



Fertility Preservation in a Premenarchal Girl

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Case Presentation

A 13-year-old girl with severe eczema and dermatitis since infancy; recurrent skin, ear, and throat infections; and plantar warts was diagnosed with DOCK8 deficiency. DOCK8 deficiency is a rare autosomal recessive form of Hyper-IgE syndrome (incidence <1:1,000,000) associated with early mortality due to sepsis and infectious complications [7]. While treatment has focused on prevention of infections, bone marrow transplantation (BMT) using myeloablative conditioning protocols has been described as a curative approach for this deadly disease [8].

Following counseling, the family desired to participate in a DOCK8 deficiency clinical trial at the National Institutes of Health using a reduced-intensity conditioning regimen with fludarabine and busulfan prior to allogeneic hematopoietic stem cell transplant [1]. As part of her BMT counseling, the patient and her family were referred by the pediatric oncology team to reproductive endocrinology to discuss fertility risk and fertility preservation options. The oncology team provided estimated cumulative doses for the conditioning regimen.

The patient and her family presented to the reproductive endocrinology team. She reported thelarche 1 year prior, adrenarche around the same time, and no menarche. Maternal age for menarche was 15. The patient reported no prior sexual intercourse and denied smoking, alcohol, and drug use. Her current medications included antibiotics and inhaled steroids and beta-agonist for asthma. Her surgical and family history was non-contributory. She verbalized her desire to have children in the future, and her mother reported that this is a longstanding wish for the patient.

58.1 Assessment and Diagnosis

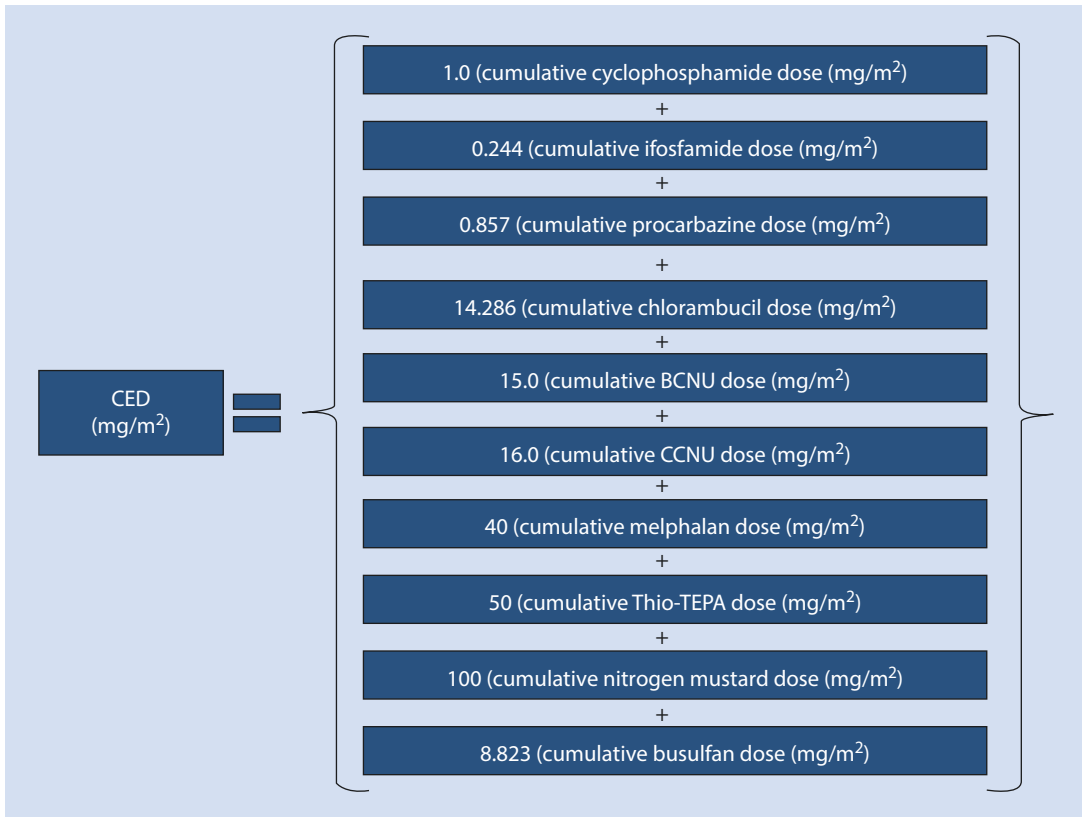
On exam, we observed a well-nourished girl weighing 48 kg with a height of 163 cm (BMI 18.2). She had Tanner stage 2–3 breast development, Tanner stage 2 pubic hair development, and normal external female genitalia. A transabdominal pelvic ultrasound demonstrated an anteverted uterus, a normal vagina, and two ovaries that were easily visible. The right ovary had an antral follicle count of 5, with the largest follicle measuring 9 mm, and the left ovary had an antral follicle count of 6. No abnormalities were observed on ultrasound. Vaginal exam and transvaginal ultrasound were not performed in this patient to minimize patient discomfort as this exam would not additionally impact her fertility preservation care. A hormone assessment revealed an AMH level of 0.95 ng/mL, an estradiol level of 65 pg/mL, a FSH level of 5.0 IU/L, and LH level of 2.9 IU/L, consistent with her state of undergoing the pubertal transition.

