Pharmacological options for the protection of ovarian function in patients undergoing chemotherapy

Botha MH, MBChB, MMed(O&G), FCOG(SA), PhD
Associate Professor and Head, Unit for Gynaecological Oncology, Department of Obstetrics and Gynaecology
Faculty of Medicine and Health Sciences, University of Stellenbosch, Tygerberg
Correspondence to: Matthys Botha, e-mail: mhbotha@sun.ac.za
Keywords: protection of ovarian function, pharmacological options, chemotherapy, radiotherapy

Abstract

Chemotherapy, particularly alkylating agents, can be toxic to germ cells, and may lead to treatment amenorrhoea in young women. The age at which chemotherapy is administered is a strong predictor of subsequent premature ovarian failure, with older patients being at the highest risk. Smaller primordial follicles may survive the insult of chemotherapy better, and the suppression of follicle development may protect the germ cell pool. The suppression of follicle development by gonadotrophin-releasing hormone agonists (or antagonists) or combined oral contraceptives has been reported in the literature. The results are promising, but conflicting reports on the protection makes further studies essential.

℗ Peer reviewed. (Submitted: 20112-04. Accepted: 2015-02-17.) © SASGO South Afr J Gynaecol Oncol 2015;7(1):27-33

Introduction

Over the last few decades, major improvements have been made in the treatment of childhood and adolescent malignancies. However, treatment modalities may have serious harmful effects on ovarian function. Also, the incidence of premature ovarian failure (POF) in patients who have received chemotherapy and/or radiotherapy is much increased when compared to the general population.[1,2] The late effects of cancer treatment have been a popular topic for discussion in the medical literature, and various attempts has been made to protect young women against the potential harm of chemotherapy and radiotherapy.

Freezing germ cells has improved significantly in the last two decades. Oocyte cryopreservation before oncotherapy may offer hope, although the success of this technique is often limited by the number of available oocytes after stimulation, and the availability of time for ovarian stimulation.[3] Embryo freezing may be another option in women with a male partner at the time.

Ovarian tissue cryopreservation may be the only option for some young women before they undergo aggressive chemotherapy or a combination of chemotherapy and radiotherapy.[4] This technique is still regarded by many experts as experimental, but successful pregnancies have been reported from thawed, previously cryopreserved, ovarian tissue.[5]

This review will focus on hormonal manipulation during chemotherapy.

Age at treatment

The age of the patient at the time of treatment with chemotherapy remains one of the most important determinants of the incidence of ovarian failure. The risk increases with age of exposure, and increasing age is as an important predictive factor for chemotherapy-induced amenorrhoea.[6-8]

Many theories have been put forward to explain the age phenomenon. A popular theory is that the absolute number of primordial follicles decreases with age, as demonstrated by the Faddy-Gosden differential equation.[9-11] It is argued that a constant fraction of the primordial follicles is damaged during a particular cycle of chemotherapy or radiotherapy. Therefore, the remaining number is directly proportional to the initial population. The initial pretreatment population of

oocytes decreases at a logarithmic rate with increasing age. The total number of oocytes in the ovary is a combination of primordial, small primary and intermediary follicles.[12]

Prepubescent girls are less likely to develop POF after chemotherapy than older patients. This finding has led to the assumption that the creation of an artificial prepubertal environment might lead to ovarian protection. This argument has not been conclusively shown in experimental models. The reason why younger girls may have less obvious functional damage is likely to be owing to the fact that the total number of available follicles is much higher at a younger age. This argument is supported by the long-term follow-up of young girls after receiving chemotherapy, who eventually developed premature menopause.[13] The relative risk of early menopause in the third decade was 9.2 after treatment with alkylating agents in a large post-treatment follow-up study.[14]

Gonadotrophin-releasing hormone agonists

Almost 30 years ago, Glode, Robinson and Gould tested the possibility of testicular protection against cyclophosphamide-induced damage in male mice, and found a protective effect of gonadotrophin-releasing hormone (GnRH) agonists.[15]Ataya demonstrated that GnRH agonists may protect against chemotherapy-induced ovarian compromise in a murine model as early as 1985. Other animal studies seemed to confirm this work.[16-19] However, conflicting results from Letterie did not show an improvement in outcome.[20] Mice treated with cyclophosphamide also received combined oestrogen and progesterone or leuprolide in this study. Neither method of ovulation suppression protected the mice against gonadal toxicity. Despite the promising work in rats where GnRH agonists inhibited chemotherapy-induced ovarian damage, many authors remain sceptical of its application in humans.[21,22]

It was demonstrated in further animal studies on rhesus macaque monkeys (Macaca mulatta) treated with GnRH agonists during chemotherapy that more primordial follicles remained after treatment in those receiving a GnRH agonist.[23]

The possible protective mechanisms postulated were:

- Suppression of the levels of gonadotrophins in the ovary.
- A direct influence of the GnRH agonists mediated through receptors in the ovary.
- A reduction in the metabolism of the ovary, resulting in a decrease in the blood flow to the ovary.

