Does subcutaneous Granulocyte Colony Stimulating Factor (G-CSF) improve ovarian reserve in women with premature ovarian insufficiency?

1.1 Principal Investigator: Dr Robert Casper, TRIO Fertility

1.2 Study Site: TRIO Fertility

HC protocol number TF005

Background

Women are born with all the eggs they will have in their lifetime which is estimated to be about two million. By puberty there are about four hundred thousand quiescent follicles remaining of which a small number initiate growth each month during a woman's reproductive life. Depletion of this primordial pool of follicles occurs over time, however the rate at which this occurs depends both on genetics (for instance accelerated in women with FMR-1 mutations) and environment (ie. exposure to chemotherapy or radiation therapy in the setting of cancer). Over time these women with premature ovarian insufficiency (POI), like women approaching age 50, can develop resistance to the signals from the brain that are involved in ovulation, disrupting their cycle and ultimately rendering fertility treatments such as IVF not effective.

Premature ovarian insufficiency (otherwise known as premature menopause or premature ovarian failure) is defined as a state of elevated gonadotropins, hypoestrogenism and anovulatory cycles below 40 years old and impacts about 1 % of women [1]. Interestingly, patients who develop premature ovarian insufficiency post chemotherapy have been known to achieve spontaneous pregnancy after a bone marrow transplant [2,3,4]. This is thought to be related to bone marrow derived stem cells that could lead to recruitment of pre-existing quiescent follicles through various mechanisms including differentiation into oocyte-support cells (granulosa/theca/cumulus or stromal cells) and subsequent improved follicular environment, probable paracrine action via growth factor release and allowing for neovascularization and facilitation of blood perfusion to primordial follicles [5,6].

In fact, there is much evidence to suggest this is the case in both mouse models and human studies [6, 9-14]. In these studies, hematopoietic stem cells are either collected via direct aspiration from a patient's bone marrow (BM) or via a less invasive technique using granulocyte colony stimulating factor (G-CSF), which mobilizes stem cells from the BM into the peripheral blood. In the latter procedure, BM derived stem cells (BMDSC) are then collected through peripheral blood plasmapheresis using flow cytometry. In both instances, BMDSCs are either infused in the ovarian stroma of women with POI via laparoscopy or alternatively infused into the ovarian artery.

In Herraiz et al 2018 [7], women who had diminished ovarian response (DOR) and considered as poor responders to gonadotropins were treated with G-CSF for 5 days at a mean dose of 600 mcg/d and peripheral blood was collected to isolate BMDSC and subsequently infused in one ovarian artery using the other ovary as control. The authors were able to show that autologous stem cell ovarian transplantation (ASCOT) post G-CSF treatment led to improvement in antral follicle count

and AMH levels in 81% of patients (13/16 patients). Overall comparing pre and post ASCOT IVF cycles they noted improvement in AFC, reduced cycle cancellation rate and better fertilization rates [7]. Noteworthy a total of 5 pregnancies occurred post ASCOT treatment (3 spontaneous) [7]. Interestingly, although they only infused stem cells into one ovarian artery they noted an effect on the control ovary as well suggesting that perhaps these stem cells and/or growth factors were able to travel through the blood stream and exert their function elsewhere. Hence, this raises the question of whether ovarian artery infusion is required at all, and if simply mobilizing BMDSC from BM into peripheral blood is sufficient to see these positive outcomes. As a result, the same group performed a follow-up study or 10 women with POI undergoing ASCOT compared to 10 women receiving only GCS-F (mean dose 600 mcg/d for 5 days) without apheresis and intraovarian artery infusion of hematopoietic stem cells (8). Both groups had similar results with 6/10 women in each group having visible basal antral follicles, 2 in each group undergoing IVF and 1 patient having a single embryo transferred in each group. This study suggests that intra-ovarian artery infusion of stem cells is not required for folliculogenesis. Since both bone marrow aspiration and ASCOT are quite demanding and invasive procedures, we would like to confirm that treatment with G-CSF will lead to stem cell mobilization from the BM and this process alone will allow for improvement in markers of ovarian reserve in POI patients. G-CSF treatment is safe and has been routinely used in healthy donors for decades for the purpose of allogeneic stem cell transplants. We plan to use a lower dose of GCS-F (480 mcg/d for 4 days) 1 month apart for 2 or 3 treatments.

