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The radiosensitivity of the human oocyte: a revised estimate using data from the CCSS on women who had developed premature ovarian failure following a known dose of radiation to the ovary as part of their treatment of cancer.

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Background & Rationale

It is known that irradiation to an area that includes the pelvis in girls is associated with an adverse impact on reproductive function in the survivors (Kalapurakal et al. (2004), Wallace et al. (1989) Critchley et al. (1992)). An improved understanding of the radiosensitivity of the human oocyte will allow the early prediction of ovarian failure and will help physicians manage young women through puberty and premature menopause. It will also allow the early commencement of sex steroid replacement therapy, and provide guidance to patients and their physicians with regard to fertility preservation and future reproductive potential.

The human ovary contains a fixed number of primordial follicles (oocytes), maximal at five months of gestational age. These are progressively lost with increasing age in a bi-exponential fashion, culminating in the menopause at an average age of 50-51 years. The rate of oocyte decline increases around age 37 years when there are approximately 25,000 primordial oocytes, and precedes the menopause by 12-14 years when approximately 1000 oocytes remain. Both chemotherapy and radiotherapy will accelerate oocyte depletion, leading to a premature menopause. Knowledge of the radiosensitivity of the human oocyte will allow an accurate prediction of the age at which permanent, premature ovarian failure will occur in survivors of cancer who have received radiation to a field that includes their ovaries.

In 1989 we estimated the radiosensitivity (LD₅₀: Lethal dose to kill 50% of cells) of the human oocyte to be less than 4Gy (Wallace et al, 1989). The estimate was derived from eighteen females treated with 30Gy of fractionated radiotherapy to the whole abdomen in childhood for a Wilms' tumour. The methodology used was to assume a primordial follicle population of about 2,000 at ovarian failure (the same population as an average 50—51 year old at menopause), then calculate the probable surviving fraction of primordial follicles after treatment. Averaging the

surviving fractions gave a dose-response model for population loss; solving this for 50% gave the LD₅₀ as less than 4Gy.

The study had several shortcomings. The model used for primordial follicle decline was a simple exponential decay, this model has now been substantially revised (Faddy and Gosden, 1996). Furthermore the ages at diagnosis of ovarian failure were older than expected due to limitations in the clinical follow up methodology.

In 2003 we published a revised study that addressed these issues (Wallace et al, 2003). We estimated the LD₅₀ of the human oocyte to be less than 2Gy based upon the application of the solution of the Faddy-Gosden mathematical model of ovarian follicle decline to six patients with ovarian failure secondary to total body irradiation as part of the treatment for childhood cancer (see figs one and two). The revised LD₅₀ is now estimated to be less than 2 Gy.