Oral contraceptives also may reduce gonadotrophic levels in the follicular environment, and therefore also create a similar environment for protection.[24]

A single oocyte within a dominant follicle must develop for ovulation to occur during a particular menstrual cycle. This whole process starts when a group of primordial follicles starts to develop. A single follicle is selected from this group to eventually effect ovulation. Growing follicles make up less than 10% of the total number of follicles, but after initiation of the growth process, either ovulation or atresia is inevitable.[25] The initial growth is independent of gonadotrophins.[12] It is not a biologically plausible argument that GnRH agonists work as a protective mechanism by keeping follicles outside of the growth phase because the initial process is gonadotrophin independent. It was found in some interesting work that GnRH agonist receptors are absent in the preovulatory follicle and the *corpus luteum* in human ovarian cells. Therefore, GnRH agonist stimulation is not possible in the primordial and early follicular stages of development.[26,27]

A protective effect of Buserelin was demonstrated in an in vitro study on cultured human granulosa cells exposed to doxyrubicine chemotherapy.[28] This study provided support for the protective action of GnRH agonists on stromal cell damage. Doxorubicin caused irreversible decreases in oestrogen production, but the addition of GnRH agonists before or during treatment protected the granulosa cells.

The cortex of the ovary is a relatively avascular area, and GnRH agonist downregulation owing to a lack of pituitary stimulation may further reduce blood flow in the ovary. Ultrasound studies have confirmed reduced perfusion during diminished pituitary function.[29,30] By reducing blood flow, the exposure of follicles to harmful chemotherapy may be decreased. However, there is conflicting evidence that ovarian perfusion may not significantly change after treatment with GnRH agonists, when measured by a Doppler study. Significant changes in antral follicle count, stromal blood flow or ovarian volume were not demonstrated in a three-dimensional Doppler study on 85 women treated with short-term Buserelin.[31]

GnRH agonists may upregulate the production of antiatoptotic molecules, such as sphingosine 1-phosphate.[32] They may also affect molecules, such as transforming growth factor beta (TGF-β).[33] Other growth factors in the TGF-β superfamily may also be influenced by GnRH analogues. However, the complex interaction of all these growth factors is difficult to measure and control in an experimental model. Some work suggests that the transition from the primordial to the primary follicle may be influenced by ligands produced by growing follicles.[34] Through this secondary pathway, downregulation of the growing follicles by an absence of follicle-stimulating hormone (FSH) stimulation may lead to a reduced number of follicles being recruited.

Table I: The incidence of ovarian failure in different studies with and without gonadotrophin-releasing hormone agonist administration

References	GnRH agonist (%)	Control (%)	Follow-up (years)
Waxman et al[35]	50.0	67.0	2.3
Blumenfeld et al[36]	0.0	66.0	-
Pereyra Pacheco et al[37]	0.0	100.0	< 5.0
Blumenfeld and Eckman[38]	7.0	54.0	
Franke, Smit and Vermes[39]	20.0	-	< 1.0
Dann et al[40]	0.0	17.0	5.3
Somers et al[41]	5.0	30.0	-
Elis et al[42]	0.0	8.7	-
Del Mastro et al[43]	3.0	-	1.0
Recchia et al[44]	33.0		6.3
Giuseppe et al[45]	0.0	47.0	2.4
Castelo-Branco et al[46]	7.0	77.0	-
Blumenfeld et al[24]	3.0	37.0	8.0
Huser et al[47]	21.0	71.0	1.0
Badawy, Elnashar, El-Ashry, Shahat[48]	11.0	67.0	< 1.0
Gerber et al[49]	0.0	0.0	2.0

GnRH: gonadotrophin-releasing hormone

Clinical studies on ovarian suppression and protection against chemotherapy-induced ovarian damage

