	√	√
- Side effects of each FP technique	√	√	√	√
- Advantages of each FP technique	√	√	√	√
- Disadvantages of each FP technique	√	√	√	√
- Costs of each FP technique	√	√	√	√
Late FP options[1]	√	√	√	
Ethical issues associated with embryo cryopreservation	√	√	√	√
4) Cryopreservation and storage of cryopreserved material				
Maximum time for cryopreservation	√	√	√	√
Costs of cryopreservation	√	√	√	√
5) Infertility and fertility treatments				
Infertility and Medically assisted reproduction treatments	√	√	√	√
6) Pregnancy				
Risk of disease recurrence due to pregnancy	√	-	-	-
Risks/benefits of having children after cancer/other diseases	√	√	-	-
Effects of disease/treatments on future children	√	√	√	-
Obstetric risks	√	√	√	√
7) Childbearing/Parenting options				
Reproductive planning after disease/treatment/other situations	√	√	√	√
Other options to achieve pregnancy/parenting	√	√	√	√

[1] Including FP after completion of cancer treatment or other treatments for non-malignant diseases. For transgender men, this implies FP options after the start of gender-affirming hormone therapy

- here is no direct evidence on the information needs of patients at risk of infertility due to other medical situations, gender reassignment therapy or oocyte cryopreservation for age-related fertility loss, but it seemed relevant to expand the recommendations to be also applicable to these patient groups
- Although the information can be stressful and difficult to handle by some patients, being informed about the possibility of FP is associated with better outcomes (the benefits seem to outweigh the harms).
- **It is recommended to provide decision aids to patients who are considering FP.**

Partc: Patient selection and pre-FP assessment

- **Intrinsic factors**
- □ Health status of patient
- o Surgical/anaesthetic risk, including thrombosis, infection, and mediastinal masses
- o Malignant contamination of the ovary

- □ The need to obtain fully informed consent (patient/parent)
- □ Age (upper and lower limits for safety and efficacy)
- □ Assessment of ovarian reserve

- **Extrinsic factors**
- □ Nature of predicted treatment o High/medium/low/uncertain risk of POI/infertility
- o Other risks relating to pregnancy e.g. cardiac toxicity
- o Uterine radiotherapy

- □ Time, availability of local resources, expertise, and local criteria/funding
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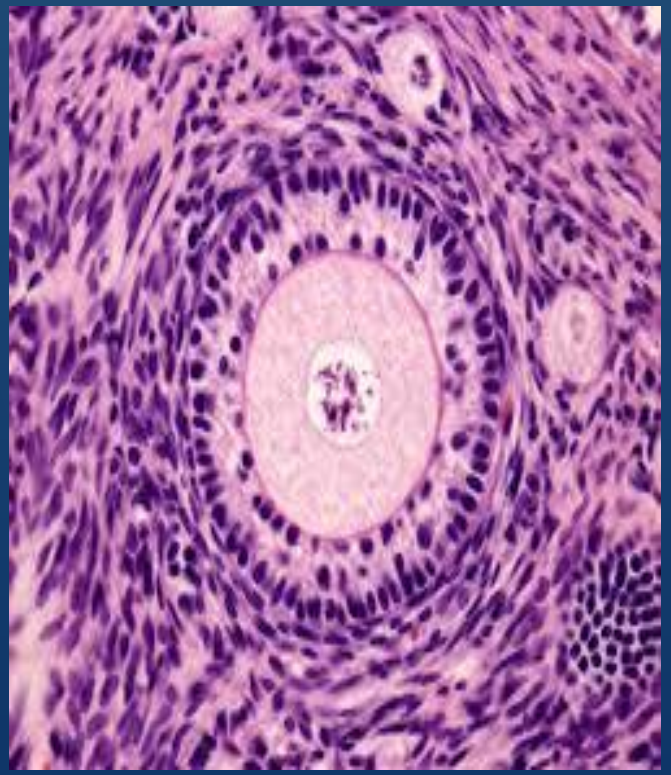
Ovarian reserve testing

- AFC and serum levels of AMH appear to be the most promising markers mainly due to their low intercycle variation and ease of measure.

