

Appendices

APPENDIX 1 Reference values and semen nomenclature

A1.1 Reference values

Measurements made on semen samples need to be compared with reference values to allow decisions to be made about patient management and thresholds for clinical trials or investigations. The reference values given here have been generated from the results of several prospective, cross-sectional studies of semen quality and fertility. They were obtained by direct, retrospective selection of fertile men, defined as men whose partner conceived within 12 months after stopping use of contraception (Cooper et al., 2010).

- Only complete semen samples—one per man (the first where several were given), obtained following 2–7 days of abstinence—were included in this analysis.
- Semen volume was measured using methods recommended by WHO at the time, namely, weighing or transferring to pipettes or graduated vessels. Total sperm number was calculated from concentrations measured by haemocytometer on fixed, diluted samples. Total motility (PR+NP), progressive motility (PR), non-progressive motility (NP) and immotile sperm (IM) were measured at room temperature or at 37 °C. Data on normal sperm morphology were taken only from laboratories that provided values not exceeding the anticipated maximum level for the strict categorization (Tygerberg) method (approximately 35% normal forms). Vitality was determined by exclusion of vital dye (eosin) from sperm head membranes.
- Statistical tradition is to take the 2.5th centile from a two-sided reference interval as the threshold below which values may be considered to come from a different population. However, a one-sided reference interval was considered to be more appropriate for semen parameters, since high values are unlikely to be detrimental to fertility. The 5th centile lower reference limits are given in Table A1.1, and the complete frequency distributions are given in Table A1.2.

Comment 1: The reference distributions in Table A1.2 provide a description of the semen characteristics of recent fathers, whose partner became pregnant within 12 months of stopping use of contraception.

Comment 2: Fathers constitute a select group of individuals and their semen parameters may be different from those of the general population of healthy men.

Comment 3: Semen characteristics are highly variable, both within and among men, and are not the sole determinants of a couple's fertility; the ranges therefore provide only a guide to a man's fertility status.

Comment 4: Semen parameters that lie within the 95% reference interval do not guarantee fertility.

Comment 5: Men whose semen characteristics fall below the lower limits given here are not necessarily infertile; their semen characteristics are below the reference range for recent fathers—as are, by definition, those of 5% of the fertile men who provided data used in the calculation of the reference range.

Comment 6: A man's semen characteristics need to be interpreted in conjunction with clinical information.

Comment 7: There may be regional differences in semen quality, or differences between laboratories; laboratories should consider preparing their own reference ranges, using the techniques described in this manual.

Comment 8: Time to pregnancy is also affected by the female partner's fertility status.

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 ($\mu\text{mol}/\text{ejaculate}$)	≥ 2.4
Seminal fructose ($\mu\text{mol}/\text{ejaculate}$)	≥ 13
Seminal neutral glucosidase (mU/ejaculate)	≥ 20

Table A1.2 Distribution of values for semen parameters from men whose partners became pregnant within 12 months of discontinuing contraceptive use

Parameter (units)	N	Centile								
		2.5	5	10	25	50	75	90	95	97.5
Semen volume (ml)	1941	1.2	1.5	2.0	2.7	3.7	4.8	6.0	6.8	7.6
Total sperm number (10 ⁶ per ejaculate)	1859	23	39	69	142	255	422	647	802	928
Sperm concentration (10 ⁶ per ml)	1859	9	15	22	41	73	116	169	213	259
Total motility (PR + NP, %)	1781	34	40	45	53	61	69	75	78	81
Progressive motility (PR, %)	1780	28	32	39	47	55	62	69	72	75
Non-progressive motility (NP, %)	1778	1	1	2	3	5	9	15	18	22
Immotile spermatozoa (IM, %)	1863	19	22	25	31	39	46	54	59	65
Vitality (%)	428	53	58	64	72	79	84	88	91	92
Normal forms (%)	1851	3	4	5.5	9	15	24.5	36	44	48

Source: Cooper et al., 2010.

A1.2 Nomenclature

This manual retains the nomenclature introduced to describe deviations from reference semen values, using words rather than numbers (see Table A1.3), although some have argued for the abandonment of such terminology (Grimes & Lopez, 2007). The nomenclature simply classifies the quality of the semen and does not suggest any biological cause (Eliasson et al., 1970). These terms are used to describe samples with values lying outside the reference range, and therefore possibly originating from a different population. Much of the semen nomenclature relates to a single parameter. However, normozoospermia refers to three sperm parameters—number, motility and morphology. Thus deviations from the reference range for each parameter can be described individually.

References

- Cooper TG et al. (2010). World Health Organization reference values for human semen characteristics. *Human Reproduction Update*, 16:231-245.
- Grimes DA, Lopez LM (2007). "Oligozoospermia", "azoospermia", and other semen-analysis terminology: the need for better science. *Fertility and Sterility*, 88:1491-1494.
- Eliasson R et al. (1970). Empfehlungen zur Nomenklatur in der Andrologie. *Andrologia*, 2:1257.