

Mature oocyte cryopreservation: a guideline

The Practice Committees of the American Society for Reproductive Medicine and the Society for Assisted Reproductive Technology

Society for Reproductive Medicine and Society for Assisted Reproductive Technology, Birmingham, Alabama

There is good evidence that fertilization and pregnancy rates are similar to IVF/ICSI with fresh oocytes when vitrified/warmed oocytes are used as part of IVF/ICSI for young women. Although data are limited, no increase in chromosomal abnormalities, birth defects, and developmental deficits has been reported in the offspring born from cryopreserved oocytes when compared to pregnancies from conventional IVF/ICSI and the general population. Evidence indicates that oocyte vitrification and warming should no longer be considered experimental. This document replaces the document last published in 2008 titled, "Ovarian Tissue and Oocyte Cryopreservation," *Fertil Steril* 2008;90:S241-6. (*Fertil Steril*® 2013;99:37-43. ©2013 by American Society for Reproductive Medicine.)

Earn online CME credit related to this document at www.asrm.org/elearn

Discuss: You can discuss this article with its authors and with other ASRM members at <http://fertilityforum.com/goldsteinj-mature-oocyte-cryopreservation-guideline/>



Use your smartphone to scan this QR code and connect to the discussion forum for this article now.*

* Download a free QR code scanner by searching for "QR scanner" in your smartphone's app store or app marketplace.

Mature oocyte cryopreservation (OC) is a method to preserve reproductive potential in women of reproductive age. This document outlines the current technology, clinical outcomes, and risks of mature oocyte cryopreservation and provides recommendations for clinical applications.

HISTORY OF CRYOPRESERVATION TECHNOLOGY

Cryopreservation refers to the cooling of cells and tissues to sub-zero temperatures in order to stop all biologic activity and preserve them for future use. The science of cryobiology can be traced as far back as 2500 BC, when early civilizations used cold for medicinal purposes. However, cryopreservation of cells and tissues did not become a reality until the mid-20th century. Initial efforts at cryopreservation were ineffective because simple cooling techniques led to cellular dam-

age from changing concentration of solutes within the cells, intra- or extracellular ice formation, and excessive dehydration. In the 1940s, it was discovered that glycerol could protect sperm from damage during cryopreservation and thawing.

The first human birth from frozen sperm was reported in 1953 (1). In the 1970s other cryoprotectants such as propanediol, ethylene glycol (EG), and dimethyl sulfoxide (DMSO) were identified and found to minimize cellular damage. In addition, slow-freeze techniques using programmable freezers were developed to allow for freezing to occur at a slow enough rate to permit sufficient cellular dehydration to minimize intracellular ice formation. These improvements led to the first human birth from a frozen embryo, reported in 1984 (2). In 1986, the first human birth from a frozen oocyte was reported (3).

Over the past decade, an alternative to slow-freeze, vitrification, has been

developed. Vitrification is the process of cryopreservation using high initial concentrations of cryoprotectant and ultra-rapid cooling to solidify the cell into a glass-like state without the formation of ice. Vitrification is currently being applied to the cryopreservation of embryos, oocytes, and ovarian tissue. While various methods of slow-freeze and vitrification have been used, for the purpose of this document the terms slow-freeze and vitrification will be used to summarize the data.

MATURE OOCYTE CRYOPRESERVATION TECHNOLOGY

Historically, overall success with respect to oocyte survival, fertilization rates, and pregnancy rates was low (4) and has only recently improved (5). Initial success was limited by the fragility of the metaphase-II (M-II) oocyte related to its large size, water content, and chromosomal arrangement.

In mature oocytes (M-II), typically retrieved after superovulation, the metaphase chromosomes are lined up by the meiotic spindle along the equatorial plate. Studies have documented that the spindle apparatus may be damaged by intracellular ice formation

Received September 19, 2012; accepted September 20, 2012; published online October 22, 2012.

No reprints will be available.

Correspondence: Practice Committee, American Society for Reproductive Medicine, 1209 Montgomery Hwy., Birmingham, AL 35216 (E-mail: ASRM@asrm.org).

Fertility and Sterility® Vol. 99, No. 1, January 2013 0015-0282/\$36.00

Copyright ©2013 American Society for Reproductive Medicine, Published by Elsevier Inc. <http://dx.doi.org/10.1016/j.fertnstert.2012.09.028>

during the freezing or thawing process (6, 7), and these abnormalities may be dependent on patient age and cryopreservation technique and may vary by time after thaw (8).

Modifications in cryopreservation methods over the past few years may be responsible for improved survival of cryopreserved mature oocytes. For example, modifications in the combination and composition of cryoprotectants in slow-freeze protocols have improved the survival rate of frozen M-II oocytes (9–12). Numerous studies also have reported improved oocyte survival by modifications of slow-freeze cryopreservation techniques such as changing the initial temperature of the cryoprotectant (13), the seeding temperature (14), and timing in relation to the oocyte retrieval (15).

