MALE INFERTILITY AND

SURGICAL SPERM RETRIEVAL / ICSI

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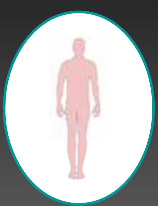
MALE INFERTILITY

- 80 millions infertile patients (WHO)
- 1/6 couples are infertile
- 1/10 in ART
- Up to 50% "male" factor
- 2-10% "infertile men"
- 10% infertile men with <1M/ml
- 10-15% infertile men with azoospermia (1% among all men)
- France : 30 000 ICSI / 1800 surgical sperm retrieval

De Kretzter DM, Lancet 1997

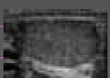
EVALUATION



Anamnesis
- Relevant medical history
- Lifestyle and environment

Physical examination
- BMI
- Testis
- Complete clinical examination

Ultrasonography



- Color flow Doppler ultrasonography
- Transrectal ultrasonography
- Scrotal ultrasonography
- Renal tract evaluation

Hormones
- ✓Serum total testosterone
- ✓FSH, LH, PRL
- ✓ Serum inhibin B, AMH

Sperm

- ✓≥ 2 ejaculates – 3 months
- ✓No sperm after centrifugation
- ✓Microbiological evaluation
- ✓Seminal biochemical markers

Genetics

- ✓Karyotype
- ✓Yq microdeletion
- ✓CFTR mutations
- ✓FISH on gametes

Surgical exploration
Diagnosis
MESA/TESE

EVALUATION

Anamnesis

- ❑Prior fertility
- ❑Childhood illnesses and disorders such as viral orchitis ; cryptorchidism
- ❑Genital trauma or pelvic or inguinal surgery
- ❑Infectious such as epididymitis or urethritis
- ❑Exposure to gonadotoxins such as radiation or chemotherapy, recent fevers or heat exposure or current or recent medications
- ❑Family history of birth defects, mental retardation, reproductive failure, or cystic fibrosis

EVALUATION

Physical examination

- ❑Presence of inguinal or scrotal scars
- ❑Testis size (normal volume > 19 mL) and consistency
- ❑Secondary sex characteristics including body habitus, hair distribution, and gynecomastia
- ❑Presence and consistency of the vasa deferentia
- ❑Consistency of the epididymes
- ❑Presence of varicoceles
- ❑Masses palpable on digital rectal examination

EVALUATION

Measurement of selected hormones

TABLE 2

Basal hormone levels in various clinical states.

Clinical condition	FSH	LH	Testosterone	Prolactin
Normal spermatogenesis	Normal	Normal	Normal	Normal
Hypogonadotropic hypogonadism	Low	Low	Low	Normal
Abnormal spermatogenesis*	High/Normal	Normal	Normal	Normal
Complete testicular failure/Hypergonadotropic hypogonadism	High	High	Normal/Low	Normal
Prolactin-secreting pituitary tumor	Normal/Low	Normal/Low	Low	High

* Many men with abnormal spermatogenesis have a normal serum FSH, but a marked elevation of serum FSH is clearly indicative of an abnormality in spermatogenesis.

ASRM Practice Committee. Optimal evaluation of the infertile male. Fertil Steril 2006.

EVALUATION

Azoospermia or cryptozoospermia ?

- ❑ Extended sperm preparation with careful examination of the pellet droplets ESP
- ❑ Surgical sperm retrieval SSR
- ❑ Two semen analyses performed at least 3 months apart

TABLE 4

Summary of findings of extended sperm preparation (ESP) in azoospermic men.

	Present study	Ron et al. 1997	Timm et al. 2005
Sample size, n	87	49	27
Spermatozoa seen, n (%)	19 (22%)	17 (35%)	10 (37%)
Spermatids seen, n (%)	N/A	N/A	11 (41%)

Note: N/A = not available.

Swanton. Extended sperm preparation in azoospermia. Fertil Steril 2007.

CAUSES….

❑ Pre-testicular causes (rare) : secondary testicular failure

- Endocrine abnormalities : hypogonadotropic hypogonadism

Treatable

❑ Testicular causes +++ : primary testicular failure

- External factors
- Infection
- Malformation
- Genetics
- Idiopathic male infertility….

