Fertility preservation in children and young people with cancer – what are the remaining challenges ?

Professor W Hamish B Wallace

Consultant Paediatric Oncologist,

Royal Hospital for Sick Children,

Edinburgh

Scotland, UK

hamish.wallace@nhs.net

* Oncofertility Consortium, Chicago, 2012

Disclosure slide

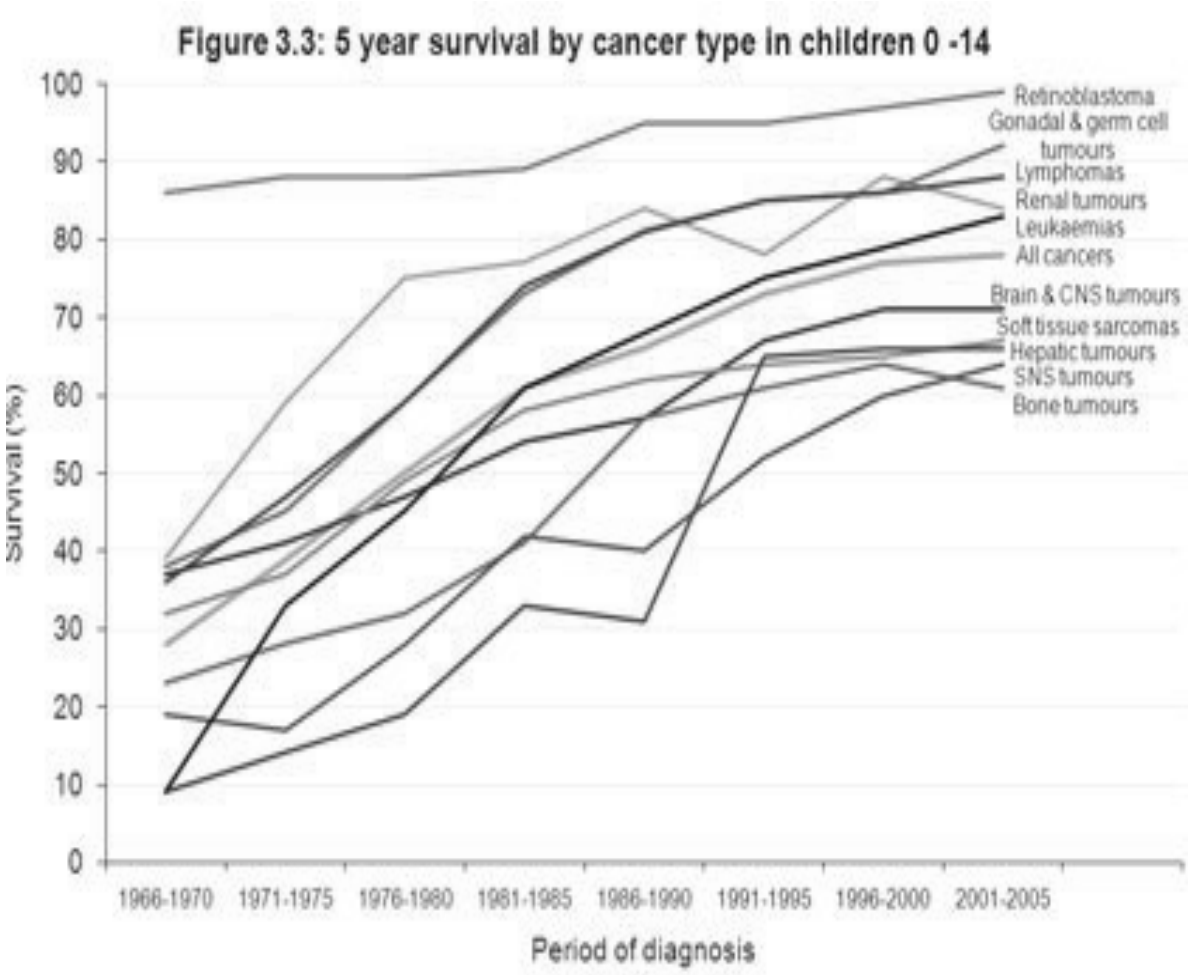
* No financial or competing interests to declare

Summary of Talk

* A young girl with advanced Hodgkin's lymphoma
* Fertility risk assessment
* Assessment of Ovarian Reserve
* What can be offered to those at high risk of infertility ?
* Our Edinburgh experience of ovarian cryopreservation
* In vitro growth and maturation

Improved survival rates

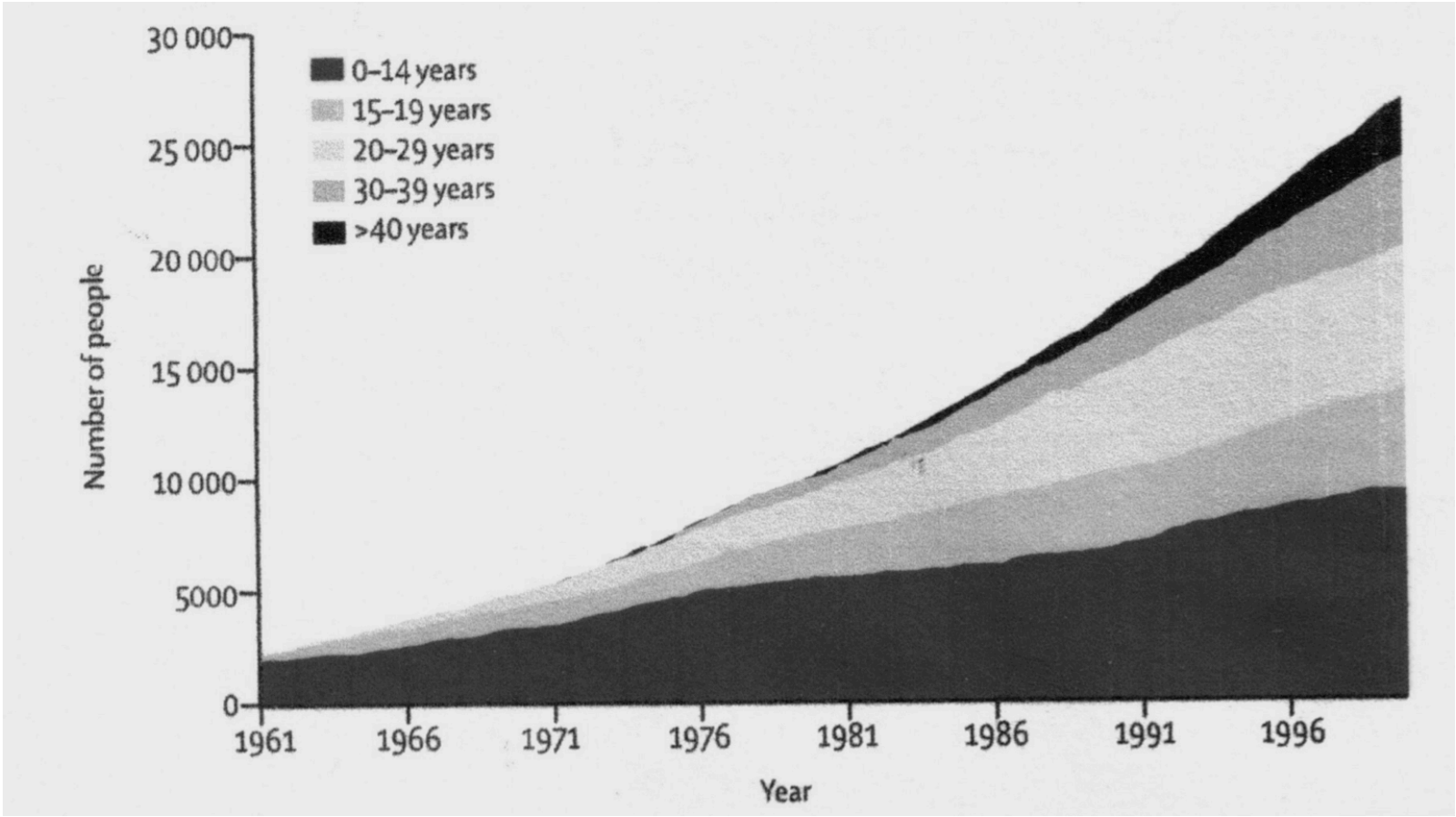
- Paediatric oncology units
- Clinical trials
- Intensifying treatment
- Supportive care



Epidemiology of Childhood Cancer

- Cumulative Risk of childhood cancer: 1 in 444 boys; 1 in 594 girls (1500 cases/yr in UK)
- >75% of children with cancer will survive five years, 70% are ten year survivors
- 1 in 570 young adults (20-34 years) is a childhood cancer survivor in UK
- By 2010, one in 715 of the adult population was a long term survivor of childhood cancer in UK

Increasing numbers of five year UK survivors by current age



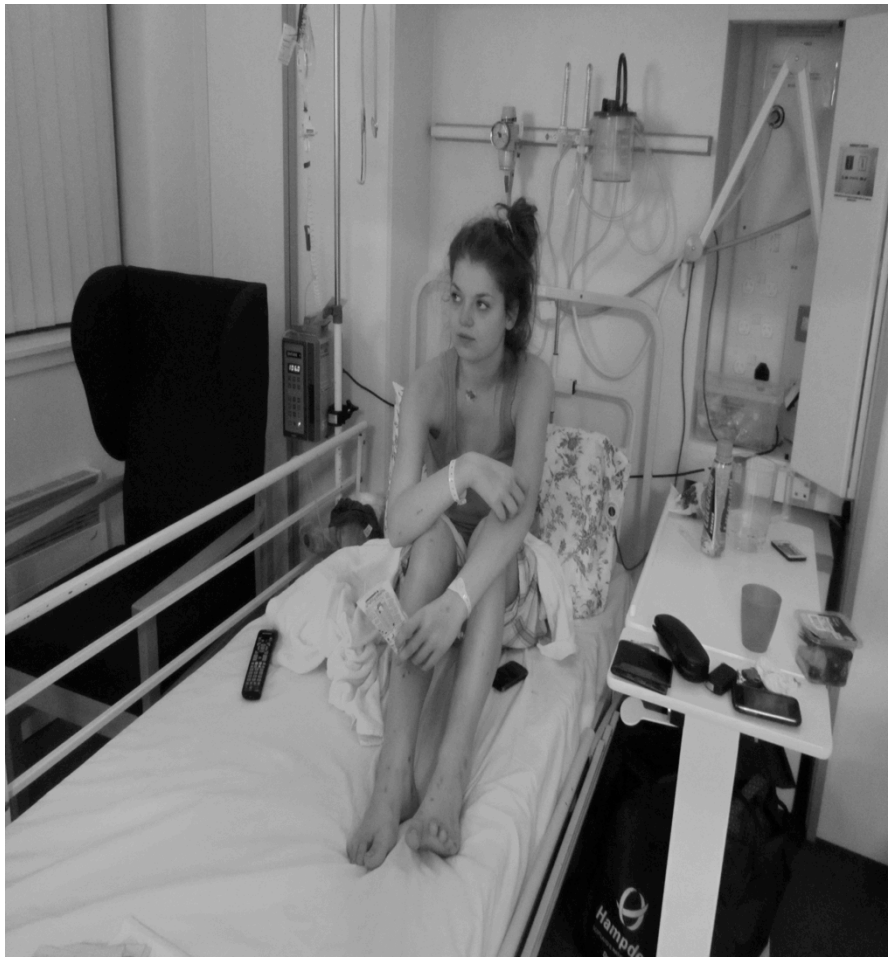
Skinner et al, Lancet Oncology, 2006

Cure at a cost

Sustain survival rates

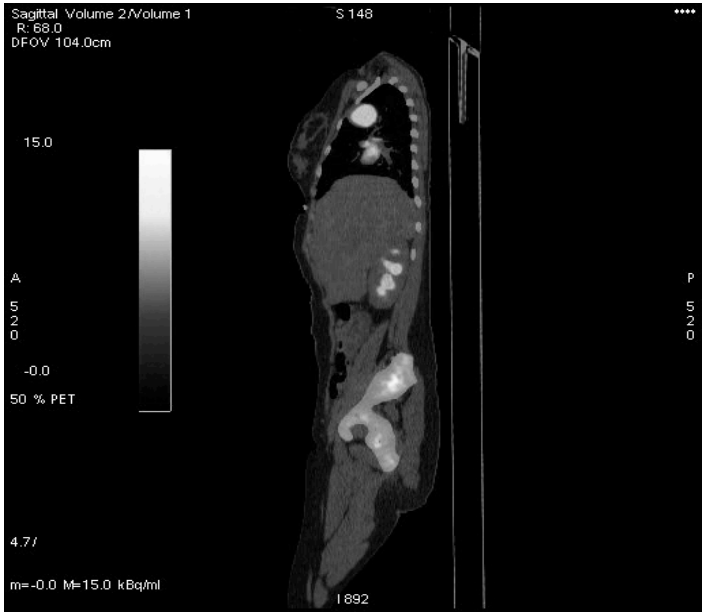
Minimise late effects

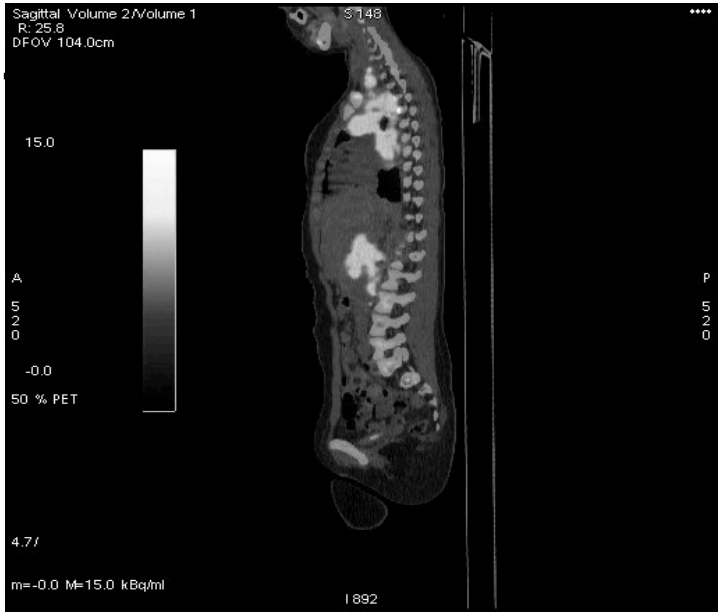
Laura Age 15

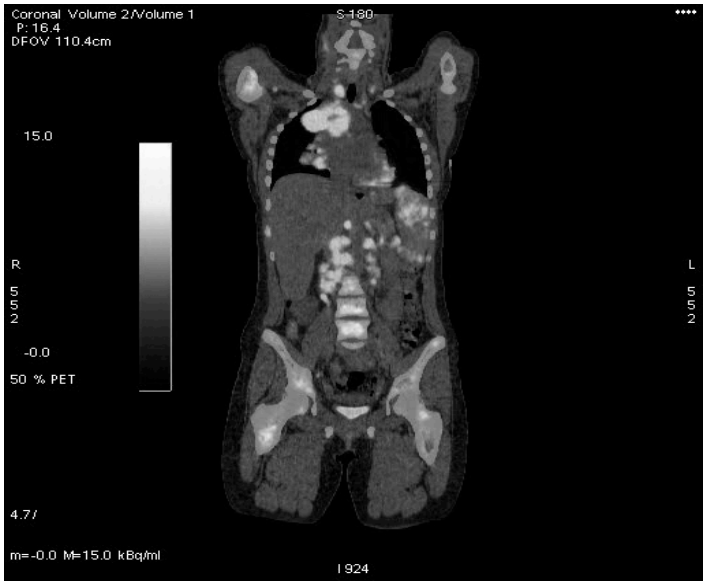


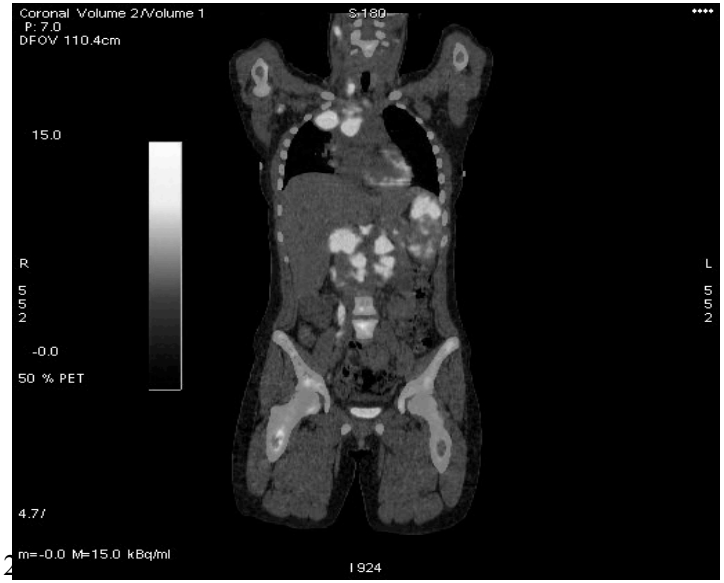
* Nodular sclerosing Stage IVB Hodgkin lymphoma
* Bilateral supraclavicular, right axillary, anterior mediastinal, paratracheal, right hilar, posterior mediastinal, coeliac, portal, retroperitoneal, internal and external iliac and deep inguinal lymph nodes
* Bone marrow infiltration

en


Sagittal Volume 2/Volume 1
S 148
R: 68.0
DFOV 104.0cm
15.0
A
P
-0.0
50 % PET
4.7/
m=-0.0 M=15.0 kBq/ml
I 892


