Evidence tables male fertility preservation

Who should be informed about potential infertility risk?

Gupta et al. Testicular biopsy for fertility preservation in prepubertal boys with cancer: Identifying preferences for procedure and reactions to disclosure practices Journal of Urology 2016; 196 (1):219-224

Study design & Main study objective	Participants and relevant characteristics	Relevant results (per outcome)	Additional remarks
<u>1. Study design</u>	<u>1. Type and number of participants</u>	1. Outcome(s) definition	<u>1. Strengths</u>
Cross sectional multi-center	153 parents of pre-pubertal boys with	Outcome 1: Measure of desire for information about TBx	- Use of a novel approach to
study with in-depth	cancer	for FP and Reactions to practices related to disclosure of	assessing the acceptability of an
interviews	77 male survivors of childhood cancer	information	as yet experimental procedure
	30 pediatric oncology health professionals	Outcome 2: Measure of relative willingness of each group	that may meet needs otherwise
2. Main study objective		to accept risk associated with TBx and Predictors of	unmet
To measure and compare	2. Age (at diagnosis) of participants	relative willingness	
parent, male cancer survivor	Parents (age of their child at diagnosis):		- 3 relevant subgroups of
and health professional	≤12 years, median 4 years	2. Results outcome 1	participants with good number in
willingness to accept the risk		Desire for information about testicular biopsy for fertility	each
of TBx in pre-pubertal boys	Survivors:	preservation	
and to identify reactions to	≤12 years, median 5 years		- 3 institutions in Canada included
disclosure practices regarding		90% survivors and 94% parents would have wanted	in the study (multicenter)
biopsy	Health providers:	information about testicular biopsy prior to	
	NA	commencement of therapy regardless of whether or not	2. Limitations
3. Additional study		testicular biopsy was available at treating institution	 Lack of ethnic/cultural diversity
characteristics, if relevant	3. Number of participants per diagnosis		in participant groups (identified
Interviews conducted	 Parents (diagnosis of their child): 	Parents reported the preference of having information	by authors)
between July 2012 and	106(69.3%) leukaemia/lymphoma:	about testicular biopsy regardless the risk of infertility	
September 2013	11(7.2%) sarcoma		- Risk of selection bias: number of
	17(11.1%) brain tumour	3. Results outcome 2	those approached who declined
'Threshold technique' used is	19(12.4%) other	Barrier to testicular tissue cryopreservation	to participate is given, but not
clearly described in			reason for non-participation
appendices and measured	Survivors:	Parents and patients perceived a >30% risk of infertility, a	
willingness to accept the risks	53(69.7%) leukaemia/lymphoma	>25% chance of complications of testicular biopsy, a	- Study undertaken outside of the
associated with TBx with	10(13.2%) sarcoma	>\$500 per year storage cost, and a >14% chance that	'real life' situation in which
	4(5.3%) brain tumour		decisions around fertility

reference to 4 relevant	9(11.8%) other	technology will evolve as barriers for testicular tissue	preservation are made (identified
considerations: • Health providers:		cryopreservation	by authors)
Risk of infertility	NA		
Risk of complications		Health professionals perceived a >29% risk of infertility, a	- Risk of interviewer induced bias
from bx	4. Additional participants characteristics, if	>13.5% chance of complications, a >14% chance that that	
Likelihood of	<u>relevant</u>	technology will evolve, and >\$391 storage cost per year as	- Risk of reporting bias
technology	- Parents:	barriers for testicular tissue cryopreservation	
developing	38(24.8%) Male		
sufficiently to allow		4. If applicable, results per additional outcomes	
successful future use	103(67.3%) White	Predictors:	
of tissue	26(17%) Asian	- Survivors more likely to accept TBx with lower risk of	
Requirement for	5(3.3%) Hispanic	infertility or lower chance of technology evolving as they	
family to cover costs	19(12.4%) Other	aged (p= 0.05)	
of storage of tissue			
until used	1. Survivors:	- Greater household income associated with a lower	
	Boys received at least 2 months of cancer	minimum infertility risk (p= 0.05), and higher yearly costs	
In-depth interviews were also	therapy and either still receiving therapy,	(p= 0.04)	
conducted with a subset of	or post-therapy		
each participant group to		- No demographic variables were associated with TBx	
explore information	62(80.5%) White	desirability scores for HP	
disclosure practices.	5(6.5%) Asian		
	2(2.6%) Hispanic	Choose TBx vs. no biopsy overall:	
Threshold technique followed	8(10.4%) Other	110(72%) parents	
by indepth guided interview		52(67%) survivors	
of subgroup	1. Healthcare providers:	22(73%) HP	
	15(50%) Female		
	29(96.7%) White		
	25(83%) physician		
	2(6.7%) NP		
	2(6.7%) RM		
	1(3.3%) SW		

TBx: testicular biopsy; NA: not applicable; NP: nurse practitioners; SW: social worker; HP: health provider

Who should be informed about potential infertility risk?

Gupta et al. Assessing information and service needs of young adults with cancer at a single institution: the importance of information on cancer diagnosis, fertility preservation, diet and exercise. Supportive Care in Cancer 2013; 21:2477-2484

Study design & Main study objective	Participants and relevant characteristics	Relevant results (per outcome)	Additional remarks
<u>1. Study design</u>	1. Type and number of participants	1. Outcome(s) definition	<u>1. Strengths</u>
Single center cross-	243 cancer patients receiving	Outcome 1: Importance of information on fertility effects	- The questionnaire used was an
sectional (survey) study	treatment, or within 5 years of	from treatment and fertility preservation	existing published item, adapted to
	completion of treatment	Outcome 2: Importance of information on treatments for	reflect the study site, and piloted
2. Main study objective		infertility and other options for having children	
To identify the	61.3% male		- Participants were representative
information and service	40.1% currently receiving cancer	2. Results outcome 1 and outcome 2	of a wide range of diagnoses
needs of young adults	treatment	Desire for information in fertility preservation discussion	relevant in this age group, and of
with cancer to inform a			both on and off treatment groups
program development	2. Age (at diagnosis) of participants	Survey question: Information about effects of cancer	
	NR	treatment on your ability to have children in the future and	2. Limitations
3. Additional study	Age at study: median 28 years (17-	how to preserve your fertility before starting treatment	- Single center study (results have
characteristics, if	35 years)		low external validity)
<u>relevant</u>		Median 10 (range 1-10); mean (SD) 8.77 (2.23)	
Survey conducted	3. Number of participants per	males: mean 8.45 (2.34)	- Risk of selection bias: no
November 2010	diagnosis	females: mean 9.28 (1.94)	information on how the 243
	23(9.5%) brain tumour		patients were selected
Adapted existing survey	19(7.8%) breast, ovarian, cervical	Survey question: Information on treatment for infertility and	
to use Likert Scale of	cancers	other options for having children (i.e. artificial insemination,	- Convenience sample (survey
importance (1-10)	46(18.9%) leukaemia	in vitro fertilization, surrogacy, adoption etc)	administered to those attending
	69(28.4%) lymphoma		ambulatory care centre of
Study participants were	21(8.6%) sarcoma	Median: 9 (range 1-10); mean (SD) 7.81 (2.85)	Canadian adult tertiary cancer
asked how important it	40(16.5%) testicular cancers	males: mean (SD) 7.50 (2.90)	centre)
was to them	25(10.5%) colon, other cancers	females: mean (SD) 8.30 (2.72)	
to get information on a			 No report of ethnicity of
certain resource as part	4. Additional participants	3. If applicable, results per additional outcomes	participants
of a program	characteristics, if relevant	- Females rated information on FP methods (p=0.004) and	
	162 (66.7%) Single/never married	risk of infertility (p=0.033) as more important than did males	 Actual questions of survey not
	68 (28%) Married/common-law		included in report

for young adult cancer survivors, or have it included in the	49 (20.3%) Have existing children	- Presence of existing children did not significantly impact on importance of information regarding risk to fertility from cancer treatment (n=0.65)	 Risk of reporting bias (as use of survey)
program			
Fertility items were 2 out of 18 questions		 Those who had completed active therapy showed a trend towards rating receiving information about fertility as more important than those on active treatment p=0.052 	
Item responses averaged for entire sample			

NR: not reported; SD: standard deviation; FP: fertility preservation; SD: standard deviation

Who should be informed about potential infertility risk?

Wyns et al. Fertility preservation in the male pediatric population: factors influencing the decision of parents and children. Hum Reprod 2015;30:2022-30.

Study design & Main study	Participants and relevant	Relevant results	Additional remarks
objective	characteristics	(per outcome)	
1. Study design Cross sectional study Single-center survey from Belgium	<u>1. Type and Number of</u> <u>Participants</u> Prepubertal boys and adolescents aged 0-18 years diagnosed with cancer between May	 <u>1. Outcome definitions</u> Factors influencing the FP decision Feelings of patients and their parents, with a view to better fulfilling their expectations 2. Results 	1. Strengths A large study. Closed-ended questionaire followed by response options to minimize random errors in the data collection process and allow
2.Main study objective To critically analyse the multidisciplinary collaborative care pathway (MCCP) in the pediatric population,	2005 and May 2013. Eligible patients: 348 of which 120 returned questionnaire; only 78 questionnaires included responses to Part 2.	Response by patients, parents or both: - Parents considered their child (91.4% of adolescents and 26.2% of children aged <12 years, but >7yr) capable of understanding and participating in the decisional process - Reasons for not understanding and participating in the decision process vere immaturity of the child (5.7%), poor general health (2.9%)	quantitative interpretation. <u>2. Limitations</u> Recall bias due to the to the time interval between the actual FP procedure and the survey. Single-center survey, thus does not allow generalizability of results to other places.

focusing on factors	Parents gave their	 No discrepancy between patient and parent 	There is no availability of
influencing the	answers for 22 patients	decisions was noted, indicating that decisions were	preexisting validated
decision, and to	under 12 years of age	essentially made jointly	questionnaires or gold standard for
elucidate and	and 3 patients aged 12–	 Information was provided mainly by 64/78 	this type of study.
characterize the	18yrs	(82%) oncologist, 7/78 (8.9%) GP, 5/78 (6.4%)	
feelings of patients		specialist and 2/78 (2.5%) by nurses; Although	3. Risk of bias
and their parents,	2. Age at diagnosis	nurse support was limited in this study, it appeared	1. Selection bias: high risk
with a view to	Mean ± SD:	to be relevant for 16.6% of adolescents	Reason: 120/348 (34.5%) eligible
better fulfilling	6.05 ± 3.74 years (range		patients returned their
their expectations.	0.1–143 months) for	Emotional state of parents during discussion of FP (barrier)	questionnaires (44 patients died,
	boys aged <12 yr	- 52% of adolescents and 23.5% of children felt anxious	14 lost to fup, 8 declined to
3. Study years		at the time of discussion	participate, some did not return
May 2005 to May	14.41 ± 1.5 years (range	- Reasons were concern about future fertility, rather than	their questionnaire.)
2013	144–212 months) for	the method of FP, 46% of boys aged 12–18 years considered	
	boys aged 12–18 yr	the FP method challenging because of poor	2. Attrition bias: High risk
4. Follow-up		general health, lack of experience with masturbation and	Reason: A total of 78/120 (65%)
Mean ±SD 3.4 ± 2.3	3. Number of responded	its taboo or embarrassing nature	gave information on FP issues and
years (ie) Time	participants per	- 76% of children and 48% of adolescents considered their	have responded to questions on
from diagnosis to	<u>diagnosis</u>	health to be more important than the ability to have a family	communication, emotional
the time of the	Acute lymphoblastic	- Family support was considered important for 75% of	state and perceptions during
survey was	Leukemia	adolescents and 58% of children, and medical support was	discussion of FP, reasons for refusal
	33(27.7%)	considered important for 50% of adolescents and 42% of	etc.
	Acute myeloid leukemia	children; Nursing support was relevant for 16.6% of	
	2(1.68%)	adolescents.	3. Detection bias: Unclear
	Non-Hodgkin's		Reason: Unclear if outcome
	lymphoma 13 (10.9%%)	Understanding information: (facilitator)	assessors were blinded.
	Hodgkin's lymphoma	- Majority of boys aged >12 years reported information to	
	6(5.0%)	be clear (72%), complete (80%) and understandable (90.9%)	4. Confounding: High risk
	Medulloblastoma 3	 Only 33.3% of boys aged <12 years were able to 	Reason: did only bivalent analysis.
	(2.5%)	comprehend the information, the youngest being 11 years	Thus did not adjust for
	Nephroblastoma 4 (3.3%)	old (although, respectively, 71.4 and 57.9% of subjects found	confounders.
	Neuroblastoma 9 (7.6%)	it to be complete and clear)	
	Osteosarcoma 9 (7.6%)		
	Retinoblastoma 7 (5.9%)	Satisfaction with information:	
	Ewing's sarcoma 6 (5.0%)	 19% was not satisfied with the fertility 	
	Rhabdomyosarcoma 6	preservation information content (completeness)	
	(5.0%)		

Hepatoblastoma 4(3	.4%) Acceptance and refusal rate: (barrier)	
Brain tumor 7 (5.9%)) - One-third of the patients lack information	
Astrocytosis 2(1.68%	b) about FP options when seen by the oncologist	
Ependymoma 1(0.89	 FP acceptance rates were 74% for boys aged 	
Benign pathologies	7 <12 and 78.6% for boys 12-18 years	
(5.9%)	- 6/78 (7.7%) adolescents and 13/78 (16.7%)	
	children under the age of 12 years refused to	
4. Additional patient	undergo FP procedures	
characteristics, if rel	evant - Reasons for refusal were the urgency of	
42 patients (35%) die	d not cancer treatment, diminished general health, the FP	
receive information	on procedure not being a priority or the experimental	
FP issues	status of FP before puberty	
	 Wishing to avoid an additional procedure 	
	was not an issue for FP acceptance	
	- Satisfaction about completeness of	
	information provided to patients and parents	
	positively impact decision to preserve fertility	
	(p=0.04)	
	 Hope for future parenthood positively 	
	impact decision to preserve fertility (p<0.01)	
	- Timing of FP information, healthcare	
	provider who proved the FP information and anxiety	
	were not significantly associated with decision to	
	preserve fertility	

Who should be informed about potential infertility risk?

Quinn et al. Fertility Preservation and Adolescent/Young Adult Cancer Patients: Physician Communication Challenges. J Adolesc Health 2009;44(4):394-400

Study design & Main study objective	Participants and relevant characteristics	Relevant results (per outcome)	Additional remarks
<u>1. Study design</u>	<u>1. Type and number</u>	<u>1. Outcome(s) definition</u>	<u>1. Strengths</u>
with qualitative	24 Dediatric	Outcome 1: Reditical e system barriers	discussing ED in padiatric appalagy
somistructured in depth	24 Feulatine	information	implying that now mathods of
intonvious	in 15 clinics in Florida	Outcome 2: Awareness of ED resources	communication between all parties
Interviews		Outcome 4: Patient characteristics that may impact EP	must be examined and utilized
2 Main study objective	(03)	discussions	
To examine barriers	Response rate: 41%	Outcome 5: Issues unique to adolescent patients	2. Limitations
experienced by	participated (59		 Results cannot be generalized to
physicians in discussing	asked to participate)	2. Results outcome 1:	other pediatric hematology/oncology
cancer-related fertility		- Perceptions that the financial costs of FP were too high	physicians or other populations
issues with patients aged	2. Age (at diagnosis)	for most families (FP not covered by insurance)	
12-18yrears	of participants	- Combination of lack of resources and lack of training or	 Authors state that interview may
	NA	guidelines for having discussions	have limited the amount of in-depth
3. Additional study			discussion on any one topic
characteristics, if	<u>3. Number of</u>	3. Results outcome 2:	
<u>relevant</u>	participants per	- About half of physicians said the cancer diagnosis is such	- Risk of selection bias: responders
- Study used a subset of	<u>diagnosis</u>	a shock that an issue like fertility is often put on the "back	more interested in the topic and
data from a larger study	NA	burner"	more likely to engage in discussions
examining knowledge,		- Other half thought that parents and teens do want this	about and/or encourage FP might
attitudes, and behaviors	4. Additional	information but are either to embarrassed to discuss it or	have been participants
of pediatric oncologists	participants	have no background on the topic and do not know how to	
	characteristics, if	begin a discussion	- Risk of interviewer induced bias
- All interviews were	relevant		
tape recorded and	NA	4. Kesuits outcome 3:	
transcribed. The		- One third of physicians were aware of sperm banking	
transcripts were read		Tacinues	

through and the content	- Remainder said their facility had no FP resources or they	
analyzed	were unaware of resources for females (except	
through intuitive	oophoroypexy)	
analysis. Key	- Physicians typically had low levels of knowledge about	
themes were identified	resources to refer patients to for FP procedures or	
	consultations	
- Author used theoretical	- Few pediatric oncologsist reported that the nationally	
saturation, in which each	distributed educational brochure they used was not	
new participant	always relevant to the local level and needed	
we recruited refined	improvement	
new theoretical		
constructs. Midway	5. Results outcome 4:	
in the data analysis we	- Most were comfortable in a general sense	
ascertained no new	- However, many experienced barriers related to patient	
information	specific diagnosis or socioeconomic situation (ranged	
was emerging; thus, we	from perceived cultural or religious differences to	
perceived we had	knowing a family could not afford FP)	
reached theoretical		
saturation and made no	6. Results outcome 5:	
further attempts to	- All found that it is an important issue to address for	
recruit additional	teens who have reached puberty	
physicians	 Most agreed that these conversations were awkward 	
	because resources were usually limited and there was a	
	fine line between establishing a sense of trust with the	
	patient, while not excluding parents	
	 Conversations about fertility were related to issues of 	
	sexuality, and this was a source of embarrassment for	
	both the patient and parents	
	4. If applicable, results per additional outcomes	