For counseling on her fertility risk, we assessed the impact of her treatment exposures, which included busulfan, an alkylating agent, and fludarabine, a purine analog, on ovarian reserve and uterine function. Antimetabolites such as fludarabine are hypothesized to impact only dividing cells and hence would pose less risk to the ovarian follicle pool. Alkylating chemotherapy has been shown to adversely impact ovarian reserve, increasing risks of infertility. In contrast to pelvic

radiation, chemotherapy alone has not been associated with increased spontaneous abortion or intrauterine growth restriction [2].

In order to individualize her risk from busulfan, we calculated the Cyclophosphamide Equivalent Dose (CED) using an equation derived from multiple observational studies of toxicities associated with cyclophosphamide and other alkylating agents (■ Fig. 58.1) [3]. This equation allows for the conversion of different alkylating agents into an equivalent dose of cyclophosphamide, for which there are data on risks of primary ovarian insufficiency (POI) and infertility [3, 4]. Our patient's CED was calculated to be 3.7 g/m². Based on data from the Childhood Cancer Survivor Study (CCSS), children receiving <4 g/m² CED did not have higher rates of POI compared to children who did not receive alkylating agents (risk ratio 0.56, 95% CI 0.07–4.27, *p* = 0.58) [3]. For children receiving <8 g/m² CED, the hazard ratio of POI was 1.55 (95% CI 0.77–3.11, *p* = 0.10), compared to children who did not receive alkylating agents [5]. Within the CCSS, the risk of clinical infertility in females was reported to be 13%. Relative to girls who did not receive alkylating agents, those in the lowest tertile of alkylating agents exposure had a relative risk of infertility of 0.91 (95% CI 0.69–1.20, *p* = 0.51) [4].

Based on these estimates, the patient and her family were counseled that her fertility risk from these exposures was limited, but it is still not possible to predict individual risk. We then



■ Fig. 58.1 Cyclophosphamide equivalent dose (CED) calculation

discussed fertility preservation options for premenarchal girls are all experimental, as there is a dearth of data on treatment efficacy in this population. The options that we discussed included expectant management, ovarian stimulation for oocyte or embryo freezing, ovarian tissue freezing, and ovarian suppression with GnRH agonist. On oocyte retrieval, we discussed transvaginal approach once deep sedation was achieved. Following counseling, the patient and her family verbalized understanding of risks and wished to proceed with oocyte freezing.

58.2 Management

In preparation for oocyte retrieval, the patient signed oocyte freezing assents, and her parents signed consents. The family met with the treating anesthesiologist prior to stimulation. Random start ovarian stimulation was initiated 2 months prior to patient's planned BMT (■ Table 58.1).

