Fertility preservation in females with malignant disease-1: causes, clinical needs and indications

Malignitesi olan kadınlarda doğurganlığın korunması-1: nedenler, klinik gereksinimler ve endikasyonlar

Murat Sönmezer, Sinan Özkavukçu
Department of Obstetrics and Gynecology, IVF Unit, Ankara University School of Medicine, Ankara, Turkey

Abstract

Cancer incidence is progressively increasing in parallel with an increase in the rate of cancer survivors with the help of advanced treatment modalities. By the year 2010, it is estimated that one in every 250 persons will have survived a childhood malignancy. The increased rates of survival bring about complications related to reproductive health. Cytotoxic treatments due to chemo- and radiotherapy or bone marrow transplantation suppress or irreversibly harm not only female ovarian reserve but also male testicular sperm production. In this review, cryopreservation of gametes and gonads with fertility preservation options and indications prior to cancer treatments are discussed. *(Turk J Hematol 2009; 26: 106-13)*
Key words: Fertility preservation, cancer, chemotherapy, radiotherapy, cryopreservation

Received: December 23, 2008 Accepted: April 8, 2009

Özet

Kanser insidansındaki artışa paralel olarak, gelişen tedavi modalitelerinin yardımıyla, bu hastalıktan sağkalım oranları da her geçen gün artmaktadır. 2010 yılında her 250 kişiden birinin çocukluk çağı kanserlerinden tedavi olmuş bir birey olacağı hesaplanmaktadır. Sağkalım oranlarının artışı, üreme sağlığını etkileyen komplikasyonları da beraberinde getirmektedir. Yüksek kemoterapi ve radyoterapi ya da kemik iliği transplantasyonuna bağlı sitotoksik tedaviler gerek over rezervini, gerekse testiküler sperm üretimini baskılayan ya da geri dünüşümsüz olarak hasara uğratan tedavi seçenekleridir. Bu derlemede, bahsedilen tedaviler öncesinde hastaların gonad ya da gametlerinin kriyoprezervasyonu ve kür sonrası fertilitenin geri kazandırılmasıyla ilgili seçenekler ve endikasyonlar ele alınmıştır. *(Turk J Hematol 2009; 26: 106-13)*
Anahtar kelimeler: Doğurganlığın korunması, kanser, kemoterapi, radyoterapi, kriyoprezervasyon

Geliş tarihi: 23 Aralık 2008 Kabul tarihi: 08 Nisan 2009

Address for Correspondence: Dr. Murat Sönmezer, Ankara University School of Medicine, Department of Obstetric and Gynecology, IVF Unit, Ankara Turkey
Phone: +90 312 595 70 27 E-mail: msonmezer@gmail.com

Introduction

Cancer incidence is gradually increasing while deaths caused by malignant diseases decrease. When all female cancers are considered, despite an increase in the cancer incidence by 0.3% annually from 1987 to 1999, the death rates for all cancers combined decreased by 0.6% annually from 1992 to 1999 as a result of improvements in current treatment modalities including surgical techniques, radiation therapy, multi-agent chemotherapy, and hematopoietic stem cell transplantation (HSCT) [1,2]. By 2010, one in every 250 persons is estimated to have survived childhood malignancies. Thus, more patients survive cancer every year but face the challenging long-term side effects, especially related to their reproductive system. The risk of ovarian failure may increase up to nine-fold in female cancer survivors receiving cyclophosphamide-based combination chemotherapy [3,4], and ovarian failure is almost inevitable in patients undergoing preconditioning with chemoradiation before HSCT [5]. It is also reported that an ovarian radiation dose of more than 6 Gy usually results in permanent infertility [6]. Chemo-/radiotherapy sequelae may impair the quality of the pregnancy as well, with an increased risk of early pregnancy loss, premature labor, and low birth weight, even if the patients were not sterilized after treatments [7-9].

The introduction of assisted reproduction and its current worldwide utilization has resulted in the development of successful cryopreservation techniques for surplus embryos. The techniques created for embryo cryopreservation have further been applied to the unfertilized mature and immature human oocyte [10,11] and ovarian tissue [12-17]. Currently, embryo cryopreservation is the preferred method to preserve future fertility because of reasonable post-thaw survival, implantation, and delivery rates, if the patient has a life-partner or access to a sperm donation program. However, oocyte or ovarian cryopreservation can be the solitary option for young or unmarried female patients. Pre-pubertal children, on the grounds of ethical concerns regarding ovulation induction and oocyte retrieval, and women who cannot delay cancer treatment for the 2-4 week period necessary to perform ovulation induction, are not candidates for embryo cryopreservation.

Numerous non-neoplastic diseases are also treated with cytotoxic chemotherapy and radiation. In some of them, chemotherapy or radiation therapy is also used in extreme doses to ablate the preexisting bone marrow in HSCT. The indications for HSCT that extend beyond cancer now include some autoimmune diseases unresponsive to immunosuppressive therapy, diseases associated with genetically abnormal stem cells (hemoglobinopathies and enzyme deficiency disorders), and those associated with the deficiency of bone marrow stem cell products [18-23]. In this article, the current indications and techniques of fertility preservation will be reviewed from the gynecologic point of view, which mainly focuses on embryo, oocyte and ovarian cryopreservation.

