Médecine de la Reproduction 2018 ; 20 (1) : 11-8

Oocyte cryopreservation to anticipate age-related fertility decline

AGE banking ou la prévention de l'infertilité liée à l'âge

Michel De Vos
Christophe Blockeel

Centre for Reproductive Medicine, Universitair Ziekenhuis Brussel, Vrije Universiteit Brussel, Belgium
<Michel.devos@uzbrussel.be>

Abstract. In contrast to men who produce new sperm cells up to old age, women are born with a limited number of oocytes that will decrease with age. No new eggs are added, and oocyte quality is reduced with advancing age. As female reproduction is strongly influenced by a society in change, women may have various reasons to postpone childbearing, mostly because the preconditions for having a child are not yet fulfilled. Awareness rising has led to an increased demand for oocyte cryopreservation although there is controversy among bioethicist whether this technology will emancipate women or whether it may disempower them instead. The technique of vitrification has outgrown the experimental stage for some time now. Although follow-up of the health of the children who conceive after cryopreservation of oocytes has yielded reassuring results, the take-home baby rate of women who embark on social freezing is yet unknown and the danger of false hope is lurking behind optimistic advertisements from commercial oocyte cryopreservation companies. Indeed, the chances of having a child with vitrified oocytes are significantly reduced beyond the age of 36 (age of cryopreservation). Ovarian stimulation protocols for oocyte cryopreservation have been adopted from the ART clinic and are focused on maximised output of mature oocytes, whilst optimising the safety profile and burden for those who seek elective oocyte vitrification.

Key words: oocyte cryopreservation, social freezing, ovarian stimulation

Résumé. Contrairement à l'homme, qui continue à produire de nouveaux spermatozoïdes jusqu'à un âge avancé, la femme naît avec un nombre d'ovocytes limité, et qui diminue avec l'âge. La qualité des ovocytes diminue, par surcroît, elle aussi avec le temps. Les choix opérés par les femmes en matière de reproduction sont fortement affectés par les mutations de la société ; ainsi ont-elles aujourd'hui tendance à différer leur maternité, notamment dans l'attente des conditions plus favorables pour avoir un enfant. Corrélativement, la demande de cryocongélation des ovocytes connaît, depuis quelques années, une nette hausse. Cette technologie est l'objet d'une controverse parmi les bioéthiciens, qui se demandent si elle permet une émancipation des femmes ou si elle exerce au contraire un effet psychosocial négatif. La technique de vitrification a aujourd'hui dépassé sa phase expérimentale, et le suivi des enfants conçus après cryocongélation des ovocytes a donné des résultats rassurants quant à leur santé. Pour autant, la communication optimiste des sociétés de cryocongélation d'ovocytes semble susceptible de donner naissance à de faux espoirs. En effet, les chances d'avoir un enfant avec des ovocytes vitrifiés sont significativement réduites au-delà de l'âge de 36 ans (âge de cryocongélation). Les protocoles de stimulation ovarienne pour la cryocongélation des ovocytes adoptés par les cliniques d'assistance médicale à la procréation sont axés sur une production maximale d'ovocytes matures, tout en optimisant le profil de sécurité et la convivialité pour ces femmes qui se tournent vers la vitrification ovocytaire élective.

Mots clés :, congélation d'ovules, raisons sociales, stimulation ovarienne

Oocyte cryopreservation using the technology of vitrification to prevent age-related fertility decline is becoming increasingly popular, at least in countries where current legislation allows this practice. We will here focus on the scope of this emerging procedure, on the debate among health stakeholders, researchers and the public as to whether this practice is ethically acceptable, on the legal context in Belgium, and on current clinical approaches to ovarian stimulation in the setting of oocyte cryopreservation.

Definition

Although cryopreservation of oocytes in a woman in order to preserve her fertility until when she is older is commonly referred to as "social" freezing in social media,

Médecine de la Reproduction

Tirés à part : M. De Vos

doi:10.1684/mte.2018.0681

there is a rather negative connotation to this label. The term social freezing refers to oocyte cryopreservation for non-medical "social" reasons as opposed to oocyte freezing for medical reasons such as cancer and is regarded as a technological solution for postponed childbearing. Opponents of the term "social" freezing advocate that oocyte freezing for non-medical reasons is more than just an answer to a societal problem and that the term is probably too restrictive. Instead, bio-ethicists as well as reproductive medicine professionals have coined the term "AGE" banking for oocyte freezing for "Anticipated Gamete Exhaustion" [1], since there is no doubt that the physiological decline of oocyte quality and quantity inevitably leads to reduced fecundity; in this regard, oocyte cryopreservation should be regarded as an anticipating measure in view of an impending medical condition. In analogy with the existence of gamete banks, the term "AGE banking" was launched.