- According to the Bologna criteria for the definition of "poor ovarian response" (POR), ***cut-off levels for AFC are less than 5 to 7 follicles, and AMH levels below 0.5-1.1 ng/ml***



ovarian reserve

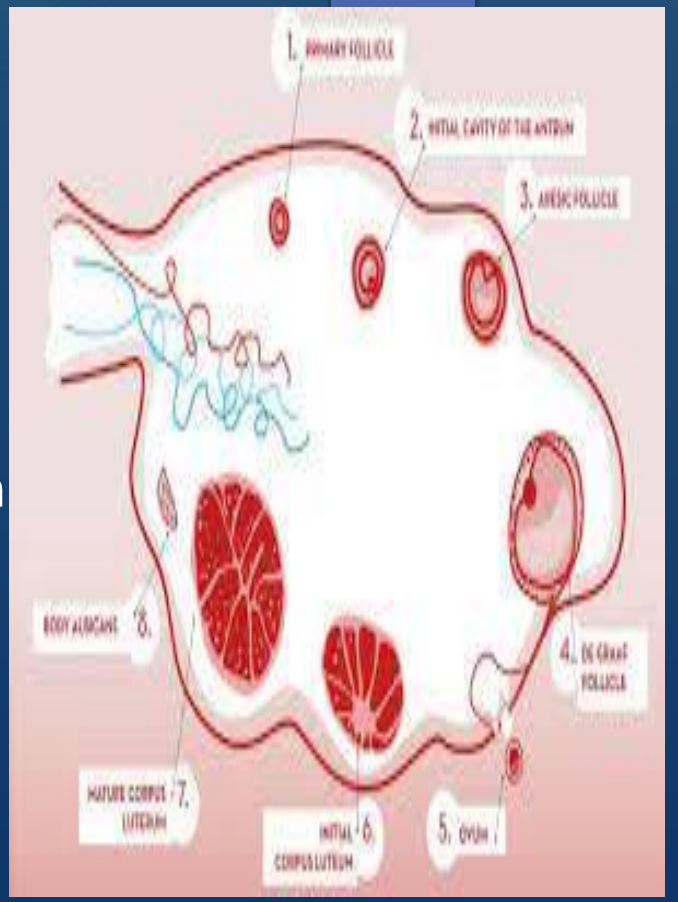
- testing has mainly been performed on infertile populations, with little data on the distribution in the normal fertile population.
- Ovarian reserve testing cannot be used to predict infertility or time to infertility; therefore, its application to the general population as a screening tool is untested. Most studies have used these tests to try to predict a woman's ovarian response and prognosis with fertility treatment and IVF.
- Overall, markers of ovarian reserve have been shown to correlate with egg quantity and response to ovarian stimulation, but not with egg quality.

- The most commonly used test of ovarian reserve is the cycle day 3 or basal FSH level. An elevated basal FSH level (> 14 IU/L) is the first sign of ovarian aging that can be detected in women, and usually occurs in women aged 35 to 40. Physiologically, the follicular pool is reduced to approximately 10% of the levels present at puberty.
- However, basal FSH levels have been shown to be predictive for poor response to ovarian stimulation and for non-pregnancy only when the levels are extremely elevated. Although a high threshold may improve the usefulness of the test in predicting a poorer prognosis, only a small number of women will have abnormal tests at this threshold.
- In addition, it has been associated with a false positive rate of 5%.
- Elevated basal FSH levels are also less predictive of pregnancy for women < age 35.

- Ovarian reserve testing may be considered in women > age 35 to screen for age-related infertility, although its results may be useful only for counselling and to aid women in their decision-making process.
- Testing in women < 35 years may be considered if they have risk factors for

decreased ovarian reserve, such as a single ovary, previous ovarian surgery, poor response to FSH, previous exposure to chemotherapy or radiation, or unexplained infertility.

- Although markers of ovarian reserve are not good predictors of pregnancy rate with ART for women < 35, identification of these women may prompt shorter delay to infertility investigations and treatment



IS IT RELEVANT TO DO OVARIAN RESERVE TESTING, AND FOR WHOM? (Women requesting oocyte cryopreservation for age-related fertility loss)

- Both **ovarian reserve** and **age** are the important patient features that determine the ovarian response to stimulation.
- There is a clear correlation between AFC and serum AMH levels with oocyte yield to stimulation
- Therefore, ovarian reserve testing is commonly used to tailor ovarian stimulation strategies and maximize follicular recruitment if a poor response is anticipated.
- However the ability of ovarian testing using AMH levels to predict embryo quality and chances to conceive has not been demonstrated.
- **Therefore, ovarian reserve testing should not be performed for making FP decisions.**

PART D: Fertility preservation interventions

- **EMERGENCY AND NON-EMERGENCY?**
- cryopreservation of oocytes, embryos or ovarian tissue
- **oocyte cryopreservation is the method of choice for women undergoing treatment for age-related fertility loss**, and for most women undergoing fertility preservation for medical indications.
- Embryo cryopreservation is even more widely available and long-established part of assisted reproduction
- Ovarian tissue cryopreservation is an important option either through choice, or if there is insufficient time for ovarian stimulation

Choice of cryopreservation of oocytes versus embryos ?

