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Review

FERTILITY PRESERVATION IN YOUNG WOMEN WITH EARLY-STAGE BREAST CANCER

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SUMMARY – Although breast cancer (BC) occurs more often in older women, it is the most commonly diagnosed malignancy in women of childbearing age. Owing to the overall advancement of modern medicine and the growing global trend of delaying childbirth until later age, we find ever more younger women diagnosed and treated for BC who have not yet completed their family. Therefore, fertility preservation has emerged as a very important quality of life issue for young BC survivors. This paper reviews currently available options for fertility preservation in young women with early-stage BC and highlights the importance of a multidisciplinary approach to fertility preservation as a very important quality of life issue for young BC survivors. Pregnancy after BC treatment is considered not to be associated with an increased risk of BC recurrence; therefore, it should not be discouraged for those women who want to achieve pregnancy after oncologic treatment. Currently, it is recommended to delay pregnancy for at least 2 years after BC diagnosis, when the risk of recurrence is highest. However, BC patients of reproductive age should be informed about the potential negative effects of oncologic therapy on fertility, as well as on the fertility preservation options available, and if interested in fertility preservation, they should be promptly referred to a reproductive specialist. Early referral to a reproductive specialist is an important factor that increases the likelihood of successful fertility preservation. Embryo and mature oocyte cryopreservation are currently the only established fertility preservation methods but they require ovarian stimulation (OS), which delays initiation of chemotherapy for at least 2 weeks. Controlled OS does not seem to increase the risk of BC recurrence. Other fertility preservation methods (ovarian tissue cryopreservation, cryopreservation of immature oocytes and ovarian suppression with gonadotropin-releasing hormone agonists) do not require OS but are still considered to be experimental techniques for fertility preservation.

Key words: *Fertility preservation – methods; Breast neoplasms; Cryopreservation – methods; Ovulation, induction; Gonadotropin-releasing hormone – agonists; Quality of Life*

Introduction

Although the incidence of breast cancer (BC) increases with age, it is the most commonly diagnosed malignancy in women of childbearing age with 10.5% of new cases diagnosed every year in patients younger than 45 years[1]. Younger women present more often with more aggressive BC requiring gonadotoxic che-

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motherapy and subsequently endocrine therapy for hormone receptor (HR)-positive BC, which can significantly impair fertility[2]. Advances in BC awareness, early detection, diagnosis and treatment options have increased BC survival rates. Because of the global trend of delayed childbearing, there is a growing population of young women diagnosed with BC before they have completed their family[3].

Data show that at the time of diagnosis, approximately 50% of young women are concerned about becoming infertile after BC treatment, but only a minority (10%) used fertility preservation strategies[4]. Therefore, fertility preservation in young BC patients has emerged as a very important survivorship issue regarding the quality of life for these women and it should be discussed early upon diagnosis with all BC patients of reproductive age. A recent meta-analysis showed that BC survivors who had received adjuvant systemic therapy for BC had a 14% chance of becoming pregnant with the pregnancy rate by 40% lower than the pregnancy rate in the general population.[5]

Therefore, BC patients of reproductive age should be informed about the potential negative effects of oncologic therapy on fertility and fertility preservation options, and if interested in fertility preservation, they should be referred to a reproductive specialist to further discuss the currently available fertility preservation options[3,6].

Currently, evidence suggests that pregnancy in BC survivors is not associated with an increased risk of recurrence regardless of hormonal status, therefore induction of abortion for therapeutic purposes is unjustified[7,8]. Although optimal timing of pregnancy after BC is not clear, patients are advised to delay pregnancy for at least 2 years after diagnosis when the risk of recurrence is highest[2]. The ongoing prospective POSITIVE trial (Pregnancy Outcome and Safety of Interrupting Therapy for Women with Endocrine Responsive BC, ClinicalTrials.gov Identifier: NCT02308085) is conducted to determine whether temporary interruption of endocrine therapy for young HR-positive BC patients who want to attempt pregnancy is associated with an increased risk of BC recurrence and to evaluate pregnancy success and offspring outcome. The aim of this review is to point out the importance of fertility preservation as an important quality of life issue for young BC survivors and to analyze the currently available options for fertility preservation in young women with early-stage BC.

Factors Influencing Fertility Preservation

Young women interested in fertility preservation should be referred to a reproductive specialist as soon as possible. Several factors should be discussed with BC patients who are faced with gonadotoxic therapy, i.e. type of cancer, patient age, reproductive history, comorbidities, type, dose, expected benefits and adverse effects (AE) of planned oncologic therapy, need for endocrine therapy, risk of infertility after planned oncologic treatment, fertility preservation options, and delay of cancer treatment[6,9]. Early referral gives young BC patients a chance to undergo multiple cycles of embryo/oocyte cryopreservation if desired, which increases the chance of successful fertility preservation because of the increased number of embryos/oocytes stored[10].

Early referral to reproductive center before surgery allows women who will undergo OS and fertility preservation to start adjuvant chemotherapy around 3 weeks earlier than those women who were referred after surgery[11]. Chemotherapy-induced ovarian damage depends on the patient age at BC diagnosis, individual ovarian reserve, and type and dose of planned chemotherapy. The risk of ovarian failure is greater in older patients, especially in patients older than 40 years, even at lower doses of gonadotoxic chemotherapy, primarily due to natural reduction in the number of primordial follicles that comes with aging[12,13]. Chemotherapeutic agents can cause damage to developing follicles because granulosa cells are proliferating cells and therefore cause transient amenorrhea. Induced ovarian damage is associated with a reduction in the follicle number, follicular apoptosis, damage to ovarian stroma, impaired blood flow, and increased follicular recruitment[12]. Alkylating agents cause deoxyribonucleic acid (DNA) double-strand breaks and they are considered to be the most gonadotoxic chemotherapeutic agents. As alkylating agents are non-cell-cycle specific, they can cause damage to both resting and growing follicles[12,13].