Sagittal Volume 2/Volume 1
S 148
R: 25.8
DFOV 104.0cm
15.0
A
P
-0.0
50 % PET
4.7/
m=-0.0 M=15.0 kBq/ml
I 892


Coronal Volume 2/Volume 1
S 180
P: 16.4
DFOV 110.4cm
15.0
R
L
-0.0
50 % PET
4.7/
m=-0.0 M=15.0 kBq/ml
I 924


Coronal Volume 2/Volume 1
S 180
P: 7.0
DFOV 110.4cm
15.0
R
L
-0.0
50 % PET
4.7/
m=-0.0 M=15.0 kBq/ml
I 924

Risk assessment for Fertility preservation

* Intrinsic factors
  * Health status of patient
  * Consent (Patient/Parent)
  * Assessment of ovarian reserve
* Extrinsic factors
  * Nature of predicted treatment
    * High/Medium/Low/Uncertain Risk
  * Time available
  * Expertise available

Wallace WH et al. JCO, 2012

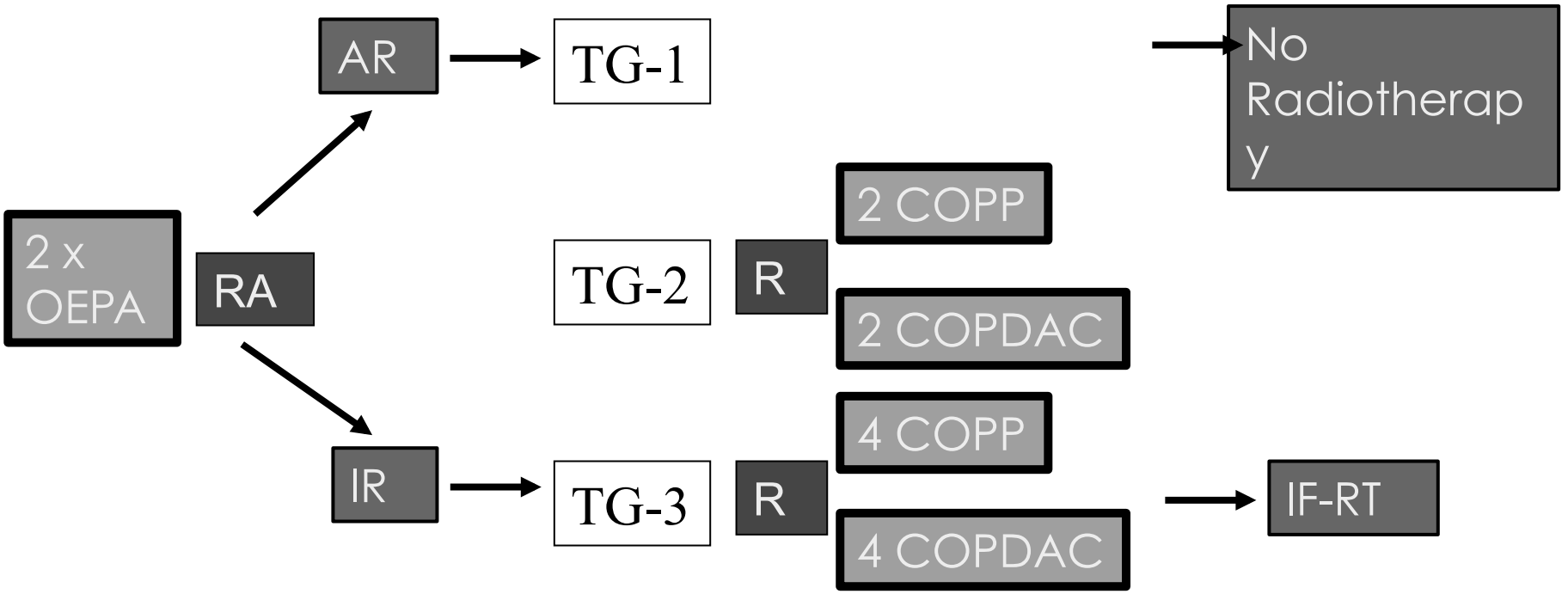
Laura Age 15

* Surgical procedures

* Mediastinal lymph node biopsy
* Insertion of double lumen portacath

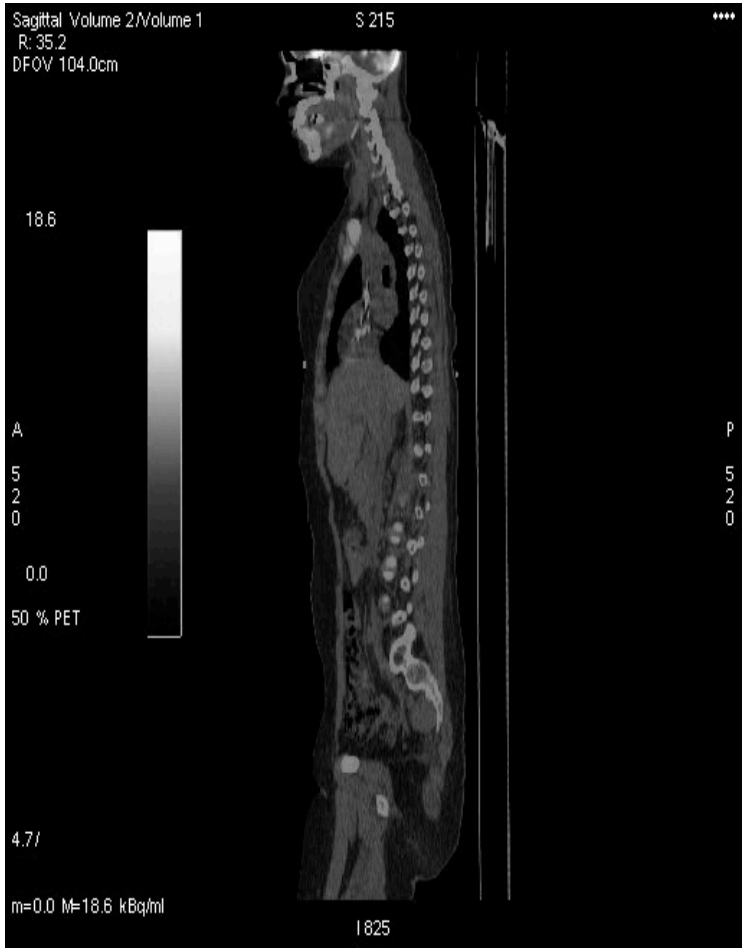
* Laparoscopic ovarian biopsy and cryopreservation of ovarian cortical strips

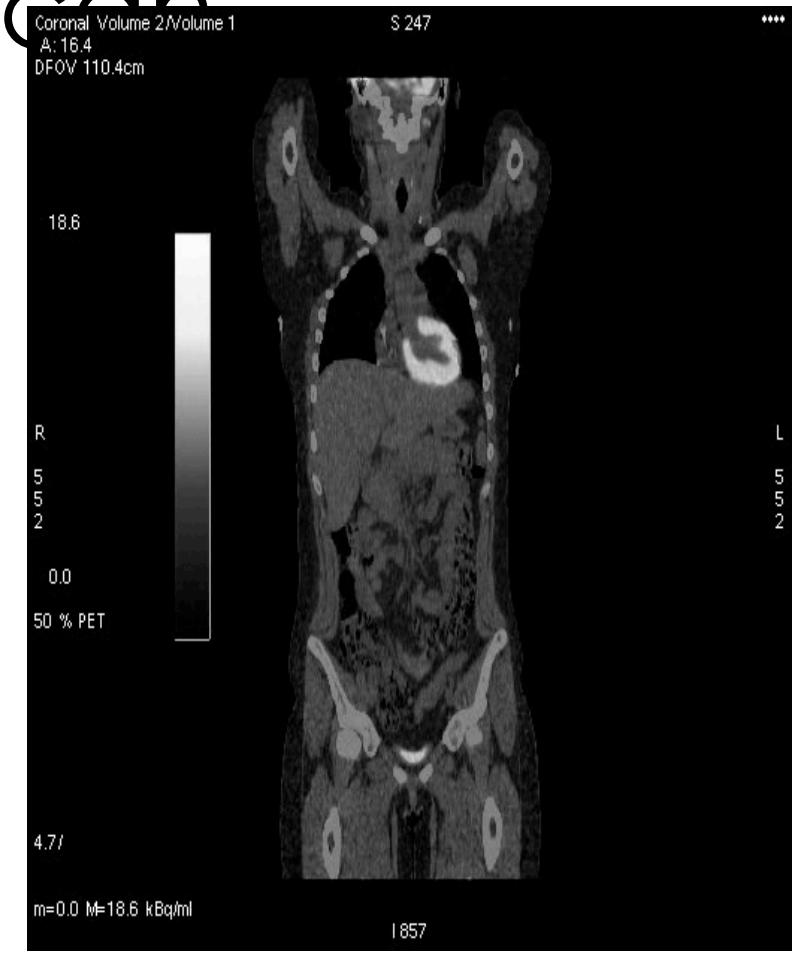
EuroNet-PHL-C-1
Chemotherapy randomisation



Laura: During Chemotherapy on the TCT Unit at RHSC

Response assessment PET scan



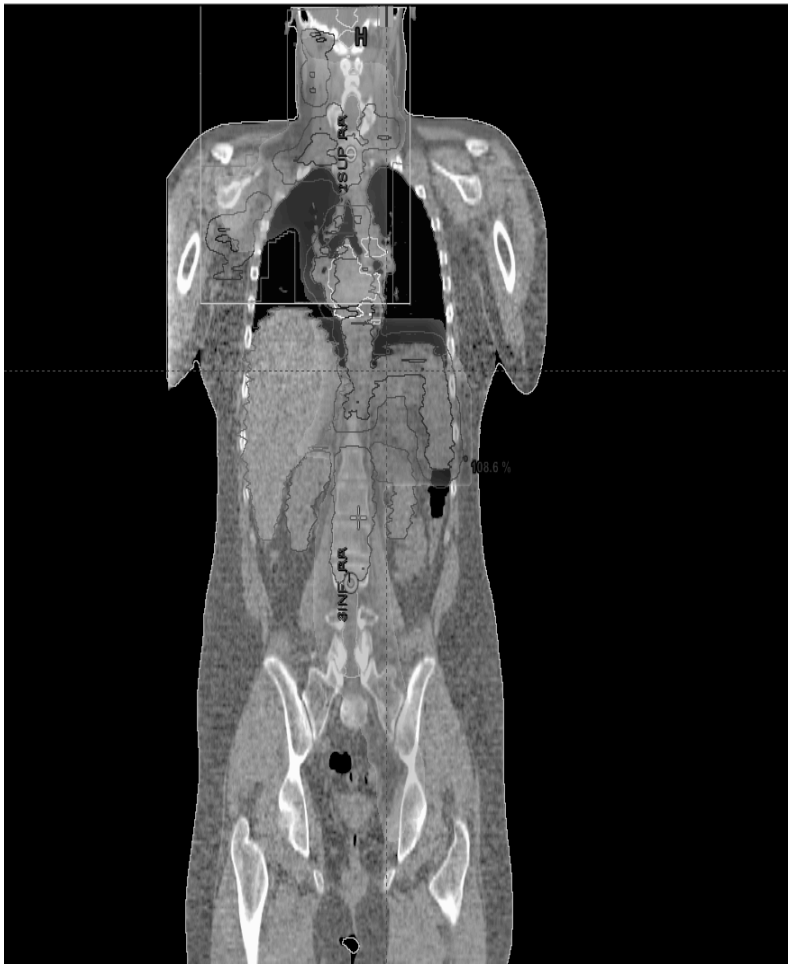


Laura: Response assessment

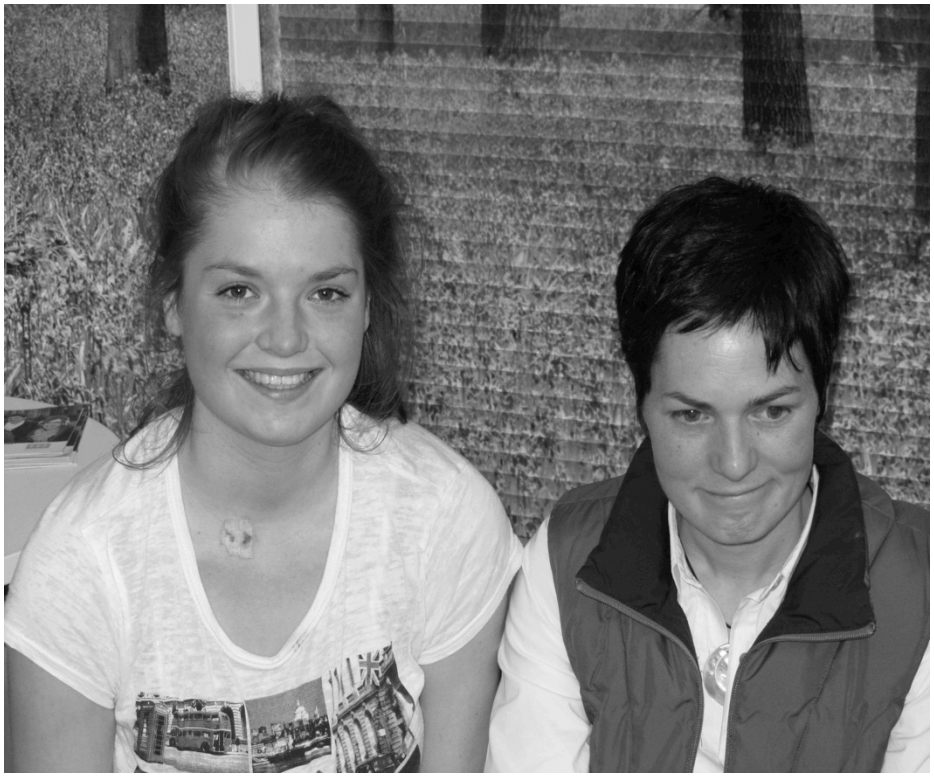
* > 50% decrease in the volume of all involved areas
* Largely PET scan negative
* -but still positive in a few small areas on central review
* Involved Field Radiotherapy

Estimated dose to organs at risk

Organs at risk		
	Maximium dose received	Mean Dose
- spinal cord	2139.7 cGy	1916.2 cGy
- heart	2116.1 cGy	1701.4 cGy
- left kidney	2169.1 cGy	1439.8 cGy
- right kidney	2022.2 cGy	639.3 cGy
- lung	2148.5 cGy	1168.9 cGy
- right breast	2195.1 cGy	476.7 cGy
- left breast	2156.4 cGy	654.6 cGy
- liver	2153.4 cGy	830.2 cGy
- thyroid	2047.2 cGy	1999.0 cGy



After Treatment:
with a famous guest!