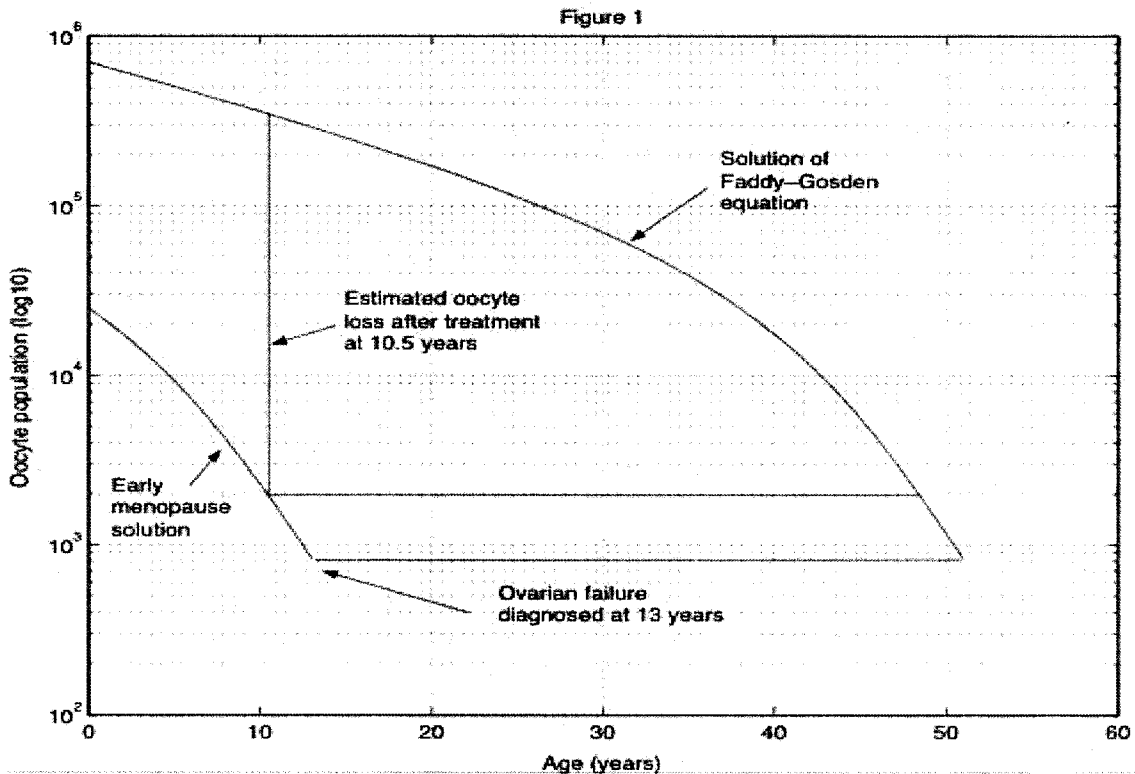


Figure 1. The solution of the Faddy-Gosden equation enables the size of the oocyte pool to be determined for any given age from birth to menopause, at an estimated age of 51 years. Calculation of the estimated surviving fraction for a patient (Aged 10.5 years at treatment) is shown above: ovarian failure occurred at age 13 years, by applying the Faddy-Gosden model we can determine the size of the surviving fraction following radiotherapy at age 10.5 years to be 0.56%.

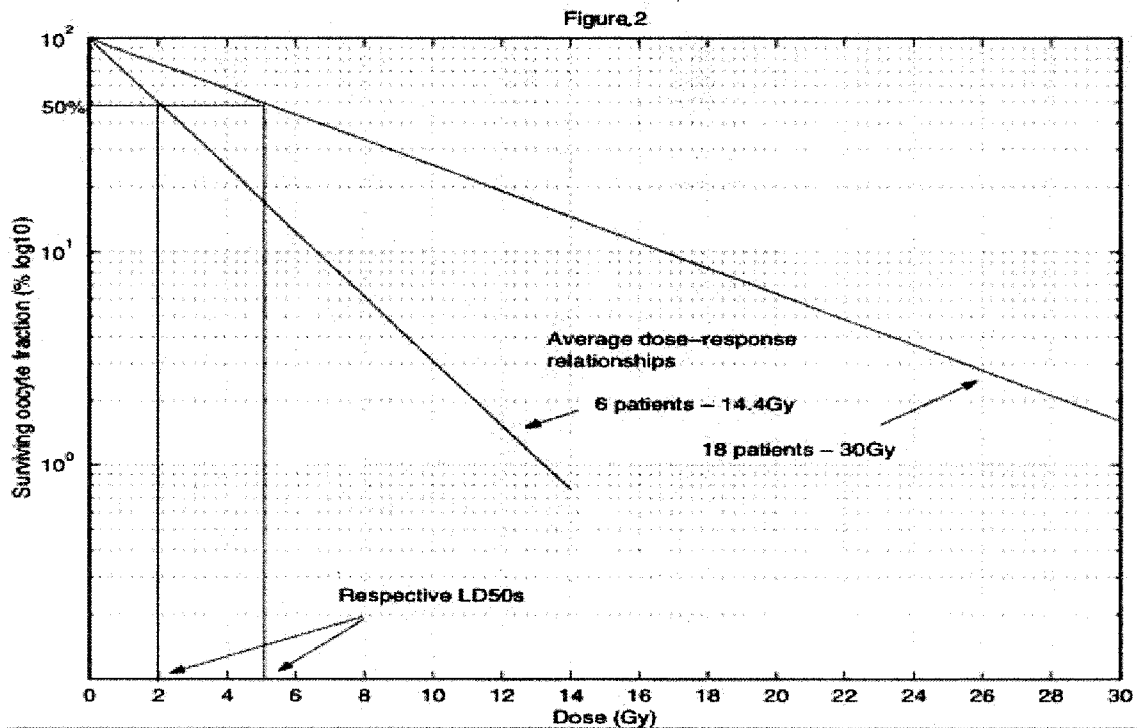


Figure 2. Dose-response relationships for the human oocyte. The mean surviving fraction of oocytes for each patient has been calculated and plotted against the dose of radiation received (i) for the six patients who received 14.4 Gy, and (ii) for the 18 patients who received 30 Gy. These lines represent the estimated (fractionated) dose-response relationship for the human oocyte. The LD₅₀ is given by the dose required to leave a surviving fraction of 50%. Our revised LD₅₀ of <2 Gy is taken from the relationship obtained by data from the cohort of six patients.