The dose of chemotherapy is also an important factor. The CMF (cyclophosphamide, methotrexate, 5-fluorouracil) protocol seems to be more gonadotoxic than the AC (doxorubicin, cyclophosphamide) protocol, probably because of a higher cumulative dose of cyclophosphamide reached with CMF compared to AC protocol[14]. Significantly lower rates of amenorrhea (34% *vs.* 69%) have been reported with the AC protocol com-

Table 1. Fertility preservation methods for young women with early-stage breast cancer

Fertility preservation method	Advantages	Disadvantages
Embryo cryopreservation	• Well established technique • Possibility of PGD	• Need for OS • 2-5 week delay of oncologic treatment • Requires male partner/sperm donor • Expensive • Ethically questionable • Legal issues
Cryopreservation of mature oocytes	• Well established technique • No need for male partner/sperm donor • Possibility of PGD	• Need for OS • 2-5 week delay of oncological treatment • Expensive
Cryopreservation of ovarian tissue	• No need for OS • Menstrual cycle independent method • No need for delay in oncologic treatment • Restoration of endocrine function • Fertility preservation method for young women who already started gonadotoxic chemotherapy • Combination with *in vitro* maturation • No need for male partner/ sperm donor	• Experimental method • Potential reintroduction of malignant cells within ovarian tissue • Expensive • Need for surgery • Success is highly dependent on ovarian reserve, it is not recommended for patients older than 35 years • Available only in highly specialized centers
Cryopreservation of immature oocytes	• No need for OS or short OS lasting for 3-5 days • Menstrual cycle independent method • Shortened period to initiation of cancer treatment compared to standard cryopreservation methods • No need for male partner/sperm donor	• Experimental method • Expensive • Technically demanding • Implantation and pregnancy rates significantly lower than with standard cryopreservation methods
GnRHa	• Simple administration • Noninvasive • No need for assisted reproductive technologies • No need to delay start of chemotherapy • No need for male partner/sperm donor	• Experimental method • Conflicting efficacy data • Slightly more grade 2 adverse events (hot flushes, headache)

PGD = preimplantation genetic diagnosis; OS = ovarian stimulation; GnRHa = gonadotropin-releasing hormone agonists

pared to CMF protocol[15]. It has been reported that chemotherapy protocols such as AC may result in a 10-year loss of oocyte reserve[16]. The addition of taxane and trastuzumab to AC protocol or dose-dense approach did not seem to significantly increase the risk of amenorrhea compared to AC protocol[17]. An update from a single arm adjuvant paclitaxel-trastuzumab (APT) study showed a lower amenorrhea rate (28% *vs*. 95% CI; 18%-41%) among premenopausal women treated with adjuvant paclitaxel and trastuzumab compared to standard BC regimens with alkylating agent[18].

Furthermore, surgeons, radiologists and psychosocial providers should be informed about this important issue and included in the management of BC patients. Early referral to a multidisciplinary team offers selection of the most required fertility preservation method for fertility preservation, between the standard methods such as embryo or oocyte cryopreservation and experimental options including ovarian tissue cryopreservation, cryopreservation of immature oocytes and ovarian suppression with gonadotropin-releasing hormone agonists (GnRHa)[19]. An overview of available fertility preservation methods, including their advantages and disadvantages, is shown in Table 1.

Embryo Cryopreservation

Embryo cryopreservation is the most established fertility preservation method for BC patients who have male partners, or for those women who are using

donor sperm, but it has several ethical and legal concerns such as prohibition of this method in some countries, posthumous reproduction or divorce[20]. This method combines ovarian stimulation (OS), oocyte retrieval and *in vitro* fertilization (IVF). The success of this method is highly dependent on the number of stored oocytes or embryos and patient age. OS with gonadotropins is needed to gain more than one oocyte *per* cycle and it is very important for successful IVF, especially because most BC patients usually have only one opportunity to undergo IVF protocol before starting gonadotoxic treatment[10,21]. Controlled OS for fertility preservation is considered not to be associated with an increased risk of BC recurrence[22]. The process of embryo cryopreservation delays oncologic treatment for 2 to 5 weeks, therefore, it is not recommended for patients who cannot delay BC treatment[21].

In order to reduce the risk of short-term exposure to very high estrogen levels in BC patients, alternative and safer protocols for OS using aromatase inhibitors and tamoxifen with gonadotropins have been developed to reduce estrogen production[21,23,24]. The preferred option for OS in fertility preservation cycles for women with HR-positive BC are stimulation protocols using letrozole with gonadotropins because they are safe, more effective than tamoxifen protocols, and are associated with a higher number of oocytes retrieved and fertilized. Use of these protocols resulted in overall live birth *per* embryo transfer of 45%, which is comparable to those in a non-cancer population undergoing IVF because of infertility[22,25,26].

Random-start OS protocols for emergency fertility preservation are initiated in the late follicular or luteal phase of the menstrual cycle and they allow OS to start anytime during the menstrual cycle. Random-start protocols have shown to be efficient for fertility preservation with similar numbers of retrieved oocytes, mature oocyte yield, and fertilization rates. With the use of these protocols, there is no need to wait for the onset of the next menstrual cycle because they reduce the waiting period for egg retrieval to about 2 weeks, which allows earlier start of oncologic treatment[19,27,28].