A review published in 2008 on the topic of ovulation suppression to protect against chemotherapy-induced ovarian toxicity and amenorrhoea concluded that the currently available information in the literature is not enough for a definite conclusion to be drawn that GnRH agonists are beneficial in protecting against chemotherapy-induced damage.[33] The authors argue that on the basis of the current evidence, it is possible to draw the wrong conclusions. There are many drawbacks to the published studies, which are summarised in Table I. Very often, the studies were not controlled or historical controls were used. There were many treatment protocols, many different indications for chemotherapy, and the definition of ovarian function was not always the same. Various parameters were included, like the presence or absence of menstruation, hormone levels, the ultrasound appearance of the ovaries, and even cycle regularity. A clear indication of what is meant by "preserved ovarian function" and "ovarian failure" is not given in most of the published data.

There were unexpectedly high incidences of POF in the control group in two larger studies by Castelo-Branco et al and Huser et al.[46,47] The expected rate of POF in the population not treated with GnRH agonists is usually in the region of 32-37%, as found in other studies on Hodgkin's lymphoma survivors.[50,51] The high POF rate in the control group in the two larger studies influences any meta-analysis currently performed.

A slightly more positive conclusion was recorded in another review published in the same journal.[52] However, the authors also concluded their review by stating that there was not enough convincing statistical evidence concerning the reduction of POF when treating young women with GnRH agonists. More large prospective randomised studies are needed.

Thirty-nine patients received GnRH and 39 chemotherapy only in a prospective randomised study on 80 patients with breast cancer.[48] The rate of POF was significantly lower in the treatment arm of the study. This study used goserelin for ovarian suppression, while previous work with Buserelin was not beneficial. It is argues that Buserelin may perhaps not adequately suppress pituitary function, and this may explain the difference in outcome.

A study from Italy evaluated ovarian reserve in 29 patients treated for Hodgkin's disease.[45] The serum levels of FSH, luteinising hormone (LH), inhibin B, anti-Mullerian hormone (AMH) and the ultrasound antral follicular count were used as markers of ovarian reserve after chemotherapy. The patients were randomly given or not given GnRH agonists at the time of chemotherapy. It is not clear from the article which specific analogue was used. None of the 14 women who received an GnRH agonist developed amenorrhoea. Seven of the 15 (46%) who did not receive a GnRH agonist showed the clinical symptoms of ovarian failure, "suggesting the possible protective action of GnRH agonists". However, when the results of the ovarian reserve parameters were compared, those treated with a GnRH agonist had similar results.

A different Italian group treated 61 women with triptorelin monthly during chemotherapy for patients with Hodgkin's lymphoma.[53] The majority (81%) recovered normal menses, while 6% had irregular cycles. The remaining 12% had premature ovarian failure. The authors felt confident enough about the results to recommend that all young women should receive GnRH agonist co-treatment from the onset of chemotherapy.

A report from Korea investigated the incidence of chemotherapy-related amenorrhoea in premenopausal women with breast cancer.[54] Two hundred and thirty-eight women out of a total of 326 received adjuvant endocrine therapy for oestrogen and progesterone receptor positivity. The adjuvant endocrine therapy included selective ooestrogen-receptor modulators (toremifene citrate and tamoxifen), aromatase inhibitors (letrozole and anastrozle),

and LH-releasing hormone. The two important predictors for amenorrhoea were older age at treatment and the use of endocrine treatment. Interestingly "the use of adjuvant endocrine therapy was more likely to result in permanent chemotherapy-related amenorrhoea, than no use" (p-value 0.006).

The German Hodgkin Study Group prematurely stopped a prospective study on hormonal treatment with combined oral contraception or GnRH owing to a lack of effectiveness. The AMH levels after 12 months showed no benefit of GnRH or combined oral contraception.[55]

A meta-analysis of nine studies that included data on 366 women found a 68% increase in the rate of preserved ovarian function in those women receiving a GnRH agonist.[56] Similarly, a Cochrane review came to the conclusion that intramuscular or subcutaneous GnRH agonists seemed "to be effective in protecting ovaries during chemotherapy." [57]

Potential problems with gonadotrophin-releasing hormone agonist treatment

Treatment with GnRH agonists is not without complications. The side-effects include postmenopausal symptoms, such as hot flushes and decreased bone mineral density. It is also of importance to note that GnRH receptors have been found in certain tumour cell lines, and the effects that GnRH agonists may have on tumour progression have not been studied carefully.[21,58]