❑ Post- testicular causes (40%) :

- Ejaculatory dysfunction
- Ductal obstructions

Correctable

de Kretser 1997; Irvine 1998; ASRM 2008

External factors

Microscopic level :

- ❑ **Concentration**
- ❑ **Motility**
- ❑ **Morphology**

Treatable

Molecular level :

- ❑ **Aneuploïdy**
- ❑ **Oxidative stress**
- ❑ **DNA fragmentation**

- ❑ **Drugs**
- ❑ **Endocrine disruptors**
- ❑ **Smoking**
- ❑ **Alcohol**
- ❑ **Recreational drugs**
- ❑ **Genital heat stress**
- ❑ **Psychological stress**
- ❑ **Cellular telephone use**
- ❑ **Weight and nutrition**
- ❑ **….**

GENETIC TESTING

		France	ESHRE	Italy	USA
Men	NOA	Karyotype Y microdeletion	Karyotype Y microdeletion	Karyotype Y microdeletion	Karyotype Y microdeletion
	Oligospermy	Karyotype Y microdeletion (<5M/ml)	Karyotype Y microdeletion (<5M/ml)	Karyotype Y microdeletion (<10M/ml)	Karyotype Y microdeletion (<5-10M/ml)
	CBAVD	CFTR mutations	CFTR mutations	CFTR mutations	CFTR mutations
Women	Pre -ICSI	Discussed	Karyotype if medical history	Karyotype	Discussed
	Men CBAVD	CFTR mutations	CFTR mutations	CFTR mutations	CFTR mutations

GENETIC TESTING

- **Among couples** in ICSI for male infertility : **17% with genetic male factor**

Meschede et al, 1996

- **100 azoospermic patients:**

29% : genetic anomaly *(karyotype, CFTR, Y microdeletion)*
22% : external factor or illness
27 % : cryptochidism
22 % : idiopathic

Fedder, 2004

- **Human spermatogenesis : > 4000 genes !**

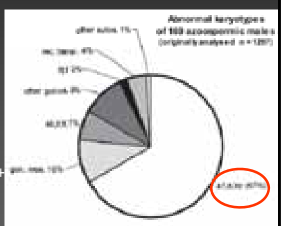
AZOOSPERMIA

13 % : chromosomal anomalies

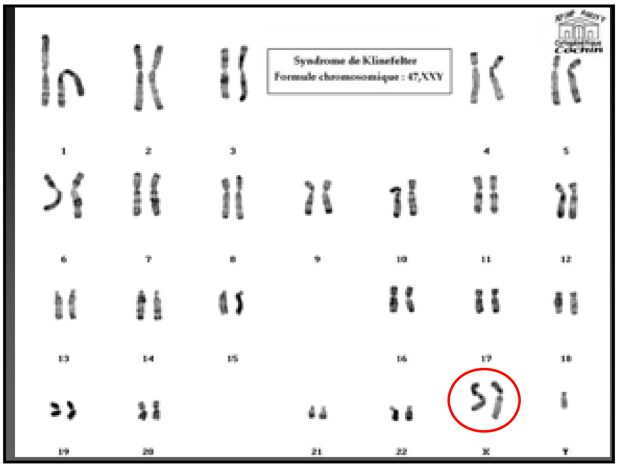
(21 X male newborn)

93% : sex chromosomal abnormalities

47,XXY Klinefelter syndrome KS 67% ++
and mosaic 10%
Structural defects X and Y
Men 46,XX 7%