Recent studies suggest that vitrification for oocyte cryopreservation significantly improves oocyte survival and pregnancy rates. In humans, most studies suggest that post-thaw survival rates of vitrified oocytes are superior to those that have undergone slow-freeze protocols (4, 16, 17). It should be noted that successful thawing of viable oocytes continues to improve with both vitrification and slow-freeze techniques. In addition, a study reported that meiotic spindle recovery was faster in oocytes that had been vitrified rather than cryopreserved with a slow-freeze technique (18). Most vitrification protocols use an “open” system, in which oocytes are directly exposed to liquid nitrogen to maximize ultra-rapid cooling and minimize ice crystal formation. A theoretical concern regarding such “open” systems is their potential to expose oocytes to infectious organisms present in contaminated liquid nitrogen. While infectious transmission has never been observed in reproductive tissues, methods to sterilize liquid nitrogen are being developed such as micro-filtration or ultraviolet (UV) radiation (19). As of September, 2012, the open loop technique was not FDA approved in the

United States (US). Closed systems also exist, but it is not clear whether they are associated with equivalent success rates.

Cryopreservation protocols usually involve removing cumulus cells from oocytes in order to assess oocyte maturity. Because removing cumulus cells may reduce fertilization following standard insemination and because zona hardening has been reported after thawing cryopreserved oocytes, intracytoplasmic sperm injection (ICSI) is generally used for fertilizing previously cryopreserved oocytes (20). While some studies suggest that the use of ICSI may improve fertilization rates and overcome changes in the zona pellucida after freezing (21, 22), it is not clear whether ICSI is necessary for fertilization of frozen thawed oocytes (23).

REVIEW METHODS

To evaluate the efficacy and safety of mature oocyte cryopreservation the Committee performed a systematic literature search using the MEDLINE site up to April 2012. In order to compare the efficacy (clinical pregnancy and live birth rates) of embryo transfers using fresh or cryopreserved/thawed oocytes, the search utilized combinations of medical subject headings “oocyte,” “cryopreservation,” “vitrification,” “frozen,” “birth,” “delivery,” and “pregnancy.” In order to assess the safety of oocyte cryopreservation, the search included the terms “safe,” “risk,” “birth defect,” “karyotype,” and “abnormal” to the search. Only English language articles were selected, and the search was restricted to published articles. Review articles were included. The relevance of included articles was assessed by an epidemiologist with subsequent consultation by the Committee. A total of 981 articles on oocyte cryopreservation efficacy was identified initially and 80 were determined to be relevant. Three hundred seventy-seven articles were initially identified in the oocyte cryopreservation

TABLE 1

Summary of randomized controlled trials comparing fresh versus vitrified oocytes.

	Cobo 2008 (24)	Cobo 2010 (26)	Rienzi 2010 (25)	Parmegiani 2011 (19)
Patient population	Oocyte donors	Oocyte donors	Infertile patients <43 years of age requiring ICSI with >6 mature oocytes	Infertile patients <42 years of age requiring ICSI with >5 mature oocytes
No. patients	30 vitrification 30 fresh	295 vitrification 289 fresh	40 vitrification 40 fresh	31 vitrification 31 fresh
Mean age at retrieval	26	26	35	35
No. oocytes	231 vitrification 219 fresh	3286 vitrification 3185 fresh	124 vitrification 120 fresh	168 vitrification NA fresh
No. oocytes per retrieval	18.2	11	13	NA
Survival	96.9%	92.5%	96.8%	89.9%
Fertilization rate	76.3 vitrification 82.2 fresh	74% vitrification 73% fresh	79.2% vitrification 83.3% fresh	71% vitrification 72.6% fresh
No. transferred vitrification vs. fresh	3.8 vitrification 3.9 fresh	1.7 vitrification 1.7 fresh	2.3 vitrification 2.5 fresh	2.5 vitrification 2.6 fresh
Day of transfer	3	3	2	2–3
Implantation rate	40.8% vitrification 100% fresh	39.9% vitrification 40.9% fresh	20.4% vitrification 21.7% fresh	17.1% vitrification NA fresh
CPR/transfer vitrification vs. fresh	60.8% (23 vitrification transfers) 100% (1 fresh transfer)	55.4% vitrification 55.6% fresh	38.5% vitrification 43.5% fresh	35.5% vitrification 13.3% fresh
CPR/oocyte thawed	6.1%	4.5%	12%	6.5%

Note: All used vitrification with Cryotop, 15% EG + 15% DMSO + 0.5M sucrose. CPR = clinical pregnancy rate.

Practice Committee. Oocyte cryopreservation. Fertil Steril 2013.

safety search and 32 were found to be relevant. All relevant articles were reviewed and the level of evidence was determined for each article.