1.3 Primary Hypothesis

We hypothesize that treatment of premature ovarian insufficiency patients with G-CSF to mobilize bone marrow hematopoietic stem cells will allow for improved ovarian reserve markers including antral follicle count, anti-mullerian hormone (AMH) levels and gonadotropin (FSH) levels. Our aim is to confirm and expand the results of Pellicer et al (8).

Significance of the study:

If this hypothesis is correct, G-CSF subcutaneous injection could be a simple, safe treatment for patients with POI to restore ovulation and improve fertility outcomes.

Study Objectives:

To determine if treatment with G-CSF will allow improvement in markers of ovarian reserve.

Primary outcome –Decreased serum FSH and increased AMH levels and u/s measurement of increased antral follicle count (AFC)

Secondary outcome – Improved ovarian response in IVF cycles if BAFs develop, and spontaneous or IVF pregnancy.

Design:

A pilot study.

1.4 Inclusion Criteria

1) Women ages 25-40

2) Woman who meet criteria for POI defined as AFC < 5, AMH < 3 pmol/L and FSH >30 IU/L. There may also be associated symptoms of the menopause such as hot flushes, night sweats, insomnia and vaginal dryness.

3) Women who are not taking any other medical or fertility treatments except natural estrogen to stop hot flushes.

4) Informed consent

1.5 Exclusion Criteria

1) Women with age > 40

2) Women with history of autoimmune disorders

3) Women with a history of hematopoietic cell malignancies

4) Women with sickle cell disease

5) Women with any other comorbidities that would preclude infertility treatment and pregnancy such as HIV/AIDS, hepatitis B or C, breast cancer or body mass index (BMI) >40.

1.6 Recruitment

Subjects will be identified by the investigators from the clinics at TRIO Fertility. Eligible study participant(s) will be given the consent form and approached in person by study personnel at the clinic. Subjects who express initial interest will be provided with detailed verbal and written information and they will be encouraged to consider the information at home also. They will be followed up by telephone call if they agree to being followed in this manner.

Power Calculation: This is a pilot study, and we will recruit a total of 30 study participant(s). This number is based on the study of Pellicer et al (2020, ref 8) in which 10 women received G-CSF infusion alone without intra-arterial infusion of BM-derived stem cells and only one woman had an embryo transfer in IVF. We hope that 30 subjects will give us a better chance to see at least a 10% response as in the previous study or better with the two monthly treatments.

Study Methods and Procedures

1.7 Ethics

This study will be conducted with adherence to the ethical principles that have their origins in the Declaration of Helsinki, the applicable guidelines for Good Clinical Practices (GCP), and the applicable laws and the Health Canada investigational testing regulations.

1.7.1 Independent Ethics Committee (IEC) or Institutional Review Board (IRB)

Approval from VERITAS IRB will be obtained before the recruitment of study subjects begins. All protocols (and applicable amendments, if any), including the subject information and consent form must be approved by the IRB before implementation. Appropriate reports on the progress of the study will be made by the investigator to the IRB, including any information on the completion or termination of the study.