Mau-Holzmann, 2005



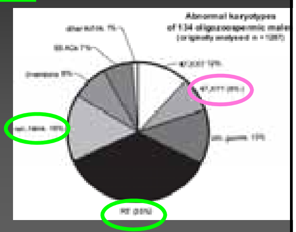
OLIGOZOOSPERMIA

4.6 % : chromosomal anomalies

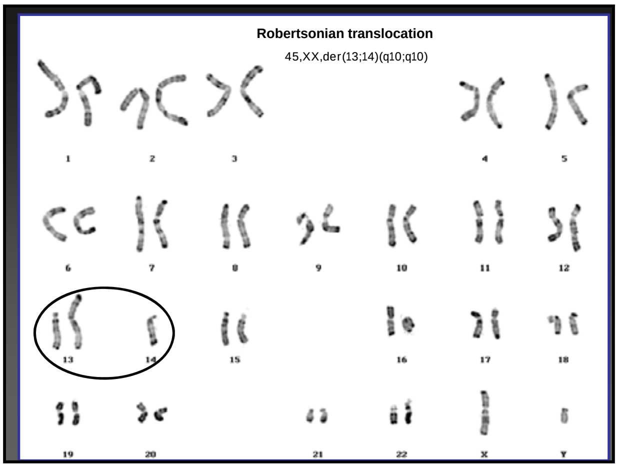
(7 X male newborn)

- 67% autosomal translocation and inversion
  - Robertsonian T 35%
  - Eciprocal autosomal T 16%

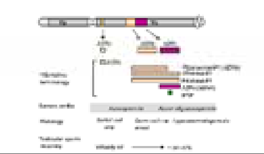
- 33% sex anomalies
  - KS 12%
  - 47, XYY 8%



Mau-Holzmann, 2005. Cytogenet Genome Research



Yq MICRODELETIONS

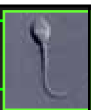


- ❑Non obstructive male infertility (*de novo*)
- ❑Azoospermia AND < 1 M SPZ /ml
- ❑FSH high or normal
- ❑ 10-15% azoospermia, 5-10% oligozoospermia

Krausz and Degl'Innocenti 2006

SFGH, ESHRE, ASRM, EAA...HAS

Yq MICRODELETIONS



❑Correlation genotype – sperm concentration

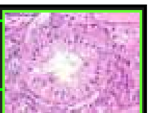
Table 2: Distribution of the 64 men according to the average sperm concentration and the site deletion.

Deletion site	Number of men	Sperm concentration (10^6/ml)			
		0	CA	0> and ≤1	1>
		n (%)	*n* (%)	*n* (%)	*n* (%)
AZFa	1	1	0	0	0
AZFb	7	7 (100%)	0	0	0
AZFc	46	22 (47.8%)	3 (6.6%)	19 (41.3%)	2 (4.3%)
AZFb + c	9	9 (100%)	0	0	0
AZFa +b + c	1	1 (100%)	0	0	0
TOTAL	**64**	**40 (62.5%)**	**3 (4.7%)**	**19 (29.7%)**	**2 (3.1%)**

CA : crypto azoospermia

Patrat et al., 2008

Yq MICRODELETIONS



❑Correlation genotype – testicular histology

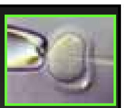
Table 3: Testicular histology and/or testicular sperm extraction according to location of Y deletion in 27 azoospermic patients

Y deletion	Number of patients	Testicular histology	Testicular spermatozoa recovered
AZFb	2	SCO	0
	3	MASc	0
	1	MASt	0
AZFc	9	SCO	0
	1	MASc	0
	5	HS	+
	1	NS	+
	2	NA	0
AZFb+c	2	SCO	0
AZFa+b+c	1	NA	0

HS: hypo-spermatogenesis; MASc: maturation arrest at the spermatocyte I stage; MASt: maturation arrest at the spermatid stage; SCO: Sertoli Cells Only; NS: normal spermatogenesis (see definition in material and methods); NA: not available

Patrat et al., 2008

❑ Genetic counseling



ICSI can be performed but risk of transmission of the Y deleted chromosome to the offspring

- **More nullisomic gametes for sex chromosomes**

  ***Turner syndromes**

- **Among 12 46,XY/45,X men : 3 AZFc deletions**

Y CHROMOSOME INSTABILITY

PGD Sex selection ?

Patsalis et al. Lancet 2002; Siffroi et al., 2000

❑ Future fertility

Possible decline in spermatogenesis over time in AZFc deleted men :

- Patient : sperm cryopreservation for future fertility!
- ICSI - conceived sons : follow-up and sperm cryopreservation in early adulthood ?