NA: not applicable; FP: fertility preservation

Who should be counselled about fertility preservation?

van Beek et al. Inhibin B is superior to FSH as a serum marker for spermatogenesis in men treated for Hodgkin lymphoma with chemotherapy during childhood. Human Reprod 2007;22:3215-3222

Study design				
Treatment era	Participants	Treatment	Main outcomes	Additional remarks
Years of follow-up				
<u>1. Study design</u>	<u>1. Type and number of</u>	1. Chemotherapy	1. Outcome definitions	<u>1. Remarks</u>
Multi-centre	participants	- ABVD or EBVD: 56 (100%);	- Oligozoospermia: sperm cell density <20 x	3 men reported 5 spontaneously
cohort study	56 male survivors of	Adriamycin 25mg/m ² or	10°/mL	conceived pregnancies. 2 men
	childhood Hodgkin	epirubicin 30 mg/m ² ,	- Severe oligozoospermia: sperm cell	treated without MOPP each
2. Treatment era	lymphoma	bleomycin 10 mg/m²,	density <5 x 10 ⁶ /mL	fathered 1 child. 1 man treated
1974-1998		vinblastine 6 mg/m ² ,	 Decreased inhibin B: <150 ng/L 	with 6 MOPP cycles fathered 1
	2. Diagnoses	dacarbazine 250 mg/m ² (days	- Increased FSH: >7.0 U/L	child and reported 2 spontaneous
<u>3. Follow-up</u>	56 (100%) Hodgkin	1 and 8)		abortions.
Median 15.5 (5.6-	lymphoma	- MOPP: 40 (71.4%) of whom	2. Results	
30.2) years after		14 (25.0%) 3-4 cycles, 26	Sperm concentration (n=21 assessed):	2. Risk of bias
treatment	3. Age at diagnosis	(46.4%) ≥6 cycles;	- MOPP ⁻ : 49.1 (28-63) x 10 ⁶ /mL (n=4)	A. Selection bias
	Median 11.4 (3.7-15.9)	Mechlorethamine	- MOPP ⁺ : 1.1 (0-72) x 10 ⁶ /mL (n=17)	High risk
	years	6mg/m ² (days 1 and 8),	p<0.05	Reason: 56/100 (56%) eligible
		vincristine 2 mg/m ² (days 1	- MOPP ⁻ : 0/4 (0.0%) azoospermia or	patients included in this study,
	4. Age at follow-up	and 8), prednisone 40	oligozoospermia (all 4 normospermia)	however, there were no
	Median 27.0 (17.7-42.6)	mg/m²/day (days 1 – 14),	- MOPP ⁺ : 9/17 (52.9%) azoospermia; 1/17	differences in age, disease
	years	procarbazine 100 mg/m ² /day	(5.9%) oligozoospermia; 3/17; (17.6%)	characteristics and treatment
		(days 1 – 14)	severe oligozoospermia	between the included 56 male
	5. Controls			survivors and the 44 not
	No controls	2. Radiotherapy	Inhibin B (n=38 assessed):	included.
		- Involved field irradiation: 7	- MOPP ⁻ : 144.0 (93.0-274.0) ng/L (n=12)	
		(12.5%)	- MOPP ⁺ : 16.5 (0.0-173.0) ng/L (n=26)	B. Attrition bias
		- Abdominal irradiation: 0	p<0.01	High risk
		(0.0%)		Reason: 21/56 (37.5%) of patients
		- Pelvic irradiation: 0 (0.0%)	FSH (n=56 assessed):	had sperm concentrations
			- MOPP ⁻ : 3.0 (1.7-6.0) U/L (n=16)	assessed.
			- MOPP ⁺ : 16.8 (1.3-51.0) U/L (n=40)	
			p<0.01	C. Detection bias
				Unclear

Risk factors for decreased sperm	Reason: unclear if the outcome
concentration in multivariable analysis:	assessors were blinded for
- Number of MOPP cycles: beta -6.25	important determinants related
(p<0.05)	to the outcome.
- Age at diagnosis: beta -6.18 (p<0.05)	
- Number of EBVD/ABVD cycles,	D. Confounding
radiotherapy (mantle or mediastinal),	Low risk
puberty at diagnosis, presence of B-	Reason: Multiple linear
symptoms and follow-up duration	regression analyses were
(p>0.05) (no effect measures reported)	performed, including number of
	MOPP cycles, age at diagnosis,
Risk factors for decreased inhibin B in	number of EBVD/ABVD cycles,
multivariable analysis:	radiotherapy (mantle or
- Number of MOPP cycles: beta -21.59	mediastinal), puberty at
(p<0.05)	diagnosis, presence of B-
- Other factors not significant	symptoms and follow-up
	duration.
Risk factors for increased FSH in	
multivariable analysis:	
- Age at diagnosis; beta 1.4 (n<0.05)	
- Number of MOPP cycles: beta 2.57	
(n<0.01)	
- Other variables not significant	

Abbreviations: FSH, follicle-stimulating hormone.

Who should be counselled about fertility preservation?							
Brignardello et al. Gonadal status in long-term male survivors of childhood cancer. J Cancer Res Oncol 2016;142:1127-32.							
Study design Treatment era Years of follow-up	itudy design Treatment era Participants Treatment Treatment Main outcomes Additional remarks Treatment era follow-up						
<u>1. Study design</u> Betrospective	<u>1. Type and number of</u>	<u>1. Chemotherapy</u> - Apy: 187 (94.0%)	<u>1. Outcome definitions</u> - Hypogonadism: testosterone <3.0	<u>1. Strengths</u> Cohort size long follow-up duration			
cohort study	199 male CCS aged <18 yr at cancer diagnosis	- Alkylating agents: 147 (73.9%)	ng/dl; further subclassified as primary or central, depending on	2. Limitations			

2. Treatment era	with ≥1 clinic visit after	- Alkylating agents and	gonadotropin levels (no further	- It is not stated how many participants
1985-2007	age 18 yr	platinum agents: 23 (11.6%)	information reported)	were treated with both radiotherapy
		2. Radiotherapy	- Spermatogenesis damage: FSH >10.0	and chemotherapy. The result that
3. Follow-up	2. Diagnoses	- Any: 125 (62.8%)	UI/I and inhibin B <100.0 pg/ml	"the risk of gonadal dysfunction was
Median 14.01 (IQR	Hematological	- TBI: 33 (16.6%)		higher in patients treated with
10.08-17.76) yr	malignancies n=145	- Cranial: 38 (19.1%)	<u>2. Results</u>	radiotherapy" may be biased.
	(ALL n=72, HL n=40,	<u>3. Surgery</u>	Abnormal gonadal function:	- Definitions of primary or central
	NHL n=21, AML n=12)	Number patients treated with	- Normal gonadal function: 102/194	hypogonadism are not given.
	brain tumours n=28,	surgery not given, but in	(52.6%)	 Testosterone assay not described,
	sarcomas n=15, other	discussion mentioned that	 Spermatogenesis damage: 68/199 	time of assessment not given
	n=11	some were treated with	(34.2%) and confirmed in 41 patients	(Testosterone fluctuates during the
		surgical excision	in whom semen analysis was	day and may give false negative
	3. Age at diagnosis		performed	results when assessed in afternoon
	0-4 yr: n=45; 5-10 yr:	4. Other treatments	- Primary hypogonadism: 16/199 (8.0%)	and not twice).
	n=57; 10-18 yr: n=97	HSCT: 48 (24.1%)	 Secondary hypogonadism: 13/199 	 Primary hypogonadism (n=16) will
			(6.5%)	always result in impaired
	4. Age at follow-up			spermatogenesis (n=68). When
	>18 yr		- 33/33 (100%) treated with TBI had	considering spermatogenesis damage
			abnormal gonadal function:	the results are displayed incorrectly.
	<u>5. Controls</u>		spermatogenesis damage n=17	 Reference values for normal semen
	No controls		primary hypogonadism n=13	analysis not given.
			central hypogonadism n=3	 Assumption bias: men with FSH >10
			 46/48 (95.8%) treated with HSCT had 	U/I and inh B <100 ng/I levels may
			abnormal gonadal function	have compensated spermatogenesis
			Risk factors for spermatogenesis	and may father children without
			damage and primary hypogonadism in	assisted reproduction. Even so, the
			multivariable logistic regression analysis:	prognostic value for sperm
			 Alkylating + platinum agents vs. 	concentration/progressive motility
			alkylating agents only: OR 9.22 (95% Cl	and morphology for fecundity is poor.
			2.17-39.23)	Result without semen analysis
			 Other chemotherapy or none vs. 	parameters should be interpreted
			alkylating agents only: OR 0.19 (95% Cl	with caution.
			0.05-0.76)	- Cohort unclear: total cohort 199, in
			- Any radiation vs. none: OR 8.72 (95%	text endocrine levels available at last
			CI 3.94-19.30)	visit n=194, Table 2 cohort n=186;
				gonadal dysfunction n=84 (elsewhere
				described as n=92): unclear.

	- Period of first cancer diagnosis 1990-	3. Risk of bias
	1999 vs. 1985-1989: OR 1.48 (95% Cl	A. Selection bias
	0.43-5.10)	Unclear
	- Period of first cancer diagnosis 2000-	Reason: "all patients referred to the
	2007 vs. 1985-1989: OR 1.24 (95% Cl	transition unit for CSS in Turin, Italy" the
	0.32-4.87)	protocol for referral is not described: it
	- Age at cancer diagnosis 5-9 vs. 0-4 yr:	is unclear if pre-selection for referral
	OR 1.08 (95% CI 0.40-2.93)	was made.
	 Age at cancer diagnosis ≥10 vs. 0-4 yr: 	
	OR 0.64 (95% CI 0.25-1.68)	B. Attrition bias
	- Brain tumours vs. hematological	Low risk
	malignancies: OR 0.98 (95% CI 0.36-	Reason: of the referred eligible cohort
	2.63)	only 11 males were lost to follow up and
	- Sarcomas vs. hematological	in at least 194 patients reproductive
	malignancies: OR 3.69 (95% CI 1.11-	hormone levels were available (97.5%).
	12.22)	
	- Other tumours vs. hematological	C. Detection bias
	malignancies: OR 1.13 (95% CI 0.33-	Unclear
	3.89)	Reason: unclear if the outcome
		assessors were blinded for important
		determinants related to the outcome.
		D. Confounding
		Low risk
		Reason: analyses were adjusted for age
		at cancer diagnosis, period of cancer
		diagnosis and treatment.

Abbreviations: ALL, acute lymphoblastic leukaemia; AML, acute myeloid leukaemia; CCS, childhood cancer survivors; IQR, inter quartile range; HL, Hodgkin lymphoma; HSCT, hematopoietic stem cell transplantation; NHL, non-Hodgkin lymphoma; TBI, total body irradiation.

Who should be counselled about fertility preservation?

Chemaitilly et al. Leydig Cell Function in Male Survivors of Childhood Cancer: A Report From the St Jude Lifetime Cohort Study. J Clin Oncol 2019;37:3018-3031.

Study design Treatment era Years of follow-up	Participants	Treatment	Main outcomes	Additional remarks
1. Study design	1. Type and number of	1. Chemotherapy	1. Outcome definitions	1. Remarks
Single-center	participants	- Alkylating agents: 898 (59.2%)	- Levdig cell failure: morning serum levels	Participants with hormone levels
cohort study	1.516 adult male CCS	- CED >0-<4.000 mg/m ² : 133	of total testosterone < 250 ng/dL (or	indicative of Levdig cell failure
	aged ≥18 years who	(8.8%)	8.67 nmol/L) and LH > 9.85 IU/L	before replacement and those
2. Treatment era	survived ≥5 years since	- CED ≥4,000-<8,000 mg/m ² : 269	- Leydig cell dysfunction: morning serum	without pretreatment laboratory
1970-2002	diagnosis	(17.7%)	levels of total testosterone ≥ 250 ng/dL	data but whose medical records
		- CED ≥8,000-<12,000 mg/m ² : 245	and LH > 9.85 IU/L	specifically documented Leydig
3. Follow-up	2. Diagnoses	(16.2%)		cell failure as the reason for
Median 22.0	Leukaemia 520 (34.3%),	- CED ≥12,000 mg/m ² : 251 (16.6%)	2. Results	treatment were considered to
(range 7.5-49.8)	lymphoma 337 (22.2%),		Leydig cell function at most recent SJLIFE	have Leydig cell failure.
years since	bone and soft tissue	2. Radiotherapy	visit vs. controls:	
diagnosis	sarcomas 223 (14.7%),	- Testicular radiotherapy: 123	- Point prevalence Leydig cell failure: 104	These found associations of
	Wilms tumour 85	(8.1%)	(6.9%; 95% CI 5.6%-8.2%) vs. 8 (4.8%;	identified risk factors and Leydig
	(5.6%), CNS tumour 148	- 0-11.9 Gy: 65 (4.3%)	95% Cl 1.5%-8.0%); p=0.30	cell failure remained significant
	(9.8%), neuroblastoma	- 12-19.9 Gy: 39 (2.6%)	 Point prevalence Leydig cell 	after the exclusion of nine
	68 (4.5%),	- ≥20 Gy: 19 (1.3%)	dysfunction: 223 (14.7%; 95% Cl 13.0%-	survivors who were receiving
	retinoblastoma 45		16.5%) vs. 4 (2.4%; 95%Cl 0.1%-4.7%);	treatment for Leydig cell failure
	(3.0%), carcinomas 23	<u>3. Surgery</u>	p>0.001	but lacked laboratory data
	(1.5%), germ cell	Unilateral orchiectomy: 35 (2.3%);		supporting the diagnosis.
	tumour 20 (1.3%),	Survivors with a bilateral	Risk factors for Leydig cell failure in	
	other 47 (3.1%)	orchiectomy were excluded from	multivariable logistic regression analysis:	2. Risk of bias
		the study	- Testicular radiation dose >0-11.9 Gy vs.	A. Selection bias
	3. Age at diagnosis		none: OR 3.1; 95% Cl 1.4-7.2 (p=0.007)	High risk
	Range 0-≥15 years		- Testicular radiation dose 12-19.9 Gy vs.	Reason: Out of 2,880 potentially
			none: OR 97.3; 95% Cl 29.2-323.6	eligible male survivors 1,701
	4. Age at follow-up		(p<0.001)	(59.1%) agreed to participate in
	Mean 33.8 ± 9.2 years;		- Testicular radiation dose ≥20 Gy vs.	the study.
	Median 30.8 (range 18		none: OR 220.0; 95% Cl 26.0-1,858.8	
	1-63.8) years		(p<0.001)	B. Attrition bias
				Low risk

5. Controls	- CED >0->4,000 mg/m ² vs. none: OR 0.5;	Reason: All patients had an
168 age- and sex-	95% CI 0.2-1.7 (p=0.28)	outcome assessment.
matched community	- CED 4,000-<8,000 mg/m ² vs. none: OR	
controls recruited	3.4; 95% Cl 1.7-6.8 (p<0.001)	C. Detection bias
among friends and	- CED 8,000-<12,000 mg/m ² vs. none: OR	Unclear
non-first degree	2.9; 95% Cl 1.4-6.0 (p=0.005)	Reason: unclear if the outcome
relatives of current and	- CED ≥12,000 mg/m ² vs. none: OR 5.6;	assessors were blinded for
former patients	95% CI 2.8-10.9 (p<0.001)	important determinants related
	- Unilateral orchiectomy yes vs. no: OR	to the outcome.
	2.4; 95% Cl 0.5-10.7 (p=0.25)	
	- Age at diagnosis 5-9.9 years vs. 0-4.9	D. Confounding
	years: OR 1.8; 95% Cl 1.0-3.3 (p=0.06)	Low risk
	- Age at diagnosis 10-14.9 years vs. 0-4.9	Reason: important prognostic
	years: OR 1.1; 95% Cl 0.6-2.2 (p=0.73)	factors (i.e. age at diagnosis, age
	- Age at diagnosis ≥15 years vs. 0-4.9	at assessment and gonadotoxic
	years: OR 0.8; 95% Cl 0.4-1.8 (p=0.66)	treatment) were taken
	- Age at study 26-35.9 years vs. 18-25.9	adequately into account.
	years: OR 2.5; 95% Cl 1.1-5.7 (p=0.026)	
	- Age at study 36-45.9 years vs. 18-25.9	
	years: OR 3.7; 95% Cl 1.6-8.6 (p=0.003)	
	- Age at study ≥46 years vs. 18-25.9	
	years: OR 5.3; 95% Cl 2.0-13.6 (p<0.001)	
	 Non-Hispanic black ethnicity vs. non- 	
	Hispanic white ethnicity: OR 1.8; 95% Cl	
	1.0-3.4 (p=0.06)	
	 Other ethnicity vs. non-Hispanic white 	
	ethnicity: OR 1.3; 95% Cl 0.4-4.5	
	(p=0.69)	
	- Body mass index <18.5 kg/m² vs. ≥18.5-	
	24.9 kg/m ² : OR 1.1; 95% CI 0.2-5.5	
	(p=0.90)	
	- Body mass index 25-29.9 kg/m ² vs.	
	≥18.5-24.9 kg/m²: OR 1.1; 95% CI 0.6-	
	2.0 (p=0.82)	
	- Body mass index ≥30 kg/m² vs. ≥18.5-	
	24.9 kg/m ² : OR 1.8; 95% CI 1.0-3.3	
	(p=0.06)	