Although a woman has the potential to produce "fertilizable" oocytes from puberty to menopause, her fecundity (the ability to produce offspring) declines with age. Societal changes that were introduced in the sixties of last century and that were catalyzed by the contraceptive pill, have gradually reduced the fertility rate (number of births per woman). The postponement of pregnancy pushes an increasing number of women into the category of women who have a higher risk of needing medically assisted procreation to have a child. As a woman gets older, there is a decline of the number and the quality of her oocytes [2]. The gradual loss of oocytes starts as early as during fetal development. The approximately 300,000 primordial follicles available at the time of menarche will be gradually recruited until there are only around a thousand left at the time of the menopause. When a woman seeks reproductive treatment, her ovarian reserve is usually examined by measuring the serum level of Anti-Mu¨llerian Hormone (AMH) and by counting the number of antral follicles on the third day of the menstrual cycle. Women with polycystic ovary syndrome (PCOS) have increased levels of AMH, whereas AMH levels are reduced after ovarian surgery and toxic treatment. Although numerous studies have shown a correlation between AMH and oocyte yield in assisted reproduction, the ability to predict pregnancy after infertility treatment and natural conception is poor, while a nomogram integrating serum AMH in a protocol of ovarian stimulation is only useful for avoiding poor or excessive response. While AMH may predict age of menopause and remaining reproductive lifespan only very limited data suggest that AMH levels is related to the present fecundability after natural conceptions, although a prospective study of women in their thirties found a significantly reduced fecundability in those with a low AMH [3]. A recent Danish cohort study of young women in their early twenties found no association between lower AMH levels and fecundability, which illustrates the wide range of AMH among women with a normal fertility potential [4]. Furthermore, it is crucial to be cautious when interpreting AMH in oral contraception users as AMH levels may be almost 20% lower in this population [5].

Therefore, caution is needed with regard to AMH testing in a preconception setting; this test should only be offered after proper counselling and assessment of the past medical history and family history. Whether AMH testing can be used as a tool to assist women with their decision to embark on oocyte cryopreservation is a challenging question that merits further scrutiny.

Population

Many women delay childbirth until their fourth and fifth decades. Fertility data across the European Union show that the average age at first delivery has crossed the 30-years mark and continues to rise[1]. The reasons for this trend are multifactorial and include postponement of identification of a suitable partner, longer education, pursuing career goals, the desire to secure financial and housing stability, and other social factors, as well as advances in assisted reproductive technologies (ART) [6]. Interest in fertility preservation for women who defer childbearing for non-medical reasons, as listed above, is increasing [7]; this interest has been fueled by the removal, in 2012, of the experimental label on oocyte cryopreservation by the American Society of Reproductive Medicine (ASRM) in view of the advances of oocyte vitrification as compared to the old technique of slow-freezing of oocytes which has been largely abandoned [8]. Indeed, birth rates following in vitro fertilization (IVF) with the use of cryopreserved oocytes are similar to rates with the use of fresh oocytes [9]. However, because there is a lack of data to support the cost-efficiency of oocyte cryopreservation to prevent age-related fertility decline, oocyte cryopreservation is the subject of debate among fertility specialists and public health stakeholders; in 2013 the ASRM have issued a statement that oocyte cryopreservation should not be recommended to circumvent reproductive aging because of lack of data regarding the cost-efficiency, as well as the safety, the efficacy, and the psychosocial impact of the procedure (ASRM Practice Committee 2013). Their European counterpart ESHRE have also pronounced their concerns, and advised potential candidates for non-medical oocyte cryopreservation to do so before the age of 35 (ESHRE 2012). On a similar note, bio-ethicists have focused on the ethical acceptability of oocyte cryopreservation, claiming that this emerging practice may be driven by commercial and economic factors, and by the questionable assumption that women may become empowered by the extension

[1] http://ec.europa.eu/eurostat/statisticsexplained/index.php/Fertility_statistics

of their reproductive window [10-13]. Indeed, in spite of the increasing utilization of cryopreserved oocytes, data regarding the efficiency of the technique appear to indicate dichotomous results, with encouraging outcomes in women until the age of 35, but markedly reduced live birth rates in women who were 36 or older when they had their oocytes cryopreserved [14, 15]. Therefore, the opponents of oocyte cryopreservation warn against a potential misconception of the public that oocyte cryopreservation and fertility treatments can make up for the natural decline in fertility [13, 16, 17]. Therefore, there is a need for correct education of women regarding age-related fertility decline and the concurrent increasing risk of miscarriage [18], which should assist women in making better informed decisions regarding timely elective fertility preservation [19, 20]. In this regard, questionnaire-based studies in women who have undergone fertility preservation demonstrate that the majority of these patients acknowledge that they should have undergone oocyte cryopreservation at an earlier age, if they had been properly educated [21, 22].