1.7.2 Subject Information and Consent

The details of the study and consent forms will be provided by one of the study investigators who do not previously know the study participant(s). Information must be given in both verbal and written form. The subject consent form advises the participant that they are free to refuse their participation in or withdraw from the study at any time, without prejudice to the physician-patient relationship or medical care provided. The participant will be encouraged to ask questions. The investigator will obtain and document informed consent from the participant. If additional medical releases are required for the access to diagnostic reports, these too will be obtained and maintained in the study files. The participant will be given copies of their subject information and consent form. The consent form will outline possible side effects of the G-CSF injections. See attached letter from Dr Keating who has 30 years' experience using G-CSF in cancer patients and in healthy bone marrow donors. He describes bone pain, headache, nausea, fatigue and muscle pain as the most common side effects and these usually resolve with over-the-counter ibuprofen products. Dr Keating indicates that G-CSF is safe to use in healthy women although there have been reported rare but serious side effects such as splenic enlargement and rupture and inflammatory vascular complications like aortitis.

1.7.3 Ethical Concerns

Confidentiality – Dr. Casper will be custodian of patient data collected and will ensure that this data is appropriately protected with encryption and coded for analysis. No unnecessary patient information will be collected.

Financial considerations – There will be no compensation for participating in this study.

1.8 Study Timetable

It is anticipated the study will commence pending ethics approval and be completed within 1 year.

1.8.1 Study Procedures

Patients will be recruited in the study as outlined above. They will be given a SC injection of 0.5 ml of G-CSF or filgrastim (Neupogen, Amgen, USA) at a concentration of 300 micrograms/day for 4 consecutive days. This is a typical dose used in healthy volunteers to allow for mobilization of stem cells in the setting of allogeneic stem cell transplantation. We have consulted Dr Armand Keating, head of hematologic oncology at Princess Margaret Hospital in Toronto who confirmed that the most common side effects reported from G-CSF treatment in healthy volunteers are aches/bone pains, headache and flu-like symptoms [15, 16]. Typically, these symptoms are mild to moderate and easily treated with over-the-counter analgesics (i.e.. ibuprofen or acetaminophen). G-CSF administration has been shown to cause a slight splenic enlargement which self-resolves in a few weeks [15]. We will monitor with abdominal ultrasound and routinely assess for persistent splenomegaly. In addition, leukocytosis is expected along with a mild decrease in platelet count and potassium, increase in uric acid, alkaline phosphatase, transaminases and lactate dehydrogenase levels in patients post G-CSF treatment [16] and hence we will monitor these levels as well. The first SC injection will be given in the office by our nursing staff. Patients will be kept under observation for 60 mins. Subsequent injections can be self-administered by the patient at home. Neupogen comes in prefilled syringes of 0.5 ml containing 300 mcg of filgrastim. The syringes can be kept at room temperature for 14 days and will be provided in an opaque envelop to prevent light exposure. The patients will be shown how to administer the Neupogen subcutaneously by the nurse (most subjects have experience with subcutaneous injections from their infertility treatments). They will be instructed to inject the full syringe contents daily at the same time as their first injection for three days and then return to TRIO for monitoring the following day. The procedure, including monitoring for WBC count, FSH, AMH, pelvic u/s for basal antral follicle count and splenic u/s and 4 days of SC injections of Neupogen will be repeated in one month.

Monitoring

Baseline monitoring prior to G-CSF injections will include CBC, liver function tests, electrolytes, abdominal and pelvic u/s, and endocrine screening including LH, FSH, E2 and progesterone plus AMH. All tests except AMH and BAF count will be repeated at each visit to TRIO and after each set of 4 injections. AMH and BAF counts will be repeated at the end of 2 months. (See uploaded Excel spreadsheet for monitoring schedule).

Patients will be offered two rounds of G-CSF treatment one month apart. If no response is seen (improvement in gonadotropin and anti-mullerian levels as well as antral follicle count) then we will provide the patient with the option of a third treatment a month later. Follow-up by blood assessment of AMH and FSH and u/s measurement of BAF count will continue for 3months after the last GCS-F infusion.

