Retrieval of immature oocytes after chemotherapy for Hodgkin’s disease and prolonged ovarian down-regulation with gonadotropin-releasing hormone agonist

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Objective: To describe isolation and in vitro maturation of primary oocytes from the ovarian cortex in the presence of hypothalamic pituitary down-regulation.

Design: Case report.

Setting: Tertiary care university-affiliated hospital.

Patient(s): An 18-year-old patient was given treatment with the ABVD (Adriamycin, bleomycin, vinblastine, and dacarbazine) protocol for Hodgkin’s disease. She underwent ovarian tissue cryopreservation while being cotreated with GnRH agonist because of disease relapse.

Intervention(s): Laparoscopic oophorectomy, ovarian tissue cryopreservation, and in vitro maturation of primary oocytes.

Main Outcome Measure(s): Maturation of primary oocytes isolated from the medium used for preparation of ovarian tissue.

Result(s): Twenty-one immature germinal vesicle–stage oocytes were isolated from the medium of dissection. All were incubated in in vitro maturation medium, and five were matured and frozen.

Conclusion(s): The fact that germinal vesicle–stage oocytes were present in our patient’s medium despite hormonal down-regulation demonstrates that GnRH agonist does not completely inhibit antral follicle development.

(Gernotropin-releasing hormone agonist (GnRH-a) is used before and during chemotherapy to preserve ovarian function and fertility in patients with cancer. Several possible mechanisms have been suggested to explain the beneficial effects of the GnRH-a including [1] inhibition of FSH secretion, which prevents the recruitment of preantral follicles and protects them from gonadotoxic effects, [2] decrease of the utero-ovarian perfusion, [3] direct protective effect on the ovary, [4] up-regulation of the levels of sphingosine-1-phosphate (a potent protective against gonad toxicity), and [5] protection of the undifferentiated germline stem cells. There has been, however, considerable criticism of these mechanisms, especially because of the lack of supporting evidence (1–4). Furthermore, the utility of GnRH-a administration during chemotherapy is controversial, mainly because several studies have not been able to demonstrate its effectiveness (5). We recently reported that cotreatment with GnRH-a had limited efficacy in the ovarian protection of patients with Hodgkin’s disease (1).

CASE REPORT

An 18-year-old woman had a diagnosis of Hodgkin’s disease and received treatment with the ABVD (Adriamycin, bleomycin, vinblastine, and dacarbazine) protocol and a cotreatment of monthly injections of 3.7 mg triptorelin (Decapeptyl; Ferring, Kiel, Germany). Two months later, she began to have amenorrhea, hot flushes, and dyspareunia, which persisted throughout the treatment period. Her baseline hormonal profile was as follows: FSH 2.07 mIU/mL, LH 0.4 mIU/mL, and E2 < 20 pmol/L. She was receiving daily treatment of oral E2 2 mg and norethisterone acetate 1 mg. After six courses of chemotherapy and GnRH-a, she was scheduled for

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CO2 in air with high levels of humidity. After maturation in the Fertility and Sterility laboratory. The ovarian cortex was cut into 5- to 2-mm thickness (Fig. 1).

Unexpectedly, 21 immature germinal vesicle (GV)–stage oocytes were retrieved and isolated from the medium used for dissecting and preparing the ovarian cortex. These GV oocytes were incubated in a culture dish containing 1.0 mL of the SAGE in vitro maturation medium (Cooper Surgical Company, Trumbull, CT) supplemented with 75 mIU/mL FSH and 75 mIU/mL LH at 37°C in an atmosphere of 5% CO2 in air with high levels of humidity. After maturation in culture for 24 hours, the cumulus cells were denuded, and five oocytes were matured to the metaphase II stage by 40 hours after in vitro maturation. These oocytes were cryopreserved in modified human tubal fluid medium (Irvine Scientific, Santa Ana, CA) supplemented with 30% synthetic serum substitute (Irvine Scientific), 1.5 mol/L 1,2-propanediol, and 0.2 mol/L sucrose following the protocol of Fabbri et al. (6). Ovarian slices were precooled for 30 minutes in phosphate-buffered saline medium supplemented with 20% synthetic serum substitute and 1.5 mol/L dimethyl sulfoxide and then cryopreserved as previously described (7).

DISCUSSION

We believe that this is the first report of primordial follicles continuing to grow and develop despite cotreatment with GnRH-a in a patient with Hodgkin’s disease who had received treatment according to the ABVD protocol. The precise mechanism of growth initiation of primordial follicles is not known, but it is obviously FSH independent. Indeed, FSH receptors are expressed after growth at the multilayer stages (3).

Our current clinical experience may help to clarify some of the issues in this topic. If it were true that cotreatment with GnRH-a prevents the growth of new follicles, immature GV oocytes should not be present in the medium. That they were present in our patient’s medium despite hormonal down-regulation demonstrates that GnRH-a does not inhibit antral follicle development completely.

This finding is in accordance with the observation that preantral follicles are present in ovaries of prepubertal monkeys and humans (8). Likewise, primordial follicles continue to initiate growth through hypogonadal states, such as puberty, and during GnRH-a therapy (9). Our current findings cannot, however, refute other possible mechanisms of GnRH-a action, such as a direct protective effect or a decrease of the utero-ovarian perfusion (3, 4).

The effectiveness of GnRH-a is still controversial, and two affiliated groups from the same faculty currently have reached opposite conclusions regarding the efficacy of GnRH-a in fertility preservation (2, 5). The conclusion of the editor was that there is a need for a prospective randomized trial to elucidate this topic (10).

REFERENCES