



Abstracts from the 2019 ISFP meeting in NYC

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A MODIFIED HYDROSTATIC MICROFLUIDIC PUMPLESS DEVICE: A PRELIMINARY STUDY AS AN ALTERNATIVE APPROACH FOR IN VITRO FOLLICLE ACTIVATION IN MOUSE MODEL

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Introduction: In vitro follicle activation (IVA) theoretically involves with ovarian cortical culture for period of time. Fundamental in vitro tissue culture protocol, static culture system, with sequential culture media changed every day two consecutive days has been generally performed. However, this culture system propagates through missing of natural blood circulation mimicking leading to cell to cell interaction receding. This study aimed to compare efficacy of conventional (static culture system) and non-conventional (modified hydrostatic microfluidic pumpless device; MHMPD) systems on IVA using mouse model. **Methods:** Ovarian tissues were retrieved from four wild-type mice aged 2–3 months under approval by Institutional Animal Care and Use Committee, Faculty of Medicine, Chulalongkorn University. Ovarian cortical pieces (0.2 cm x 0.2 cm x 0.1 cm; width x length x thickness) from each animal were divided into two groups; I) conventional culture system (control) and II) non-conventional system with MHMPD (fabricated from standard soft lithography technique using Polydimethylsiloxane connected with modified media

tank and calculated for legitimated flow rate; 0 – 10 hours: 0.0003 mL/min and 11 – 20 hours: 0.00083 mL/min and constant flow rate after 20 hours). Ovarian tissues were cultured for four days and histologically evaluated for follicle morphology, follicle development growth rate and follicle density. **Results:** Our preliminary data demonstrated that mean (SD) percentage of normal follicle morphology (primordial to secondary stage) in fresh tissues and control were 78.1 ± 1.9 and 73.4 ± 3.9 whereas 69.2 ± 4.7 in MHMPD group. Growth rates from primordial to primary follicles were 2.2% and 1.9% in control and MHMPD groups, respectively. For follicle density (primordial to secondary stage), conventional culture system also yielded higher result than MHMPD. Nonetheless, non-conventional system with our current MHMPD prototype could not advocate a better outcome. **Conclusions:** The next generation platform should be further developed as alternative tool for IVA and fertility preservation in cancer patients. **Support:** The present study was financially supported by Ratchadaphiseksomphot Endowment Fund (Grant number RA61/066) and Research Unit of Reproductive Medicine and Fertility Preservation, Faculty of Medicine, Chulalongkorn University, Thailand. The authors are grateful to Dr. Triruk Pisitkun (MD); Head of System Biology Center – Chulalongkorn University, for laboratory animal support. **Disclosures:** Nothing to disclose
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ACTIVATION OF HUMAN OVARIAN STROMAL CELL PROLIFERATION VIA SIGNAL TRANSDUCTION AKT PATHWAY: A PRELIMINARY STUDY FOR ARTIFICIAL OVARY CONSTRUCTION FROM OVARIAN TISSUE CULTURE

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Introduction: Ovarian stromal cells have been considered one of key regulation factors during early follicle development in vivo. Therefore, their integrity and proliferation may accommodate follicle development during in vitro follicle activation by ovarian tissue culture which is imperative step prior to artificial ovary construction. The present study aimed to investigate influence of growth factors on stromal cell proliferation after long-term ovarian tissue culture. **Methods:** Ovarian tissues were collected from two premenopausal patients, age 39 and 48 years old, diagnosed with breast cancer and uterine fibroid, respectively. Use of human biological materials was approved by Institutional Review Board, Faculty of Medicine, Chulalongkorn University. Cortical tissues (average size; 0.5 x 0.5 x 0.1 cm) from each patient were randomly allocated into five groups; I) fresh, II) 10 day-tissue culture, III) 10 day-tissue cultured with 10 ng/mL epidermal growth factor (EGF), IV) 10 day-tissue cultured with 100 ng/mL Insulin, 55 ng/mL Transferrin and 0.5 ng/mL Sodium Selenite (ITS) and V) 10 day-tissue cultured with EGF and ITS. The evaluation criteria performed at day 0 and 10 were as following; cell proliferation (Ki-67: immunofluorescence assay), AKT signal transduction (western blot analysis) and ATP synthase gene expression (conventional PCR). **Results:** Preliminary results revealed that proliferation marker present in stromal cells could be visualized after 10-d culture. The highest positive Ki-67 cell staining per total 100 cell count was observed in cultured samples supplemented with ITS. Collectively, mean relative band intensity of signal transduction AKT and ATP synthase gene expression in ITS supplementation group was higher than the others (AKT = 0.019, 0.021, 0.019, 0.033 and 0.030; ATP synthase gene expression = 0.53, 0.52, 0.32, 0.62 and 0.55 respectively). **Conclusion:** The present findings preliminary contributed to possibility of stromal cell proliferation and activation by ITS via AKT signal transduction pathway which will benefit for further artificial ovary construction. **Support:** The authors are grateful for Assistant Prof. Tul Sittisomwong (MD); the operator in the second patient. The present study was financially supported by Research Unit of Reproductive Medicine and Fertility Preservation, Faculty of Medicine, Chulalongkorn University, Thailand. **Disclosures:** None

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ACTUAL TREATMENT RESULTS AND PREGNANCY OUTCOMES OF YOUNG PATIENTS WITH BREAST CANCER IN JAPAN

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Introduction: Chemotherapy (CT) and 5–10 years of endocrine therapy (ET) are essential treatments for young patients with breast cancer. However, subsequent declining fertility is an important issue in this population. We investigated the treatment results and pregnancy outcomes of young patients with breast cancer. **Methods:** We conducted a retrospective review involving 538 patients aged <40 years with Stage 0-IIIc breast cancer treated between 2007 and 2011. **Results:** At the time of surgery, 157 patients (29.2%) desired to get pregnant, 164 patients (30.5%) did not, and the desire of 217 patients (40.3%) was not known. Fertility preservation (FP) was performed in only 5 patients (3.2% of those desired pregnancy) prior to CT or ET. Based on the tumor status, CT was prescribed 289 patients, ET was prescribed 342 patients. Due to the desire for pregnancy, 2 patients refused CT, 12 refused ET, and 23 patients discontinued ET. During a median follow-up of 8.1 years, breast cancer recurrence was observed in 106 patients (19.7%) and 38 patients (14.4%) died due to breast cancer. Regarding childbirth after breast cancer, 73 patients gave birth (13.6% of all patients; 29.3% of those who desired pregnancy). Factors that significantly correlated with childbirth after breast cancer were a desire for pregnancy, younger age, early stage cancer, no lymph node metastasis, and absence of chemotherapy. There was no difference between childbirth after breast cancer and breast cancer recurrence or mortality. **Conclusion:** Most young patients with breast cancer prioritized cancer treatment over future pregnancy but only a few underwent FP. Young patients with high-risk breast cancer who desired pregnancy should consider undergoing FP before systemic treatment and subsequently attempting to become pregnant after completing the treatment. An adequate FP system must be established to support patients' decision making by providing appropriate medical information. **Support:** None **Disclosures:** None

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ADIPOSE TISSUE-DERIVED STEM CELLS TO ENHANCE EARLY REVASCULARIZATION AND FOLLICLE SURVIVAL RATES IN HUMAN OVARIAN TISSUE LONG-TERM XENOTRANSPLANTS

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Introduction: Our team recently developed a strategy to shorten hypoxia and ischemia in grafted human ovarian tissue (OT) using adipose tissue-derived stem cells (ASCs), which were peritoneally grafted 14 days prior to OT transplantation (OTT). Improved revascularization in the grafting site and higher follicle survival rates were

achieved 7 days post-transplantation with this 2-step grafting procedure. Indeed, ASCs exert a proangiogenic effect, differentiating into vessels and expressing growth factors. **Objective:** We aimed to investigate whether ASCs have long-term benefits in terms of folliculogenesis and primordial follicle pool preservation in 2-step OTT. **Methods:** Fourteen severe combined immunodeficient (SCID) mice were ovariectomized and xenografted with human OT from 7 patients using either the 2-step (ASCs+OT group, n=7) or standard (OT group, n=7) procedure. Monthly blood tests evaluated anti-Müllerian hormone (AMH) levels by enzyme-linked immunosorbent assays (ELISA). All OT fragments were retrieved after 6 months for histology and AMH immunolabeling, and compared with non-grafted controls. **Results:** Follicle survival rates fell significantly to 40% in both grafted groups. Primordial follicle density was significantly lower in the OT group than in non-grafted controls (9.65 ± 17.6 vs 124.7 ± 140 , $p=0.01$), but no significant difference was detected in the ASCs+OT group (41.86 ± 28.35) compared to non-grafted controls. Primordial follicle proportions (mean percentage $\pm$ SD) were significantly lower in the OT group (13.9 ± 16.14) than in non-grafted controls (68.43 ± 36.7) and the ASCs+OT group (51.07 ± 18.25), while secondary follicle proportions were significantly higher in the OT group (55.03 ± 31.68) than in non-grafted controls (9.24 ± 14.1) and the ASCs+OT group (16.74 ± 13.8). No differences were observed in AMH kinetics over time and AMH staining showed greater expression in both grafted groups, with a significant increase in secondary follicles. **Conclusion:** Although follicle development to the antral stage was demonstrated with both OT procedures, the primordial follicle reserve appears to be better preserved with 2-step OTT using ASCs. **Support:** None **Disclosures:** No conflicts of interest to disclose

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AGE AND SERUM ANTIMULLERIAN (AMH) HORMONE LEVELS AS PREDICTORS OF TIME TO RETURN TO MENSES AFTER CHEMOTHERAPY.

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Introduction: Iatrogenic ovarian damage occurs after chemotherapy for cancer and for some non-malignant conditions such as conditioning for stem cell transplant in haematological diseases. Younger age and higher pre-chemotherapy serum AMH levels are two favourable prognostic factors for ovarian recovery after gonadotoxic treatment. The aims were to determine the relationship between age and time to return to

menses after chemotherapy treatment and to determine the association between baseline serum AMH levels prior to chemotherapy and time to return to menses. **Methods:** A retrospective analysis of 145 fertility preservation patients seen at the Reproductive Services Unit of the Royal Womens' Hospital, Melbourne was performed. Stata 7.3 statistical software was used to assess pairwise correlation between time to return to menses with age and serum AMH levels. Spearman's rank correlation coefficient and p values are reported. **Results:** Majority of the cancers, were breast (46%), and Hodgkins/Non Hodgkins lymphoma (19%). The rest were bowel, gynaecological, brains and others (sarcoma, melanoma). Pairwise correlation of the baseline serum AMH with the time to return to menses suggests that patients with a higher baseline AMH will have an earlier return to menses. The Spearman's rank correlation coefficient was -0.29 and the result was statistically significant with $p=0.036$. Pairwise correlation of the age at diagnosis and the time to return to menses suggests that there was no correlation observed between age and time to return to menses. The Spearman's rank correlation coefficient was 0.13 and the result did not achieve statistical significance, $p=0.28$. **Conclusions:** There was a negative correlation between baseline serum AMH level and time to return to menses. Women with higher baseline AMH had an earlier return to menses after chemotherapy. No correlation was seen in our data between age at the time of chemotherapy and return to menses. Serum AMH levels continued to rise up to 12 months post chemotherapy. **Support:** None **Disclosures:** None

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ALTERED EXPRESSION OF YAP, PYAP, AND CTGF PROTEINS MAY PLAY A ROLE IN PRIMORDIAL FOLLICLE LOSS AFTER OVARIAN TISSUE CRYOPRESERVATION AND RE-TRANSPLANTATION

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Introduction: In the ovary, Hippo signaling specifically prevents activation of primordial follicles. Disruption of this pathway promote follicle growth via decreased phospho-YAP (pYAP) levels in concert with increased nuclear localization of YAP and leads to increased CCN growth factors like connective tissue growth factor (CTGF) which is required for normal follicle development. The aim of this study is to evaluate the expression of YAP, pYAP, and CTGF proteins after ovarian tissue cryopreservation (OTC) and re-transplantation. **Methods:** Four groups were established in rats: fresh-control (FC), frozen/thawed (FT), fresh-transplanted (T), and frozen/thawed/transplanted (FTT). After vitrification and thawing, OTs were auto-transplanted into the back muscle and grafts were harvested after 2 weeks. Expression of YAP, pYAP, and CTGF proteins was evaluated in all groups by

immunohistochemistry. Results are presented as cytoplasmic and nuclear expressions both in oocytes and granulosa cells of primordial and growing follicles. **Results:** In primordial follicles; expression of YAP increased in nucleus of granulosa cells in T and FTT groups (Fig. 1). Nuclear pYAP expression increased in FT and T groups whereas cytoplasmic and nuclear expression of pYAP decreased in FTT group (Fig. 2). CTGF expression increased in FTT group (Fig. 3). In growing follicles; nuclear YAP expression increased in FT, T, and FTT groups (Fig. 1). Cytoplasmic and nuclear pYAP expression increased in granulosa cells in FT and T groups whereas cytoplasmic expression of pYAP decreased in FTT group (Fig. 2). CTGF expression increased in FT, T, and FTT groups (Fig. 3). **Conclusions:** Expression of Hippo signaling pathway proteins are altered following OTC, particularly after re-transplantation. Therefore, disruption of Hippo pathway may lead to follicle activation and subsequent loss of follicle reserve after OTC. **Keywords:** ovarian tissue cryopreservation, transplantation, Hippo signaling, follicle reserve, fertility preservation **Support:** None **Disclosures:** The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript. The authors have declared that no competing interests exist.

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APPLICATION OF MULTISPECTRAL IMAGING (MSI) TO EVALUATE OVARIAN TISSUE RE OXYGENATION FOLLOWING TRANSPLANTATION IN A MOUSE MODEL.

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Introduction: Ovarian tissue cryopreservation and transplantation (OTCP-TP) is an experimental procedure used to treat patients with a high risk of ovarian failure after cancer treatment. However, despite its success OTCP-TP is severely limited by significant follicular loss that takes place following transplantation. Much of the post-transplant damage has been attributed to the ischemic injury that occurs during the time it takes for the ovarian microvessels to mature. Efforts to improve post transplantation revascularization have been attempted. However, as yet real-time intra-operative measurements of perfusion to evaluate these methods has not yet been established. Consequently, in situ quantification of improved tissue revascularisation remains a challenge. The aim of this study was to evaluate the feasibility of using laparoscope multispectral imaging (MSI) to monitor perfusion of oxygenated blood and quantify microvessel regeneration following

OTCP-TP in an animal model. **Methods:** The system was used to monitor OTCP-TP oxygen saturation (SaO_2) before and after transplantation. A feature-based registration algorithm was used to correct for misalignment induced by breathing or peristalsis in the tissues of interest prior to analysis. An absorption spectrum was calculated at each spatial pixel location using reflectance data from a reference standard, and the relative contributions from oxy- and deoxyhaemoglobin were calculated using a least squares regression algorithm. **Results:** To validate the use of MSI on ovarian tissue, StO_2 and total haemoglobin values were compared against follicle and vessel density, more traditional markers of evaluating OTCP-TP success. Values obtained using MSI suggested a slightly lower proportion of microvessel maturation than evident by the immunohistochemical analysis. **Conclusions:** Transplanted ovarian tissue is inherently less than 3mm thick and relatively translucent, yet the current MSI model was unable to make provision for depth resolution. This is thought to be the reason for the discrepancy between the results. Consequently, whilst promising, improved sensitivity to the model is required to allow for accurate intraoperative oxygen saturation monitoring of OTCP-TP. **Support:** None **Disclosures:** Nothing to Disclose

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BASELINE ANTI-MÜLLERIAN HORMONE (AMH) LEVELS IN YOUNG WOMEN WITH BREAST CANCER (YWBC): A COMPARISON OF THE ANSH AND THE ELECSYS® ASSAYS

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Introduction: GYPSY^a is one of 5 components of the pan-Canadian RUBY^b research for YWBC with its goal to predict the effect of treatment on their future fertility. This study evaluates the baseline AMH in YWBC using 2 different assays. **Methods:** RUBY participants had blood taken at enrollment for AMH. Samples were prepared as per the protocol. The serum was placed into aliquots, frozen, and shipped to the RUBY biobank. AMH was analysed at CRaTE Fertility Centre. One aliquot from each participant was thawed and transferred into two tubes for analysis in parallel using: a) ANSH Labs Elisa Kit using Dynex DSX, and b) Roche Elecsys® using Cobas e411 analyzers. Chi-squared, ANOVA, and Pearson correlation tests were used for analyses. **Results:** The mean age of participants at diagnosis was 35.8 ± 3.8. The mean ANSH and Elecsys® AMH results of the 238 samples are 17.7 ± 17.6 (range 0–95.3) and 15.0 ± 12.3 (range 0.1–66.7) pmol/L, respectively. A significant positive correlation of the AMH results is found for the 2 assays ($r = .97$, $p < .001$) (Figure 1) and in all age groups ($p < .001$) (Table 1).

