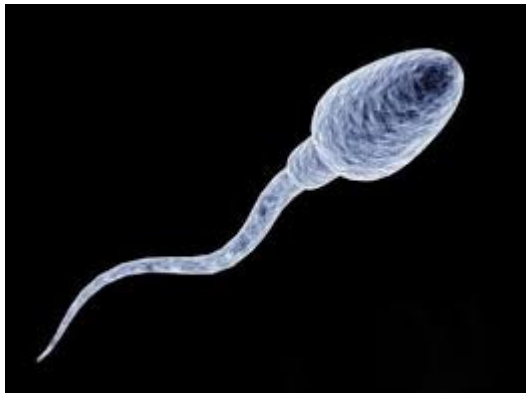
Semen analysis report

2nd symposium of Croatian Society of Clinical Embryologist and Andrology Workshop, Opatija, 28-30 November, 2014



- No single test or sperm parameter was found to be absolute in its prediction of male fertility or infertility
- The correct evaluation of the basic semen parameters still remains the most cost-effective diagnostic tool for male fertility
- The importance of the spermatozoon's contribution to embryo genesis, haploid genome, the centrosome, and the signal to initiate oocyte activation, cannot, however, be underestimated

(Coetzee et al. Hum Reprod Update 1998)

WHO Laboratory Manual for the Examination and processing of Human Semen, 5th edn. Geneva: World Health Organization, 2010.

Table A1.1 Lower reference limits (5th centiles and their 95% confidence intervals) for semen characteristics

Parameter	Lower reference limit
Semen volume (ml)	1.5 (1.4–1.7)
Total sperm number (10^6 per ejaculate)	39 (33–46)
Sperm concentration (10^6 per ml)	15 (12–16)
Total motility (PR + NP, %)	40 (38–42)
Progressive motility (PR, %)	32 (31–34)
Vitality (live spermatozoa, %)	58 (55–63)
Sperm morphology (normal forms, %)	4 (3.0–4.0)
Other consensus threshold values	
pH	≥7.2
Peroxidase-positive leukocytes (10^6 per ml)	<1.0
MAR test (motile spermatozoa with bound particles, %)	<50
Immunobead test (motile spermatozoa with bound beads, %)	<50
Seminal zinc (μmol/ejaculate)	≥2.4
Seminal fructose (μmol/ejaculate)	≥13
Seminal neutral glucosidase (mU/ejaculate)	≥20

Record form for semen

Name:			
Code:			
Date (day/month/year)			
Collection (1, at laboratory; 2, at home)			
Collection time (hour : minute)			
Sample delivered (hour : minute)			
Analysis begun (hour : minute)			
Patient			
Abstinence time (days)			
Medication			
Difficulties in collection			
Semen			
Treatment (e.g. bromelain)			
Complete sample? (1, complete; 2, incomplete)			
Appearance (1, normal; 2, abnormal)			
Viscosity (1, normal; 2, abnormal)			
Liquefaction (1, normal; 2, abnormal) (minutes)			
Agglutination (1–4, A–E)			
pH [≥7.2]			
Volume (ml) [≥1.5]			
Spermatozoa			
Total number (10^6 per ejaculate) [≥39]			
Concentration (10^6 per ml) [≥15]			
Error (%) if fewer than 400 cells counted			
Vitality (% alive) [≥58]			
Total motile PR+NP (%) [≥40]			
Progressive PR (%) [≥32]			
Non-progressive NP (%)			
Immotile IM (%)			
Normal forms (%) [≥4]			
Abnormal heads (%)			
Abnormal midpieces (%)			
Abnormal principal pieces (%)			
Excess residual cytoplasm (%)			
Direct MAR-test IgG (%) (3 or 10 minute) [<50]			
Direct MAR-test IgA (%) (3 or 10 minute) [<50]			
Direct IB-test IgG (% with beads) [<50]			
Direct IB-test IgA (% with beads) [<50]			
Non-sperm cells			
Peroxidase-positive cells, concentration (10^6 per ml) [<1.0]			
Accessory gland function			
Zinc (µmol per ejaculate) [≥2.4]			
Fructose (µmol per ejaculate) [≥13]			
α-Glucosidase (neutral) (mU/ejaculate) [≥20]			
Technician:			

Table A1.3 Nomenclature related to semen quality

aspermia	no semen (no or retrograde ejaculation)
asthenozoospermia	percentage of progressively motile (PR) spermatozoa below the lower reference limit
asthenoteratozoospermia	percentages of both progressively motile (PR) and morphologically normal spermatozoa below the lower reference limits
azoospermia	no spermatozoa in the ejaculate (given as the limit of quantification for the assessment method employed)
cryptozoospermia	spermatozoa absent from fresh preparations but observed in a centrifuged pellet
haemospermia (haematospermia)	presence of erythrocytes in the ejaculate
leukospermia (leukocytospermia, pyospermia)	presence of leukocytes in the ejaculate above the threshold value
necrozoospermia	low percentage of live, and high percentage of immotile, spermatozoa in the ejaculate
normozoospermia	total number (or concentration, depending on outcome reported)* of spermatozoa, and percentages of progressively motile (PR) and morphologically normal spermatozoa, equal to or above the lower reference limits
oligoasthenozoospermia	total number (or concentration, depending on outcome reported)* of spermatozoa, and percentage of progressively motile (PR) spermatozoa, below the lower reference limits
oligoasthenoteratozoospermia	total number (or concentration, depending on outcome reported)* of spermatozoa, and percentages of both progressively motile (PR) and morphologically normal spermatozoa, below the lower reference limits
oligoteratozoospermia	total number (or concentration, depending on outcome reported)* of spermatozoa, and percentage of morphologically normal spermatozoa, below the lower reference limits
oligozoospermia	total number (or concentration, depending on outcome reported)* of spermatozoa below the lower reference limit
teratozoospermia	percentage of morphologically normal spermatozoa below the lower reference limit

*Preference should always be given to total number, as this parameter takes precedence over concentration.