Chemotherapy- and radiotherapy-associated gonadal damage

Multiagent chemotherapy constitutes the basis of the modern cancer treatment. Ovaries, which are stocked with irreplaceable follicles, are extremely sensitive to most cytotoxic drugs [23,24]. The end result of the chemotherapy can range from damage to steroid-producing cells and/or oocytes of developing ovarian follicles (granulosa and theca cells), which can cause temporary amenorrhea, to apoptotic death of primordial follicles, which results in premature ovarian failure (POF). Ultrastructurally, ovarian exposure to chemotherapeutics is associated with marked follicle loss [25]. Factors that can potentially modify the risk of chemotherapy-induced ovarian failure are summarized in Table 1.

Some chemotherapeutic agents are more commonly associated with permanent and irreversible gonadal damage, such as cyclophosphamide, chlorambucil, melphalan, busulfan, nitrogen mustard, procarbazine, ifosfamide, and thiotepa [23,26-30]. Among the moderately gonadotoxic agents are cisplatin and adriamycin, while bleomycin, actinomycin D, vincristine, methotrexate, and 5-fluorouracil are associated with mild or no gonadotoxicity (Table 2). Although there is limited evidence, paclitaxel may also be gonadotoxic, but this remains to be verified [31]. In Table 2, we classify the gonadal risk of commonly used chemotherapeutic agents after a comprehensive literature search [32].

Cyclophosphamide is the most recognized agent to cause damage to oocytes and granulosa cells. In a recent mouse study, cyclophosphamide-induced follicular damage occurred in a dose-dependent manner, even at low doses of 20 mg/kg [33]. Relative risk of POF was reported to be between 4 and 9.3 in patients receiving cyclophosphamide [34,35]. During the last 10-15 years before the onset of menopause, primordial follicle loss is accelerated, which is reflected by a constant decrease in inhibin B levels and increase in follicle-stimulating hormone (FSH) levels. As a consequence, a smaller number of follicles that are more prone to cell division errors begin to grow each cycle, until menopause occurs, when the number of follicles falls below 1.000. Because of this, older women with a low primordial follicle pool have a higher risk of developing ovarian failure compared with young women with higher primordial follicle numbers. Consistent with this biological fact, earlier studies demonstrated that a cumulative cyclophosphamide dose of 5.2 g caused amenorrhea in women in their forties, 9.3 g in women in their thirties, and 20.4 g in women in their twenties [27].

Table 1. Factors that can modify the risk of chemotherapy-related gonadal failure

Age of the patient
Type of chemotherapeutic agents
Cumulative dose of alkylating agent
Concomitant use of abdominopelvic radiation therapy
Ovarian reserve
Schedule of implementation

Table 2. The degree of gonadotoxicity associated with chemotherapeutic agents

High ovarian failure risk
Cyclophosphamide
Chlorambucil
Melphalan
Busulfan
Nitrogen mustard
Procarbazine
Moderate ovarian failure risk
Cisplatin
Adriamycin
Paclitaxel
No or low ovarian failure risk
Methotrexate
5-Fluorouracil
Vincristine
Actinomycin D
Bleomycin
Commonly used novel agents with as yet undetermined risk
Irinotecan
Imatinib

Hematopoietic stem cell transplantation and the risk of ovarian failure

There has been a dramatic increase in the survival of childhood cancer patients in recent years as a result of HSCT. On the contrary, high-dose chemotherapy used for conditioning before HSCT is extremely gonadotoxic, as has been consistently demonstrated in the previous studies. In the acute ovarian failure of childhood cancer survivor study [36] that included 3309 childhood cancer survivors, exposure to more than 1000 cGy ovarian radiation, age and treatment with cyclophosphamide or procarbazine were found as independent risk factors for development of POF in a multivariable logistic regression model. An important finding from the previous studies is that despite timely menarche, FSH concentrations show a tendency to rise to menopausal levels in children exposed to high-dose chemotherapy during the pre-pubertal period. This highlights the fact that occurrence of timely menarche does not guarantee preserved ovarian function. Brachet et al. [37] found that seven of 10 children with sickle cell disease receiving busulfan (14 or 16 mg/kg) and cyclophosphamide (200 mg/kg) as preconditioning before HSCT developed POF. In the remaining three who had spontaneous puberty, serum FSH levels were very high at the time of puberty and slowly normalized thereafter. It is important to underline that three girls with ovarian function recovery differed from the seven others by the lower busulphan dose of the conditioning regimen they received (14 rather than 16 mg/kg). In a survey including 2819 childhood cancer survivors, Sklar et al. [38] demonstrated that children who receive chemotherapy are at an extremely high risk for POF. In that study, the authors followed a cohort of children who were diagnosed with a malignancy before the age of 21 and were menstruating for at least five years afterward. The patients in the study group were compared with their 1065 siblings. The median age at diagnosis was 7 (range, 0-20) and the median age at study was 29 (range, 18-50). The risk of developing ovarian failure was found 13.2-fold increased (range, 3.26-53.51) in those exposed to chemotherapy compared with their siblings. Although rare, resumption of menstruation years after the diagnosis of POF has also been reported [39].

How to assess post-chemotherapy gonadal function?