Oncofertility Consortium, Chicago, 2012

Risk assessment for Fertility preservation

* Intrinsic factors
  * Health status of patient
  * Consent (Patient/Parent)
  * Assessment of ovarian reserve
* Extrinsic factors
  * Nature of predicted treatment
    * High/Medium/Low/Uncertain Risk
  * Time available
  * Expertise available

Wallace WH et al. JCO, 2012

Risk of infertility

Low risk (<20%)	Medium risk	High risk (>80%)
ALL Wilms' tumour Brain tumour Sx, RT < 24Gy Soft tissue sarcoma (stage1) Hodgkin's Lymphoma HL(Low stage)	AML Osteosarcoma Ewing's sarcoma STS: stage II/III Neuroblastoma NHL Brain tumour RT>24Gy HL (High Stage)	Total Body Irradiation Pelvic/testes RT Chemo pre BMT Metastatic Ewing's HL (Pelvic RT)

Infertility - Risk Factors

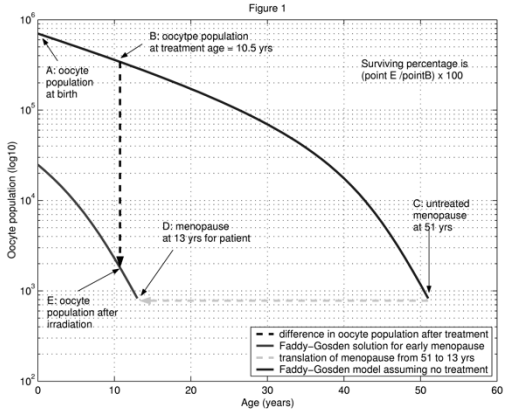
- RT to HPA or a field that includes testes/ovaries
- Busulphan
- BCNU
- CCNU
- Cyclophosphamide
- Ifosfamide
- Melphalan
- Mustine
- Nitrogen mustard
- Procarbazine
- Thiotepa
- Chlorambucil
- Cytarabine

The pre-pubertal gonad is not protected

Radiation-induced ovarian damage

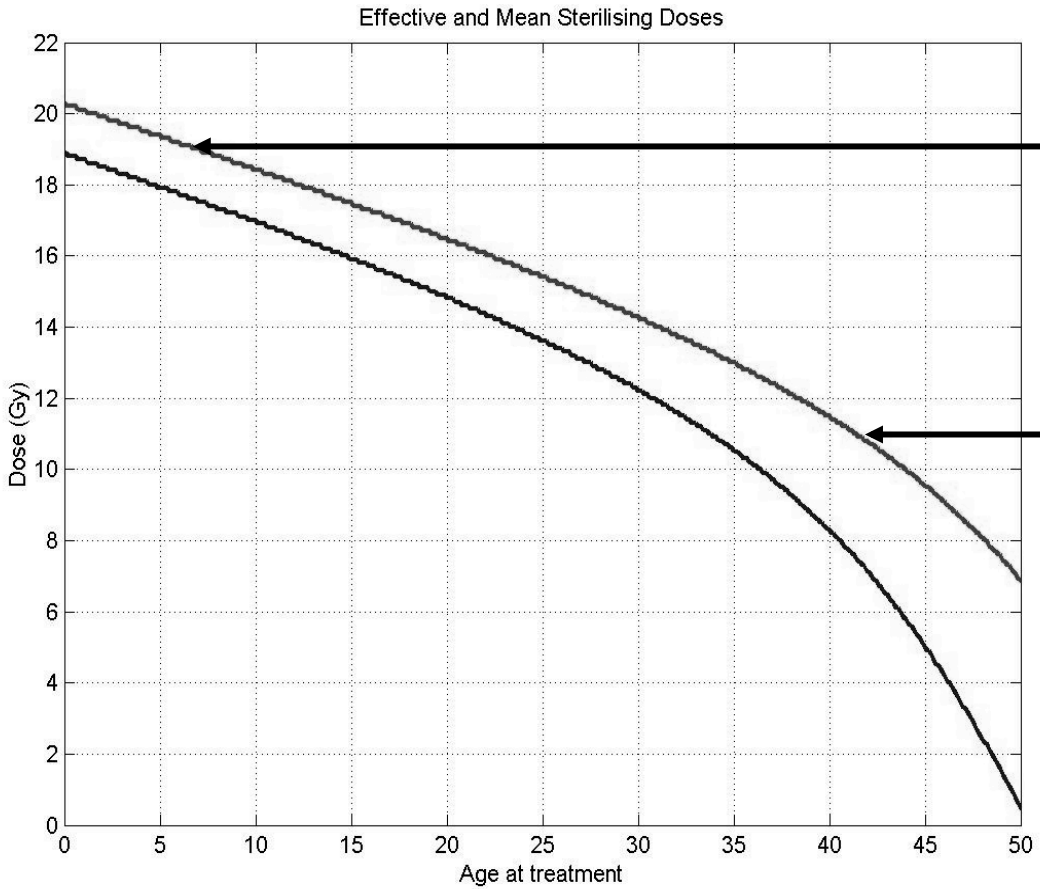
Human oocyte
(Primordial follicle)

* $LD_{50} < 2$ Gy



Wallace et al. (2003) Hum Reprod.

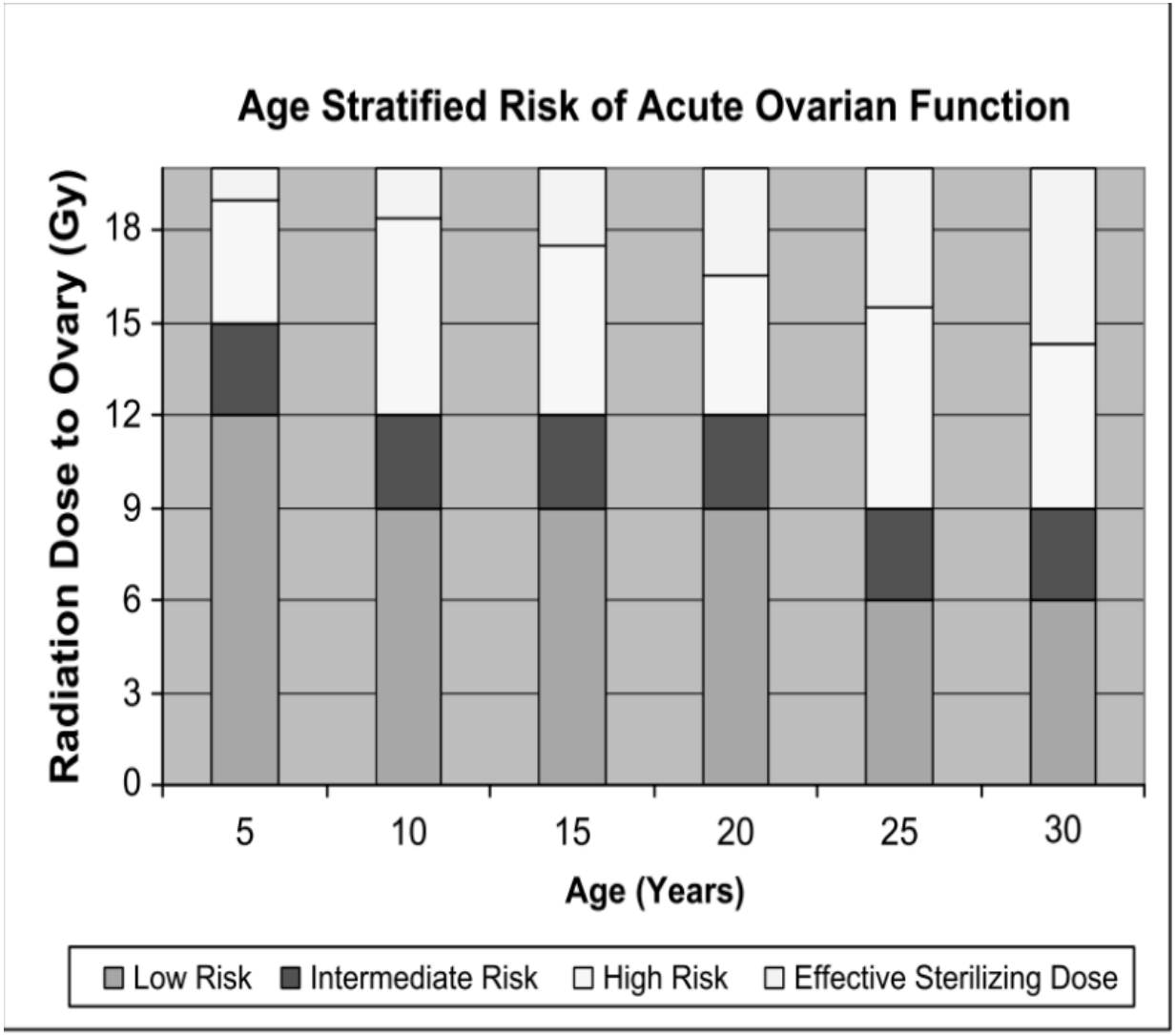
Effective and mean ovarian sterilizing doses of radiotherapy at increasing age



19 Gy will sterilize at 7 years

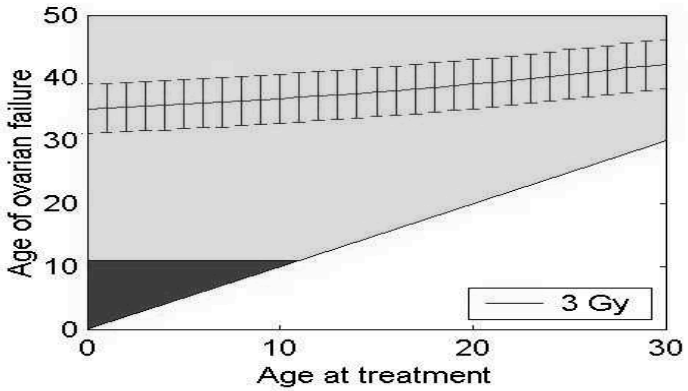
11 Gy will sterilize at 42 years

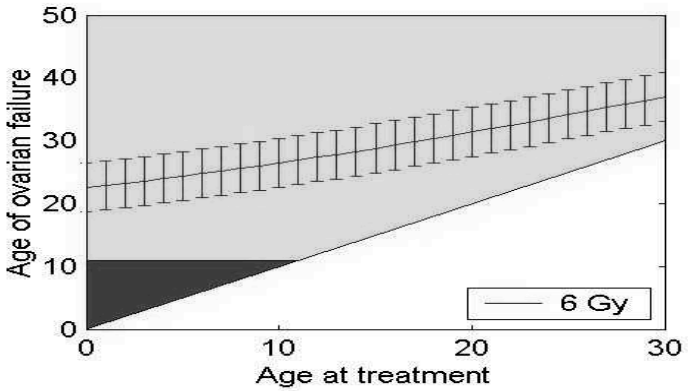
Wallace WH et al. IJRBP (2005)

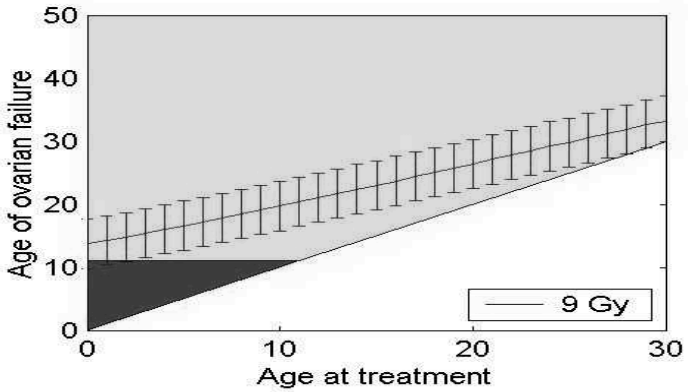


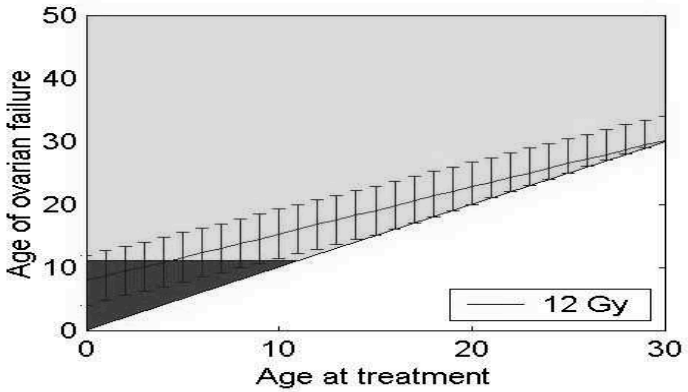
Wallace WH et al.
IJRBP (2005)

Age prediction for development of ovarian failure after known dose of radiotherapy

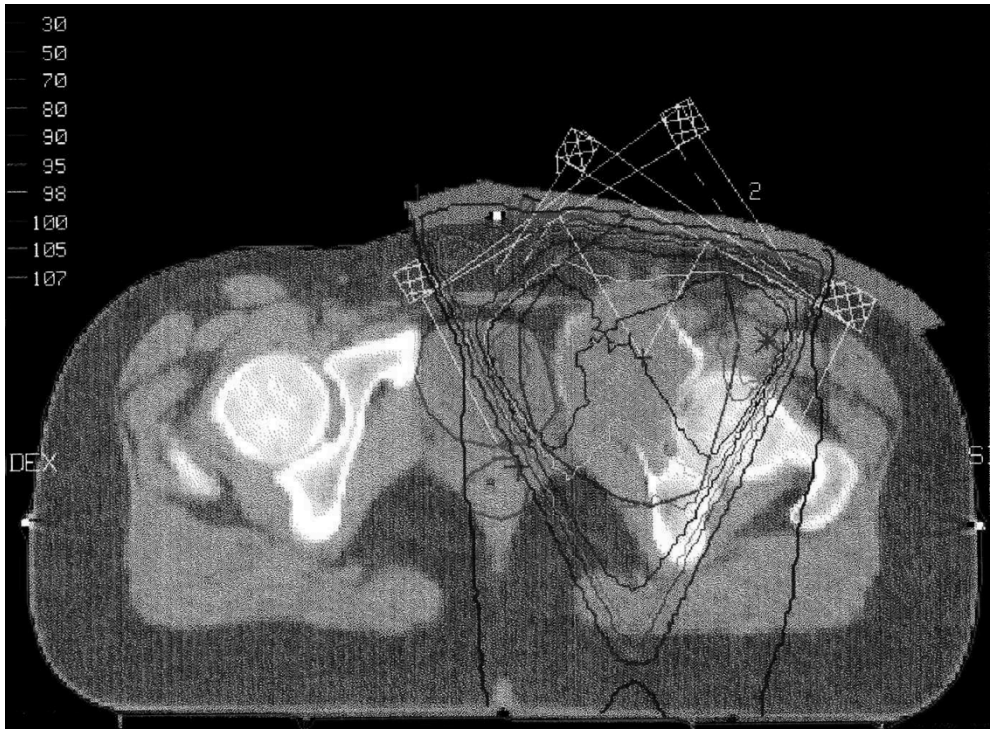








CT planning for pelvic Ewing's sarcoma



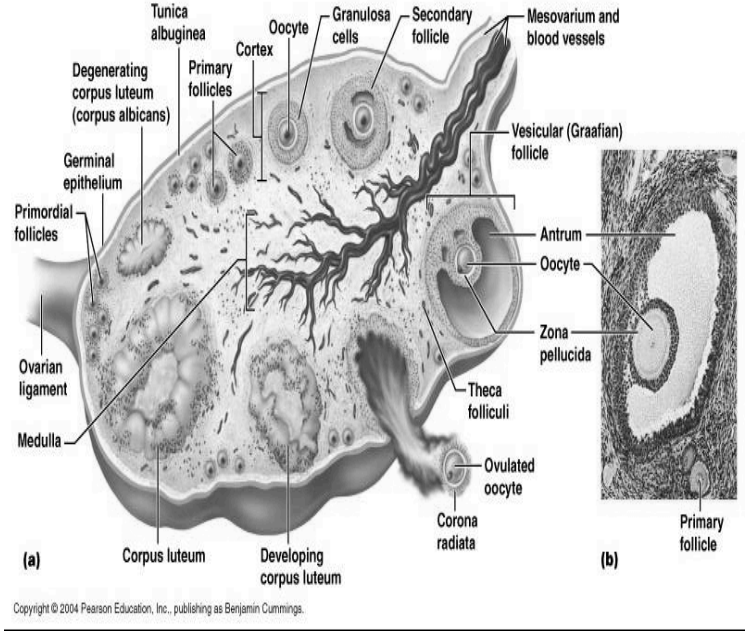
Oncofertility Consortium, Chicago, 2012

Risk assessment for Fertility preservation

* Intrinsic factors
  * Health status of patient
  * Consent (Patient/Parent)
  * Assessment of ovarian reserve
* Extrinsic factors
  * Nature of predicted treatment
    * High/Medium/Low/Uncertain Risk
  * Time available
  * Expertise available