BILATERAL CONGENITAL ABSENCE OF VASA DEFERENTIA

- ❑ 1-2% male infertility
- ❑ 99% if cystic fibrosis
- ❑ 25% OA
  - o CF (AR) 1:2500 birth incidence
  - o BCAVD in isolation

Test the female partner (4%)

Genetic counseling

CFTR* mutations

***Cystic fibrosis transmembrane - conductance regulator (CFTR) gene**

*** Positive detection in 50 - 80% of men with BCAV (4% general pop.)**

CF/BCAV : check -up

- Atrophy during the fetal life of the Wolffian duct derivatives (seminal vesicles, ejaculatory ducts, vasa, epididymal body/tail)
- OA with normal testis volume, thin/absent scrotal vasa
- Ejaculate : low volume, low fructose, acidic ejaculation
- Renal tract anomalies in 10% of BCAV patients

SPERM RETRIEVAL

INDICATIONS

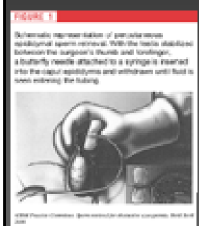
AZOOSPERMIA

- Non obstructive NOA C/A
- Obstructive OA C/A

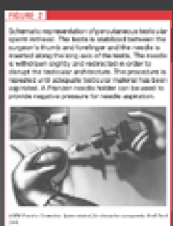
CRYPTOZOOSPERMIA
NECROZOOSPERMIA
IMMOTILE SPERMATOZOA / KARTAGENER
DNA FRAGMENTATION

SPERM-RETRIEVAL

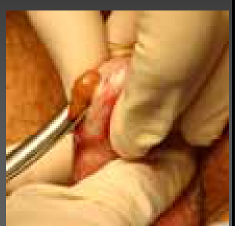
TECHNIQUES



Percutaneous epididymal sperm retrieval PESA



Percutaneous biopsy



Testicular sperm extraction TESE

TABLE 2

Advantages and disadvantages of sperm retrieval techniques.

	Advantages	Disadvantages
MESA	Best clinical pregnancy rates Large number of sperm retrieved Excellent results with cryopreservation Reduced risk of hematoma	Requires microsurgical expertise Increased cost General or local anesthesia Incision required Postoperative discomfort
TESE	No microsurgical expertise required Local or general anesthesia Few instruments Fast and repeatable	Relatively few sperm retrieved Limited risk of testicular atrophy (with multiple biopsies)
PESA	No microsurgical expertise required Local anesthesia Few instruments Fast and repeatable Minimal postoperative discomfort	Few sperm retrieved Risk of hematoma Damage to adjacent tissue
PercBiopsy, TESA, TEFNA	No microsurgical expertise required Local anesthesia Few instruments Fast and repeatable Minimal postoperative discomfort	Few sperm retrieved Risk of testicular atrophy Risk of hematoma

Note: TESE = testicular sperm extraction; PercBiopsy = percutaneous testicular biopsy; TESA = testicular sperm aspiration; TEFNA = testicular fine-needle aspiration.

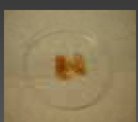
ASRM Practice Committee. Sperm retrieval for obstructive azoospermia. Fertil Steril 2008.

SURGICAL SPERM RETRIEVAL

Transport in culture medium

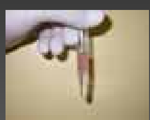
Measurement and washing

SURGICAL SPERM RETRIEVAL



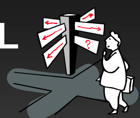
Dilaceration

Extraction



Centrifugation

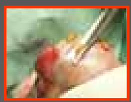
SPERM-RETRIEVAL

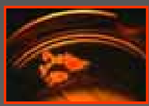


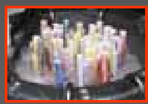
RESULTS

(NOA) C/A ? | Testis ? | Fresh ?

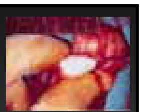
(OA) C/A ? | Epididymides ? | Frozen ?