- Cryopreserved oocytes will always belong to the woman. If a couple is being treated, resulting in embryo storage, the embryos belong to the couple
- A recent prospective study of FP investigating trends in patients' choices found that more than half of the women with a partner chose either not to fertilize their oocytes aiming at cryopreservation of oocytes only or to share obtained oocytes attempting both cryopreservation of oocytes and cryopreservation of embryos
- Women should receive information on the relevant legal issues
- More accurate data on CLBR after oocyte and embryo vitrification for FP for medical indications would also be of value to inform patients who have a choice as to what to cryopreserve.

Current guidelines on oocyte cryopreservation for age-related fertility loss

- International guidelines **generally support** oocyte cryopreservation for age-related fertility loss but recommend that it should be used with caution.
- The ESHRE Ethics taskforce paper states: '**It is concluded that the arguments against allowing this application of the technology are not convincing**' . However, they stress the need, '**for adequate information of women interested in oocyte cryopreservation, to avoid raising false hopes**.
- Recent guidance from the American Society for Reproductive Medicine (ASRM) states: 'The Committee concludes that planned oocyte cryopreservation may allow women who, in earlier times, would have faced infertility and childlessness to potentially have a child to whom they are genetically linked. **Planned oocyte cryopreservation is an ethically permissible medical treatment that may enhance women's reproductive autonomy and promote social equality**' (2018).
- A recent Royal College of Obstetricians and Gynaecologists (RCOG) Scientific Opinion piece on the topic stated that " **elective egg freezing for non-medical reasons provides an opportunity for women to mitigate the decline in their fertility with age**, but highlight that women undertaking oocyte cryopreservation should only do so with a full understanding of the likelihood of success, as well as costs and risks

Debates over oocyte cryopreservation for age-related fertility loss

- Arguments that are supportive point to the possible benefits the procedure might produce. It can be seen as a useful procedure that can extend women's fertility options and in doing so enhance an individual's reproductive autonomy.

- reproductive choices are a very important, central aspect of peoples' lives.
- It has also been argued that it can possibly alleviate the gender inequality created by women and men having different age-related biological fertility decline, by allowing women to extend their reproductive years.

- In their summary of the arguments the ASRM state: ‘Planned oocyte cryopreservation may also promote social justice by reducing the obstacles women currently face because their reproductive window is smaller than men's.’ The cost of oocyte cryopreservation for age-related fertility loss may conversely increase social inequality as it is only available to women who can afford the significant financial outlay.

- It is possible that oocyte cryopreservation for age-related fertility loss could be better for any future child, as this technology gives people more time to prepare, become financially secure, and women will not rush into reproducing when they are not ready or they have not met the ‘right’ partner.

objections to oocyte cryopreservation for age-related fertility loss

- A key objection is that it further **medicalizes reproduction**, by offering a medical solution to what is arguably a societal problem. It could also lead to greater commercialization of reproduction, with the use of inappropriate high-pressure sales practices.
- However, both ESHRE and ASRM have concluded that oocyte cryopreservation for age-related fertility loss does not produce substantial harms and there are no convincing arguments to restrict its use or only employ it for fertility loss due to particular medical conditions and their treatment.
- highlight the need for women to be fully informed of the likelihood of success, as well as costs and risks to mitigate any possible harms.
- The RCOG also highlights the need for education on the age-related impact on fertility. It is also worth noting that how far oocyte cryopreservation for age-related fertility loss delays having a child depends on the situation of the individual woman. Given common age restrictions on IVF treatment, often no older than 45, it may not offer a significant number of 'extra' childbearing years.

Issues with oocyte cryopreservation for age-related fertility loss

- Success rates: oocyte cryopreservation had a very high cumulative live birth rate (CLBR) for those who froze before they were 35 years old approaching 95% in cases with 24 or more utilized thawed oocytes (with a 42.8% CLBR from 10 oocytes)
- However, a maximum CLBR of 50% was achieved by those who froze when they were over 35, after using 20 or more thawed oocytes,(with CLBR of 25.2% with 10 oocytes frozen).
- Thus, age at cryopreservation is key and patients should be made aware of this