Additional potential benefits of gonadotrophin-releasing hormone treatment

GnRH agonists may be beneficial in patients with early receptor-positive breast carcinoma, where the suppression of oestrogen production could lead to a better outcome. It was found in a recent Cochrane review on GnRH agonists in an adjuvant therapy setting for early breast cancer in premenopausal women that the therapy may be of clinical benefit. The authors concluded that the current data strongly support the continuation of further trials on LH-releasing hormone agonists in early breast cancer treatment.[59]

Other potential benefits of GnRH agonists unrelated to ovarian function include a reduction in menorrhagia during chemotherapy, especially at the time when myelosuppression may be an important complication of chemotherapy. A systematic review found that GnRH agonist therapy is highly effective in preventing excessive uterine bleeding in the treatment of haematological malignancies.[60]

Hormonal contraceptives

The suppression of follicle development during the administration of chemotherapy may reduce the risk of follicular damage. A group of 31 young women who received neoadjuvant chemotherapy for the treatment of osteosarcoma were observed for chemotherapy-induced menopause.[61] All of these patients received high-dose ifosfamide, methrotrexate, adriamycin and cisplatinum, and were treated with oral contraceptives for the duration of chemotherapy.

Three of the 19 patients in the treatment group developed early menopause. They were compared with a historical control group, in which three of the 71 patients developed chemotherapy-induced menopause. The authors concluded that oral contraceptives during chemotherapy do not protect ovarian function in patients receiving high-dose alkylating chemotherapy, and that the age at treatment and the total dose of alkylating agents are the most important predictors of ovarian failure.[61]

An earlier retrospective report on the reproductive outcome of women treated for Hodgkin's disease included a total of 44 women followed-up after chemotherapy [mustine, vinblastine, procarbazine and prednisolone (MVPP)].[6] Nine of these patients took combined oral contraceptives throughout the administration of chemotherapy, of whom four subsequently developed amenorrhoea, and three more oligomenorrhoea. It appears as if oral contraceptives did not protect these patients against chemotherapy-induced ovarian damage.

A report published in 1981 evaluated the use of oral contraceptives in six young women treated with MVPP for Hodgkin's disease.[62] They were followed-up between four and 12 months, and underwent post-therapy ovarian biopsies and menstrual history to determine their fertility potential. Five of the six patients had normal menstruation after discontinuation of the chemotherapy. The three cases who underwent ovarian biopsies were aged 18, 19 and 28 years, respectively. The 19-year-old woman had more than 1 000 follicles per section after chemotherapy. It is difficult to know whether or not a histological evaluation of follicle numbers can predict eventual menstrual function, but despite the small numbers in this study, the authors concluded "that the suppression of ovarian function by combined oral contraceptives protects the ova against otherwise certain injury by the chemotherapeutic drugs".[62]

A group from Israel reported on the post-treatment fertility status of women after receiving aggressive treatment for non-Hodgkin's lymphoma. It was found that age was the most important risk factor. Older women were at the highest risk of chemotherapy-related amenorrhoea.[42] Fertility-preserving measures, in the form of GnRH agonists, were used in three patients and nine received combined oral contraceptives. When the patients who received ovarian suppression were compared to the rest of the group, a significant difference in the rate of ovarian failure was not found.

The best evidence, so far, for the potential benefit of oral contraceptives during chemotherapy derives from a large retrospective report from Germany.[63] A total of 405 women answered a questionnaire on their menstrual status after therapy for Hodgkin's lymphoma. These patients were all aged 40 years and younger at treatment. The rate of amenorrhoea was significantly higher in women not taking oral contraceptives, compared to that in the women on oral contraceptives during chemotherapy (44.1% versus 10.1%) (p-value < 0.0001). A multivariate analysis was performed, including age, chemotherapy regime, stage of disease, the use of oral contraceptives during chemotherapy and the effect on amenorrhoea.

There was enough information on 214 women, from a total of 405, for them to be included in the multivariate analysis, and the following were found to be significant predictors of amenorrhoea:

- Receiving eight cycles of dose-escalated bleomycin, etoposide, adriamycin (doxorubicin), cyclophosphamide, oncovin (vincristine), procarbazine and prednisone (BEACOPP), when compared to other less toxic regimes.
- Amenorrhoea was statistically significantly higher in women with advanced stage cancer (p-value < 0.0001).
- Being older than 30 years of age at treatment (p-value 0.0065).
- Not taking oral contraceptives during chemotherapy (p-value 0.0002).

This study provides the strongest evidence so far for the potential positive effect of oral contraceptives during chemotherapy.