Risk factors for Leydig cell dysfunction in	
multivariable logistic regression analysis:	
- Testicular radiation dose >0-11.9 Gy vs.	
none: OR 20; 95% Cl 1.0-3.8 (p=0.05)	
- Testicular radiation dose 12-19.9 Gy vs.	
none: OR 17.7; 95% CI 5.8-50.1	
(p<0.001)	
- Testicular radiation dose ≥20 Gy vs.	
none: OR 38.9; 95% CI 4.2-358.8	
(p=0.001)	
- CED >0->4,000 mg/m ² vs. none: OR 0.8;	
95% CI 0.3-1.7 (p=0.52)	
- CED 4,000-<8,000 mg/m ² vs. none: OR	
3.3; 95% CI 2.0-5.3 (p<0.001)	
- CED 8.000-<12.000 mg/m ² vs. none: OR	
3.4; 95% Cl 2.1-5.5 (p=0.005)	
- CED ≥12.000 mg/m ² vs. none: OR 6.4:	
95% CI 4.0-10.0 (p<0.001)	
- Unilateral orchiectomy ves vs. no: OR	
4.1: 95% CI 1.7-9.5 (p=0.01)	
- Age at diagnosis 5-9.9 years vs. 0-4.9	
vears: OR 1.4: 95% CI 0.9-2.1 (p=0.15)	
- Age at diagnosis 10-14.9 years vs. 0-4.9	
vears: OB 1.3: 95% CI 0.8-2.0 (p=0.27)	
- Age at diagnosis >15 years vs. 0.49	
vears: OR 1.3: 95% CI 0.8-2.1 (p=0.23)	
- Age at study 26-35 9 years vs. $18-25.9$	
vears: OR 1 7: 95% CI 1 1-2 7 (n=0.028)	
- Age at study 36-45 9 years vs. 18-25 9	
vears: OR 1 9: 95% CI 1 1-3 3 (n=0.014)	
- Age at study >46 years vs. $18-25.9$	
vears: OR 3.0: 95% CI 1.6-5.3 (n<0.001)	
- Non-Hispanic black ethnicity vs. non-	
Hispanic white ethnicity: OR 1 5: 95% Cl	
1 0-2 3 (n=0 07)	

 Other ethnicity v ethnicity: OR 1.2 (p=0.74) Body mass index 24.9 kg/m²: OR 1 (p=0.26) Body mass index ≥18.5-24.9 kg/m² 0.9 (p=0.007) Body mass index 24.9 kg/m²: OR 0 (p=0.002) 	s. non-Hispanic white ; 95% Cl 0.5-2.7 <18.5 kg/m ² vs. ≥18.5- 6; 95% Cl 0.7-3.6 25-29.9 kg/m ² vs. ² : OR 0.6; 95% Cl 0.4- ≥30 kg/m ² vs. ≥18.5- 0.5; 95% Cl 0.3-0.8
Among 683 prospe survivors, progress function to Leydig Leydig cell failure (significantly associa (p=0.025)	actively followed ion from normal cell dysfunction or n=25) was ated with higher CEDs

Abbreviations: CCS, childhood cancer survivors; CED, cyclophosphamide equivalent dose.

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Green et al. Cumulative alkylating agent exposure and semen parameters in adult survivors of childhood cancer: a report from the St Jude Lifetime Cohort Study. Lancet Oncol 2014;15:1215-1223

Study design Treatment era Years of follow-up	Participants	Treatment	Main outcomes	Additional remarks
<u>1. Study design</u>	<u>1. Type and number of</u>	<u>1. Chemotherapy</u>	1. Outcome definitions	<u>1. Remarks</u>
Single-center	<u>participants</u>	- Alkylating agents: 214 (100%)	- Azoospermia: sperm concentration 0	Unable to identify threshold dose
cohort study	214 adult male CCS	- Cyclophosphamide: 195 (91.1%)	- Oligospermia: sperm concentration >0 -	(substantial overlap). Impaired
	treated with alkylating	- Ifosfamide: 26 (12.1%)	<15 x 10 ⁶ /mL	spermatogenesis unlikely when
2. Treatment era	agent therapy, but no	- Procarbazine: 2 (0.9%)	- Normospermia: sperm concentration	CED <4,000 mg/m ² (88.6%
1970-2002	radiotherapy,	- Chlormethine	≥15 x 10 ⁶ /mL	normospermia in when CED
	vasectomy or bilateral	(mechlorethamine): 1 (0.5%)		<4,000 mg/m2).
3. Follow-up	orchiectomy	- Chlorambucil: 1 (<1%)	2. Results	
Median 21.0 years		- Busulfan: 3 (1.4%)	Spermatogenesis and CED:	2. Risk of bias
since diagnosis	2. Diagnoses	- Cyclophosphamide equivalent	- Azoospermia: 53 (24.7%); mean CED	A. Selection bias
(10.5-41.6)	ALL 70 (33%), AML 5	dose (CED): Median 7,400 (1,000-	10,830 mg/m ² (SD 7,274)	High risk
	(2%), NHL 53 (25%), HL	41,311) mg/m ²	- Oligospermia: 59 (27.6%); mean CED	Reason: Of the 549 men eligible,
	2 (1%), NB 26 (12%),	- Cisplatin/carboplatin: 44 (20.6%)	8,480 mg/m ² (SD 4,264)	226 (41%) agreed to participate;
	osteosarcoma 32	- Dacarbazine: 3 (1.4%)	- Normospermia: 102 (47.7%); mean CED	demographic and treatment
	(15%), other 26 (12%)		6,626 mg/m ² (3,576)	characteristics were not equally
		2. Radiotherapy	- Correlation CED and sperm	distributed among the
	3. Age at diagnosis	0 (0%)	concentration: r=-0.37 (p<0.0001)	participants and non-participants.
	Median 7.7 (0.01-20.3)			
	yr	<u>3. Surgery</u>	Risk factors for azoospermia as compared	B. Attrition bias
		Bilateral orchiectomy: 0 (0%)	to normospermia in multinomial logistic	Low risk
	4. Age at follow-up		regression analysis:	Reason: Of the 226 men who
	Median 29.0 (18.4-		- CED per 1,000 mg/m ² :	agreed to participate, 214
	56.1) yr		OR 1.22; 95% CI 1.11-1.34 (p<0.0001)	(94.7%) produced semen
			 Age at diagnosis per years: 	specimen.
	5. Controls		OR 0.97; 95% Cl 0.91-1.05 (p=0.45)	
	No controls		 Age at assessment per years: 	C. Detection bias
			OR 0.99, 95% Cl 0.94-1.05 (p=0.8)	Unclear
				Reason: unclear if the outcome
				assessors were blinded for

	Risk factors for oligospermia as compared	important determinants related
	to normospermia in multinomial logistic	to the outcome.
	regression analysis:	
	- CED per 1,000 mg/m ² :	D. Confounding
	OR 1.14; 95% CI 1.04-1.25 (p=0.006)	Low risk
	 Age at diagnosis per years: 	Reason: important prognostic
	OR 0.95; 95% Cl 0.89-1.02 (p=0.13)	factors (i.e. age at diagnosis and
	- Age at assessment per years:	age at assessment) were taken
	OR 0.97, 95% Cl 0.92-1.03 (p=0.28)	adequately into account.

Abbreviations: ALL, acute lymphoblastic leukaemia; AML, acute myeloid leukaemia; CED, cyclophosphamide equivalent dose; HL, Hodgkin lymphoma; NB, neuroblastoma; NHL, non-Hodgkin lymphoma; OR, odds ratio; 95% CI, 95% confidence interval; SD, standard deviation.

Who should be counselled about fertility preservation?

Green et al. Effect of cranial radiation on sperm concentration of adult survivors of childhood acute lymphoblastic leukaemia: a report from the St. Jude Lifetime Cohort Study. Human Reproduction 2017;32:1192-1201.

Study design				
Treatment era	Participants	Treatment	Main outcomes	Additional remarks
Years of follow-up				
1. Study design	1. Type and number of	<u>1. Chemotherapy</u>	1. Outcome definitions	<u>1. Remarks</u>
Single-center	<u>participants</u>	 Cyclophosphamide: 241 (100%) 	- Azoospermia: sperm concentration 0	Large cohort study.
cohort study	241 adult male	 Mechlorethamine: 1 (0.4%) 	- Oligospermia: sperm concentration >0 -	
	childhood ALL survivors	 Cyclophosphamide equivalent 	<15 x 10 ⁶ /mL	2. Risk of bias
2. Treatment era	treated with alkylating	dose (CED) (mg/m ²) CRT patients:	- Normospermia: sperm concentration	A. Selection bias
1970-2002	agent therapy, but no	>0-<4000: 7 (7.7%)	≥15 x 10 ⁶ /mL	High risk
	vasectomy, patients	≥4000-8000: 17 (18.7%)		Reason: Of the 380 men eligible,
3. Follow-up	receiving androgen	≥8000 -12000: 50 (54.9%)	2. Results	241 (63.4%) participated;
CRT: Mean 26.3 ±	replacement therapy,	≥12000: 17 (18.7%)	Spermatogenesis (n=173):	treatment characteristics were
6.3 yr	testicular radiation or	 CED (mg/m²) no CRT patients: 	- Normospermia: 62 (35.8%)	not equally distributed among
No CRT: Mean 18.7	CRT >26 Gy	>0-<4000: 10 (12.2%)	CRT: 32 (35.2%)	the participants and non-
± 6.0 yr		≥4000-8000: 18 (22.0%)	No CRT: 30 (36.6%)	participants.
	2. Diagnoses	≥8000 -12000: 50 (61.0%)	- Azoospermia: 65 (37.6%)	
	241 (100%) ALL	≥12000: 4 (4.9%)	- Oligospermia: 46 (26.6%)	B. Attrition bias
				High risk
	3. Age at diagnosis	2. Radiotherapy		Reason: Of the 241 men who
	CRT: Mean 6.6 ± 4.4 yr			agreed to participate, 173

No CRT: Mean 7.5 ± 5.0	Hypothalamic-pituitary radiation	Risk factors for azoospermia or	(71.8%) underwent semen
yr	>0-20 Gy: 81 (33.6%)	oligospermia in univariable log-binominal	analysis.
	>20-26 Gy: 53 (22.0%)	regression analysis:	
4. Age at follow-up		- CED (mg/m ²) ≥4000-8000 vs. >0-<4000:	C. Detection bias
CRT: Mean 32.9 ± 7.8 yr	<u>3. Surgery</u>	RR 1.46 (95% Cl 0.71-2.99)	Unclear
No CRT: Mean 26.2 ±	Bilateral orchiectomy: 0 (0%)	- CED (mg/m ²) ≥8000-1200 vs. >0-<4000:	Reason: unclear if the outcome
5.6 yr		RR 1.98 (95% CI 1.03-3.82)	assessors were blinded for
		- CED (mg/m ²) ≥12000 vs. >0-<4000:	important determinants related
5. Controls		RR 2.29 (95% CI 1.17-4.51)	to the outcome.
No controls		- CED per 1000 mg/m ² :	
		RR 1.01 (95% CI 1.00-1.02)	D. Confounding
		- CRT >0-20 Gy vs. 0:	Low risk
		RR 0.99 (95% CI 0.70-1.28)	Reason: important prognostic
		- CRT ≥20-26 Gy vs. 0:	factors (i.e. age at diagnosis and
		RR 1.09 (95% CI 0.81-1.46)	age at assessment) were taken
		- Age at diagnosis (yr) 5-9 vs. <4:	adequately into account (only the
		RR 1.3 (95% CI 1.05-1.61)	significant factors in univariable
		- Age at diagnosis (yr) ≥10 vs. <4:	analysis were included in the
		RR 0.92 (0.69-1.23)	multivariable analysis).
		- Age at semen analysis (yr) 26-35 vs. 18-	
		25: RR 0.82 (95% Cl 0.60-1.04)	
		- Age at semen analysis (yr) ≥35 vs. 18-25:	
		RR 0.94 (95% Cl 0.69-1.26)	
		- Time from diagnosis to semen analysis	
		per yr: RR 1.00 (95% Cl 0.98-1.01)	
		Risk factors for azoospermia or	
		oligospermia in multivariable log-	
		binominal regression analysis:	
		- CED (mg/m ²) ≥4000-8000 vs. >0-<4000:	
		RR 1.42 (95% CI 0.70-2.89)	
		- CED (mg/m ²) ≥8000-1200 vs. >0-<4000:	
		RR 2.06 (95% CI 1.08-3.94)	
		- CED (mg/m²) ≥12000 vs. >0-<4000:	
		RR 2.12 (95% CI 1.09-4.12)	
		- Age at diagnosis (yr) 5-9 vs. <4:	
		RR 1.3 (95% CI 1.05-1.61)	

	- Age at diagnosis (yr) ≥10 vs. <4: RR 0.92 (0.69-1.23)	

Abbreviations: ALL, acute lymphoblastic leukaemia; CED, cyclophosphamide equivalence dose; CRT, cranial radiotherapy.

Who should be counselled about fertility preservation?

Isaksson et al. High risk of hypogonadism in young male cancer survivors. Clinical Endocrinology 2018;88:432-441

Study design Treatment era Years of follow-up	Participants	Treatment	Main outcomes	Additional remarks
<u>1. Study design</u>	1. Type and number of	1. Treatment subgroups	1. Outcome definitions	<u>1. Remarks</u>
Multi-centre	participants	- Chemotherapy: 29 (23%) of	Biochemical hypogonadism:	Treatment subgroup other
cohort study	125 male childhood	whom 16 (13%) alkylating	 Primary hypogonadism: total 	radiotherapy and chemotherapy:
	cancer survivors	agents	testosterone <10 nmol/L, LH and FSH	12/21 (57%) received alkylating
2. Treatment era		- Cyclophosphamide	both >10 IU/L with FSH > LH or total	agents with a median CED of
1970-2002	2. Diagnoses	equivalent dose (CED) >4000	testosterone <10 nmol/L, LH \leq 10 IU/L and	9593 mg/m ² . One case received
	27(22%) leukaemia, 28	mg/m ² : 10 (8.0%)	FSH >10 IU/L	high-dose chemotherapy
<u>3. Follow-up</u>	(22%) intracranial	 Radiotherapy to testes: 5 	 Secondary hypogonadism: total 	followed by autologous BMT.
Mean 24.3 (±7.1)	tumour, 21 (17%)	(4.0%)	testosterone <10 nmol/L, LH and FSH	
years after	lymphoma, 6 (4.8%)	 Cranial radiotherapy: 12 	both ≤10 IU/L	2. Risk of bias
treatment	testicular cancer, 8	(9.6%)	 Compensated hypogonadism: total 	<u>A. Selection bias</u>
	(6.4%) Wilms tumour, 6	- Cranial radiotherapy and	testosterone ≥10 nmol/L, LH >10 IU/L	High risk
	(4.8%) bone tumour, 29	chemotherapy: 16 (13%)	 Or ongoing androgen replacement 	Reason: 125/427 (29%) eligible
	(23%) other	- Other radiotherapy: 5 (4.0%)	therapy	survivors included in this study.
		- Other radiotherapy and		
	3. Age at diagnosis	chemotherapy: 23 (18%)	2. Results	B. Attrition bias
	Median 9.6 (5.4-15.0)	- Brain surgery: 15 (12%)	Mean (SD) testosterone levels childhood	Low risk
	years	- Surgery other than brain	cancer survivors vs. controls:	Reason: 121/125 (97%) of
		surgery: 15 (12%)	- Total testosterone: 15.4 (6.22) vs. 15.5	included survivors had an
	4. Age at follow-up		(6.01), p=0.87	outcome measure.
	Median 33.7 (30.2-40.1)		- Free testosterone: 0.313 (0.104) vs. 0.311	
	years		(0.099), p=0.86	C. Detection bias
				Unclear
	<u>5. Controls</u>			Reason: unclear if the outcome
				assessors were blinded for

125 age-matched	Hypogonadism childhood cancer survivors	important determinants related
controls from the	(n=121 assessed) vs. controls (n=122	to the outcome.
Swedish Population	assessed):	
Register	- 31 (26%) vs. 17 (14%)	D. Confounding
	- OR 2.1 (95% CI 1.1-4.1)	Low risk
	- Survivors: primary hypogonadism n=7.	Reason: survivors matched to
	secondary hypogonadism n=9.	controls.
	compensated hypogonadism n=2.	
	ongoing testosterone replacement	
	therapy n=13	
	- Controls: primary hypogonadism n=1,	
	secondary hypogonadism n=16	
	, ,, ;	
	Risk factors for hypogonadism in bivariate	
	logistic regression analysis (childhood	
	cancer survivors vs. controls):	
	- Chemotherapy: OR 1.1 (95% CI 0.34-3.8)	
	- CED >4000 mg/m ² : OR 2.0 (95% CI 0.36-	
	11.0)	
	- Radiotherapy to testes: OR 28.0 (95% CI	
	2.9-279.0)	
	- Cranial radiotherapy: OR 4.4 (95% Cl 1.1-	
	18.0)	
	 Cranial radiotherapy and chemotherapy: 	
	OR 1.7 (95% CI 0.42-6.7)	
	- Other radiotherapy: OR 0.92 (95% CI 0.09-	
	9.5)	
	 Other radiotherapy and chemotherapy: 	
	OR 3.7 (95% CI 1.3-10.0)	
	 Brain surgery: OR 0.94 (95% CI 0.19-4.7) 	
	 Surgery other than brain surgery: OR 1.0 	
	(95% CI 0.26-3.9)	

Who should be counselled about fertility preservation?