Previous surveys examining the willingness of the public to embark on oocyte cryopreservation have shown that one in three respondents would consider themselves as potential candidates for this procedure [13, 22, 23], although women were more inclined to undergo oocyte cryopreservation if they found themselves at the lower end of the ovarian reserve range. Again, these reported intentions are a source for concern, as they point towards a paradox: women who are excellent candidates for oocyte freezing in terms of the cost/oocyte yield ratio may turn down the option of cryopreservation just because of their good ovarian reserve, whereas women with poor ovarian biomarker results may express a higher tendency towards oocyte freezing but will face a much higher cost per oocyte and will end up having suboptimal numbers of cryopreserved oocytes, with reduced efficiency rates. Nevertheless, fertility decline in women deteriorates after the age of 35 [24]. This decline is mainly accounted for by a decrease in follicle number and oocyte quality [25], with older women having a lower probability to conceive, either spontaneously of after medically assisted reproduction, and if they do conceive, they will have an increased risk of miscarriage and fetal loss [26]. Not only does the shift towards postponement of conception result in increased risks of age-related subfertility, this tendency also contributes to reduced fecundity and declining birth rates in highly industrialized countries, in parallel with the negative impact of male reproductive disorders on total fertility rates [27]. Concerted efforts by researchers in reproductive medicine, environmental health sciences, and demography are urgently required to address the role of fertility education, and the potential role of oocyte cryopreservation to mitigate the impact of age-related fertility decline on a couple's failure to realize their desired family size [28].

Debate

One of the main arguments against the cryopreservation of oocytes in anticipation of age-related fertility decline is that the approach constitutes an unjustifiable medicalization of procreation [29]. Oocyte cryopreservation creates patients whereas reproduction should be something as natural as possible. With oocyte cryopreservation, fertile women are presumably offered a medical solution for potential infertility that they may never face. Moreover, the technology fosters the idea that procreation is entirely controllable. This idea is misleading and it has been fueled by marketing strategies developed by commercial oocyte cryopreservation companies; oocyte cryopreservation has been advocated as a way for women to extend the biological clock, and vitrified oocytes have been marketed as an insurance against childlessness, although marketeers do not generally focus on the expense of oocyte cryopreservation or the potential side-effects of hormone treatment and oocyte retrieval procedures. Most importantly, there is a dramatic lack of figures reporting the chances of a live birth after oocyte cryopreservation. Although information is scarcely available, recent data appear to indicate that women at the age of 34, 37 and 42 would have to freeze ten, twenty, and even more than sixty oocytes, respectively, to have a 75 percent likelihood of having at least one live birth [30]. Furthermore, oocyte cryopreservation contributes to maintaining a society dominated by man. This kind of medicalization of fertility may not be liberating nor empowering women. Instead, the option of oocyte cryopreservation may exert undue pressure on women and encourage them to rely on oocyte cryopreservation over other reproductive options when it is far from guaranteed that these vitrified oocytes (particularly in women with below-average ovarian reserve at the time of oocyte cryopreservation) will ultimately lead to successful pregnancies and births.

In view of all this, the American College of Obstetricians and Gynecologists advised that although oocyte cryopreservation "represents an appealing option for those women who wish to defer childbearing until later in life", women should be counseled on the "risks, costs, and alternatives". ACOG emphasized that it did not "recommend oocyte preservation for the sole purpose of circumventing reproductive aging in healthy women."[31]

Use of cryopreserved oocytes

In general, cryopreserved oocytes can be used for the purpose of achieving one of three objectives:

1. By default, vitrified oocytes are cryopreserved for autologous use: IVF and embryo transfer. Regardless of

age, the transfer of an embryo can be scheduled in a natural cycle, a managed natural cycle with hCG ovulation triggering or an artificial endometrial preparation cycle. The optimal clinical strategy for embryo transfer is the subject of ongoing debate [32].

2. Donation to other recipients. Recipients include women with premature ovarian insufficiency or infertility based on advanced age, or carriers of a genetic risk (e.g. mitochondrial DNA mutations). The oocytes may be donated by women who had them cryopreserved for themselves and who no longer wish to use these oocytes for themselves.

3. Donation for scientific research purposes. In a number of countries, including Belgium, there is the option of carrying out scientific research on oocytes and on embryos resulting from in vitro fertilization. In Belgium, research on oocytes and embryos is regulated according to the Law of 19th December 2008 on the procurement and use of human biological material intended for medical human applications or for scientific research.

Legal context in Belgium

From the legal point of view, there is no provision in Belgian law which prohibits the oocyte cryopreservation in anticipation of age-related infertility. Medically assisted procreation is defined, by the Belgian law of 6 July 2007, as "all procedures and conditions for implementation related to medical techniques for assisted reproduction in which the following interventions are performed: 1) artificial insemination, 2) the in-vitro fertilisation techniques, in which, at any given moment during the procedure, access is provided to the oocyte and/or to the embryo".

According to this legislation, the originator of the parental project is "any person who has taken the decision to become a parent by means of medically assisted procreation, whether or not it is carried out using his or her own gametes or embryos". In Belgium, unlike other countries, no criteria have been established regarding the profile of the individuals seeking access to medically assisted procreation. It is therefore, by law, not necessary to be married or to be living as a couple, whether heterosexual or homosexual. Oocyte retrieval and requests for embryo transfer or oocyte insemination are available to adult women until and including the age of 45. Embryo transfer and oocyte insemination cannot be carried out in women aged over 47 years and 364 days. Nevertheless, the collection of gametes, gonads or fragments of gonads for cryopreservation may be carried out, if medically indicated, on a minor. The time limit for cryopreservation of embryos for the purpose of a parental project is five years, with effect from the day of cryopreservation, and this time limit is ten years for gametes.