However, oral contraceptives have never been adequately tested in a randomised controlled trial.[64] A randomised phase II trial performed by the German Hodgkin Study Group, the PROtecting Ovaries and Fertility During Chemotherapy (PROOF), that evaluated the use of GnRH agonists and oral contraceptives in patients receiving treatment for Hodgkin's lymphoma, was stopped early after only 23 patients enrolled. The reason for the early termination of the study was mainly because most women had already received GnRH analogues, as recommended by their gynaecologists. Twelve patients received oral contraceptives and 11 GnRH analogues. The respective infertility rates were 90% for contraceptives and 100% with the GnRH agonists. The conclusion of the authors was that treatment with GnRH agonists or oral contraceptives conferred no meaningful ovarian protection.

Medroxiprogesterone acetate

The only report on the effect of medroxyprogesterone acetate on ovarian protection during chemotherapy derives from a small descriptive study from Italy.[65] Twelve women were included, of whom four received no chemotherapy or MPA, four received no chemotherapy and 250 mg MPA, and the last four received chemotherapy and 250 mg MPA per month. Ovarian biopsies were taken, and electron microscopy studies performed. Despite the very small number of cases, this did not prevent the authors from stating that "the results presented here demonstrate that the administration of MPA to patients with Hodgkin's disease protects the ovary against an acute effect of chemotherapy". It is not clear from the discussion how the authors concluded that there was a protective effect because the studied follicles of patients receiving chemotherapy and MPA had substantial morphological damage. At present, there is no strong argument in the literature to suggest a protective effect of MPA against chemotherapy-induced amenorrhoea.

Conclusion

It is very difficult to reach a conclusion on the use of ovarian suppression during chemotherapy in an attempt to prevent subsequent chemotherapy-induced ovarian failure. Most of the available evidence in the literature derives from non-randomised case reports, often with historical controls or no controls. Various chemotherapy regimens were used, the doses of GnRH agonists and combined oral contraceptives were not similar, and in general, the number of patients was low. Also, a randomised controlled trial has not been conducted to evaluate combined oral contraceptives in this clinical scenario. The authors of a Cochrane review were cautious when concluding that GnRH analogues seemed "to be effective", but recommended that they should be given before or during treatment.[57]

The only way to reach consensus would be to test combined oral contraceptives and GnRH agonists in properly designed, large, prospective randomised trials. The numbers of young patients with cancer are usually low in individual institutions, and it is important to combine data from multiple sites to obtain useful results.

At present, the indication for oral contraceptives or GnRH agonists may be stronger for other non-fertility benefits. GnRH agonists and oral contraceptives may be used to reduce menstrual blood loss, which is often increased during treatment because of abnormal clotting and platelet function. When hormones are contraindicated owing to hormone-sensitive tumours, GnRH agonists can be very useful. There is some evidence that GnRH agonist administration may improve the outcome in oestrogen receptor-positive breast carcinoma.

References

1. Lo Presti A, Ruvolo G, Gancitano RA, Cittandi E. Ovarian function following radiation and chemotherapy for cancer. Eur J Obstet Gynecol Reprod Biol. 2004;113 Suppl 1:S33-S40.
2. Chiarelli AM, Marrett LD, Darlington G. Early menopause and infertility in females after treatment for childhood cancer