Jahnukainen et al. Semen quality and fertility in adult long-term survivors of childhood acute lymphoblastic leukemia. Fertility and Sterility 2011; 96(4): 837-42.

Study design Treatment era Years of follow-up	Participants	Treatment	Main outcomes	Additional remarks
1. Study design	1. Type and number of	<u>1. Chemotherapy</u>	1. Outcome definitions	<u>1. Strengths</u>
Single-centre	<u>participants</u>	Cyclophosphamide: 26 (51.0%)	- Semen analysis	- Semen analysis
cohort study	51 male ALL survivors	1-6 g/m2: 13 (25.5%) 7-10 g/m2: 8 (15.7%)	- Hormone levels: testosterone, LH, FSH	- Control group
2. Treatment era	2. Diagnoses	>20 g/m2: 5 (9.8%)	2. Results	2. Limitations
1970-1995	51 (100%) ALL		Median (IQR) sperm concentration	- Small sample size especially
		2. Radiotherapy	(10 ⁶ /mL) CCS vs. controls:	when separating by treatment.
3. Follow-up:	3. Age at diagnosis	Cranial radiation: 38 (74.5%)	- Controls (n=56): 50 (27-66)	- Multiple treatments in a given
Median 20 (11-30)	Median 5 (1-15) yr	18 Gy: 5 (9.8%)	 No cyclophosphamide, no testicular 	group make it difficult to link a
years after		24 Gy: 31 (60.8%)	irradiation (n=16): 41 (29-74); p>0.05	given predictor to outcomes.
treatment	4. Age at follow-up	>24 Gy: 2 (3.9%)	 ≤10 g/m² cyclophosphamide, no 	
	Median 29 (26-38) yr	Testicular radiation: 18 (35.3%)	testicular irradiation (n=12): 35 (24-42);	<u>3. Risk of bias</u>
		10 Gy: 2 (3.9%)	p>0.05	A. Selection bias:
	<u>5. Controls</u>	24 Gy: 16 (31.4%)	 >20 g/m² cyclophosphamide, no 	High risk
	56 age-matched males	Spinal radiation (6 Gy): 1 (2.0%)	testicular irradiation (n=5): 1 (0-17);	Reason: Of 164 treated for ALL,
	median 30 (25-36) yr		p<0.05	only 77 were alive and 2 moved.
		<u>3. Surgery</u> 0 (0%)	 Testicular irradiation (n=10) ± cyclophosphamide (n=8): 0; p<0.05 	51/77 enrolled (66.2%).
			- Same results for total sperm count	B. Attrition bias
			- No significant differences in percentage	Low risk
			of motile sperm and morphologic	Reason: 47/51 (92.2%) survivors
			normal sperm, semen volume	provided semen.
			Median (IQR) testosterone levels (pmol/L)	<u>C. Detection bias:</u>
			$\frac{\text{CCS vs. controls:}}{\text{CCS vs. controls:}}$	Unclear
			- Controls (n=56): 18.4 (14.7-24.0)	Reason: unclear if the outcome
			- NO CYCIOPNOSPNAMICE, NO TESTICULAR	assessors were blinded for
			irradiation (n=16): 18.3 (13.6-20.1);	determinants related to
			p>0.05	outcomes
1	1			

	- ≤10 g/m² cyclopnospnamide, no	D. Confounding:
	testicular irradiation (n=12): 12.7 (12.2-	Low risk
	16.6); p<0.05	Reason: survivors matched to
	 >20 g/m² cyclophosphamide, no 	controls
	testicular irradiation (n=5): 13.4 (7.7-	
	17.5); p<0.05	
	- Testicular irradiation (n=10) ±	
	(n=8): 1.4 (0.9-8.9):	
	n<0.05	
	þ (0.05	
	Median (IOP) I H levels (III/I) CCS vs	
	sontrole	
	$\frac{\text{controls.}}{(n-F(k), 2, 2, (2, 8, 4, 2))}$	
	- Controls (11=50): 3.3 (2.8-4.2)	
	- No cyclophosphamide, no testicular	
	irradiation (n=16): 3.5(2.7-4.6); p>0.05	
	 ≤10 g/m² cyclophosphamide, no 	
	testicular irradiation (n=12): 3.8 (3.0-	
	4.5); p>0.05	
	 >20 g/m² cyclophosphamide, no 	
	testicular irradiation (n=5): 5.3 (2.6-7.0);	
	p>0.05	
	 Testicular irradiation (n=10) ± 	
	cyclophosphamide (n=8): 6.4 (5.6-14.6);	
	p<0.05	
	•	
	Median (IOR) FSH levels (IU/L) CCS vs.	
	controls:	
	- Controls $(n=56)$: 3.2 (1.9-4.1)	
	- No cyclophosphamide, no testicular	
	irradiation $(n-16)$: 2.5 (2.1-4.2): n>0.05	
	$\sim < 10 \text{ g/m}^2$ cyclophosphamida, no	
	- 210 g/m cyclophosphalmuc, 10	
	(1-12), 4.7 (2.0-	
	7.4); $P < 0.05$	
	- >20 g/m² cyclopnospnamide, no	
	testicular irradiation (n=5): 11.1 (5.5-	
	20.8); p<0.05	

25.01, 010.05

Abbreviations: ALL, acute lymphoblastic leukaemia; CCS, childhood cancer survivors; FSH, follicle stimulating hormone; IQR, inter quartile range; LH, luteinizing hormone.

Who should be counselled about fertility preservation?					
Lopez Andreu et al.	Lopez Andreu et al. Persistent altered spermatogenesis in long-term childhood cancer survivors. Pediatr Hematol Oncol 2000;17:21-30				
Study design Treatment era Years of follow-up	Participants	Treatment	Main outcomes	Additional remarks	
<u>1. Study design</u> Single-centre	<u>1. Type and number of</u> <u>participants</u>	<u>1. Chemotherapy</u> Cyclophosphamide: 9 (20.9%);	<u>1. Outcome definitions</u> - Moderate oligozoospermia:	<u>1. Remarks</u> A forward-stepping logistic	
cohort study	43 male CCS >16 years of age at follow-up	1200-27,200 mg/m ²	- Severe oligozoospermia:	regression analysis was performed. In addition to	
<u>2. Treatment era</u> Treatment era not	2. Diagnoses	- Cranial irradiation:	- Moderate asthenozoospermia:	dose, FSH level was included as	
3 Follow-up	ALL (n=21), AML (n=1), NB (n=8), GNB (n=1), GN	- Spinal irradiation:	- Severe asthenozoospermia:	volume was excluded from the	
Mean 13.6 (3.9-	(n=9), mesoblastic	treated with cranial radiation)	- Infertile: azoospermia or severe oligo-	up duration were not included in	
treatment	2 Age at diagnosis	1 (2.3%); 30 Gy (also treated		1 survivor was on replacement	
	Range 0.0-9.4 years	radiation)	<u>Spermatogenesis:</u>	therapy after 30 Gy testicular	
	<u>4. Age at follow-up</u> Mean 20.2 (16.1-30.6)	2 (4.7%); 30 Gy	- Moderate oligozoospermia: 1 (2.3%)	cyclophosphamide.	
	years		- Moderate asthenozoospermia: 8 (18.6%)	2. Risk of bias	
	5. Controls		- Moderate oligo-asthenozoospermia: 2	Unclear Reason: Unclear how many	
	aged ≤30 years; 373		- Severe oligo-asthenozoospermia: 2 (4.6%)	patients were included in the	
	consulting for infertility		asthenozoospermia: 3 (7.0%)		

	- Infertile: 10 (23.2 %)	B. Attrition bias:
		Low risk
	Risk factors for infertility in multivariable	Reason: 90% of the eligible
	analyses.	survivors were evaluated
	Cumulative cyclophosphamide doso	survivors were evaluated.
	- Culturative cyclophosphannue dose	C Detection bios
	significant (no effect measure reported)	C. Detection blas:
	- FSH level significant (no effect measure	Unclear
	and p-value reported)	Reason: unclear if the outcome
		assessors were blinded for
	Univariable results cyclophosphamide:	important determinants related
	- 5/9 (55.6%) treated with	to the outcome.
	cyclophosphamide azoospermic and 1/9	
	(11.1%) severe oligozoospermic	D. Confounding
	- Cumulative cyclophosphamide dose	High risk:
	negatively correlated with sperm count.	Reason: Models did not include
	r_{-0} (n=0.004)	age radiation therapy other
	- Cumulative cyclophosphamide dose	chemo agents therapy
	- cumulative cyclophosphannue dose	duration /follow up time
	negatively correlated with sperm motility.	duration/follow-up time.
	r=-0.45 (p=0.002)	
	Univariable results cranial radiotherapy:	
	 No correlation with sperm count, sperm 	
	motility, testicular volume, FSH level	

Abbreviations: ALL, acute lymphoblastic leukaemia; AML, acute myeloid leukaemia; FSH, follicle-stimulating hormone; GNB, ganglioneuroblastoma; GN, ganglioneuroma; NB, neuroblastoma.

Who should be counselled about fertility preservation?

Mackie et al. Gonadal function following chemotherapy for childhood Hodgkin's disease. Med Pediatr Oncol 1996;27:74-78

Study design Treatment era Years of follow-up	Participants	Treatment	Main outcomes	Additional remarks
1. Study design	1. Type and number of	1. Chemotherapy	<u>1. Outcome definitions:</u>	<u>1. Remarks</u>
National multi-	participants	ChIVPP: 58 (100%); 6-8	- Leydig cell dysfunction:	- Relatively narrow range of
centre trial cohort	58 postpubertal male	cycles;	LH >10 IU/L	cumulative doses
UK CCSG study	survivors of childhood	Chlorambucil 504-672	Testosterone <10 nmol/L	- LH and testosterone only described
	Hodgkin disease treated	mg/m ² , vinblastine,	- Abnormal gonadotropin levels:	clearly in survivors with increased FSH
2. Treatment era	according to UKCCSG HD	prednisolone,	FSH >10 IU/L	levels.
Not reported	Trial 8201	procarbazine 8,400-		- Note concerns about outcome
		11,200 mg/m ²	2. Results	definitions: Leydig cell dysfunction
3. Follow-up	2. Diagnoses	_	Leydig cell dysfunction (个 LH) (n=41 assessed):	based on high LH rather than low
Median 6 (2.5-	58 (100%) Hodgkin	2. Radiotherapy below	10 increased (24.4%); range of these	testosterone;
11.1) years from	disease	<u>diaphragm</u>	increased LHs, 10.3-18 IU/L	 Cutoff of 10 may be too high
diagnosis		0 (0.0%)		 High LH definition not
	3. Age at diagnosis		$\sqrt{1}$ testosterone levels (n=37 assessed):	appropriate if patient received
	Mean 12.2 (8.2-15.3)	3. Surgery	5 decreased (13.5%); range of all	cranial RT (not specified in this
	years	0 (0.0%)	testosterones, 4.3-37.8 nmol/L	paper albeit unlikely)
				- Azoospermia present in 7 survivors in
	4. Age at follow-up		<u>↑ FSH levels (n=46 assessed):</u>	whom semen analyses were
	Not reported		41 increased (89.1%); range of increased	performed. All progressed
			FSHs, 10.8-40.7 IU/L	spontaneously through puberty.
	5. Controls			- Unclear how "amount of
	No controls		Risk factors for Leydig cell dysfunction in	chemotherapy" is defined.
			multivariable regression analyses:	- Multiple regression performed for
			- Amount of chemotherapy, NS	gonadotropins and Leydig cell
			- Age at treatment, NS	dysfunction. However, methodology of
			- Follow-up duration, NS	testosterone analysis not clear.
			(no effect measures reported)	
				2. Risk of bias
			<u>Risk factors for ↑ FSH in multiple regression</u>	A. Selection bias
			analyses:	High risk

1			1
		- Age or pubertal status at time of treatment,	Reason: 101 out of 168 (60.1%) eligible
		NS	male and female CCS included in this
		- Follow-up duration, NS	study.
		(no effect measures reported)	
			B. Attrition bias
			High risk
			Reason: 41 out of 58 (70.7%) CCS had
			I H measured and 37 out of 58 (63 8%)
			had testosterone measured
			had testosterone medsared
			C Detection bias
			<u>C. Detection bias</u>
			Unclear
			Reason: unclear if the outcome
			assessors were blinded for important
			determinants related to the outcome.
			D. Confounding
			Low risk
			Reason: analyses were adjusted for
			age at treatment and follow-up
			duration.
			aaracioni

Abbreviations: FSH, follicle-stimulation hormone; LH, luteinizing hormone; NS, not significant.

Who should be counselled about fertility preservation?

Romerius et al. Hypogonadism risk in men treated for childhood cancer. Clin Endocrinol Metab 2009;94:4180-4186

Study design Treatment era Years of follow-up	Participants	Treatment	Main outcomes	Additional remarks
<u>1. Study design</u>	<u>1. Type and Number of</u>	<u>1. Chemotherapy</u>	1. Outcome definition	<u>1. Remarks</u>
Single-centre	Participants	Chemotherapy alone, n=38 (26.4%)	Hypogonadism:	 Blood sampling was done
cohort study	144 male childhood	Of whom n=21 (14.6%) alkylating	 testosterone levels <10 nmol/L and/or 	between 0800 and 1600 h; the
	cancer survivors who	agents	LH >10 IU/L	proportion of truly hypogonadal
2. Treatment era	were >18-45 years of		 or receiving androgen replacement 	men maybe lower than the
1970-2002	age	2. Radiotherapy	therapy	reported 23%.
		 Radiotherapy to testes, n=6 		- The definition of hypogonadism
3. Follow-up	2. Diagnoses	(4.2%) (mean dose 21 Gy)	2. Results	does not include free
Mean 20 years	Leukaemia: 26 (18.1%)	 Radiotherapy alone 	Hypogonadism, survivors vs. controls:	testosterone; BMI (mean 25 in
since diagnosis (±	Brain tumours: 31	(nontesticular), n=17 (11.8%)	- n=33 (22.9%) vs. n=6 (4.3%)	the study group) can influence
7.3)	(21.5%)	- Mean cranial irradiation dose 38	- OR 6.7 (95% CI 2.7-17.0)	SHBG resulting in false low total
	Lymphoma: 32 (22.2%)	Gy and mean dose directly on		testosterone.
	Testicular cancer: 9	testis 21Gy	Risk factors for hypogonadism in binary	- The clinical relevance of
	(6.3%)	3. Chemotherapy and radiotherapy	logistic regression analysis; survivors vs.	testosterone deficiency is difficult
	Wilms' tumour: 11	n=49 (34.0%)	<u>controls:</u>	to interpret because of the lack
	(7.6%)		 Leukaemias (31% hypogonadal): 	of information concerning
	Others: 35 (24.3%)	<u>4. Surgery</u>	OR 10.0 (95% CI 3.1-32.0)	complaints of hypogonadism.
		 Brain surgery: n=17 (11.8%) 	 Brain tumours (19% hypogonadal): 	
	3. Age at diagnosis	 Surgery alone (except brain 	OR 5.4 (95% CI 1.6-18.0)	2. Risk of bias
	Median 10 (0.1-17)	surgery): n=17 (11.8%)	 Lymphomas (31% hypogonadal): 	A. Selection bias
	years		OR 10.0 (95% CI 3.4-31.0)	High risk
			 Testicular cancer (22% hypogonadal): 	Reason: 151 out of 397 (38.0%)
	4. Age at follow-up		OR 6.4 (95% CI 1.1-38.0)	eligible survivors included in the
	Median 30 (20-46)		• Wilms' tumour (9.1% hypogonadal):	study.
	years		OR 2.3 (95% CI 0.25-21.0)	
			• Others (17% hypogonadal):	B. Attrition bias
	5. Controls		OR 4.7 (95% CI 1.4-15.0)	Low risk
	141 healthy fertile men		- Chemotherapy alone (26%	Reason: hormonal analyses were
	(none on any fertility		hypogonadal):	assessed in all 144 survivors.
	treatment)			

	OR 8.0 (95% CI 2.7-24.0)	C. Detection bias
	 Radiotherapy to testes (83%) 	Unclear
	hypogonadal):	Reason: unclear if the outcome
	OR 110.0 (95% CI 11.0-1100.0)	assessors were blinded for
	- Radiotherapy alone (nontesticular) (12%	important determinants related
	hypogonadal):	to the outcome.
	OR 3.0 (95% CI 0.56-16.0)	
	 Combination chemotherapy and 	D. Confounding
	radiotherapy (22% hypogonadal):	High risk
	OR 6.5 (95% CI 2.3-19.0)	Reason: controls were not
	 Brain surgery (29% hypogonadal): 	matched to survivors and no
	OR 9.4 (95% CI 2.5-35.0	correction for confounding
	 Surgery alone (except brain surgery) 	factors in analyses.
	(0% hypogonadal): OR 1.0	

Abbreviations: OR, odds ratio; 95% CI, 95% confidence interval.