Social and psychological aspects

The demographics of parenthood have undergone a tremendous change in most industrialised countries and the utilization of in-vitro fertilisation is steadily increasing.

Some of the literature mentions a negative effect on the children due to the older age of their parents. However, we have little empirical data on this subject. Recent studies have suggested a relationship between autism and parental age [33]. On the other hand, studies have shown that parenthood at a more advanced age may also offer advantages for the parents, including a smaller loss of income and shorter career breaks for mothers [34] and increased financial stability [35], although there is no evidence of any effect of an older parental age on their children. Nevertheless, advanced parental age may be associated with a more stable couple relationship between the parents, which may be beneficial for the well-being of the child [36]. On the other hand, children of older parents may lack contact with (older) grandparents. A questionnaire-based investigation of the association between the age of the mother of children born through IVF and the well-being of these children has shown reassuring results: the well-being of the children did not appear to be impacted by maternal age [37].

Health economics

Oocyte cryopreservation and related procedures including ovarian stimulation, oocyte retrieval, storage, warming, in vitro fertilization and embryo transfer is expensive. Two models have been developed to evaluate these costs. According to Van Loenderschoot et al. oocyte cryopreservation was more cost-effective than IVF if at least 61% of the women intended to actually use their vitrified oocytes, provided that one was willing to pay around 20,000 euro for an additional birth. In this model, three strategies were compared: cryopreservation at the age of 35 and IVF at 40, natural conception, and IVF at 40 without cryopreservation [38]. According to another study oocyte vitrification was not cost-effective, when the following strategies were compared: no action at the age of 25, oocyte vitrification at 25 and ovarian tissue freezing [39]. Further studies are required to reach a conclusion about cost-efficiency of oocyte cryopreservation to prevent age-related fertility decline.

Ovarian stimulation protocols in the context of cryopreservation of oocytes

Oocyte cryopreservation has expanded the scope of assisted reproductive technology: the application of

ovarian stimulation in healthy women has emphasized the need for simpler and more patient friendly protocols. On the other hand, ovarian stimulation in a cryopreservation setting is primarily focused on oocyte number and quality, whereas endometrium receptivity is not an issue.

Dosage of gonadotropins is based on serum AMH levels and/or antral follicle count as biomarkers of ovarian reserve, but usually commenced in a dose range between 150-300IU urinary or recombinant FSH per day. Clinical trials comparing different gonadotropins and GnRH analogue protocols have largely focused on live birth rates after fresh embryo transfer and are not entirely relevant for oocyte cryopreservation protocols [40]. An emerging gonadotropin is long acting FSH (corifollitropin alfa). This molecule has a similar action to recombinant FSH and is devoid of LH activity. Because of its carboxyterminal component containing four O-linked oligosaccharides it has a prolonged half-life compared to recombinant FSH [41], and in view of this, corifollitropin alpha can eliminate the need for daily subcutaneous injections which simplifies the stimulation process and reduces patient discomfort [42]. Multiple dose finding studies comparing the efficacy of a single bolus of corifollitropin alpha versus daily recombinant FSH have shown that corifollitropin alpha is equally effective with respect to the number of oocytes collected and ongoing pregnancy rates after fresh embryo transfer [43]. The higher incidence of OHSS in high responders and women with polycystic ovaries, again after fresh embryo transfer [44], led to caution in these subgroups. This is probably much less relevant in the setting of oocyte cryopreservation, as long as a GnRH agonist trigger is administered for final oocyte maturation in these women, which requires the use of a GnRH antagonist protocol.

When appraising the published clinical trials that have compared the long GnRH agonist and the GnRH antagonist protocol, one has to be aware that these studies have been performed in infertile couples with an aim of achieving a live birth after embryo transfer. These studies, including meta-analyses, have shown no conclusive evidence of a difference in live birth rate between GnRH antagonist and long course GnRH agonist [45] and demonstrated a significant advantage for GnRH antagonist protocols for the incidence of OHSS. Nevertheless, average oocyte retrieval rates were higher after long GnRH agonists compared to GnRH antagonists according to a systemic review by Kolibianakis et al. [46] which may potentially be relevant for women who request oocyte cryopreservation, although the incidence of estrogen deprivation side-effects and the obligatory use of hCG for final oocyte maturation after a long GnRH agonist protocol reduce the appeal for the latter protocol in the setting of oocyte cryopreservation.