1.8.2 Study Visits

Baseline testing will include CBC, endocrine screen (FSH, LH, estrogen, progesterone), markers of ovarian reserve (US for AFC count, AMH levels) and monitoring of liver function tests (ALT, AST, ALP, LDH), kidney function (creatinine, sodium, potassium, uric acid) and lastly US to

check for splenic enlargement. This will be done prior to the first Neupogen injection and on day five (one day after the fourth SC G-CSF injection).

Patients will return one month after the first series if injections (day 30 of study) and repeat all the testing. Another round of G-CSF treatment will be administered (4 days of SC injections) and testing will occur again on day 35. Patient will again return one month after (day 60 of the study) and repeat all testing. If no response is seen, then the patient will be offered a third round of G-CSF injections with testing on day 65 and day 90 of study.

1.8.3 Outcome Measures

Primary outcomes –Success of the treatment will be assessed by a reduction in serum FSH, an increase in AMH (measure of ovarian reserve) and u/s measurement of an increased number of antral follicles (AFC). If an increased number of basal antral follicles is seen in association with an FSH level below 20 IU/L, the subjects will be offered a cycle of IVF to see if oocytes and subsequently embryos can be obtained.

Secondary outcomes – improved ovarian response in IVF cycles if BAFs develop, and spontaneous or IVF pregnancy

Significance

Women with POF are young and most want to have a family. At present, there is no alternative for these women to conceive except to use donor eggs, either from a friend or relative, or more commonly, from an anonymous egg donor bank or service in the United States or Europe since payment for donor eggs in Canada is illegal. If the results of this study are positive for development of new follicles in the ovaries of women with POF, it might be possible for them to use their own eggs in order to conceive, either naturally or through IVF. This opportunity would represent a major benefit both emotionally and financially.

Discontinuation Criteria

The following issues will result in a subject being withdrawn from the study and all existing study data including that obtained from withdrawn subjects will be retained.

1) *Late determination of ineligibility* – Subjects who are subsequently determined not to be eligible for inclusion based on the study protocol inclusion/exclusion criteria set for that study stage or withdraw their consent will be excluded.
2) Patient request.

1.9 Safety Parameters

1.9.1 Risk-Benefit Estimate

The time commitment for subjects will be minimal, because the consent forms will be signed during the regular planned visits at the clinic. Study participant(s) may benefit regarding their fertility potential if a positive response is seen with G-CSF treatment. We plan to use a lower dose of G-CSF (300 mcg/d x 4 days) compared to the study of Pellicer et al (8) (600 mcg/d x 5 days) but with repeated treatments monthly for 2-3 months. If subjects complain of any worrisome side effects, they will be withdrawn from the study after the first month. The product monograph outlining side effects is uploaded with the application.

Knowledge Transfer

All results will be disseminated in the form of peer–reviewed or open-access publications as well as presentations at national and international expert meetings.

Essential Documentation

The Principal Investigator will sign the protocol to document their willingness to adhere to this protocol and to conduct the study in accordance with local legal requirements and the International Conference on Harmonization (ICH) guidelines for GCP.

References:

1. *Guideline on the management of premature ovarian insufficiency*. (n.d.). https://www.eshre.eu/Guidelines-and-Legal/Guidelines/Management-of-premature-ovarian-insufficiency.
2. Hershlag, A., & Schuster, M. W. (2002). Return of fertility after autologous stem cell transplantation. *Fertility and Sterility*, *77*(2), 419–421.
3. Merloni, A., Hawley, J., Levy, W. C., Gooley, T., Cd, B., Deeg, H. J., Doney, K., Storb, R., Sullivan, K. M., Witherspoon, R. P., & A. (1996). Pregnancies following high-dose cyclophosphamide with or without high- dose busulfan or total-body irradiation and bone marrow transplantation. *Blood*, *87*(7), 3045–3052.
4. Salooja, N., Szydlo, R., Socié, G., Rio, B., Chatterjee, R. M., Ljungman, P., Van Lint, M. T., Powles, R. L., Jackson, G., Hinterberger-Fischer, M., Kolb, H., & Apperley, J. F. (2001). Pregnancy outcomes after peripheral blood or bone marrow transplantation: a retrospective survey. *The Lancet*, *358*(9278), 271–276.
5. Tandulwadkar, S., & Karthick, M. S. (2020). Combined use of autologous bone marrow-derived stem cells and platelet-rich plasma for ovarian rejuvenation in poor responders. *Journal of Human Reproductive Sciences*, *13*(3), 184.

