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1. INTRODUCTION

1.1 Background to the guidance

Liquid based cytology (LBC) technology has been in use since 1996 in the USA and parts of Europe. Now, following the successful completion of the implementation pilots in England, the National Institute for Clinical Excellence (NICE) has recommended that LBC techniques are introduced across the NHS Cervical Screening Programme (NHSCSP).\(^1\) The implementation is part of the wider modernisation of the NHSCSP and will take up to five years to complete. It is a challenge for all cytopathology laboratories and will have a major effect on their working practices. The following advice incorporates experience gained during the UK implementation pilots and is based on guidance used in the Scottish and English LBC pilot laboratories. This has subsequently been supported in the LBC evaluation report on the English implementation pilot.\(^2\)

This interim advice is produced to assist laboratories in the introduction of LBC into their routine cervical screening practice. It will be revised as wider experience becomes available.

1.2 LBC technologies

1.2.1 Technical requirements

Advice on the technical requirements for LBC systems can be found in LBC Implementation Guide No 1.\(^3\) This is based on the two LBC technologies evaluated as part of the LBC pilots in England. These are the SurePath™ LBC system supplied by Medical Solutions PLC and the ThinPrep® system supplied by Cytyc UK Ltd.

1.2.2 SurePath

When using the SurePath system, the cervical sample is collected using a plastic broom device and placed into a vial of preservative fluid (CytoRich). The head of the device is detached and left in the vial, which is then capped and transported to the laboratory. Once in the laboratory, the vials are vortex mixed to resuspend sedimented cells and an aliquot is placed into a centrifuge vial using the PrepMate device. The aliquot is treated through a density gradient centrifugation process to remove unwanted materials and a concentrated pellet of cervical cells is produced. Next, the pellet is resuspended and the PrepStain slide processor transfers an aliquot to a settling chamber mounted on a microscope slide. Cells sediment onto the slide to form a thin layer and the excess fluid and cells are discarded. The slides are routinely stained as part of the automated process.

1.2.3 ThinPrep

The head of the sampler is rinsed thoroughly into a vial of preservative fluid (PreservCyt) rather than detached. Vials are then sent to the laboratory and loaded onto the instrument, individually in the case of the ThinPrep 2000 processor or in batches on the larger ThinPrep 3000 processor. The first step in the processing of the suspension is a gentle dispersion step, which thoroughly mixes the sample. Negative pressure pulses then draw the suspension through a TransCyt filter in order to produce a thin layer of cervical cells for interpretation. The rate of the flow is monitored to ensure that the amount of material collected is appropriate. The cellular material is then transferred from the filter to a glass slide and fixed. Staining is performed in the same way as the laboratory would stain a conventionally spread smear.
1.3 Commissioning requirements

1.3.1 Building requirements
Neither the SurePath system nor the ThinPrep systems require stringent building requirements, but the suitability of the area planned for the system should be discussed and agreed with the supplier before installation. Experience from the English pilot sites suggests that it is better to have a dedicated area for the LBC preparation equipment, particularly if using the SurePath system since there are a number of separate components. A full site survey should be carried out by the supplier prior to installation.

1.3.2 Site acceptance
Following installation, qualified personnel from the supplier must undertake a programme of system checks to ensure optimal performance of the device. The laboratory must have a protocol for checking that the system installed meets the technical requirements set out in LBC Implementation Guide No 1. The system must not be used for the preparation of clinical samples until the clinical head of the laboratory is satisfied that the technical requirements have been met and the system checks have been completed satisfactorily. This should be confirmed in writing to the supplier.

1.3.3 Operator training
At least two people per laboratory should be trained in the operation of the instrument(s) in order to ensure adequate cover. Training should be undertaken according to the supplier’s specifications. The supplier will provide a full on-site training programme and a comprehensive operator manual and maintenance schedule. These should form part of the laboratory standard operating procedures (SOPs) and should be available for inspection as part of the laboratory accreditation process carried out by Clinical Pathology Accreditation (CPA) (UK) Ltd. At the end of training, an operator should be able to perform any ‘front end’ tasks on the samples, operate the device, perform the required routine systems checks, carry out routine maintenance procedures and undertake minor trouble shooting tasks in line with the operator manual. The company should certify satisfactory completion of operator training for each individual.