When AMH is categorized as: “very low” (0–2.2), “low” (2.3–15.7), “medium” (15.8–28.6), “high” (28.7–48.5), “very high” (>48.5) pmol/L, 79% (188/238) of the ANSH and Elecsys® results had matched AMH categories. Among the 50 cases (21%) with assay discrepancy, 31 were in the “high” or “very high” category. For 24 samples in which the ANSH result was zero, the Elecsys® assay detected very low AMH in the range of 0.1–1.9 pmol/L. **Conclusions:** Both assays function well showing high correlation of AMH. Although a discrepancy was shown in both the “high” and “very high” categories, the clinical significance for fertility preservation (FP) counselling in YWBC is low. The increased sensitivity of the Elecsys Assay in the “very low” category may assist YWBC’s FP decisions. **Support:** None **Disclosures:** Nothing to Disclose drglass@createivf.com

BIRTH DEFECTS RISK FACTORS: A MULTI-CENTERED CASE-CONTROL STUDY IN EASTERN CHINA

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Introduction: Every year, birth defects (BDs) affect hundreds of thousands of babies and families in China. Chinese government has paid high attention to prevention and treatment of BDs, especially since the relaxation of one child policy. This study investigated the risk factors of BDs under the new two child policy in China. **Method:** A case-control study was conducted in four women and children’s hospitals in Zhejiang Province, China. All BDs (including live births, early fetus losses, still births, and infant deaths) identified during Jan. to Mar. 2019 were recruited. The control group were births without BDs and matched by maternal age, birth hospital and birth day. The association between risk factors and BDs occurrence was assessed by adjusted odds ratios (aORs) with conditional logistic regression model adjusted for potential confounders. AORs for BDs were calculated for socioeconomic status, pre-pregnancy BMI, parity, gravidity, smoking, pre-existing diseases and conditions in pregnancy. **Result:** There were 398 BDs and 398 without controls included in the analysis. Every standard deviation increase in pre-pregnancy BMI was positively associated with BDs in offspring (aOR 1.29, 95%CI 1.08 to 1.54). Compared to normal weight, overweight and obesity were both positively associated with BDs in offspring (overweight: aOR 1.91, 95%CI 1.16 to 3.17; obesity: aOR 2.15, 95%CI 1.19 to 3.90). The risk for BDs was significantly associated with maternal report of flu

(aOR 2.95, 95%CI 1.19 to 7.33) and maternal exposure to housing renovations (aOR 14.70, 95%CI 1.71 to 126.76) from 3 months before pregnancy to end of first trimester. **Conclusion:** Higher pre-pregnancy BMI, maternal report of flu and maternal exposure to housing renovations during the period of 3 months before pregnancy to end of first trimester may increase the risk of giving birth to fetuses with BDs. However, large-population research is needed to clarify the effects seen here. **Support:** This work was supported by grants from the Cyrus Tang Foundation [419600-11102], the China Medical Board (CMB) Collaborating Program [12-108 and 15-216], and the National Key Research and Development Plan [2018YFC1002700 and 2018YFC1002702]. **Disclosures:** None

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CHALLENGES IN A FERTILITY PRESERVATION UNIT

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Objective: to describe the difficulties of accessing to a proper treatment of fertility preservation and their possible solutions. **Methods:** We included 274 consecutive patients assisted in our programme in order to preserve their reproductive potential in the last 5 years. All of them, received assessment by an oncofertility specialist prior to a gonadotoxic treatment. The setting corresponded to the tertiary center Hospital Italiano de Buenos Aires. Fortunately, the number of patients who access to the full strategies proposed, increased every year. Nevertheless, there is still a constant proportion of near the 20% of patients, who couldn’t achieve a fertility preservation treatment, even when they were properly counselled. During these years of follow up, 20.63% of the patients did not complete any treatment. **Results:** When we analyze this subgroup, we found that 32.6% did not carry out any treatment for personal decision, 19.6% couldn’t access for poor clinical condition, 19.6% for health insurance issues, 15.2% for late derivation and in 13% we did not found the reason. These subgroups show evitable reasons and need to perform different approaches to avoid lost of opportunities in fertility preservation. One of them is intensify the follow up of the patients, try to resolve health insurance problems, and optimize the derivations protocols in each case. **Support:** None **Disclosures:** We have no conflicts of interest mariano.uzal@hospitalitaliano.org.ar

CHALLENGING CHOICES: UNDERSTANDING THE UNIQUE FERTILITY BARRIERS FOR ONCOLOGIC PROVIDERS IN LOW RESOURCE SETTINGS

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Introduction: The American Society of Clinical Oncology guidelines recommend that oncologic providers address potential treatment-related infertility early in the course of diagnosis and treatment planning and refer all interested/uncertain patients for fertility preservation counseling prior to initiation of cancer treatment. Pre-treatment discussion of fertility preservation improves quality of life, lowers decisional regret, and promotes informed decision-making. Although barriers to fertility counseling have been previously identified in the general population (time constraints, poor knowledge, and demographic variables), little is known about barriers encountered in low-resource settings. **Methods:** Semi-structured interviews with oncology providers caring for reproductive-age women with cancer at a safety-net oncology clinics were conducted by a trained interviewer. Interviews assessed 1) current practices, 2) unmet needs and barriers, and 3) recommendations for resources and tools. Participants were gynecologic and medical oncology providers at Lyndon B. Johnson Hospital. **Results:** Participants included 13 physicians and 2 advanced practice providers (9 female, 6 male). Dominant themes identified included: barriers to discussion (lack of financial support, low literacy, fear of causing distress), gender inequality (men having more consistent and streamlined access to counseling and services, and need for support tools/resources for both patients and providers (provider didactic session, tailored written handouts for patients, and well-defined referral protocols). **Conclusion:** Oncology providers in low-resource settings also face multiple challenges to discussing fertility preservation. Provider and patient education materials that are tailored to address the unique needs of this population are needed to optimize opportunities for discussion and referral. **Support:** None **Disclosures:** None

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COMPARING GYNECOLOGICAL CANCERS AND POTENTIAL NEED FOR FERTILITY PRESERVATION IN A TERTIARY INNER-CITY SAFETY-NET HOSPITAL TO NATIONAL AVERAGES

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Introduction: Fertility preservation (FP) in reproductive aged women with gynecological cancers is a critical aspect of healthcare. Our community is dependent upon government assistance, thus limiting ART. We compared our community rates to national averages regarding who could be candidates for FP/ART. **Methods:** De-identified data from Tumor Registry was analyzed. Women <27 years old (community average age of conception) and age <35 with cervical, endometrial, ovarian, and uterine cancer were defined as “probably” and “possibly” candidates for FP. Stage 1 diagnoses were used as surrogates to represent patients “possibly” benefiting from FP. Data was then compared to SEER, determining if our

community was reflective of national data. **Results:** Between 2014–18 there were a total of 810 cases. Of these, 375 (46.3%) were cervical (including CIS), 203 (25.1%) endometrial, 88 (10.9%) ovarian, and 30 (3.7%) uterine. For cervical 10.9% of these patients were <27, 46.9% were <35; for endometrial, 0.49% <27 and 3.0% <35; for ovarian, 1.1%, <27 and 6.8% <35; for uterine, no cases under 27 or 35 were noted. For women <35, SEER data rates were: 13.5% cervical (including CIS), 5.3% ovarian, and 1.5% “uterine” (combined endometrial/uterine). Of 495 cancers with known staging, 328 (66.3%) were stage 0 or 1; with cervical= 251 (76.5%); endometrial =66 (20.2); ovarian =7 (2.1%); and uterine =4 (1.2%) **Conclusion:** Local rates of “probable” or “possible” FP candidates with cervical cancer were higher than nationally; with similar rates for ovarian and uterine cancers. The proportion of women with cervical cancer <35, highlights needed FP for this group. The majority of women (66.3%) had early stage cancers, suggesting better success rates and potential of FP although we recognize all patients may benefit from these treatments. **Support:** None **Disclosures:** None

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CONTROLLED OVARIAN STIMULATION IN BREAST CANCER PATIENTS: DOES RECEPTOR STATUS MAKE A DIFFERENCE?

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Introduction: Fertility preservation (FP) protocols include tamoxifen for controlled ovarian stimulation (COS) in patients with estrogen receptor-positive (ER+) breast cancer, to reduce the breast tissue exposure to estrogen. The aim was to evaluate COS outcomes, such as oocyte yield, mature eggs frozen, recombinant FSH dose and duration of stimulation between the ER+ and estrogen receptor negative (ER-) groups, due to tamoxifen exposure. **Methods:** In this retrospective cohort analysis, 204 breast cancer patients (146 ER+, 58 ER-) presenting for FP between 2013–2018 were studied. Eighty of these patients underwent embryo freezing, while mature egg freezing was performed in the remainder (124 patients). The patients underwent antagonist cycles, with tamoxifen administered for the duration of stimulation in the ER+ group. **Results:** The patients had a mean age of 35.1 and 33.3 years in the ER+ and ER- groups. We observed no significant differences in the ER+ and ER- groups, with respect to mean eggs collected (13.2 and 11.8, respectively), FSH starting dose (253.3 and 251.9 IU), duration of FSH (13.8 and 13.2 days). Interestingly, there was a statistical difference in the mean number of mature eggs frozen (12.3 and 9.4, p-value 0.05), favouring the ER+ group and cannot be attributed to age. The mean number of embryos frozen was the same for the two groups (4.9 and 4.6). **Conclusions:** While a higher number of mature eggs were frozen in the ER+ group, for those patients undergoing embryo freezing, the mean number of embryos frozen were the same for the two groups. COS outcomes

were similar for patients with breast cancer, irrespective of receptor status and co-treatment with tamoxifen. **Support:** None **Disclosures:** None

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CURRENT STATUS OF FERTILITY PRESERVATION FOR CHILD AND ADOLESCENT CANCER PATIENTS IN ASIAN COUNTRIES

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Introduction: Although “fertility preservation (FP)” has become popular as a new field of reproductive medicine. However, FP for child and adolescent (C-A) patients is still developing, even in advanced countries. The aim of present study was to assess the barriers to FP for C-A patients by investigating the current status of FP for C-A patients in Asian countries, which just have started FP activities. **Method:** A questionnaire survey of founding members of the Asian Society for Fertility Preservation (ASFP) was conducted in November 2018, under permission of IRB-SMU. **Results:** Of the 14 countries, 11 representatives replied to this survey. FP for C-A patients is still developing in Asian countries, even in Australia, Japan, Korea, and India, which have organizations specialized for FP. In all countries that replied to the present survey, the patients can receive embryo cryopreservation, oocyte cryopreservation, and sperm cryopreservation as FP option. While, testicular tissue cryopreservation is an uncommon option because of its still extremely experimental status (7 of 11 countries provide it). Most of countries can provide FP for C-A patients in terms of medical technology, but most insufficient experiences and an unestablished system due to barriers that inhibit promoting. In particular, “How to provide FP treatment for C-A” is a major barrier. Also, low recognition in society and among medical staffs are major issues. There is also big problem with cooperative systems with pediatricians. To achieve high-quality FP for C-A

patients, a multidisciplinary approach is vital, but, according to the present study, few paramedical staff can participate in FP for C-A patients in Asia. **Conclusion:** The present study demonstrated the developing status of FP for C-A patients in Asian countries. More intensive consideration and discussion are needed to provide FP based on the local cultural and religious needs of patients. **Support:** None **Disclosures:** We don't have any COI on this research. Also we performed this investigation under permission of IRB committee of SMU.

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DECISION MAKING AND MOTIVATIONS FOR FERTILITY PRESERVATION AMONG TRANSGENDER MEN AND WOMEN

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Introduction: Patients undergoing gender reassignment therapies are offered the possibility to undergo fertility preservation (FP) to allow biological parenthood, although studies have documented low use of these procedures. This study analyzed the motivations to undergo FP in transgender patients referred for FP counselling. **Methods:** Since 2018, 27 transgender patients were referred for FP counselling. Motives for choosing to undergo or to decline FP were assessed. **Results:** Of the 27 patients who received FP counselling, 9 were transgender women (TW) and 18 were transgender men (TM) and 16 (59%) had already started hormonal therapy (HT, $M = 2.9$ years, $SD = 2.00$, 6 months to 7 years). Of the 27 patients, 14 (51.9%) decided to perform FP techniques, 12 (44.4%) had no desire or intention to perform FP and 1 (3.7%) had not yet made a decision. There were no significant differences in the decision between patients undergoing or not HT at the time of FP ($p = .303$). No differences were found between TM and TW regarding FP decision ($p = .416$). However, among TW ($n = 6$) who decided to undergo FP, those who were on HT ($n = 3$) failed to collect sperm. The main reasons for performing FP were 1) the enhancement of genetic ties in parenting ($n = 11$, 40.7%) and 2) keeping this option open ($n = 4$, 26.7%). The main reasons for not wanting to perform FP were 1) not valuing the biological ties in parenting ($n = 3$, 11.1%), 2) fears related with the procedures ($n = 2$, 7.4%) 3) not wanting to postpone or retrocede in gender transition ($n = 5$, 18.5%) and 4) envisioning difficulties in accomplishing parental roles using FP ($n = 1$, 3.7%). **Conclusions:** The discussion of FP options in transgender patients is a key component in gender reassignment and should be considered and

discussed with patients as early as possible to enable informed and thoughtful decision making. **Support:** None **Disclosures:** No conflict of interest to declare

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DEVELOPMENT OF A HYDROGEL FROM DECELLULARIZED BOVINE OVARIAN TISSUE FOR MOUSE OVARIAN FOLLICLE SURVIVAL IN VITRO

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Background: One of the main requirements for an artificial ovary is creation of a biomimetic 3D matrix to encapsulate isolated ovarian follicles and cells. Our goal was to develop a hydrogel based on bovine ovarian decellularized matrix (boECM) to support survival and development of isolated mouse preantral follicles. **Methods:** Bovine ovarian tissue was decellularized and lyophilized before hydrogel preparation and characterization. Preantral follicles (n=235) were isolated from mouse ovaries (n=3) and in vitro-cultured in the boECM hydrogel. Seven days later, they were investigated for viability and growth. Decellularization was evaluated by quantification of dsDNA (Quant-iT™ PicoGreen). Hydrogel ultrastructure and gelation kinetics were assessed by scanning electron microscopy (SEM) and rheology (storage [G'] and loss [G''] modulus measurement) respectively. Glycosaminoglycans (GAGs) were determined using the 1,9-dimethyl-methylene blue (DMMB). Collagen content was calculated by quantifying hydroxyproline. **Results:** Based on dsDNA concentrations (8.6±0.17 ng/mg), we confirm that decellularization was successful. Regarding hydrogel characterization, G' and G'' of the pre-gel solution increased ~ 5 minutes after the temperature rose, indicating the start of gelification. G' was always higher than G'', reflecting formation of a solid hydrogel. Hydrogel samples showed a fibrillary structure with homogeneous fiber size (SEM). GAG content was 0.3±0.03% and collagen content 89.49±5.82 %, comparable to human decellularized ovarian cortex (88–98%). After in vitro culture, 80% of follicles were recovered and 70% of them were viable. Mean follicle diameter did not change during culture (44.9±22.0 µm on D0 and 43.2±21.2 µm on D7). **Conclusions:** Our results showed that it is possible to develop a boECM hydrogel able to support follicles in vitro.

