DIAGNOSTIC SEMEN ANALYSIS
GP USER MANUAL

CONTENTS

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GP USER MANUAL

1 INTRODUCTION

This manual describes the policies, procedures and repertoire of the Andrology Laboratory at Homerton Fertility Centre, Homerton University Hospital.

This manual follows the standards outlined by the WHO Laboratory manual for The Examination and Processing of Human Semen, 2021.

Semen Analysis is performed only after receiving a referral and then booking an appointment with the Fertility Centre Laboratory. Samples brought in without an appointment will not be accepted or analysed.

Patients **must use only the sterile specimen pot provided by the Fertility Centre**, which must be collected from the Fertility Clinic before their booked appointment. GPs should ensure that patients re aware of these points and not provide the patient with any other sample pot.

2 CONTACT DETAILS

Homerton Fertility Centre
Homerton University Hospital
Homerton Row
London
E9 6SR
Email: huh-tr.fertility.unit@nhs.net
Phone: 020 8510 7660

3 OPENING TIMES

The Andrology Laboratory at Homerton Fertility Centre is open 08:30 – 16:00 Monday to Friday (except bank holidays) only.

4 ROUTINE TESTS PROVIDED

The Andrology Laboratory provides a range of Andrology tests and follows recommendations made by the World Health Organisation (WHO Laboratory manual for The Examination and Processing of Human Semen, 2021), Laboratory Guidelines for Post Vasectomy Analysis 2016 and The Association of Reproductive and Clinical Scientists (ARCS).

Routine tests include:

- Diagnostic fertility semen analysis (including assessment of retrograde ejaculation)
- Post-vasectomy semen analysis

5 PROTECTION OF PATIENT INFORMATION

The information that you give to us regarding your patients is protected by Homerton Healthcare NHS Foundation Trust policies. The following link will direct you to more information if required: https://www.homerton.nhs.uk/about-us

DIAGNOSTIC SEMEN ANALYSIS
GP USER MANUAL

6 HOW TO REQUEST A SEMEN ANALYSIS

6.1 External referrals

Semen analysis appointments for the Andrology Laboratory at Homerton Fertility Centre are made through the NHS e-Referral Service (E-RS) RAS.

6.1.1 Receipt of referral

On receipt and confirmation of a referral, the patient, patient's partner or a relative/friend authorised by the patient should visit Homerton Fertility Centre to collect following:

a. Semen collection pot and sample bag
b. Semen analysis instruction and consent form

6.2 Internal referrals

Appointments will be made through EPR and IDEAS database by the admin team. The sample collection kit comprising of the Semen collection pot, bag, and semen production instruction & consent form can be picked up from the sample collection kit bin located at the front of the reception desk.

7 SEMEN SAMPLES PRODUCTION

Patients are advised to carefully follow the instructions on the semen production instruction form to optimise the accuracy of the diagnostic outcome of their sample. The instructions should be read in advance of the appointment due to abstinence requirements for this test. Patients should be advised to only use the collection pot provided by the laboratory and the sample should be produced by masturbation only.

If they are unable to ejaculate by masturbation, special male factor paks for use at home may be provided in exceptional circumstances. However, the entire ejaculate will not be available for examination, and the specimen is likely to be contaminated by contact with the skin of the penis and to some extent also vaginal fluid and cells on the outside of the male factor pak.

8 TRANSPORTATION OF SEMEN SAMPLES

Patient can produce sample at home or at the facility available at the Homerton clinic. If the sample produced at home, the sample pot should be kept close to the body under clothes during transportation (e.g., pocket) and delivered to the laboratory within maximum 40 minutes after collection. If the sample is not delivered within the mentioned specified time frame, the patient may need to repeat this process. If preferable, the patient's partner can deliver the sample on the patient's behalf.

9 CRITERIA FOR SAMPLE ACCEPTANCE/REJECTION

Any patient who attends the laboratory without a prior referral will not be accepted under any circumstances including the following:

- If an unlabeled (name, DOB, and date) sample pot is dropped off with an incomplete semen analysis form.
- If the semen analysis form is not clearly signed and filled completely.