2. TRAINING IN THE SCREENING AND REPORTING OF LBC SLIDES

2.1 Role of the cytology training centres
The training centres involved in the English pilot sites will offer training to the staff of all NHSCSP approved cytology training centres as part of the first phase of LBC implementation. When all training centre staff have successfully completed their LBC training, there must be a six month period during which staff consolidate their skills by gaining experience in routine screening work. Following this, the training centre can then offer LBC implementation training to laboratory staff. While each training centre may offer training on both current systems, the trainers delivering system specific interpretive training must be currently undertaking routine screening work using the relevant system. Trainers
should not offer implementation training on systems which they employ outside routine screening, eg systems used primarily for colposcopy samples.

Should training centre staff convert between LBC systems, they must repeat the six month period to gain experience of the new preparations before offering LBC implementation training to laboratory staff.

Each training centre will be given a ‘starter kit’ of LBC slide sets for the purpose of training and testing along with advice on the composition of additional slide sets. Experienced staff from the pilot sites may be available to provide training assistance during the roll out of training to laboratory staff. The marking of training and test sets has been a major undertaking during the pilot training programme, and training centres are advised to consider how best to manage this. The NHSCSP approved cytopathology training centres are listed in Appendix 1.

2.2 Objectives of LBC implementation training

LBC implementation training is designed for staff (medical and scientific) already trained in conventional cytology, and should be regarded as an additional skill of already competent staff. The objectives of the implementation training are as follows:

- to understand the principles of LBC and how it may be integrated into a laboratory
- to learn how to assess LBC samples microscopically
- to achieve competency in LBC (system specific) microscopic evaluation.

All staff must comply with the appropriate NHSCSP requirements for training, certification, continuing education and internal quality control for conventional microscopic screening before undertaking LBC implementation training.

2.3 Organisation of LBC implementation training

LBC implementation training comprises three stages:

- an induction course at an NHSCSP approved cytology training centre
- a consolidation stage in the laboratory
- a performance review phase in the laboratory.

It is preferable for groups of staff of different grades from each laboratory to be trained together rather than on an individual basis, as this provides support and encourages discussion of cases. Where laboratories do not have multiheaded microscopes, it may be possible to borrow one from the training centre to cover the period of laboratory based training. This should be coordinated by the training centre. It is recommended that the departmental training officer be among the first group of staff to be trained in order that he/she can adequately support and supervise staff during the consolidation and performance review stages of their training. Ideally, planned leave should be avoided during the minimum training period.
2.4 LBC training requirements

2.4.1 LBC training log
Details of the LBC training requirements for non-medical laboratory staff are specified in the Liquid Based Cervical Cytopathology Training Log. Completion of the training log is mandatory for scientific staff. It forms part of the assessment for the NHSCSP Certificate of Completion in Liquid Based Cervical Cytopathology Training.

2.4.2 Induction course
The LBC induction course is designed for medical and scientific staff of all grades already trained in conventional cervical cytopathology. A suggested programme for the course is shown in Appendix 2. Staff must attend an induction course at an NHSCSP approved cytology training centre. It is recommended that medical staff from a laboratory attend an induction course with their scientific staff. It is preferable for each induction course to include staff from more than one laboratory to encourage discussion. The purpose of the induction course is to provide an introduction to LBC. The course covers the basics of LBC that are common to all available commercial LBC systems as well as system specific morphological training. Each course consists of a minimum 20 hour programme over three days, comprising lectures, workshops, test sets and discussion of cases on a multiheaded microscope. All participants will be allocated a personal identification number that will be used throughout their training. The training centre director will provide the scientific cytology lead in each laboratory with a written assessment of the performance of scientific staff at the end of the induction course.

2.4.3 Consolidation stage
Following completion of an induction course, medical and scientific staff must undertake an in-house consolidation stage. This lasts up to four weeks, during which participants are expected to fully screen a total of 200 unmarked LBC slides compiled into 10 training sets. These training sets are system specific so that each individual will perform this part of the training on the system that their laboratory will employ. The training sets will include inadequate samples, negative samples, samples with borderline nuclear changes and samples with all grades of dyskaryosis, microorganisms, hormonal effects and other important morphological changes. Each participant must record his/her own opinion of each slide and be blinded to any previous opinions. Care must be taken to ensure that the slides passed to another participant are unmarked. The response sheets for each participant will be marked by the training centre in accordance with the marking scheme set out in the LBC training log.