Interestingly, this matrix does not appear to induce large-scale follicle activation and growth, which is usually the case after culture or grafting. However, more studies are needed to understand these findings and their impact on folliculogenesis.

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DISCORDANT RESULTS OF MINIMAL RESIDUAL DISEASE BETWEEN CRYOPRESERVED CORTEX AND MEDULLAR OVARIAN FRAGMENTS COLLECTED IN PATIENTS WITH ACUTE LEUKEMIA IN REMISSION

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Introduction: Cryopreserved ovarian tissue transplantation (OTT) could restore ovarian function and lead to natural pregnancies and births. OTT is rarely performed in patients cured for leukemia due to the potential infiltration of ovaries by leukemic cells. We reported the evaluation of minimal residual disease (MRD) in cryopreserved ovarian cortex and medulla harvested in leukemic patients in remission. **Methods:** Patients with acute leukemia in remission after chemotherapies who underwent ovarian tissue cryopreservation (OTC) before hematopoietic stem cell transplantation as part of fertility preservation program were included. MRD was assessed by quantitative PCR of clonal rearrangements of immunoglobulin/T-cell receptor genes or oncogenic fusion genes in a cortical and/or a medullar fragment and in bone marrow (BM) at the time of ovariectomy. Consents were obtained. Institutional review board approved the study. **Results:** Thirty-six patients were included (32 acute lymphoblastic leukemia, 4 acute myeloid leukemia). Eight patients (22%) had

positive MRD in ovarian samples (8 ALL). On the 24 patients tested in cortex and medulla: 18 were negative in both, 3 positive in both and 3 discordant with positive medulla and negative cortex. MRD in BM was available in 30 patients and positive in 30%. Eight patients (27%) had discordant results between ovarian samples and BM: 4 with positive MRD in ovarian samples while negative in BM and vice-versa.

Conclusions: Our study underlies the interest of performing OTC in leukemic patients in remission to decrease the risk of leukemic infiltration in ovarian samples (22% versus nearly 60% in all published cases, mostly with OTC at diagnosis) and possibly allow OTT for a greater number of patients. MRD in the BM doesn't systematically predict infiltration of ovarian tissue. Discordant MRD results may exist between cortex and medulla. Consequently, MRD evaluation in both a cortex and a medullar fragment seems necessary for a better risk assessment. **Support:** None **Disclosures:** No disclosure

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DISSEMINATION AND IMPLEMENTATION OF A FERTILITY PRESERVATION DECISION AID WEBSITE FOR WOMEN WITH CANCER: RECOMMENDATIONS FROM TEN EXPERTS IN THE FIELD

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Introduction: The American Society for Clinical Oncology guidelines recommend discussing fertility preservation with all reproductive-age women with cancer; however, adherence remains suboptimal. The *Pathways* fertility preservation patient decision aid provides information and support to help women make well-informed decisions about fertility preservation. Studies show it successfully improves women's fertility preservation knowledge, decisional conflict, and satisfaction with decisions; however, concerns arose about the feasibility and dissemination when scaled across varying types of cancer care sites. **Objective:** To assess practice patterns and recommendations for feasibility and scaling of *Pathways* at diverse cancer care sites across the United States. **Methods:** Oncologists were recruited through the Cancer Care Delivery Research (CCDR) alliance. A trained interviewer provided participants with a summary of the *Pathways* patient decision aid research protocol and conducted semi-structured phone interviews assessing: 1) existing practice patterns, and 2) recommendations for feasibility and scalability at their site. **Results:** Oncologists (n = 10) responded from 9 states, including 5 public hospitals, 3 private hospitals, and 2 comprehensive cancer centers. All oncologists supported the importance of fertility preservation and the potential usefulness of a patient decision aid delivered

alongside the consultation. Oncologists confirmed potential feasibility but noted three consistent barriers – awareness, time, and assumed costs. Top recommendations included: 1) the provider introducing the tool in person first, 2) a member of the care team follow up by phone/email with details, and 3) also distributing pamphlets in person for visual reinforcement.

Conclusion: Fertility preservation remains an important, but often difficult, topic to introduce across many cancer care settings. Dissemination of a patient decision aid website may be feasible and scalable, provided it is introduced by the provider, facilitated by a navigator, and viewed after the consultation time. **Support:** None **Disclosures:** The authors declare they have no conflict of interest.

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DOES EE/CPA INCREASE BREAST CANCER RISK FOR WOMEN WITH PCOS UNDERGOING IVF?

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Introduction: Progesterone receptor membrane component 1 (PGRMC1) has been found to be highly expressed in the tissue of breast cancer patients. In our previous studies, Certain synthetic progestin can stimulate the proliferation of breast cancer cells which express PGRMC1 in vitro and in vivo. In this study, we aim to investigate the expression of PGRMC1 in PCOS patients treated with IVF, and estimate the risk of breast cancer in PCOS patients undergoing IVF.

Methods: According to whether they were PCOS patients and level of estrogen, patients were classified, 14 cases in group A (PCOS patients with high level of estrogen, HCG day), 8 cases in group B (PCOS patients) and 42 cases in group C (non-PCOS patients with high level of estrogen, HCG day). Group D: 28 cases (control group). There are 18 cases in EE/CPA pretreatment group and 38 cases in control group. The concentration of PGRMC1 was measured by ELISA. **Results:** The level of PGRMC1 in group A was significantly higher than that in group B. The level of PGRMC1 in group C was significantly higher than that in group D. There was no significant difference in the level of PGRMC1 between group A and group B. There was no significant difference in the level of PGRMC1 between group B and group D. There was no statistical difference of PGRMC1 between EE/CPA pretreatment group and control group. **Conclusion:** There is no significant difference in level of PGRMC1 between PCOS patients and control group. The

level of PGRMC1 increased at high level of E2. Short-term use of EE/CPA may not increase the risk of breast cancer in IVF patients, but breast cancer risk should be vigilan by patients with high level of estrogen. **Support:** This work was supported by the National Natural Science Foundation of China (grant number 81671411). **Disclosures:** None

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EFFECT OF PELVIC ARTERY EMBOLIZATION FOR POSTPARTUM HEMORRHAGE ON SUBSEQUENT REPRODUCTIVE FUNCTION

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Introduction: Pelvic artery embolization (PAE) for treatment of postpartum hemorrhage (PPH) can remain the uterus and preserve reproductive function. We review the effect of PAE for PPH on fertility preservation, including menstruation, ovarian preservation and subsequent pregnancy outcomes. **Methods: Results:** About 91% - 100% patients with an average rate of 93% after PAE for PPH had menstrual recovery. Few articles reported acute ovarian failure and uterine stroke after PAE. The subsequent pregnancy rate is generally low after PAE for PPH. A 38 literatures review including 1072 cases treated with PAE shown the pregnancy rate was 23.2%. Endometrial ischemic injury caused by PAE may be the cause of subsequent low pregnancy rate. Patients do not want to conceive or fear PPH again may contribute the low pregnancy rate, so the low pregnancy rate may be underestimated to the actual pregnancy rate. **Results:** The recurrence rate of PPH in the subsequent pregnancy after PAE was significantly high with an average rate of 14-23.3% and 16.7% with abnormal placenta needing undergoing hysterectomy. However, the effect of PAE on placenta formation in subsequent pregnancies remains unclear. It has been suggested that PAE may lead fetal growth restriction, but Fiori reported that 60% of pregnant women can deliver healthy, normal-weight newborns through the vagina after PAE. Soro reported a study including 61 newborns after PAE, with an average birth weight of 3250g and an average gestational age of 38 weeks. Data indicate that PAE has no direct effect on placental blood supply and fetal growth in subsequent pregnancy. **Conclusions:** PAE does not affect the recovery of menstruation, but the subsequent pregnancy rate is generally low. The incidence of PPH in subsequent

pregnancy is significantly higher than that in normal population. In addition, there is no significant correlation between PAE and fetal growth restriction in subsequent pregnancies. **Support:** None **Disclosures:** Authors have no conflict interest for this paper.

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EFFECT OF REPEATED VITRIFICATION AND WARMING PROCEDURES ON BLASTOCYSTS DERIVED FROM FROZEN OOCYTES AND PREGNANCY OUTCOMES

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Introduction: This study compares fresh versus frozen blastocyst transfer in embryos resulting from frozen oocytes to address if repeated vitrification and warming procedures affect clinical outcomes. It is a multicenter retrospective study. **Methods:** 472 frozen oocytes were warmed and ICSI was performed within 4 hours post warming. Surplus blastocysts were cryopreserved by vitrification. Blastocyst transfer was done with either fresh blastocyst(s) or frozen blastocyst(s). For pregnancy outcome, 30 fresh and 14 frozen blastocyst transfer cycles were analyzed. The statistical analysis was performed using t-test, chi-square test or Fisher's exact test. A *p* value of <0.05 was considered statistically significant. **Results:** The survival rate of frozen oocytes was 80.9%, fertilization rate was 84.1%, and blastocyst rate was 45.8%. Pregnancy outcomes are summarized in Table 1. There were no significant differences between fresh versus frozen blastocyst transfer in embryos resulting from frozen oocytes. **Conclusions:** In fertility preservation or oocyte donation cycles, repeated vitrification and warming procedures may be required for frozen oocytes and the embryos which result from them. There is no detectable effect in this retrospective study of fresh versus frozen blastocysts resulting from warmed vitrified oocytes on multiple markers of clinical IVF success including ongoing pregnancy/live birth rates. **Support:** None **Disclosures:** Nothing to disclose

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EVALUATING THE EFFECT OF CHAMOMILE EXTRACT IN COMPARISON WITH AMPICILLIN ON REPRODUCTIVE HORMONES IN MALE MICE TREATED WITH E. COLI LIPOPOLYSACCHARIDE

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Introduction: The objective of this study was to evaluate the effects of chamomile extract in comparison with ampicillin on parameters of the reproductive system in male mice treated

with *E. coli* LPS. **Methods:** In this study, 65 adult male mice were divided into 6 groups. Control, LPS, LPS+ampicillin, LPS+chamomile extract 50 mg/kg, LPS+chamomile extract 100 mg/kg and LPS+chamomile extract 200 mg/kg. IP injections were performed for 20 days and the fertility of each group was evaluated. The results of hormonal tests of each group were compared with the control group. **Results:** The results showed no significant difference between the mean FSH level of the treatment groups and that of control group ($p < 0.05$). The mean LH level of LPS group decreased significantly compared to that of control group ($P \leq 0.05$). The mean level of testosterone in chamomile extract group 200 mg/kg and ampicillin group was significantly higher than that in control group ($p \leq 0.05$). **Conclusions:** Given these results, while LPS can reduce male fertility potential, treatment of samples with chamomile extract in all three doses of 50, 100 and 200 can reduce the effects of LPS and increase the fertility potential. It seems that the use of dose-dependent chamomile extract, as an anti-bacterial, anti-inflammatory and anti-oxidant agent, to be effective in improving infection compared to ampicillin antibiotic and in increasing male sex hormones.

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EXAMINATION OF OVARIAN STIMULATION BY THE RANDOM START METHOD IN CANCER PATIENTS IN OUR HOSPITAL

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Introduction: Recently, controlled ovarian stimulation (COS) has been performed by a random start (RS) method for oocyte and embryo cryopreservation in cancer patients. To obtain sufficient oocytes, COS may be performed in a continuous cycle. Though it was started from the fifth day of oocyte retrieval (OCR) in the past report, the second start time has not become clear. We retrospectively examined. **Methods:** From August 2017 to December 2018, 13 patients underwent COS by RS method, and oocytes and embryos were frozen. The mean age was 33.9 years. OCR was performed twice in a cycle in four patients. Each stimulation was examined. **Results:** The first COS was performed over 9.0 days and the second over 11 days. The dose of HMG was higher in the second COS (1253 mIU/ml vs 1860 mIU/ml). The number of OCRs was 15.2 and 12.0 for the first and second time, respectively. However, when COS was conducted twice separately, the number of oocytes collected after the first cycle was 6.4, while that after the second was 12.0. In cases of ovarian hyperstimulation, the second round of stimulation was initiated 4 days after the first OCR, and a 15-day COS period was required before the second OCR. Therefore, if the second COS was initiated on day 11 after the first OCR, 11.5 days were

required before the second OCR. In contrast, in cases of mild stimulation, OCR could be performed 6 days after the second COS, even if it was performed on the 1st day after the 1st OCR. **Conclusions:** The COS period and injection volume for the second COS was larger than that of the first. By identifying the optimum stimulation start time, the second injection volume can be reduced and a sufficient number of oocytes can be obtained from cancer patients. **Support:** None **Disclosures:** The authors have no financial conflicts of interest to disclose concerning the study.

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EXAMINATION OF THE USEFULNESS OF OOCYTES COLLECTED AT THE TIME OF OVARIAN FREEZING AS A FERTILITY-PRESERVING THERAPY

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Introduction: Cryopreservation of oocytes, embryos, and ovarian tissue is being implemented as fertility-preserving therapy for young cancer patients. Although ovarian freezing has the advantage of preserving many temporary follicles in a short time period, the possibility of cancer cell contamination cannot be denied. Oocytes may be collected at the time of ovarian freezing treatment; if oocytes can be used, there is no possibility of transplantation of cancer cells. Herein, we investigated the number and maturation rate of oocytes that could be recovered at the time of ovarian freezing, to examine the possibility of a low-risk fertility-sparing therapy. **Methods:** We included 7 patients who had ovarian freezing at our hospital from January 2018 to December 2018. One side of the ovary was removed by laparoscopy, and if the follicle was confirmed, follicular fluid and the oocyte were collected. The ovarian freezing treatment was performed in m-HTF solution, and the remaining oocytes were collected. The collected oocytes were determined to be matured or immature, and the matured oocytes were frozen. The immature oocytes were cultured in IVM medium for a maximum of 48 hours and frozen after confirming maturation. **Results:** The number of oocytes recovered was 5.0 ± 6.4 (0–20). The number of matured oocytes immediately after collection was 0, and the number of eggs matured by 48 hours after collection was 2.6 ± 3.2 (up to 10). Moreover, oocytes could be recovered from an 8-year-old prepubertal patient and matured oocytes could be frozen. **Conclusions:** The pregnancy rate per oocyte is said to be 4.5–12%; in some patients, the number of oocytes required for pregnancy could be frozen. In addition, this technique is thought to be useful as a minimally invasive fertility-preserving treatment because mature oocytes could be frozen even in prepubertal patients. **Support:** None **Disclosures:** The authors have no conflicts of interest directly relevant to the content of this article.