6. Buigues, A., Marchante, M. C. F., De Miguel-Gómez, L., Martinez, J. A., Cervelló, I., Pellicer, A., & Herraiz, S. (2021). Stem cell–secreted factor therapy regenerates the ovarian niche and rescues follicles. *American Journal of Obstetrics and Gynecology*, *225*(1), 65.e1-65.e14.
7. Herraiz, S., Romeu, M., Buigues, A., Rodríguez, S., Diaz-Garcia, C., Gómez-Seguí, I., Martínez, J. L., Pellicer, N., & Pellicer, A. (2018). Autologous stem cell ovarian transplantation to increase reproductive potential in patients who are poor responders. *Fertility and Sterility*, *110*(3), 496-505.e1.
8. Pellicer, N, Herraiz, S, Romeu, M, Martinez, S, Buiges, A, Gomez-Seguí, I, Martínez, J, Pellicer, A., 2020. Bone marrow derived stem cells restore ovarian function and fertility in women with POI: Interim report of a randomized trial comparing mobilization versus ovarian injection. 36th Annual Meeting of European Society Reproduction and Embryology (ESHRE).
9. Andaloussi, A. E., Igboeli, P., Amer, A., & Al-Hendy, A. (2018). Intravenous Infusion of Nucleated Peripheral Blood Cells Restores Fertility in Mice with Chemotherapy-Induced Premature Ovarian Failure. *Biomedicines*, *6*(3), 93.
10. Edessy, M., Hosni, H. N., Shady, Y. A., Waf, Y., Bakr, S., & Kamel, M. M. (2015). Autologous stem cells therapy, The first baby of idiopathic premature ovarian failure. *Acta Medica International*, *3*(1), 19.
11. Gupta, S., Lodha, P., Karthick, M. S., & Tandulwadkar, S. (2018). Role of autologous bone marrow-derived stem cell therapy for follicular recruitment in premature ovarian insufficiency: Review of literature and a case report of world's first baby with ovarian autologous stem cell therapy in a perimenopausal woman of age 45 year. *Journal of Human Reproductive Sciences*, *11*(2), 125.
12. He, Y., Chen, D., Yang, L., Qiaoni, H., Ma, H., & Xu, X. (2018). The therapeutic potential of bone marrow mesenchymal stem cells in premature ovarian failure. *Stem Cell Research & Therapy*, *9*(1), 263.
13. Mashayekhi, M., Mirzadeh, E., Chekini, Z., Ahmadi, F., Eftekhari-Yazdi, P., Vesali, S., Madani, T., & Aghdami, N. (2021). Evaluation of safety, feasibility and efficacy of intra-ovarian transplantation of autologous adipose derived mesenchymal stromal cells in idiopathic premature ovarian failure patients: non-randomized clinical trial, phase I, first in human. *Journal of Ovarian Research*, *14*(1).
14. Herraiz, S., Buigues, A., Diaz-Garcia, C., Romeu, M., Rodríguez, S., Gómez-Seguí, I., Simón, C., Hsueh, A. J. W., & Pellicer, A. (2018). Fertility rescue and ovarian follicle growth promotion by bone marrow stem cell infusion. *Fertility and Sterility*, *109*(5), 908-918.e2.
15. Yang, B., & Kido, A. (2011). Pharmacokinetics and Pharmacodynamics of Pegfilgrastim. *Clinical Pharmacokinectics*, *50*(5), 295–306.
16. Hölig, K. (2013). G-CSF in Healthy Allogeneic Stem Cell Donors. *Transfusion Medicine and Hemotherapy*, *40*(4), 225–235.