CLINICAL OUTCOMES

Success of In Vitro Fertilization (IVF) with Cryopreserved Oocytes Compared with Fresh Oocytes

The literature search identified only four randomized controlled trials comparing outcomes with cryopreserved and fresh oocytes in IVF/ICSI cycles (19, 24–26) (Table 1). All studies used a similar open vitrification protocol (Cryotop device, 15% EG + 15% DMSO + 0.5 M sucrose) and were conducted in Europe. Two of these studies were conducted in egg donor/recipient cycles, and 2 were conducted in infertile couples with supernumerary oocytes available to vitrify and warm only if pregnancy was not achieved in the fresh cycle. Overall, oocyte survival after vitrification and warming ranged between 90%–97%, fertilization rates were between 71%–79%, implantation rates were 17%–41%, and clinical pregnancy rates per transfer ranged from 36%–61%. The clinical pregnancy rate (CPR) per thawed oocyte ranged from 4.5%–12%. The largest and most compelling RCT compared the use of fresh versus vitrified donor oocytes in 600 recipients. The investigators found that 92.5% of vitrified oocytes survived warming, and that there were no significant differences in fertilization rates (74.2 vitrified vs. 73.3 fresh), implantation rates (39.9 vs. 40.9) and pregnancy rates per transfer (55.4 vs. 55.6) between groups, with a mean of 1.7 embryos transferred (26). These studies and a recent meta-analysis (5) suggest that fertilization and pregnancy rates are similar to IVF/ICSI with fresh oocytes when vitrified/warmed oocytes are used as part of IVF/ICSI. In summary, there is good evidence that fertilization and pregnancy rates are similar to IVF/ICSI with fresh oocytes when vitrified/warmed oocytes are used as part of IVF/ICSI in young patients.

However, given the limited number of randomized controlled trials, it is not clear that these data are generalizable. Indeed, it is likely that only programs with the highest pregnancy rates conduct and publish such studies, limiting the generalizability of their results to other clinical programs. In addition, the majority of these data derives from experience using oocytes obtained from healthy, young oocyte donors under the age of 30 years, which have been vitrified for a limited duration. Therefore, such data cannot be extrapolated to other clinics, different patient populations (particularly older women), and to programs that utilize different cryopreservation protocols.

Observational Studies

Given these limitations, it is useful also to consider the results of observational studies comparing success rates using fresh and cryopreserved oocytes. The largest of these observational studies have been conducted in Italy, where Italian law limited the number of oocytes that may be fertilized as part of IVF. For this reason, programs in Italy have been offering OC to couples with additional oocytes available at retrieval for many years. These data are important because they reflect success rates at various clinical programs rather

than only at programs with particular expertise. A large Italian multi-center prospective cohort study of infertile couples with supernumerary oocytes (>3 oocytes retrieved) cryopreserved using a slow-freeze protocol demonstrated that success with fresh oocyte cycles (2,209 retrievals) was superior to that of frozen oocyte cycles (940 thaws) (27). The overall oocyte survival was 55.8% fresh or frozen; other studies comparing the fertilization rate (72.5% vs. 78.3%), implantation rate (10.1% vs. 15.4%), pregnancy rate per transfer (17% vs. 27.9%), and delivery rate per transfer (11.6% vs. 21.6%) were all significantly lower in frozen oocyte cycles compared with fresh cycles when an average of 2 embryos were transferred. It should be recognized that the lower success rates observed in this study may be due in part to selection bias as selection of superior-appearing oocytes for fresh insemination may lead to falsely lower success for oocyte cryopreservation and because regulations limit the number of thawed oocytes that may be fertilized. In addition, different cryopreservation protocols (slow-freeze) and variable clinic specific experience may contribute to these findings as well. Similarly, an analysis of Italian national register data from 193 IVF centers and over 120,000 IVF cycles from 2005 to 2007 also demonstrated that implantation rates (13.5% vs. 6.9%; odds ratio [OR] 2.12; 95% confidence interval [CI], 1.99–2.26) and pregnancy rates per transfer (24.9% vs. 12.5%; OR 2.32; 95% CI, 2.16–2.49) were higher with fresh oocyte cycles compared to frozen oocyte cycles. In addition, while frozen embryo cycles were limited, they found that implantation (OR 1.31; 95% CI, 1.17–1.46) and pregnancy rates (OR 2.37; 95% CI, 1.21–1.55) were higher when frozen embryos were used compared to frozen oocytes (28). In summary, results from large observational studies of clinical practice in Italy where supernumerary oocytes are cryopreserved suggest that implantation and pregnancy rates may be lower when frozen oocytes are used compared to fresh or frozen embryos.

Because IVF practices in Europe differ considerably from those in the United States, it is also relevant to summarize recent observational data on the success of oocyte cryopreservation in the US even though samples sizes are limited in these reports. Several studies have been conducted in young infertile and fertile populations that demonstrate excellent success rates. A retrospective cohort study of 19 women less than 37 years of age undergoing either slow-freeze or vitrification of oocytes reported an oocyte survival rate of 89%, a fertilization rate of 78%, an implantation rate of 45%, and a live-birth rate per transfer of 58% (29). The same group previously reported the results of oocyte cryopreservation/thaw cycles in 22 infertile women and oocyte donors and reported similar results (92% survival, 42% implantation rate, 57% clinical pregnancy rate [CPR] per transfer, and 4% CPR per oocyte thaw) (30). A study of oocyte vitrification in 19 fertile women 35 years of age or younger with a prior tubal ligation demonstrated a survival rate of 81%, fertilization rate of 72.3%, implantation rate 45% and CPR of 80%, and live birth rate per transfer of 65% (31). Overall, the CPR per oocyte warmed was 5.1% in this study. Finally, another group reported similar success rates in a small study of 10 oocyte donors and 20 recipients (89% oocyte survival rate, 87%

fertilization rate, 75% CPR per transfer) (32). In summary, published data in the United States are limited to a few clinics but demonstrate acceptable success rates in young highly selected populations. It is important to recognize that success rates may not be generalizable, and clinic-specific success rates should be used to counsel patients whenever possible.