diagnosed in 1964-1988 in Ontario, Canada. Am J Epidemiol. 1999;150(3):245-254.
3. Martinez M, Rabadan S, Dominigo J, et al. Obstetric outcome after oocyte vitrification and warming for fertility preservation in women with cancer. Reprod Biomed Online. 2014;29(6):722-728.
4. Practice Committee of American Society for Reproductive Medicine. Ovarian tissue cryopreservation: a committee opinion. Fertil Steril. 2014;101(5):1237-1243.
5. Macklon KT, Jensen AK, Loft A, et al. Treatment history and outcome of 24 deliveries worldwide after autotransplantation of cryopreserved ovarian tissue, including two new Danish deliveries years after autotransplantation. J Assist Reprod Genet. 2014;31(11):1557-1564.
6. Whitehead E, Shalet SM, Blackledge G, et al. The effect of combination chemotherapy on ovarian function in women treated for Hodgkin's disease. Cancer. 1983;52(6):988-993.
7. Petrek JA, Naughton MA, Case LD, et al. Incidence, time course, and determinants of menstrual bleeding after breast cancer treatment: a prospective study. J Clin Oncol. 2006;24(7):1045-1051.
8. Goldhirsch A, Gelber RD, Castiglione M. The magnitude of endocrine effects of adjuvant chemotherapy for premenopausal breast cancer patients. The International Breast Cancer Study Group. Ann Oncol. 1990;1(3):183-188.
9. Brougham MF, Wallace WH. Subfertility in children and young people treated for solid and haematological malignancies. Br J Haematol. 2005;131(2):143-155.
10. Wallace WH, Thomson AB, Kelsey TW. The radiosensitivity of the human oocyte. Hum Reprod. 2003;18(1):117-121.
11. Wallace WH, Kelsey TW. Ovarian reserve and reproductive age may be determined from measurement of ovarian volume by transvaginal sonography. Hum Reprod. 2004;19(7):1612-1617.
12. Gougeon A. Regulation of ovarian follicular development in primates: facts and hypotheses. Endocr Rev. 1996;17(2):121-55.
13. Sklar CA, Merlens AC, Mitty P, et al. Premature menopause in survivors of childhood cancer: a report from the childhood cancer survivor study. J Natl Cancer Inst. 2006;98(13):890-896.
14. Byrne J, Fears TJ, Gail MH, et al. Early menopause in long-term survivors of cancer during adolescence. Am J Obstet Gynecol. 1992;166(3):788-793.
15. Glode LM, Robinson J, Gould SF. Protection from cyclophosphamide-induced testicular damage with an analogue of gonadotropin-releasing hormone. Lancet. 1981;1(8230):1132-1134.
16. Ataya KM, McKanna JA, Weintraub AB, et al. A luteinizing hormone-releasing hormone agonist for the prevention of chemotherapy-induced ovarian follicular loss in rats. Cancer Res. 1985;45(8):3651-3656.
17. Ataya K, Moghissi K. Chemotherapy-induced premature ovarian failure: mechanisms and prevention. Steroids. 1989;54(6): 607-626.
18. Ataya K, Ramahi-Ataya A. Reproductive performance of female rats treated with cyclophosphamide and/or LHRH agonist. Reprod Toxicol. 1993;7(3):229-235.
19. Bokser L, Szende B, Schally AV. Protective effects of D-Trp6-luteinising hormone-releasing hormone microcapsules against cyclophosphamide-induced gonadotoxicity in female rats. Br J Cancer. 1990;61(6):861-865.
20. Letterie GS. Anovulation in the prevention of cytotoxic-induced follicular attrition and ovarian failure. Hum Reprod. 2004;19(4): 831-837.
21. Bohlmann MK, von Wolff M, Strowitzki T. Comment on the symposium article "Fertility after treatment for Hodgkin's disease", by Z. Blumenfeld, E. Dann, I. Avivi et al. (Ann Oncol 2002;13 Suppl 1:138-147). Ann Oncol. 2003;14(3):499.
22. Oktay K, Sönmezer M, Oktem O, et al. Absence of conclusive evidence for the safety and efficacy of gonadotropin-releasing hormone analogue treatment in protecting against chemotherapy-induced gonadal injury. Oncologist. 2007;12(9):1055-1066.