Wallace WH et al. JCO, 2012

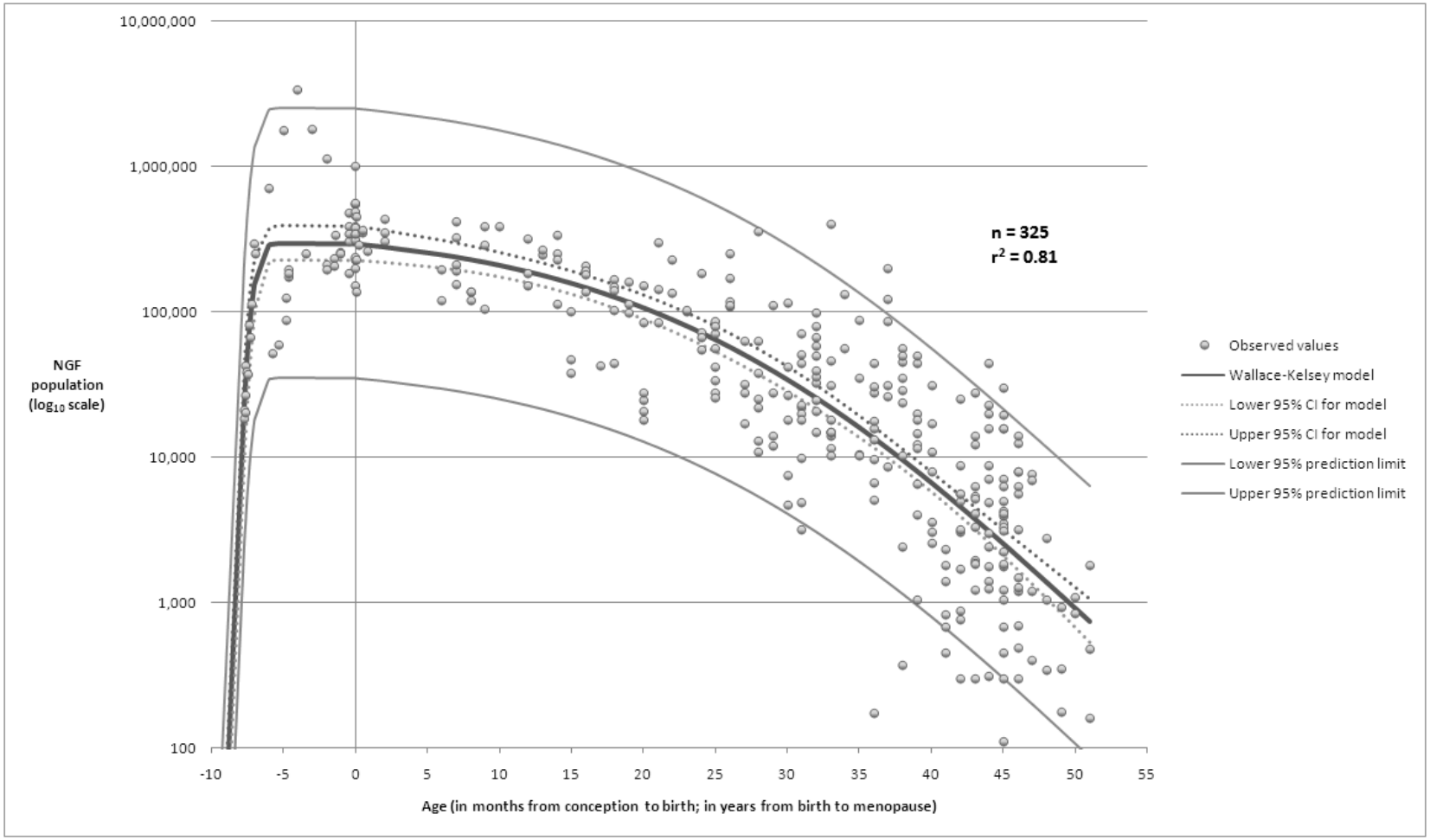
The Egg Store in Quito, Ecuador!



Data set:
Eight quantitative histological studies

Study			Statistics			
Number	First author	Year	No. ovaries	Min. age	Max. age	Median age
1	Bendsen	2006	11	−0.6	−0.6	−0.6
2	Baker	1963	11	−0.6	7.0	−0.2
3	Forabosco	2007	15	−0.5	0.5	−0.3
4	Block	1953	19	−0.2	0.0	0.0
5	Hansen	2008	122	0.1	51.0	38.0
6	Block	1951	86	6.0	44.0	28.0
7	Gougeon	1987	52	25.0	46.0	39.5
8	Richardson	1987	9	45.0	51.0	46.0
Overall			325	−0.6	51.0	32.0

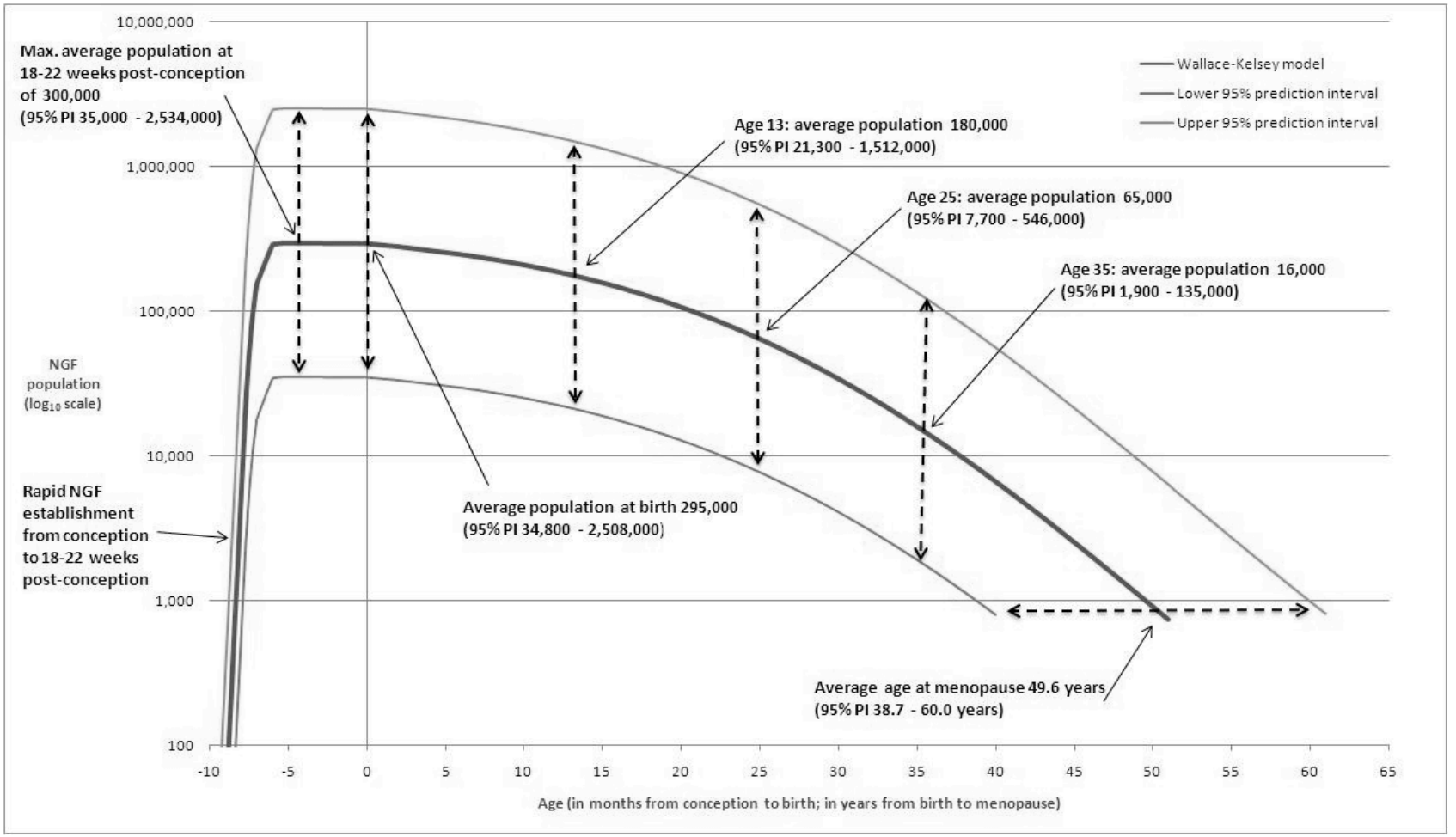
The Wallace-Kelsey Model



Oncofertility Consortium, Chicago, 2012

Wallace &Kelsey (2010) PloS ONE

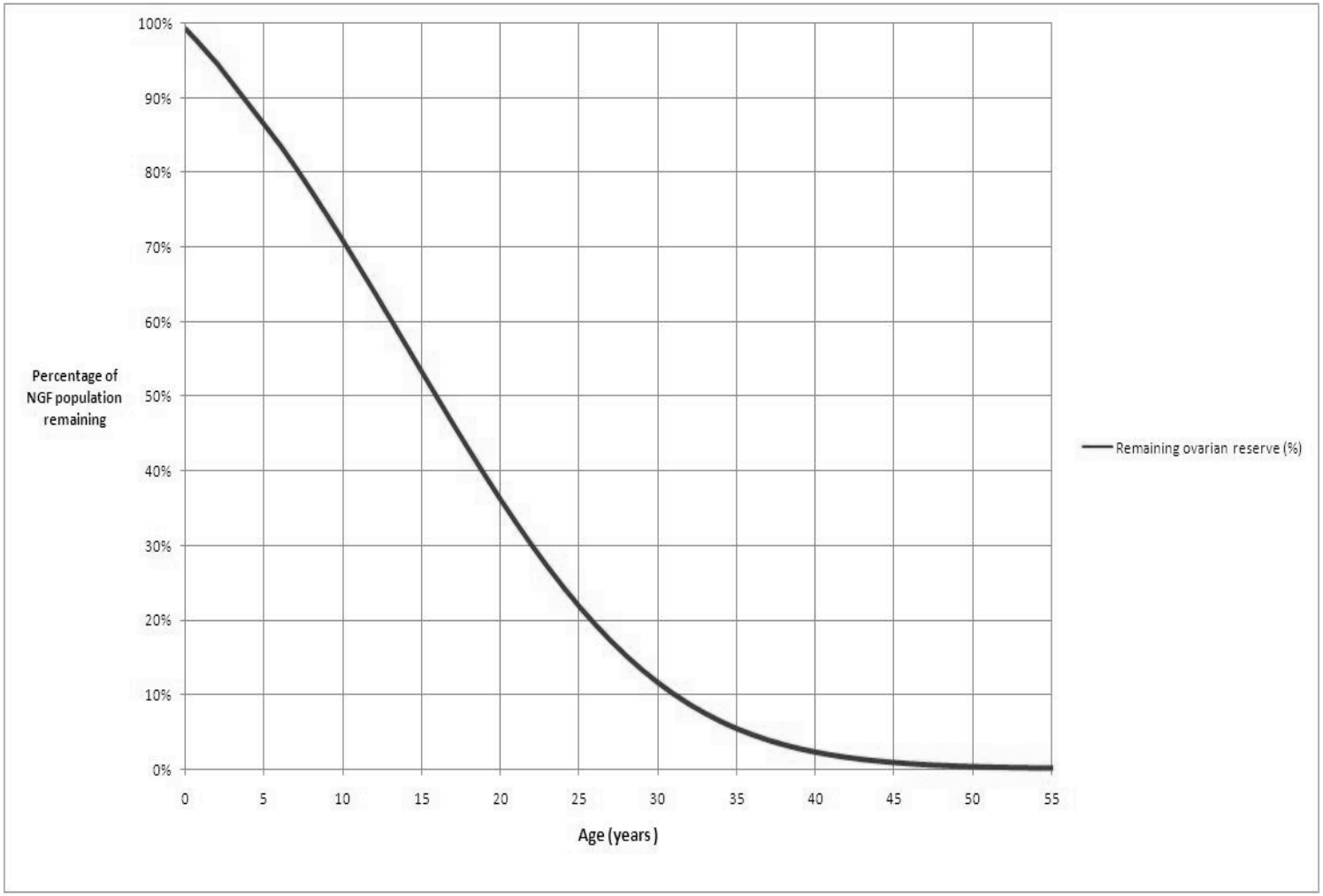
Ovarian reserve: Conception to Menopause (NGF population)



Oncofertility Consortium, Chicago, 2012

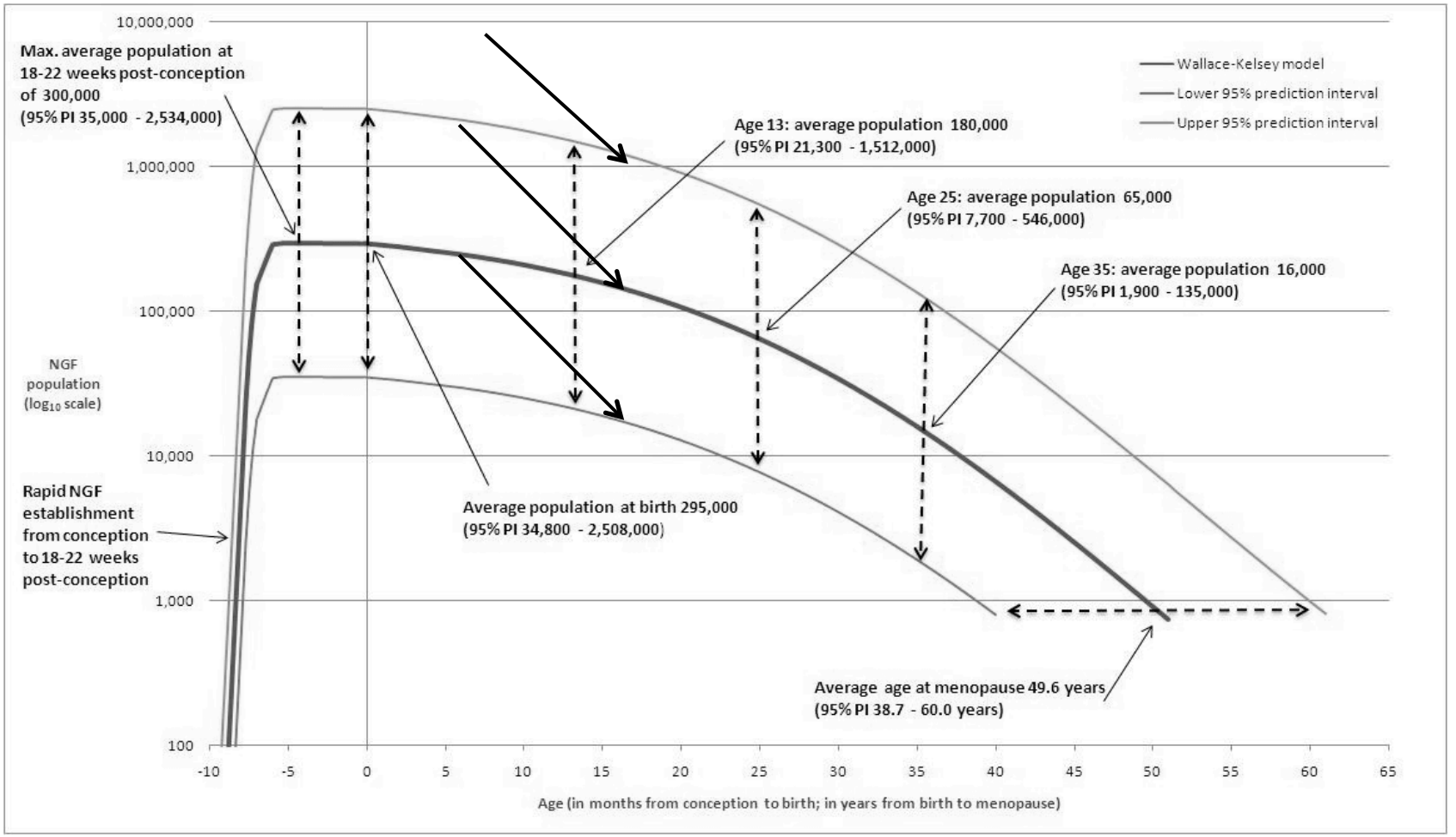
Wallace &Kelsey (2010) PloS ONE

Percentage of NGF population remaining with increasing age



Wallace &Kelsey (2010) PloS ONE

Ovarian reserve:
A Validated model from Conception to Menopause
(NGF population)