TRIO Fertility

CONSENT FORM

CONSENT TO PARTICIPATE IN A RESEARCH STUDY

Title: Does subcutaneous Granulocyte Colony Stimulating Factor (G-CSF) improve ovarian reserve in women with premature ovarian insufficiency?

Investigator: Dr R. Casper

The investigator can be contacted by phone: 416-506-0804 x3228 or Fax: 416-972-0036

Introduction

You are being asked to participate in this study because you meet pre-determined study criteria that define premature ovarian insufficiency. Please read this explanation about the study and its risks and benefits before you decide if you would like to take part. You should take as much time as you need to make your decision. You should ask the study doctor or study staff to explain anything that you do not understand and make sure that all your questions have been answered before signing this consent form. Before you make your decision feel free to talk about this study with anyone you wish. Participation in this study is voluntary.

Background

Women are born with all the eggs they will have in their lifetime which is estimated to be about two million. By around puberty, there are about 400,000 follicles left (follicle refers to the egg and surrounding cells). Once you reach puberty, several follicles are recruited each month of which you only typically ovulate one. During a woman's reproductive life, follicle are used up over time and at menopause there about 1000 or fewer follicles remaining.

In premature ovarian insufficiency, this process occurs much faster for reasons unknown. In addition, women can develop resistance to the signals from the brain that are involved in ovulation hence disrupting their cycle.

Patients who developed premature ovarian insufficiency after chemotherapy have been known to achieve spontaneous pregnancy after a bone marrow transplant. This observation is thought to be related to bone marrow derived stem cells producing factors that could lead to the waking up of pre-existing dormant or unresponsive follicles (eggs).

Granulocyte colony stimulating factor (G-CSF) mobilizes certain stem cells from the bone marrow in the blood and is routinely used in healthy patients who will be donors before a stem cell transplant. It is possible that mobilizing bone marrow stem cells with G-CSF could work the same way as a bone marrow transplant to wake up resting follicles in the ovary. If you agree to participate in this study, you will have baseline blood work and ultrasound and four G-CSF injections under the skin each month for three months. You will have monitoring by blood draw and ultrasound before and after every series of 4 daily injections. You will be a part of a group of 30 patients participating in the study.

Purpose

The purpose of this study is to compare your ovarian follicle count (reflecting the presence of eggs that could be stimulated by fertility drugs) and measures of markers of active ovarian follicles before and after treatment with G-CSF.

Procedures

You are being asked to participate in this study because you meet pre-determined study criteria that define premature ovarian insufficiency. If you agree to participate, you will receive 4 G-CSF injections under the skin each month for 2 or possibly 3 months. You will receive the first injection in the office and you will be trained to do subsequent injections at home by one of our nurses if you are comfortable with doing the injections. You will be shown by a nurse how to administer the G-CSF by needle under the skin. The G-CSF comes in preloaded syringes for ease of administration. The syringes can be stored at room temperature in the provided envelop to protect the solution from light exposure. Each injection should be done daily at about the same time as the first injection and for three days. Alternatively, a nurse at your closest Trio location could do the injection for you. You will be monitored with blood work and ultrasound before and after treatment each month. You will have access to the study investigator if you have any concerns with regards to the study or think you are experiencing side effects.

Risks

G-CSF has been developed as a treatment for patients with blood cancers. However, it is now standard practice to administer G-CSF to healthy individuals, both men and women, in order to get stem cells to leave the bone marrow and enter the bloodstream where they can be collected and used for a bone marrow transplant for another person who is ill. In the present study, we will be using the G-CSF for the same reason that it is given to heathy bone marrow donors. In this study, we want to get your bone marrow stem cells into the blood so they can interact with your ovaries and possible awaken resting eggs. The most common side effects reported from G-CSF treatment in healthy subjects are muscle aches, bone pains, fatigue, headache and flu-like symptoms.