The Impact of Maternal Age on Oocyte Cryopreservation Success

Several observational studies have assessed the impact of age on the success of oocyte cryopreservation. As with fresh oocytes, there is an expected decline in success with increased age. There are no comparative trials assessing success with cryopreserved vs. fresh oocytes by age. However, several studies using slow-freeze protocols suggest that success rates are lower with advanced maternal age. In the large Italian cohort study described above, oocyte survival was similar among women of different ages and women over 38 years of age had lower implantation rates (6.5% vs. 10.9%, $P = .012$) and pregnancy rates (10.1% vs. 18.7%, $P = .02$) compared to younger women (27). Another Italian study of 342 infertile patients cryopreserving supernumerary oocytes using a slow-freeze protocol reported pregnancy rates in three groups of women by age (12). Implantation rates were 16.7%, 11.6%, and 10.8%; pregnancy rates per thaw cycle were 24.3%, 18.9%, and 16.1%; and pregnancy rates per embryo transfer were 27.7%, 21.4%, and 17.6% in women ≤ 34 years, 35–38 years, and over 38 years, respectively. While success appeared to be lower in older women, differences did not reach statistical significance.

Several studies also have observed decreased success with oocyte vitrification in women of advanced age. A large Italian retrospective cohort study of 450 couples undergoing oocyte thaw cycles using previously vitrified supernumerary oocytes found that maternal age was inversely correlated with delivery rates (33). Another report also noted that ongoing pregnancy rates in 182 oocyte vitrification/warming cycles were significantly lower in women over 40 years of age (34). In this study, age-stratified CPR per transfer were: 48.6% in ≤ 34 year-olds, 24.1% in 35–37 year-olds, 23.3% in 38–40 year-olds, and 22.2% in 41–43 year-olds. In summary, success rates with oocyte cryopreservation via either slow-freeze or vitrification appear to decline with maternal age consistent with the clinical experience with fresh oocytes.

Success Rates with Slow-Freeze Compared with Vitrification

Most studies suggest that post-thaw survival rates of vitrified oocytes are superior to those that have undergone slow-freeze protocols, but there are limited studies comparing the two methods directly (4, 16). Only one RCT was identified in the literature search that compared pregnancy rates with slow-freeze vs. vitrified supernumerary oocytes and demonstrated that vitrification resulted in better oocyte survival (81% vs. 67%, $P < .001$), fertilization (77% vs. 67%, $P = .03$), and CPR per thawed oocyte (5.2% vs. 1.7%, $P = .03$) compared to slow-freeze (16). Similarly,

another RCT demonstrated improved survival, cleavage, and blastocyst development, but did not assess pregnancy as an outcome (35). Nonetheless, some clinics report equivalent success with slow-freeze and vitrification in observational studies (30), and it is likely that clinic-specific success rates may vary with different methods of cryopreservation.

Duration of Storage

Limited data exist regarding the effect of duration of storage on oocyte cryopreservation survival and pregnancy. One study was identified in the literature search that assessed oocyte cryopreservation efficacy with duration of storage. In this study, no differences in survival, fertilization, cleavage, embryo quality, implantation, and live-birth rates were observed in oocytes cryopreserved with slow-freeze and thawed after up to 48 months compared to earlier thaws (36).

Risks

While there are a limited number of established pregnancies and deliveries derived from cryopreserved oocytes, perinatal outcome data are reassuring. Despite concerns regarding spindle abnormalities in cryopreserved oocytes, the incidence of chromosomal abnormalities in human embryos obtained from cryopreserved oocytes is no different from that of control embryos as determined by fluorescence in-situ hybridization (37). A recent review of over 900 live births derived from cryopreserved oocytes, principally using slow-freeze, suggests that there is no increased risk of congenital anomalies compared to the general US population (38). In addition, a study of 200 infants born from 165 vitrified oocyte pregnancies revealed no difference in birth weight or congenital anomalies among those born from vitrified oocytes compared to children conceived after fresh IVF (39). While short-term data appear reassuring, long-term data on developmental outcomes and safety data in diverse (older) populations are lacking. As previously discussed, there also are theoretic infectious disease concerns with the use of open vitrification methods. However, infectious transmission has never been observed in reproductive tissues from this technique (40). The well-described risks associated with ovarian stimulation and oocyte retrieval also apply. Since embryo transfer is not being performed in most individuals cryopreserving oocytes, the risks of ovarian hyperstimulation syndrome (OHSS) are very low (41).

PROPOSED APPLICATIONS

Successful oocyte cryopreservation has the potential to simplify oocyte donation. Currently, oocyte donation cycles require coordination of fresh cycles between the donor and recipient, which can be inconvenient and costly. Use of cryopreserved oocytes may provide women with more choices in selecting a donor and more flexibility in timing pregnancy and potentially reduce the cost. Indeed, much of the best data to support the use of OC are in the setting of donor oocyte cycles (24, 30, 32). The largest RCT comparing fresh vs. vitrified donor oocytes in 600 recipients revealed

excellent clinical pregnancy rates, no different than in fresh cycles (26) (Table 1). However, while these data are reassuring, more widespread clinic-specific data on the safety and efficacy of oocyte cryopreservation in this population are needed before universal donor oocyte banking can be recommended.