23. Ataya K, Rao LV, Lawrence E, Kimmel R. Luteinizing hormone-releasing hormone agonist inhibits cyclophosphamide-induced ovarian follicular depletion in rhesus monkeys. Biol Reprod. 1995;52(2):365-372.
24. Blumenfeld Z, Avivi I, Eckman A, et al. Gonadotropin-releasing hormone agonist decreases chemotherapy-induced gonadotoxicity and premature ovarian failure in young female patients with Hodgkin lymphoma. Fertil Steril. 2008;89(1):166-173.
25. Sonmezer M, Oktay K. Fertility preservation in female patients. Hum Reprod Update. 2004;10(3):251-266.
26. Birnbaumer L, Shahabi N, Rivier J, Vale W. Evidence for a physiological role of gonadotropin-releasing hormone (GnRH) or GnRH-like material in the ovary. Endocrinology. 1985;116(4):1367-1370.
27. Janssens RM, Brus L, Cahill DJ, et al. Direct ovarian effects and safety aspects of GnRH agonists and antagonists. Hum Reprod Update. 2000;6(5):505-518.
28. Imai A, Sugiyama M, Furui T, et al. Direct protection by a gonadotropin-releasing hormone analog from doxorubicin-induced granulosa cell damage. Gynecol Obstet Invest. 2007;63(2):102-106.
29. Engmann L, Sladkevicius P, Agrawal R, et al. The pattern of changes in ovarian stromal and uterine artery blood flow velocities during in vitro fertilization treatment and its relationship with outcome of the cycle. Ultrasound Obstet Gynecol. 1999;13(1):26-33.
30. Dada T, Salha O, Allgar V, Sharma V. Utero-ovarian blood flow characteristics of pituitary desensitization. Hum Reprod. 2001;16(8):1663-1670.
31. Yu Ng EH, Chi Wai Chan C, Tang OS, et al. Effect of pituitary downregulation on antral follicle count, ovarian volume and stromal blood flow measured by three-dimensional ultrasound with power Doppler prior to ovarian stimulation. Hum Reprod. 2004;19(12):2811-2815.
32. Blumenfeld Z. How to preserve fertility in young women exposed to chemotherapy? The role of GnRH agonist cotreatment in addition to cryopreservation of embrya, oocytes, or ovaries. Oncologist. 2007;12(9):1044-1054.
33. Beck-Fruchter R, Weiss A, Shalev E. GnRH agonist therapy as ovarian protectants in female patients undergoing chemotherapy: a review of the clinical data. Hum Reprod Update. 2008;14(6):553-561.
34. Knight PG, Glister C. TGF-beta superfamily members and ovarian follicle development. Reproduction. 2006;132(2):191-206.
35. Waxman JH, Ahmed R, Smith D, et al. Failure to preserve fertility in patients with Hodgkin's disease. Cancer Chemother Pharmacol. 1987;19(2):159-162.
36. Blumenfeld Z, Shapiro D, Shteinberg M, et al. Preservation of fertility and ovarian function and minimizing gonadotoxicity in young women with systemic lupus erythematosus treated by chemotherapy. Lupus. 2000;9(6):401-405.
37. Pereyra Pacheco B, Mendez-Ribaz JM, Milone G, et al. Use of GnRH analogs for functional protection of the ovary and preservation of fertility during cancer treatment in adolescents: a preliminary report. Gynecol Oncol. 2001;81(3):391-397.
38. Blumenfeld Z, Eckman E. Preservation of fertility and ovarian function and minimization of chemotherapy-induced gonadotoxicity in young women by GnRH-a. J Natl Cancer Inst Monogr. 2005(34):40-43.
39. Franke HR, Smit WM, Vermes I. Gonadal protection by a gonadotropin-releasing hormone agonist depot in young women with Hodgkin's disease undergoing chemotherapy. Gynecol Endocrinol. 2005;20(5):274-278.
40. Dann EJ, Epelbaum R, Avivi I, et al. Fertility and ovarian function are preserved in women treated with an intensified regimen of cyclophosphamide, adriamycin, vincristine and prednisone (Mega-CHOP) for non-Hodgkin lymphoma. Hum Reprod. 2005;20(8):2247-2249.