Who should be counselled about fertility preservation?

Siimes et al. Prophylactic cranial irradiation increases the risk of testicular damage in adult males surviving ALL in childhood. Med Pediatr Oncol 1993;21:117-121

Study design Treatment era Years of follow-up	Participants	Treatment	Main outcomes	Additional remarks
<u>1. Study design</u>	<u>1. Type and number of</u>	<u>1. Chemotherapy</u>	1. Outcome definitions	<u>1. Remarks</u>
Multi-centre	<u>participants</u>	- Cyclophosphamide: 23	- \downarrow testosterone levels: threshold level not	3 patients treated with CRT had
(n=4)cohort study	41 male childhood ALL	(56.1%)	reported	been started on testosterone
	survivors at least 1 yr off	- Cytosine arabinoside: 9	- \uparrow LH levels: threshold level not reported	supplementation from 4 to 9
2. Treatment era	chemotherapy	(22.0%)	- Semen analysis, oligospermia: sperm	years earlier. Their mean
1966-2003		- Vincristine, prednisone, 6-	concentration <20 million/mL	testosterone concentration was
	2. Diagnoses	mercaptopurine,		lower (9.8 U/L at 2 weeks after
3. Follow-up	41 (100%) ALL	methotrexate: 41 (100%)	2. Results	the preceding testosterone
Mean 15.2 (4.0-		- Adriamycin: 21 (51.2%)	Testosterone levels:	injection) than that of the other
25.0) yr from	3. Age at diagnosis	- Asparaginase: 33 (80.5%)	- Cranial radiotherapy: Mean 17.0 (±7.5)	patients at time of the study.
diagnosis	Mean 8.5 (range 1-16) yr		- No cranial radiotherapy: Mean 20.2 (±6.7)	
		2. Radiotherapy	p=0.242	

4. Age at follow-up	- CRT 20-24 Gy: 17 (41.5%)		A forward-stepping linear
Median 21.0 (18.0-27.0)	- Radiation to fields including	LH levels:	regression analysis was used to
yr	testes: 0 (0%)	- Cranial radiotherapy: Mean 8.2 (±8.1)	identify factors independently
	- Both cyclophosphamide	- No cranial radiotherapy: Mean 6.0 (±3.4)	associated with testosterone
5. Controls	and cranial radiotherapy: 12	p=0.456	deficiency. 3 patients with
No controls	(29%)		testosterone supplementation
		Risk factors for lower (but not necessarily	excluded from analysis.
		abnormal) testosterone in multivariable	
		<u>analysis:</u>	Note limitations in interpreting
		- Chemotherapy NS	LH level in patients who have
		- Cranial radiotherapy NS	received cranial radiotherapy (LH
		- Age at diagnosis NS	response may be blunted by
			central hypogonadism.
		Semen analysis:	
		Available in 18 patients: 3 (16.7%)	2. Risk of bias
		azoospermia, 7 (38.9%) oligospermia	A. Selection bias
			Unclear
			Reason: Unclear how many
			patients were included in the
			original cohort of survivors.
			B. Attrition bias
			Low risk
			Reason: All patients had
			testosterone and LH samples
			taken.
			C. Detection bias
			Unclear
			Reason: unclear if the outcome
			assessors were blinded for
			important determinants related
			to the outcome.
			D. Confounding
			Low risk

		Reason: analysis was adjusted for
		chemotherapy, cranial
		radiotherapy and age at
		diagnosis.

Abbreviations: CRT, cranial radiotherapy; LH, luteinising hormone; NS, not significant.

Who should be counselled about fertility preservation?

Tauer et al. Long-term imatinib treatment does not cause testicular toxicity in male adolescents with chronic myeloid leukemia and in a juvenile rat model. Klin Pediatr 2014;226:169-174.

Study design Treatment era Years of follow-up	Participants	Treatment	Main outcomes	Additional remarks
1. Study designProspectivemulticenter study2. Treatment era1978-20003. Follow-upPatients monitoredover a meanperiod 39 (range 0-89) weeks andwere on imatinibprior to entering	1. Type and number of participants13 boys with CML enrolled into the CML- PEAD II trial2. Diagnoses CML n=133. Age at diagnosis Median 13.8 (range 7.9- 18.7) yr4. Age at follow-up	1. Chemotherapy Imatinib: 13 (100%) 260-300 mg/m ² imatinib orally once daily for a minimum period of at least 3 months	1. Outcome definitions Testosterone and inhibin B levels compared to age-related reference ranges 2. Results Testosterone levels: - Compared to age-related reference ranges serum testosterone showed no rising or falling pattern during the course of treatment - Patients aged 11-15 yr showed highest variability of testosterone levels during therapy but within the age-related reference ranges	1. Remarks Small study sample. 2. Risk of bias A. Selection bias Unclear Reason: unclear how many patients were included in the original cohort. B. Attrition bias Low risk Reason: 10/13 (76.9%) patients could be investigated at least 3
the study with a median of 105 (range 12-296) weeks	Not applicable <u>5. Controls</u> No controls		 <u>Inhibin B levels:</u> Compared to age-related reference ranges inhibin B showed no rising or falling pattern during the course of treatment 	time points while the remaining 3 could be investigated once. <u>C. Detection bias</u> Unclear Reason: unclear if the outcome assessors were blinded for important determinants related to the outcome.

		<u>D. Confounding</u> High risk Reason: no adjustment for
		Reason: no adjustment for
		important confounding factors.

Abbreviations: CML, chronic myeloid leukaemia.

Who should be counselled about fertility preservation?					
<i>Tromp et al.</i> Reprod	Tromp et al. Reproductive status in adult male long-term survivors of childhood cancer. Hum Reprod 2011;26:1775-1783				
Study design Treatment era Years of follow-up	Participants	Treatment	Main outcomes	Additional remarks	
1. Study design	1. Type and number of	<u>1. Chemotherapy</u>	1. Outcome definitions	<u>1. Remarks</u>	
Single-centre	participants	Alkylating agents: 336 (59.5%)	- \downarrow testosterone levels: <11 nmol/L	- Higher incidence of decreased	
cohort study	565 male childhood		- 个 LH levels: >15 U/L	testosterone levels compared to	
	cancer survivors who	2. Radiotherapy	- 个FSH levels: >10 U/L	elevated LH levels. Note higher	
2. Treatment era	survived ≥5 years from	- Pelvic/abdominal		LH threshold in this study, but	
1966-2003	diagnosis and were ≥18	irradiation only: 51 (9.0%)	2. Results	note that one third of cancer	
	years at follow-up	- Cranial irradiation only: 120	\downarrow testosterone levels (n=460 assessed):	survivors have elevated FSH	
3. Follow-up		(21.2%)	- 57 CCS (12.4%)	levels.	
Median 15 (range	2. Diagnoses	 Cranial + pelvic/abdominal 	- Mean testosterone level 17.2 nmol/L, SD 5.5	- Normal LH levels do not exclude	
5.0-39.0) yr since	Lymphoma 154 (27.3%)	irradiation: 4 (0.7%)		testosterone deficiency in this	
diagnosis	Leukaemia 125 (22.1%)	- TBI: 11 (1.9%)	↑ LH levels (n=489 assessed):	group – clinically, testosterone	
	Soft tissue sarcoma 70	- Radiation to fields including	- 14 CCS (2.9%)	and LH need be assessed	
	(12.3%)	testes: Not reported	- Median LH level 6.0 U/L (1.0-40.0)	together, especially in patients	
	Kidney tumour 64			who received cranial irradiation	
	(11.3%)	3. Surgery testicular region:	<u> ↑FSH levels (n=488 assessed):</u>	(in whom LH response may be	
	Bone tumour 53 (9.4%)	38 (6.7%)	161 (33%)	blunted by central	
	Brain/CNS tumour 47		- Median FSH level 6.0 U/L (0,1-72.7)	hypogonadism), but data not	
	(8.3%)	Treatment;		reported	
	Neuroblastoma 19 (3.3%)	Combination chemotherapy	Risk factors for lower (but not necessarily	- Difficult to interpret clinical	
	Testicular tumour 9	and surgery n=172 (30.4%)	abnormal) testosterone in linear regression	relevance of testosterone risk	
	(1.9%)	9 survivors(2.4%)	analysis:	factor analyses since most of the	
		chemotherapy not containing			

Endocrine tumour 5 (0.9%) Other 19 (4.3%) <u>3. Age at diagnosis</u> Median 7.8 years (0.0- 17.8) <u>4. Age at follow-up</u> Median 21.0 years (18.0- 46.0) <u>5. Controls</u> No controls	alkylating agents vinca alkaloid or antmetabolite	 Model 1 – adjusted for age at diagnosis and follow-up duration: TBI yes vs. no: beta -3.53 (p=0.036) Model 2 – adjusted for age at diagnosis, follow-up duration and all other variables as stated below: All treatment variables not significant: procarbazine, cyclophosphamide, other alkylating agents (busulfan, carmustine, mechlorethamine, ifosfamide, lomustine, melphalan, temozolamide – not evaluated separately), cisplatin/carboplatin, antimetabolites, vinca alkaloids, anthracyclines, other chemotherapeutic agents, cranial irradiation, pelvic/abdominal radiation, other irradiation, TBI, surgery testicular region Risk factors for elevated FSH in multivariable logistic regression analysis: Cyclophosphamide yes vs. no: OR 4.23 (95% CI 2.24-8.0) Procarbazine yes vs. no: 	testosterones were in the normal range - The clinical relevance of testosterone deficiency is also difficult to interpret because of the lack of age-specific reference values of hormone levels and the lack of information concerning complaints of hypogonadism - This study does not distinguish between hypogonadotropic hypogonadism and hypergonadotropic hypogonadism 73 men reported that their partner had become pregnant: 120 conceptions resulted in 103 live births and 14 miscarriages. 56 (77%) natural conception. No data presented to prove paternity in these pregnancies.
17.8)		- All treatment variables not significant: procarbazine, cyclophosphamide, other	values of hormone levels and the lack of information concerning
4. Age at follow-up		alkylating agents (busulfan, carmustine,	complaints of hypogonadism
Median 21.0 years (18.0-		mechlorethamine, ifosfamide, lomustine,	- This study does not distinguish
46.0)		melphalan, temozolamide – not evaluated	between hypogonadotropic
5 Controls		antimetabolites vinca alkaloids	hypergonadotronic
No controls		anthracyclines, other chemotherapeutic	hypogonadism
		agents, cranial irradiation, pelvic/abdominal radiation, other irradiation, TBI, surgery testicular region	73 men reported that their partner had become pregnant: 120 conceptions resulted in 103
		Risk factors for elevated FSH in multivariable	live births and 14 miscarriages. 56
		logistic regression analysis:	(77%) natural conception. No
		 Cyclophosphamide yes vs. no: OR 4.23 (95% CI 2.24-8.0) 	paternity in these pregnancies.
		- Procarbazine yes vs. no:	2. Risk of bias
		- Other alkylating yes vs. no:	A. Selection bias
		OR 2.14 (95% Cl 1.14-4.00)	High risk
		- Cisplatin/carboplatin yes vs. no:	Reason: 565 out of 796 (71.0%)
		OR 2.29 (95% CI 0.89-5.89)	this study
		- Antimetabolites yes vs. no:	this study.
		OR 1.15 (95% Cl 0.63-2.07)	B. Attrition bias
		- Anthracyclines yes vs. no: OR 1.06 (95% Cl 0.56-2.00)	Low risk
		- Vinca alkaloids ves vs. no:	Reason: testosterone levels and
		OR 2.80 (95% CI 1.07-7.30)	LH levels were available in 81.4%
		- Other chemo yes vs. no:	and 86.5%, respectively.
		OR 0.90 (95% CI 0.50-1.59)	C. Detection bias

	- Pelvic/abdominal radiotherapy ye	es vs. no: Unclear
	OR 2.35 (95% CI 1.03-5.37)	Reason: unclear if the outcome
	- Cranial radiotherapy yes vs. no:	assessors were blinded for
	OR 0.55 (95% CI 0.28-1.07)	important determinants related
	- TBI not included in the model; All	11 to the outcome (no information is
	survivors treated with TBI had \uparrow	FSH available concerning time of the
	- Other radiotherapy yes vs. no:	day of collection of blood
	OR 1.78 (95% CI 0.95-3.34)	samples).
	- Surgery testicular region:	
	OR 2.61 (95% CI 1.08-6.29)	D. Confounding
	- Age at diagnosis, per yr:	Low risk
	OR 1.08 (95% CI 1.02-1.16)	Reason: the analyses are adjusted
	- Follow-up duration, per yr:	for important factors (adjusted
	OR 1.06 (95% CI 1.01-1.12)	for age at diagnosis, follow-up
		duration and treatment).

Abbreviations: LH, luteinizing hormone; NS, not significant; SD, standard deviation; TBI, total body irradiation; CCS, childhood cancer survivors.

Who should be counselled about fertility preservation?				
Wilhelmsson et al. A 2014;61:1094-1100	dult testicular volume prec	licts spermatogenetic recovery after al	logeneic HSCT in childhood and adolescence. P	ediatr Blood Cancer
Study design Treatment era Years of follow-up	Participants	Treatment	Main outcomes	Additional remarks
1. Study design	1. Type and number of	<u>1. Chemotherapy</u>	1. Outcome definitions	<u>1. Remarks</u>
Two-centre cohort	<u>participants</u>	 Alkylating agents: 106 (100%) 	Azoospermia: no spermatozoa detected in	Testicular volume measured
study	106 male CCS who	 Cyclophosphamide: 84 (79.2%); 	any field of a double-chambered	with orchidometer or ruler &
	received allogeneic	120 mg/kg	haemacytometer before or after	formula. If both testes
2. Treatment era	HSCT >5 years after	- Busulfan: 18 (17.0%)	centrifugation	measured, mean of both used.
1978-2000	treatment and reached	 Cytarabine: 16 (15.1%) 		Adult testicular volume (n=74)
	puberty at follow-up	- Melphalan: 3 (2.8%)	2. Results	documented by Tanner stage 5,
3. Follow-up		- Etoposide: 2 (1.9%)	Spermatogenesis (n=31 assessed):	reached final height, or age >18
Mean 13 (4-28)	2. Diagnoses	2. Radiotherapy	- Azoospermia: 21 (67.7%)	years.
years since HSCT	ALL (n=44), AML (n=20),	- TBI: 71 (67.0%); 10-12 Gy	- TBI: 20/24 (83.3%) azoospermia, 4/24	
	SAA (n=17), other	- CRT for leukaemia: 14 (22%)	(16.7%) non-azoospermia	In the multivariable analysis the
	(n=25); 68 malignant			effect of TBI was evaluated.

HSCT (4 CML, 20 AML,	- Testicular irradiation for	 Cyclophosphamide-only: 0/3 (0%) 	However, 5 patients treated in
44 ALL),	leukaemia: 8 (12%)	azoospermia, 3/3 (100%) non-azoospermia	the no TBI group had received
38 non-malignant HSCT	- TNI: 5 (4.7%); 6 Gy	- Busulfan-based: 1/4 (25%) azoospermia,	TNI, hence potentially exposing
		3/4 (75%) non-azoospermia	the testes to radiotherapy (no
3. Age at HSCT	3. Therapy subgroups	 Leukaemia: 17/19 (89.5%) azoospermia, 	details were provided to
Mean 8.0 (1-17) years	 sTBI + cyclophosphamide: 30 	2/19 non-azoospermia (10.5%)	indicate if testicular shielding
	(28%)		was used). In addition, the no
4. Age at follow-up	 fTBI + cyclophosphamide: 20 	Predictors for active sperm production in	TBI group was also treated with
Mean 22 (12-42) years	(19%)	multivariable logistic regression analysis:	cyclophosphamide, busulfan, or
	 fTBI + cyclophosphamide + 	 No leukaemia diagnosis vs. leukaemia 	both.
5. Controls	etoposide: 2 (2%)	diagnosis: OR 19.8; 95% Cl 1.9-210.3	
No controls	 fTBI + cytarabine: 16 (15%) 	(p<0.01)	Leukaemia was associated with
	 fTBI + melphalan: 3 (3%) 	- Testicular volume ≥15 ml vs. <15 ml:	azoospermia. This might be due
	- Busulfan: 3 (3%)	OR 17.1; 95% CI 1.4-215.8 (p<0.03)	to CRT and/or testicular
	 Busulfan + cyclophosphamide: 15 	- No TBI vs. TBI:	irradiation. It is, however,
	(14%)	p>0.05 (OR not mentioned)	unclear how many patients with
	 Cyclophosphamide only: 12 	- FSH <10 IU vs. ≥10 IU:	sperm samples were treated
	(11%)	p>0.05 (OR not mentioned)	with CRT and/or testicular
	 Cyclophosphamide + TNI: 5 (5%) 		irradiation.
		Predictors for active sperm production in	
		bivariate logistic regression analysis:	2 out of 106 fathered a child.
		 No leukaemia diagnosis vs. leukaemia 	
		diagnosis:	2. Risk of bias
		OR 17.0; 95% CI 2.6-113.0 (p<0.003)	A. Selection bias
		- Testicular volume ≥15 ml vs. <15 ml:	Low risk
		OR 14.2; 95% CI 2.1-98.1 (p<0.007)	Reason: 106/123 (86.2%)
		- No TBI vs. TBI:	eligible survivors participated in
		OR 30.0; 95% CI 2.8-322.1 (p<0.005)	this study.
		- FSH <10 IU/L vs. ≥10 IU/L:	
		OR 0.8; 95% Cl 0.7-1.0 (p<0.047)	B. Attrition bias
			High risk
			Reason: 31/106 (29.2%) semen
			analysis.
			C. Detection bias
			Unclear

		Reason: unclear if the outcome assessors were blinded for important determinants related to the outcome.
		<u>D. Confounding</u> High risk Reason: Although multivariable analyses were performed, treatment with alkylating agents was not included in the models.