MEDICAL INDICATIONS

Patients Receiving Gonadotoxic Therapies for Cancer and Other Medical Diseases

The gonadotoxicity of chemotherapeutics and radiotherapy has been well documented (42). In addition, some patients may require oophorectomy for various benign and malignant conditions. Mature oocyte banking is an attractive strategy for fertility preservation in postpubertal females without a partner and who do not wish to use donor sperm. Freezing oocytes, rather than embryos, would be an option for patients unable or not wishing to cryopreserve embryos. Data on pregnancy and live births from oocyte cryopreservation in cancer patients are very limited, and success rates must be extrapolated from other populations for patient counseling. However, in this population at high risk for infertility, oocyte cryopreservation may be one of the few options available and therefore is recommended with appropriate counseling.

Genetic Conditions

Certain genetic conditions, such as BRCA mutations, are associated with a high risk of ovarian cancer, and prophylactic salpingo-oophorectomy may be recommended during the late reproductive years. Ideally, this procedure is performed after completion of childbearing. However, in the event that prophylactic oophorectomy is recommended before childbearing and pregnancy is not an option at that time, cryopreservation of oocytes or embryos may be considered.

In addition, several genetic disorders have been associated with premature ovarian failure, such as Turner syndrome, fragile X premutation, and deletions of the X chromosome. Early diagnosis of these conditions may raise the possibility of fertility preservation in these populations (43). However, the efficacy of oocyte banking in this population is not known, and the risk of chromosomal abnormalities in offspring and the safety of future pregnancy are significant concerns (44).

Failure to Obtain Sperm for IVF

Occasionally, the male partner of a couple undergoing IVF is unable to collect a semen sample for oocyte insemination on the day of the oocyte retrieval. In addition, males with severe male infertility may have insufficient sperm for fertilization of retrieved oocytes. In such instances, oocytes may be cryopreserved for insemination and embryo transfer at a later date. Two studies have reported success rates of oocyte cryopreservation in such situations (45, 46). One study assessing the success of oocyte cryopreservation in 22 infertile couples with insufficient sperm on the day of the retrieval reported a survival of 70.5%, a fertilization rate of 61.5%,

and a pregnancy rate per transfer of 33% (46). Another study reported a pregnancy rate of 53% per transfer after oocyte cryopreservation in female partners of males with nonobstructive azoospermia and failed testicular extraction (45). Therefore, oocyte cryopreservation may be considered in couples pursuing IVF with insufficient sperm on the day of retrieval.

Oocyte Cryopreservation for Those Unable to Cryopreserve Embryos

Some couples undergoing IVF cannot or wish not to cryopreserve embryos that are not transferred in a fresh cycle. While some studies suggest the use of supernumerary cryopreserved oocytes may be associated with lower success rates compared to IVF with fresh oocytes, oocyte cryopreservation can contribute to the overall cumulative pregnancy rate (26). Therefore, oocyte cryopreservation is a reasonable strategy for patients who are unable to cryopreserve embryos.

Elective Cryopreservation to Defer Childbearing

Since there is a progressive loss of oocyte quantity and quality that occurs with female aging, the prevalence of infertility and the incidence of pregnancy loss and chromosomal abnormalities increase steadily up to age 35 and more rapidly thereafter. Technologies such as OC may allow women to have an opportunity to have biologic children later in life. While this technology may appear to be an attractive strategy for this purpose, there are no data on the efficacy of oocyte cryopreservation in this population and for this indication. Data on the safety, efficacy, cost-effectiveness, and emotional risks of elective oocyte cryopreservation are insufficient to recommend elective oocyte cryopreservation. Marketing this technology for the purpose of deferring childbearing may give women false hope and encourage women to delay childbearing. In particular, there is concern regarding the success rates in women in the late reproductive years who may be the most interested in this application. As described above, success rates appear to be significantly lower for women who cryopreserve or vitrify oocytes over the age of 38 (47). Patients who wish to pursue this technology should be carefully counseled about age and clinic-specific success rates of oocyte cryopreservation vs. conceiving on her own and risks, costs, and alternatives to using this approach (48).

SUMMARY

The success of oocyte cryopreservation has improved dramatically over the past decade, and preliminary data for safety are reassuring. Therefore, this technique should no longer be considered experimental. Four randomized controlled trials of fresh vs. vitrified/warmed oocytes indicate that implantation and clinical pregnancy rates are similar. However, results from large observational studies of clinical practice where supernumerary oocytes were cryopreserved suggest that implantation and pregnancy rates may be lower when frozen oocytes are used compared with fresh or frozen embryos. Published data in the United States are limited to a few clinics but demonstrate acceptable success rates in young, highly

selected populations. It is important to recognize that success rates may not be generalizable, and clinic-specific success rates should be used to counsel patients whenever possible.