Most of the long-term follow-up studies assessing post-chemotherapy ovarian function rely on menstruation as the only surrogate marker. Even though irregular menstruation or amenorrhea is highly likely to occur during the chemotherapy, even lasting for a considerable period after completion of the chemotherapy, many patients return to a pre-chemotherapy menstrual pattern. Hormonal reversal of a hypergonadotropic state that commonly occurs during the courses of chemotherapy to a normo-gonadotropic state may also be expected [40]. However, these women will always have a high risk of developing premature menopause during their later reproductive life. The fact that ovulation may occur despite loss of half of the follicular pool in rodents indicates that indirect assessment of ovarian reserve is an unreliable tool [23]. Ovarian reserve diminishes when FSH levels on the third day of the menstrual cycle are more than 12 IU/ml or estradiol is more than 75 pg/ml, whereas ovarian failure is diagnosed when FSH is found as more than 40 mIU/ml in two measurements regardless of menstrual bleeding. Anti-Müllerian hormone (AMH) has recently been suggested as the most reliable marker of ovarian reserve [41]. In normal ovulating women, serum AMH levels are relatively constant during the menstrual cycle, serum concentrations of which show a rapid decline after 37 years. Anderson et al. [42] showed that compared with estradiol and FSH, AMH showed a more rapid and sustained change after chemotherapy. Moreover, the decrease in AMH occurred without a significant decrease in inhibin-B or increase in FSH concentrations. The severity and rapidity of the decrease in AMH concentrations compared with the partial decline in inhibin-B concentrations might reflect primordial and preantral follicles as the primary site of toxicity. This supports the observation that, even though there may be no clinical signs of ovarian failure, there is always damage to follicular reserve in proportion to the cumulative dose of chemotherapeutic agents that might not be detectable with routinely used laboratory tests. It is important to note that AMH is not influenced by confounding factors such as oral contraceptive use, day of menstrual cycle, or pregnancy. In a study assessing post-chemotherapy ovarian function [43], despite the fact that all eight breast cancer study patients resumed menstruation after chemotherapy, three had irregular menstrual cycles, and five had undetectable inhibin-B levels or FSH values more than 50

IU/ml, suggesting some degree of impairment in ovarian reserve. Another study [44] found that, compared with FSH and inhibin-B, AMH constitutes the most sensitive predictor of ovarian reserve in women treated with chemotherapy for Hodgkin's lymphoma. Furthermore, most women who reported one or more pregnancies had normal AMH levels for age at the time of the study. Similarly, it has been demonstrated that in breast cancer patients, AMH levels declined despite continued menstrual activity [45] and ovarian reserve markers were altered in those who seemingly had normal menstruation post-chemotherapy [46]. Giuseppe et al. [47] assessed FSH, luteinizing hormone (LH), AMH, inhibin-B, and antral follicle count (AFC) and suggested the combination of AFC and AMH as having the best predictive value for ovarian reserve with a high sensitivity (83%) and specificity (88%) in patients treated with chemotherapy for Hodgkin's lymphoma. In 25 patients with hematological malignancies, serum AMH concentrations were measured before and after cancer therapy and compared with normoovulatory controls. Despite having menstrual cycles and despite some patients conceiving spontaneously after chemotherapy, AMH levels and AFC were decreased, showing some degree of ovarian damage [48].

Radiotherapy

Ionizing radiation is a well-recognized cause of ovarian damage and permanent infertility. Gonadal damage occurs not only by direct exposure to radiation such as in the case of pelvic or low abdominal irradiation, but scattering of radiation may also cause considerable damage even if gonads are outside the radiation field. Radiation causes a dose-related reduction in the primordial follicle pool [49]. The human oocyte is extremely sensitive to radiation, and irradiation at ovarian dose >6 Gy usually causes irreversible ovarian failure [6]. Wallace et al. [50,51] demonstrated that <4 Gy is enough to destroy half of the oocyte population (LDL50 <4 Gy); however, very recently, using a revised mathematical model, the same authors suggested that the LDL50 of the oocytes was <2 Gy. Age at the time of exposure to radiotherapy, extent and type of radiation therapy (e.g. abdominal, pelvic external beam irradiation, intracavitary brachytherapy) and fractionation schedule are important prognostic indicators for development of ovarian failure [52-56]. In mice, radiation-induced chromosome damage in the oocytes was more evident in older compared with younger animals [54]. In general, irradiation is more toxic when given in single dose compared to fractionated doses. Stillman et al. [57] investigated the risk of ovarian failure among 182 long-term survivors of childhood cancers receiving abdominal radiotherapy. The mean follow-up was 16.4 years. Ovarian failure occurred in 68% of the patients when both ovaries were in the irradiation field, and in 14% of the patients when both ovaries were at the edge of the treatment field. None of the 122 children developed ovarian failure when one or both ovaries were outside the abdominal treatment field. In another study, failure in pubertal development or premature menopause was observed in 37 of 38 patients who received external abdominal irradiation during childhood for intra-abdominal tumors in doses ranging from 20 to 30 Gy [58]. Sanders et al. [59] reported the probability of ovarian failure in patients receiving cyclophosphamide and total body irradiation for HSCT as 1.00 at one year. Failure in pubertal development may be the first sign of ovarian failure in these patients who received radiotherapy during childhood.

Indications for fertility preservation

As a result of improvements in cancer treatment and in the ability to detect tumors in their early stages by well-established screening programs for some cancers, life expectancy has strikingly increased. Furthermore, a cure is now possible for many childhood and adult cancers. Notably, cure rates approximate 90% in certain childhood cancers. A beneficial effect of cytotoxic treatment in various non-malignant diseases has also been repeatedly demonstrated. The idea of cryopreserving ovarian tissue is based on the finding that the ovarian cortex harbors primordial follicles that are more resistant to cryo-injury than are mature oocytes. Although the clinical indications for ovarian tissue cryopreservation are almost identical to those for the oocyte, there are fewer logistical restrictions in offering this technique. Despite the limited data on successful pregnancy rates, ovarian tissue cryopreservation has broader applications and, in theory, a greater fertility potential than oocyte cryopreservation because of the far larger number of oocytes preserved. Extending indications for ovarian tissue cryopreservation are listed below. A detailed list of indications is presented in Table 3 [60].