2.4.4 LBC interim test
At the end of the consolidation stage, the primary screening sensitivity of each individual is calculated by the training centre. If this meets the national target of 95% or above for moderate dyskaryosis and above, then the individual may take the LBC interim test. This comprises a set of 20 unmarked slides of both negative and abnormal cases. Individual (anonymised) response sheets must be submitted to the training course director for marking. Individuals are required to achieve a score of 80% or more before moving to the final stage of training. Individuals who do not pass the LBC interim test are expected to identify themselves to their medical or scientific head of department. Performance will be reviewed regularly and further training designed by the training centre on an individual basis. An individual cannot move on to the next stage of training until successful completion of the LBC interim test.
2.4.5 Performance review
The performance review phase forms the final element of practical training. During this phase, staff will gain further experience and build confidence by screening a further 200 system specific LBC samples, with additional batches of 200 slides being screened as necessary to achieve the level of sensitivity required. These slides will be selected to reflect routine laboratory experience as far as possible, but will be seeded with sufficient abnormal cases to allow calculation of sensitivity. Individuals should aim to complete this stage within a four week period. Again, care must be taken to ensure that the slides passed to another participant are unmarked, and each participant must record their own opinion of each slide and be blinded to any previous opinions. Response sheets will be marked by the training centre.

2.4.6 Discordant opinions
Discordant opinions on consolidation and performance review slides must be reviewed with the individual concerned by a member of the training centre staff using a discussion microscope.

2.5 Completion of training for scientific staff
Primary screening sensitivity for moderate dyskaryosis or above must be calculated. If the individual continues to screen conventional smears these should be recorded and analysed separately. By the end of the LBC training period, the individual’s primary screening sensitivity for moderate dyskaryosis or above must be 95% or above for the most recent 400 LBC samples screened (ie initially the 200 consolidation slides and the 200 performance review slides). Training will be deemed complete when an individual has assessed a minimum of 550 LBC cases (ie 150 induction course slides, 200 consolidation slides and 200 performance review slides) and has achieved a primary screening sensitivity of 95% or above for moderate dyskaryosis and above. He/she may then submit their training log and an application for the NHSCSP Certificate of Completion in Liquid Based Cervical Cytopathology Training. Scientific staff (cytology screeners and biomedical scientists) must undertake the training programme and acquire the certificate of completion before issuing negative and inadequate results on LBC samples.

2.6 Completion of training for medical staff and advanced practitioners
Medical staff are expected to attend an LBC induction course and complete the consolidation stage of the LBC training programme before taking the LBC interim test. Medical staff who achieve the pass mark are deemed to have completed their training and will be issued with an NHSCSP Certificate of Completion in Liquid Based Cervical Cytopathology Training. Advanced biomedical scientist practitioners (ABMSPs) undertaking primary screening or checking roles should complete all three stages of training (induction course, consolidation stage and performance review) and acquire the NHSCSP Certificate of Completion in Liquid Based Cervical Cytopathology Training before issuing results on LBC samples. ABMSPs who perform neither primary screening nor checking will only be required to complete the induction course and consolidation stage, and to pass the LBC interim test. As with conventional microscopic screening, only medical staff and ABMSPs may issue borderline or abnormal results.
2.7 Conversion course

All staff switching from one LBC system to another must undertake an LBC conversion course organised by an NHSCSP approved cytology training centre. Details of the conversion course are given in the LBC training log. The course lasts for one day and is a minimum of seven hours. Course participants are encouraged to take part in a slide self-assessment exercise in their own laboratory.