As far as the choice of ovulation trigger in a GnRH antagonist protocol is concerned, the GnRH agonist displaces the GnRH antagonist from its pituitary receptor, which causes a surge in LH and FSH levels, followed by down regulation of the receptor [47]. In analogy with the physiological mid-cycle surge, the GnRH agonist trigger also induces a surge of FSH, which is supposed to have a role in completion of oocyte meiosis, cumulus expansion and induction of LH receptors on the granulosa cells [48]. The biphasic LH surge following GnRH agonist trigger is shorter than the triphasic LH surge of a natural cycle, which results in deficient luteal gonadotropin levels [49]. The early corpus luteum demise after GnRH agonist trigger results in reduced secretion of vasoactive peptides and is thus associated with reduced risk of OHSS. Furthermore, the GnRH agonist trigger is associated with improved patient comfort with lesser abdominal bloating due to reduced ovarian volumes, reduced intraperitoneal fluid and earlier onset of menses [50], features that are attractive in the setting of oocyte cryopreservation. Moreover, studies in oocyte donation cycles have demonstrated that triggering with GnRH agonists has no negative effect on oocyte maturation and/or embryo quality [51].

GnRH agonists as ovulation trigger have to be used with caution in women with a down-regulated hypothalamic-pituitary axis, as a suboptimal response to the ovulation trigger may be observed in a subset of women who fail to respond to the trigger with an adequate endogenous LH surge [52]. These patients characteristically have very low LH and FSH at the start of the cycle, and generally need more exogenous gonadotropins for stimulation. Women with low BMI and long term oral contraceptive pill users are at increased risk of suboptimal response to GnRH agonist trigger and response in these women may be rescued using the combination of a GnRH agonist with a bolus of hCG [53]. Whether a more universal use of this so-called dual trigger may enhance oocyte maturation rates is a subject of future investigation. Screening patients with pre-trigger LH values <0.5 IU/L may help identify women likely to elicit a suboptimal response to GnRH agonist trigger [52]. Furthermore, monitoring serum LH levels 12h after GnRH agonist trigger could identify suboptimal response and may lead to appropriate action (re-trigger with hCG) to avoid retrieval of immature oocytes. Although a cut-off value of LH for adequate response to GnRH agonist trigger has not been defined, patients with a post trigger LH <15 IU/L are at increased risk of failed maturation.

Timing of triggering final oocyte maturation

In regular IVF patients, the timing of trigger is crucial in a GnRH antagonist protocol cycle as studies have shown that delay in the trigger beyond the threshold of three

follicles of > 17mm diameter is associated with decline in pregnancy rates after fresh embryo transfer. This is probably due to rise in serum progesterone levels, which result in premature decidualization of the endometrium [54]. However, prolongation of the follicular phase does not seem to have any adverse effect on the oocyte quality nor cleavage rate in the embryo as reported in oocyte donation cycles [55]. Thus, delay of ovulation triggering in the setting of oocyte cryopreservation may be beneficial as this should lead to a higher number of mature oocytes retrieved. Nevertheless, the optimal follicle diameter threshold for ovulation triggering in oocyte cryopreservation cycles is still not established.

Oncological risks of ovarian stimulation

Female cancers such as breast cancer, uterine cancer or ovarian cancer have a known multifactorial etiology with hormonal factors playing an important role in development of most of these cancers [56]. In view of this, the safety of supra-physiological hormone levels during ovarian stimulation with regard to the potential oncological risks needs to be addressed. A meta-analysis in 2013 including a total of 746,455 patients demonstrated an increased risk of ovarian cancer in patients who had undergone IVF treatment (relative risk = 1.59) [57]. Similar findings were published in another meta-analysis done in the same year [58]. However, the increased risk in these patients existed only when comparing with the general population and was not present when comparing IVF patients with other subfertile women who never had IVF. A recent analysis confirmed the existence of a potential association between IVF treatment and risk of ovarian cancer even after adjustment for confounding factors such as maternal age and obesity [59].

Unopposed estradiol exposure is a known risk factor for the development of endometrial cancer – in view of this, an increased risk of uterine cancer after more than 6 cycles of fertility treatment with the use of gonadotropins has been reported by a Danish study in 2009 [60]. Similar associations between uterine cancer and IVF treatment have since been reported [59, 61]. Multiple meta-analyses have evaluated the risk of breast cancer in women who had undergone IVF treatment but failed to identify such association [59, 62]. Nevertheless, the majority of cancer association studies to date have evaluated the oncological risks among infertile women after IVF. It is important to emphasize that factors causing infertility may independently affect the risk of developing malignancy.

In conclusion, whether the availability of oocyte cryopreservation programs could empower women needs to be established. The procedure may provide an additional option for deferred motherhood using assisted reproduction but appropriate counseling is mandatory in view of the lack of cost-efficiency studies and the reduced live birth rates after oocyte vitrification when cryopreservation is performed after the age of 36 years. When ovarian stimulation is used as an elective procedure in apparently healthy women it is of vital importance that the stimulation is simplified and that potential health risks are as much as possible reduced. Ovarian hyperstimulation syndrome incidence has been dramatically reduced since the introduction of GnRH agonist triggering in IVF practice and should no longer exist in the setting of oocyte cryopreservation. A small subset of patients who develop an inadequate response to the GnRH agonist trigger. Thus, maturation trigger with GnRH agonist still deserves further scrutiny with well-designed randomized controlled trials.