Knowledge of the dose of radiation received by both ovaries or the ovary furthest from the radiation field in a large cohort of women who have developed premature ovarian failure will allow a revised and more accurate estimate of the radiosensitivity of the human ovary.

Based on our previous determination of the radiosensitivity of the human oocyte to be <2 Gy we were able to calculate the surviving fraction of the follicle pool for any given dose of radiotherapy (Wallace et al, 2005). The effective sterilising dose (ESD: dose of fractionated RT [Gy] at which premature ovarian failure occurs immediately following treatment in 97.5% of patients) decreases with increasing age at treatment. ESD at birth is 20.3Gy; at 10 yrs 18.4Gy; at 20 yrs 16.5Gy, and at 30 yrs 14.3Gy(see fig three). We have calculated 95% confidence limits for age at premature ovarian failure for estimated radiation doses to the ovary from 1 Gy to the ESD from birth to 50 years.

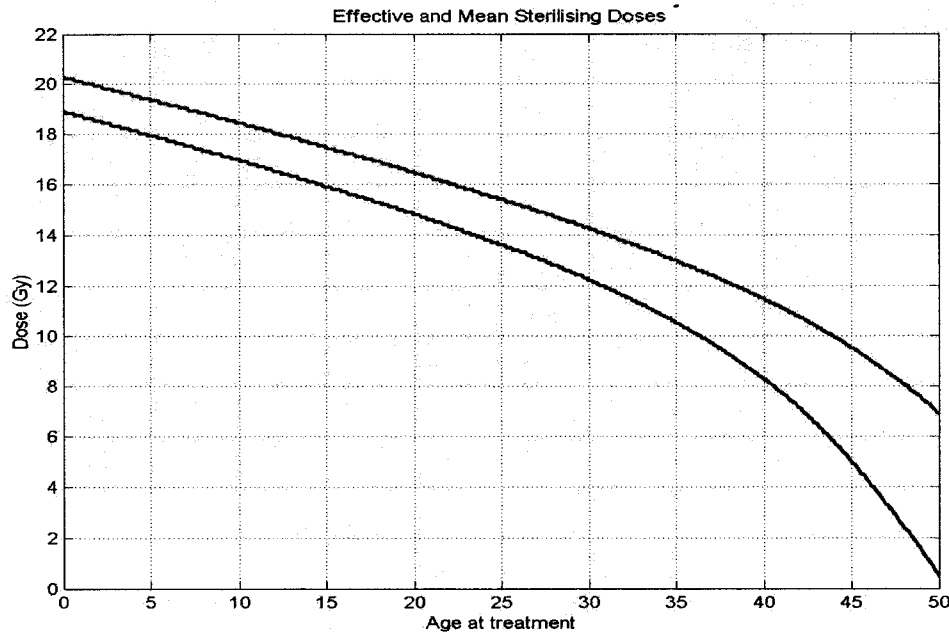


Figure three

The effective (red, upper) and mean (blue, lower) sterilizing dose of radiation for a known age at treatment

With no biochemical markers available to predict premature ovarian failure, a model that determines the extent of radiotherapy-induced damage and allows an accurate assessment of the 'fertile window' will have a significant impact on reproductive counselling for girls and young women with cancer.

Aims, Objectives & Research Hypotheses

Specific aims and objectives:

- Revise the determination of the radiosensitivity of the human oocyte, using our solution of the Faddy-Gosden solution of follicle decline.
- Revise our estimate of the estimated sterilising (ESD) dose for age.
- Revise our estimate of the age at premature ovarian failure for estimated radiation doses to the ovary from 1 Gy to the effective sterilizing dose from birth to 50 years.

Analysis Framework

Subject population:

All CCSS cases who have received radiation therapy to a field that includes the ovaries and are known to have developed premature ovarian failure. The range of diagnoses includes, brain tumours (Medulloblastoma, Ependymoma) receiving cranio-spinal radiotherapy, Hodgkins Lymphoma, Wilms' tumour, Leukaemia, Neuroblastoma and Osteosarcoma. So far 245 cases have been identified who have developed premature ovarian failure all of whom have detailed estimates of dose of radiation received by the ovaries using the methodology of Stovall et al. (2004)

The exploratory variables are:

- (i) age at treatment
- (ii) diagnosis, age at follow up, time since diagnosis, doses of chemotherapeutic drugs, alkylating agent score.
- (iii) total dose of fractionated radiation received by either both ovaries or ovary furthest from radiation field. Estimation of ovarian dose received.
- (iv) age at ovarian failure for each member of the subject population.