Also, patients may commonly experience redness, swelling, or itching at the site of injection.

Typically, these symptoms are mild to moderate and easily treated with over-the-counter medications such as the pain-relievers ibuprofen or acetaminophen. G-CSF administration may cause slight spleen enlargement which usually resolves in a few weeks and hence we will also monitor you with an abdominal ultrasound to measure the size of your spleen before and after treatment. There have been reported some rare complications listed below:

Possible serious side effects from using NEUPOGEN

These are not all the possible side effects that you may experience when taking NEUPOGEN. A table with other very rare but serious side effects is attached at the end of this consent form. If you experience any side effects whether listed here or not, please let Dr Casper know.

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• **Spleen Rupture.** Your spleen may become enlarged and can rupture while taking NEUPOGEN. A ruptured spleen can cause death. The spleen is located in the upper left section of your stomach area. Call your doctor right away if you or your child has pain in the left upper stomach area or left shoulder tip area. This pain could mean you or your child's spleen is enlarged or ruptured.
• **Serious Allergic Reactions.** NEUPOGEN can cause serious allergic reactions. These reactions can cause a rash over the whole body, shortness of breath, wheezing, dizziness, swelling around the mouth or eyes, fast pulse, and sweating. If you or your child starts to have any of these symptoms, stop using NEUPOGEN and call your doctor or seek emergency care right away. If you or your child has an allergic reaction during the injection of NEUPOGEN, stop the injection right away.
• **A serious lung problem called acute respiratory distress syndrome (ARDS).** Call your doctor or seek emergency care right away if you or your child has shortness of breath, trouble breathing or a fast rate of breathing.
• **Sickle Cell Crisis.** Call your doctor or seek emergency care right away if you experience severe pain in bones, chest, gut or joints.
• **Kidney injury (glomerulonephritis)** has been seen in patients who received NEUPOGEN. Call your doctor right away if you experience puffiness in your face or ankles, blood in your urine or brown coloured urine, or if you notice that you urinate less often than usual.

Benefits

You may not receive any direct benefit from being in this study. In the case our hypothesis is correct, you may have a greater number of follicles as well as improved ovarian markers of egg count in the ovary. If there are an increased number of follicles seen on ultrasound and if your FSH levels are less than 20 IU/L, you will be offered an IVF cycle if you wish. You may also be able to get pregnant naturally. In addition, information learned from this study may help other people with infertility in the future.

Voluntary Participation

Your participation in this study is voluntary. If you consent to participate, you can withdraw your participation at any time.

If any other research information becomes available which may be relevant to your care, you will be informed of this in a timely manner.

Alternatives to Being in the Study

You do not have to join this study to receive the regular treatment and refusing to participate in the study will not influence any current treatment decisions. The standard infertility treatment for premature ovarian insufficiency is the use of donor eggs in order to get pregnant.

Confidentiality

If you agree to join this study, the study doctor and his/her study team will look at your personal health information and collect only the information they need for the study. Personal health information is any information that could be used to identify you and includes your:

- name, address, date of birth, and new or existing medical records,that includes types, dates and results of medical tests or procedures.

The information that is collected for the study will be kept in a locked and secure area by the study doctor for 15 years. Only the study team or the people or groups listed below will be allowed to look at your records. Your participation in this study also may be recorded in your medical record at TRIO.

All information collected during this study, including your personal health information, will be kept confidential and will not be shared with anyone outside the study unless required by law. The data will be stored in an electronic medical record that is password protected and only your doctor and research staff will have access to the data. As with any virtual platform of data storage, there is a potential risk of a data breach. You will not be named in any reports, publications, or presentations that may come from this study.

If you decide to leave the study, the information about you that was collected before you left the study will still be used. No new information will be collected without your permission.

Expenses Associated with Participating in the Study

No compensation will be provided for any routine costs experienced by you during the study.