- **Likelihood of using the oocytes:** 12.1% of women returned to use their oocytes, with a mean storage time of 2.1 years.
- 29.2% of women indicated that they currently consider the use of frozen oocytes less likely than anticipated at time of oocyte pick up.
- **Medical harms**: These risks are expected to be low as women cryopreserving oocytes for age-related fertility loss are likely to be healthy.
- **Obstetric risks**: Importantly, there are risks of due to older age at pregnancy. These risks increase after the age of 45.
- **Long-term data:** Studies on the long-term effects on both safety and efficacy of cryopreserved oocytes are lacking due to the relative novelty of these techniques.
- **Risks to the future child**: There could be long-term consequences of oocyte cryopreservation on health of the child and possible, as yet unspecified, psychological effects

- **Funding of these procedures:** It is unlikely that these 'elective' techniques for healthy women will be funded by state health provision/insurance. Funding availability will depend on the healthcare system, but it is unlikely that any system will provide adequate funding for all those who might want to access oocyte cryopreservation for age-related fertility loss. Women should be informed of all costs involved, including for ongoing storage and later use of their oocytes.

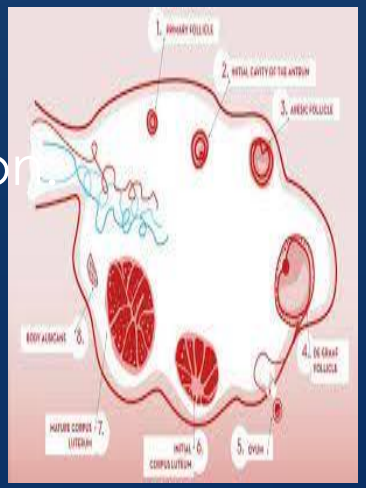
- **Company sponsored oocyte cryopreservation**: There are also ethical issues raised by companies offering to pay for women to cryopreserve their oocytes, such as coercion and manipulation, that might make women may feel that they are not able to take time off to have children

- **Consent and counselling procedures**

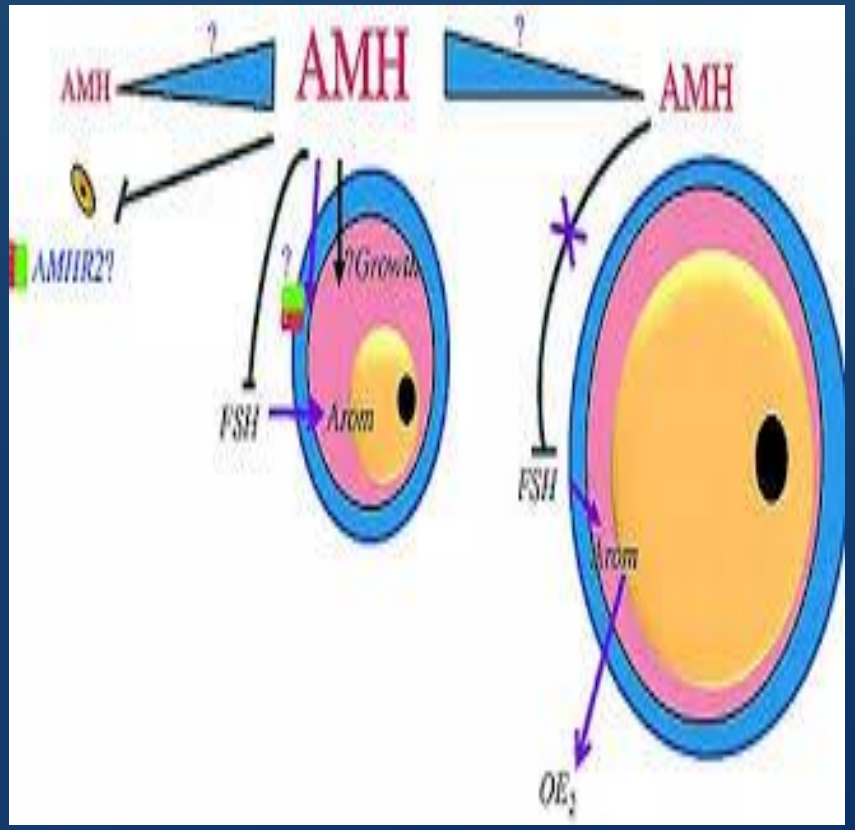
- **Psychological counselling** (in addition to counselling specific to FP) is usually offered to fertility patients and should be routinely offered to those considering oocyte cryopreservation for age-related fertility loss.