Double OS protocols are developed to allow double stimulation in both follicular and luteal phase of the same menstrual cycle in order to increase the number of obtained oocytes with subsequent improvement in birth rate, without delaying cancer treatment. These protocols could be suitable for patients of advanced maternal age and reduced ovarian reserve but they need to be further investigated[29,30].

Oocyte Cryopreservation

Oocyte cryopreservation is an alternative method to embryo cryopreservation for those women without a partner or those who do not want to use donor sperm and in the countries where embryo cryopreservation is forbidden by law[19]. This technique also requires controlled OS and oocyte retrieval, thus having the same disadvantages as embryo cryopreservation. Vitrification has significantly improved live birth rates, with a higher live birth rate reported for women younger than 35 years (live birth rate 50% *vs*. 22.9% in women older than 36 years)[31].

Cryopreservation of Immature Oocytes

Cryopreservation of immature oocytes or oocytes matured *in vitro* is a promising experimental fertility preservation option for BC patients. This method is menstrual cycle independent, does not require OS, although short OS lasting for 3-5 days can be performed, which will result in a shortened period from BC diagnosis to initiation of cancer treatment[14]. Upon retrieval, immature oocytes can be either cryopreserved after *in vitro* maturation or cryopreserved at the immature stage and matured *in vitro* after thaw. *In vitro* maturation (IVM) before cryopreservation is reported to be a better option because it results in higher maturation and survival rates than post-thaw maturation (63.8% *vs*. 33.3%, $p<0.05$)[32]. Data from studies using IVM oocytes are still limited, the implantation and pregnancy rates are significantly lower than with standard IVF, the use of this method should be considered experimental, and it should be performed only in specialized centers[33].

Ovarian Tissue Cryopreservation

Ovarian tissue cryopreservation (OTC) is an experimental fertility preservation method in which the ovarian cortex, which contains a lot of primordial follicles, is removed surgically and then cryopreserved. Upon completion of oncologic treatment, the ovarian tissue can be thawed and transplanted back into the patient, either to orthotopic (into the pelvic cavity; on the atrophic ovary, pelvic peritoneum) or heterotopic sites (outside of the pelvis; subcutaneous regions such as the forearm, abdominal wall)[34].

This method has several advantages, i.e. it can be performed at any time during the menstrual cycle, there is no need for OS, it does not require delay in the initiation of oncologic treatment or male partner/sperm donor, it results in storage of a large number of primordial follicles and restores endocrine function[34,35]. OTC is an option for those BC patients who require immediate start of gonadotoxic therapy and do not have enough time to undergo OS for embryo/oocyte cryopreservation. After reimplantation, ovarian function is expected to be recovered after 4-5 months in the majority of cases, and ovarian function is restored in more than 90% of patients with the mean duration of ovarian function after reimplantation of 4-5 years[36]. Several factors can affect ovarian graft longevity, including advanced age at the time of OTC, previous chemotherapy exposure, graft size and method of cryopreservation, inhomogeneous distribution of follicles in ovarian graft, and post-transplantation ischemia[36]. Age is an important factor that should be taken into account because the success of OTC is highly dependent on ovarian reserve, which decreases with age. Suggested selection criteria for OTC are age younger than 35 years, a realistic chance of 5-year survival, and at least 50% risk of premature ovarian insufficiency[34]. Post-transplantation ischemia can cause high follicular loss after transplantation (up to 70% of follicles)[37]. It is desirable to retrieve ovarian tissue for cryopreservation before initiation of gonadotoxic treatment, but this method can be performed in those young women who normally have a high number of primordial follicles in their ovaries and have already started chemotherapy[38]. The reported live birth and ongoing pregnancy rate of 37.7% for OTC with endocrine restoration rate of 63.9% has reached promising levels[39]. It is also suggested that a combination of oocyte vitrification and OTC could increase the live birth rate to 50%-60%[34]. Only one (twin) pregnancy has been reported when ovarian tissue was transplanted to a heterotopic site[40]. The greatest concern about this method is safety of the procedure because for fear of potential reimplantation of malignant cells, mostly in patients with hematologic malignancies. Ovarian tissue should be appropriately examined (histology, immunohistochemistry, polymerase chain reaction) to exclude malignant involvement of ovarian tissue in cancer patients undergoing OTC[35].

When there is a risk of transferring malignant cells, ovarian follicles can be isolated and matured *in vitro*, and then fertilized and transferred to the patient. Another option is the formation of an artificial ovary where isolated follicles are encapsulated into a scaffold, which allows them to grow and develop in an ovarian-like environment and to be grafted to the patient[34]. Additional potential improvements of this method could be the use of robotic surgery and protective agents such as AS101 or sphingosine-1-phosphate (S1P)[34].

Ovarian Suppression with Gonadotropin-Releasing Hormone Agonists

Administration of GnRHa is an attractive option for fertility preservation in BC patients because it is noninvasive, easy to administer, does not require the usage of assisted reproductive technologies, and does not require delay in chemotherapy initiation[9]. Because of a flare effect at the beginning of the treatment with GnRHa, which lasts for about one week, GnRHa should be administered at least one week before chemotherapy. The idea for the usage of GnRHa ensued from the hypothesis that induced gonadal quiescence during chemotherapy could reduce chemotherapy-induced damage[41]. There are several possible mechanisms of ovarian protection with the usage of GnRHa, including decreased ovarian perfusion and delivery of chemotherapy to the ovaries, prevention of the increased recruitment of primordial follicles by the increased follicle stimulating hormone (FSH) concentration induced through apoptosis of the growing follicles, up-regulation of antiapoptotic pathways within the ovary and protection of ovarian germline stem cells[42]. More advanced follicles are gonadotropin-responsive and they secrete growth factors (such as TGF-β, BMP, activin) that cause growth of primordial and primary follicles. Burnout theory suggests that the gonadotoxic effect of chemotherapy leads to increased concentration of FSH because it causes death of the follicles and decreased levels of estrogen and inhibin. GnRHa decreases FSH levels; therefore, it could cause decreased recruitment of primordial follicles[43]. On the other hand, primordial follicles do not express gonadotropin receptors; therefore, GnRHa cannot have a direct effect on ovarian reserve[44].