Oncofertility Consortium, Chicago, 2012

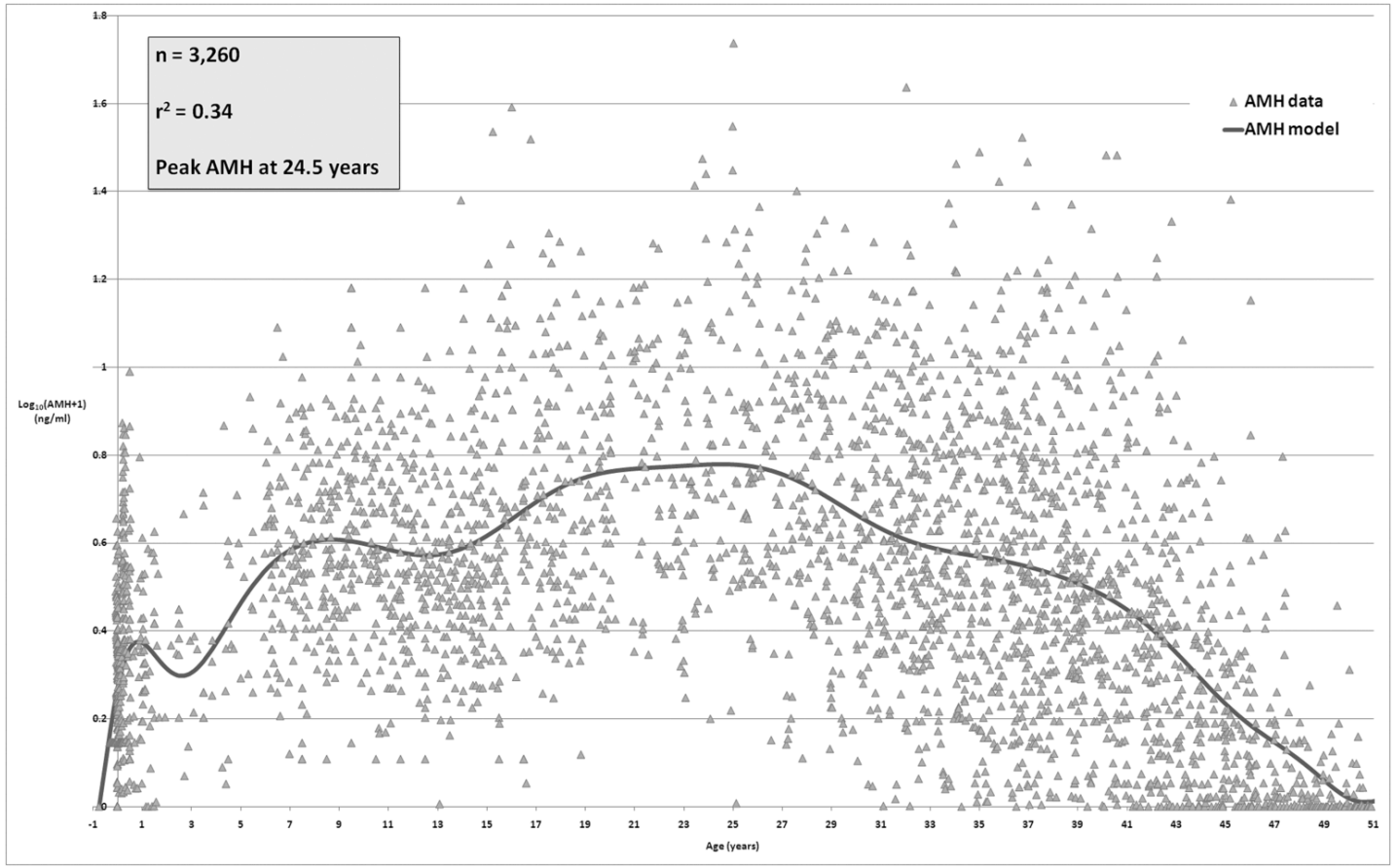
Wallace &Kelsey (2010) PloS ONE

Prediction of ovarian reserve

* Anti Mullerian Hormone (AMH) is an important product of the adult ovary, produced by the granulosa cells of small growing follicles
* AMH has little variation across and between menstrual cycles
* AMH is the best currently available marker of the number of small-growing follicles in the ovary

A validated model of serum anti-Mullerian hormone from conception to menopause

(a single data set of healthy females (n=3260) from twenty different sources)

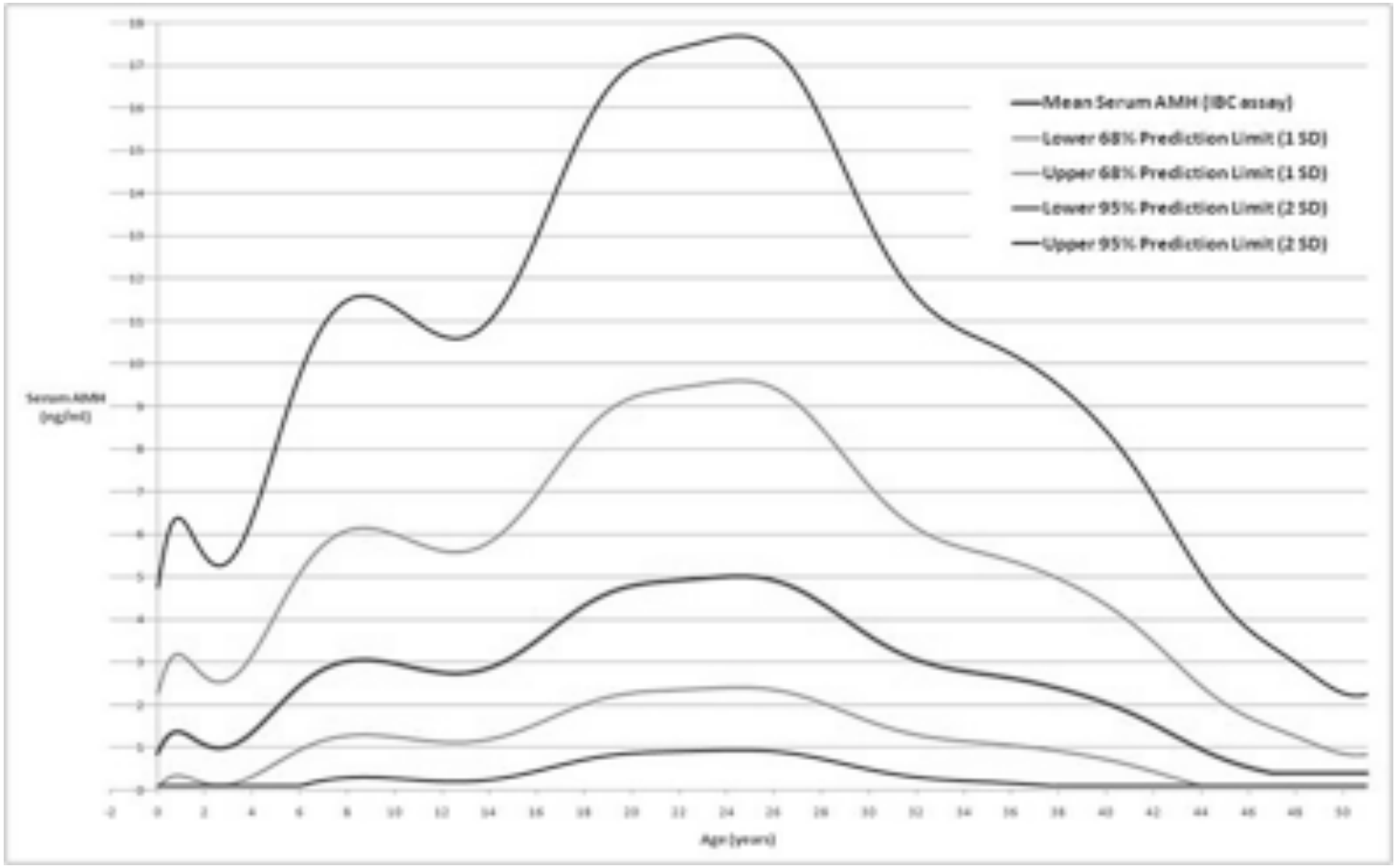


Oncofertility Consortium, Chicago, 2012

Kelsey et al. PLoS ONE 2011

AMH: Normogram from birth to menopause

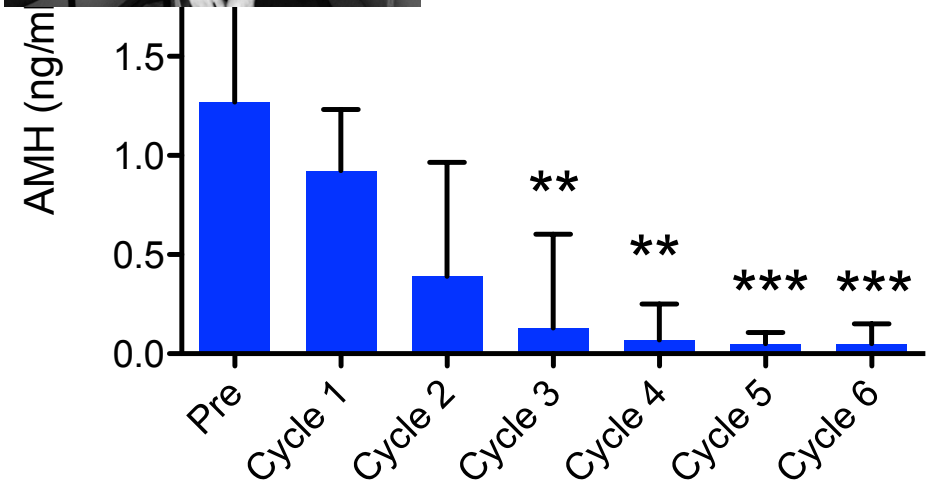
The green and blue lines are the 68% and 95% prediction limits for the model



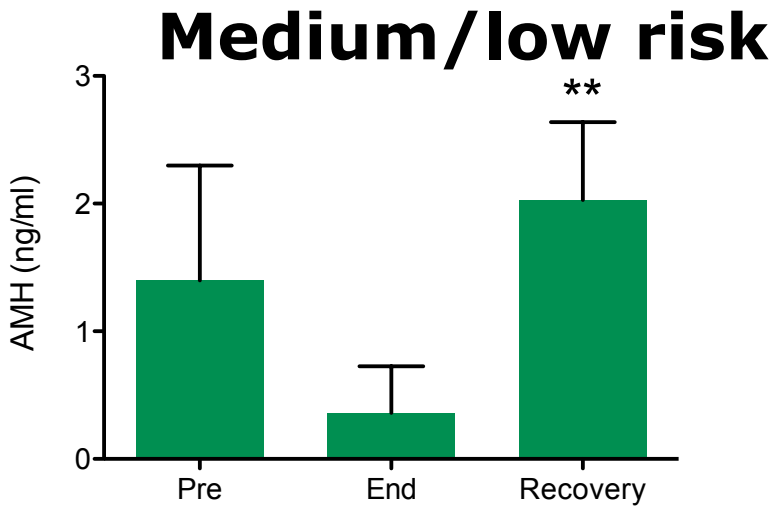
Oncofertility Consortium, Chicago, 2012

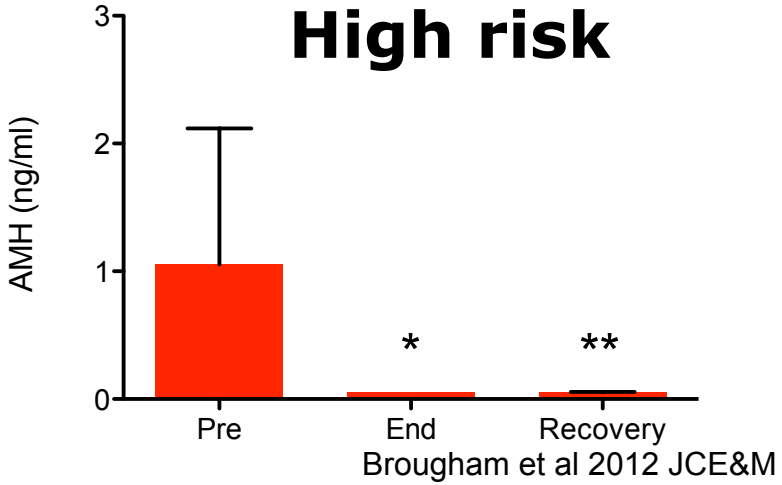
Kelsey et al. PLoS ONE 2011

AMH in childhood cancer



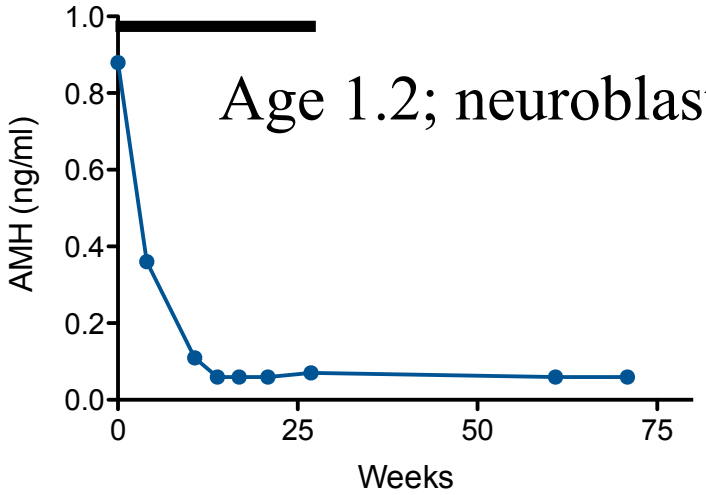
22 girls age 0.3-15yr
17 prepubertal



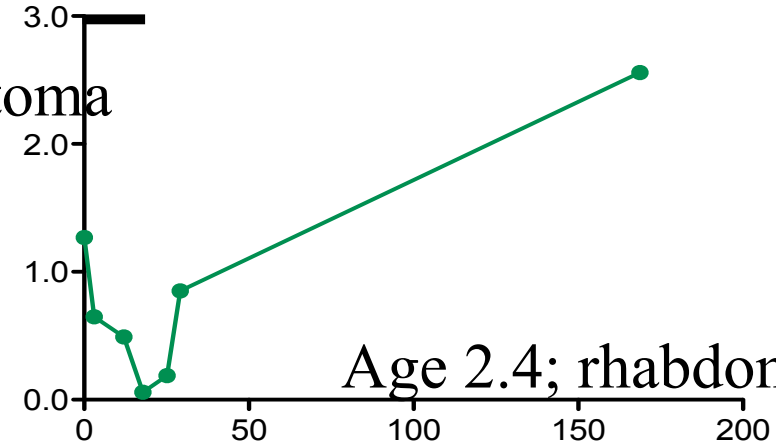


Brougham et al 2012 JCE&M

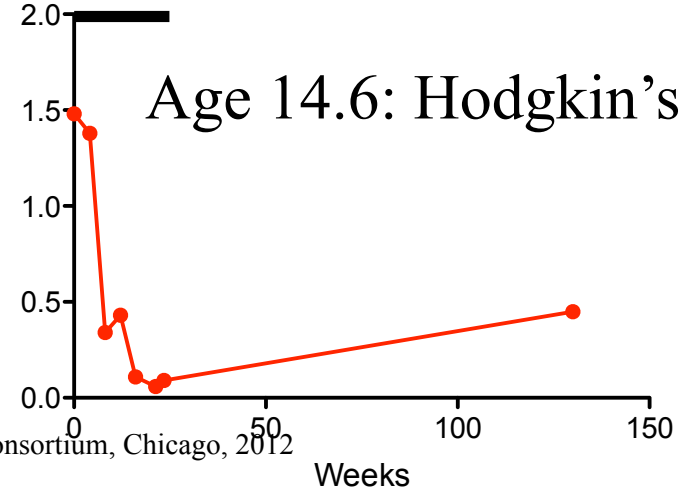
AMH in 3 girls with cancer



Age 1.2; neuroblastoma



Age 2.4; rhabdomyosarco



Age 14.6: Hodgkin's lymphoma

Brougham et al 2012 JCE&M

Summary

- AMH is detectable before puberty
- AMH falls rapidly during cancer treatment in both pre-pubertal and pubertal girls
- AMH levels recover in those patients at low/medium risk of gonadotoxicity
- AMH fails to recover in those at high risk. This could be indicative of future reproductive impairment
- AMH is therefore a clinically useful marker of damage to the ovarian reserve in girls aiding assessment and information provision