- Oocyte cryopreservation for age-related fertility loss is recognized internationally as an acceptable option to offer women with appropriate cautions and safeguards

- An ovarian antral follicle count can be performed early in the menstrual cycle.
- The number of antral follicles seems to correlate with the number of primordial follicles in the ovary, with a decline in primordial follicles being reflected in a lower number of antral follicles.
- In later reproductive years, the proportion of antral follicles to total follicles may increase as the ovary allows a higher proportion of follicles to be selected.
- The decline in AFC may not be as steep as the decline in fertility. Although decline in AFC is correlated with both the menopause transition and ovarian response to stimulation, it is not a good predictor of pregnancy
- An AFC of fewer than 3 to 4 follicles has a sensitivity of 9% to 73% and a specificity of 73% to 100% for predicting poor response to ovarian stimulation



- Antimüllerian hormone is produced by the granulosa cells of pre-antral and small antral follicles
- AMH levels decrease with decreasing AFC, which in turn is a marker of the primordial follicle count.
- Levels remain consistent throughout the menstrual cycle and become undetectable in women after menopause.
- Although AMH provides moderate value in prediction of ovarian response in IVF, it is a poor predictor of pregnancy.
- Cutoff values between 0.2 and 0.7 ng/mL have a sensitivity of 40% to 97% and a specificity of 78% to 92% in predicting poor response to ovarian stimulation.



- The clomiphene challenge test is performed by administering 100 mg of clomiphene daily from day 5 to day 9 of the cycle. FSH is measured on day 3 and on day 10.
- If an adequate response to clomiphene is generated, the rise in FSH will be suppressed by the release of estradiol and inhibin-B by developing follicles.
- Systematic reviews have not shown a benefit to the clomiphene challenge test over basal FSH or AFC.
- Inhibin-B and basal estradiol have not been shown to be more useful predictors of poor response or pregnancy than basal FSH. However, basal estradiol levels are often screened in conjunction with FSH and can confirm correct timing in the menstrual cycle. An elevated estradiol level may also falsely suppress FSH levels
- The CCCT has higher sensitivity (13%–66%) but lower specificity (67%–100%) than basal FSH values alone

Age at which to freeze oocytes

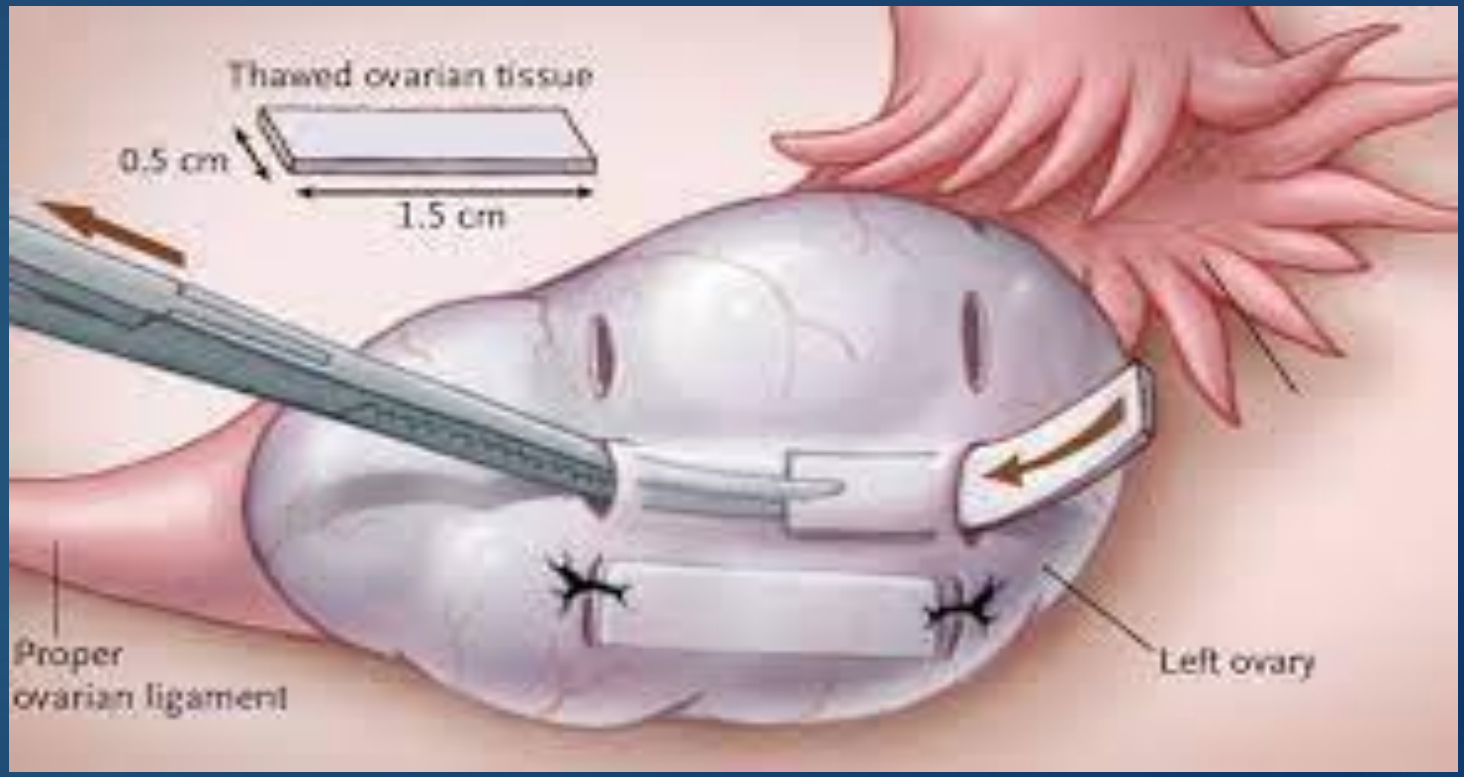
- Although no good answer exists as every patient has unique biological and social circumstances, there is some evidence available to guide the discussion. Similar to IVF, success rates following OC may be related to the age at which the oocytes were frozen.
- A retrospective multicenter study evaluating 1468 women undergoing elective oocyte cryopreservation for reasons other than a diagnosis of cancer revealed that 191 (13%) presented to thaw and inseminate their oocytes . Of those that did, the oocyte survival rate in women ≤35 y/o and ≥ 36 y/o were 94.6% vs. 82.4%, respectively .
- Additionally, differences in live birth rates for women undergoing OC ≤ 35 y/o and ≥ 36 y/o were significantly different, with 50% and 22.9%, respectively