The usage of GnRH agonist in fertility preservation is still considered experimental. Although a recent meta-analysis has suggested that GnRHa reduces the risk of premature ovarian failure (adjusted OR 0.38; 95% CI, 0.26-0.57; p<0.001), the data need to be further investigated[45]. The majority of clinical studies that showed benefit from GnRHa used amenorrhea as a primary outcome, which is an inappropriate surrogate for ovarian function and reproductive potential[44]. Ovarian reserve might be severely diminished despite the resumption of menstruation and those women are more likely to develop early menopause[46]. Also, young non-menstruating women may still be fertile despite the low ovarian reserve because of the high quality of the remaining oocytes[44]. Other limitations of GnRHa studies were the lack of randomization and appropriate controls, retrospective design of studies, and limited long-term data[44].

The best primary outcome for fertility preservation is successful pregnancy. Although the number of pregnant patients in a recent meta-analysis was relatively small (37 pregnancies in patients treated with GnRHa (10.5%) *vs.* 20 pregnancies in control group (5.5%), incidence rate ratio 1.83; 95% CI, 1.06-3.15; p=0.030), it suggests a higher chance for pregnancy with the use of GnRHa[45].

Besides pregnancy rate, fertility preservation can be assessed through ovarian reserve biomarkers such as antimüllerian hormone (AMH), inhibin B levels, or antral follicle count[12]. AMH is considered to be the most sensitive marker of ovarian reserve as it is produced by granulosa cells of primary follicles and is involved in the regulation of primordial follicle recruitment. Decreased AMH levels are associated with diminished ovarian reserve following chemotherapy[12]. A meta-analysis which included 10 trials where ovarian reserve parameters including AMH were evaluated showed no benefit from GnRHa treatment[47].

Also, it is important to note that the addition of GnRHa to chemotherapy does not seem to increase the incidence of serious (grade 3) adverse effects (AE), although the rates of grade 2 AE were reported to be increased (48% *vs.* 24%, p<0.001), mostly the increased risk of hot flushes and headaches[48]. The use of GnRHa is still considered to be an experimental fertility preservation method but GnRHa can be considered when it is not possible to perform embryo/oocyte cryopreservation, or in addition to these established fertility preservation techniques[19].

Other Noninvasive Fertility Preservation Strategies

Usage of new fetoprotective agents could protect ovaries from chemotherapy-induced damage. Burnout theory suggests that the gonadotoxic effect of chemotherapy leads to activation of dormant follicle growth[49]. Co-administration of the AS101 immunomodulator acts on the PI3K/PTEN/Akt pathway, which is important in the activation of dormant follicles. AS101 prevents follicle activation, which leads to reduced follicular loss and improved reproductive outcomes in mice treated with cyclophosphamide[50]. AMH is also important in the regulation of follicle activation as an inhibitor of recruitment of primordial follicles. The use of recombinant human AMH showed reduced preclinical follicle loss[49]. S1P is a ceramide-induced apoptotic inhibitor, which can prevent chemotherapy induced apoptotic follicle loss in preclinical models, as well as imatinib and granulocyte colony-stimulating factor (G-CSF)[49]. Although these agents are promising noninvasive strategies for fertility preservation, it is important to note that the major concern for clinical usage of these agents is the potential interaction with oncologic treatments and reduced therapeutic effects[10,49].

Neoadjuvant Chemotherapy and Fertility Preservation

Young BC patients are often candidates for neoadjuvant chemotherapy because they present more often with larger, node-positive tumors and more aggressive disease (triple-negative, Her-2 positive BC). In neoadjuvant setting there is a desire to start oncologic treatment as soon as possible, but there are several obstacles that are delaying this process such as referral time to oncology specialists, additional oncology consultation, additional diagnostic work-up (MRI, additional biopsies, staging scans, echocardiogram)[51]. In the meantime, patients could also have the opportunity to see a reproductive specialist, discuss their fertility preservation options, and if desired undergo fertility preservation.

There is a concern regarding delay and safety of OS in neoadjuvant settings and its potential influence on tumor growth and spread. Random-start protocols with letrozol do not seem to increase the risk of BC recurrence, but reduce the risk of short-term exposure

to very high estrogen levels, allow earlier start of anticancer treatment by reducing the waiting period for egg retrieval, and can be performed in neoadjuvant settings as long as there is prompt referral[27,28,52]. There is the need to further evaluate safety of the established fertility preservation methods in the neoadjuvant setting. The neoadjuvant setting is a good setting for still experimental fertility preservation methods such as OTC and cryopreservation of immature ovarian cells because these methods do not require delay in the initiation of oncologic treatment[13]. Good collaboration among medical oncologists, surgeons and reproductive specialists is crucial for successful fertility preservation, especially in a neoadjuvant setting.