Although a variety of clinical applications have been proposed for the use of oocyte cryopreservation, data on the success of oocyte cryopreservation are limited to donor populations and infertile couples with supernumerary oocytes. While pregnancy and live-birth rates appear to be similar using vitrified and fresh donor oocytes in select clinics, more widespread clinic-specific data on the safety and efficacy of oocyte cryopreservation in this population are needed before universal donor oocyte banking can be recommended. The existing literature supports the use of oocyte cryopreservation to improve cumulative pregnancy rates in couples who are unable to cryopreserve embryos. In the case of patients who are facing infertility due to chemotherapy or other gonadotoxic therapies, oocyte cryopreservation may be one of the few options available and therefore is recommended under these circumstances with appropriate counseling. On the other hand, there are not yet sufficient data to recommend oocyte cryopreservation for the sole purpose of circumventing reproductive aging in healthy women because there are no data to support the safety, efficacy, ethics, emotional risks, and cost-effectiveness of oocyte cryopreservation for this indication.

In addition, while data are reassuring at this point, it is too soon to conclude that the incidence of anomalies and developmental abnormalities of children born from cryopreserved oocytes is similar to those born from cryopreserved embryos. Oocyte cryopreservation will need to be studied in adequate numbers of patients for a sufficient length of time to determine whether the development of children is comparable to those conceived from other established assisted reproduction techniques. While oocyte cryopreservation has been shown to be safe and effective in select populations, more data are needed before this technology should be used routinely.

In conclusion, there is good evidence that fertilization and pregnancy rates are similar to IVF/ICSI with fresh oocytes when vitrified/warmed oocytes are used as part of IVF/ICSI in young infertility patients and oocyte donors. No increases in chromosomal abnormalities, birth defects, or developmental deficits have been noted in the children born from cryopreserved oocytes. This technique should no longer be considered experimental.

RECOMMENDATIONS

- In patients facing infertility due to chemotherapy or other gonadotoxic therapies, oocyte cryopreservation is recommended with appropriate counseling (Level B).
- More widespread clinic-specific data on the safety and efficacy of oocyte cryopreservation in donor populations are needed before universal donor oocyte banking can be recommended (Level B).
- There are not yet sufficient data to recommend oocyte cryopreservation for the sole purpose of circumventing reproductive aging in healthy women (Level B).
- More data are needed before this technology should be used routinely in lieu of embryo cryopreservation (Level B).

Acknowledgments: This report was developed under the direction of the Practice Committee of the American Society for Reproductive Medicine (ASRM) in collaboration with the Society for Assisted Reproductive Technology (SART) as a service to its members and other practicing clinicians. Although this document reflects appropriate management of a problem encountered in the practice of reproductive medicine, it is not intended to be the only approved standard of practice or to dictate an exclusive course of treatment. Other plans of management may be appropriate, taking into account the needs of the individual patient, available resources, and institutional or clinical practice limitations. The Practice Committees and the Board of Directors of ASRM and SART have approved this report. It has been reviewed by the SART presidential chain and edited based on their comments.

This document was reviewed by ASRM members and their input was considered in the preparation of the final document. The following members of the ASRM Practice Committee participated in the development of this document. All Committee members disclosed commercial and financial relationships with manufacturers or distributors of goods or services used to treat patients. Members of the Committee who were found to have conflicts of interest based on the relationships disclosed did not participate in the discussion or development of this document.

Samantha Pfeifer, M.D.; Jeffrey Goldberg, M.D.; R. Dale McClure, M.D.; Roger Lobo, M.D.; Michael Thomas, M.D.; Eric Widra, M.D.; Mark Licht, M.D.; John Collins, M.D.; Marcelle Cedars, M.D.; Catherine Racowsky, Ph.D.; Michael Vernon, Ph.D.; Owen Davis, M.D.; Clarisa Gracia, M.D., M.S.C.E.; William Catherino, M.D., Ph.D.; Kim Thornton, M.D.; Robert Rebar, M.D.; Andrew La Barbera, Ph.D.

REFERENCES

1. Sherman J. Synopsis of the use of frozen human semen since 1964: state of the art of human semen banking. *Fertil Steril* 1973;24:397–412. Level III.
2. First baby born of frozen embryo. *New York Times*; 1984. Level III.
3. Chen C. Pregnancy after human oocyte cryopreservation. *Lancet* 1986;1:884–6. Level III.
4. Oktay K, Cil AP, Bang H. Efficiency of oocyte cryopreservation: a meta-analysis. *Fertil Steril* 2006;86:70–80. Level III.
5. Cobo A, Diaz C. Clinical application of oocyte vitrification: a systematic review and meta-analysis of randomized controlled trials. *Fertil Steril* 2011;96:277–85. Level III.
6. Shaw JM, Oranratnachai A, Trounson AO. Fundamental cryobiology of mammalian oocytes and ovarian tissue. *Theriogenology* 2000;53:59–72. Level III.
7. Baka SG, Toth TL, Veeck LL, Jones HW Jr, Muasher SJ, Lanzendorf SE. Evaluation of the spindle apparatus of in-vitro matured human oocytes following cryopreservation. *Hum Reprod* 1995;10:1816–20. Level II-2.
8. Bromfield JJ, Cotichio G, Hutt K, Sciajno R, Borini A, Albertini DF. Meiotic spindle dynamics in human oocytes following slow-cooling cryopreservation. *Hum Reprod* 2009;24:2114–23. Level II-2.
9. Fabbri R, Porcu E, Marsella T, Rocchetta G, Venturoli S, Flamigni C. Human oocyte cryopreservation: new perspectives regarding oocyte survival. *Hum Reprod* 2001;16:411–6. Level II-2.
10. Stachecki JJ, Willadsen SM. Cryopreservation of mouse oocytes using a medium with low sodium content: effect of plunge temperature. *Cryobiology* 2000;40:4–12. Level II-3.