Methodology:

The Faddy-Gosden model for primordial follicle decline with age is the solution of $dy/dx = -y[0.0595 + 3716/(11780 + y)]$, where x denotes age and y denotes follicle population, with initial value (population at birth) $y(0) = 701200$.

Our underlying modelling assumptions are:

- (i) ovarian failure occurs when primordial follicle population falls below about 1,000, at around age 51 in average healthy women.
- (ii) The Faddy-Gosden model for primordial decline with age holds both before and after cyto-toxic insult . In other words, women and girls are assumed to have primordial follicle populations close to that predicted by the Faddy-Gosden model at age of treatment, chemo- and/or radiotherapy reduces the population, population decline then follows a Faddy-Gosden trajectory with ovarian failure occurring before the average expected age of 51 years.
- (iii) Later (respectively earlier) than average age of menopause corresponds to a higher (resp. lower) initial value for the Faddy-Gosden differential equation.

The first calculations will be analogous to those performed in (Wallace et al., 2003): effects of chemotherapy will be discounted. For each patient surviving fractions will be estimated by subtracting the age at premature ovarian failure from 51 -- this value being the increase in reproductive age due to radiotherapy -- and obtaining the Faddy-Gosden solution for age 51 less age at ovarian failure plus age at treatment. This population after treatment as a proportion of the pre-treatment

population gives the surviving fraction. The dose of radiation received by each ovary will be estimated from dummies (Stovall et al.,2004). A dose response relationship for each patient will be constructed, and the LD₅₀ calculated, as described in Figure 2. The LD₅₀ values will provide a mean and standard deviation.

For the second stage we segregate the cohort in terms of the likely impact of any chemotherapy received. For example, **minimal** could correspond to an estimated increase in reproductive age of 0—3 years, **moderate** to 4—6 years and **severe** to 7—10 years. Given these estimates, we can then revise our estimate for each LD₅₀ calculated in stage 1 by reducing the primordial follicle population before treatment by a factor that takes chemotherapy into account.

Special Consideration

There are a number of important assumptions and confounders inherent in our proposed analysis:

- The role of chemotherapy, particularly alkylating agents, in contributing to the development of ovarian failure will make our assessment of the radiosensitivity of the human oocyte an upper limit.
- The age at which premature ovarian failure occurs can be reliably determined from a questionnaire study, without confirmation by measuring gonadotrophins.
- The use of the oral contraceptive pill will mask the subjects appreciation of the age at which ovarian failure occurs. Therefore women on the oral contraceptive pill, who are experiencing regular withdrawal bleeds may have developed premature ovarian failure unbeknown to themselves
- How accurate is the determination of the dose received by the ovary? The exact location of each ovary during fractionated radiotherapy is both unknown and may vary from day to day.

References:

Critchley HOD, Wallace WHB, Mamtora H, Higginson J, Shalet SM, Anderson DC. Ovarian failure after whole abdominal radiotherapy: The potential for pregnancy. *British Journal of Obstetrics and Gynaecology* (1992) 99: 3392-394.

Faddy MJ, Gosden RG. A model confirming the decline in follicle numbers to the age of menopause in women. *Hum Reprod* 1996;11:1484-1486.

Kalapurakal JA, Peterson S, Peabody EM et al. Pregnancy outcomes after abdominal irradiation that included or excluded the pelvis in childhood Wilms tumour survivors: A report from the National Wilms tumour study. *Int. J Radiation Oncology Biol. Phys.*,2004;58:1364-1368.

Stovall M, Donaldson SS, Weathers RE et al. Genetic effects of radiotherapy for childhood cancer: gonadal dose reconstruction. *Int. J Radiation Oncology Biol. Phys.*,2004; 60: 542-552.

Wallace WHB, Shalet SM, Crowne EC, Morris-Jones PH, Gattamaneni HR. Ovarian failure following abdominal irradiation in childhood: Natural history and prognosis. *Clinical Oncology* (1989) 1: 75-79.

Wallace WHB, Shalet SM, Hendry JH, et al. Ovarian failure following abdominal irradiation in childhood: The radiosensitivity of the human oocyte. *Br J Radiol* 1989;62: 995-998

Wallace WHB, Thomson AB, Kelsey TW. The radiosensitivity of the human oocyte. *Hum Reprod* 2003;18: 117-121.

Wallace WHB, Thomson AB, Saran F, Kelsey TW. Predicting ovarian failure after radiation to a field that includes the ovaries. *Int J Radiation Oncology Biol. Phys.* 2005;62(3):738-744.