Cancers in children

Adult and childhood cancers are the most common indication for fertility preservation. Even though cancer is still the second leading cause of death in children, there has been remarkable improvement in the cure rates of many childhood cancers over the last three decades [1]. Among the most common cancers encountered during childhood are leukemia, Hodgkin's and non-Hodgkin's lymphomas, tumors of the central nervous system, soft tissue sarcomas, and renal tumors [61-66]. Acute lymphoblastic leukemia is the most common childhood cancer, with more than 2.100 new cases and 2,000 long-term survivors each year [66,67]. The five-year survival rate for all childhood cancers is approximately 80%, with a higher percentage for lymphomas (94%) and Wilms' tumor (91%) [66-69]. Although many children survive cancer because of improved treatment modalities, they are certainly not immune to the gonadotoxic effects of various cancer treatments. Ovarian tissue cryopreservation may be the only acceptable method for any pre-pubertal or pre-menarchal female patients receiving chemotherapy, pelvic radiotherapy, HSCT, or oophorectomy for benign disease or prophylaxis [23]. The greatest benefit from the procedure is expected in children, since they have the highest number of primordial follicles [16]. With ovarian tissue freezing, no ovarian stimulation is needed; therefore, time restrictions for cancer therapy are fewer, and there is no risk of stimulating estrogen-sensitive cancer following ovarian stimulation [23]. Additionally, it avoids ethical concerns regarding ovarian stimulation and oocyte retrieval in children.

Table 3. Indications of fertility preservation

Cancer in children
Hodgkin and non-Hodgkin lymphoma
Leukemias
Ewing's sarcoma
Neuroblastoma
Wilms' tumor
Pelvic osteosarcoma
Genital rhabdomyosarcoma
Breast cancer*
Infiltrative ductal histological subtype
Stage I–III
Cancer of the cervix
Squamous cell carcinoma
Adeno/adenosquamous carcinoma
Autoimmune and hematological diseases
Systemic lupus erythematosus
Behçet's disease
Steroid-resistant glomerulonephritis
Inflammatory bowel disease
Sickle cell disease
Rheumatoid arthritis
Progressive systemic sclerosis
Pemphigus vulgaris
Juvenile idiopathic arthritis
Multiple sclerosis
Autoimmune thrombocytopenia
Aplastic anemia
Benign ovarian disease
Endometriosis
Benign ovarian lesions requiring repeated surgeries
Patients receiving pelvic radiation
Solid organ tumors presenting in the pelvis
Osteosarcoma
Ewing's sarcoma
Tumors of the spinal cord
Retroperitoneal sarcoma
Rectal cancer
Benign bone tumors
Vanishing bone disease
Prophylactic oophorectomy
BRCA-I and II germline mutation carriers
Hematopoietic stem cell transplantation
Malignant diseases
Genetic, hematological, and autoimmune disorders
Patients undergoing surgery for gynecological cancers**

* Ovarian cryopreservation is not recommended in advanced stage breast cancer. Compared to infiltrative ductal histologic subtype, infiltrative lobular breast cancer is more prone to metastasize to the ovaries in the early stages

**Fertility-preserving surgery includes conization or trachelectomy for early stage cervical cancer, fertility-preserving surgery for early stage ovarian cancer and hormonal treatment for endometrial cancer

Cancers in adults

The death rates from cancer in women have fallen, despite increased incidence during the 1990s. Approximately 8% of these cancers occur in reproductive aged women under the age of 40 years. Breast cancer, the most common cancer in women during the reproductive years, afflicted approximately 216,000 women in the United States in 2004 [66]. The five-year survival rate in breast cancer now approaches 90%. Most of the patients with breast cancer are subjected to cyclophosphamide-based gonadotoxic chemotherapy. In breast cancer, fortunately, unlike other malignant diseases, there is approximately a six-week hiatus between the initial surgery and chemotherapy. These patients may resort to assisted reproductive technologies during this time period. However, in theory, conventional ovarian stimulation protocols are thought to affect the growth of breast cancer as a result of supraphysiological estrogen concentrations. Novel stimulation protocols with tamoxifen and aromatase inhibitors are suggested as safer protocols in these patients. In addition, since occult ovarian metastasis is extremely rare, with the exception of stage IV disease and lobular carcinoma, these patients may resort to ovarian tissue cryopreservation [23,70]. Cancer of the cervix is a serious health problem afflicting 500.000 women worldwide each year, with almost half of them under the age of 35 [71]. Patients with advanced stage disease and those with early stage disease who are found to have high risk factors receive pelvic or pelvic/paraaortic radiation therapy. Squamous cell cancer of the cervix, which is the most often encountered subtype, rarely metastasizes to the ovaries, whereas this may occur at a rate as high as 12% for adenocarcinoma and adenosquamous carcinoma. Ovarian transposition might be performed; however, success rates vary greatly because of damage to the vasculature during the procedure. Ovulation induction might be risky since there is risk of bleeding from the fragile cervix during oocyte retrieval. Ovarian tissue can be removed in selected patients for cryopreservation during primary cancer surgery. Another group of patients that are potential candidates for ovarian cryopreservation are those carrying BRCA I and II mutations. Despite the fact that the risk of peritoneal cancer cannot be totally eliminated in BRCA-positive patients, prophylactic oophorectomy is suggested as soon as childbearing is completed or by the age 35-40 years to decrease the risk of ovarian and breast cancer [72,73]. Cortical pieces of ovarian tissue in those with a desire for fertility can be frozen for future use.