ART WITH TESTICULAR SPERM



INDICATION

Better results in obstructive azoospermia

	FRESH/FROZEN	NOA		OA	
		Fertilization rate %	Clinical pregnancy rate %	Fertilization rate %	Clinical pregnancy rate %
Palermo et al., 1999	T FRESH + FROZEN	57.0* (n=53)	49.1	80.5* (n=14)	57.1
De Croo et al., 2000	T FRESH	67.8 * (n=54)	36.7	74.5* (n=139)	36.8
Vernaeve et al., 2003	T FRESH + FROZEN	48.5* (n=306)	15.4*	59.7* (n=605)	24.0*
Kanto et al; 2008	T FRESH	33.2 (n=17)	34.2	30.0 (n=18)	32.8
Ishikawa et al., 2008	T FROZEN	57.6* (n=75)	30.9	63.0* (n=184)	29.9

FRESH/FROZEN

- FIVNAT : 2001-2005

	SPERMATOZOA						
	Fresh			Frozen			
	Cycles	Pregnancy/Cycle		Cycles	Pregnancy/Cycle		p
	N	N	%	N	N	%	
Testicular	1082	243	22,5	1006	204	20,3	0,22
Epididymal	602	183	30,4	2202	517	23,5	0,01
p	0,001			0,04			

- Neri et al., 2008

	Spermatozoa			
	Epididymal		Testicular	
	Fresh	Frozen/Thawed	Fresh	Frozen/Thawed

- **Fresh >>Frozen/Thawed**, : chi2=11,0 ; p<0,001
- **Epididymal >>Testicular**, : chi2=11,0 ; p<0,001

OBSTRUCTIVE AZOOSPERMIA

EPIDIDYMAL/TESTICULAR ?

Better results with epididymal sperm !

- Buffat et al., 2007; FIVNAT

N = 171 OA (368 ICSI)	Testicular	Epididymal
Fertilization %	51.9	58.9
Clinical pregnancy %	24.3	22.1
Spontaneous abortion SA %*	35.7	12.5
Malformations	1	3

Better results with testicular sperm !

- Dozortsev et al., *2006*

N = 265 OA	Testicular	Epididymal
Fertilization %	67.5	77.2
Clinical pregnancy %	51.3	37
Implantation %	32.8	20.8
SA %	25.0	34.7

NON OBSTRUCTIVE AZOOSPERMIA

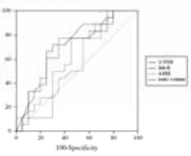
PREDICTIVE VALUE FOR TESE OUTCOME :

- Testis volume
- Serum FSH, inhibin B, AMH
- Seminal inhibin B, AMH

Controversy ! No ideal marker ! Combined markers

FIGURE 2

Receiver operating characteristic curves of 1/FSH, INHB, AMH, and volume of the larger testis for sperm retrieval in testicular FNA.



Goulis. Serum inhibin B and AMH in testicular FNA. Fertil Steril 2008.

TABLE 3

Experimental parameters according to testicular sperm extraction (TESE) outcome.

TESE outcome	Seminal inhibin B concentration Mean (SEM) (ng/L)	Seminal AMH concentration (pmol/L) Mean (SEM)	Serum FSH concentration Mean (SEM) (UI/L)	Serum inhibin B concentration Mean (SEM) (ng/L)
Positive	45.7 ± 54.7	8.64 ± 20.96	11.4 ± 7.5	66.25 ± 58.8
Negative	33.3 ± 24.6	4.52 ± 4.7	29.5 ± 14.0	22.2 ± 19.6

Note: ANOVA after logarithmic transformation: $P = .0008$ for serum FSH and $P = .0129$ for serum inhibin B; $P = .9158$ for seminal inhibin B and .9433 for seminal AMH.

Duvilla. Inhibin B and AMH in seminal plasma. Fertil Steril 2008.