Brougham et al 2012 JCE&M

Oncofertility Consortium, Chicago, 2012

Key features of the 3 options for fertility preservation for women

Technique	Main advantages	Main disadvantages
Embryo cryopreservation	Established technique	May incur delay Sperm required: partner or donor Fixed potential for future fertility
Oocyte cryopreservation	Does not require sperm	May incur delay Not appropriate for pre-pubertal child Limited numbers of eggs can be stored in time available
Ovarian tissue cryopreservation	Minimal delay No lower age limit Allows for spontaneous and repeated conception Greater allowance for future developments	Requires surgical procedure Malignant contamination in some conditions precludes reimplantation In vitro follicle growth unlikely to be available for several years.

Oncofertility Consortium, Chicago, 2012

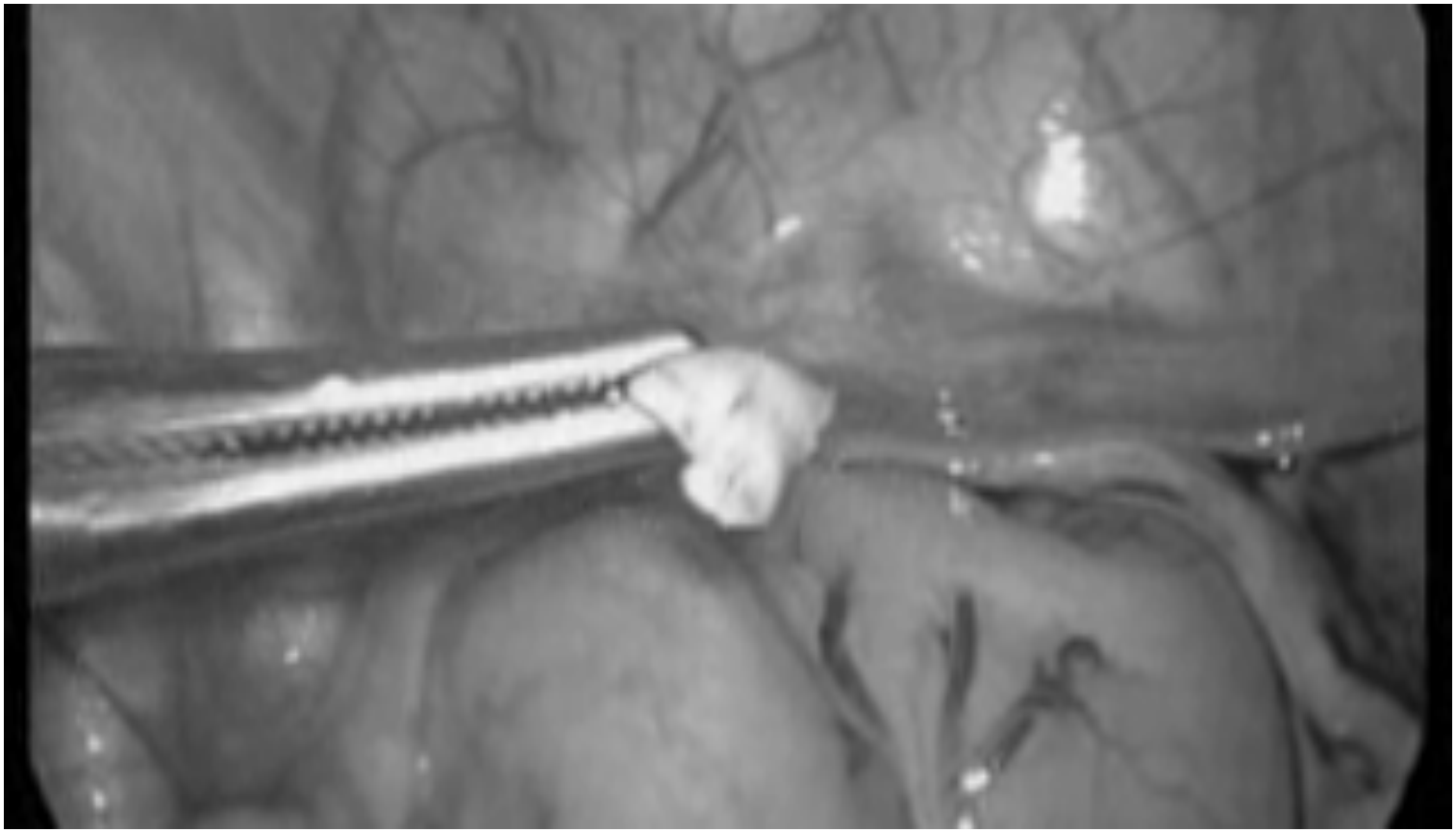
Ovarian cortical strips

- rich in primordial follicles
- survive cryopreservation
- technique validated in sheep

Live births following cryopreservation of ovarian tissue and transplantation

Diagnosis	Age (yrs)	Surgical method	Reimplantation	Pregnancy	Reference
Hodgkin's Lymphoma	25	Unilateral ovarian biopsy	Orthotopic	Spontaneous, live birth	Donnez, 2004
Non-Hodgkin's Lymphoma	28	Unilateral ovarian biopsy (after 1st course chemo)	Orthotopic (Both ovaries)	IVF, live birth	Meirow 2005; 2007
Hodgkin's Lymphoma	31	Unilateral ovarian biopsy (after 1st course chemo)	Ortho and heterotopic	Spontaneous, miscarriage then livebirth	Demeestere 2007
Hodgkin's lymphoma	27	Whole ovary	Orthotopic	Livebirth male Week 37 B.Wt 2.6 Kg	Andersen et al 2008
Ewings Sarcoma	36	Whole ovary	Orthotopic	Livebirth Female Term B Wt 3.2 Kg	Andersen et al 2008

Ovarian biopsy at laparoscopy



Oncofertility Consortium, Chicago, 2012

Cryopreservation: World-wide experience

* At least 20 pregnancies worldwide after othotopic reimplantation of frozen–thawed ovarian cortex
* Success rate is unclear as the denominator is unknown
* No pregnancies reported following the reimplantation of ovarian tissue harvested pre-pubertally
* Young children are potentially ideal candidates

Ovarian transplantation: World-wide experience

* Silber et al. have also extensively reported their experience of successful fresh ovarian transplantation in identical twins discordant for premature ovarian failure

* 12 pregnancies and eight healthy babies have been reported from nine homozygotic transplants

Silber et al. MHR 2012

Cryopreservation: World-wide experience

* Recent report of three women who have experienced long-term (> 7 years) duration of function of ovarian cortical tissue grafts.

* Birth of eight healthy babies in total following a single graft per patient.

Andersen et al. 2012 RBMonline

Ethical issues

* Ethical considerations for children are different and more challenging from those involving adults
  * who are assumed to be competent
* interventions in children can only be ethical if they can be considered to be therapeutic and in the best interests of the minor

HRT and pubertal induction

- An intriguing question remains: Should ovarian tissue that has been harvested and frozen be reimplanted to provide HRT?
- or even pubertal induction in the young patient with premature ovarian failure?
  - Poirot et al., Lancet 2012
- Ovarian grafts will survive for up to 7 years
  - Andersen et al ., 2012
- several groups have reimplanted ovarian tissue once the initial graft has failed
  - Silber et al., 2008
- Our view is that this precious tissue should only be reimplanted if fertility is requested

Technology or evidence led?

* In the field of fertility preservation there is a dearth of well-designed studies to fully evaluate exciting new techniques

* Unlikely to be feasible or ethical to perform an RCT in a well characterized group of young women to test laparoscopic collection of ovarian cortex versus either dummy laparoscopy or no intervention

* It is highly unlikely that IRBs would pass such a study, or that such a randomized study would be able to recruit sufficient patients

Technology or evidence led?

* When there is uncertainty about a new experimental procedure, it is important for it to be evaluated in IRB-approved clinical trial

* the ASCO guideline recommends that ovarian cryopreservation and transplantation procedures should only be performed in centres with the necessary expertise under IRB-approved protocols that include follow-up for recurrent cancer

Lee et al. JCO 2006, 24(18):2917-31

Edinburgh experience in children (< 18 yrs) 1996-2012

- Ovarian cryopreservation & ovarian function

Cryopreservation of ovarian cortical tissue – Edinburgh criteria

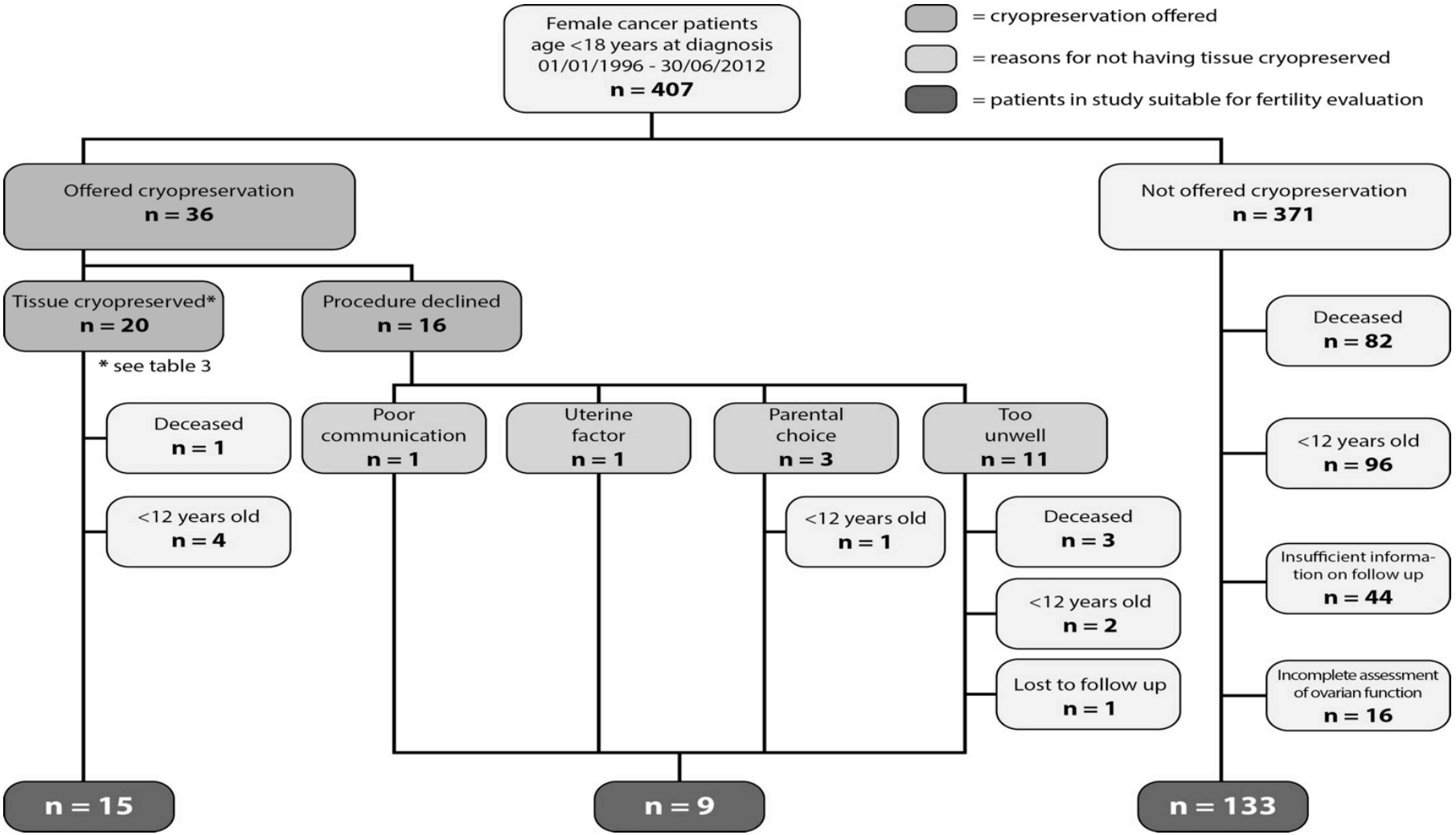
Selection criteria (1995,modified 2000)

* Age < 30 years
* No previous chemotherapy/radiotherapy if age >15 yeqrs
* Mild, non gonadotoxic chemotherapy if < 15 years
* A realistic chance of surviving five years
* A high risk of ovarian failure
* Informed consent (Parent and where possible Patient)
* Negative HIV and Hepatitis serology
* No existing children

Consent

- We emphasize in the information sheet that the procedure is voluntary and experimental, and not part of routine practice
- We obtain informed consent for disposal of ovarian tissue if it is no longer required or the patient dies
- If consent has been obtained, it may be used for ethically approved research studies
- Separately, we ask if an additional small amount can be taken at the time of collection for research studies
- Our practice constitutes research and has been approved by the local institutional review board (IRB)