Conflict of Interest

Dr. Casper is a founding partner of TRIO Fertility and benefits financially from all activities at the clinic including IVF. You should not feel pressured or coerced to join this study.

In Case You Are Harmed in this Study

If you become ill, injured or harmed as a result of taking part in this study, you will receive care. The costs of any medical treatment not covered by your Provincial Health Insurance plan or Medical Insurance plan will be covered. No other form of reimbursement for study-related injury or illness is offered. In no way does signing this consent form waive your legal rights nor does it relieve the investigators, sponsors or involved institutions from their legal and professional responsibilities. You do not give up any of your legal rights by signing this consent form. This study has been reviewed and approved by Veritas Independent Review Board (IRB). If you have any questions about your rights as a research participant or the Investigator's responsibilities, you may contact the Manager of Veritas IRB 24 hours per day and 7 days per week at 514-337-0442 or toll-free at 1-866-384-4221. An IRB is a group of scientific and non-scientific individuals who perform the initial and ongoing ethical review of the research study with the subject's rights and welfare in mind. If you have any study-related comments, complaints or concerns, you should first contact the study investigator. Please call the IRB if you need to speak to a person independent from the Investigator and the research staff, and /or if the Investigator and the research staff could not be reached.

Questions

If you have any questions, concerns or would like to speak to the study team for any reason, please call 416 506-0804 x2353.

Consent

This study has been explained to me and any questions I had have been answered. I know that I may leave the study at any time. I agree to take part in this study.

_____	_____	_____
Print Study Participant's Name	Signature	Date

_____	_____	_____
Witness's Name	Signature	Date

(You will be given a signed copy of this consent form)

My signature means that I have explained the study to the participant named above. I have answered all questions.

_____	_____	_____
Print Name of Person Obtaining Consent	Signature	Date

Serious side effects and what to do about them			
Symptom / effect	**Talk to your healthcare professional**		**Stop taking drug and get immediate medical help**
	Only if severe	**In all cases**	
VERY RARE < 0.01%			
Splenomegaly (including the following symptoms: pain in the left upper stomach area or left shoulder tip area)		√	
Osteoporosis in children with severe chronic neutropenia (including decreased bone density, making them weak, more brittle and likely to break)		√	
Allergic reactions (including the following symptoms: rash over the whole body, shortness of breath, a drop in blood pressure (usually causing dizziness or lightheadedness), swelling around the mouth or eyes, fast pulse, weakness, sweating; severe redness or swelling or itching at injection site)		√	√
Cutaneous Vasculitis (including the following signs: inflammation of the blood vessels in the skin)		√	
Sweet's Syndrome (including the following symptoms: plum-coloured, raised, painful sores on the limbs and sometimes the face and neck with a fever)		√	
Kidney Injury (glomerulonephritis) (including the following symptoms: puffiness in the face or ankles, blood in urine or brown coloured urine, or urinating less often than usual).		√	√
***FREQUENCY NOT KNOWN**			
Splenic rupture (including the following symptoms: left upper abdominal pain or pain at the tip of your shoulder)		√	
Pseudogout (including the following symptoms in patients treated for cancer: pain and swelling of the joints, similar to gout)		√	

Serious side effects and what to do about them			
Symptom / effect	**Talk to your healthcare professional**		**Stop taking drug and get immediate medical help**
	Only if severe	**In all cases**	
**Abnormal number of immature bone marrow cells (myelodysplastic syndrome) that could lead to a type of cancer (acute myeloid leukemia) (including the following symptoms: fever, bone pain, bruising, difficulty breathing, bleeding and a general feeling of tiredness).		√	√

*Reported in the post-marketing setting where the incidence is not known.

**Adverse events in breast and lung cancer patients receiving chemotherapy and/or radiotherapy

If you have a troublesome symptom or side effect that is not listed here or becomes bad enough to interfere with your daily activities, tell your healthcare professional.