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FERTILITY OUTCOMES IN WOMEN WITH ATYPICAL ENDOMETRIAL HYPERPLASIA AND ENDOMETRIAL CANCER GRADE I

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Introduction: In available literature there is no generalized information about condition of the reproductive system of young women suffering from cancer, management tactics and the time to restore the implantation properties of the endometrium after the treatment. The objective was the presentation of the first experience treatment of patients with endometrial cancer grade I or atypical endometrial hyperplasia directed to preserve oocytes or embryos. **Methods:** 77 women with endometrial carcinoma grade I or atypical endometrial hyperplasia, directed by oncologist for fertility preservation or pregnancy achievement. Among 28 women, performed IVF programs, 6 of them underwent ovulation stimulation, collection and cryopreservation of oocytes / embryos for the delayed reproductive function, after which the treatment of the underlying disease was started or continued. Twenty patients underwent an IVF program with embryo transfer in the “fresh cycle” after completing the treatment of cancer and achieving stable remission. 37 patients were excluded from the study due to different medical issues. **Results:** PCOS with severe endocrine-metabolic is diagnosed in 51% of cases, which makes it necessary to develop programs aimed at preserving reproductive material and treatment methods that reduce the risk of cancer recurrence. It was shown that the best results were obtained with preliminary cryopreservation programs in patients with normal ovarian reserve, also it has been demonstrated that IVF programs 3–4 months after completion of treatment are associated with better outcomes. **Conclusions:** There is an increasing need for rehabilitation of the reproductive function of patients with atypical hyperplasia and stage I endometrial cancer, which makes this problem relevant. It is advisable to carry out preliminary cryopreservation of oocytes/embryos before treatment of the underlying disease due to the time spent on treatment, especially with initially reduced rates of ovarian reserve, fewer and worse quality of oocytes obtained after completion of treatment. **Support:** None **Disclosures:** Presentation of the first experience treatment of patients with endometrial cancer grade I or atypical endometrial hyperplasia directed to preserve oocytes or embryos.

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FERTILITY PRESERVATION IN PATIENTS WITH BREAST CANCER

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Objective: To address the preservation of fertility in patients with breast cancer. **Methods:** We retrospectively investigated the medical records of 45 patients with breast cancer who visited our institution for fertility preservation from February 2011 to May 2019. Random-start therapy with letrozole was used. Women aged >40 years were excluded from the protocol of fertility preservation. **Results:** The median age of the 45 patients was 36 years (range, 22–44 years). Oocyte or embryo cryopreservation was performed in 30 patients (66.7%), and 15 did not choose fertility preservation because priority was given to the treatment of breast cancer. The 11 women who were >40 years received counseling, and five (age range, 40–42 years) received fertility-preservation therapy. The median number of oocytes/embryos that could be cryopreserved was 2 (range, 0–6). In two patients, embryo transfer was performed after cancer treatment, but it failed to result in pregnancy. Total 10 patients (33%) who underwent oocyte/embryo cryopreservation before neoadjuvant chemotherapy included six, two, and one patient in cancer stages I, II, and IV, respectively. The cancer subtype was triple negative in four patients and luminal in six. The median number of days from the first visit to oocyte retrieval was 24 days (range, 13–59 days), and the median observation period after oocyte retrieval was 14 months (range, 3–50 months). One woman died after the main treatment. Two patients with polycystic ovarian syndrome (PCOS) and low estrogen value because of letrozole therapy developed severe OHSS and required hospitalization for 5 and 9 days, respectively. **Conclusions:** The administration of fertility-preservation therapy before neoadjuvant chemotherapy is generally unacceptable; therefore, their indications require thorough discussion with an oncologist. It should be noted that breast cancer patients with PCOS can develop severe OHSS, even in the presence of a low E2 value from letrozole therapy. **Support:** None **Disclosures:** There are no conflicts of interest.

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FERTILITY PRESERVATION OF TWO BREAST CANCER PATIENTS WITH BRCA GENE TEST MUTATIONS

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Introduction: In our clinic, fertility preservation with oocytes and embryos has been performed since 2003, and our first ovarian tissue preservation was performed in 2017. Recently, we experienced fertility preservation in two breast cancer patients who were found to have mutations in the BRCA gene test. Here, we present case reports of each preservation. [Case 1] AI, 36 years old, single. **Methods:** She was diagnosed with stage 1 right-side breast cancer; hormone receptor positive; HER2 negative; Ki-67, 47%; BRCA1 negative; and BRCA2 positive. Surgery was completed without lymph node dissection. Pathological results showed anticancer drugs were unrequired. Hormonal therapy was given with tamoxifen and leuporelin. Family History: father, prostate cancer; mother, cancer; siblings, younger brother. Fertility preservation: She started random-start ovarian stimulation, with uMG 150 IU and uFSH 150 IU for 11 days combined with luteal and letrozole. Lucrin 1 mg and uCG 5000 IU were administered after 10 days as a trigger. **Results (case 1):** After egg retrieval, 23 MII oocytes were cryopreserved. [Case 2] NI, 35 years old, married. Treatment: She was diagnosed with stage 1 right-side breast cancer; triple negative; Ki-67, 30.9%; BRCA1 positive; and BRCA2 negative. Family history: mother and aunt, breast cancer; grandmother, possibly cancer; siblings, older brother, older sister (2 daughters). Fertility preservation: She started random-start ovarian stimulation using uMG 150 IU and rFSH 150 IU for 7 days. Lucrin 1 mg and uCG 5000 IU were administered as a trigger after 6 days. **Results (case 2):** After egg retrieval and ICSI, 3 embryos of D2 and 1 blastocyst of D5 were cryopreserved. Breast cancer surgery is scheduled for the near future. [Conclusion] Fertility preservation was performed in 2 cases with BRCA mutation. Genetic counseling, including BRCA genetic testing for their siblings and nieces will be required for further study. **Support:** None **Disclosures:** The authors declare no conflicts of interest associated with this manuscript.

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FIRST ONGOING PREGNANCY FOLLOWING FROZEN-THAWED OVARIAN TISSUE RE-TRANSPLANTATION IN TURKEY IN A PATIENT WITH CURED ACUTE LYMPHOBLASTIC LEUKEMIA

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Introduction: Infertility, a major side effect due to cytotoxic treatments, is most commonly manifested by premature ovarian failure (POF) and early menopause. Unlike oocyte freezing, ovarian tissue cryopreservation (OTC) allows the preservation of hundreds of primordial follicles at once, without the need for controlled ovarian hyperstimulation (COH). In pre-pubertal girls, as there is no other

option for fertility preservation (FP), OTC is the gold standard procedure. Here, we would like to present the first ongoing pregnancy in Turkey following frozen-thawed ovarian tissue re-transplantation, after hematopoietic stem cell transplantation (HSCT) due to acute lymphoblastic leukemia (ALL-L2, Ph+). To the best of our knowledge, it is also the second reported pregnancy in the world for ALL. **Methods:** Patient was referred to our center in April 2010 for FP. A year before her consultation she had received vincristine, daunorubicin, L-asparaginase, methotrexate, and afterward 6-mercaptopurine, cyclophosphamide and imatinib were added to the BFM protocol. At her referral, serum AMH level was measured 1.5 ng/ml and 15 antral follicles were counted on ultrasound. Unilateral ovarian wedge resection was performed by laparoscopic surgery and $\frac{3}{4}$ of the left ovary was removed. Following tissue dissection, 0.5x1 cm-sized, 20 cortical strips were slow frozen. After HSCT, the patient developed POF with amenorrhea and an FSH rise up to 179 mIU/mL. Seven years after the initial diagnosis, she applied to our center for pregnancy. Upon decision to re-transplant frozen ovarian tissues, one of the cortical strips was thawed in advance, to uncover any residual cancerous cells. No malignant contamination was detected in sample tissue through immunohistochemistry and molecular qPCR analysis. Therefore, nine additional cortical strips were thawed and re-transplanted orthotopically in December 2017. **Results:** In her second attempt for COH-IVF she achieved pregnancy using frozen thawed embryo; a singleton ongoing healthy pregnancy is on the 24th week of follow-up without any complications. **Support:** None **Disclosures:** None

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FOLLICLE POPULATIONS AND VASCULARIZATION IN PREPUBERTAL OVARIAN TISSUE

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Introduction: Our aim was to characterize cryopreserved ovarian tissue from prepubertal patients, focusing on follicle populations, ovarian vascularization and follicle development after long-term xenografting. **Methods:** Frozen-thawed ovarian tissue fragments from 7 prepubertal girls (age 1–10 years) and 7 women (age 20–35 years) were xenografted to immunodeficient mice for 20 weeks. Ovarian tissue pieces (frozen-

thawed controls and grafts) were processed for follicle classification and evaluation of ovarian vessel density and endothelial area by double immunofluorescence for anti-Von Willebrand factor and anti-smooth muscle actin. **Results:** In the prepubertal group, quiescent-stage follicles (primordial and intermediate) made up the largest proportion of the follicle pool (71.2%) in non-grafted tissue, and also after grafting (69.6%). Moreover, growing follicles at the final stage of development (secondary and antral) were significantly more numerous after grafting than in non-grafted tissue (secondary follicles: 0% vs 15.6%, $p=0.004$; antral follicles: 0% vs 2.2%, $p=0.01$; before and after grafting respectively) [Figure 1]. Abnormal and atretic follicles were also detected in non-grafted tissue from prepubertal patients, but their numbers decreased after grafting (abnormal follicles: 11.5% vs 4.2%, p value not significant; atretic follicles: 16% vs 5.2%, $p=0.009$; before and after grafting respectively). Interestingly, a negative correlation was found between the number of abnormal ($p=0.02$, $r=0.60$) and atretic ($p=0.02$, $r=0.38$) follicles and age [Figure 2]. Concerning vascularization, more immature vessels were observed in prepubertal ovarian tissue than in adult ovarian tissue ($p=0.03$), while mature vessels were larger in adult patients compared to prepubertal patients ($p=0.008$). **Conclusions:** Follicles in transplanted ovarian tissue from very young prepubertal patients can grow and develop to secondary and antral stages. However, high rates of abnormal and atretic follicles were encountered in frozen-thawed prepubertal ovarian tissue, with significant loss upon grafting and aging. Prepubertal ovarian tissue vascularization was also documented for the first time.

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FREEZE ALL STRATEGY FOR PATIENTS WITH ADVANCED MATERNAL AGE

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Introduction: Women delay their reproduction nowadays. The number of patient in the group with advanced maternal age (AMA) increases constantly and we need effective methods for them to achieve pregnancy and live birth. There are data that freeze all program gives better results than fresh ET for specific patients groups. **Objective:** Our aim was to analyse the effectiveness of freeze all strategy in patient with AMA and compare the results to these of fresh ET. **Methods:** Analysed were 134 embryo transfers with autologous oocytes. All patients were with AMA. Their age was 36 - 46 years. They were divided in two groups. Group I included 53 cases of freeze all program where all developing embryos were vitrified without fresh ET. Later they were used for vitrified embryo transfers (VET). Group II contained 81 fresh embryo transfers (ET). **Results:** Clinical pregnancy rates (CPR) were 41,5 % in group I (freeze all) and 42% in group II (fresh ET)

$p=1$. Live birth rates (LBR) were 26,4% vs 24,7% in group I and II, respectively ($p=0.8$). When we compare day 5 to day 3 embryos the results for CPR in group I were 51.6% vs 27.3% ($p=0.08$). In group II they were 51,7% vs 14.3% ($p=0.003$). LBRs were 32.3% vs 18.2% when compare day 5 to day 3 ($p=0.3$) in group I. Group II had the following results - 30% vs 9.5%- day 5 to day 3 ($p=0.06$). **Conclusion:** After applying freeze all strategy for AMA women CPR and LBR were very good. But there was no statistically significant difference when compare CPR and LBR in freeze all group and fresh ET group. We found increased LBR (although not statistically significant) when we culture embryos to day 5 versus day 3 for both groups - freeze all program and fresh ET. **Support:** None **Disclosures:** No conflict of interest

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FRESH OVARIAN AUTOTRANSPLANTATION TO PRESERVE OVARIAN ENDOCRINE FUNCTION AND DELAY PREMATURE MENOPAUSE: A DEMONSTRATION OF THE TECHNIQUE

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Introduction: Ovarian tissue cryopreservation and transplantation has been successfully used for fertility preservation in patients suffering from cancer. In our centre, we are offering fresh ovarian autotransplantation to prevent iatrogenic premature ovarian insufficiency (POI) as a new technique for patients undergoing bilateral salpingo-oophorectomy (BSO) for benign gynaecological conditions. We aim to assess the endocrine function of patients who have received the fresh ovarian autografts and provide an overview of the technique (using visual demonstration). **Methods:** The tissue is retrieved laparoscopically. Vasopressin is injected into the ovarian cortex followed by a stay stitch to manipulate the ovary. Curved micro scissors are then used to remove the cortex. The medulla is carefully dissected, and cortex trimmed to 1x1cm (1mm thick). The tissue is transplanted back to pelvic side wall using two to four simple inverted stitches. Patients are followed up at 6 months and 12 months and transplant function is tested using clinical symptoms, FSH, AMH and pelvic ultrasound. **Results:** Over 20 patients have had fresh autografts in our centre. The graft function is seen to be better in younger patients (evident by their ovarian reserve markers and follicles on ultrasound scans). It is important to note however that some patients may have a better reserve than others prior to the surgery, because of which the graft yields better results. **Conclusions:** Ovarian tissue transplantation should be considered in patients undergoing BSO for non-cancer indications such as chronic pelvic pain, benign ovarian cysts and endometriosis to prevent POI. Further consideration needs to be given to the use of transplants in delaying menopause in women to prevent osteoporosis and heart disease as well as provision of endogenous hormone production. **Support:** None **Disclosures:** None

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FUNCTIONAL PRESERVATION AFTER WHOLE OVARY VITRIFICATION IN A PORCINE MODEL

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Introduction: Infertility remains one of the most common and life-altering complications in young female cancer survivors. With improved survival rates, the demand for fertility preservation has increased dramatically. While cryopreservation of embryos and oocytes are effective, currently, ovarian tissue cryopreservation is the only proven option for future fertility in prepubertal patients and those who require immediate cancer treatment. Compared to ovarian tissue cryopreservation, whole ovary cryopreservation offers longer graft lifespan and immediate blood supply to the ovarian follicles after transplantation with vascular anastomosis. However, methods for cryopreserving whole ovaries are suboptimal and require new and improved technologies. **Methods:** In this study, porcine ovaries were perfused with increasing concentrations of cryoprotective agents (CPAs) at constant pressure (40 mmHg) for 50 minutes under dynamic temperature profile (20 to 4°C, as CPA concentration increases). CPA-loaded ovaries were 1) examined for ice formation using a differential scanning calorimeter, 2) perfused with CPA washout solutions (CPA load/unload; L/U), or 3) cooled to, and stored at -145°C, followed by re-warming (RT bath) and CPA washout (vitrified, VIT). Fragments of fresh, L/U and VIT ovaries (n=5 per group) were cultured in media containing bromodeoxyuridine (BrdU) for 48 hours at 37°C in 5% CO₂, fixed and processed for histology and BrdU immunohistochemistry. Estradiol (E) and progesterone (P) concentrations were measured in culture media. DSC thermograms revealed that ice was absent during cooling but minimally present (2.36±0.79% in cortex and 1.86±0.84% in medulla) during re-warming. **Results:** After L/U and VIT, different classes of ovarian follicles demonstrated typical morphology; however, possible cryo-induced damage was observed in the stroma (abnormal space). Interestingly, ovaries in all groups exhibited BrdU uptake (proliferation) in growing follicles and produced E and P. **Conclusions:** Whole ovary cryopreservation, once available, will have a significant impact in the care provided for patients at high risk of ovarian failure from cancer treatments. **Support:** None **Disclosures:** I have nothing to disclose.