Results



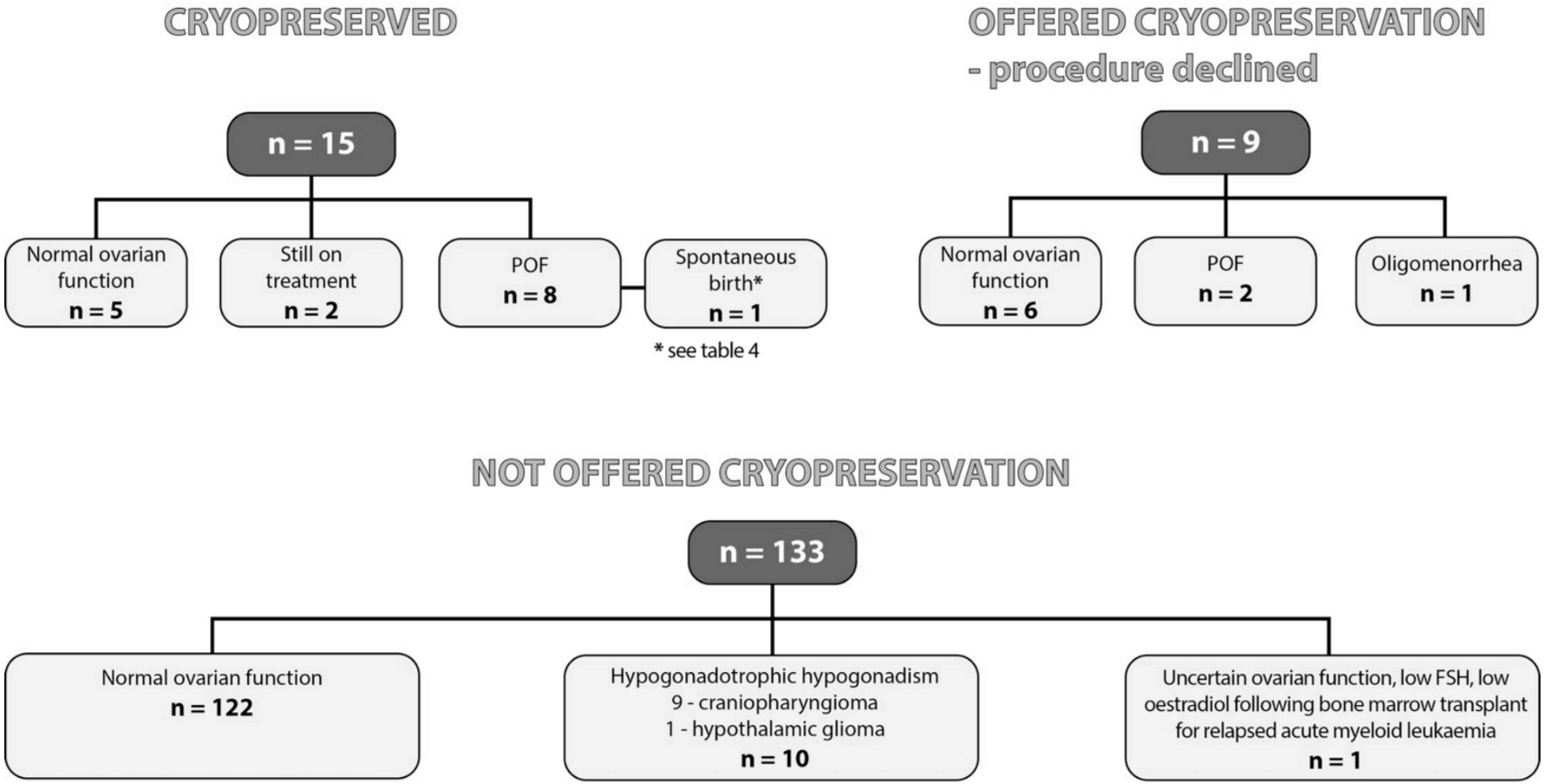
Oncofertility Consortium, Chicago, 2012

Edinburgh Paediatric Experience

Table 3: Patients that had ovarian tissue cryopreserved

Patient No.	Diagnosis	Age at procedure	Method	Complications
1	Hodgkin's lymphoma¤	14.9	Laproscopic Cortical Strip	None
2	Ewing's sarcoma of pubic bone	14.9	Laproscopic Cortical Strip	None
3	Sacral ependymoma	11.3	Laproscopic Cortical Strip	None
4	Hodgkin's lymphoma	13.7	Laproscopic Cortical Strip	None
5	Hodgkin's lymphoma	11.0	Laproscopic Cortical Strip	None
6	Chronic granulocytic leukaemia	9.9	Laproscopic Cortical Strip	None
7	Rhabdomyosarcoma	5.3	Laproscopic Cortical Strip	None
8	Ewing's sarcoma (pelvic)	9.8	Laproscopic Cortical Strip	None
9	Uterine Cervix Rhabdomyosarcoma*	16.5	Laproscopic Cortical Strip	None
10	Hodgkin's lymphoma°	14.1	Laproscopic Cortical Strip	None
11	Abdominal embryonal Rhabdomyosarcoma	7.9	Laproscopic Cortical Strip	None
12	Ewing's sarcoma	12.1	Laproscopic Cortical Strip†	None
13	Hodgkin's lymphoma	12.7	Laproscopic Cortical Strip	None
14	Metastatic Medulloblastoma	8.1	Laproscopic Cortical Strip	None
15	Hodgkin's lymphoma	15.2	Laproscopic Cortical Strip	None
16	Alveolar Rhabdomyosarcoma	10.5	Laproscopic Cortical Strip	None
17	Embryonal Rhabdomyosarcoma	3.0	Oophorectomy	None
18	Ewing's Sarcoma	12.0	Laproscopic Cortical Strip	None
19	Undifferentiated Sarcoma	12.3	Laproscopic Cortical Strip†	None
20	Wilm's Tumour	1.2	Oophorectomy	None

Results


CRYOPRESERVED
n = 15
Normal ovarian function
n = 5
Still on treatment
n = 2
POF
n = 8
Spontaneous birth*
n = 1
* see table 4
OFFERED CRYOPRESERVATION
- procedure declined
n = 9
Normal ovarian function
n = 6
POF
n = 2
Oligomenorrhea
n = 1
NOT OFFERED CRYOPRESERVATION
n = 133
Normal ovarian function
n = 122
Hypogonadotrophic hypogonadism
9 - craniopharyngioma
1 - hypothalamic glioma
n = 10
Uncertain ovarian function, low FSH, low oestradiol following bone marrow transplant for relapsed acute myeloid leukaemia
n = 1

Conclusion

- Ovarian cryopreservation was offered to 9% of our patients, and performed in 5%
- The procedure was safe and without complications
- No patients have asked for re-implantation of their tissue – to date (15.7 [1.3-30.9] yrs)
- All patients who have thus far developed ovarian failure were identified
- The Edinburgh Selection Criteria have proved to be helpful (only one patient not offered cryopreservation who has uncertain ovarian function)

Reimplantation?

* It is important to be aware that reimplantation of ovarian cortical tissue is a separate procedure at a time distant from the treatment of the original cancer
* Consent for harvesting ovarian tissue from children often will have been obtained from their parents
* Informed consent for reimplantation can be obtained from the patients at a much later date when they are competent to assess the complex issues themselves.

Case history:KB

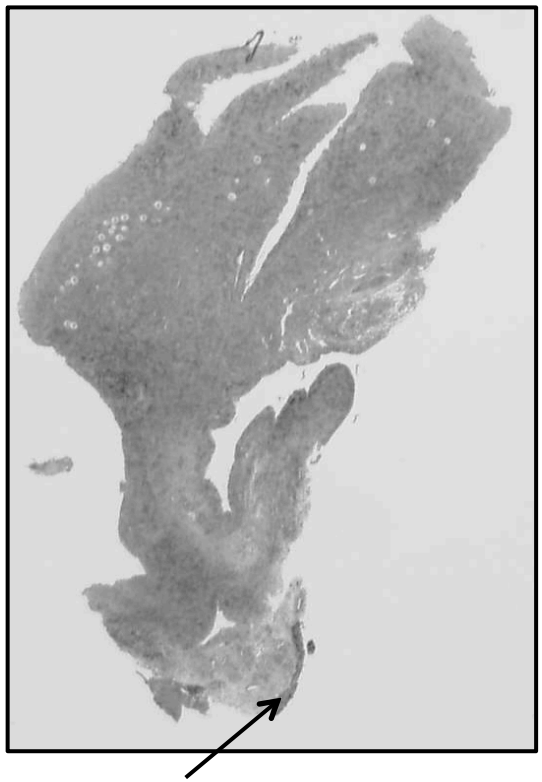
- Ewing's sarcoma L sup pubic ramus, non metastatic 07.96, Age 15
- EICESS,92; 14 courses of ifos based CT and RT (55Gy)
- Lap cortical strips before treatment
- Completed Rx 04.97
- Premature ovarian failure: FSH 23.6, LH 19.5 E2<37 11.97
- Radiation cystitis and vaginitis
- HRT complicated by breakthrough bleeding
- Uterus 4.7x4.5x2.7cm, normal hysteroscopy

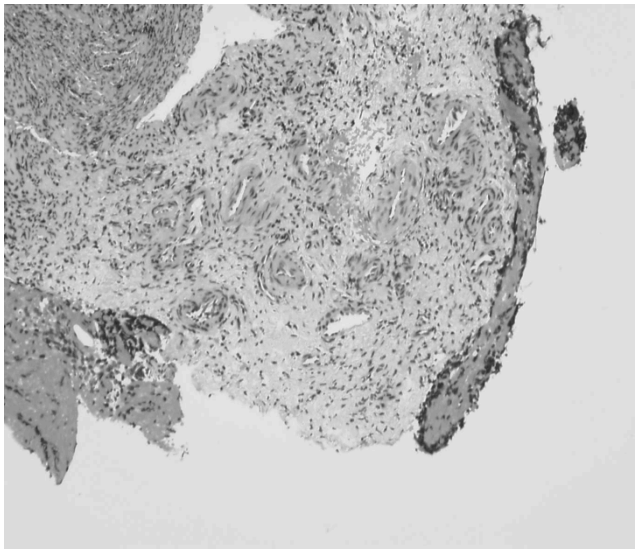
Case history: KB

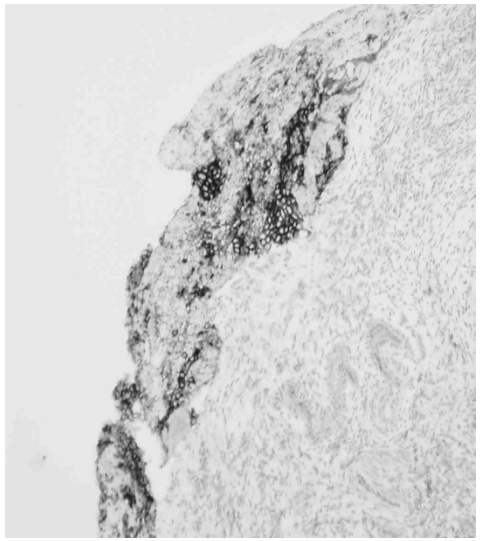
- Became pregnant on HRT!!
- MRI: distortion of pelvic inlet
- Elective section at 37 weeks, uncomplicated
- Male infant 2.94 Kg

Bath et al. Human Reproduction 2004

Ewings sarcoma localised T 7 Vertebrae (Age 12) – Unexpected Contamination of ovarian biopsy







CD99

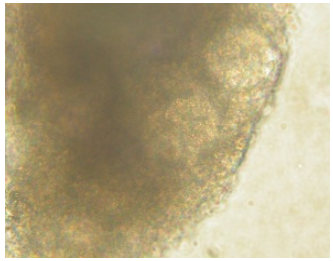
Oncofertility
Consortium, Chicago,
2012

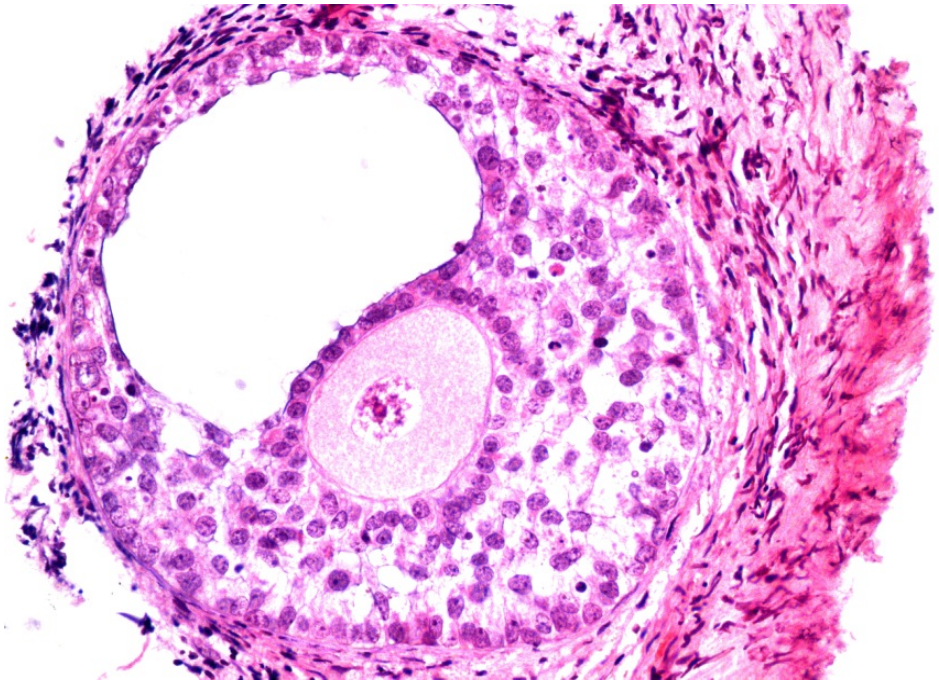
Re-implantation or IVG and maturation?

* Contamination of the cryopreserved tissue with malignant cells, particularly in haematological malignant disease – shown in a rodent lymphoma model – to cause recrudescence of the original disease

* Oocyte maturation in vitro, followed by IVF, would eliminate this risk

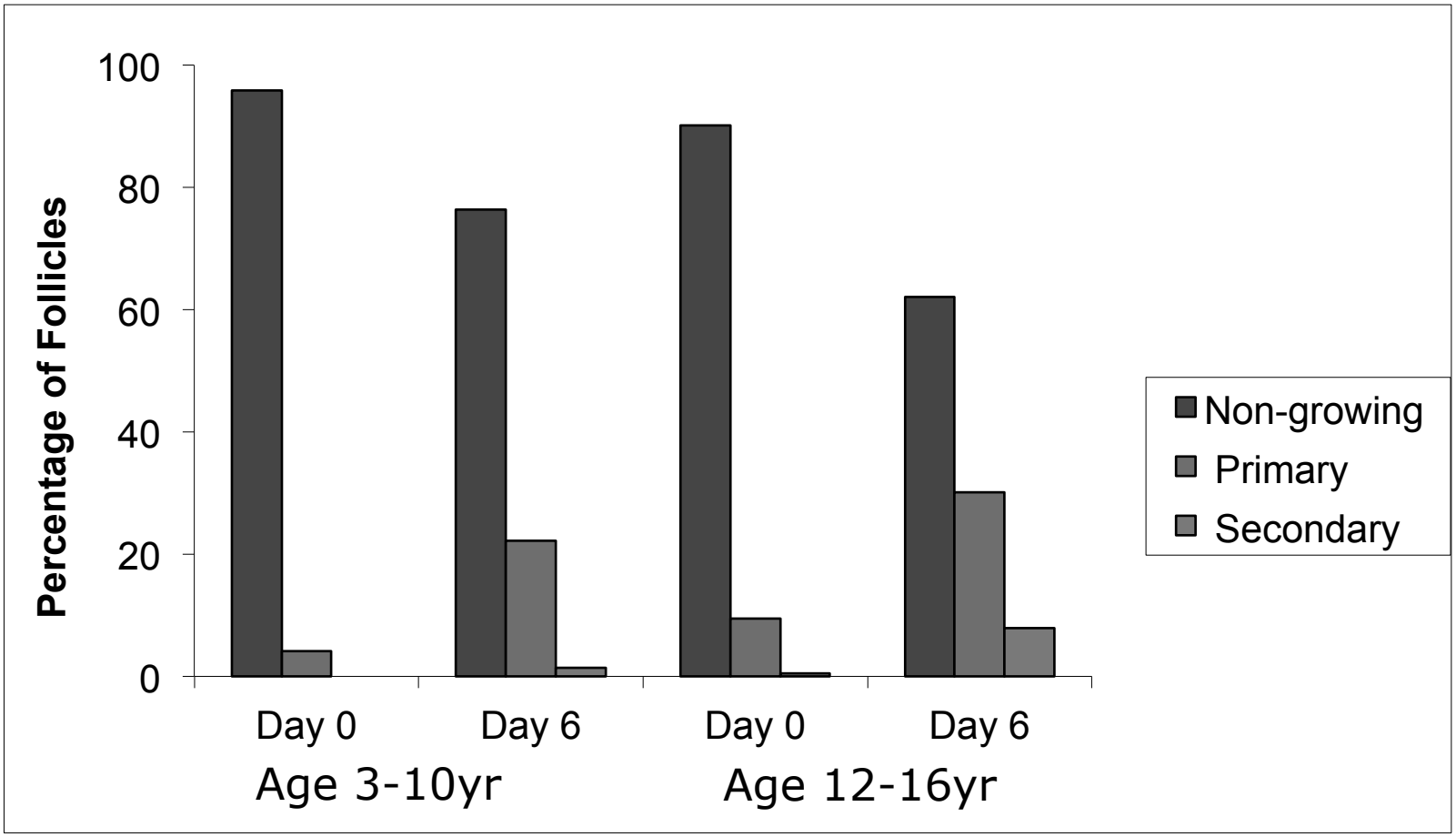
Antral development from *in vitro* grown human primordial follicles within 10 days