- If the sample has been collected into a non- issued laboratory container.
- If the sample has leaked into the specimen bag.

Samples with the following conditions may be accepted and analysed; however, the accuracy of the results cannot be guaranteed, and a repeat analysis will be recommended:

- Abstinence period does not meet recommended guidelines.
- Sample delivered more than 40 minutes after collection, preventing analysis within the optimal 60-minute window post-ejaculation.

In cases where the sample is accepted for analysis but there is any deviation away from normal semen parameters, which may have been caused by the failure of the patient to follow the instructions provided, the report will indicate that a repeat analysis is recommended. It is up to the referring clinician to action a referral for this repeat analysis.

10 DIAGNOSTIC SEMEN ANALYSIS

We use the computer-Aided Sperm Analysis (CASA) software and manual assessment when necessary to conduct the analysis. The report will include macroscopic and microscopic observations of the following parameters.

Parameter	Explanation
Appearance	The visual appearance of the ejaculate e.g. cream-grey, Clear-colourless etc.
Semen volume	The volume of the ejaculate (ml).
Liquefaction	Time taken for the sample to liquefy and be a homogenous fluid mixture. This is usually between 15-30minutes.
pH	The pH of the ejaculate is measured to assess the function of the seminal vesicles.
Viscosity	The consistency of the ejaculate e.g., viscous, or non-viscous.
Presence of agglutination	An assessment of any motile sperm sticking to each other by their heads, tails, or mid-pieces. The degree of agglutination is reported. Please see page 22 of the WHO 2021 manual for further information WHO laboratory manual for the examination and processing of human semen
Presence of Aggregation	Sperm aggregation is non-specific. An assessment of adherence either of immotile spermatozoa to each other or of motile spermatozoa to mucous strands or debris. The degree is reported as none, some, or many as suggested in the WHO 2021 Semen analysis template. Please see pages 21 and 234 of the WHO laboratory manual for the examination and processing of human semen.
Presence of non-sperm cells	This assessment measures the number of non-sperm cells in the ejaculate. Note that no distinction is made between immature germ cells, and leukocytes which are generally called round cells. The concentration of non-sperm cells is reported as an estimated number of millions of cells per millilitre of ejaculate. Please see page 22 of the WHO

	laboratory manual for the examination and processing of human semen
Debris	This is the assessment of the presence of non-cellular particulate matter in the semen sample, which may include fragments of cells, mucus, or other organic and inorganic material. It is reported on a semi-quantitative scale and graded as a percentage in proportion to the number of sperm cells represented as pluses.
Sperm concentration	The number of sperm per ml of ejaculate (millions/ml).
Total sperm number	The total number of sperm in the whole ejaculate (millions).
Sperm motility	The motility of sperm (at 37°C) expressed as the percentage of rapidly progressive, slowly progressive, non-progressive and immotile sperm
Sperm morphology	Percentage of sperm with 'ideal' morphology. *SpermBlue morphology staining method currently used is not prescribed by the WHO. *

The time of analysis is documented in the report and indicates the duration (in hour: minutes format) from the onset of macroscopic analysis of the semen sample following ejaculation and receipt of sample. The method of analysis, either manual or CASA, will be recorded according to the characteristics of the sample.

10.1 Lower fifth percentile values

Data characterising the semen parameters of a reference population (from men in couples with a time to pregnancy of 12 months or less) have been updated since the previous edition of the WHO Manual in 2010, leading to a revision of the lower fifth percentile values (see table below). It is important to note that these percentiles do not delineate distinct thresholds between fertile and sub-fertile men; therefore, caution is required to prevent over-interpretation.