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GAP JUNCTION PROTEIN CONNEXIN-43 EXPRESSION BEFORE AND AFTER VITRIFICATION OF ISOLATED BOVINE PRE-ANTRAL FOLLICLES

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Introduction: Reprotoxic cancer treatments often threaten future reproductive capacity. Currently, ovarian tissue cryopreservation is the only option to preserve fertility for pre-pubertal girls and women who cannot undergo hormonal stimulation. However, the reintroduction of malignant cells after reimplantation of a frozen-thawed tissue strip is a huge concern. Cryopreservation of isolated pre-antral follicles (PAFs) might therefore represent a promising alternative. So far, it is not quite clear if vitrification safeguards membrane proteins such as connexins (Cx), that constitute gap junctions and mediate intercellular communication, essential for follicle development. Therefore, we evaluated the expression of Cx43 in vitrified isolated bovine (as a model for human) PAFs. **Methods:** Mechanically isolated bovine PAFs (Ø 30-70µm) were cultured in DMEM/Ham's F12 (38,5°C, 5%CO₂). At D2 of culture, category 1 follicles (i.e. intact basement membrane and intact connection between oocyte and granulosa cells, Jorssen et al., 2015) were selected for Neutral Red staining and fixation or vitrification. Vitrification was carried out in mini cell strainers (25µm, nylon, Funakoshi, Japan), using the IrvineScientific® Vitrification Kit for human oocytes and embryos (Alere Health BV, Tilburg, NL). Follicles were warmed the same day and cultured. On D3, viability was assessed by the non-toxic vital dye Neutral Red (Langbeen et al., 2014) and follicles were fixed for immunofluorescence staining of Cx43. Follicles were incubated overnight with rabbit anti-Cx43, 1h with the appropriate secondary antibody conjugated with FITC and counterstained with Hoechst. **Results:** Preliminary data showed expression of Cx43, present between granulosa cells, in 100% of cultured PAFs (n=10) and in 72,7% of vitrified-warmed PAFs (n=11). All evaluated PAFs stained Neutral Red positive. **Conclusions:** Cx43 was, although slightly reduced, preserved in vitrified PAFs. This study reports for the first time the expression of connexins in isolated cultured and vitrified PAFs, which is a difficult procedure considering the small follicle size and multiple transfer steps. **Support:** None **Disclosures:** The authors declare that they have no conflict of interest.

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HOPE USING TELEMEDICINE, TRANSPORTATION, AND SLOW FREEZING OF HUMAN OVARIAN TISSUE IN JAPAN

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Introduction: Human Ovarian-tissue Preservation Enterprise (HOPE) was established in Tokyo on November 1st, 2016. HOPE utilizes telemedicine, the safe proven transportation at the proper temperature, and the slow freezing method of ovarian tissues for fertility preservation (FP) of female cancer patients as in Denmark and FertiPROTECT. Using this system, women can be treated for cancer in their local hospitals, and optional treatments for FP can be consisted, decided and operated without physically moving. Only the removed ovarian tissues are safely transported from any place of Japan within 24h. to HOPE to cryopreserve. This network system makes each expertise in the field work more proficiently without holding all the work on their hands. **Methods:** We utilized telemedicine for communication among cancer patients with their family, counsellors, nurses, oncologists, and reproductive doctors, before reaching a decision on fertility preservation. Removal of one entire ovary or half resection of bilateral ovaries was performed by laparoscopy under general anesthesia. 8x4x1 mm-sized slices were cryopreserved by proven slow freezing. We summarized the data of HOPE from May 2017 to July 2019. **Results:** We cryopreserved ovarian tissue of *fifteen female cancer patients (breast cancer, leukemia, malignant lymphoma, etc.)* in Japan. Necessary information for decision-making was provided by telemedicine. 2 of 15 cases were completed all procedures up to ovarian tissue cryopreservation by telemedicine. The average age was 34.6±6.6. Transportation took 1–19 hours. The average number of cryopreserved slices was 22.2±7.2. We confirmed live primordial follicles in all patients by viability test. **Conclusions:** Customizing the model of Denmark and FertiPROTECT suitable for cancer patients in Japan and related institutions, we will firmly establish this network system. In the near future, we would also like to introduce a successful case of pregnancy after transplanting cryopreserved ovarian tissues. **Support:** None **Disclosures:** The authors declare no conflicts of interest associated with this manuscript.

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IS OVARIAN TISSUE ABLE TO WITHSTAND REFREEZING-RETHAWING ?

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Introduction: Several years ago, whole ovary cryopreservation with its vascular pedicle was achieved in our center by perfusing the ovarian artery with heparinized solution prior to freezing medium. The aim of the present study was to evaluate the capacity of ovarian tissue to withstand freezing-thawing of whole ovaries followed by refreezing-rethawing of cortical strips. **Methods:** Cryopreserved whole ovaries with their vascular pedicles were donated by four patients aged 25–34 years for research purposes. Whole ovaries were thawed by perfusing the vascular pedicle with different solutions containing decreasing concentrations of sucrose, before being cut into strips. Some strips were assigned to the control group (frozen-thawed; FT), while the rest were refrozen by the slow-freezing technique and subsequently rethawed, constituting the refrozen-rethawed (RR) group. Strips from each group (FT and RR) were either directly fixed in formaldehyde after thawing or xenografted to mice for 21 days. In order to compare tissue viability between groups, follicle density, follicle stage, proliferation and fibrosis were analyzed. **Results:** Healthy preantral follicles (follicles/mm³, mean ± SD) were observed in all four subgroups: 338 ± 445.9 in FT non-grafted, 246.5 ± 327.6 in FT grafted, 207.9 ± 134.5 in RR non-grafted and 30.8 ± 30.8 in RR grafted. Follicle density showed a similar decline after grafting in both FT and RR tissue. Moreover, follicle classification dynamics were comparable in both grafted groups (50.1 ± 5.3% vs 45.2 ± 50.7% primordial follicles and 49.9 ± 5.3% vs 21.4 ± 37.1% growing follicles in the FT and RR grafted groups respectively, mean ± SD). We did not encounter any significant increase in the proportion of fibrotic areas after grafting. **Conclusions:** Refreezing-rethawing does not appear to alter follicle survival or dynamics more than standard freezing-thawing. Further analyses are ongoing. **Support:** None **Disclosures:** Nothing to disclose

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JAPANESE NATIONWIDE SURVEY OF SPERM BANKING FOR YOUNG CANCER PATIENTS IN 2016

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Introduction: Because of advances in therapy, survival rates among AYA patients with cancer have improved. However, many of chemotherapies are toxic to germ cells, they have been known to reduce spermatogenic potential and cause male infertility. Fertility preservation is currently the only tool to achieve this. However, unlike egg or embryo cryopreservation systems, the sperm cryopreservation system in Japan is not a registration system. To clarify the status of sperm banking in Japan, we conducted this survey regarding to sperm bank in 2016. **Methods:** We mailed questionnaires to 695 institutes that advocated sperm banking for either ART or Oncofertility. The institutes' directors were asked to report included type of clinic and department, use and renewal rate of preserved sperm, the average number of patients per year, and the number of patients who banked sperm in 2015. **Results:** In total 329(47.3%) directors returned completed questionnaires. Of these, 152 had sperm bank before chemotherapy. According to the classification of department, 117 gynecological departments and 35 urology. Of those who responded that they had opened sperm banks, 136 responded that was 1 - 10 patients; the most frequent. The usage rate of preserved sperm was 17.8%. In total, 820 patients: 237 with testicular; 383, hematological; 46, bone and soft tissue; 20, brain; and 134 with other malignancy consulted the responding institutes for sperm banking. 105 (27.4%) patients with hematological malignancy underwent chemotherapy before banking. The rate of sperm banking failure in the hematological malignancy group was the highest in all groups. **Conclusions:** This investigation was the first nationwide survey on sperm banking in Japan. Our survey revealed that, in Japan, about 60% of the agencies responsible for banking are private clinics, and about three fourth are gynecological departments. The number of patients whose sperm was preserved seemed small considering the number of cancer patients. In Japan, information regarding sperm banking seems to be lacking. **Support:** We conducted this national survey with the assistance of the Ministry of Health, Labor and Welfare in Japan. **Disclosures:** We have no COI and we conducted this national survey with the assistance of the Ministry of Health, Labor and Welfare in Japan.

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N-ACETYL CYSTEINE IMPROVES FOLLICLE SURVIVAL AND UPREGULATES ANTIOXIDANT DEFENSE IN XENOGRAFTED HUMAN OVARIAN TISSUE

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Introduction: Transplantation of cryopreserved ovarian tissue result in a great loss of ovarian follicles due to an initial period of ischemia followed by reperfusion injury and oxidative stress. Murine studies have shown that the antioxidant N-acetylcysteine (NAC) can improve follicular survival after grafting. The aim of this study was to evaluate effects of NAC on follicular survival and vascularization in xenografted and cultured human ovarian tissue. **Methods:** Donated ovarian cortex (5x5x1mm; n=24) from 12 women undergoing ovarian tissue cryopreservation were thawed and transplanted subcutaneously into immunodeficient mice, which were allocated to 2 treatment groups receiving daily intraperitoneal injections with; saline (control) or 150 mg/kg NAC for 12 days post-transplantation. Grafts were retrieved after 4 weeks and histologically processed for revascularization by immunohistochemistry using murine CD31, fibrosis by Masson's Trichrome and follicular counts and morphological evaluation. Ovarian cortex pieces (2.5x2.5x1mm; n=48) from 3 women were allocated to 4 groups with increasing concentrations of NAC (0,5,25,75mM) and cultured for 4 days. Gene expression analysis were performed for vascular endothelial growth factor (VEGF) and antioxidant defense markers; HMOX1, CAT, and SOD1. **Results:** Follicle density was highest in the NAC-treated group (16.3±4.8) (mean±SEM) compared to control (10.2±3.6) but the groups did not differ significantly. Fibrosis was significantly decreased in the NAC-group compared to control (P<0.005). Interestingly, the percentage of CD31 positive area was decreased in the NAC-group (0.53±0.09) compared to control (0.73±0.1) (P=0.06) which was supported by *in vitro* data showing a significant decrease in VEGF expression with increasing concentrations of NAC (P<0.005). A significant increase in HMOX1, CAT, and SOD1 gene expression with increasing concentrations of NAC was found (P<0.001). **Conclusions:** Results indicates that NAC increase follicle survival in xenografted human ovarian tissue and upregulates transcription of antioxidant defense markers but reduce revascularization in xenografted tissue by downregulating VEGF expression. **Support:** None **Disclosures:** None

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ONCOLOGIC AND REPRODUCTIVE OUTCOMES IN PATIENTS WITH CERVICAL CANCER AFTER RADICAL TRACHELECTOMY

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Objective: To investigate oncologic and reproductive outcomes after radical trachelectomy for cervical cancer in our hospital. **Methods:** We retrospectively assessed patients who were scheduled radical trachelectomy in Gifu Univ. Hosp. from 2011 to 2019. A total of 20 patients with cervical cancer were scheduled radical trachelectomy. The median of age was 35 (range 27–40). There were 16 patients with squamous cell carcinoma, 3 patients with adenocarcinoma and 1 adenosquamous carcinoma in histological diagnosis. Clinical Stage (FIGO) of these patients were 2 of IA1, 5 of IA2, 12 of IB1 and 1 of IIA1. Seventeen patients underwent radical trachelectomy and pelvic lymphadenectomy, whereas 3 underwent radical hysterectomy because of 2 patients with wider spread of disease and 1 patient with positive nodes. Complications (urinary tract injury and postoperative cervical stenosis) occurred in 2 patients. The median of follow-up duration was 40.5 months (range 7–98). None showed recurrence following radical trachelectomy. **Results:** In 9 patients with planned pregnancy after treatment, 4 patients had 9 pregnancies. The median age was 32 (range 29–35) in pregnant group and 34 (range 28–37) in non-pregnant group. The median follow-up duration was 73 (range 18–98) in pregnant group and 67 (range 13–85) in non-pregnant group. The breakdown of these 9 pregnancies were 5 spontaneous pregnancies, 2 of artificial insemination of husband sperm and 1 of assisted reproductive treatment. There were 1 of full-term birth, 2 of premature births, 2 of ongoing pregnancy, 3 of early miscarriage and 1 of tubal pregnancy. In 5 non-pregnant group, 2 patients with complications were included. Reproductive treatment period is still short in non-pregnant group. **Conclusions:** Patients underwent radical trachelectomy in our hospital had good oncologic outcomes. Although cervical stenosis was the cause of infertility and operative complication delayed to start reproductive treatment, our reproductive outcomes were favorable. **Support:** None **Disclosures:** We have no disclosure.

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OOCYTE VITRIFICATION IN ADOLESCENT WOMEN FOR FERTILITY PRESERVATION

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Introduction: Fertility preservation (FP) by means of oocyte vitrification (OV) is nowadays a universally-accepted technique. Some concerns remain regarding its use in adolescent women due to their recent maturation of the pituitary axis thus affecting ovarian stimulation outcomes, especially in

light of the scarce results available. The aim of the study is to describe our experience with OV in adolescent women who preserve their fertility for medical reasons. **Methods:** Fertility Preservation Unit of La Fe University Hospital. Forty-nine cycles of oocyte vitrification were performed from June 2013 to August 2019 in a total of 33 patients. **Results:** Mean age of the group was 17.76 ± 1.76 (13 to 19 years inclusive). The main indication for FP was Hodgkin Lymphoma (N=17, 51.52%), followed by ovarian surgery (N=6, 18.18 %). Average AMH was $19.62 \text{ pMol/L} \pm 15.74$ and average antral follicle count at the beginning of the stimulation was 15.22 ± 9.75 . Cancellation rate due to poor ovarian response was 12.12 % (N=6). Short antagonist protocol was used in 45 cycles (91.84%). Start of cycle was in initial follicular phase in 33 cycles (67.35%), random start in 9 (18.37%) and luteolysis in 7 (14.29%). Average doses per initiated cycle were FSH $2219.47 \text{ IU} \pm 909.88$ FSH and $910.65 \text{ IU} \pm 1065.29$ hMG, with average duration 10.12 ± 3.46 days. Per follicular aspiration, average number oocytes retrieved and metaphase II were 13.69 ± 10.00 and 10.28 ± 8.08 respectively. The oocyte maturity rate was 67.76% and follicular output rate (FORT) was 79.53%. **Limitations:** Descriptive study, small sample size. **Conclusions:** OV in adolescent women seems to yield optimal results which are comparable to those of adult women. These results along with other coming from more studies should be reassuring for clinicians and patients. OV appears to be a good FP technique in this age group. **Support:** None **Disclosures:** Authors do not have any conflict of interest to disclose.

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ORGANISATIONAL ISSUES AND OUTCOME OF DELIVERING FERTILITY PRESERVATION SERVICE FOR CHILDREN OVER A 10-YEAR PERIOD IN STRASBOURG UNIVERSITY HOSPITALS

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Introduction: In last decades, survival of children with malignancies and those undergoing hematopoietic cell transplantation (HCT) has been increased significantly. One of the long term side effect of radio- and chemotherapy concern difficulties in conception in adult life. Fertility preservation techniques are employed in order to prevent infertility at adult stage. Children and their parents face a complex medical care when considering different methods for fertility preservation (FP). Multidisciplinary teams need to be available and trained to provide the suitable FP technique in reasonable time. We highlight here our issues and outcome of 10-year experience

of ovarian and testicular tissue cryopreservation. **Methods:** We performed ovarian tissue cryopreservation for 26 pre-pubertal and 6 post-pubertal girls undergoing chemotherapy and testicular tissue cryopreservation for 9 pre-pubertal boys. **Conclusions:** Continuous information of medical teams, parents and public should be maintained in order to improve the availability of these FP techniques to concerned patients. **Support:** None **Disclosures:** The authors declare no conflict of interest.