Autoimmune diseases

Autoimmune diseases can also affect women of reproductive age. There have been an increasing number of reports regarding the use of cytotoxic treatment, especially with cyclophosphamide, in autoimmune diseases, including systemic lupus erythematosus, steroid- resistant glomerulonephritis, Behçet's disease, inflammatory bowel diseases, and pemphigus vulgaris [74-78]. Pieces of ovarian tissue may be harvested for possible future use in these patients in order to retain fertility.

Experimental approaches

When the risk of ovarian involvement with cancer cells is high, some other experimental options may be considered. It has been possible to isolate primordial follicles from human ovarian tissue, but there has been no success in growing them

in vitro to get a healthy offspring [79]. Early stage preantral follicles can only be grown for brief periods of time in three-dimensional culture systems. Another potential approach is xenografting human ovarian tissue in immunodeficient mice, where human follicles can be grown to antral stages and ovulated. However, the applicability of xenografting in the clinical setting has not been determined due to the risk of trans-species viral infections [80,81]. The mechanism of age-related as well as chemo- or radiotherapy-induced loss in the ovarian germ cell population is proposed to be mediated by programmed cell death, i.e. apoptosis. Sphingosine-1-phosphate (S1P), a bioactive sphingolipid metabolite, is an important lipid mediator and has many actions both inside and outside the cell. It was demonstrated that wild-type mice treated with S1P resisted both developmental and cancer therapy-induced apoptosis. Radiation-induced oocyte loss could be completely prevented by S1P therapy in wild-type mice, and no genomic damage in mice pretreated with S1P before receiving ionizing radiation could be demonstrated [82]. Another experimental approach has become a current issue with a report by Silber et al. [83], in which a transplantation of ovarian cortical tissue took place between 24-year-old monozygotic twins, one of whom suffered POF. The question arises of whether allogeneic ovarian transplantation is possible in the future for females with ovarian dysfunction following cancer therapies.

Conclusion

Fertility preservation requires a multimodality approach. Depending on a patient's age, the type of cancer treated, time constraints, availability of a partner, and whether there is ovarian involvement, a different procedure may be needed for each cancer survivor facing treatment-related infertility. Physicians should take a comprehensive approach in counseling their patients regarding fertility preservation procedures.

References

1. Reis LAG, Percy CL, Bunin GR. Introduction. In: Ries LAG, Smith MA, Gurney JG, Linet M, Tamra T, Young JL, Bunin GR, editors. Cancer Incidence and Survival Among Children and Adolescents: United States SEER Program 1975-1995, National Cancer Institute, SEER Program. Bethesda, MD: NIH Pub. No. 99-4649, 1999: 1-15.
2. Jemal A, Tiwari RC, Murray T, Ghafoor A, Samuels A, Ward E, Feuer EJ, Thun MJ. Cancer statistics, 2004. CA Cancer J Clin. 2004;54:8-29.
3. Byrne J, Fears TR, Gail MH, Pee D, Connelly RR, Austin DF, Holmes GF, Holmes FF, Latourette HB, Meigs JW. Early menopause in long-term survivors of cancer during adolescence. Am J Obstet Gynecol 1992;166:788-93.
4. Meirow D, Nugent D. The effects of radiotherapy and chemotherapy on female reproduction. Hum Reprod Update 2001;27:535-43.
5. Sanders JE, Buckner CD, Amos D, Levy W, Appelbaum FR, Doney K, Storb R, Sullivan KM, Witherspoon RP, Thomas ED. Ovarian function following marrow transplantation for aplastic anemia or leukemia. J Clin Oncol 1988;6:813-8.
6. Howell S, Shalet S. Gonadal damage from chemotherapy and radiotherapy. Endocrinol Metab Clin North Am 1998;827:927-43.
7. Sanders JE, Hawley J, Levy W, Gooley T, Buckner CD, Deeg HJ, Doney K, Storb R, Sullivan K, Witherspoon R, Appelbaum FR. Pregnancies following high-dose cyclophosphamide with or without high-dose busulfan or total body irradiation and bone marrow transplantation. Blood 1996;87:3045-52.
8. Chiarelli AM, Marrett LD, Darlington GA. Pregnancy outcomes in females after treatment for childhood cancer. Epidemiology 2000;11:161-6.
9. Green DM, Whitton JA, Stovall M, Mertens AC, Donaldson SS, Ruymann FB, Pendergrass TW, Robison LL. Pregnancy outcome of female survivors of childhood cancer: a report from the Childhood Cancer Survivor Study. Am J Obstet Gynecol 2002;187:1070-80.
10. 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.
11. 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.
12. Bahadur G, Steele SJ. Ovarian tissue cryopreservation for patients. Hum Reprod 1996;11:2215-6.
13. Oktay K, Karlikaya G. Ovarian function after transplantation of frozen, banked autologous ovarian tissue. N Engl J Med 2000;342:1919.
14. Oktay K, Aydin BA, Karlikaya G. A technique for laparoscopic transplantation of frozen-banked ovarian tissue. Fertil Steril 2001;75:1212-6.
15. Oktay K, Economos K, Kan M, Rucinski J, Veeck L, Rosenwaks Z. Endocrine function and oocyte retrieval after autologous transplantation of ovarian cortical strips to the forearm. J Am Med Assoc 2001;286:1490-3.
16. Poirot C, Vacher-Lavenu MC, Helardot P, Guibert J, Brugieres L, Jouannet P. Human ovarian tissue cryopreservation: indications and feasibility. Hum Reprod 2002;17:1447-52.
17. Gook DA, Edgar DH, Borg J, Archer J, McBain JC. Diagnostic assessment of the developmental potential of human cryopreserved ovarian tissue from multiple patients using xenografting. Hum Reprod 2005;20:72-8.
18. Oktay K, Kan MT, Rosenwaks Z. Recent progress in oocyte and ovarian tissue cryopreservation and transplantation. Curr Opin Obstet Gynecol 2001;13:263-8.
19. Slavin S, Nagler A, Aker M, Shapira MY, Cividalli G, Or R. Nonmyeloablative stem cell transplantation and donor lymphocyte infusion for the treatment of cancer and life-threatening non-malignant disorders. Rev Clin Exp Hematol 2001;5:135-46.
20. Burt RK, Traynor AE, Craig R, Marmont AM. The promise of hematopoietic stem cell transplantation for autoimmune diseases. Bone Marrow Transplant 2003;31:521-4.