NON OBSTRUCTIVE AZOOSPERMIA

CRYO-TESE-ICSI : CRUDE CUMULATIVE RATE

	Sertoli Cell Only	Maturation arrest	Hypospermatogenesis
N° spermatozoa/straw	8 (-10)	15 (3-50)	25 (10-80)
Fertilization rate FR %	61*	49*	69*
Implantation rate IR %	17.1	25.3	24.0

[illegible]	1	2	3	4
No. of ICSI procedures	[illegible]	[illegible]	[illegible]	7
Mean maternal age (years)	[illegible]	[illegible]	[illegible]	[illegible]
No. of deliveries	13	12	2	2
Delivery rate per cycle (%)	[illegible]	[illegible]	[illegible]	[illegible]
Cumulative delivery rate (%)	25	49	53	57

53 61

De Croo et al., 2000; Giorgetti et al., 2005

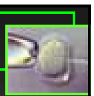
Y MICRODELETION

Table 5 : Outcome of ICSI cycles in couples with Y deletion

Study	Couple (n)	ICSI cycle (n)	Pregnancy [a] (n) PR per cycle (%)	Deliveries (n) DR per cycle (%)	Children born (n)
Mulhall *et al.* (1997)	3	6	1 (16.6%)	1 (16.6%)	2
Van Golde *et al.* (2001)	8	19	3 (15.8%)	3 (15.8%)	5
Oates *et al.* (2002)	26	48	13 (27.1%)	13 (27.1%)	18
Choi *et al.* (2004)	17	27	9 (33.3%)	7 (25.9%)	8
Stouffs *et al.* (2005)	16	40	7 (17.5%)	3 (7.5%)[b]	3
Our study	23	42	13 (30.9%)[c]	8 (19.1%)[b]	12
TOTAL	**93**	**182**	**46**	**35**	**48**

PR: pregnancy rate; DR: delivery rate; [a] Including clinical pregnancies obtained after only fresh embryos transfers; [b] + 2 and 1 ongoing pregnancies respectively; [c] additional clinical pregnancies were obtained after frozen-thawed embryo transfers, leading to the birth of a healthy boy.

KLINEFELTER SYNDROME



Reference	n	Sperm retrieval, %	Type of spermatozoa used for successful fertilisation	Transferred embryos	Clinical pregnancies (as defined by fetal heartbeat)	Liveborn children	Karyotype of conceptus or neonate
91, 92	20	50	Fresh	31	3 (singleton)	3	46,XY (2); 46,XX
93, 94			Frozen-thawed	8	1 (singleton)	*	
95	2	..	Fresh	9	2 (1 singleton, 1 twin)	3	46,XY (2); 46,XX
96	7	57	Fresh	4	1 (singleton)	1	46,XY
97	1	..	Fresh	3	1 (twin)	2	46,XY (2)
98	1	..	Fresh	3	1 (singleton)	1	46,XY
99	1	..	Frozen-thawed†	10	2 (twin; in 3 treatment cycles)	2	46,XY (2)
100	1	..	Fresh	3	1 (triplet)	2	46,XX; 46,XY, 1 47,XXY aborted
101	52	..	Fresh	..	1 (singleton)	1	46,XX
102	20	40	Fresh	..	4 (2 singleton, 1 twin, 1 triplet)	7	46,XY (4); 46,XX (3)
103	1	..	-	..	..	1	46,XX
104	12	42	Fresh	15	3 (2 singleton and 1 triplet)	4	4 healthy neonates; 1 47,XXY aborted
			Frozen-thawed	18	2 (1 twin, 1 abortion)	2	46,XY (2)
105	1	..	Fresh	3	1 (twin)	2	46,XY (2)
106	2	100	Fresh	6	2 (singleton)	2	46,XX; 46,XY
24	19	21	Fresh	..	4 (3 singleton, 1 miscarriage)	Not known	..
107	1	..	Frozen-thawed	4	1 (singleton)	1	46,XY
108	24	50	Fresh	..	4 (3 singleton, 1 twin)	5	46,XY (2); 46,XX (3)
109	12	55	Fresh	25	2 (singleton)	1	46,XX
110	8	..	-	..	4 (2 singleton, 2 twin)‡	3	..
Total	185	52	-	142	40	43	..

*Child stillborn at 23 weeks of gestation. †From same patient reported by Ron-El et al. ‡Two pregnancies (one singleton, one 1 twin) were still under way when the paper was published.