Liens d'intérêt : Les auteurs déclarent n'avoir aucun lien d'intérêt en rapport avec cet article.

Références

1. Stoop D, Van der Veen F, Deneyer M, Nekkebroeck J, Tournaye H. Oocyte banking for anticipated gamete exhaustion (AGE) is a preventive intervention, neither social nor nonmedical. *Reprod Biomed Online* 2014 ; 28 : 548-51.

2. Broekmans F, Soules M, Fauser B. Ovarian aging: mechanisms and clinical consequences. *Endocr Rev* 2009 ; 30 : 465-93.

3. Steiner AZ, Herring AH, Kesner JS, *et al*. Antimullerian hormone as a predictor of natural fecundability in women aged 30-42 years. *Obstet Gynecol* 2011 ; 117 : 798-804.

4. Hagen CP, Vestergaard S, Juul A, *et al*. Low concentration of circulating antimullerian hormone is not predictive of reduced fecundability in young healthy women: a prospective cohort study. *Fertil Steril* 2012 ; 98 : 1602-8.

5. Birch Petersen K, Hvidman HW, Forman JL, *et al*. Ovarian reserve assessment in users of oral contraception seeking fertility advice on their reproductive lifespan. *Hum Reprod* 2015 ; 30 : 2364-75.

6. Cil AP, Turkgeldi L, Seli E. Oocyte cryopreservation as a preventive measure for age-related fertility loss. *Semin Reprod Med* 2015 ; 33 : 429-35.

7. Garcia-Velasco JA, Domingo J, Cobo A, Martìnez M, Carmona L, Pellicer A. Five years' experience using oocyte vitrification to preserve fertility for medical and nonmedical indications. *Fertil Steril* 2013 ; 99 : 1994-9.

8. Practice committee of the american society for reproductive medicine, Society for assisted reproductive technology. Mature oocyte cryopreservation: a guideline. *Fertil Steril* 2013 ; 99 : 37-43.

9. Cobo A, Kuwayama M, Pérez S, Ruiz A, Pellicer A, Remohí J. Comparison of concomitant outcome achieved with fresh and cryopreserved donor oocytes vitrified by the Cryotop method. *Fertil Steril* 2008 ; 89 : 1657-64.

10. Zoll M, Mertes H, Gupta J. Corporate giants provide fertility benefits: have they got it wrong? *Eur J Obstet Gynecol Reprod Biol* 2015 ; 195 : A1-2.

11. Mertes H, Pennings G. Social egg freezing: for better, not for worse. *Reprod BioMed Online* 2011 ; 23 : 824-9.

12. Goldman KN, Grifo JA. Elective oocyte cryopreservation for deferred childbearing. *Curr Opin Endocrinol Diabetes Obes* 2016 ; 23 : 458-64.

13. Daniluk JC, Koert E. Childless women's beliefs and knowledge about oocyte freezing for social and medical reasons. *Hum Reprod* 2016 ; 31 : 2313-20.

14. Cobo A, Garcia-Velasco JA, Coello A, Domingo J, Pellicer A, Remohi J. Oocyte vitrification as an efficient option for elective fertility preservation. *Fertil Steril* 2016 ; 105 : 755-64.

15. Doyle JO, Richter KS, Lim J, Stillman RJ, Graham JR, Tucker MJ. Successful elective and medically indicated oocyte vitrification and warming for autologous in vitro fertilization, with predicted birth probabilities for fertility preservation according to number of cryopreserved oocytes and age at retrieval. *Fertil Steril* 2016 ; 105 : 459-66.

16. Birch Petersen K, Hvidman HW, Sylvest R, *et al*. Family intentions and personal considerations on postponing childbearing in childless cohabiting and single women aged 35-43 seeking fertility assessment and counselling. *Hum Reprod* 2015 ; 30 : 2563-74.

17. Yu L, Peterson B, Inhorn MC, Boehm JK, Patrizio P. Knowledge, attitudes and intentions toward fertility awareness and oocyte cryopreservation among obstetrics and gynecology resident physicians. *Hum Reprod* 2016 ; 31 : 403-11.

18. Harper J, Boivin J, O'Neill HC, *et al*. The need to improve fertility awareness. *Reprod Biomed Online* 2017 ; 4 : 18-20.

19. Ter Keurst A, Boivin J, Gameiro S. Women's intentions to use fertility preservation to prevent age-related fertility decline. *Reprod Biomed Online* 2016 ; 32 : 121-31.

20. Milman L, Senapati S, Sammel M, Cameron K, Gracia C. Assessing reproductive choices of women and the likelihood of oocyte cryopreservation in the era of elective oocyte freezing. *Fertil Steril* 2017 ; 107 : 1214-22.

21. Hodes-Wertz B, Druckenmiller S, Smith M, Noyes N. What do reproductive-age women who undergo oocyte cryopreservation think about the process as a means to preserve fertility? *Fertil Steril* 2013 ; 100 : 1343-9.