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OUTCOMES AND BARRIERS OF IMPLEMENTING A FERTILITY PRESERVATION PATIENT DECISION AID IN CLINICAL PRACTICE: LESSONS FROM THE CANCER, FERTILITY AND ME STUDY, UK

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Introduction: Women with cancer of child-bearing age often need to make time-sensitive fertility preservation decisions whilst simultaneously planning and starting cancer treatment. We report our experiences of developing a fertility preservation patient decision aid (PtDA). The purpose of this PtDA was to better support teenage and adult women (aged 16 years +) and diagnosed with *any* cancer in the UK, to make fertility preservation choices. **Methods:** In a prospective mixed-method, three stage study, the PtDA was: 1) developed following the IPDAS criteria, involving a large steering group, 2) user (*alpha*) tested to gather feedback on its content and format from patients (n=12), cancer and fertility healthcare professionals (HCPs) (n=10) and other key stakeholders (n = 5), and finally, 3) field (*beta*) tested to evaluate its acceptability when integrated into routine cancer care pathways at diagnosis. Forty-one women were given the resource and completed questionnaires to measure aspects of decision-making and quality of life. Thirty women took part in an in-depth interview. Further interviews (n=3) and a focus group with HCPs were also undertaken. **Results:** Patients who received the PtDA, described a positive impact on their ability to make fertility preservation decisions and support them at a stressful

time. However, many women (>300) were not given the PtDA and there were contrasting views between patients and their clinical teams relating to the content in the resource, who should receive it, and at what stage in the cancer pathway. Lack of clarity amongst the oncology team about whose role it is to have a FP discussion also hindered its use. **Conclusions:** PtDAs have the potential to address a great unmet need for patients. However, much work is needed to ensure implementation in routine care. Future research could utilise these findings to strengthen the successful adoption and integration of fertility preservation PtDAs within a clinical setting. **Support:** None **Disclosures:** None

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OVARIAN TISSUE CRYOPRESERVATION IN FRANCE: DATA FROM THE GRECOT (RESEARCH AND STUDY GROUP FOR OVARIAN AND TESTICULAR CRYOPRESERVATION) REGISTER.

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Introduction: Since the first publication reporting an ovarian tissue cryopreservation (OTC) in human (1996), this technique followed by an auto-transplantation, has now proven its efficacy in preserving female fertility, so it is important to better characterize the patients who can benefit from. The purpose of our study was to characterize the profile of French patients who are accessing OTC, the types of treatments indicating OTC and to compare them to other published cohorts. **Methods:** The clinical characteristics of the patients and the types of treatments considered after the OTC were extracted from the GRECOT cohort register. Anonymous clinical information for each patient was collected from voluntary fertility preservation centers. **Results:** Twenty eight university centers have sent their OTC data to the GRECOT register. From 1995 to December 2018, 2585 patients underwent an OTC. The average age of the patients at OTC was 17.9±9.8 years, 27% were under 12 years old at the time of OTC. The largest indication for OTC was oncologic conditions (87% of cases), including 52.3% of malignant hematological diseases (acute leukemia 19.8%; Hodgkin lymphoma 19.4%) and 47.7% of solid tumors (10.8% sarcoma; 7.5% breast cancer; 8.6% neuroblastoma). Among non-malignant diseases, 39.2% of patients suffered from hemoglobinopathy and 14.5% had Turner's syndrome. When data were available, 54.4% of patients had chemotherapy before OTC. In 55.2% of cases, patients had OTC before autologous or allogeneic hematopoietic stem-cell transplantation (HSCT). **Conclusion:** In France, patients are on average younger than in the other published cohorts. As a result, there are more diseases affecting young girls (e.g. neuroblastoma). The percentage of patients with breast cancer is much lower than in other series. In our series, the majority of patients had OTC before HSCT. Differences between cohorts are important to understand in order to improve patient selection for OTC. **Support:** None **Disclosures:** None

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OVARIAN TISSUE TRANSPLANTATION IN FRANCE. DATA FROM THE GRECOT (RESEARCH AND STUDY GROUP FOR OVARIAN AND TESTICULAR CRYOPRESERVATION) REGISTER.

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Introduction: Ovarian tissue cryopreservation (OTC) is one option for fertility preservation. In France, the registry of the GRECOT reports during the period of 1995 to 2018, 2585 patients with OTC. In France, the cryopreserved ovarian cortex is implanted when the patient is cured and desires to be childbearing. The objective of this study was to report the French activity of cryopreserved ovarian tissue transplantations (OTT) from 2002 up to December 31th, 2018; using the cohort of patients of the centers affiliated to the GRECOT. In France, OTT began in 2002, and was organized since 2008 by the French bioethics' law, and the orders defined the rules of best practices of ART. Sixteen centers in France perform OTT. One hundred twenty-six OTT were performed in 114 patients. At the time of the OTC 105 patients presented cancerous diseases (82% of hematological malignancies and 18% of solid cancerous diseases); only 9 patients had not cancerous diseases. **Methods:** The graft was performed in orthotopic position in 90% of the cases. The restoration of the ovarian function occurred for 89 % of the patients and was obtained 5.7 ± 2.4 months after the graft. **Results:** At the end of 2018, 33 patients (28.9%) have been pregnant (92% with initial cancerous disease). Thirteen patients obtained more than one pregnancy, with a total of 46 pregnancies for the cohort. Twenty four patients (21.0%) delivered of one child and 5 patients have more than one delivery. Eighty % pregnancies were spontaneous and only 20% required Assisted Reproductive Technology. **Conclusions:** OTT, with control of the risk of reintroduce cancer cells, can be considered as an established technique in these indications. These data confirmed the previous studies of the Danish and Israeli teams. Cases with the risk of reintroducing malignancy, needed to work for the development of alternative techniques to the OTT. **Support:** None **Disclosures:** no conflict of interest

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POTENTIAL FOR FERTILIZATION AND BLASTOCYST FORMATION OF IN VITRO MATURED OOCYTES RECOVERED FROM EXCISED OVARIES IN ONCOLOGICAL PATIENTS

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Introduction: With the increased rate of stable remission after gonadotoxic cancer treatment, new methods of fertility preservation are required for oncological patients. Oocytes obtained from the ovaries *ex vivo* have the potential for maturation and can be vitrified for fertility preservation. However, little is known about the competence of such oocytes to fertilization and euploid embryos formation. **Methods:** In our study, we included 17 cancer patients aged 24-43(mean 33,5) who underwent ovariectomy in our research center (July 2018 - June 2019). Along with ovarian tissue preparation for cryopreservation, the cumulus-oocyte complexes were retrieved from visible follicles by aspiration and from the remained fluid after ovarian tissue dissection. COCs were cultured in the commercial IVM medium supplemented with 0,75 IU/ml HP-hMG for 48-72 hours. **Results:** 82 mature oocytes were fertilized by ICSI. 18 hours after fertilization 23 zygotes (28%) had 2PN, 29 (35%) had 1 or 3PN, the remaining oocytes had no signs of fertilization. The cleavage rate was 96% (22/23), and the blastocyst formation rate was 13% (3/23). All 3 blastocysts were obtained from one patient diagnosed with T1N1M0 breast cancer. PGT-A was performed and found all 3 blastocysts euploid and suitable for embryo transfer. **Conclusions:** Our study demonstrates for the first time, that euploid blastocysts can be obtained from ovarian tissue oocytes. These embryos could be screened for aneuploidies and inherited mutations and then be vitrified in order to provide the best fertility preservation strategy for women with cancer. However, altogether, oocytes recovered from excised ovaries have a low potential for fertilization and blastocyst formation. It might be due to the suboptimal conditions of IVM or the effect of the patient's anamnesis and nosology on the oocyte quality. In order to introduce this method into routine practice, wider studies investigating the clinical predictors for successful IVM programs of ovarian tissue oocytes are needed. **Support:** None **Disclosures:** n/a

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PRESERVATION OF FERTILITY IN ONCOLOGICAL PATIENTS: EXPERIENCE IN A PUBLIC MEDICAL CENTER IN MÉXICO CITY

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Introduction: 5-year survival rate has been increased over the years in children and adolescents with cancer. Patients undergoing chemotherapy have a Relative Risk (RR) of premature ovarian failure of 4 in case of having presented cancer during adolescence and a RR of 24 in case of presenting cancer at 21–35 years. Radiation therapy doses at risk of inducing azoospermia/amenorrhea include >25 Gy in adults, >6 Gy in prepubertal children, also it depends of the location, >6Gy for abdominal or pelvic therapy in adult women, >15 Gy in prepubertal goatients and > 10 Gy in postpubertal patients. **Methods:** From the data base of spermatobioscopies of reproduction laboratory of Medical Center “20 de Noviembre”, all the patients of the fertility preservation program were included from 2012 to May 2019. **Results:** 44 male patients from 13 to 45 years old were included in the study. The most frequent diagnosis was testicular cancer with a prevalence of 43%. 88% of the patients with EBD previous chemotherapy were abnormal. The most common alterations were oligozoospermias and oligoasthenozoospermias. The girls included in the fertility preservation program were only 8 patients, the diagnoses were Ewing's sarcoma and Osteosarcoma, all of them received GnRH agonist. **Conclusions:** 88% of spermatobioscopy prior to chemotherapy or radiotherapy resulted in alterations in sperm quality which leads us to conclude that cancer and probably other factors related to treatment trigger testicular dysfunction. **Support:** None **Disclosures:** 88% of spermatobioscopy prior to chemotherapy or radiotherapy resulted in alterations in sperm quality

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PROFESSIONALS' BARRIERS IN FEMALE ONCOFERTILITY CARE AND STRATEGIES FOR IMPROVEMENT

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Background: The potential loss of fertility is one of the most important undesirable side effects of cancer treatment in women of reproductive age. Unfortunately, despite guideline recommendations, not all patients are informed about their fertility risks and referred for fertility preservation counseling. Insight into barriers for discussing fertility preservation and appropriate referral is necessary before improvements can be made. **Methods:** The aim of this study was to identify barriers and gather improvement suggestions through semi-structured in-depth interviews conducted with 24 professionals working in oncofertility care. Oncological professionals were recruited from the three Dutch expertise hospitals for female fertility preservation and their affiliated hospitals. Subsequently, an expert panel meeting was held to reach consensus on a set of improvement strategies. The expert panel consisted of six healthcare professionals, five survivors and two researchers.

In the Dutch setting, financial aspects do not play a role in oncofertility care. **Results:** Barriers were identified and categorized into the patient level (e.g. focus on surviving cancer), the professional level (e.g. lack of awareness, knowledge, time, and attitude), or the organizational level (e.g. unavailable written information, disagreement on who is responsible for discussing infertility risks). The expert panel reached consensus on essential elements for a multifaceted improvement program: development of information materials (leaflets, online decision aid), education of professionals, a role for specialized oncology nurses in informing patients and patient navigators at the fertility department to facilitate referral and counseling, medical record reminders, standard consultations with a gynecologist, and agreement on responsibility. **Conclusions:** Professionals perceived barriers in knowledge, attitude and organization of oncofertility care and suggested strategies to improve oncofertility care. This study forms the basis for the development of a multifaceted oncofertility program, which is essential to increase adherence to the national clinical guideline. **Support:** This work was supported by the Radboud University Medical Center. **Disclosures:** Prof. Dr. Braat reports unrestricted grants from Ferring BV, Serono, and Goodlife, outside the submitted work.

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PURGING HUMAN OVARIAN CORTEX FRAGMENTS OF CONTAMINATING LEUKEMIC CELLS BY INDUCING MITOTIC CATASTROPHE

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Objective: Increasing the safety of ovarian cortex autotransplantation by *ex vivo* elimination of metastasized leukemic cells with an Aurora kinase inhibitor. **Methods:** Human ovarian cortex fragments were micro-injected with three acute myeloid leukaemia (AML) and three chronic myeloid leukaemia (CML) human cell lines followed by culturing for three days to induce small tumour foci. Subsequently, these fragments were treated *ex vivo* with an Aurora kinase inhibitor for 24h to eradicate tumour foci. After treatment the fragments were cultured for an additional six days to allow any remaining tumour cells to form new foci. We performed (immuno)histochemistry to detect any residual cancer cells after *ex vivo* treatment. The effect of the Aurora kinase inhibitor exposure on the viability of ovarian cortex tissue and follicles was determined by histology, glucose uptake assay, follicular viability assay and *in vitro* growth of small follicles. **Results:** Mitosis of metastasized AML and CML cells in ovarian cortex tissue was severely affected by a 24h *ex vivo* treatment with an Aurora kinase inhibitor, leading to the formation of large multi-nuclear syncytia and large scale apoptosis. Only the megakaryocytic CML cell line MEG-01, which is known for its aberrant Aurora-B kinase expression, was not

affected. Ovarian tissue viability was not compromised by treatment, as no statistically significant difference was observed regarding the percentage of morphologically normal follicles, follicular viability, glucose uptake or *in vitro* growth of small follicles between the ovarian cortex treated with Aurora kinase inhibitor and the control. **Conclusion:** Pharmacological inhibition of Aurora kinase *ex vivo* effectively eradicates experimentally induced AML and CML tumour foci. Viability of the ovarian tissue was not compromised by this treatment. This indicates that purging of ovarian cortex tissue prior to autotransplantation using an Aurora kinase inhibitor is a promising strategy for enhancing the safety of this fertility preservation option. **Support:** Unrestricted grant Merck **Disclosures:** None

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REGIONAL ONCOFERTILITY PROGRAM: 5 YEARS OF EXPERIENCE IN THE RURAL SOUTHEASTERN UNITED STATES

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Background: Infertility is a devastating consequence of cancer therapies for reproductive age patients. Although oncofertility is standard of healthcare in the US, barriers to fertility preservation (FP) remain. The oncofertility program at Augusta University was initiated as a multi-disciplinary approach to overcome FP barriers by providing prompt access to FP, cost reduction strategies, and subsidized storage costs. **Objectives:** To present a five year experience of an oncofertility program in the rural Southeastern US, and demonstrate a cost-effective model. The study was an IRB-approved retrospective cohort study. **Methods:** Data were collected on patients referred to the oncofertility program from 2014 to 2019. Characteristics of study participants such as age, sex, and ethnicity, time to consultation, malignancy type, and outpatient/inpatient status, choice of FP method, insurance status, and pregnancies after FP utilization were analyzed. Statistical analysis was performed using logistical regression models in STATA, Pearson's Chi square and t-test (p values < 0.05 was significant). **Results:** 40 females and 29 males utilized FP. Female patients underwent medical ovarian suppression (n=17), assisted reproductive technology (ART), embryo or oocyte cryopreservation (12), ovarian transposition (1), combined FP methods (4), or declined treatment (6). Fifteen male patients underwent semen cryopreservation, 3 had unsuccessful attempts, and 11 declined treatment. Females with gynecologic malignancies have a 9 times greater odds of proceeding with FP than patients with hematologic malignancies. Females with hematologic malignancies were less likely to proceed with combined FP methods. Federally insured and uninsured patients were less likely to proceed with

ART due to financial barriers, despite discounted services. Four patients utilized their cryopreserved gametes or embryos, resulting in live births. **Conclusion:** We demonstrate effective integration of a regional oncofertility program in the rural Southeastern US. Advocacy on behalf of cancer patients for insurance coverage of FP is a key step towards elimination of oncofertility disparities in the U.S. **Support:** None **Disclosures:** None