Abbreviations: ALL, acute lymphoblastic leukaemia; AML, acute myeloid leukaemia; CRT, cranial radiotherapy; FSH, follicle stimulating hormone; fTBI, fractionated TBI; HSCT, haematopoietic stem cell transplantation; OR, odds ratio; SAA, severe aplastic anaemia; sTBI, single fraction TBI; TBI, total body irradiation; TNI, total nodal irradiation; 95% CI, 95% confidence interval.

What male reproductive preservation methods should be offered?						
Chan et al. Testicular Sperm Extraction Combined with Intracytoplasmic Sperm Injection in the Treatment of Men with Persistent Azoospermia						
Postchemotherapy.	Postchemotherapy. Cancer 2001;92:1632-37					
Study design Treatment era Years of follow-up	Participants	Intervention	Main outcomes	Additional remarks		
1. Study design	1. Type and Number of	1. Fertility Preservation	1. Outcome definitions	<u>1. Strengths</u>		
Retrospective	Participants	<u>method</u>	- Sperm retrieval rate: percentage of	This is one of two papers		
study			patients that underwent testicular biopsy	published that describes TESE-ICSI		
	17 male patients previously	17 patients produced	and had live sperm obtained	outcomes in patients that		
2. Treatment era	treated with CT	semen sample		received chemotherapy and are		
NM			- Fertilization rate: percentage of	azoospermic		
Fertility	Original cohort: 198 patients	Method of semen	embryos obtained by ICSI with two			
preservation	who underwent TESE-ICSI	collection	distinct pronuclei and two polar bodies	2. Limitations		
method: June 1995	for non-obstructive			- Very small number of patients		
to August 2000	azoospermia	Testicular sperm	- Clinical pregnancy rate: percentage of	- No controls		
		extraction (TESE)	pregnancies with the presence of fetal	- Retrospective		
	2. Diagnoses	(After 1997,	heartbeats determined by transvaginal	- Two different biopsy techniques		
3. Follow-up:	16/17 (94%) Malignant	microdissection TESE.	ultrasonography approximately 32 days	used		
	diseases:		after embryo transfer	- Varying chemotherapy/XRT		

Mean 16.3 years	6 (35%) HL	Prior to 1997, random		- One non-cancer patient
after CT	4 (24%) NSGCT	biopsy)	- Live birth rate: not defined	
completion	2 (12%) NHL			<u>3. Risk of bias</u>
	2 (12%) Leukaemia	Also combined with	- Complications intervention	
	1 (6%) Wilm's	intracytoplasmic sperm		A. Selection bias
	1 (6%) Mediastinal GCT	injection (ICSI)	2. Results	Unclear
	Other:		Sperm retrieval rate	Reason: 17/198 (8.6%) patients
	1 (6%) Nephrotic syndrome	9 patients who	9/20 (45%) patients (mean 1.2 attempts	included (inclusion those patients
		underwent TESE-ICSI had	per patient)	that received CT), unclear reasons
	3. Age at diagnosis	sperm retrieval		for exclusion
	NM		Clinical pregnancy rate	
	Age at 37.4 yrs at study	Timing of intervention	3/9 (33%) patients	B. Attrition bias
		After CT		Low risk
	4. Age at follow-up		Live birth rate	Reason: Outcomes assessed in all
	Mean 37.4 years (range 28-		2/9 (22%) patients who had TESE-ICSI	patients
	54)		fathered 3 live births	
				C. Detection bias
	5. Controls (if applicable)		1/9 (11.1%) pregnancies who had TESE-	Unclear
	No controls included		ICSI did not result in live birth	unclear if outcome assessors
				were blinded for important
	6. Additional study		Complication of intervention (TESE)	determinants related to the
	characteristics, if relevant		All patients discharged home on same day	outcome
	All pts azoospermic based		No postoperative complication	
	on 2 semen analyses			D. Confounding
			No correlation noted between TESE-ICSI	High risk / not applicable
	7. Chemotherapy		outcome and the underlying conditions	Reason: Although the authors
	- Varying CT regimens		treated with CT	noted that no correlation was
	- Total cumulative dose of			detected with the chemotherapy
	gonadotoxic agents not			received, the sample size was too
	provided			small for this determination to be
				significant. No multivariate
	8. Radiotherapy			analysis performed.
	4 patients received XRT in			
	addition to CT			
	9. Surgery			

17/17 tes sperm ex	ticular biopsy for traction	
<u>10. Other</u> -	treatments	

Abbreviations: NM: not mentioned; CT: chemotherapy; NSGCT: Mediastinal GCT; NHL: Non-Hodgkin lymphoma; HL: Hodgkin Lymphoma; XRT:chest radiation; ICSI: intracytoplasmic sperm injection; TESE: testicular sperm extraction; pts: patients ; XRT: chest radiation.

Study design Treatment era Years of follow-upParticipantsInterventionMain outcomesAdditional remarks1. Study design Retrospective study1. Type and Number of Participants1. Fertility Preservation method1. Outcome definitions - Tissue dissection in pubertal patients - Complications1. Strengths2. Treatment era TTC between 2015 20172. Diagnoses Solid tumour (74%),1. Fertility Preservation method 23 (100%) TTC (unilateral wedge biopsy)1. Outcome definitions - Tissue dissection in pubertal patients - Complications1. Strengths - Stelle2. Treatment era Solid tumour (74%),Timing of intervention - 5 (21.7%) received one or two rounds of their2. Results Tissue dissection in pubertal patients before cryopreservation3. Risk of bias A. Selection bias Unclear Reason: unclear if all patients that	What male reproductive preservation methods should be offered? Corkum et al. Testicular wedge biopsy for fertility preservation in children at significant risk for azoospermia after gonadotoxic therapy. J Ped Surgery 2019;54:1901-1905				
1. Study design Retrospective1. Type and Number of Participants1. Fertility Preservation method1. Outcome definitions 	Study design Treatment era Years of follow-up	Participants	Intervention	Main outcomes	Additional remarks
Retrospective studyParticipants 23 males who will be treated with high risk gonadotoxic therapymethod 23 (100%) TTC (unilateral wedge biopsy)- Tissue dissection in pubertal patients 	<u>1. Study design</u>	<u>1. Type and Number of</u>	1. Fertility Preservation	1. Outcome definitions	<u>1. Strengths</u>
study23 males who will be treated with high risk gonadotoxic therapy23 (100%) TTC (unilateral wedge biopsy)- Complications2. Limitations2. Treatment era TTC between 2015- 2017gonadotoxic therapy- Timing of intervention - 5 (21.7%) received one or two rounds of their- Complications3. Risk of bias A. Selection bias Unclear Before cryopreservation2. 5 ll- 5 (21.7%) received one or two rounds of their- 5 (21.7%) received one or two rounds of their- 22 (06%) had normal torticular tiscup with- A. Selection bias Duclear Reason: unclear if all patients that	Retrospective	<u>Participants</u>	<u>method</u>	- Tissue dissection in pubertal patients	
2. Treatment era gonadotoxic therapytreated with high risk gonadotoxic therapywedge biopsy)3. Risk of bias 3. Risk of bias2. Treatment era 20172. Diagnoses Solid tumour (74%),7. Timing of intervention - 5 (21.7%) received one or two rounds of their2. Results Tissue dissection in pubertal patients before cryopreservation3. Risk of bias A. Selection bias Unclear Reason: unclear if all patients that	study	23 males who will be	23 (100%) TTC (unilateral	- Complications	2. Limitations
2. Treatment era gonadotoxic therapy Timing of intervention 3. Risk of bias TTC between 2015- Timing of intervention 2. Results A. Selection bias 2017 2. Diagnoses - 5 (21.7%) received one Tissue dissection in pubertal patients Unclear Solid tumour (74%), or two rounds of their - 22 (06%) had normal torticular tiscue with Reason: unclear if all patients that		treated with high risk	wedge biopsy)		
TTC between 2015- 2017Z. Diagnoses Solid tumour (74%),Timing of intervention - 5 (21.7%) received one or two rounds of theirZ. Results Tissue dissection in pubertal patients before cryopreservationA. Selection bias Unclear Reason: unclear if all patients that2. 5	2. Treatment era	gonadotoxic therapy			<u>3. Risk of bias</u>
20172. Diagnoses Solid tumour (74%),- 5 (21.7%) received one or two rounds of theirTissue dissection in pubertal patients before cryopreservationUnclear Reason: unclear if all patients that2. 5. II- 5 (21.7%) received one or two rounds of their71000000000000000000000000000000000000	TTC between 2015-		Timing of intervention	2. Results	A. Selection bias
Solid tumour (74%), or two rounds of their <i>before cryopreservation</i> Reason: unclear if all patients that	2017	2. Diagnoses	- 5 (21.7%) received one	Tissue dissection in pubertal patients	Unclear
$2 \in \mathbb{R}$		Solid tumour (74%),	or two rounds of their	before cryopreservation	Reason: unclear if all patients that
<u>3. Follow-up:</u> nematological malignancy planned chemotherapy 22 (90%) had formal testicular tissue with underwent TTC were included in	3. Follow-up:	hematological malignancy	planned chemotherapy	22 (96%) had normal testicular tissue with	underwent TTC were included in
Median 1.4 years (17%), benign hematological prior to TTC the presence of germ cells on the study group	Median 1.4 years	(17%), benign hematological	prior to TTC	the presence of germ cells on	the study group
(interquartile disease (9%) - 6 (26%) underwent TTC histopathological analysis	(interquartile	disease (9%)	- 6 (26%) underwent TTC	histopathological analysis	
range 0.9-2.2 at the time of disease <u>Complications of intervention</u> <u>B. Attrition bias</u>	range 0.9-2.2		at the time of disease	Complications of intervention	B. Attrition bias
years) since TTC 3. Age at diagnosis relapse Low risk	years) since TTC	<u>3. Age at diagnosis</u>	relapse	O(0%) introportive complications	Low risk
Median 10 (range 0.42-18)		Median 10 (range 0.42-18)		- 0 (0%) initiative complications	Reason: outcomes assessed for all
years occurred 23 patients		years		occurred	23 patients
- 1 (4.3%) developed a scrotal cellulitis		4 Ago at follow up		- 1 (4.3%) developed a scrotal cellulitis	C Detection bias
three weeks after TTC after initiation of				three weeks after TTC after initiation of	
chemo- therapy; the superficial wound Reason: unclear if the outcome				chemo- therapy; the superficial wound	Reason: unclear if the outcome
5. Controls (if applicable) infection was successfully treated with		5 Controls (if applicable)		infection was successfully treated with	assessors were blinded for
No intravenous antibiotics		No		intravenous antibiotics	

	- Median time from TTC to start of	important determinants related
6. Additional study	cancer therapy: 7 days with no	to the outcome
characteristics, if relevant	unanticipated delays in treatment	
Tanner stage 1: 18 (78%)	initiation	D. Confounding bias
Tanner stage 2: 3 (13%)		NA
Tanner stage ≥3: 2 (9%)		Reason: Only descriptive results,
		no analyses performed
7. Chemotherapy		
NM		
8. Radiotherapy		
NM		
<u>9. Surgery</u>		
NM		
<u>10. Other treatments</u>		
9 (39.1%) HSCT		

Abbreviations: NA, not applicable; NM, not mentioned; TTC: testicular tissue cryopreservation.

What male reproductive preservation methods should be offered?

Ming et al. Cryopreservation of testicular tissue in pre-pubertal and adolescent boys at risk for infertility: A low risk procedure. J Ped Urol 2018;14:274e1-e5.

Study design Treatment era Years of follow-up	Participants	Intervention	Main outcomes	Additional remarks
1. Study design	<u>1. Type and Number of</u>	1. Fertility Preservation	1. Outcome definitions	<u>1. Strengths</u>
Retrospective	Participants	<u>method</u>	Complications	
study	34 males who will be	34 (100%) TTC (unilateral		2. Limitations
	treated with high risk	open biopsy)	<u>2. Results</u>	
2. Treatment era	gonadotoxic therapy		Complications of intervention	3. Risk of bias
TTC between 2014-		Timing of intervention	- 2 (5.9%) developed complications after	A. Selection bias
2016	2. Diagnoses	- 15 (44.1%) received	biopsy: ipsilateral epididymo-orchitis	Low risk
	Solid tumour (44%),	chemotherapy prior to	(resolved with antibiotics) and an	
3. Follow-up:	leukaemia or lymphoma	ттс		

NM	 (35%), hematological disorders (21%) <u>3. Age at diagnosis</u> Mean 6.9 ± 4.4 years (range 0.7-15 years) <u>4. Age at follow-up</u> NA <u>5. Controls (if applicable)</u> No <u>6. Additional study</u> <u>characteristics, if relevant</u> Pre-pubertal: 32 (94%) Post-pubertal: 2 (6%) <u>7. Chemotherapy</u> NM <u>8. Radiotherapy</u> NM <u>9. Surgery</u> NM <u>10. Other treatments</u> 	- 1 (2.9%) medulloblastoma patient received prior cranial radiotherapy	 ipsilateral torsed appendix testis (managed conservatively) Both patients were preparing for stem cell transplant and there was no delay to transplant as a result of these complications 0 (0%) had bleeding complications nor return visits to the operating room 	Reason: all 34 consecutive patients were included in the study. <u>B. Attrition bias</u> Low risk Reason: outcomes assessed for all 34 patients <u>C. Detection bias</u> Unclear Reason: unclear if the outcome assessors were blinded for important determinants related to the outcome <u>D. Confounding bias</u> NA Reason: Only descriptive results, no analyses performed
	22 (64.7%) HSCT			

Abbreviations: NA, not applicable; NM, not mentioned; TTC: testicular tissue cryopreservation.

What male reproductive preservation methods should be offered?

Stukenborg et al. Spermatogonial quantity in human prepubertal testicular tissue collected for fertility preservation prior to potentially sterilizing therapy Human Reprod 2018;33:1677-1683

Study design Treatment era Years of follow-up	Participants	Intervention	Main outcomes	Additional remarks
<u>1. Study design</u>	1. Type and Number of	1. Fertility Preservation	1. Outcome definitions	<u>1. Strengths</u>
Retrospective	Participants	<u>method</u>	Tissue dissection in pre-pubertal patients	
study	32 pre-pubertal males who	32 (100%) TTC (unilateral		2. Limitations
	will be treated with very	open biopsy; <20% of	2. Results	Detailed information regarding
2. Treatment era	high risk gonadotoxic	testicular volume of one	Tissue dissection in pre-pubertal patients	previous medical treatments and
TTC between 2014-	therapy (HSCT or testicular	testes sampled)	before cryopreservation	testicular volumes of patients
2017	radiotherapy)		Spermatogonia per transverse tubular	included in the biobank were not
		Timing of intervention	cross-section:	available.
3. Follow-up:	2. Diagnoses	20 (62.5%) testicular	 Mean 1.7 ± 1.0 in patients treated with 	
NM	Malignant hematological	biopsy performed 1-45	non-alkylating agents (no significant	3. Risk of bias
	disease (40.6%), solid	days after a previous	difference compared to controls)	A. Selection bias
	tumour (15.6%), benign	dose of chemotherapy	- Mean 0.2 ± 0.3 in patients treated with	Unclear
	hematological disorders		alkylating agents (p<0.05 compared to	Reason: unclear if all patients that
	(43.8%)		controls and non-alkylating agent	underwent TTC were included in
			group)	the study group
	3. Age at diagnosis		- Mean 0.8 ± 0.9 in patients treated	
	Range 0.7-13.1 years		without chemotherapy (p<0.05	B. Attrition bias
			compared to controls)	Low risk
	4. Age at follow-up		- Mean 4.1 ± 4.6 in controls	Reason: outcomes assessed for all
	NA		- Among 5 boys exposed to CED ≥4000	32 patients
			mg/m ² spermatogonia values were	
	5. Controls (if applicable)		close to zero	C. Detection bias
	14 testicular samples			Unclear
	without testicular pathology			Reason: unclear if the outcome
	from the biobank of the			assessors were blinded for
	Department of Pathology,			important determinants related
	Karolinska University			to the outcome
	Hospital served as controls;			
	Mean age: 5.6 ± 5.0 years			D. Confounding bias
				High

<u>6. Additional stu</u>	dy	Reason: Analyses were not
characteristics, i	f relevant	adjusted for potential
		confounding factors
7. Chemotherap	Y	
Alkylating agent	s: 6 (18.8%)	
Non-alkylating a	gents: 8	
(25.%)		
8. Radiotherapy		
NM		
<u>9. Surgery</u>		
NM		
<u>10. Other treatm</u>	<u>nents</u>	
NM		

Abbreviations: NA, not applicable; NM, not mentioned; TTC: testicular tissue cryopreservation.