2.8 Trainee scientific staff

Approved cytology training centres and laboratories should give priority for LBC training to staff who are already qualified and signing out negative and inadequate smears. Laboratory training officers should discuss with their cytology training centre manager the optimal time during their training for current trainee screeners to convert to LBC. The decision on whether current trainees complete their training in conventional cytology and then undertake LBC implementation training, or whether trainees switch to training in LBC part way through their training, should be made on an individual basis, taking into account length of experience, expected date of sitting the examination for the NHSCSP Certificate of Completion in Liquid Based Cervical Cytopathology Training and other local factors. If a trainee chooses to sit the LBC version of the examination, they must screen at least 1000 system specific LBC preparations. The National Cervical Cytopathology Education and Training Committee (NCCETC) has agreed that LBC slides will be available on request for the screening component of the examination for appropriate candidates. Any new trainee staff appointed should be introduced to LBC slides at the beginning of their training and, in the future, Introductory Courses in Cervical Cytopathology will train new staff in LBC.

3. WORKING PRACTICES

3.1 Stock control

It is envisaged that standing orders for consumables will be set up with the supplier. The frequency of delivery will depend on the volume of work, available storage space and shelf life. Laboratories may wish to review the distribution of consumables to sample takers with local primary care trusts (PCTs) to ensure suitability for local requirements.

3.2 Transportation

The transportation of samples from GPs’ surgeries/clinics to laboratories in the pilot was via the hospital van collection system. Laboratories need to recognise the particular transport needs associated with LBC samples. LBC vials are suitable for postage in the normal postal service as long as they are appropriately packaged in compliance with packaging instruction 650 and the requirements of UN3373.
3.3 Laboratory protocols

3.3.1 Sample protocols
Laboratory protocols must be written by individuals actively involved in the use of LBC samples in the laboratory in accordance with guidance issued by CPA (UK) Ltd. Some protocols applied during the implementation phase may need to be amended once LBC samples form the majority of samples received by the laboratory.

3.3.2 Specimen reception
Vials should be accessioned to the laboratory in the usual manner. A laboratory accession number should be applied to both the request form and vial, and a check made that these correspond. Cell suspensions that are clearly unsuitable for preparation (eg unlabelled, leaking container, scanty fluid or empty) should be carefully documented and dealt with in accordance with laboratory SOPs for cytology samples. If vials are received unlabelled or mislabelled, they should be returned to the sender and an appropriate record kept by the laboratory. It is the responsibility of the sender to either amend the details or repeat the test as appropriate.

3.3.3 Slide labelling
Where suitable slide labels are not automatically generated, the patient’s name and laboratory accession number should be written in pencil on the frosted end of the slide immediately prior to producing the preparation. Prelabelling of slides in batches should be avoided. A check of the patient’s name and slide number should be made at the time of applying the laboratory slide label. Any discrepancy detected during the stages of processing should be documented according to laboratory SOPs.

SurePath specimens Adequate labelling of the SurePath specimen is necessary throughout the slide preparation process. Aliquots of the original vial sample are placed in centrifuge tubes, processed and loaded onto the PrepStain slide processor to produce the slide. Centrifuge tubes and slides should be labelled using the labelled vials as a template, necessitating the production of four labels per sample.

ThinPrep specimens The T3000 instrument will automatically print the appropriate number onto the slide it produces from the vial. Experience has shown that the T3000 instrument may not be compatible with existing barcode systems used in cytopathology laboratories. Since the barcode is the only means of identification between the vial sample and slide, laboratories will need to ensure that compatible systems are in place. With the T2000 instrument it is necessary to manually label each slide during preparation.

It may be necessary to top up a ThinPrep vial with solution if a further preparation needs to be made from any vial.

3.3.4 Processor operation and slide preparation
There should always be at least one member of staff who is fully trained in processor operation available in the laboratory during each working day. Slides should be prepared according to the laboratory protocol.
3.3.5 Staining, coverslipping and mounting
The SurePath system stains slides on the machine. For laboratories using ThinPrep, laboratory staining protocols must be reviewed and where appropriate modified to obtain optimal results for LBC samples. There may also be a short period during change over when staining protocols will need to be modified to ensure adequate staining with both LBC and conventional preparations. Care must be taken when coverslipping to ensure that all the circle of cell deposition is within the edges of the coverslip and that drift is minimised. The thin layer facilitates the use of automated coverslipping instruments.

3.3.6 Retention and disposal of residual samples after slide preparation
The residual sample should be kept until a report has been issued. Storage should be at room temperature in racks not more than 50 trays high. Disposal of residual waste material should be carried out according to locally agreed clinical waste disposal protocols, or by an appointed agent who holds the necessary waste disposal licences.