In a normal sample we will expect the following results:

Parameter	**Lower fifth percentile value (WHO 2021)**
Semen volume	1.4ml
Liquefaction	Complete after 30 minutes
Progressive motility (a+b)	30%
Total motility (a+b+c)	42%
Sperm concentration	16 (million per ml)
Total sperm number (this takes preference over sperm concentration)	39 (million per ejaculate)
Ideal sperm morphology form	4%

10.2 Interpretive comments and terminology

Each diagnostic semen analysis report will include comments to help interpret the results. These comments are based on the 5th Edition of the WHO 2010 Manual for the Examination

and Processing of Human Semen, as well as the updated reporting guidelines from the 6th Edition of the WHO 2021 Manual. Below is an example of some common comments and their corresponding interpretations. This list is not exhaustive.

WHO 2010 comment	Recommended WHO 2021 comment	Interpretation
Normozoospermia	Semen sample parameters meet WHO 2021 criteria.	Total number of sperm, and percentages of progressively motile and morphologically ideal sperm is equal to or above the lower fifth percentile values.
Oligozoospermia	Reduced/ Low sperm count	Total number of sperm and concentration below the lower fifth percentile values.
Asthenozoospermia	Reduced progressive motility	Percentage of progressively motile sperm below the lower fifth percentile values.
Teratozoospermia	Atypical morphology	Percentage of morphologically normal sperm below the lower fifth percentile values.
Oligoasthenoteratozoospermia	Reduced/Low total number of sperm, reduced progressive motility, and atypical morphology	Total number of sperm, and percentages of both progressively motile and morphologically normal sperm, below the lower fifth percentile values.
Oligoasthenozoospermia	Reduced/Low total number of sperm and reduced progressive motility.	Total number of sperm, and percentage of progressively motile sperm below the lower fifth percentile values
Oligoteratozoospermia	Reduced/Low total number of sperm and atypical morphology	Total number of sperm, and percentage of morphologically normal sperm, below the lower fifth percentile values
Asthenoteratozoospermia	Reduced progressive motility and atypical morphology	Percentages of both progressively motile and morphologically normal sperm below the lower fifth percentile values
Cryptozoospermia	Sperm cells only seen post centrifugation	Sperm not apparent in neat sample but observed in a centrifuged pellet.
Azoospermia	No sperm seen pre and post centrifugation	Complete absence of spermatozoa in the ejaculate

For a detailed interpretation and significance of semen analysis results, the Andrology Laboratory recommends referring to section 8.1 (page 211) of the WHO laboratory manual for the examination and processing of human semen

10.3 Suggested clinical decision tree.

Recommendations:
If the first semen analysis shows a reduction in some semen parameters, this test should be repeated within two to four weeks for gross spermatozoa deficiency or after 3 months for general atypical findings as suggested by the 2023 infertility management NICE guidelines.
Note: A *new referral will have to be made on E-RS on each occasion by the GP.*

Priority repeat – patient should have a repeat semen analysis within **two to four weeks** for the following.

- Oligozoospermia with total sperm number <7M
- Apparent azoospermia.

Patient should have a repeat semen analysis within **3 months** for the following.

- Asthenozoospermia with progressive motility <10%
- Teratozoospermia with 0 or 1% normal forms
- Cryptozoospermia

10.4 Probability of undetected spermatozoa

According to the WHO Laboratory Manual for The Examination and Processing of Human Semen, 2021, if no spermatozoa are detected in wet preparations or following centrifugation, there is a low probability that spermatozoa may still exist in the ejaculate, depending on its total volume. For more details and probability estimation please refer to WHO laboratory manual for the examination and processing of human semen.

11 POST VASECTOMY SEMEN ANALYSIS

The Andrology Laboratory follows the 2016 Laboratory guidelines for post-vasectomy semen analysis: Association of Biomedical Andrologists, the British Andrology Society and the British Association of Urological Surgeons (P Hancock, BJ Woodward, A Muneer, JC Kirkman-Brown 2016, http://jcp.bmj.com/content/early/2016/04/15/jclinpath-2016-203731).

A post-vasectomy semen analysis will report any observations made on a 100µm large volume fixed depth slide. This is in accordance with the 2016 PVSA special clearance guidelines and has been shown to be a reliable method for PVSA to determine whether the number of sperm in a sample is above or below 100,000 sperm per ml.