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SUCCESSFUL SURROGATE PREGNANCY AFTER PERCUTANEOUS OOCYTE RETRIEVAL FOLLOWING MODIFIED RADICAL HYSTERECTOMY WITH LEFT SALPINGOPHERECTOMY AND RIGHT OVARIAN TRANSPOSITION TO ANTERIOR ABDOMEN WALL

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Objective: To evaluate the procedure of percutaneous oocyte retrieval, outcome of IVF cycles and subsequent successful surrogate pregnancy in a patient who underwent modified radical hysterectomy with right ovarian transposition to anterior abdominal wall (subcutaneous plane) for endometrioid adenocarcinoma grade II. **Introduction:** A 33-year-old woman, known case of PCOS, who underwent modified radical hysterectomy with left salpingo-oophorectomy and right ovarian transposition to the anterior abdominal wall and was referred for assessment of reproductive potential and IVF in the year 2016. **Methods:** Three cycles of IVF were performed using GnRH antagonist in first two attempts and short protocol with GnRH agonist in third attempt, with percutaneous technique of oocyte retrieval from the transpositioned right ovary. In the third attempt in July 2017, we were able to retrieve five oocytes and subsequently freeze three embryos and one blastocyst. The surrogate underwent sequential transfer in June 2018 which resulted in a positive clinical singleton pregnancy and successfully delivered a female baby in February 2019. **Results:** A successful pregnancy and delivery. **Conclusions:** This may possibly be the first reported case of ovarian hyperstimulation and percutaneous aspiration of oocytes from a transpositioned right ovary (subcutaneous plane). Literature survey showed reports only of transabdominal retrieval. **Support:** None **Disclosures:** Nothing to disclose

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SURROGACY AS AN OPTION FOR TREATMENT OF PATIENTS WITH CRYOPRESERVED GAMETES. A CRITICAL OVERVIEW OF OUR EXPERIENCE

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Introduction: The objective of our study is to examine if the surrogacy procedure is a fertility option for patients with cancer history having undergone fertility preservation treatment. In our retrospective study Google Analytics Solutions (Google Inc, 2019) is used to gain potential patients insights from two different European countries. 28639 different website sessions were identified in thirty six months, coming from two specific European countries where surrogacy is not allowed. **Methods:** The study included 409 patients with a mean age of 40.06 ± 5.76 years and a range between 19 and 50 years from two countries in a time period of sixteen months between September 2016 and August 2019. 52.6% were women with repeated IVF failures, whereas 8.8% were patients with hysterectomy or uterine abnormalities, 7.6% patients with MRKS Syndrome, 9.8% cancer patients, 6.8% homosexuals, 6.3% patients with serious medical disorders, 4.6% patients over 45 years of age and 3.4% with recurrent miscarriages. **Results:** 27.4% of the patients had a Skype conference, whereas 17.3% visited our clinic and 16.14% decided for treatment. Both fresh as well as frozen embryos were transferred. An oocyte pickup was performed in 56.06% of the patients with a fertilization rate of 75.9%. An oocyte donation was performed in 22.7%, while frozen embryos from the home countries were provided in 21.2% of the patients. Overall, thirty five embryo transfers are performed, resulting to twenty-two single pregnancies, four twin pregnancies, seventeen deliveries of twenty-one healthy babies, four abortions and an ectopic pregnancy. **Conclusions:** Surrogacy could be considered as a last option for fertility treatment for patients with absence of functioning uterus, serious diseases, recurrent IVF failures or pregnancy losses. The number of patients interested urges the need for a regulation of the cross border reproductive care among European countries. **Support:** None **Disclosures:** No disclosure

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SURVEY OF SPERM CRYOPRESERVATION TO PRESERVE FERTILITY IN JAPANESE CANCER PATIENTS: OUR EXPERIENCE

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Introduction: The total fertility rate (TFR) has been sluggish in many countries in Southeast Asia, including Japan and is low at 1.42 in 2018. The annual number of births fell below 1 million for 3 years and was 918,000 in 2018. Iwamoto (2013) reported there is a concern that the quality of sperm in young men will deteriorate. The declining birthrate is an important issue in Japan, where the population is declining

further. The Japanese government has formulated a general outline for measures to reduce the declining birthrate. However, the treatment of male infertility is becoming assisted reproductive technology rather than investigating the cause. A nationwide survey of sperm freezing of cancer patients by the Ministry of Health, Labor and Welfare was first conducted by Yumura (2014). **Methods:** In this paper we report the actual situation of our experience. **Results:** The clinical findings at the time of sperm freezing before malignant tumor treatment in 87 cases at Tsukuba Gakuen Hospital (Ibaraki) and 36 cases at Sanno Hospital (Tokyo) were mean age: 31 and 39, semen findings: sperm concentration 26.4 million / ml, 68.6 million / ml, sperm motility: 20.7% and 42.0%, main disease: frequency of testicular tumor / leukemia: 91% and 61%, respectively. **Conclusions:** Despite a small case, it was very interesting there were significant differences at the two facilities, Sanno Hospital in the capital and Tsukuba Gakuen Hospital, where located just 40 km from Tokyo. Now that Japan has started efforts to preserve the fertility of cancer patients, it is necessary to clarify the current situation as a whole. The role of male infertility treatment is important, and we believe that raising the awareness of doctors involved in freezing sperm before treatment in male patients with malignant tumors will surely lead to the improvement of the declining birthrate problem. **Support:** None **Disclosures:** OK

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THE BLASTOCOELIC FLUID ACQUIRED BY LASER-ASSISTED IS NOT RECOMMENDED FOR ANEUPLOIDY DETECTION OF PREIMPLANTATION CHROMOSOMES

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Introduction: The purpose of this study was to investigate the effect of blastocoelic fluid acquired by laser-assisted on the detection of chromosome aneuploidy, and to compare the consistency of preimplantation genetic testing with that of trophoctoderm cells. **Methods:** 54 fresh blastocysts were subjected to shrink by laser-assisted, and the culture medium drops were collected as blastocoelic fluid, trophoblastic cell biopsies were also performed on 32 fresh blastocysts of them. All samples were subjected to detection of chromosomes aneuploidy by MALBAC (Multiple Annealing and Looping - based Amplification Cycles, MALBAC). **Results:** A total of 54 cases of blastocoelic fluid samples were detected, only 35 cases had amplification results, with a success rate of 64.8%. All the 32 samples of trophoctoderm cells had amplification results, with a success rate of 100%, among which there were only 4 cases consistent in aneuploidy results between blastocoelic fluid and trophoctoderm cells, with a consistency of only 17.4% (4/23). **Conclusions:** The method of blastocoelic fluid acquired by laser-assisted is simple, but the amplification efficiency is still low, and the consistency with trophoctoderm cell results is poor, it is not recommended for aneuploidy

detection of chromosomes before embryo implantation. **Key words:** laser assisted; blastocoelic fluid; trophectoderm cells; aneuploidy detection **Support:** None **Disclosures:** no conflict of interest disclosure

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THE DIFFERENT CLASSIFICATION OF CRYOPRESERVED OVARIAN TISSUE BETWEEN THE FIRST OVARIAN TISSUE CRYOBANK IN CHINA AND A CENTRALIZED FERTILITY PROTECTION CENTER IN EUROPE

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Introduction: Aim is to compare the different diseases proportion of ovarian tissue cryobank between China and Europe. **Methods:** Beijing Obstetrics and Gynecology Hospital, China, established the first ovarian tissue cryobank in 2012. We analyzed 160 cryopreserved ovarian tissue cases (using slow freezing) from January 01, 2016, to June 01, 2019, comparing with the database from a centralized European cryobank at the Université Catholique de Louvain (UCL) in Brussels, Belgium, which from 1997 to 2013 cryopreserved 545 ovarian tissues. **Results:** Mean patients' age for China and Europe is 32±6 years (4 years- 45 years) vs. 22.3 ±8.8 years (6 months - 39 years), respectively. The disease classification of cryopreserved ovarian tissue is as follows (China vs. Europe in % of all cryopreserved tissues): pelvic malignancy (77.7% vs. 6%) breast cancer (10.2% vs. 17%), lymphoma (1.7% vs. 23%), leukemia (1.7% vs. 9%), benign and borderline ovarian cancer (1.7% vs. 17.5%), benign hematological pathology (1.0% vs. 3.0%), neurological malignancy (0 vs. 5.0%), gastrointestinal malignancy (2.1% vs. 3%), other diseases (3.9% vs. 16.5%). **Conclusion:** The classification of the cryopreserved ovarian tissue has no difference between China and European cryobank, but the diseases proportion is big different. Breast cancer, lymphoma, leukemia and borderline ovarian cancer are more common in the Europe cryobank. Pelvic malignancy is more frequently seen in China first ovarian tissue cryobank. The results suggested that Chinese need more education of ovarian tissue cryopreservation for fertility protection. **Support:** Beijing Municipal Administration of Hospitals Clinical medicine Development of special funding support (XMLX201710), Beijing Municipal Administration of Hospitals' Ascent Plan (DFL20181401). The first batch of Beijing maternal and child health specialist demonstration units "menopausal health specialist" (2018/01-2020/121) **Disclosures:** no

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THE OUTCOME OF RANDOM-START OVARIAN STIMULATION WITH OR WITHOUT AROMATASE

INHIBITOR FOR OOCYTE CRYOPRESERVATION IN CANCER PATIENTS

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Introduction: Random-start ovarian stimulation makes it possible to retrieve oocytes as soon as possible compared to standard follicular phase-start protocols. This is important for young cancer patients who need emergency fertility preservation preceding cancer therapy. However, the number and quality of retrieved oocytes using random-start ovarian stimulation is not well confirmed. Moreover, there are few reports on ovarian stimulation using aromatase inhibitor for estrogen receptor (ER) positive patients. Therefore, the purpose is to assess the advantage and disadvantage of random-start ovarian stimulation with or without aromatase inhibitor (letrozole) in young cancer patients who underwent controlled ovarian stimulation (COS) for oocyte cryopreservation. **Methods:** We performed a retrospective observational study at St. Marianna University from October 2012 to March 2019 in Japan. The target population of this study is 59 cycles with 53 cancer patients who underwent COS for oocyte cryopreservation before cancer treatment. The average age of the subjects is 33.3 ± 5.3 years old, and the characteristics of cancer are breast cancer(n=44), blood cancer(n=5), colon cancer(n=4). 35 patients with ER positive breast cancer underwent COS with letrozole. Random-start ovarian stimulation cycles were compared with those of using standard follicular phase-start protocols with or without letrozole. The main outcome measures were duration of ovarian stimulation, total dose of gonadotropins, number of oocytes retrieved, total number of frozen oocytes, and oocyte immaturity rate. **Results:** There was no significant difference between the random-start group and the standard protocol as in total dose of gonadotropins, number of oocytes retrieved, total number of frozen oocytes and oocyte immaturity rate. In the random-start group, the duration of ovarian stimulation scored higher than the standard protocol. We obtained the same result in ovarian stimulation with or without letrozole. **Conclusion:** Random-start ovarian stimulation with letrozole is an effective protocol to retrieve oocytes in a limited time, especially for ER positive cancer patients. **Support:** None **Disclosures:** Nothing

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THE IMPACT OF COLD ISCHEMIA TIME IN DIFFERENT MEDIA FOR OVARIAN TRANSPORTATION: PRELIMINARY RESULTS

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Introduction: Ovarian tissue cryopreservation and transplantation is a widely applied approach to preserve fertility in cancer patients. While pregnancy rates are around 30–50% with this technique, little is known about the effects of transport before ovarian tissue cryopreservation. The goal of this study was therefore to analyze different parameters of media used for ovarian tissue transportation. **Methods:** Cow ovaries were obtained from a local abattoir and divided into 1x1 cm fragments, then immersed in either 0.9% NaCl solution, IVF medium or L-15 medium for 1, 4 or 24 hours at 4°C. After each period, pH, lactate dehydrogenase (LDH) activity, reactive oxygen species (ROS), glucose, lactate and pyruvate were evaluated in the media. **Results:** Results showed a lower pH in NaCl at all timepoints compared to IVF and L-15 media. LDH activity increased with time in each medium and was significantly lower ($p<0.05$) in NaCl than in IVF/L-15 at 1 and 4 hours, indicating lower cell death rates in NaCl. A small but significant ($p<0.05$) increase in ROS production was observed over time, with concentrations lower than those found before reperfusion of ovarian grafts (Cacciottola et al., 2018), and higher in NaCl than IVF/L-15 media. There was no significant difference ($p<0.05$) in glucose consumption, but there was a significant ($p<0.05$) decrease in pyruvate concentration in IVF and L-15 and increase in lactate production in all media, suggesting some cell metabolism. **Conclusions:** Our preliminary data shed new light on the impact of cold ischemia and different transportation media on ovarian tissue. The next steps will involve quantifying apoptosis, necrosis and autophagy and grafting frozen-thawed ovarian tissue in order to understand how differences between media can affect ovarian tissue survival and its population of preantral follicles. **Support:** We would like to thank Planer for sponsoring the participation of Janice Vilela in the conference and FATH (IREC, UCL) for the collaboration with the CMA600 Analyzer. **Disclosures:** None
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THE OXFORD MODEL TO EVALUATING PATIENTS SUITABLE FOR FERTILITY PRESERVATION

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Introduction: Fertility preservation is an important clinical issue with higher cancer survival rates, particularly in young adults and children. Different methodologies have been developed to help achieve this goal. Ovarian tissue cryopreservation is an emerging method of fertility preservation that is gaining momentum with recent guidance published by the British Fertility Society. Multiple small case series have reported positive outcomes with successful live births following orthotopic auto-transplantation. **Methods:** Review of current electronic evidence databases, including review articles and guidelines, to develop a generic pathway to aid clinicians to further understand the different forms of fertility preservation currently available. We aim to present a flow chart that can serve as a reference guide for clinicians faced with this question in a clinical setting. **Results:** Fertility preservation modalities currently include embryo cryopreservation, oocyte and sperm cryopreservation and ovarian tissue cryopreservation with a view to performing either orthotopic transplantation resulting in a spontaneous live birth or requiring assisted reproductive treatment with ovarian stimulation protocols to yield oocytes suitable of recovery and artificial insemination. **Conclusions:** Ovarian tissue cryopreservation is a valuable approach for fertility preservation in children, young adults and in women usually under the age of 35 undergoing gonadotoxic treatment (surgery, chemotherapy or radiotherapy). This approach has the potential to incorporate a larger cohort of patients, including those at risk of premature ovarian insufficiency (POI) (for example secondary to multiple periodic blood transfusions in transfusion dependent haemoglobinopathies) and transgender patients. The studies to date involve small case series with positive findings. In the future, cryopreserved tissue could be subjected to in vitro maturation with subsequent in vitro fertilisation (IVF) to avoid the risk of re-transplanting malignant cells, utilising the remaining pool of primordial follicles. **Support:** None
Disclosures: None

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THE RELATIONSHIP BETWEEN CUMULATIVE CISPLATIN DOSE AND REPRODUCTIVE AND SEXUAL FUNCTION IN OVARIAN DYSGERMINOMA PATIENTS WITH PLATINUM-BASED CHEMOTHERAPY

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Introduction: Malignant ovarian germ cell tumors are rare tumors that accounts for less than 5% of all ovarian cancers, but these tumors are not uncommon in young people. BEP (Bleomycin, Etoposide, Cisplatin Combination) therapy is one of the standard treatments as post-operative chemotherapy. Although it has been reported that patients with history of

BEP therapy were delivered safely, it seemed to be difficult for many of them to become pregnant. Hypogonadism may be strongly correlated with the total amount of cisplatin. Cumulative cisplatin dose of more than 400 mg/m² is listed as an intermediate risk for men in ASCO 2013 guideline, but is not described for women. Therefore, we examined the relationship between cumulative dose of cisplatin and gonadotoxicity. **Methods:** Ten young cases with ovarian germ cell tumors at Mie University Hospital from January 2010 to May 2019 were enrolled. We retrospectively examined the relationship between cumulative dose of cisplatin and gonad function, menstrual period, and pregnancy history. **Results:** The cumulative cisplatin dose of 9 patients (90%) who recovered menstruation after BEP therapy was 458 mg (300–700 mg). All 3 cases who had hope of raising a baby after treatment were delivered. The cumulative cisplatin dose in these cases was 300 to 440 mg, and the risk of hypogonadism was deemed to be low. Surprisingly, even in a patient with high dose cisplatin of 700 mg, menstruation was resumed after treatment. **Conclusions:** Reported cases of pregnancy and delivery after chemotherapy are useful information for patients. In addition, the progress of the study on cumulative dose of anticancer drugs can reduce the number of patients suffering from hypogonadism. Since this study had small number of patients because of the rare tumors, further study with multiple institutions should be conducted to prove the relationship between cumulative doses of cisplatin and gonadotoxicity. **Support:** None **Disclosures:** The authors no financial conflicts of interest to disclose concerning the study.