3.3.7 Slide archive
One slide from all LBC samples should be kept for 10 years. If more than one slide has been prepared from a sample for diagnostic purposes, each slide should be stored for the 10 year period.

3.4 Samples unsuitable for slide preparation

3.4.1 Insufficient fluid in the vial
This may be due to a leaking sample vial caused by inadequate sealing. The importance of sealing the vial correctly should be included as part of training for sample takers. Careful alignment of torque lines on the ThinPrep LBC vials will facilitate this. However, overtightening of the lid should be avoided as this may impair T3000 functioning.

3.4.2 SurePath vials without sampler head
The process for taking an LBC smear using the SurePath system involves the smear taker detaching the head of the broom type sampler into the pot of preservative fluid. When the head is not present, the sample may not have been taken or the whole or part of the sample may have been discarded with the sampler. In this situation, it is acceptable for the sample to be processed. However, anything other than an abnormal result should be reported as inadequate.

3.4.3 ThinPrep vials containing sampler head
When using the ThinPrep system, the head of the sampler should be rinsed into the preservative fluid and not detached into the vial. Where a sampler head is present in the vial, this may not have been rinsed in the fluid, causing cells to become fixed on the sampler head. It is acceptable for the sample to be processed. However, anything other than an abnormal result should be reported as inadequate.

3.5 Reporting protocols

3.5.1 Assessing specimen adequacy
Evidence is lacking in the area of the minimum acceptable cellularity of LBC specimens. This will be monitored with a view to providing guidance once sufficient data are available.
3.5.2 Endocervical cells
The current NHSCSP guidance on reporting the presence or absence of endocervical cells for conventional smears is still applicable for LBC preparations, and endocervical material is only required in the cytological follow-up of a previous glandular abnormality or after treatment for an endocervical abnormality.

3.6 Recommended protocols for clinical management

The recommendations for clinical management and follow-up are the same for LBC samples as for conventional smears.

4. QUALITY STANDARDS

4.1 Introduction of LBC

The introduction of LBC into a laboratory will have a major effect on working practices in that laboratory. Initially, conventional smears will constitute the majority of the workload. As laboratory staff and local smear takers are trained in LBC, the numbers of conventional smears will reduce significantly, but it is essential that an appropriate number of staff continue to maintain their skills for screening conventional smears. As yet there are no evidence based quality standards within the UK for liquid based cytology. In the interim, those for conventional smears should be applied to LBC samples, with the exception of the maximum number of slides primary screened per annum (see section 4.2). The standards will be reviewed as evidence becomes available.

4.2 Workload

In any 24 hour period, up to five hours may be spent on cervical cytology microscopy (primary screening or rapid screening), whether assessing LBC or conventional smears. No more than two hours should be worked in continuous screening without a break of at least 20 minutes, ideally away from the screening room. Regular microbreaks of several seconds should be taken every 10–15 minutes. The other duties required of screeners can act as breaks from microscopy. Once a laboratory is fully converted to LBC, it is anticipated that screeners undertaking primary screening may assess up to 12 000 LBC smears per annum. However, these figures will be reassessed on the basis of the first year’s activity and regularly thereafter to ensure that they reflect work place experience.

4.3 Internal quality control

Internal quality control remains an essential component of laboratory quality assurance, whether the laboratory is handling conventional smears, LBC slides or both. It is crucial that laboratories maintain the full range of internal quality control (IQC) procedures during the introduction of LBC and that appropriate modifications and/or additions are implemented before LBC is used for the reporting of clinical samples. These should be documented in SOPs according to CPA (UK) Ltd guidance. All staff must be advised of modifications to existing quality control SOPs and be fully familiar with new ones.
4.3.1 Quality control of laboratory processes and procedures

Slide labelling Protocols must be established to ensure the correct labelling of slides prepared from the vials. Quality control checks should be part of daily routines, and all should be documented.

Slide staining The quality of slide staining should be checked daily and the results recorded.

4.3.2 Internal quality control of primary screening

Rapid screening remains the method of choice for internal quality control of primary screening. All slides assessed as negative or inadequate at primary screen should be subjected to rapid screening by a different member of staff who must hold the NHSCSP Certificate of Completion in Liquid Based Cervical Cytopathology Training.