21. Vermylen C. Hematopoietic stem cell transplantation in sickle cell disease. Blood Rev 2003;17:163-6.
22. Lawson SE, Roberts IA, Amrolia P, Dokal I, Szydlo R, Darbyshire PJ. Bone marrow transplantation for beta-thalassaemia major: the UK experience in two paediatric centres. Br J Haematol 2003;120:289-95.
23. Sönmezer M, Oktay K. Fertility preservation in female patients. Hum Reprod Update 2004;10:251-66.
24. Oktay K. Ovarian tissue cryopreservation and transplantation: preliminary findings and implications for cancer patients. Hum Reprod Update 2001;7:526-34.
25. Familiari G, Caggiati A, Nottola SA, Ermini M, Di Benedetto MR, Motta PM. Ultrastructure of human ovarian primordial follicles after combination chemotherapy for Hodgkin's disease. Hum Reprod 1993;8:2080-7.
26. Warne GL, Fairley KF, Hobbs JB, Martin FI. Cyclophosphamide induced ovarian failure. N Engl J Med 1973;289:1159-62.
27. Koyama H, Wada T, Nishizawa Y, Iwanaga T, Aoki Y. Cyclophosphamide-induced ovarian failure and its therapeutic significance in patients with breast cancer. Cancer 1977;39:1403-9.
28. Fisher B, Sherman B, Rockette H, Redmond C, Margolese R, Fisher ER. 1-phenylalanine mustard (L-PAM) in the management of premenopausal patients with primary breast cancer: lack of association of disease-free survival with depression of ovarian function. National Surgical Adjuvant Project for Breast and Bowel Cancers. Cancer 1979;44:847-57.
29. Viviani S, Santoro A, Ragni G, Bonfante V, Bestetti O, Bonadonna G. Gonadal toxicity after combination chemotherapy for Hodgkin's disease. Comparative results of MOPP vs ABVD. Eur J Cancer Clin Oncol 1985;21:601-5.
30. Mackie EJ, Radford M, Shalet SM. Gonadal function following chemotherapy for childhood Hodgkin's disease. Med Pediatr Oncol 1996;27:74-8.
31. Oktay K, Libertella N, Oktem O. The impact of paclitaxel on menstrual function. Breast Cancer Res Treat 94 2005;(Suppl1):271s-2s.
32. Oktay K, Oktem O. Fertility preservation medicine: a new field in the care of young cancer survivors. Pediatr Blood Cancer 2009;53:267-73.
33. Meirow D, Lewis H, Nugent D, Epstein M. Subclinical depletion of primordial follicular reserve in mice treated with cyclophosphamide: clinical importance and proposed accurate investigative tool. Hum Reprod 1999;14:1903-7.
34. Byrne J, Fears TR, Gail MH, Pee D, Connelly RR, Austin DF, Holmes GF, Holmes FF, Latourette HB, Meigs JW, Strong LC, Myers MH, Mulvihill JJ. Early menopause in long-term survivors of cancer during adolescence. Am J Obstet Gynecol 1992;166:788-93.
35. Meirow D, Nugent D. The effects of radiotherapy and chemotherapy on female reproduction. Hum Reprod Update 2001;7:535-43.
36. Chemaitilly W, Mertens AC, Mitby P, Whitton J, Stovall M, Yasui Y, Robison LL, Sklar CA. Acute ovarian failure in the childhood cancer survivor study. J Clin Endocrinol Metab 2006;91:1723-8.
37. Brachet C, Heinrichs C, Tenoutasse S, Devalck C, Azzi N, Ferster A. Children with sickle cell disease: growth and gonadal function after hematopoietic stem cell transplantation. J Pediatr Hematol Oncol 2007;29:445-50.
38. Sklar CA, Mertens AC, Mitby P, Whitton J, Stovall M, Kasper C, Mulder J, Green D, Nicholson HS, Yasui Y, Robison LL. Premature menopause in survivors of childhood cancer: a report from the childhood cancer survivor study. J Natl Cancer Inst 2006; 98:890-6.