What male reproductive preservation methods should be offered?

Uijldert et al. Development of the testis in pre-pubertal boys with cancer after biopsy for fertility preservation. Human Reprod 2017;32:2366-2372

Study design Treatment era Years of follow-up	Participants	Intervention	Main outcomes	Additional remarks
<u>1. Study design</u>	<u>1. Type and Number of</u>	1. Fertility Preservation	1. Outcome definitions	<u>1. Strengths</u>
Prospective study	Participants	<u>method</u>	 Tissue dissection in pre-pubertal 	
	64 pre-pubertal boys who	64 (100%) TTC (unilateral	patients before cryopreservation	2. Limitations
2. Treatment era	will be treated with	biopsy never exceeding	- Testicular growth	
TTC between 2011-	gonadotoxic therapy	50% of the testicular	- Complications	<u>3. Risk of bias</u>
2017	without the ability to	volume)		A. Selection bias
	ejaculate		<u>2. Results</u>	Low risk
3. Follow-up:		Timing of intervention	Tissue dissection in pre-pubertal patients	Reason: 64 out 78 (82.1%) eligible
Range 0.08-1 year	78 boys underwent TTC, of	All patients underwent	before cryopreservation	patients were included in the
	whom 6 deceased during	TTC prior to cancer	- No spermatogonia: 1 (1.9%)	study.
	follow-up, 5 lost to follow-	treatment	- Spermatogonia only: 44 (68.8%)	
	up, 2 orchiectomy of		- Up to spermatocytes: 9 (14.1%)	B. Attrition bias
	biopsied testes and 1		- Up to spermatids: 10 (14.1%)	Low risk
	without a volume recorded			Reason: outcomes assessed for all
			Testicular growth	64 patients. Regarding testicular
	2. Diagnoses		- After an initial decrease in testis	growth, 64, 58 and 55 patients
	Solid tumour (87.5%),		volume in both the biopsied and non-	underwent the 1 month, 6 month
	haematological cancer		biopsied testis during the first 6 months	and 12 month ultrasound,
	(7.8%) <i>,</i> other (4.7%)		after surgery, an increase in mean	respectively.
			volume was observed between 6 and	
	3. Age at diagnosis		12 months after surgery in both the	C. Detection bias
	Mean 8.3 (range 0.5-15.5		biopsied and non-biopsied testis	Unclear
	years)		- Overall, biopsy had no significant	Reason: unclear if the outcome
			impact on testicular growth (p=0.519)	assessors were blinded for
	4. Age at follow-up		- No significant differences between	important determinants related
	NA		piopsied and non-piopsied testis when	to the outcome.
			looking at subgroups defined by the	
	5. Controls (if applicable)		stage of testicular development	D. Contounding bias
	NO		(spermatogonia only, development up	LOW FISK

-		
6. Additional study	until the spermatocyte stage or up until	Reason: Biopsied testes compared
characteristics, if relevant	the spermatid stage)	to non-biopsied testes.
NM		
	Acute complications of intervention	
7. Chemotherapy	 1/78 (1.3%) post-operative bleeding 	
NM	- 2/78 (2.6%) wound infection one of	
	which had a minor infection where no	
8. Radiotherapy	additional action had to be taken: the	
NM	other boy was treated with antibiotics.	
	complaints resolved within a few days	
9 Surgery	without visible testicular damage: no	
NM	second operation or orchiectomy was	
	nocossary in oither case	
10 Other treatments	necessary in entier case	
10. Other treatments	Illtracopographic apportagities at 1	
	Ultrusonographic abnormalities at 1	
	(n=64)	
	- Calcifications: 2 (3.1%) vs. 2 (3.1%)	
	- Epididymal cyst: 3 (4.7%) vs. 1 (1.6%)	
	- Hydrocele: 4 (6.3%) vs. 1 (1.9%)	
	- Extra-testicular haematoma: 5 (7 8%)	
	vs 0	
	- Intratesticular haematoma [,] 2 (3 1%) vs	
	0	
	- Fibratic lesion: 0 vs. 0	
	Illtrasonoaraphic abnormalities at 6	
	months in highsight vs. contralateral testis	
	(n=58)	
	- Calcifications: 2 (3 1%) vs. 2 (3 1%)	
	$= \text{Endidymal cyst: } 2 (3.4\%) \vee 3.2 (3.4\%)$	
	= EpicicyIIIal Cyst. 2 (3.4/0) vs. 1 (1.7/0) = Hydrocele: 2 (3.4%) vs. 0	
	Extra tosticular baomatoma: Ove O	
	- EXITA-LESTICULAR INDERNATIONAL OVS. U	
	- intratesticular naematoma: 0 VS. 0	
	- FIDROUC IESION: U VS. U	

	Ultrasonographic abnormalities at 12 months in biopsied vs. contralateral testis	
	(n=55)	
	- Calcifications: 1 (1.6%) vs. 1 (1.6%)	
	- Epididymal cyst: 0 vs. 1 (1.6%)	
	- Hydrocele: 1 (1.6%) vs. 2 (3.1%)	
	- Extra-testicular haematoma: 0 vs. 0	
	- Intratesticular haematoma: 0 vs. 0	
	- Fibrotic lesion: 4 (6.3%) vs. 0	

Abbreviations: NA, not applicable; NM, not mentioned; TTC: testicular tissue cryopreservation.

What male reproductive preservation methods should be offered?

Hagenäs et al. Clinical and biochemical correlates of successful semen collection for cryopreservation from 12 - 18-year -old patients: a single-center study of 86 adolescents. Human Reproduction 2010; 25(8):2031-2038

Study design Treatment era Years of follow-up	Participants	Intervention	Main outcomes	Additional remarks
<u>1. Study design</u>	<u>1. Type and Number of</u>	1. Fertility Preservation	1. Outcome definitions	<u>1. Strengths</u>
Single-centre	Participants	<u>method</u>	Successful semen sampling via	Study based on standard clinical
retrospective	80 male patients with		masturbation	parameters (available in every-
cohort	malignant disease (various)	86 patients produced	Successful semen sampling via electro-	day practice)
		semen sampling for	ejaculation	
	<i>Total cohort</i> : 86 male	cryopreservation	Sperm concentration	2. Limitations
2. Treatment era	patients		Motile spermatozoa	- Retrospective cohort
December 1995 -			Semen volume	- Small cohort
May 2009	2. Diagnoses	Method of semen	Determinants of successful semen	- No long-term follow-up data
	80/86 (93%) Malignant	collection	collection	
	diagnosis:	- 74 (86%) patients		3. Risk of bias
3. Follow-up:	19 (22%) HL	masturbation		A. Selection bias
No long term	16 (20%) Testicular cancer	- 11 (13%) patients	<u>2. Results</u>	unclear
follow-up	25 (31%) Leukaemia /NHL	electroejaculation	Semen analysis before cryopreservation	Reason: unclear how the 86
	20 (23%) Solid tumours	- 1 (1.2%) Penile	by masturbation	patients were selected
		vibration	65/74 (87.8%) patients with successful	
	6/80 (7%) Other:		sample collected and cryopreserved	B. Attrition bias
	2 aplastic anemia		6/74 (8.1%) patients with azoospermia	Low risk

1 disseminated sclerosis	Timing of intervention	3/74 (4%) natients with immotile sperm	Reason: outcomes assessed in all
	Cryopreservation before	S/ + (+) patients with innotic sperm	natients
1 varicocolo	trootmont	Saman analysis before envolves an ation	Partents
1 Warcoccie	lieathent	by electro eleculation	C. Detection bias
1 Wegeners granulomatosis	Timing of succession statis		Unclear
	Timing of semen analysis	6/12 (50%)patients with successful	unclear if outcome assessors
<u>3. Age at diagnosis</u>	Before cryopreservation	sample collected and cryopreserved	were blinded for important
Median 16.2 years (12.2 -		4/12 (33%) patients with azoospermia	determinants related to the
17.9)		2/12 (16.75) patients with immotile	
		sperm	outcome
<u>4. Age at follow-up</u>		Successful semen sampling(total)	D. Confounding
NA		71/86 (83%) patients with successful	D. Confounding
		sample collected and cryopreserved	High risk
5. Controls (if applicable)			Reason: associations performed
NA		10/86 (11.6%) patients with azoospermia	with Pearson's correlation but not
		5/86 (5.8%) patients with immotile sperm	with multivariate analysis
6. Additional study			
characteristics if relevant		Sperm concentration	
- FSH H inhibin B		- Median: 9.6 (range 0-284) million/ml	
testosterone		Median. 5.0 (range o 204) minion/mi	
testosterone		22/96(28.4%) nationts had 20	
7. Chomatharran		- 35/80 (38.4%) patients had 220	
<u>7. Chemotherapy</u>		million/mi sperm concentration	
intervention prior to CI		- 43/86 (50%) patients had < 20	
		million/ml sperm concentration	
8. Radiotherapy		- 10/86 (11.6%) patients had azoospermia	
NA			
		Motile spermatozoa	
<u>9. Surgery</u>		Median: 45.5% (range 0-86%)	
NA			
		71/86 (83%) patients had motile	
<u>10. Other treatments</u>		spermatozoa; 5/86 (5.8%) patients with	
NA		few immotile sperm	
		Semen volume (median)	
		HL 1.7 ml (0.1-5.9)	
		Testicular cancer 3.0ml (0.5–6.9)	
		Leukaemia /NHL 1.8 ml (0.1–6.2)	
		Solid tumours 1.5 ml (0.03–6.1)	

	<i>Total sperm count (millions)</i> HL 7 (0–243) Testicular cancer 8.7 (0.03–216) Leukaemia /NHL 46.8 (0.04–611) Solid tumours 34.1 (0.15–210)	
	Determinants of successful semen collection - Testicular volume correlated with sperm concentration (R=0.283, p=0.046), and percentage of motile spermatozoa (R= 0.410, p=0.003)	
	- Chronological age (but not reproductive hormones) correlated with sperm concentration (R=0.25, P=0.049)	

Abbreviations: NA: not applicable; NM: not mentioned; HL: Hodgkin Lymphoma; HL: Hodgkin Lymphoma; ALL: acute lymphoblastic leukaemia

What male reproductive preservation methods should be offered?

Hovav et al. Electroejaculation before chemotherapy in adolescents and young men with cancer. Fertility and Sterility 2001; 75(4): 811-13.

Study design Treatment era Years of follow-up	Participants	Intervention	Main outcomes	Additional remarks
1. Study design	<u>1. Type and Number of</u>	1. Fertility Preservation	1. Outcome definitions	<u>1. Strengths</u>
Retrospective	Participants	<u>method</u>	Successful sperm retrieval as determined	study highlights the utility of
clinical study	6 male patients with cancer		by semen analysis	electroejaculation for sperm
	diagnoses prior to		Complication of intervention	retrieval prior to chemotherapy
	chemotherapy	6 patients produced		
2. Treatment era		semen sample	<u>2. Results</u>	2. Limitations
1998-1999	2. Diagnoses		Semen analysis before cryopreservation:	- Small study
	PT1: Ewing sarcoma	Method of sample	Sperm count	- Effect of electrostimulation on
	PT2: Osteosarcoma	collection	Mean 16 x 10 ⁶ (range 0-45 x 10 ⁶)	long-term sperm viability and
3. Follow-up:	PT3: Osteogenic sarcoma			quality unknown
			Sperm motility	- No long-term follow-up data

	1		1	
No long-term	PT4: Testicular germ-cell	Electroejaculation under	Mean 14% (range 0-53%)	
tollow-up	tumour	general anesthesia to		<u>3. Risk of bias</u>
	PT5: Hodgkin lymphoma	produce semen:	Sperm count, sperm motility	
	PT6: Testicular germ-cell		PT1: 15 x 10 ⁶ ; 6%	<u>A. Selection bias</u>
	tumour	 Antegrade semen 	PT2: 24 x 10 ⁶ ; 53%	Unclear
		collected directly	PT3: 9 x 10 ⁶ ; 0%	Reason: unclear how the 6
	3. Age at diagnosis		PT4: 35 x 10 ⁶ ; 33%	patients were selected
	Mean 18 years ±3	- Retrograde semen	PT5: 45 x 10 ⁶ ; 10%	
	Range 15-22 years	collected in Ham-F10	PT6: 6.5 x 10 ⁶ ; 20%	B. Attrition bias
		medium which was		Low risk
	4. Age at follow-up	extracted from the	Complication of intervention	Reason: outcomes assessed in all
	No long-term follow-up	bladder, centrifuged, and	0/6 patients with complications	patients
		concentrated to 1-2mL	-,	
	5. Controls (if applicable)		Electroeiaculation can be performed 2-3	C. Detection bias
	none		times every 48 hours without	Unclear
		Timing of intervention	complications	Reason: unclear if outcome
	6 Additional study	Before anticancer		assessors were blinded for
	characteristics if relevant	therapy		important determinants related
	nationts failed	therapy		to the outcome
	- patients failed	Timing of comon analysis		
	vibratory stimulation to	Poforo cruoprocoruction		D. Confounding
		Before cryopreservation		High risk
	produce semen			Reason: descriptive study, no
	- 4 patients underwent			multivariate analysis performed
	more than one			, ,
	electroejaculation session			
	7. Chemotherapy			
	-			
	8. Radiotherapy			
	-			
	<u>9. Surgery</u>			
	See intervention			
	Before CT			

Abbreviations: NA: not applicable; NM: not mentioned; CT: chemotherapy; PT: patient

What male reproductive preservation methods should be offered?

Kamischke et al. Cryopreservation of Sperm From Adolescents and Adults With Malignancies. Journal of Andrology 2004; 25: 586–592

Study design Treatment era Years of follow-up	Participants	Intervention	Main outcomes	Additional remarks
<u>1. Study design</u>	<u>1. Type and Number of</u>	1. Fertility Preservation	1. Outcome definitions	<u>1. Strengths</u>
Single-center	Participants	<u>method</u>	Successful cryopreservation (defined as	 Despite reasonable exclusion
cohort study,			observation of at least a single motile	criteria 91% of possible
retrospective	300 male patients with	851 patients produced	sperm after the thawing procedure)	participants included
	malignant disease <25 years	semen sampling for	Semen analysis	- Large cohort of adolescents: 111
	of age	cryopreservation	Sperm count	(13% of total participants)
2. Treatment era			Total sperm motility	
April 1989 - March		Method of sample	Testes volume	2. Limitations
2003	Original cohort: 936 patients	collection	Live births	 No healthy controls
	with malignant disease.	Unclear (?masturbation)		- No follow-up
	Of these, 851 (91%)			
<u>3. Follow-up</u>	included:		2. Results	
NM	111 (13%) adolescents <20	Timing of intervention	Semen analysis before cryopreservation in	3. Risk of bias
	years; 740 (87%) adults <40	- Before initiation of	patients <20 years	
	years	anticancer treatment.	110/111 (99.1%) patients with successful	A. Selection bias
		 Except: 61% patients 	sample collected and cryopreserved	Low risk
	Exclusion criteria:	with testicular cancer	1/111 (0.9%) patient with azoospermia	Reason: 851/936 (91%) patients
	 Incomplete data, relapses, 	had unilateral ablation of		included, exclusion criteria
	secondary cancers, known	the testis before	Semen analysis after freezing and	reported
	bilateral testicular cancer,	cryopreservation	thawing in patients <25 years	
	unilateral testicular cancer		268/300 (89%) patients with at least a	B. Attrition bias
	in combination with		single motile sperm	Low risk
	contralateral intraepithelial	Timing of semen analysis	32/300 (10.7%) patients without motile	Reason: outcomes assessed for all
	neoplasia, or an unknown	Before cryopreservation	sperm	patients
	primary diagnosis			
			Total sperm motility (%)	C. Detection bias
	2. Diagnoses		<15 yrs: 38 ± 7	Unclear
	Testicular cancer 485 (57%)		15- <16 yrs: 48 ± 5	Reason: unclear if outcome
	28 (6%) adolescents		16- <17 yrs: 48 ± 4	assessors were blinded for
	457 (94%) adults		17- <18 yrs :57 ± 4	