how many oocytes should be frozen

- Although there is no precise answer to this question as every women presenting for OC will be at a different age and have different future childbearing desires, there is some data to guide a discussion.
- A recent mathematical model based on live birth rates from a single institution from women with presumably normal ovarian reserve undergoing intracytoplasmic sperm injection (ICSI) due to male factor infertility or tubal factor infertility, revealed that for women 34, 37, and 42 y/ o, they would need to freeze 10, 20, and 61 mature oocytes respectively to have a 75% likelihood of having at least 1 live birth

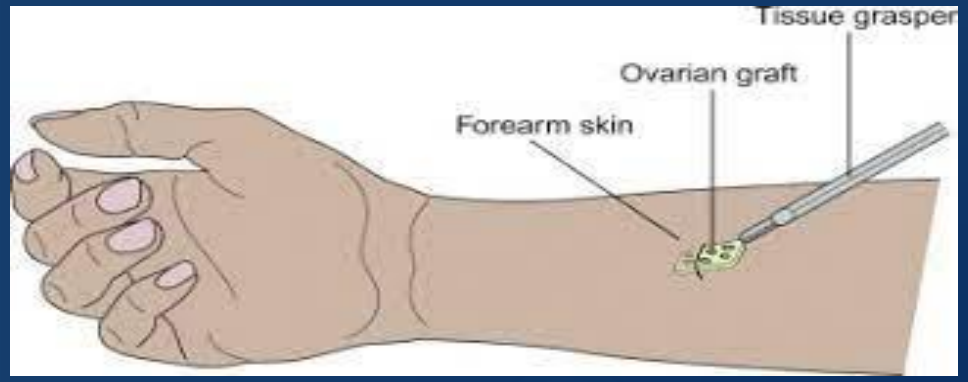
Ovarian cortex cryopreservation

- Fresh or frozen ovarian cortex transplantation might be more efficient than freezing ovarian tissue and not transplanting it back into the patient until 10 or even 20 years



Transplantation techniques

- Although almost all these pregnancies have been achieved with orthotopic ovarian tissue transplantation and most women had spontaneous pregnancies, a human patient with a heterotopic ovarian tissue transplant with in-vitro fertilization and pregnancy has been reported in Australia
- This success indicates that despite the popularity of orthotopic ovarian tissue transplants, heterotopic ovarian tissue transplantation has advantages (easier access for in-vitro fertilization and to monitor potential tumour recurrence) and could become more popular.



Embryo cryopreservation

- Embryos at the cleavage stage can be successfully cryopreserved by slow-freezing or vitrification. vitrifying day-3 embryos and blastocysts has now become the method of choice for embryo cryopreservation.
- Furthermore, transferring vitrified warmed embryos has been shown to be at the very least as efficient as fresh embryo transfer, if not better.
- Indeed, the 2015 US national report on assisted reproductive technology revealed a LBR of 48% per frozen embryo transfer compared to 46.5% per fresh embryo transfer

Ovarian tissue cryopreservation

- Ovarian tissue cryopreservation for the purposes of autotransplantation does not require ovarian stimulation and can therefore be performed immediately, with no delay in cancer treatment. Furthermore, it does not require sexual maturity, which makes it the only fertility preservation method available to prepubertal girls at risk of treatment-inducedPOI. It also restores general ovarian endocrine function.