BRCA Mutations and Fertility Preservation

Women carrying BRCA1 and BRCA2 mutations have an increased lifetime risk of developing BC, contralateral BC, and ovarian cancer. A recent prospective study reported the lifetime risk of BC to be around 70% for BRCA1 and BRCA2 carriers, and lifetime risk of ovarian cancer to be 44% for BRCA1 and 17% for BRCA2 carriers[53]. By the age of 40, the reported cumulative risk of BC development is 24% for BRCA1 and 13% for BRCA2 carriers, while the cumulative risk of ovarian cancer is 2% for BRCA1 and 0% for BRCA2 carriers[53]. Women with BRCA1/2 mutation are advised to undergo bilateral salpingo-oophorectomy before age 35-40, after they have completed childbearing to reduce the risk of developing ovarian cancer and BC[54].

Data suggest that BRCA mutation carriers may have reduced reproductive potential, i.e. diminished ovarian reserve, lower AMH levels, poorer response to controlled ovarian stimulation with letrozole protocols, more pronounced in women with BRCA1 mutation[55-57]. It is also reported that BRCA1/2 mutation carriers are more likely to experience earlier natural menopause, approximately 3-4 years earlier than healthy women[58]. Gonadotoxic effects of chemotherapy may be more pronounced in BC patients with BRCA 1/2 mutation, as deficient homologous-recombination DNA repair makes oocytes of these women more vulnerable to gonadotoxic therapy[59]. Taking into consideration the potentially reduced ovarian reserve in BC patients with BRCA1/2 mutations, using double OS protocols may be useful[29,30]. Another option for fertility preservation in BC patients with BRCA1/2 mutation is OTC, although there is a safety concern because of the risk of developing ovarian cancer[21,59]. Heterotopic OTC may be the preferred option for those women because it allows closer monitoring of ovarian tissue[16]. BRCA1/2 mutations are inherited in an autosomal dominant fashion, so there is a 50% risk of transmission of the mutated gene to the patient's offspring. Preimplantation genetic diagnosis for BRCA mutation during IVF can be performed to avoid mutation transmission to the embryo, although this is ethically questionable because BRCA mutations are not lethal mutations and their presence does not guarantee cancer occurrence[16,59,60]. Egg donation and surrogacy are the possible options for women harboring BRCA mutations in countries where those options are available and legal.

Conclusions

Fertility preservation is an emerging field in oncology that gives an opportunity for young cancer survivors to maintain reproductive health and have children after oncologic treatment. Pregnancy after BC treatment does not seem to increase the risk of BC recurrence. Unfortunately, fertility preservation is an underdiscussed issue in young BC patients, which should be discussed with every woman of reproductive age diagnosed with early-stage BC and the women interested in fertility preservation should be promptly referred to fertility specialists before starting gonadotoxic therapy. Early referral to a reproductive specialist is a crucial component of fertility preservation, which increases the chance for successful fertility preservation. Good collaboration among specialists included in the management of these patients may increase the likelihood of successful fertility preservation. Currently, embryo and oocyte cryopreservation are safe, effective and well-established options for fertility preservation in BC survivors, whereas other methods, although promising, are still considered to be experimental and controversial, and need to be further investigated.

References

1. National Cancer Institute – Surveillance, Epidemiology and End Results Program. http://seer.cancer.gov/statfacts/html/breast.html. Accessed 7 May 2017
2. Kasum M, Beketić-Orešković L, Orešković S. Subsequent pregnancy and prognosis in breast cancer survivors. Acta Clin Croat. 2014;53(3):334-41.