11. Boldt J, Tidswell N, Sayers A, Kilani R, Cline D. Human oocyte cryopreservation: 5-year experience with a sodium-depleted slow freezing method. *Reprod Biomed Online* 2006;13:96–100. Level II-3.
12. Bianchi V, Lappi M, Bonu MA, Borini A. Oocyte slow freezing using a 0.2-0.3 M sucrose concentration protocol: is it really the time to trash the cryopreservation machine? *Fertil Steril* 2012;97:1101–7. Level II-3.
13. Sathananthan AH, Trounson A, Freemann L, Brady T. The effects of cooling human oocytes. *Hum Reprod* 1988;3:968–77. Level II-2.
14. Trad FS, Toner M, Biggers JD. Effects of cryoprotectants and ice-seeding temperature on intracellular freezing and survival of human oocytes. *Hum Reprod* 1999;14:1569–77. Level II-2.
15. Parmegiani L, Bertocci F, Garelo C, Salvarani MC, Tambuscio G, Fabbri R. Efficiency of human oocyte slow freezing: results from five assisted reproduction centres. *Reprod Biomed Online* 2009;18:352–9. Level II-2.
16. Smith GD, Serafini PC, Fioravanti J, Yadid I, Coslovsky M, Hassun P, et al. Prospective randomized comparison of human oocyte cryopreservation with slow-rate freezing or vitrification. *Fertil Steril* 2010;94:2088–95. Level I.
17. Gook DA, Edgar DH. Human oocyte cryopreservation. *Human Reprod Update* 2007;13:591–605. Level III.
18. Ciotti PM, Porcu E, Notarangelo L, Magrini O, Bazzocchi A, Venturoli S. Meiotic spindle recovery is faster in vitrification of human oocytes compared to slow freezing. *Fertil Steril* 2009;91:2399–407. Level I.
19. Parmegiani L, Cognigni GE, Bernardi S, Cuomo S, Ciampaglia W, Infante FE, et al. Efficiency of aseptic open vitrification and hermetical cryostorage of human oocytes. *Reprod Biomed Online* 2011;23:505–12. Level I.
20. Gook DA, Osborn SM, Bourne H, Johnston WI. Fertilization of human oocytes following cryopreservation; normal karyotypes and absence of stray chromosomes. *Hum Reprod* 1994;9:684–91. Level II-2.
21. Porcu E, Fabbri R, Seracchioli R, Ciotti PM, Magrini O, Flamigni C. Birth of a healthy female after intracytoplasmic sperm injection of cryopreserved human oocytes. *Fertil Steril* 1997;68:724–6. Level III.
22. Polak de Fried E, Notrica J, Rubinstein M, Marazzi A, Gomez Gonzalez M. Pregnancy after human donor oocyte cryopreservation and thawing in association with intracytoplasmic sperm injection in a patient with ovarian failure. *Fertil Steril* 1998;69:555–7. Level III.
23. Fabbri R, Porcu E, Marsella T, Primavera MR, Seracchioli R, Ciotti PM, et al. Oocyte cryopreservation. *Hum Reprod* 1998;13(Suppl 4):98–108. Level II-2.
24. Cobo A, Kuwayama M, Perez S, Ruiz A, Pellicer A, Remohi J. Comparison of concomitant outcome achieved with fresh and cryopreserved donor oocytes vitrified by the Cryotop method. *Fertil Steril* 2008;89:1657–64. Level I.
25. Rienzi L, Romano S, Albricci L, Maggiulli R, Capalbo A, Baroni E, et al. Embryo development of fresh 'versus' vitrified metaphase II oocytes after ICSI: a prospective randomized sibling-oocyte study. *Hum Reprod* 2010;25:66–73. Level I.
26. Cobo A, Meseguer M, Remohi J, Pellicer A. Use of cryo-banked oocytes in an ovum donation programme: a prospective, randomized, controlled, clinical trial. *Hum Reprod* 2010;25:2239–46. Level I.
27. Borini A, Levi Setti PE, Anserini P, De Luca R, De Santis L, Porcu E, et al. Multicenter observational study on slow-cooling oocyte cryopreservation: clinical outcome. *Fertil Steril* 2010;94:1662–8. Level II-2.
28. Scaravelli G, Vigiliano V, Mayorga JM, Bolli S, De Luca R, D'Aloja P. Analysis of oocyte cryopreservation in assisted reproduction: the Italian National Register data from 2005 to 2007. *Reprod Biomed Online* 2010;21:496–500. Level II-2.
29. Hodes-Wertz B, Noyes N, Mullin C, McCaffrey C, Grifo JA. Retrospective analysis of outcomes following transfer of previously cryopreserved oocytes, pronuclear zygotes and supernumerary blastocysts. *Reprod Biomed Online* 2011;23:118–23. Level II-2.
30. Grifo JA, Noyes N. Delivery rate using cryopreserved oocytes is comparable to conventional in vitro fertilization using fresh oocytes: potential fertility preservation for female cancer patients. *Fertil Steril* 2010;93:391–6. Level II-2.