4.4 External quality assessment

4.4.1 Assessment of technical preparation

The NHSCSP is currently introducing a national external quality assessment scheme for the evaluation of Papanicolaou staining in cervical cytology (technical EQA).

4.4.2 Interpretative assessment (proficiency testing)

Regional QA teams will run separate conventional and LBC (system specific) EQA schemes in gynaecological cytopathology. Virtual regions may need to be established to ensure that sufficient participant numbers can be achieved during the period of LBC implementation. Participants continuing to report conventional smears for a period of time while their laboratory converts to LBC may need to participate in a conventional and an LBC scheme.

4.5 External monitoring during the implementation period

Laboratories will be expected to work closely with their QA teams throughout the implementation period to ensure that quality standards are maintained.
REFERENCES

5. Liquid Based Cervical Cytopathology Training Log. Implementation Training for Qualified Non-medical Laboratory Staff Working in the UK Cervical Screening Programmes. NHS Cancer Screening Programmes, 2003 (available to course participants at NHSCSP approved cytology training centres).
APPENDIX 1: NHSCSP APPROVED TRAINING CENTRES

Birmingham Cytology Training Centre
Cytology Department
Birmingham Women’s Hospital
Metchley Park Road
Edgbaston
Birmingham B15 2TG
Ms Linda Grosvenor 0121 627 2722
Dr Chris Waddell (Director)

South West Regional Cytology Training Centre
Cellular Pathology
Southmead Hospital
Westbury-on-Trym
Bristol BS10 5NB
Mrs Helen Burrell 0117 959 5649
Dr Karin Denton (Director)

South West Thames Cytology Training Centre
Pathology Department
Royal Surrey County Hospital
Egerton Road
Guildford GU2 5XX
Mrs Barbara Sayer 01483 571122
Dr Louise Daborn (Director)

Northern & Yorkshire Cytology Training Centre
Cytopathology Department
United Leeds Teaching Hospitals NHS Trust
Britannia House
Britannia Road
Morley
Leeds SL27 0BT
Mrs Diana Mera 0113 392 7836
Dr Jonathan Sutton (Director)

Liverpool Cytology Training Centre
Pathology Department
6th Floor, Duncan Building
Royal Liverpool University Hospitals NHS Trust
Daulby Street
Liverpool L69 3GA
Mrs Irene Turner 0151 706 4580
Dr Lesley Turnbull (Director)
Advice for Cytopathology Laboratories on the Implementation of Liquid Based Cytology

Manchester Cytology Training Centre
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Mrs Jenny Davies 0161 276 5114
Dr Mina Desai (Director)

London Regional Cytology Training Centre
Cellular Pathology
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Harrow
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Mr David Smith 0208 869 3314
Dr Tanya Levine (Director)

Sheffield Cytology Training Centre
Cytology Department
Royal Hallamshire Hospital
Sheffield S10 2JF
Mr Nick Dudding 0114 271 2538
Dr John Smith (Director)

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Box 235
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Hills Road
Cambridge CB2 2QQ
Mr Ian Munro 01223 216749
Dr Anne Warren (Director)
APPENDIX 2: SUGGESTED PROGRAMME FOR THE INDUCTION COURSE

Day 1
Introduction
Basic principles of LBC
(cover all available commercial systems)
The LBC training programme
Using the training log

Lecture
Normal cytology and infections

Individual microscopy
Normal cytology and infections
Test set 1 – 20 known LBC cases

Multihead review session
Test set 1

Lecture
Squamous and glandular dyskaryosis

Individual microscopy
Squamous and glandular dyskaryosis
Test set 2 – 20 known LBC cases

Multihead review session
Test set 2

Demonstration of inadequate and difficult dyskaryotic cases (20 cases)

Day 2
Individual microscopy
Test set 3 (20 cases)

Multihead review session
Test set 3

Individual microscopy
Test set 4 (20 cases)

Multihead review session
Test set 4

Rapid screening technique
(20 cases)

Day 3
Individual microscopy
Test set 5 (20 cases)

Multihead review session
Test set 5

Individual microscopy
Test set 6 (20 cases)

Multihead review session
Test set 6

General discussion and review of course

Optional – question and answer session with representative(s) from commercial company(ies)