Barratt CLR, Björndahl L, Menkveld R and Mortimer D.
Hum Reprod 26 (12) 2011: 3207-3212.

ESHRE special interest group for andrology basic semen analysis course: a continued focus on accuracy, quality, efficiency and clinical erelevance.

Critical review from authors of ESHRE Basic Semen Course

Sperm motility

- Abandons the distinction between slow- and rapid-progressive spermatozoa
- The arguments posited by the WHO5 have been refuted elsewhere (Bjorndahl, 2010; Eliasson, 2010)
- Clinical data both from manual sperm motility assessments and CASA showing showing the distinction of rapidly progressive spermatozoa to be biologically important (MacLeod i Gold, 1951; Barratt et al. 1992; Sifer et al. 2005)

Sperm morphology

- WHO5 has fully adopted the Tygerberg Strict Criteria for normal sperm morphology
- WHO5 the assessment of multiple sperm defects (teratozoospermia index, TZI) has been relegated to „Optional Procedures“
- Rowe i sur. (2000) - illustrative case with 4% normal forms indicating that if the TZI was < 1.7 succesful fertilization may be expected in vitro without ICSI, with a TZI > 1.9 ICSI may well be required in order to achieve fertilization

Nomenclature terms

- oligozoospermia - these terms simply classify the perceived quality of the semen and do not identify, or even suggest, biological cause or real fertility potential (Eliasson i sur. 1970; Eliasson 1977, 2010; Bostofte i sur. 1981)
- normal, doubtful and pathological or not normal (Bjorndahl et al. 2010)

Multiple methods and nonlinear method presentation

- alternative stains for sperm morphology assessment (e.g. Diff-Quick)
- use of eosin without a counter stain for sperm vitality assessment
- determining sperm concentration is presented in a unnecessarily complex manner

Inconsistencies and errors

- sperm vitality using eosin-nigrosin staining: the cut-off to perform a vitality assessment has been changed from >50% immotile spermatozoa (WHO, 1992, 1999) to „less than about 40% progressively motile spermatozoa“ (WHO, 2010)
- the change is illogical since non-progressively motile spermatozoa are clearly still 'live'
- eosin staining: 'light pink heads are considered alive' (WHO, 2010); the standard criterion is that degree of pink colouration indicates that a spermatozoon is not 'live' (Mortimer, 1994)

Unnecessary extra work

- It is stated that both sperm vitality and sperm morphology assessments must be made in duplicate, evaluating 200 spermatozoa in each replicate (WHO5)
- There requirements represent substantial extra work for what are unestablished improvements in accuracy

Illogical sperm preparation methods

- WHO5 still allows simple centrifugal washing of spermatozoa for „good quality“ semen samples
- Recommended density gradient method contains numerous errors
- WHO5 still recommends Ham's F10 medium for all sperm preparation methods, 15 years after a clear recommendation that it not be used for this purpose due to its iron content (Gomez and Aitken, 1996)

The delusion of suddenly changed limits between fertile and subfertile men

- WHO5 lowered reference limits calculated from results on semen provided by recent fathers and men in a general population (individuals without disorders)
- Semen samples obtained after 2-7 days of abstinence - MacLeod and Gold (1952) clearly demonstrated that ejaculte volume, and sperm concentration in particular, increase considerably with each day of increasing abstinence
- It is therefore of the utmost importance that the prescribed period of abstinence before a semen analysis should be from 3 to 4 days (Bjorndahl et al. 2010)

The handbook **A practical Guide to Basic Laboratory Andrology** (Bjorndahl i sur. 2010) is reference text for the ESHRE BSA courses.

ANALIZA KVALITETE SJEMENA

Ime i prezime	
Datum rođenja	
OIB	
ID analize	

Trajanje apstinencije (dani)	
Uzorak dan: 1 - u poliklinici; 2 - donešen	
Cijeli uzorak sakupljen: DA - NE	
Vrijeme proteklo od davanja do analize	

Sjeme	**Ref. vrijednosti**	**Izmjerene vrijednosti**
Volumen (ml)	>1,5ml	
Izgled i boja		
Viskoznost		
Likvefakcija		
Aglutinacija (1-4; A-E)		
Agregacija		
pH	≥ 7,2	
Spermiji		
Ukupan broj spermija u sjemenu (10^6)	≥ 39 x 10^6	
Koncentracija spermija (10^6/ml)	≥ 15 x 10^6 /ml	
Ukupna pokretljivost spermija (% PR + NP)	≥ 40%	
Progresivno pokretni spermiji (% PR)	≥ 32%	
Neprogresivno pokretni spermiji (% NP)	-	
Nepokretni spermiji (% IM)	-	
Vitalnost spermija (% živih spermija)	≥ 58%	
Morfološki pravilni spermiji (%)	≥ 4%	
Leukociti (10^6/ml)	< 1 x 10^6	
Drugi funkcionalni testovi		
MAR IgG (% vezanih spermija)	< 50%	
MAR IgA (% vezanih spermija)	< 50%	
DNA fragmentacija (% frag. spermija)	< 30%	
HBA test (% pokretnih vezanih sperm.)	>80%	

Dijagnoza	
Napomena	

**WHO Laboratory Manual for the Examination and Processing of Human Semen (WHO 2010)*

HVALA !