22. Stoop D, Nekkebroeck J, Devroey P. A survey on the intentions and attitudes towards oocyte cryopreservation for non-medical reasons among women of reproductive age. *Hum Reprod* 2011 ; 26 : 655-61.

23. Meissner C, Schippert C, von Versen-Höynck F. Awareness, knowledge and perceptions of infertility, fertility assessment and assisted reproductive technologies in the era of oocyte freezing among female and male university students. *J Assist Reprod Genet* 2016 ; 33 : 719-29.

24. Sozou PD, Hartshorne GM. Time to pregnancy: a computational method. *PLoS One* 2012 ; 7 : e46544.

25. Faddy MJ, Gosden RG, Gougeon A, Richardson SJ, Nelson JF. Accelerated disappearance of ovarian follicles in mid-life: implications for forecasting menopause. *Hum Reprod* 1992 ; 7 : 1342-6.

26. Nybo Anderson A, Wohlfahrt J, Christens P, Olsen J, Melbye M. Maternal age and fetal loss: population based register linkage study. *BMJ* 2000 ; 320 : 1708-12.

27. Skakkebaek NE, Rajpert-De Meyts E, Buck Louis GM, *et al*. Male reproductive disorders and fertility trends: influences of environment and genetic susceptibility. *Physiol Rev* 2016 ; 96 : 55-97.

28. Habbema JDF, Eijkemans MJC, Leridon H, Te Velde ER. Realizing a desired family size: when should couples start? *Hum Reprod* 2015 ; 30 : 2215-21.

29. Reis E, Reis-Dennis S. Freezing eggs and creating patients: moral risks of commercialized fertility. *Hastings Cent Rep* 2017 ; 47 : S41-5.

30. Goldman RH, Racowsky C, Farland LV, Munné S, Ribustello L, Fox JH. Predicting the likelihood of live birth for elective oocyte cryopreservation: a counseling tool for physicians and patients. *Hum Reprod* 2017 ; 32 : 853-9.

31. American college of obstetricians and gynecologists, committee on gynecologic practice, *Committee opinion: oocyte cryopreservation*, no. 584, 2014, http://www.acog.org/Resources_And_Publications/Committee_Opinions/Committee_on_Gynecologic_Practice/Oocyte_Cryopreservation.

32. Mackens S, Santos-Ribeiro S, van de Vijver A, *et al*. Frozen embryo transfer: a review on the optimal endometrial preparation and timing. *Hum Reprod* 2017 ; 32 : 2234-42.

33. Frans EM, Sandin S, Reichenberg A, *et al*. Autism risk across generations. *JAMA Psychiatry* 2013 ; 70 : 516-21.

34. Miller AR. Motherhood delay and the human capital of the next generation. *Am Econ Rev* 2009 ; 99 : 154-8.

35. Mills M, Rindfuss RR, McDonald P, te Velde E, *et al*, on behalf of the ESHRE Reproduction Society Task Force. Why do people postpone parenthood? Reasons and social policy incentives. *Hum Reprod Update* 2011 ; 17 : 848-60.

36. Sobotka T. Oocyte cryopreservation as an insurance strategy: a socio-demographic viewpoint. In : Proceedings of the 1st international symposium on social egg freezing. Barcelona 2013: 5-28.

37. Boivin J, Rice F, Hayle D, *et al*. Associations between maternal older age, family environment and parent and child wellbeing in families using assisted reproductive techniques to conceive. *Soc Sci Med* 2009 ; 68 : 1948-55.

38. van Loendersloot L, Moolenaar L, Mol B, Repping S, van der Veen F, Goddijn M. Expanding reproductive lifespan: a cost-effectiveness study on oocyte freezing. *Humn Reprod* 2011 ; 26 : 3054-60.

39. Hirshfeld-Cytron J, Grobman W, Milad M. Fertility preservation for social indications: a cost- based decision analysis. *Fertil Steril* 2012 ; 97 : 665-70.

40. Westergaard LW, Bossuyt PM, Van der Veen F, van Wely M. Human menopausal gonadotropin *versus* recombinant follicle stimulation hormone for ovarian stimulation in assisted reproductive cycles. *Cochrane Database Syst Rev* 2011 ; 16 : CD003973.

41. Boime I, Ben-Menahem D. Glycoprotein hormone structure-function and analog design. *Recent Prog Horm Res* 1999 ; 54 : 271-88.

42. Fauser B, Mannaerts B, Devroey P, Leader A, Boime I, Baird D. Advances in recombinant DNA technology: corifollitropin alfa, a hybrid molecule with sustained follicle-stimulating activity and reduced injection frequency. *Hum Reprod Update* 2009 ; 15 : 309-21.

43. Griesinger G, Boostanfar R, Gordon K, Gates D, Sisk CM, Stegmann BJ. Corifollitropin alfa *versus* recombinant follicle-stimulating hormone: an individual patient data meta-analysis. *Reprod Biomed Online* 2016 ; 33 : 56-60.