31. Kim TJ, Laufer LR, Hong SW. Vitrification of oocytes produces high pregnancy rates when carried out in fertile women. *Fertil Steril* 2010;93:467–74. Level II-2.
32. Nagy ZP, Chang CC, Shapiro DB, Bernal DP, Elsner CW, Mitchell-Leef D, et al. Clinical evaluation of the efficiency of an oocyte donation program using egg cryo-banking. *Fertil Steril* 2009;92:520–6. Level II-2.
33. Rienzi L, Cobo A, Paffoni A, Scarduelli C, Capalbo A, Vajta G, et al. Consistent and predictable delivery rates after oocyte vitrification: an observational longitudinal cohort multicentric study. *Hum Reprod* 2012;27:1606–12. Level II-2.
34. Ubaldi F, Anniballo R, Romano S, Baroni E, Albricci L, Colamaria S, et al. Cumulative ongoing pregnancy rate achieved with oocyte vitrification and cleavage stage transfer without embryo selection in a standard infertility program. *Hum Reprod* 2010;25:1199–205. Level II-2.
35. Cao YX, Chian RC. Fertility preservation with immature and in vitro matured oocytes. *Semin Reprod Med* 2009;27:456–64. Level III.
36. Parmegiani L, Garelo C, Granella F, Guidetti D, Bernardi S, Cognigni GE, et al. Long-term cryostorage does not adversely affect the outcome of oocyte thawing cycles. *Reprod Biomed Online* 2009;19:374–9. Level II-2.
37. Cobo A, Rubio C, Gerli S, Ruiz A, Pellicer A, Remohi J. Use of fluorescence in situ hybridization to assess the chromosomal status of embryos obtained from cryopreserved oocytes. *Fertil Steril* 2001;75:354–60. Level II-2.
38. Noyes N, Porcu E, Borini A. Over 900 oocyte cryopreservation babies born with no apparent increase in congenital anomalies. *Reprod Biomed Online* 2009;18:769–76. Level II-3.
39. Chian RC, Huang JY, Tan SL, Lucena E, Saa A, Rojas A, et al. Obstetric and perinatal outcome in 200 infants conceived from vitrified oocytes. *Reprod Biomed Online* 2008;16:608–10. Level II-3.
40. Cobo A, Bellver J, de los Santos MJ, Remohi J. Viral screening of spent culture media and liquid nitrogen samples of oocytes and embryos from hepatitis B, hepatitis C, and human immunodeficiency virus chronically infected women undergoing in vitro fertilization cycles. *Fertil Steril* 2012;97:74–8. Level II-3.
41. Gera PS, Tatpati LL, Allemand MC, Wentworth MA, Coddington CC. Ovarian hyperstimulation syndrome: steps to maximize success and minimize effect for assisted reproductive outcome. *Fertil Steril* 2010;94:173–8. Level II-2.
42. Lee SJ, Schover LR, Partridge AH, Patrizio P, Wallace WH, Hagerty K, et al. American Society of Clinical Oncology recommendations on fertility preservation in cancer patients. *J Clin Oncol* 2006;24:2917–31. Level III.
43. Borgstrom B, Hreinsson J, Rasmussen C, Sheikhi M, Fried G, Keros V, et al. Fertility preservation in girls with turner syndrome: prognostic signs of the presence of ovarian follicles. *J Clin Endocrinol Metab* 2009;94:74–80. Level III.
44. Practice Committee of the American Society for Reproductive Medicine. Increased maternal cardiovascular mortality associated with pregnancy in women with Turner syndrome. *Fertil Steril* 2012;97:282–4. Level III.
45. Song WY, Sun YP, Jin HX, Xin ZM, Su YC, Chian RC. Clinical outcome of emergency egg vitrification for women when sperm extraction from the testicular tissues of the male partner is not successful. *Syst Biol Reprod Med* 2011;57:210–3. Level II-3.
46. Virant-Klun I, Bacer-Kermavner L, Tomazevic T, Vrtacnik-Bokal E. Slow oocyte freezing and thawing in couples with no sperm or an insufficient number of sperm on the day of in vitro fertilization. *Reprod Biol Endocrinol* 2011;9:19. Level II-2.
47. Borini A, Levi Setti PE, Anserini P, De Luca R, De Santis L, Porcu E, La Sala GB, Ferraretti A, Bartolotti T, Coticchio G, Scaravelli G. Multicenter observational study on slow-cooling oocyte cryopreservation: clinical outcome. *Fertil Steril* 2010;94:1662–8. Level II-2.
48. Practice Committee of Society for Assisted Reproductive Technology; Practice Committee of American Society for Reproductive Medicine. Essential elements of informed consent for elective oocyte cryopreservation: a Practice Committee opinion. *Fertil Steril* 2008;90(5 Suppl):S134–5. Level III.