- Certain selection criteria clearly need to be applied before contemplating OTC. the most important being age less than 35 years
- The amount of ovarian tissue to be harvested for cryopreservation purposes varies according to the risk of POI and existing ovarian volume. Oophorectomy is usually indicated in case of pelvic radiotherapy or total body irradiation and in very young girls due to the small size of the ovaries. Otherwise, four to five fragments of tissue (10 × 5 × 1 mm) are typically recovered by laparoscopy and then processed for freezing. With regard to biopsy thickness, based on the results of earlier studies, it is recommended that a 1–1.5-mm-thick piece of ovarian cortex be taken. This is considered to be of paramount importance, since superficial or very thin biopsies may not contain follicles in the removed cortex, as primordial follicles are generally found at a distance of 0.8 mm from the mesothelium.

- Orthotopic transplantation involves grafting ovarian cortical fragments to the exposed medulla of the denuded ovary or a specially created peritoneal site. Orthotopic transplantation takes advantage of the anatomical position of the ovaries for possible natural conception if other conditions are met: permeable fallopian tubes, no male factor issues and restoration of ovarian function after transplantation.
- The majority of reported orthotopic transplantations are carried out by minimally invasive surgery. The choice of grafting site and decision to graft in one or more locations depend on whether or not the patient previously underwent complete unilateral or bilateral oophorectomy. Indeed, if at least one ovary is present, ovarian tissue may be transplanted both to the ovary after decortication and to a newly created peritoneal window. On the other hand, if no ovaries remain or what is left is severely atrophic or nonfunctional (due to the effects of high doses of radiotherapy), the only alternative for orthotopic transplantation is use of a peritoneal window.

There are currently different approaches that may be applied depending on the presence or not of the ovaries, and the amount of tissue available for thawing and reimplantation:

- If at least one ovary is present, decortication of the ovary should be performed first. A relatively large piece of ovarian cortex (1–2 cm) is removed by means of scissors to have access to the medulla and its vascular network. Strictly adhering to microsurgical techniques and principles, ovarian cortical pieces are then simply placed on the medulla and covered by Interceed, the edges of which are fixed with stitches or fibrin glue

- If both ovaries are absent, a peritoneal window may be created in two or one steps to induce angiogenesis before the grafting procedure. The incision for this peritoneal window is made on the anterior leaf of the broad ligament in an area where a vascular network is visible (retroperitoneal vessels). Vessels may be easily localized by transillumination using the optical light of the instrument. Fragments are placed in the window and subsequently covered with Interceed, the edges of which are fixed with fibrin glue

- A third option for patients with one or two ovaries still in place is grafting the tissue to both orthotopic sites simultaneously (if there is enough ovarian tissue), namely to the denuded ovary and the peritoneal window. With this type of transplantation, it is of utmost importance to be circumspect with amounts of tissue used, in case further reimplantations are needed in the same patient. It is recommended that only on third of a patient's cryopreserved tissue be thawed and grafted for each transplant.

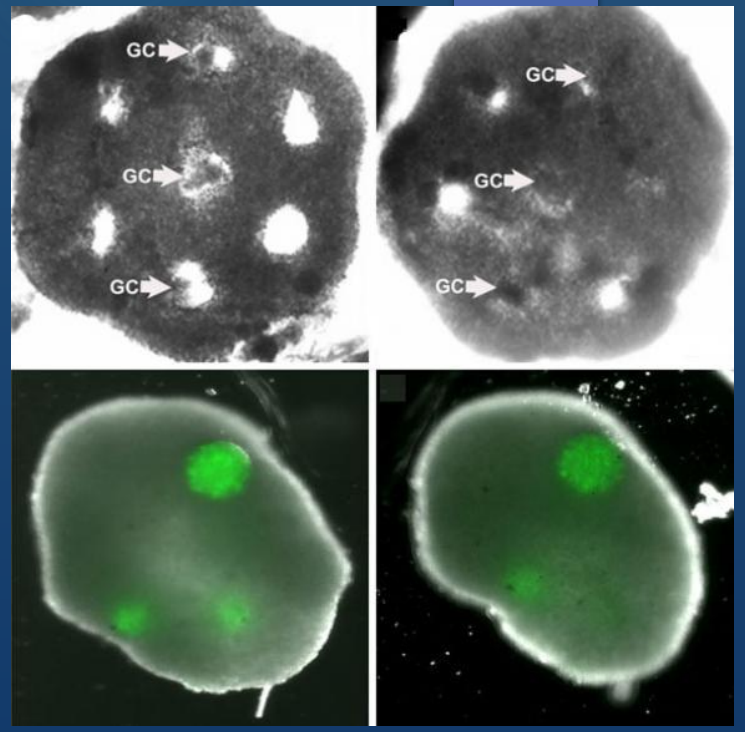
- Heterotopic OTT Common sites: for heterotopic transplantation are the abdominal wall, forearm and rectus muscle, among others. While heterotopic ovarian transplantation has debatable clinical value, as it may not provide an optimal environment for follicle development, restoration of endocrine function and embryo development have been demonstrated consistently after this procedure. However, its effectiveness for restoration of fertility remains questionable, as the success rate in terms of live births is extremely low.