44. Devroey P, Boostanfar R, Koper NP, Mannaerts BM, Ijzerman-Boon PC, Fauser BC, ENGAGE Investigators. A double-blind, non-inferiority RCT comparing corifollitropin alfa and recombinant

FSH during the first seven days of ovarian stimulation using a GnRH antagonist protocol. *Hum Reprod* 2009 ; 24 : 3063-72.

45. Al-Inany HG, Youssef MA, Ayeleke RO, Brown J, Lam WS, Broekmans FJ. Gonadotrophin-releasing hormone antagonists for assisted reproductive technology. *Cochrane Database Syst Rev* 2016 ; 29 : 4.

46. Kolibianakis EM, Collins J, Tarlatzis BC, Devroey P, Diedrich K, Griesinger G. Among patients treated for IVF with gonadotrophins and GnRH analogues, is the probability of live birth dependent on the type of analogue used? A systematic review and meta-analysis. *Hum Reprod Update* 2006 ; 12 : 651-71.

47. Engmann L, Benadiva C, Humaidan P. GnRH agonist trigger for the induction of oocyte maturation in GnRH antagonist IVF cycles: a SWOT analysis. *Reprod Biomed Online* 2016 ; 32 : 274-85.

48. Yding Andersen C. Effect of FSH and its different isoforms on maturation of oocytes from pre-ovulatory follicles. *Reprod Biomed Online* 2002 ; 5 : 232-9.

49. Humaidan P, Papanikolaou E, Tarlatzis B. GnRHa to trigger final oocyte maturation: a time to reconsider. *Hum Reprod* 2009 ; 24 : 389-94.

50. Garcia-Velasco JA, Motta L, Lopez A, Mayoral M, Cerrillo M, Pacheco A. Low-dose human chorionic gonadotropin *versus* estradiol/progesterone luteal phase support in gonadotropin releasing hormone agonist-triggered assisted reproductive technique cycles: understanding a new approach. *Fertil Steril* 2010 ; 94 : 2820-3.

51. Shapiro BS, Daneshmand ST, Garner FC, Aguirre M, Ross R. Comparison of human chorionic gonadotropin and gonadotropin-releasing hormone agonist for final oocyte maturation in oocyte donor cycles. *Fertil Steril* 2007 ; 88 : 237-9.

52. Meyer L, Murphy LA, Gumer A, Reichman DE, Rosenwaks Z, Cholst IN. Risk factors for a suboptimal response to gonadotropin-releasing hormone agonist trigger during *in vitro* fertilization cycles. *Fertil Steril* 2015 ; 104 : 637-42.

53. Lu X, Hong Q, Sun L, *et al*. Dual trigger for final oocyte maturation improves the oocyte retrieval rate of suboptimal responders to gonadotropin-releasing hormone agonist. *Fertil Steril* 2016 ; 106 : 1356-62.

54. Kolibianakis E, Albano C, Camus M, Tournaye H, Van Steirteghem A, Devroey P. Prolongation of the follicular phase in *in vitro* fertilization results in a lower ongoing pregnancy rate in cycles stimulated with recombinant follicle-stimulating hormone and gonadotropin-releasing hormone antagonists. *Fertil Steril* 2004 ; 82 : 102-7.

55. Melo MA, Meseguer M, Garrido N, Bosch E, Pellicer A, Remohí J. The significance of premature luteinization in an oocyte-donation programme. *Hum Reprod* 2006 ; 21 : 1503-7.

56. Bernstein L. Epidemiology of endocrine-related risk factors for breast cancer. *J Mammary Gland Biol Neoplasia* 2002 ; 7 : 3-15.

57. Li LL, Zhou J, Qian XJ, Chen YD. Meta-analysis on the possible association between *in vitro* fertilisation and cancer risk. *Int J Gynecol Cancer* 2013 ; 23 : 16-24.

58. Siristatidis C, Sergentanis TN, Kanavidis P, *et al*. Controlled ovarian hyperstimulation for IVF: impact on ovarian, endometrial and cervical cancer – a systematic review and meta-analysis. *Hum Reprod Update* 2013 ; 19 : 105-23.

59. Kessous R, Davidson E, Meirovitz M, Sergienko R, Sheiner E. The risk of female malignancies after fertility treatments: a cohort study with 25-year follow-up. *J Cancer Res Clin Oncol* 2016 ; 142 : 287-93.

60. Jensen A, Sharif H, Kjaer SK. Use of fertility drugs and risk of uterine cancer: results from a large Danish population-based cohort study. *Am J Epidemiol* 2009 ; 170 : 1408-14.

61. Lerner-Geva L, Liat LG, Rabinovici J, *et al*. Are infertility treatments a potential risk factor for can- cer development? Perspective of 30 years of follow-up. *Gynecol Endocrinol* 2012 ; 28 : 809-14.

62. Sergentanis TN, Diamantaras AA, Perlepe C, Kanavidis P, Skalkidou A, Petridou ET. IVF and breast cancer: a systematic review and meta-analysis. *Hum Reprod Update* 2014 ; 20 : 106-23.