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THE STEPPED VITRIFICATION FOR HUMAN OVARIAN TISSUE CRYOPRESERVATION

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Introduction: The advantage of stepped vitrification (SV) is promoting vitrification, thus avoiding ice crystal nucleation, while decreasing the toxic effects of high cryoprotectant concentrations. We aimed to test this method for human ovarian tissue cryopreservation. **Methods:** Ovarian cortex (n=7) was taken from fertile adult women undergoing surgery for benign gynecological conditions. Samples were fragmented to 1x1x1 mm pieces, cultured *in vitro* for 24h and fixed as controls. Fragments of 5x5x1 mm were subjected to the vitrification curve performed in a programmable controlled-rate freezing machine (Asymptote, VIA Freeze Research) adapted with a 'liquidus tracking' device, which allowed sample transfer to ever higher concentrations of dimethyl sulfoxide (DMSO) as the temperature was reduced. The final concentration of DMSO was 50%, while the final temperature of the curve was -40°C. Then, samples were placed in liquid nitrogen vapor (-150°C) before being submerged and stored (-196°C) until the warming protocol was performed. Warmed tissue was cultured *in vitro* for 24h in order to re-establish tissue activity and metabolism and observe the cell response to the cryopreservation protocol. **Results:** Histological evaluation of the vitrified-warmed tissue showed large numbers of degenerated follicles after 24 hours of *in vitro* culture. Thus, we evaluated DMSO perfusion rate by X-ray computed tomography, ice crystal formation by freeze-substitution, and cell toxicity by transmission electron microscopy, seeking possible reasons why follicles degenerated. Although cryoprotectant perfusion was considered normal and no ice crystals were formed in the tissue, ultrastructural analysis detected typical signs of DMSO toxicity, such as mitochondria degeneration, alterations in chromatin condensation, cell vacuolization and extracellular matrix swelling, in both stromal and follicular cells. **Conclusions:** The findings indicated that the method failed to preserve follicles due to the high concentrations of DMSO applied. However adaptations can be made to avoid cryoprotectant toxicity to follicles. **Support:** FAPESP (process number 2016/22947-8) and Siemens Healthcare S.L.U. **Disclosures:** Nothing to disclose.

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TUMOR INDUCING POTENTIAL OF TWO LEUKEMIC CELL LINES IN A XENOGRAFTING MODEL

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Background: Reimplantation of cryopreserved ovarian tissue can successfully restore ovarian function in young cancer patients after gonadotoxic treatment, but for patients with leukemia, there is a risk of malignant cell transmission. **Aim:** To assess the tumor-inducing potential of two different leukemic cell lines xenografted to immunodeficient mice. **Methods:** Fifty female severe combined immunodeficient (SCID) mice were grafted with chronic myeloid leukemia in blast crisis (BV-173 cells) or relapsed acute lymphoblastic leukemia (RCH-ACV cells). One hundred, 200, 500, 1,000 and 10,000 BV-173 or RCH-ACV cells were embedded inside a fibrin matrix along with 50,000 ovarian stromal cells and xenografted to the animals. Two mice implanted with a fibrin matrix without any leukemic cells served as negative controls. Clinical signs of disease were monitored for up to 20 weeks, before the livers and masses were collected for macroscopic analysis and gene expression of BCR-ABL1 and E2A-PBX1 fusion transcripts present in BV-173 and RCH-ACV cells respectively. **Results:** *BV-173 cell line:* Mice grafted with 100, 200 or 500 cells showed no sign of disease 20 weeks after transplantation and were negative for BCR-ABL1. However, 3 of the 5 animals grafted with 1,000 cells and all those grafted with 10,000 cells developed disease, expressing the BCR-ABL1. *RCH-ACV cell line:* Of 4 mice grafted with 100 cells, 2 developed disease and were E2A-PBX1-positive. All those grafted with 200, 500, 1,000 or 10,000 cells showed clear signs of disease, and all but one were E2A-PBX1-positive. **Conclusions:** The present work proves that when leukemic cells are xenografted to SCID mice peritoneum, the disease-inducing potential varies between cell lines. Indeed, it depends not only on the number of leukemic cells, but also the status, type and cytogenetic profile of the disease when ovarian tissue is harvested. **Keywords:** Fertility preservation, leukemia, ovarian tissue cryopreservation, ovarian tissue transplantation, artificial ovary. **Support:** None **Disclosures:** No conflict of interest to disclose

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VITRIFICATION OF HUMAN OVARIAN TISSUE AND DEVELOPMENTAL POTENTIAL OF THE IN VITRO MATURED OOCYTES RETRIEVED FROM OVARIES

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Introduction: This is a study including five patients affected by oncological diseases and enrolled in fertility preservation program from January to August 2019. **Methods:** After obtaining written informed consent, ovaries were laparoscopically retrieved from women between the ages of 29 and 35 years who underwent oophorectomy due to cervical or breast cancer. Ovarian tissue was suspended in HEPES buffered saline and transferred to the laboratory within 45 minutes. Ovarian cortex was cut into pieces measuring 10x10x1mm. Sections of ovarian cortexes were frozen by vitrification technique. Vitrification was performed in an open system with Kitazato device and media, according to protocol. To assess the follicle survival rate neutral red staining was performed followed by histological evaluation in each patient. We got the overall follicle survival rate over 70%. In two patients in vitro maturation (IVM) was performed. The cumulus oocyte complexes (COCs) were retrieved from the visible follicles by aspiration and from the remained fluid after ovarian tissue dissection. COCs were cultured in the Sage medium (CooperSurgical) supplemented with 0,75 IU/ml FSH/hCG (Meriofert, IBSA) for 48–72 hours. **Results:** In first patient only two COCs were retrieved, one oocyte reached the MII stage after IVM and was fertilized by husband's sperm, but no signs of fertilization. In second patient 54 COCs were retrieved, 21 reached the MII stage after IVM, 17 oocytes arrested at the GV or MI stages and 16 oocytes degenerated. 10 mature oocytes were vitrified and 11 - fertilized with donor sperm. 5 normally fertilized zygotes stopped developing on 3–8 cell stage. **Conclusions:** Our study suggests that ovarian cortex vitrification is one of the promising freezing techniques in order to maintain reproductive function. Immature oocytes can be retrieved successfully from the visible antral follicles of excised ovaries, matured in vitro, however, such oocytes have low potential for fertilization and blastocyst formation.

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VITRIFIED MACAQUE OVARIAN TISSUE PRESERVES OVARIAN FUNCTION AND PRODUCES FERTILIZABLE OOCYTES AFTER TRANSPLANTATION TO HETEROTOPIC SITES

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Introduction: Ovarian tissue cryopreservation is the only fertility preservation option available to girls who survive cancer. A randomized, longitudinal, in vivo study was performed to determine which heterotopic transplantation sites produce

competent oocytes from vitrified ovarian cortical tissue. **Methods:** Ovarian cortical pieces from 4 adult and 4 peripubertal rhesus macaques were vitrified in closed straws using glycerol:ethylene glycol containing non-permeating polymers. Tissue was transplanted to subcutaneous (sc) sites in the arm; sc abdomen; sc abdomen + omentum; retroperitoneal + omentum; and bilateral mesosalpinx sites. Transplant vasculature was imaged by contrast enhanced ultrasound (CEUS) and quantified for blood volume (BV) and vascular flow (VF). Ovarian steroids were determined during one year post-transplantation. When the preovulatory follicle reached 5–6mm, hCG was given, follicles were aspirated, and mature oocytes were inseminated in vitro. All animals resumed ovarian cyclicity within 6 months post-transplantation. At all sites, BV increased during weeks 1–2 (2.7-fold) and 1–16 (4.2-fold). BV and VF were greater in omental sites than sc arm. The total number of preovulatory follicles seen in sc arm (2) and sc abdomen (2) was less than in sc abdomen + omentum (14), retroperitoneal + omentum (10) and mesosalpinx (11). The number of oocytes collected were similar between ages. Of 38 total follicles, 31 were aspirated yielding 21 oocytes (68%); 52% metaphase II, 24% metaphase I, 24% degenerated. **Results:** Three oocytes fertilized, and developed to the 4-cell (mesosalpinx), 8-cell (retroperitoneal + omentum), and morula (sc abdomen + omentum) stage. Vascular dynamics in vitrified ovarian cortical tissue transplanted to heterotopic sites in primates can be quantified by CEUS. Retroperitoneal or sc abdominal sites containing omentum and mesosalpinx sites consistently developed a single preovulatory follicle monthly and yielded competent oocytes. **Conclusions:** Efforts to produce offspring are ongoing. Heterotopic transplantation of vitrified ovarian tissue is promising for young cancer survivors seeking fertility. **Support:** R01HD083930, P51OD011092 **Disclosures:** Nothing to disclose.

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WHAT IS THE POTENTIAL OF COLLAPSED BLASTOCYST IN VITRIFIED-WARMED CYCLES?

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Introduction: In IVF vitrified blastocysts transfer is increasingly used, and for a successful outcome blastocyst viability is crucial. Expanded blastocysts tend to collapse during thawing and do not always re-expand until transfer. **Aims:** To assess the potential of vitrified-warmed blastocysts which are assessed as collapsed at the time of transfer, to implant. **Methods:** A total of 278 vitrified-full and expanded blastocysts were thawed between Nov. 2017 and Dec. 2018. PGD Blastocysts were excluded from the analysis. Vitrified-warmed blastocysts were divided into four groups according to their appearance immediately after thawing and at the time of transfer. Group 1 – collapsed after thawing and at transfer. Group 2 – collapsed after thawing and re-expanded before transfer.

Group 3 – expanded after thawing but collapsed before transfer, and group 4 – expanded after thawing and at the time of transfer. IVF outcomes were compared between the groups.

Results: Patient age and blastocyst grade were not different between the groups ($p>0.05$). Our results demonstrate that transfer of expanded-warmed blastocysts (groups 2 and 4) results in 30% pregnancy rates. Interestingly, pregnancy rates of collapsed blastocysts (group 1 and 3) were not different from expanded ones (groups 2 and 4 ($P>0.05$)). The time of incubation between thawing and transfer ranged between 2–3 hours.

Conclusions: Vitrified-warmed blastocysts can be transferred at 2–3 hrs after thawing. Transfer of collapsed blastocysts that didn't re-expand till transfer results in the same pregnancy rates as those of expanded blastocyst. **Support:** None **Disclosures:** No conflict of interest

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WHEN PATIENTS HAVE NO EMBRYOS FOR PGT-A BIOPSY ON DAY 5 OR 6 OF DEVELOPMENT, DOES EXTENDING THE CULTURE TO DAY 7 GIVE ANY REAL HOPE OR JUST FALSE HOPE?

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Objective: Vastly improved embryo culture conditions have extended the standard time frame an embryo can survive in culture, be biopsied and survive vitrification. While it is understood that the optimal days for trophectoderm biopsy would be Day 5 or 6, there are a limited number of patients whose embryos only develop on Day 7. To determine the success rates for this subset of patients, we analyzed one year of IVF cycles for cases where blastocysts were only biopsied Day 7 of culture. **Design:** A retrospective cohort study at a private fertility center. **Methods:** Cycles in 2018 that had trophectoderm biopsy performed only on Day 7 were included in this study ($n=22$). Analysis of the euploidy rate of these embryos and pregnancy outcomes after transfer were performed. **Results:** Only 22 (2.4%) of 904 total patients had biopsy performed on only Day 7. There were 26 embryos biopsied, with a mean age of 38.9 years. Biopsies were analyzed using Next Generation Sequencing, resulting in 7 euploids (26.9%). Three Day 7 only Embryo Transfers resulted in no pregnancies. In the same time period, an additional 6 transfers with other Day 7 biopsied embryos all resulted in negative pregnancies too. **Conclusion:** It is the goal for patients to get an embryo from each IVF cycle, although, extending culture to Day 7 for patients who have not had biopsy performed on Days 5 or 6, may not provide success. The euploidy rate of these embryos is low (26.9%) and our studies showed a 0% pregnancy rate. Let's do all we can to give hope, but not false hope. We suggest that programs analyze and

determine their outcomes with these highly extended embryo culture embryos to accurately inform patients of the extremely low probability of a successful cycle. **Support:** None

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WHO AND WHEN ARE THE YOUNG WOMEN RETURNING FOR USING THEIR CRYOPRESERVED MATERIAL AFTER FERTILITY PRESERVATION FOR CANCER TREATMENT? A 10-YEAR EXPERIENCE OF AN ONCOFERTILITY CENTER

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Introduction: Fertility preservation (FP) is increasingly being offered to women facing cancer treatments. However, the use of the cryopreserved material is still scarce. In this study, we present data of a FP center describing who and when are female cancer patients returning to use their cryopreserved material. **Methods:** Patients referred to FP counselling were included. Returning rates and time to return to attempt

pregnancy were examined in a subsample of women in child-bearing age (> 25 years old). **Results:** Between 2009 and 2019, 340 women were counselled for FP (mean age 30.31 ± 6.27, 12–43). Breast (59.1%) and hematological (19%) cancers were the most frequent. Of those, 245 (72.1%) chose to undergo FP, although only 204 (60%) patients had cryopreserved material (69.3%: oocytes; 3.5%: embryos and 27.1%: ovarian tissue). Among the group of patients who survived and had material cryopreserved ($n = 192$), 9.8% returned to use the cryopreserved material. A detailed analysis was performed in a sample of women currently aged > 25 and FP until 2018 ($n = 163$). Nineteen women returned to use the cryopreserved material (returning rate of 11.66%). Mean age at the time of FP was 32.95 ± 3.73 and mean age at the time of the return was 36.47 ± 3.58. Mean time between time of FP and return was 3.5 years. Expectedly, women who returned were significantly older than women who did not (34.48 vs 38.21, $p < 0.001$). No differences were found in the number of MII oocytes between the women who and who did not return (7.91 vs 6.00, $p = 0.159$). Of the total of 19 women who return to attempt pregnancy, 3 achieved pregnancy after treatment (15.7%), and 3 are still in treatment. **Conclusions:** The use of cryopreserved material is still low but increasing with women's ageing. Patients should be assisted in their decision making to attempt pregnancy after FP. **Support:** None

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