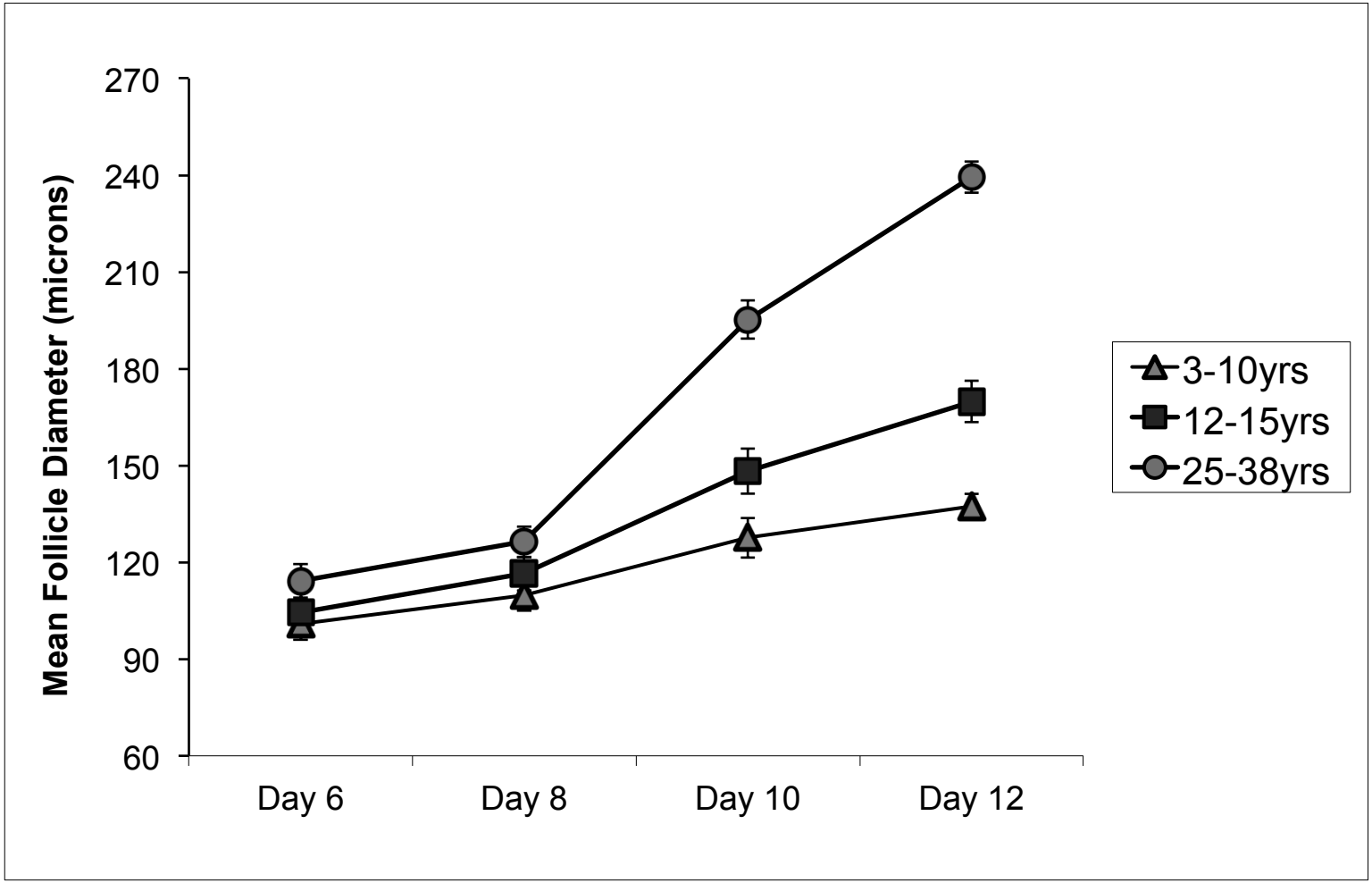
Telfer et al., 2008: A two step serum free culture system supports development of human oocytes from primordial follicles in the presence of activin. **Human Reproduction** 23: 1151-1158

Follicles initiate growth *in vitro* at all ages



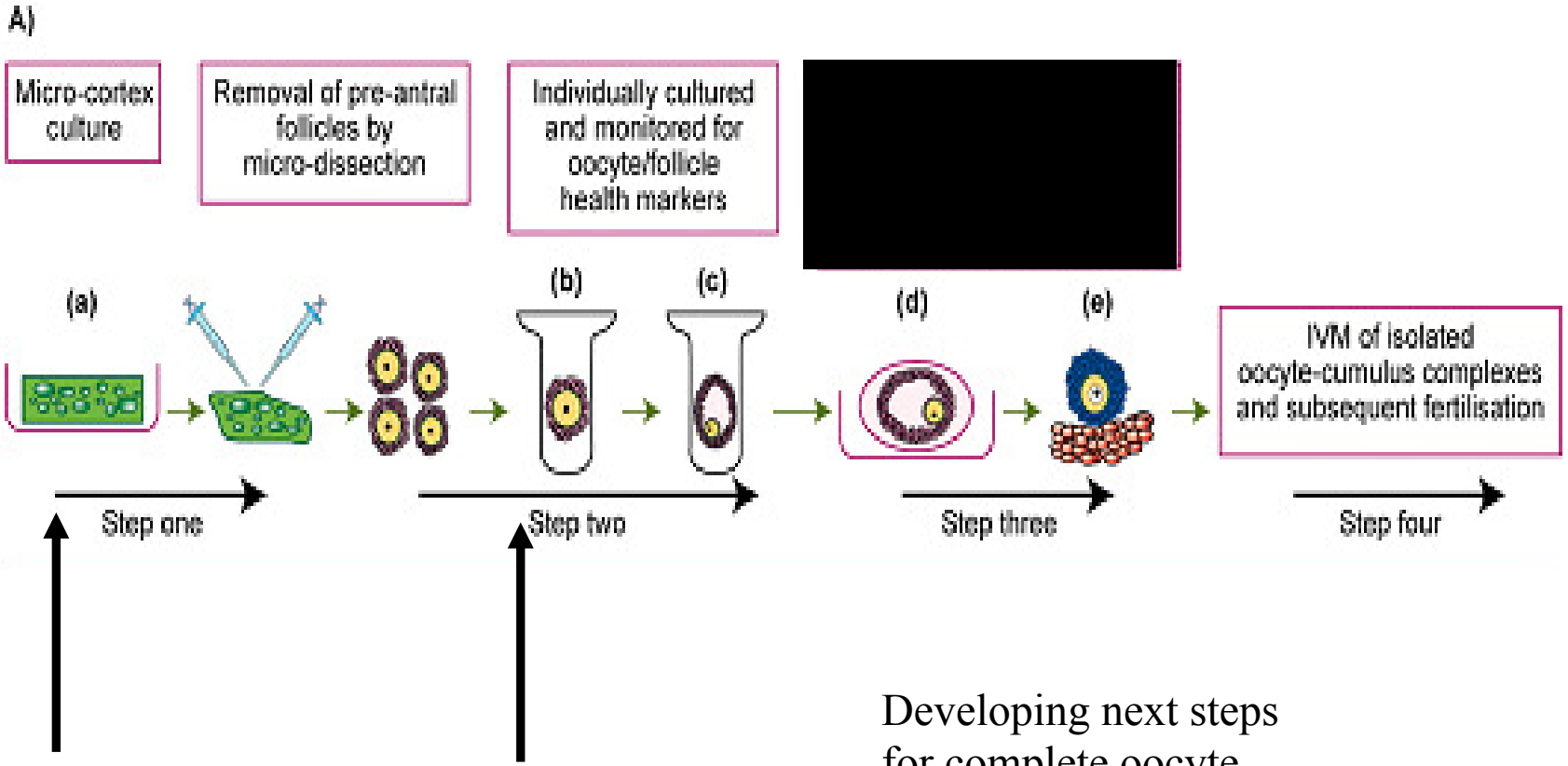
Greater initiation in older girls?

Comparison with adult follicles



Adult follicles (n=44); from Caesarian section
(Antral cavities form at approx 200μm: 30% in adults)

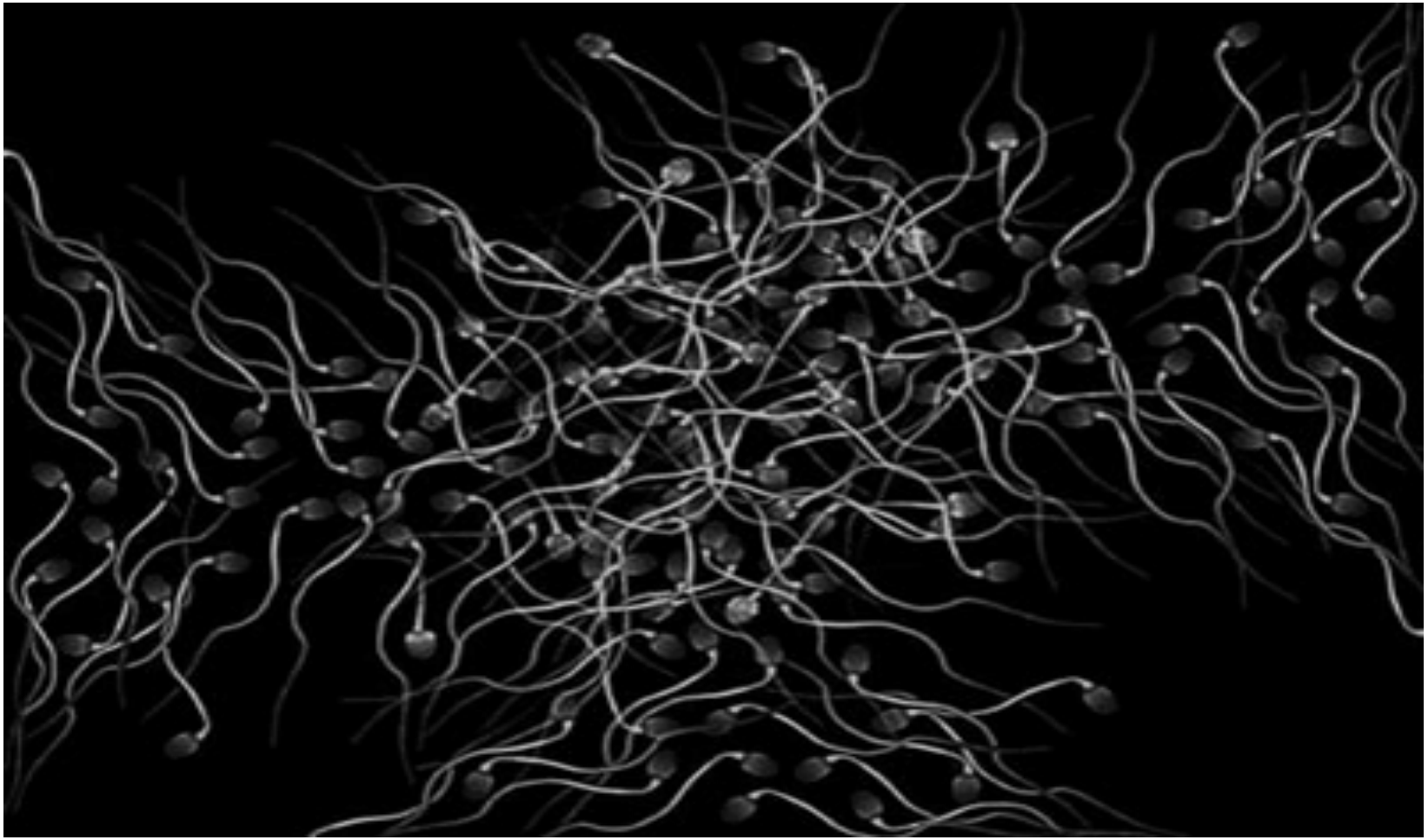
Multi-step Culture system to support human oocyte development



Telfer et al., 2011

Isolated human sperm cells (1500x)

Albert Tousson – Nikon Small world



Oncofertility Consortium, Chicago, 2012

Strategies for fertility preservation in males undergoing treatment for cancer

- Clinical practice
  - Sperm banking
    - Ejaculation
    - Rectal electrostimulation?
    - Testicular/epididymal aspiration

Males: Fertility preservation

- Young men who can produce semen should have the opportunity of sperm banking before treatment begins
- Sperm retrieval should be considered if the chances of infertility are high and the testes are >10mls
  - Storage of gametes is governed by the HFE act 1990
  - Written informed consent from a competent male is required
- There is currently no option to preserve fertility in the prepubertal boy

Pilot interviews with adolescent males (Glaser, Crawshaw et al.)

- 7 young men aged 14 to 17 at diagnosis
  - with cancer
  - offered sperm banking
  - (Aged: 16 to 20 at interview)
- Focus on retrospective perceptions of:
  - communication
  - decision-making
  - management of sperm banking

‘

and down the corridor until I saw a doctor…..It was a bit weird explaining to them what I’d just been through and what do I do with it…and then they said just leave it in the room……I didn’t know if it was safe or not because the doctor… walked off in a different direction…’

‘ ”

were to die”….I had just found out I was diagnosed with it and the question comes up “if you were to die”……’

‘to be honest, it went in one ear and out the other. It was just a load of jargon. I didn’t understand it at all. All I understand was if I didn’t sign this, it, the sperm, wouldn’t be stored’

Oncofertility Consortium, Chicago, 2012

‘I haven’t had a girlfriend since I was diagnosed…I think if I did get a girlfriend, having to tell them that I’m not going to be able to have kids or anything, that’s going to be a bit of a shock to them isn’t it, so… Relationships, yeh, that’s affected me getting in a relationship, getting the confidence….’

Oncofertility Consortium, Chicago, 2012

Summary

Males

* Sperm banking must be considered in all males before treatment that carries a risk of long-term gonadal damage

* There is currently no option to preserve fertility in the pre-pubertal boy (more research required)

Summary

Females

* It remains difficult to predict which patients are at high risk of a premature menopause
* Cryopreservation of ovarian tissue before treatment is the best option for girls and young women
* Orthotopic reimplantation works but so far there have been very few live births.
* Acccelerated IVG of human oocytes is likely to become a realistic possibility.

Challenges

* Provide fertility counseling to all young patients with cancer
* Cryopreserve ovarian tissue from the right patients
* Define the success rate of the procedure
* Develop IVG/M as a safe alternative to reimplantation


joe-ks.com
Seven Dwarfs
of menopause
Itchy
Bitchy
Sweaty
Sleepy
Bloated
Forgetful
All-dried
Up

Acknowledgements

- Tom Kelsey
- Richard Anderson
- Evelyn Telfer
- Marie McLaughlan
- Alice Grove Smith
- Phoebe Wright
- George Galea
- Louise Bath
- Chris Kelnar
- Angela Edgar
- Mark Brougham
- Fraser Munro
- Scott Nelson
- Richard Fleming

Oncofertility Consortium, Chicago, 2012

Thank You