41. Somers EC, Marder W, Christman GM, et al. Use of a gonadotropin-releasing hormone analog for protection against premature ovarian failure during cyclophosphamide therapy in women with severe lupus. Arthritis Rheum. 2005;52(9):2761-2767.
42. Elis A, Tevet A, Yerushalmi R, et al. Fertility status among women treated for aggressive non-Hodgkin's lymphoma. Leuk Lymphoma. 2006;47(4): 623-627.
43. Del Mastro L, Catzeddu T, Boni I, et al. Prevention of chemotherapy-induced menopause by temporary ovarian suppression with goserelin in young, early breast cancer patients. Ann Oncol. 2006;17(1):74-78.
44. Recchia F, Saggio G, Amiconi G, et al. Gonadotropin-releasing hormone analogues added to adjuvant chemotherapy protect ovarian function and improve clinical outcomes in young women with early breast carcinoma. Cancer. 2006;106(3):514-523.
45. Giuseppe L, Attilio G, Eduordo DN, et al. Ovarian function after cancer treatment in young women affected by Hodgkin disease (HD). Hematology. 2007;12(2):141-147.
46. Castelo-Branco C, Nomdedeu B, Camus A, et al. Use of gonadotropin-releasing hormone agonists in patients with Hodgkin's disease for preservation of ovarian function and reduction of gonadotoxicity related to chemotherapy. Fertil Steril. 2007;87(3):702-705.
47. Huser M, Crha I, Ventruba P, et al. Prevention of ovarian function damage by a GnRH analogue during chemotherapy in Hodgkin lymphoma patients. Hum Reprod. 2008;23(4):863-868.
48. Badawy A, Elnashar S, El-Ashry M, Shahat M. Gonadotropin-releasing hormone agonists for prevention of chemotherapy-induced ovarian damage: prospective randomized study. Fertil Steril. 2009;91(3):694-697.
49. Gerber B, von Minkwitz G, Stehle H, et al. Effect of luteinizing hormone-releasing hormone agonist on ovarian function after modern adjuvant breast cancer chemotherapy: the GBG 37 ZORO study. J Clin Oncol. 2011;29(17):2334-2341.
50. Meirow, D., Ovarian injury and modern options to preserve fertility in female cancer patients treated with high dose radio-chemotherapy for hemato-oncological neoplasias and other cancers. Leuk Lymphoma, 1999. 33(1-2): p. 65-76.
51. Haukvik UK, et al. Treatment-related premature ovarian failure as a long-term complication after Hodgkin's lymphoma. Ann Oncol. 2006;17(9):1428-1433.
52. Blumenfeld Z, von Wolff M. GnRH-analogues and oral contraceptives for fertility preservation in women during chemotherapy. Hum Reprod Update. 2008;14(6):543-552.
53. Falorio S, Angrilli F, Fioritoni G. Gonadotropin-releasing hormone analog treatment for the prevention of treatment-related ovarian failure and infertility in women of reproductive age with Hodgkin lymphoma. Leuk Lymphoma. 2008;49(6):1087-1093.
54. Lee S, Wil WJ, Chun M, et al. Chemotherapy-related amenorrhea in premenopausal women with breast cancer. Menopause. 2009;16(1):98-103.
55. Behringer K, Wildt L, Mueller H, et al. No protection of the ovarian follicle pool with the use of GnRH-analogues or oral contraceptives in young women treated with escalated BEACOPP for advanced-stage Hodgkin lymphoma. Final results of a phase II trial from the German Hodgkin Study Group. Ann Oncol. 2010;21(10):2052-2060.
56. Clowse ME, Behera MA, Anders CK, et al. Ovarian preservation by GnRH agonists during chemotherapy: a meta-analysis. J Womens Health (Larchmt). 2009;18(3):311-319.
57. Chen H, Li J, Cui T, Hu L.. Adjuvant gonadotropin-releasing hormone analogues for the prevention of chemotherapy induced premature ovarian failure in premenopausal women. [Cochrane review]. In: The Cochrane Library, Issue 11, 2011. Oxford: Update Software.
58. Lee SJ, Schover LR, Partridge AH, et al. American Society of Clinical Oncology recommendations on fertility preservation in cancer patients. J Clin Oncol. 2006;24(18):2917-2931.
59. Goel S, Sharma R, Hamilton A, et al. LHRH agonists for adjuvant therapy of early breast cancer in premenopausal women. [Cochrane review]. In: The Cochrane Library, Issue 4, 2009. Oxford: Update Software.
60. Quaas AM, Ginsburg ES. Prevention and treatment of uterine bleeding in hematologic malignancy. Eur J Obstet Gynecol Reprod Biol. 2007;134(1):3-8.
61. Longhi A, Pignotti E, Versari M, et al. Effect of oral contraceptive on ovarian function in young females undergoing neoadjuvant chemotherapy treatment for osteosarcoma. Oncol Rep. 2003;10(1):151-155.
62. Chapman RM, Sutcliffe SB. Protection of ovarian function by oral contraceptives in women receiving chemotherapy for Hodgkin's disease. Blood. 1981;58(4): 849-851.
63. Behringer K, Breuer K, Reineke T, et al. Secondary amenorrhea after Hodgkin's lymphoma is influenced by age at treatment, stage of disease, chemotherapy regimen, and the use of oral contraceptives during therapy: a report from the German Hodgkin's Lymphoma Study Group. J Clin Oncol. 2005;23(30):7555-7564.
64. Behringer K, Wildt L, Mueller, et al. No reduction of ovarian failure with the use of GnRH-analogues or oral contraceptives in young women treated with escalated BEACOPP for Advanced-Stage Hodgkin Lymphoma (HL). Final results of the PROFE Trial, German Hodgkin Study Group (GHSG). Washington: American Society for Hematology, 2009.
65. Familiari G, Caggiati A, Nottola SA, et al. Ultrastructure of human ovarian primordial follicles after combination chemotherapy for Hodgkin's disease. Hum Reprod. 1993;8(12):2080-2087.