3. Kasum M, Beketić-Orešković L, Peddi PF, Orešković S, Johnson RH. Fertility after breast cancer treatment. Eur J Obstet Gynecol Reprod Biol. 2014;173:13-8. doi: 10.1016/j.ejogrb.2013.11.009
4. Ruddy KJ, Gelber SI, Tamimi RM, Ginsburg ES, Schapira L, Come SE, *et al*. Prospective study of fertility concerns and preservation strategies in young women with breast cancer. J Clin Oncol. 2014;32:1151-6. doi: 10.1200/JCO.2013.52.8877
5. Gerstl B, Sullivan E, Ives A, Saunders C, Wand H, Anazodo A. Pregnancy outcomes after a breast cancer diagnosis: a systematic review and meta-analysis. Clin Breast Cancer. 2018;18 (1):e79-e88. doi: 10.1016/j.clbc.2017.06.016
6. Paluch-Shimon S, Pagani O, Partridge AH, Abulkhair O, Cardoso M-J, Dent RA, *et al*. ESO-ESMO 3rd International Consensus Guidelines for Breast Cancer in Young Women (BCY3). Breast. 2017;35:203e17. doi: 10.1016/j.breast.2017.07.017.
7. Azim HA, Kroman N, Paesmans M, Gelber S, Rotmensz N, Ameye L, *et al*. Prognostic impact of pregnancy after breast cancer according to estrogen receptor status: a multicenter retrospective study. J Clin Oncol. 2013;31:73-9. doi: 10.1200/JCO.2012.44.2285
8. Azim HA Jr, Santoro L, Pavlidis N, Gelber S, Kroman N, Azim H, *et al*. Safety of pregnancy following breast cancer diagnosis: a meta-analysis of 14 studies. Eur J Cancer. 2011;47(1):74-83. doi: 10.1016/j.ejca.2010.09.007
9. Lambertini M, Del Mastro L, Pescio MC, Andersen CY, Azim HA Jr, Peccatori FA, et al. Cancer and fertility preservation: international recommendations from an expert meeting. BMC Med. 2016;14:1. doi: 10.1186/s12916-015-0545-7
10. Kasum M. Očuvanje plodnosti bolesnica liječenih od karcinoma dojke. In: Beketić-Orešković L, Šantek F, eds. Karcinom dojke. Zagreb: Medicinska naklada, 2018; p. 203-11. (in Croatian)
11. Lee S, Ozkavukcu S, Heytens E, Moy F, Oktay K. Value of early referral to fertility preservation in young women with breast cancer. J Clin Oncol. 2010;28(31):4683-6. doi: 10.1200/JCO.2010.30.5748
12. Bedoschi G, Navarro PA, Oktay K. Chemotherapy-induced damage to ovary: mechanisms and clinical impact. Future Oncol. 2016;12(20):2333-44. doi: 10.2217/fon-2016-0176
13. Waks AG, Partridge AH. Fertility preservation in patients with breast cancer: necessity, methods, and safety. J Natl Compr Canc Netw. 2016;14(3):355-63.
14. Taylan E, Oktay K. Current state and controversies in fertility preservation in women with breast cancer. World J Clin Oncol. 2017;8(3):241-8. doi: 10.5306/wjco.v8.i3.241
15. Bines J, Oleske DM, Cobleigh MA. Ovarian function in premenopausal women treated with adjuvant chemotherapy for breast cancer. J Clin Oncol. 1996;14:1718-29. doi:10.1200/JCO.1996.14.5.1718
16. Rodriguez-Wallberg KA, Oktay K. Fertility preservation and pregnancy in women with and without BRCA mutation-positive breast cancer. Oncologist. 2012;17(11):1409-17. doi: 10.1634/theoncologist.2012-0236
17. Abusief ME, Missmer SA, Ginsburg ES, Weeks JC, Partridge AH. The effects of paclitaxel, dose density, and trastuzumab on treatment-related amenorrhea in premenopausal women with breast cancer. Cancer. 2010;116:791-8. doi: 10.1002/ cncr.24835
18. Ruddy KJ, Guo H, Barry W, Dang CT, Yardley DA, Moy B, *et al*. Chemotherapy-related amenorrhea after adjuvant paclitaxel-trastuzumab (APT trial). Breast Cancer Res Treat. 2015; 151:589-96. doi: 10.1007/s10549-015-3426-z
19. Oktay K, Harvey BE, Partridge AH, Quinn GP, Reinecke J, Taylor HS. Fertility preservation in patients with cancer: ASCO Clinical Practice Guideline Update. J Clin Oncol. 2018 Apr 5:JCO2018781914. doi: 10.1200/JCO.2018.78.1914
20. Bedoschi G, Oktay K. Current approach to fertility preservation by embryo cryopreservation. Fertil Steril. 2013;99(6): 1496-502. doi: 10.1016/j.fertnstert.2013.03.020
21. Kasum M, von Wolff M, Franulić D, Čehić E, Klepac-Pulanić T, Orešković S, *et al*. Fertility preservation options in breast cancer patients. Gynecol Endocrinol. 2015;31(11):846-51. doi: 10.3109/09513590.2015.1081684
22. Rodriguez-Wallberg KA, Eloranta S, Krawiec K, Lissmats A, Bergh J, Liljegren A. Safety of fertility preservation in breast cancer patients in a register-based matched cohort study. Breast Cancer Res Treat. 2018;167(3):761-9. doi: 10.1007/s10549-017-4555-3
23. Reddy J, Oktay K. Ovarian stimulation and fertility preservation with the use of aromatase inhibitors in women with breast cancer. Fertil Steril. 2012;98:1363-9. doi: 10.1016/j.fertnstert.2012.09.022
24. Meirow D, Raanani H, Maman E, Paluch-Shimon S, Shapira M, Cohen Y, *et al*. Tamoxifen co-administration during controlled ovarian hyperstimulation for *in vitro* fertilization in breast cancer patients increases the safety of fertility-preservation treatment strategies. Fertil Steril. 2014;102(2):488-95.e3. doi: 10.1016/j.fertnstert.2014.05.017
25. Azim AA, Costantini-Ferrando M, Oktay K. Safety of fertility preservation by ovarian stimulation with letrozole and gonadotropins in patients with breast cancer: a prospective controlled study. J Clin Oncol. 2008;26:2630-5. doi: 10.1200/JCO.2007.14.8700
26. Oktay K, Turan V, Bedoschi G, Pacheco FS, Moy F. Fertility preservation success subsequent to concurrent aromatase inhibitor treatment and ovarian stimulation in women with breast cancer. J Clin Oncol. 2015;33(22):2424-9. doi: 10.1200/JCO.2014.59.3723
27. Cakmak H, Rosen MP. Random-start ovarian stimulation in patients with cancer. Curr Opin Obstet Gynecol. 2015;27: 215-21. doi: 10.1097/GCO.0000000000000180
28. Danis RB, Pereira N, Elias RT. Random start ovarian stimulation for oocyte or embryo cryopreservation in women desiring fertility preservation prior to gonadotoxic cancer therapy. Curr Pharm Biotechnol. 2017;18(8):609-13. doi: 10.2174/1389201018666170808122531

29. Kuang Y, Chen Q, Hong Q, Lyu Q, Ai A, Fu Y, *et al*. Double stimulations during the follicular and luteal phases of poor responders in IVF/ICSI programmes (Shanghai protocol). Reprod Biomed Online. 2014;29:684-91. doi: 10.1016/j.rbmo.2014.08.009

30. Ubaldi FM, Capalbo A, Vaiarelli A, Cimadomo D, Colamaria S, Alviggi C, *et al*. Follicular *versus* luteal phase ovarian stimulation during the same menstrual cycle (DuoStim) in a reduced ovarian reserve population results in a similar euploid blastocyst formation rate: new insight in ovarian reserve exploitation. Fertil Steril. 2016;105:1488-95. doi: 10.1016/j.fertnstert.2016.03.002