Restoration of endocrine function

- Restoration of ovarian endocrine function is measured by either follicle growth or recurrence of menstruation. Several teams have found that around 95% of transplanted women experiences return of menses and back-to-normal hormone levels.
- The period from the first OTT to observed follicular function is on average 4 months and ovarian activity usually persists for more than 5 years. Moreover, by repeating the procedure, ovarian activity could be restored for longer depending on the amount of stored tissue and patient age at the time of OTC.These encouraging results on ovarian function restoration lead us to speculate that in the future, OTC at a young age, followed by reimplantation at menopause, could well provide an alternative to hormone replacement therapy

Artificial Ovary:

- One alternative to obtaining mature oocytes would be using the so-called transplantable artificial ovary . Isolating primordial follicles and transferring them onto a scaffold to create this artificial organ would serve to eliminate the risk of transmission of malignant cells. Recent developments in the isolation technique, involving washing the follicles three times, have proved successful. Growing antral follicles were observed after autografting primordial follicles inside a fibrin scaffold in a mouse model and after xenografting human primordial follicles in mice with severe combined immunodeficiency



An engineered honeycomb of cultured theca cells (top row) envelopes spheres of granulosa cells (GC). The bottom row shows the tissue after 48 hours (left) and after five days.*Carson Lab / Brown University*

In Vitro Development of Primordial Follicles:

- A dynamic multistep culture system is needed to support each of the transitional stages of follicles. This multistep approach to in vitro follicle growth must meet the changing requirements of the developing oocyte and its surrounding somatic (granulosa) cells in order to maintain interactions between these cells. The challenges, such as acquisition of meiotic and developmental competence as well as genome imprinting, are numerous.

Ovarian Stem Cells

- The discovery of ovarian stem cells has challenged the theory that production of germ cells in female mammals ceases before birth
- However, in vitro derivation from ovarian stem cells might interfere with the complex genomic imprinting and epigenetic mechanisms required fo

New Avenues of Research

- Allografting The first live birth to occur after ovarian-tissue transplantation between two genetically different sisters was reported in 2011. Since this is an acceptable practice with monozygotic twins, there is no apparent reason to refrain from using it with genetically different sisters, especially if one of the sisters previously received bone marrow from the other,leading to complete chimerism (HLA compatibility) between the sisters and obviating the need for immunosuppressive treatment. This approach allows for natural conception, which could be important on moral, ethical, or religious grounds.

- Ovarian tissue cryopreservation remains the technique of choice in preserving fertility in prepubertal females or postpubertal females in whom controlled ovarian stimulation (COS) with gonadotropins is contraindicated.
- Ovarian tissue cryopreservation may hold promise in the future for FP in the premenarchal girls, even though it is still labeled as experimental by the Practice Committee of the American Society for Reproductive Medicine. Approximately 100 live births have been reported via transplantation of cryopreserved ovarian tissues.
- The process of OTC involves surgical removal of the whole, or part, of an ovary and dissection of the tissue to prepare fragments of the ovarian cortex for cryopreservation. It is worth mentioning that a new trend, which involves freezing of whole ovarian tissue for its later transplantation through reanastomosis, has been elucidated.

- The main limitation of ovarian cortical grafting is the reduced graft's life span, as the graft generally loses two-thirds of its follicle reserve because of posttransplantation ischemic injury. Data from animal studies suggest that whole ovary transplantation can overcome this problem.
- Sound surgical transplantation techniques and well established cryopreservation protocols that can ensure the diffusion of cryoprotectants through the entire organ are key issues for success


Thank you
for your
attention