Lymphomas 187 (22%)	18- <19 yrs: 56 ± 5	important determinants related
36 (19%) adolescents	19- <20 yrs: 48 ± 5	to the outcome
151 (81%) adults	20- <25 yrs: 49 ± 2	
		D. Confounding
Leukaemia's 68 (8%)	Sperm concentration (million/mL)	High risk
13 (19%) adolescents	<15 yrs: 38.9 ± 13.9	Reason: multivariate analysis not
55 (81%) adults	15- <16 yrs: 56.0 ± 24.4	performed
	16- <17 yrs: 46.6 ± 15.0	
Bone cancer 65 (8%)	17- <18 yrs : 49.9 ± 20.1	
20 (31%) adolescents	18- <19 yrs: 36.6 ± 11.4	
45 (69%) adults	19- <20 yrs: 34.7 ± 6.5	
	20- <25 yrs: 34.8 ± 3.7	
Other cancers 46 (5%)		
14 (30%) adolescents	Ejaculate volume (mL)	
32 (70%) adults	<15 yrs: 1.5 ± 0.3	
	15- <16 yrs:1.7 ± 0.3	
3. Age at diagnosis (years)	16- <17 yrs: 3.4 ± 0.5	
NM	17- <18 yrs: 2.4 ± 0.2	
<25 years	18- <19 yrs: 3.0 ± 0.3	
	19- <20 yrs: 3.0 ± 0.4	
111/300 (37%) patients <20	20- <25 yrs: 3.6 ± 0.1	
years at cryopreservation		
	Sperm count (million/ejaculate)	
<15: 11 (1.3%)	<15 yrs: 38 ± 10.2	
15- <16: 15 (1.8%)	15- <16 yrs:73 ± 23.6	
16- <17:19 (2.2%)	16- <17 yrs: 174.4 ± 58.7	
17- <18:21 (2.5%)	17- <18 yrs :113.2 ± 35.6	
18- <19: 19 (2.2%)	18- <19 yrs: 119.7 ± 38.4	
19- <20:26 (3%)	19- <20 yrs: 106.9 ± 20.5	
20- <25:189 (22.2%)	20- <25 yrs: 135 ± 18.9	
25- <30: 271 (31.8%)		
30- <35: 191 (22.4%)	- Testicular volume significantly	
35- <40: 89 (10.5%)	correlated with age ($r = 0.24$, $P = .0096$),	
	ejaculate volume ($r = 0.26$, $P = .0058$),	
Split into diagnose groups:	testosterone (P = .0048), and sperm count	
- Testicular cancer	(P = .0064)	
Adolescents 17.6 ± 0.3		

Adults 29.2 ± 0.2	 No significant correlation between 	
- Lymphomas	testicular volume and FSH	
Adolescents 17.5 ± 0.3		
Adults 27.5 ± 0.4	Pregnancy in patients <20 years	
- Leukaemias	1/1 (100%) patient who had IVF-ICSI	
Adolescents 17.5 ± 0.5	achieved pregnancy but resulted in early	
Adults 27.1 ± 0.7	abortion	
- Bone cancer		
Adolescents 17.0 ± 0.4	Pregnancy in adults >20 years	
Adults 27.0 ± 0.8	9 clinical pregnancies resulted in 11 live	
- Other cancers	births (including 3 sets of twins) and 1	
Adolescents 17.1 ± 0.4	abortion from 11 adult patients	
Adults 30.1 ± 0.9	·	
4. Age at follow-up		
NM		
5. Controls (if applicable)		
-		
6. Additional study		
characteristics, if relevant		
-		
7. Chemotherapy		
NM		
8. Radiotherapy		
NM		
9. Surgery		
NM		
10. Other treatments		
Not reported		

Abbreviations: NA: not applicable; NM: not mentioned; yrs: years; IVF/ICSI: in vitro fertilization/ intracytoplasmic sperm injection; ART: assisted reproduction

What male reproductive preservation methods should be offered?

Kliesch et al. Cryopreservation of Semen From Adolescent Patients With Malignancies. Med Pediatr Oncol. 1996;26:20-7

Study design Treatment era Years of follow-up	Participants	Intervention	Main outcomes	Additional remarks
<u>1. Study design</u>	<u>1. Type and Number of</u>	1. Fertility Preservation	1. Outcome definitions	<u>1. Strengths</u>
Retrospective	<u>Participants</u>	<u>method</u>	Testicular volumes	Adequate semen analysis.
cross- sectional	28/239 (11.7%) male		Semen analysis (Sperm concentration,	
study	patients with malignant	239 patients produced	total sperm number, motility and	2. Limitations
	diseases (various) aged < 20	semen sampling for	morphology of sperm prior to	 No long follow-up data on
	years	cryopreservation	cryopreservation)	semen quality after thawing and
2. Treatment era			Live births	pregnancy rate.
Unclear	Total cohort: 239 male	Method of sample		- No control group
	patients:	collection	2. Results	
	210 patients aged >20 years;	Unclear (?masturbation)	Semen analysis before cryopreservation	3. Risk of bias
3. Follow-up:	29 patients aged 14-20 years		28/29 (97%) patients with successful	A. Selection bias
NM		Timing of intervention	sample collected and cryopreserved	unclear
	2. Diagnoses	Before cancer treatment	1/29 (3%) patient did not produce	Reason: unclear how the 239
	Testicular cancer		ejaculate (15-year old osteosarcoma	patients were selected
	Hodgkin's disease	Timing of semen analysis	patient)	
	Leukaemia	Before cryopreservation		B. Attrition bias
	Other tumours		Sperm motility before vs after freezing	Low risk
	(osteosarcoma, melanoma,		and thawing	Reason: outcomes assessed for
	astrocytoma,		14-17 years: mean 30 ± 7 vs. mean 18 ± 6	all patients
	rhabdomyosarcoma of		18-20 years: mean 45 ± 5 vs. mean 22 ± 4	
	epididymis, Ewing's			<u>C. Detection bias</u>
	sarcoma)		Semen analysis before cryopreservation	unclear
	Other diseases (non-cancer)		Total sperm number	Reason: unclear if outcome
			Group 1: mean 157±94, median 46	assessors were blinded for
			Group 2: mean 127±33, median 72	important determinants related
	3. Age at diagnosis (years)			to the outcome
	NM		Total sperm number (mill/ejaculate)	
			Group 1: mean 157 ± 94, median 46	D. Contounding
				High risk

29/239 (12%) patients: 14-	Group 2: mean 127 ± 33, median 72	Reason: no multivariate analysis
20 years at study		performed
4. Age at cryopreservation	Sperm concentration (mill/mL)	
3 groups:	Group 1: mean 44 ± 21, median 13	
Group 1 age 12-17 (12	Group 2: mean 40 ± 8, median 31	
patients; median age = 15.9		
years)	Testes volume, right and left (mL)	
Group 2 age 18-20 (17	Group 1: mean 29.6 ± 2.2	
patients; median age = 19.5	Group 2: mean 32.6 ± 2.4	
years)		
Group 3 age > 20 (210	Live births	
patients; median age = 28.9	5/239(2%) patients' partners had 13	
years)	inseminations	
4. Age at follow-up	3/13 (23%) inseminations resulted in	
NM	pregnancies	
	2/13 (15%) inseminations in patients'	
5. Controls (if applicable)	partners produced live births (twins)	
-	1/13 (7.7%) inseminations in patients'	
	partners resulted in abortion	
6. Additional study		
characteristics, if relevant	(unclear if these patients were <20 years	
-	at study)	
7. Chemotherapy		
NM		
8. Radiotherapy		
NM		
9. Surgery		
NM		
10. Other treatments		
NM		

Abbreviations: NA: not applicable; NM: not mentioned

What male reproductive preservation methods should be offered?

Ho et al. A short report on current fertility preservation strategies for boys. Clin Endocrinol (Oxf). 2017;87:279-285

Study design Treatment era Years of follow-up	Participants	Intervention	Main outcomes	Additional remarks
<u>1. Study design</u>	<u>1. Type and Number of</u>	1. Fertility Preservation	1. Outcome definitions	<u>1. Strengths</u>
Retrospective	Participants	<u>method</u>	- Tissue dissection in pubertal patients	Study present new fertility
study	20/44(C0) male nationts	14 patients had testioular	- Complications of intervention	preservation method
2 Treatment ora	30/44(68%) male patients	44 patients had testicular		
<u>2. freatment era</u> 1987 - 2015	(various)	collected	2. Results	2 Limitations
1907 -2019	(vanous)	conceled	Tissue dissection in pubertal patients	2. Limitations
3. Follow-up:	Original cohort: 46 patients	Timing of intervention	before cryopreservation	3. Risk of bias
No long-term	(2 declined)	Prior to anticancer	3/11 (27.2%) azoospermic	A. Selection bias
follow-up		treatment	8/11 (72.7%) mature sperm found	Unclear
	2. Diagnoses		(testicular size >10ml and Tanner stage 3;	Reason: unclear how the selection
4 pts died	30/44(68%) malignant	Timing of semen analysis	all Cr haive)	for the 46 patients was done
40 pts still alive	diagnosis	Prior to anticancer	Complications of intervention	
	malignant diagnosis	treatment	1/44 (2.3%) patient suffered scrotal	B. Attrition bias
			wound dehiscence occurring 2 weeks	Low risk
	<u>3. Age at diagnosis</u>		after procedure (patient with aplastic	Reason: outcomes assessed for all
	NM		anaemia)	44 patients
	Range 0.3-16.8 years		0/44 patients had delay in treatment	
				C. Detection bias
	Prepubertal: 33/44(75%)			Unclear
	Pupertal: 11/44(25%)			Reason: unclear if the outcome
	Patients with tissue only			important determinants related
	stored (n=33, prepubertal)			to the outcome
	Age at study 0.3-11.3			
				D. Confounding bias

Patients with tissue only		High risk
store (n=3, pubertal)		Reason: univariate analysis (not
Age at study 12.7-16.8		multivariate analysis used) so this
		study did not control for
Pationts with tissue and		confounding factors
Patients with tissue and		
sperm stored (n=4,		
pubertal):		
Age at study 12.7-16.2		
Patients with sperm stored		
(n=4, pubertal):		
Age at study 13.0-15.9		
, ge dtotddy 10:0 10:5		
1 Age at follow-up		
5. Controls (if applicable)		
-		
6. Additional study		
characteristics, if relevant		
- Prior to 2013 only sperm		
cryopreservation was		
offered		
- From October 2013		
tosticular tissuo		
cryopreservation was		
offered to boys where a		
moderate		
to high risk to future fertility		
was anticipated (>30% risk)		
7. Chemotherapy		
7/44(15.9) pts received		
previous CT		
Rest CT naive		
Act of have		

8. Radiotherapy NM		
<u>9. Surgery</u> NM		
<u>10. Other treatments</u> NM		

Abbreviations: NM: not mentioned; CT: chemotherapy

What male reproductive preservation methods should be offered?

Adank et al. Electroejaculation as a method of fertility preservation in boys diagnosed with cancer: a single-center experience and review of the literature. Fertil Steril. 2014;102:199-205.e1

Study design Treatment era Years of follow-up	Participants	Intervention	Main outcomes	Additional remarks
1. Study design	1. Type and Number of	1. Fertility Preservation	1. Outcome definitions	1. Strengths
Retrospective	Participants	<u>method</u>	1. Successful semen cryopreservation	Focus on electroejacualation in
cohort study	114 boys diagnosed with	81 patients produced	(based on the first attempt, defined as	the context of patients who
	cancer had sperm	semen sampling for	any motile spermatozoa present)	attempt masturbation but fail to
2. Treatment era	cryopreservation offered:	cryopreservation		produce adequate sample
From January 1998			2. Semen analysis	
to March 2013	106 patients with	Method of sample		2. Limitations
semen	cryopreservation attempt by	collection		- Small cohort
cryopreservation	masturbation	78 patients via	2. Results	- No long-term follow-up data
was offered		masturbation	1. Successful semen cryopreservation via	
	11 patients had		masturbation	
3. Follow-up:	cryopreservation attempt by	3 patients via	78/106 (68%) patients with successful	3. Risk of bias
No applicable	electroejaculation:	electroejaculation (under	sample collected and cryopreserved	A. Selection bias
	8 patients were offered	general anesthesia)		unclear
Electroejaculation	electroejaculation		18/106 (16%) patients with immotile	Reason: unclear how many
was offered from	primarily; 3 patients were	Timing of intervention	spermatozoa or absent spermatozoa	patients were included in original
2003 (when	offered electroejaculation	Before start of cancer		cohort
		treatment	10/106 (9%) patients were not able to	
			produce an ejaculate	

masturbation was	secondarily (failed to			B. Attrition bias
not possible)	produce adequate semen by	Timing of semen analysis	Successful semen cryopreservation via	Low risk
	masturbation)	At time of	electroejaculation	Reason: outcomes in 106/114
		cryopreservation /	3/11(27%) patients with successful	(93%) patients
	2. Diagnoses	Before start of cancer	sample collected and cryopreserved	
	Various	treatment		
			2. Semen analysis in patients with	C. Detection bias
	In 11 patients who were		successful sample collected and	Unclear
	offered electroejaculation:		cryopreserved via electroejaculation	Reason: unclear if the outcome
	Leukaemia/NHL:4			assessors were blinded for
	HL: 5		Volume (x10 ⁶ mL): 0.4 (0.4-0.4)	important determinants related
	Sarcoma/PNET:1		Concentration (x10 ⁶ /mL): 2.0 (0.1-5.5)	to the outcome
	Diagnosis not reported in 1		Motility (%): 3.0 (2.0-4.0)	
	patient.		рН: 7.9	D. Confounding bias
				High risk
	3. Age at diagnosis		Semen analysis in patients without	Reason: Descriptive, no
	16.5 years (10.8-18.9)		successful sample collected and	multivariate analysis performed
			cryopreserved via electroejaculation	
	4. Age at follow-up			
	Not applicable		Volume (x10 ⁶ mL): 0.4 (0.02-3.0)	
			Concentration (x10 ⁶ /mL): 2.0 (0.1-14.5)	
	5. Controls (if applicable)		Motility (%): 0	
	Not applicable		pH: 7.0 (6.4-8.0)	
	6. Additional study			
	characteristics, if relevant			
	Tanner:			
	Genital development 4.5 (3-			
	5)			
	Pubic hair development 4.5			
	(3-5)			
	Testicular volume (mL):			
	Left 14.3 (8.9-20.0)			
	Right 13.5 (8.0-20.0)			
	7. Chemotherapy			

Not mentioned		
Patients with previous		
gonadotoxic therapy as well		
as patients diagnosed with a		
brain tumour were excluded		
8 Radiotherapy		
Not mentioned		
Not mentioned		
9. Surgery		
Not mentioned		
<u>10. Other treatments</u>		
Not mentioned		

What male reproductive preservation methods should be offered?

Müller et al. Cryopreservation of semen from pubertal boys with cancer. Med Pediatr Oncol. 2000;34:191-4

Study design Treatment era Years of follow-up	Participants	Intervention	Main outcomes	Additional remarks
1. Study design	1. Type and Number of	1. Fertility Preservation	1. Outcome definitions	<u>1. Strengths</u>
Cohort study	Participants	<u>method</u>	1. Successful semen cryopreservation	Focus on electroejaculation as
	21 patients delivered semen	21 patients produced	2. Semen analysis	well as masturbation
2. Treatment era	sample	semen sampling for		
January 1 1995 –		cryopreservation	2. Results	2. Limitations
July 31 1998	45 patients with cancer		1. Successful semen cryopreservation via	- Small cohort
	eligible for cryopreservation	Method of sample	masturbation	- No long-term follow-up data
3. Follow-up:	(4 patients were not able to	collection	17/19 (89.5%) patients with successful	
Not applicable	produce sperm; 20 patients	18 patients via	sample collected and cryopreserved	<u>3. Risk of bias</u>
	time did not allow attempt	masturbation		A. Selection bias
	of cryopreservation, or		Successful semen cryopreservation via	Unclear
	patient was not assessed to		electroejaculation	

be mature enough to deliver	2 patients via	2/2 (100%) patients with successful	Reason: unclear how the 45
semen sample or the	electroejaculation (under	sample collected and cryopreserved	patients were selected
procedure was not	general anesthesia)		
accepted)		2. Semen analysis in patients with	B. Attrition bias
	1 patient via vibration	successful sample collected and	High risk
<u>2. Diagnoses</u>		cryopreserved via masturbation	Reason: outcomes in 21/45 (47%)
ALL: 7 (2 with relapse)		Median percentage of motile sperm: 50%	patients
Non-Hodgkin lymphoma: 2	Timing of intervention	(range 9-86%)	
AML: 1	Before start of cancer		C. Detection bias
Hodgkin lymphoma: 6	treatment (2 patients	Semen analysis in patients with successful	Unclear
Osteosarcoma: 2	had received	sample collected and cryopreserved via	Reason: unclear if the outcome
Testicular cancer: 1	chemotherapy before)	electroejaculation	assessors were blinded for
CNS tumour: 1		Patient 1:	important determinants related
Wilms tumour: 1	Timing of semen analysis	Volume 0.8 mL	to the outcome
	Before start of cancer	Concentation 75 x 10 ⁶ /mL	
<u>3. Age at diagnosis</u>	treatment (2 patients	Motility 38%	D. Confounding bias
Mean 14.5 years (13-18)	had received		High risk
	chemotherapy before)	Patient 2:	Reason: Descriptive, no
<u>4. Age at follow-up</u>		Volume 3.2 mL	multivariate analysis performed
Not applicable		Concentration 4.0 x 10 ⁶ /mL	
		Motility 10%	
5. Controls (if applicable)			
Not applicable			
6. Additional study			
characteristics, if relevant			
2 patients suffered from			
relapse and had received			
before chemotherapy for			
standard-			
risk ALL (vincristine,			
daunorubicin, MTX, and			
prednisolone)			
7. Chemotherapy			
Not mentioned			

<u>8. Radiotherapy</u>		
Not mentioned		
9. Surgery		
Not mentioned		
10. Other treatments		
Not mentioned		