31. Cobo A, García-Velasco JA, Coello A, Domingo J, Pellicer A, Remohí J. Oocyte vitrification as an efficient option for elective fertility preservation. Fertil Steril. 2016;105(3):755-64.e8. doi: 10.1016/j.fertnstert.2015.11.027

32. Lee JA, Sekhon L, Grunfeld L, Copperman AB. *In-vitro* maturation of germinal vesicle and metaphase I eggs prior to cryopreservation optimizes reproductive potential in patients undergoing fertility preservation. Curr Opin Obstet Gynecol. 2014;26(3):168-73. doi: 10.1097/GCO.0000000000000062

33. Practice Committees of the American Society for Reproductive Medicine and the Society for Assisted Reproductive Technology. *In vitro* maturation: a committee opinion. Fertil Steril. 2013;99(3):663-6. doi: 10.1016/j.fertnstert.2012.12.031

34. Donnez J, Dolmans MM. Fertility preservation in women. N Engl J Med. 2017;377:1657-65. doi: 10.1056/NEJMra1614676

35. Rosendahl M, Timmermans Wielenga V, Nedergaard L, Kristensen SG, Ernst E, Rasmussen PE, *et al*. Cryopreservation of ovarian tissue for fertility preservation: no evidence of malignant cell contamination in ovarian tissue from patients with breast cancer. Fertil Steril. 2011;95(6):2158-61. doi: 10.1016/j.fertnstert.2010.12.019

36. Donnez J, Dolmans MM, Pellicer A, Diaz-Garcia C, Sanchez Serrano M, Schmidt KT, *et al*. Restoration of ovarian activity and pregnancy after transplantation of cryopreserved ovarian tissue: a review of 60 cases of reimplantation. Fertil Steril. 2013;99(6):1503-13. doi: 10.1016/j.fertnstert.2013.03.030

37. Salama M, Winkler K, Murach KF, Seeber B, Ziehr SC, Wildt L. Female fertility loss and preservation: threats and opportunities. Ann Oncol. 2013;24(3):598-608. doi: 10.1093/annonc/mds514

38. Rodriguez-Wallberg KA, Oktay K. Fertility preservation during cancer treatment: clinical guidelines. Cancer Manag Res. 2014;6:105-17. doi: 10.2147/CMAR.S32380

39. Pacheco F, Oktay K. Current success and efficiency of autologous ovarian transplantation: a meta-analysis. Reprod Sci. 2017 Aug;24(8):1111-20. doi: 10.1177/1933719117702251. Epub 2017 Jul 13

40. Stern CJ, Gook D, Hale LG, Agresta F, Oldham J, Rozen G, *et al*. First reported clinical pregnancy following heterotopic grafting of cryopreserved ovarian tissue in a woman after a bilateral oophorectomy. Hum Reprod. 2013;28:2996-9. doi: 10.1093/humrep/det360

41. Rivkees SA, Crawford JD. The relationship of gonadal activity and chemotherapy-induced gonadal damage. JAMA. 1988; 259:2123-5.

42. Blumenfeld Z. How to preserve fertility in young women exposed to chemotherapy? The role of GnRH agonist cotreatment in addition to cryopreservation of embryo, oocytes, or ovaries. Oncologist. 2007;12(9):1044-54. doi: 10.1634/theoncologist.12-9-1044

43. Garrido-Oyarzún MF, Castelo-Branco C. Controversies over the use of GnRH agonists for reduction of chemotherapy-induced gonadotoxicity. Climacteric. 2016;19(6):522-5. doi:10.1080/13697137.2016.1225713

44. Oktay K, Bedoschi G. Appraising the biological evidence for and against the utility of GnRHa for preservation of fertility in patients with cancer. J Clin Oncol. 2016;34(22):2563-5. doi: 10.1200/JCO.2016.67.1693

45. Lambertini M, Moore HCF, Leonard RCF, Loibl S, Munster P, Bruzzone M, *et al*. Gonadotropin-releasing hormone agonists during chemotherapy for preservation of ovarian function and fertility in premenopausal patients with early breast cancer: a systematic review and meta-analysis of individual patient-level data. J Clin Oncol. 2018; JCO2018780858. doi: 10.1200/JCO.2018.78.0858

46. Azim HA Jr, Davidson NE, Ruddy KJ. Challenges in treating premenopausal women with endocrine-sensitive breast cancer. Am Soc Clin Oncol Educ Book. 2016;35:23-32. doi: 10.14694/EDBK_159069

47. Elgindy E, Sibai H, Abdelghani A, Mostafa M. Protecting ovaries during chemotherapy through gonad suppression: a systematic review and meta-analysis. Obstet Gynecol. 2015;126: 187-95. doi: 10.1097/AOG.0000000000000905

48. Moore HCF, Unger JM, Phillips KA, Boyle F, Hitre E, Porter D, *et al*. Goserelin for ovarian protection during breast-cancer adjuvant chemotherapy. N Engl J Med. 2015;372:923-32. doi: 10.1056/NEJMoa1413204

49. Roness H, Kashi O, Meirow D. Prevention of chemotherapy-induced ovarian damage. Fertil Steril. 2016;105(1):20-9. doi: 10.1016/j.fertnstert.2015.11.043

50. Kalich-Philosoph L, Roness H, Carmely A, Fishel-Bartal M, Ligumsky H, Paglin S, *et al*. Cyclophosphamide triggers follicle activation and "burnout"; AS101 prevents follicle loss and preserves fertility. Sci Transl Med. 2013;5(185):185ra62. doi: 10.1126/scitranslmed.3005402.