39. Rahhal SN, Eugster EA. Unexpected recovery of ovarian function many years after bone marrow transplantation. J Pediatr 2008;152:289-90.
40. Oktay K, Sönmezer M. Chemotherapy and amenorrhea: risks and treatment options. Curr Opin Obstet Gynecol 2008;20:408-15.
41. Kwee J, Schats R, McDonnell J, Themmen A, de Jong F, Lambalk C. Evaluation of anti-Mullerian hormone as a test for the prediction of ovarian reserve. Fertil Steril 2008;90:737-43.
42. Anderson RA, Themmen AP, Al-Qahtani A, Groome NP, Cameron DA. The effects of chemotherapy and long-term gonadotrophin suppression on the ovarian reserve in premenopausal women with breast cancer. Hum Reprod 2006;21:2583-92.
43. Schmidt KLT, Andersen CY, Loft A, Byskov AG, Ernst E, Andersen AN. Follow-up of ovarian function postchemotherapy following ovarian cryopreservation and transplantation. Hum Reprod 2005;20:3539-46.
44. van Beek RD, van den Heuvel-Eibrink MM, Laven JS. Anti-Mullerian hormone is a sensitive serum marker for gonadal function in women treated for Hodgkin's lymphoma during childhood. J Clin Endocrinol Metab 2007;92:3869-74.
45. Oktay K, Oktem O, Reh A, Vahdat L. Measuring the impact of chemotherapy on fertility in women with breast cancer. J Clin Oncol 2006;24:4044-6.
46. Reh A, Oktem O, Oktay K. Impact of breast cancer chemotherapy on ovarian reserve: a prospective observational analysis by menstrual history and ovarian reserve markers. Fertil Steril 2007;Dec 29 [Epub ahead of print].
47. Giuseppe L, Attilio G, Edoardo DN, Loredana G, Cristina L, Vincenzo L. Ovarian function after cancer treatment in young women affected by Hodgkin disease (HD). Hematology 2007;12:141-7.
48. Lie Fong S, Lugtenburg PJ, Schipper I, Themmen AP, de Jong FH, Sonneveld P, Laven JS. Antimullerian hormone as a marker of ovarian function in women after chemotherapy and radiotherapy for haematological malignancies. Hum Reprod 2008;23:674-8.
49. Gosden RG, Wade JC, Fraser HM, Sandow J, Faddy MJ. Impact of congenital or experimental hypogonadotrophism on the radiation sensitivity of the mouse ovary. Hum Reprod 1997;12:2483-8.
50. Wallace WH, Shalet SM, Hendry JH, Morris-Jones PH, Gattamaneni HR. Ovarian failure following abdominal irradiation in childhood: the radiosensitivity of the human oocyte. Br J Radiol 1989;62:995-8.
51. Wallace WH, Thomson AB, Kelsey TW. The radiosensitivity of the human oocyte. Hum Reprod 2003;18:117-21.
52. Fisher B, Cheung AY. Delayed effect of radiation therapy with or without chemotherapy on ovarian function in women with Hodgkin's disease. Acta Radiol Oncol 1984;23:43-8.
53. Lushbaugh CC, Casarett GW. The effects of gonadal irradiation in clinical radiation therapy: a review. Cancer 1976;37:1111-25.
54. Tease C, Fisher G. The influence of maternal age on radiation-induced chromosome aberrations in mouse oocytes. Mutat Res 1991;262:57-62.
55. Morice P, Juncker L, Rey A, El-Hassan J, Haie-Meder C, Castaigne D. Ovarian transposition for patients with cervical carcinoma treated by radiosurgical combination. Fertil Steril 2000;74:743-8.
56. Meirow D, Nugent D. The effects of radiotherapy and chemotherapy on female reproduction. Hum Reprod Update 2001;7:535-43.
57. Stillman RJ, Schinfeld JS, Schiff I, Gelber RD, Greenberger J, Larson M, Jaffe N, Li FP. Ovarian failure in long-term survivors of childhood malignancy. Am J Obstet Gynecol 1981;139:62-6.