Estradiol and progesterone levels on the day stimulation started were <20 pg/mL and <0.2 ng/mL , respectively. She received 150 IU of menotropin and 75 IU of follitropin daily. Estradiol levels were followed every other day starting on day 3 of stimulation. Serial abdominal ultrasound was performed to monitor follicle growth. GnRH antagonist was initiated on day 6 when the lead follicle was at least 14 mm in diameter. On stimulation day 10, her lead follicles were 21 mm by size, a total of six follicles greater than 10 mm were observed, estradiol level was 2312 pg/mL , and 10,000 IU of hCG was administered intramuscularly to trigger oocyte maturation.

On the day of oocyte retrieval, the patient was brought to the operating room, and her parents were able to accompany her while the IV catheter was placed. Then, they were escorted to the waiting room, and deep sedation was administered to the patient. She was positioned in dorsal lithotomy, and vaginal preparation was performed. The transvaginal ultrasound probe was introduced, and the oocyte retrieval was then completed with-

Table 58.1 Ovarian stimulation monitoring

Stimulation day	1	2	3	4	5	6	7	8	9	10
Menotropin dose (IU)	150	150	150	150	150	150	150	150	150	
Follitropin dose (IU)	75	75	75	75	75	75	75	75	75	
hCG (IU)										10,000
Estradiol level (pg/mL)	<20		70		207		760	1194		2312
Progesterone level (ng/mL)	<0.2									
Lead follicle diameters(mm)	7, 9				11, 13		15, 16, 16	19, 18, 16, 15, 13, 11		21, 21, 19, 17

out difficulty. Care was taken during preparation and retrieval to minimize tear to the hymen, which remained intact after the procedure. Ten oocytes were retrieved (eight metaphase II and two metaphase I). All ten oocytes were vitrified. The patient tolerated the procedure well and was discharged home 2 hours after surgery without complications. Onset of menses started 2 weeks following the procedure. She then underwent BMT as scheduled.

58.3 Outcome

This case demonstrates several key aspects of fertility risk counseling and fertility preservation procedures in adolescents. First, patient education of fertility risk was initiated by the treating oncology team, in line with professional society guidelines to address the possibility of infertility with patients and parents prior to cancer therapy. The oncology team had a longstanding partnership with the reproductive endocrinology team and the fertility preservation program at a National Cancer Institute-designated Comprehensive Cancer Center, facilitating a streamlined referral. Importantly, the oncology team provided the planned cumulative doses for chemotherapy to enable the reproductive endocrinology team to help estimate risk for counseling.

Second, the fertility preservation team inclusive of the reproductive endocrinologist, anesthesiologist, nurse, embryologist, administrative staff, and operating room staff had prior experience with treating adolescents. In addition, a thorough literature search was performed to provide up-to-date evidence on fertility risk and review the few

published experiences with ovarian stimulation in pre-menarchal girls [6]. Then, the fertility preservation team was able to discuss coordinated care of this patient prior to stimulation start. The reproductive endocrinologists reviewed the patient's pubertal stage, gonadotropin and estradiol levels, and size of antral follicles and surmised that these antral follicles should be responsive to exogenous gonadotropins for ovarian stimulation. Her normal body habitus enabled ultrasound monitoring of follicular development abdominally in conjunction with estradiol levels. The anesthesiologist and operating room staff worked to confirm that equipment would be suitable for a patient of this size. The administrative team worked with the patient's family to determine insurance coverage and help apply for donated medications. The nurse helped to support the family through injection teaching and communication during stimulation. We met with the patient and her family on two occasions prior to ovarian stimulation and talked with the patient individually without her parents to ascertain that she understood and wished to undergo this procedure.