If motile sperm are present in the ejaculate, analysis will continue as a diagnostic semen analysis. If a low number of immotile sperm are observed, this number will be included in the report (in relation to 100,000 sperm per ml) to assist clinicians in giving ‘special clearance’. It is left to the clinical judgement of the referring clinician to decide whether this should be granted. Clinical advice can be provided on request.

There are cases where the sample is not suitable for deep chamber analysis, including samples that are non-homogenous, samples that are highly viscous or have a high level of background debris, the coverslip and centrifugation method is used. This method will be stated on the report.

DIAGNOSTIC SEMEN ANALYSIS
GP USER MANUAL

11.1 Post-Vasectomy Interpretive comments

Below are some examples of comments that may be included on a post vasectomy semen analysis report. This list is not exhaustive.

Large volume fixed depth slide
No sperm seen on 25µl large volume deep chamber slide.
x non-motile sperm seen on 25µl large volume deep chamber slide. This equates to approx. x non-motile sperm per ml.
>200 non-motile sperm seen on 25µl large volume deep chamber slide. No motile sperm observed. Concentration then performed. Concentration of approximately x M/ml determined.

For more detailed interpretation and significance of results reported in a semen analysis, the Andrology Laboratory recommends referring to the 2016 Laboratory guidelines for post vasectomy semen analysis: Association of Biomedical Andrologists, the British Andrology Society and the British Association of Urological Surgeons. https://pubmed.ncbi.nlm.nih.gov/27083211/

11.2 Suggested clinical decision tree

Interpretive comment	HUH Suggestion
'No sperm seen on 25µl large volume deep chamber slide.	Patient can be given clearance provided recommendation 5 of the 2016 Laboratory guidelines for post vasectomy semen analysis is met. '**Recommendation 5: Assessment of a single sample is acceptable to confirm vasectomy success if all recommendations and laboratory methodology are met and no sperm are observed. Clearance can then be given.'**
'x non-motile sperm seen on 25µl large volume deep chamber slide. This equates to approx. x non-motile sperm per ml.	Patient can be given clearance provided recommendation 6 of the 2016 Laboratory guidelines for post vasectomy semen analysis is met. **Recommendation 6: The level for special clearance should be <100,000/ml non-motile sperm. Special clearance cannot be provided if any motile sperm are observed and should only be given after assessment of two samples in full accordance with these guidelines.**
>200 non-motile sperm seen on 25µl large volume deep chamber slide. No motile sperm observed. Concentration then performed. Concentration of approximately x M/ml determined.	If concentration of x is >0.1, clearance is recommended not to be given as per 2016 Laboratory guidelines for post vasectomy semen analysis.
Motile sperm observed in sample.	If any level of motile sperm is observed, clearance is recommended not to be given as per 2016 Laboratory guidelines for post vasectomy semen analysis.

12 REPORTING OF RESULTS

A 'Semen Analysis Report' is generated by the Andrology laboratory of the Homerton Fertility 'IDEAS' database and sent via a secure email by the fertility admin within 10 working days of the sample receipt date.

KINDLY BE ADVISED VERBAL RESULTS WILL NOT BE GIVEN OUT UNDER ANY CIRCUMSTANCES.

12.1 TURNAROUND TIME

The Andrology Laboratory endeavours to return results within 10 working days of patient attendance.

13 ADVICE, COMMENTS, FEEDBACK AND COMPLAINTS

Clinical advice on any aspect of the diagnostic services provided by the Andrology Laboratory can be obtained by sending e-mail enquiry to the huh-tr.fertility.unit@nhs.net. The Andrology Laboratory welcomes feedback from their patients and service users and shall be analysed and used to improve laboratory activities and services to users.

13.1 Patients

Patients attending for semen analysis appointments are sent a friends and family survey after their appointment.

13.2 Referrers

Clinicians that refer into our services are encouraged to provide feedback via email to the general fertility email address.

13.3 Complaints

Complaints should be directed to the Laboratory Director at the Homerton Fertility Centre. This should be done using the email addresses provided above.