58. Wallace WH, Shalet SM, Crowne EC, Morris-Jones PH, Gattamaneni HR. Ovarian failure following abdominal irradiation in childhood: natural history and prognosis. Clin Oncol (R Coll Radiol) 1989;1:75-9.
59. Sanders JE, Buckner CD, Amos D, Levy W, Appelbaum FR, Doney K, Storb R, Sullivan KM, Witherspoon RP, Thomas ED. Ovarian function following marrow transplantation for aplastic anemia or leukemia. J Clin Oncol 1988;6:813-8.
60. Sönmezer M, Oktay K. Assisted reproduction and fertility preservation techniques in cancer patients. Curr Opin Endocrinol Diabetes Obes 2008;15:514-22.
61. Nussbaum Blask AR, Nicholson HS, Markle BM, Wechsler-Jentzch K, O'Donnell R, Byrne J. Sonographic detection of uterine and ovarian abnormalities in female survivors of Wilms' tumor treated with radiotherapy. AJR Am J Roentgenol 1999;172:759-63.
62. Arndt CA, Donaldson SS, Anderson JR, Andrassy RJ, Laurie F, Link MP, Raney RB, Maurer HM, Crist WM. What constitutes optimal therapy for patients with rhabdomyosarcoma of the female genital tract? Cancer 2001;91:2454-68.
63. Franchi-Rezgui P, Rousselot P, Espie M, Briere J, Pierre Marolleau J, Gisselbrecht C, Brice P. Fertility in young women after chemotherapy with alkylating agents for Hodgkin and non-Hodgkin lymphomas. Hematol J 2003;4:116-20.
64. Ozaki T, Flege S, Kevric M, Lindner N, Maas R, Delling G, Schwarz R, von Hochstetter AR, Salzer-Kuntschik M, Berdel WE, Jürgens H, Exner GU, Reichardt P, Mayer-Steinacker R, Ewerbeck V, Kotz R, Winkelmann W, Bielack SS. Osteosarcoma of the pelvis: experience of the Cooperative Osteosarcoma Study Group. J Clin Oncol 2003;21:334-41.
65. Rodl RW, Hoffmann C, Gosheger G, Leidinger B, Jurgens H, Winkelmann W. Ewing's sarcoma of the pelvis: combined surgery and radiotherapy treatment. J Surg Oncol 2003;83:154-60.
66. Jemal A, Murray T, Samuels A, Ghafoor A, Ward E, Thun MJ. Cancer statistics, 2003. CA Cancer J Clin 2003;53:5-26.
67. Brenner H, Kaatsch P, Burkhardt-Hammer T, Harms DO, Schrappe M, Michaelis J. Long-term survival of children with leukemia achieved by the end of the second millennium. Cancer 2001;2:1977-83.
68. Pui CH, Cheng C, Leung W, Rai SN, Rivera GK, Sandlund JT, Ribeiro RC, Relling MV, Kun LE, Evans WE, Hudson MM. Extended follow-up of long-term survivors of childhood acute lymphoblastic leukemia. N Engl J Med 2003;349:640-9.
69. Robison LL, Bhatia S. Late-effects among survivors of leukaemia and lymphoma during childhood and adolescence. Br J Haematol 2003;122:345-59.
70. Oktay KH, Yih M. Preliminary experience with orthotopic and heterotopic transplantation of ovarian cortical strips. Semin Reprod Med 2002;20:63-74.
71. Waggoner SE. Cervical cancer. Lancet 2003;361:2217-25.
72. Kauff ND, Satagopan JM, Robson ME, Scheuer L, Hensley M, Hudis CA, Ellis NA, Boyd J, Borgen PI, Barakat RR, Norton L, Castiel M, Nafa K, Offit K. Risk-reducing salpingo–oophorectomy in women with a BRCA1 or BRCA2 mutation. N Engl J Med 2002;346:1609-15.
73. Rebbeck TR, Lynch HT, Neuhausen SL, Narod SA, Van't Veer L, Garber JE, Evans G, Isaacs C, Daly MB, Matloff E, Olopade OI, Weber BL; Prevention and Observation of Surgical End Points Study Group. Prophylactic oophorectomy in carriers of BRCA1 or BRCA2 mutations. N Engl J Med 2002;346:1616-22.
74. Russell AI, Lawson WA, Haskard DO. Potential new therapeutic options in Behcet's syndrome. BioDrugs 2001;15:25-35.
75. Langford CA, Talar-Williams C, Barron KS, Sneller MC. Use of a cyclophosphamide-induction methotrexate-maintenance regimen for the treatment of Wegener's granulomatosis: extended follow-up and rate of relapse. Am J Med 2003;114:463-9.
76. Nousari CH, Brodsky R, Anhalt GJ. Evaluating the role of immunoablative high-dose cyclophosphamide therapy in pemphigus vulgaris. J Am Acad Dermatol 2003;49:148-50.
77. Stallmach A, Wittig BM, Moser C, Fischinger J, Duchmann R, Zeitz M. Safety and efficacy of intravenous pulse cyclophosphamide in acute steroid refractory inflammatory bowel disease. Gut 2003;52:377-82.
78. Katsifis GE, Tzioufas AG. Ovarian failure in systemic lupus erythematosus patients treated with pulsed intravenous cyclophosphamide. Lupus 2004;13:673-8.
79. Picton HM, Harris SE, Muruvi W, Chambers EL. The in vitro growth and maturation of follicles. Reproduction 2008;136:703-15.
80. Dolmans MM, Yuan WY, Camboni A, Torre A, Van Langendonckt A, Martinez-Madrid B, Donnez J. Development of antral follicles after xenografting of isolated small human preantral follicles. Reprod Biomed Online 2008;16:705-11.
81. Lan C, Xiao W, Xiao-Hui D, Chun-Yan H, Hong-Ling Y. Tissue culture before transplantation of frozen-thawed human fetal ovarian tissue into immunodeficient mice. Fertil Steril. 2008;22. [Epub ahead of print].
82. Kaya H, Desdicioglu R, Sezik M, Ulukaya E, Ozkaya O, Yilmaztepe A, Demirci M. Does sphingosine-1-phosphate have a protective effect on cyclophosphamide- and irradiation-induced ovarian damage in the rat model? Fertil Steril. 2008;89:732-5.
83. Silber SJ, Lenahan KM, Levine DJ, Pineda JA, Gorman KS, Friez MJ, Crawford EC, Gosden RG. Ovarian transplantation between monozygotic twins discordant for premature ovarian failure. N Engl J Med. 2005;353:58-63.