Third, it was important to estimate the magnitude of her risk, as cancer treatments from chemotherapy to radiation to surgery pose differential risks to fertility outcomes. Even with exposure to alkylating agents, fertility outcomes, particularly in girls, can be normal without fertility preservation procedures [4]. Hence, as part of informed consent, it is critical to convey the magnitude of risk to facilitate discussions in light of significant physical, emotional, and financial costs of undergoing fertility preservation in an adolescent girl. Ultimately, this patient clearly verbalized her desire to prevent infertility if possible, and there is

still a lack of tools to predict precise risk of infertility for an individual. Hence, in line with supporting patient autonomy, we proceeded with ovarian stimulation and oocyte cryopreservation.

Clinical Pearls and Pitfalls

- Fertility and ovarian failure risk from alkylating chemotherapy can be estimated using the CED. There are also risk estimates for abdominal and pelvic radiation [3–5].
- In the pubertal transition, antral follicle count (AFC) and AMH levels are lower than at peak reproductive age. Low AMH and AFC prior to cancer therapy in this population likely do not indicate decreased ovarian reserve.
- There are cases of successful ovarian stimulation in pre-menarchal girls that have resulted in cryopreservation of mature oocytes. In conjunction with estradiol levels, abdominal ultrasounds can be used to monitor stimulation.
- Quality fertility preservation care involves a multi-disciplinary team inclusive of oncology, reproductive endocrinology, anesthesia, nursing, and administration.

Review Questions and Answers [1, 2]

- ❓ Q1. True or False? Alkylating chemotherapy always poses high risk to fertility in girls.
- ✔️ A1. False, fertility risk is dependent on cumulative dose, which can be calculated using the CED. The CED can then be used to estimate risks of POI and infertility based on data from the Childhood Cancer Survivor Study [3–5].
- ❓ Q2. True or False? Fertility preservation using controlled ovarian stimulation and oocyte cryopreservation is possible in pre-menarchal girls.
- ✔️ A2. False, fertility preservation using controlled ovarian stimulation and oocyte cryopreservation is possible in pre-menarchal girls as demonstrated by our successful case and previously published report [6].
- ❓ Q3. True or False? Monitoring of ovarian stimulation must be performed using a transvaginal ultrasound.
- ✔️ A3. False, abdominal ultrasound monitoring in conjunction with estradiol levels can be used in select individuals of low-normal BMI if their clinical history precludes transvaginal monitoring (e.g., virginal status or vaginismus).

References

1. [ClinicalTrials.gov](https://clinicaltrials.gov). Available from URL: <https://clinicaltrials.gov/ct2/show/NCT01176006?cond=DOC K8+Deficiency&rank=1>.
2. Shliakhtsitsava K, Suresh D, Hadnott T, Su HI. Best practices in counseling young female cancer survivors on reproductive health. *Semin Reprod Med*. 2017;35:378–89.
3. Green DM, Nolan VG, Goodman PJ, et al. The cyclophosphamide equivalent dose as an approach for quantifying alkylating agent exposure: a report from the Childhood Cancer Survivor Study. *Pediatr Blood Cancer*. 2014;61:53–67.
4. Barton SE, Najita JS, Ginsburg ES, et al. Infertility, infertility treatment, and achievement of pregnancy in female survivors of childhood cancer: a report from the Childhood Cancer Survivor Study cohort. *Lancet Oncol*. 2013;14:873–81.
5. Chemaitilly W, Li Z, Krasin MJ, et al. Premature ovarian insufficiency in childhood cancer survivors: a report from the St. Jude Lifetime Cohort. *J Clin Endocrinol Metab*. 2017;102:2242–50.
6. Reichman DE, Davis OK, Zaninovic N, Rosenwaks Z, Goldschlag DE. Fertility preservation using controlled ovarian hyperstimulation and oocyte cryopreservation in a premenarcheal female with myelodysplastic syndrome. *Fertil Steril*. 2012;98:1225–8.

Suggested Reading

- Online Mendelian Inheritance in Man. An online catalog of human genes and genetic disorders. Available from URL: <http://omim.org/entry/243700>.
- Uygun DFK, Uygun V, Reisli İ, et al. Hematopoietic stem cell transplantation from unrelated donors in children with DOCK8 deficiency. *Pediatr Transplant*. 2017;21:e13015.