51. Chien AJ, Chambers J, Mcauley F, Kaplan T, Letourneau J, Hwang J, *et al*. Fertility preservation with ovarian stimulation and time to treatment in women with stage II-III breast cancer receiving neoadjuvant therapy. Breast Cancer Res Treat. 2017;165(1):151-9. doi: 10.1007/s10549-017-4288-3

52. Letourneau JM, Sinha N, Wald K, Harris E, Quinn M, Imbar T, *et al*. Random start ovarian stimulation for fertility preservation appears unlikely to delay initiation of neoadjuvant chemotherapy for breast cancer. Hum Reprod. 2017;32(10):2123-9. doi: 10.1093/humrep/dex276

53. Kuchenbaecker KB, Hopper JL, Barnes DR, Phillips KA, Mooij TM, Roos-Blom MJ, *et al*. Risks of breast, ovarian, and

contralateral breast cancer for BRCA1 and BRCA2 mutation carriers. JAMA. 2017;317(23):2402-16. doi: 10.1001/jama.2017.7112

54. Paluch-Shimon S, Cardoso F, Sessa C, Balmana J, Cardoso MJ, Gilbert F, *et al*. ESMO Guidelines Committee. Prevention and screening in BRCA mutation carriers and other breast/ovarian hereditary cancer syndromes: ESMO Clinical Practice Guidelines for cancer prevention and screening. Ann Oncol. 2016; 27(Suppl 5):103-10. doi: 10.1093/annonc/mdw327
55. Turan V, Bedoschi G, Emirdar V, Moy F, Oktay K. Ovarian stimulation in patients with cancer: impact of letrozole and BRCA mutations on fertility preservation cycle outcomes. Reprod Sci. 2018;25(1):26-32. doi: 10.1177/1933719117728800
56. de la Noval BD. Potential implications on female fertility and reproductive lifespan in BRCA germline mutation women. Arch Gynecol Obstet. 2016;294(5):1099-103. doi: 10.1007/s00404-016-4187-6
57. Lambertini M, Goldrat O, Ferreira AR, Dechene J, Azim HA Jr, Desir J, *et al*. Reproductive potential and performance of fertility preservation strategies in BRCA-mutated breast cancer patients. Ann Oncol. 2018 Jan 1;29(1):237-43. doi: 10.1093/annonc/mdx639
58. Lin WT, Beattie M, Chen LM, Oktay K, Crawford SL, Gold EB, *et al*. Comparison of age at natural menopause in BRCA1/2 mutation carriers with a non-clinic-based sample of women in northern California. Cancer. 2013;119(9):1652-9. doi: 10.1002/cncr.27952
59. Peccatori FA, Mangili G, Bergamini A, Filippi F, Martinelli F, Ferrari F, *et al*. Fertility preservation in women harboring deleterious BRCA mutations: ready for prime time? Hum Reprod. 2018;33(2):181-7. doi: 10.1093/humrep/dex356
60. Daum H, Peretz T, Laufer N. BRCA mutations and reproduction. Fertil Steril. 2018 Jan;109(1):33-8. doi: 10.1016/j.fertnstert.2017.12.004

Sažetak

OČUVANJE PLODNOSTI U MLADIH ŽENA S RANIM RAKOM DOJKE

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Premda se karcinom dojke češće javlja u starijoj životnoj dobi, to je i najučestaliji malignitet u žena reproduktivne dobi. Zbog sveukupnog napretka moderne medicine i rastućeg globalnog trenda odgađanja rađanja djece za kasniju dob suočavamo se sa sve više mladih žena s dijagnosticiranim i liječenim karcinomom dojke koje još nisu kompletirale obitelj. Stoga je područje očuvanja plodnosti postalo jako bitno u očuvanju kvalitete života mladih žena koje su preboljele karcinom dojke. Ovaj rad iznosi trenutno dostupne metode za očuvanje plodnosti u mladih žena s ranim karcinomom dojke i ističe važnost multidisciplinarnog pristupa u očuvanju plodnosti kao bitnog čimbenika kvalitete života tih žena. Smatra se da trudnoća nakon karcinoma dojke nije povezana s povišenim rizikom od recidiva pa stoga ne treba obeshrabriti žene koje žele ostvariti trudnoću nakon provedenog onkološkog liječenja. Danas se preporuča pričekati s trudnoćom barem 2 godine nakon postavljene dijagnoze za vrijeme kada je rizik od povrata bolesti najveći. No, isto tako bi bolesnice reproduktivne dobi trebalo obavijestiti o mogućem negativnom učinku onkološke terapije na plodnost te o dostupnim metodama očuvanja plodnosti i u slučaju zainteresiranosti za očuvanje plodnosti bolesnice treba žurno uputiti reproduktivnom specijalistu. Rano upućivanje reproduktivnom specijalistu je bitan čimbenik koji povećava izglede za uspješno očuvanje plodnosti. Krioprezervacija embrija i zrelih oocita su trenutno jedine standardne metode očuvanja plodnosti koje zahtijevaju stimulaciju ovarija kojom se odgađa početak kemoterapijskog liječenja barem 2 tjedna. Smatra se da kontrolirana stimulacija ovarija ne povećava rizik od povrata karcinoma dojke. Druge metode očuvanja plodnosti (krioprezervacija tkiva jajnika, krioprezervacija nezrelih oocita, ovarijska supresija GnRH agonistima) ne zahtijevaju primjenu ovarijske stimulacije, ali se i dalje smatraju eksperimentalnim metodama za očuvanje plodnosti.

Ključne riječi: *Plodnost, očuvanje – metode; Dojka, tumori; Kriokonzerviranje – metode; Ovulacija, indukcija; Gonadotropin-otpuštajući hormon – agonisti; Kvaliteta života*