

Semen collection and physical examination

Semen analysis

Spermatozoa were first described by Leeuwenhoek in the 17th century but it was not until 1928 that the sperm count was found to be associated with fertility potential. Since that time a variety of sperm tests and semen parameters have been developed with the hope of clarifying whether or not a man could impregnate his partner.

MacLeod (1942), MacLeod and Gold (1953), Eliasson (1971) and Hellinga (1949,1976) have led the scientific basis of conventional analysis of spermatozoa and the techniques recommended by them are still considered the reference for more advanced methods.

Semen analysis comprises a set of descriptive measurements of spermatozoa and seminal fluid parameters that help to estimate semen quality.

Conventional semen analysis includes measurement of particular aspects of spermatozoa such as concentration, motility and morphology and of seminal plasma. Quantification and identification of non-spermatozoidal cells and detection of antisperm antibodies are also part of basic semen analysis.

Sample collection and delivery

The following instructions for sample collection and delivery are based on WHO recommendations. The subject should be provided with clearly written or oral instructions concerning the collection and, if required, transport of the semen sample.

1. The sample should be collected after a minimum of 48 hours and no longer than 7 days of sexual abstinence. The name of the man, period of

abstinence, date and time of collection should be recorded. The time interval between the last ejaculation and sample collection should be well defined and preferentially as constant as possible in order to allow a reliable interpretation of the results of, in particular, sperm concentration and motility. When the duration of abstinence is more than 7 days, sperm motility, i.e. the proportion of spermatozoa with rapid progressive motility, may decline. If the duration of abstinence is <48h, sperm concentration may be reduced, but motility will probably not be affected.

2. Two semen samples should be collected for initial evaluation. The interval of time between the collections will depend on local circumstances but should not be less than 7 days or more than 3 months apart. If the results of these assessments are remarkably different, additional semen samples should be tested because marked variations in sperm output may occur within the same individual. Analysis of multiple semen specimens provides a reliable screen in the evaluation of male factor infertility. Information and support are important since semen analysis cause a moderate amount of stress.
3. Ideally the sample should be collected in the privacy of a room near the laboratory. If not, it should be delivered to the laboratory within 1h after collection.
4. The sample should be obtained by masturbation and ejaculated into a clean, wide-mouthed glass or plastic container. If plastic is used, it should be checked for lack of toxic effects on spermatozoa. The container should be warm to minimize the risk of cold shock.
5. Ordinary condoms must not be used for semen collection because they may interfere with the viability of spermatozoa. In cases in which masturbation is not possible or against an individual's values, the specimen can be collected in a non-spermicidal condom following intercourse. It has been shown that semen samples collected during intercourse using a special

plastic condom or a silastic collection device tend to have better parameters. Other authors, referring to their experience, hold the view that the quality of the specimen when collected in this way is generally compromised. This way of collection should be considered for a second sample if the first one shows a relatively low volume. Coitus interruptus is not acceptable as a means of collection because it is possible that the first portion of the ejaculate, which contains the highest concentration of spermatozoa, will be lost. Moreover, there will be cellular and bacteriological contamination of the sample and the acid pH of the vaginal fluid will adversely affect sperm motility.

6. Incomplete samples should be not analyzed, particularly if the first portion of the ejaculate is lost. The sample should be protected from extremes of temperature (not less than 20°C and not more than 40°C) during transport to the laboratory. The sample should be examined immediately after liquefaction and certainly within 1h of ejaculation.

Laboratory technicians should be aware that semen samples may contain harmful viruses (e.g., HIV and viruses causing hepatitis and herpes) and should therefore be handled with due care.

Examination is carried out after liquefaction of semen that occurs usually within 20-30 minutes of ejaculation.

1. VISUAL APPEARANCE

Normal semen is viscous and opaque gray-white in appearance. After prolonged abstinence, it appears slightly yellow.

2. VISCOSITY

Immediately following ejaculation, normal semen is thick and viscous. It becomes liquefied within 30 minutes by the action of

proteolytic enzymes secreted by prostate. If liquefaction does not occur within 60 minutes, it is abnormal. The viscosity of the sample is assessed by filling a pipette with semen and allowing it to flow back into the container. Normal semen will fall drop by drop. If droplets form 'threads' more than 2 cm long, then viscosity is increased. Increased semen viscosity affects sperm motility and leads to poor invasion of cervical mucus; it results from infection of seminal vesicles or prostate.

3. VOLUME

Volume of ejaculated semen sample should normally be > 2 ml. It is measured after the sample has liquefied. Volume < 2.0 ml is abnormal, and is associated with low sperm count.

4. pH

A drop of liquefied semen is spread on pH paper (of pH range 6.4-8.0) and pH is recorded after 30 seconds. Normal pH is 7.2 to 8.0 after 1 hour of ejaculation. The portion of semen contributed by seminal vesicles is basic, while portion from prostate is acidic. Low pH (< 7.0) with absence of sperms (azoospermia) suggests obstruction of ejaculatory ducts or absence of vas deferens. Low pH is usually associated with low semen volume (as most of the volume is supplied by seminal vesicles).