Table 4: Reported pregnancies after ICSI treatment with testicular spermatozoa of patients with non-mosaic Klinefelter's syndrome

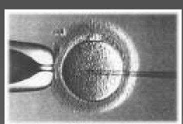
Lanfranco et al., 2004; Schiff et al., 2005; Kyono et al., 2007; Chantot-Bastaraud et al., 2008

CRYO - ONCO-TESE - ICSI

CASE REPORT

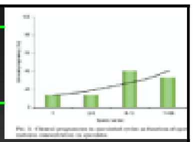
Testicular sperm extraction in a single cancerous testicle in patients with azoospermia: A case report

<u>A SINGLE CANCEROUS TESTIS AND AZOOSPERMIA</u>



Descombes et al., 2007

CRYPTOZOOSPERMIA



❑EJACULATED/TESTICULAR (NOA)

TABLE 2. *ICSI outcome in couples with discordant sperm origin*

	No. Ejaculated		No. TESE	
Cycles	16		16	
MII	165		192	
2 PN (%)*	80	(48.5)	117	(60.9)
Embryos replaced (mean ± SD)	2.8 ± 0.8		3.3 ± 0.6	
Implantation/total (%)	2/37	(5.4)	10/45	(22.2)
Deliveries (%)	2	(14.3)	7	(50.0)

There were 14 transfers per group.
* Effect of sperm origin on fertilization 2 × 2, 1 df, chi-square p <0.05.

Better results in TESE

Bendikson et al., 2008

❑POOR EMBRYO QUALITY AND REPEATED IMPLANTATION FAILURES

Better results in TESE

Weissman et al., 2008

DNA FRAGMENTATION

Table I. Basic sperm parameters of the patients involved in this study and the incidence of DNA fragmentation in their ejaculated and testicular sperm samples

Patient	Basic sperm parameters			% Spermatozoa with fragmented DNA	
	Concentration (×10⁶/ml)	Motility (%)	Normal forms (%)	Ejaculate	Testis
1	6	11	2	20	3
2	21	32	3	13	3
3	38	71	20	24	4
4	35	60	11	21	3
5	3	19	9	27	2
6	25	65	17	31	6
7	79	42	49	29	4
8	22	7	8	25	1
9	26	15	6	22	10
10	3	14	7	25	9
11	19	29	23	26	4
12	91	41	48	21	3
13	12	65	11	37	6
14	24	32	80	19	5
15	1	42	22	17	6
16	20	21	13	28	4
17	17	56	10	26	5
18	46	44	58	19	5

Better results in TESE

EJACULATED VS TESTICULAR SPERM (NOA)

Table III. Implantation and pregnancy after ICSI with ejaculated and testicular spermatozoa

Sperm source	Attempts	Embryos transferred	Clinical pregnancies[a]	Pregnancy rate[a]	Gestational sacs[a]	Implantation rate[a]
Ejaculate	18	56	1	5.6%[a]	1	1.8%[a]
Testis	18	58	8	44.4%[a]	12	20.7%[a]

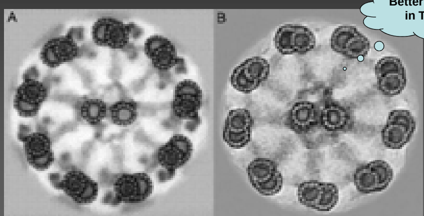
SA

Greco et al., 2005

IMMOTILE SPERMATOZOA

Birth after intracytoplasmic sperm injection with use of testicular sperm from men with Kartagener or immotile cilia syndrome

HOS TEST

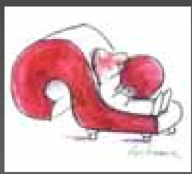


Better results in TESE

Westlander et al., 2003; Kaushal et al., 2007

CHILDREN

Patients ?



Malformations ?

CHILDREN….

❑ 252 TESE (227) AND MESA (25)

No influence of sperm !

Ludwig et al, 2003

❑ 412 TESE (318) and MESA (94)

More girls 45.4% vs 53.1% IVF (P < 0.005)

Ludwig et Katalinic, 2002 ; Fedder et al, 2007

❑ 737 TESE (195) and MESA (542)

More malformations and chromosomal abnormalities TESE 6.48% vs MESA vs 2.38% vs ICSI 3.17% (p<0.0001)

Bajirova et FIVNAT et al, 2001

Malformations

FIVNAT

Major malformations (%)	TESE	MESA	EJACULATED ICSI	EJACULATED IVF
Bonduelle et al	6/206 (2.9)	4/105 (3.8)	84/2477 (3.4)	112/2955 (3.8)
Kallen et al	3/147 (2.0)	5/135 (3.7)	139/4248 (3.3)	284/10116 (2.8)
Ludwig and Katalinic	21/229 (9.2)	1/26 (3.8)	248/2944 (8.4)	_
Palermo et al	1/87 (1.1)	4/198 (2.0)	33/1774 (1.9)	30/176 (1.7)
Wennerholm et al	0/31 (0.0)	3/69 (4.3)	39/934 (4.2)	_
Our study	2/176 (1.1%)	8/266 (3.0%)***	185/5250 (3.5%)	144/4537 (3.2%)

Woldringh et al, Aout 2009

CHROMOSOMAL ABNORMALITIES IN MISCARRIAGES

- ❑ No difference **ART (63.2%)** versus **natural conception (71.5%)**
- ❑ No difference **ICSI (61.5%)** versus **IVF (54.5%)**
- ❑ **ICSI TESE (80% abnormal)** : **50% tri / tetraploidy**

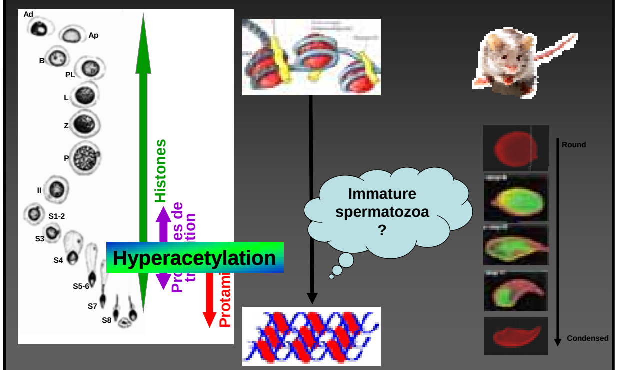
	Autosomal trisomy n (%)	Autosomal double trisomy n (%)	Triploidy n (%)	Tetraploidy n (%)	Monosomy X n (%)	Structural anomalies[a] n (%)
IVF	13 (72.2)	1 (5.6)	1 (5.6)	1 (5.6)	2 (11.0)	–
ICSI	23 (57.5)	4 (10.0)	4 (10.0)	3 (7.5)	4 (10.0)	2 (5.0)
ICSI-TESE	4 (50.0)	–	2 (25.0)[b]	2 (25.0)[c]	–	–
cryo-ICSI	4 (66.7)	–	–	–	–	2 (33.3)
IUI	8 (66.7)	3 (25.0)	–	1 (8.3)	–	–
Total	52 (61.9%)	8 (9.5%)	7 (8.3%)	7 (8.3%)	6 (7.2%)	4 (4.8%)

[a] de novo: ICSI: 45,XX,t(13;14)(q10;q10); 46,XY,del(2)(q24q34); cryo-ICSI: mos 46,XY,add(14)(p12)/46,XY; mos 46,XX,add(22p)/46,XX.
[b] 69,XXY; 68,XXX,der(14;21)(q10;q10).
[c] 94,XXXX,+16,+16; mos 94,XXXX,+14,+14[5]/47,XX,+14[5].

- o immature diploid sperm ?
- o incorrect oocyte activation ?
- o incorrect oocyte maturation ?
- o post-zygotic abnormality ?

Bettio et al, 2008

EPIGENETIC RISK ?



EPIGENETIC RISK?

❑**Epigenetic alterations of IgF2 – H19 in spermatozoa from infertile men**

Genomic imprinting in disruptive spermatogenesis
Cristina Joana Marques, Filipa Carvalho, Mário Sousa, Alberto Barros

❑**DNA methylation errors at imprinted loci after ART originate in the parental sperm**

Direct inheritance from the father's sperm : a source of imprinting error in ART

Marques et al, 2004 et 2008; Kobayashi et al., 2007; 2009; Chalas et al., 2010


THANKS !