Remote intra-operative diagnosis in neurosurgery

The introduction of digital slide scanning for remote reporting has restored an important service for patient healthcare at a Scottish hospital

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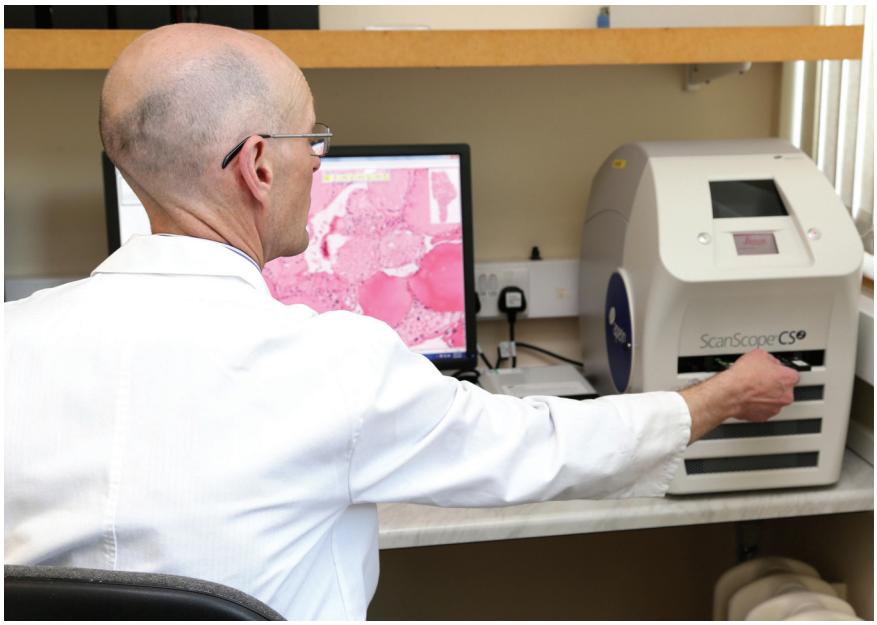


Telepathology is the process of digitising histological images for transmission electronically to remote centres, often for the purposes of diagnosis. Some telepathology systems employ robotic microscopes, which can be controlled remotely in order to select microscopic fields and to adjust magnification and focussing. This type of system may suffer from delays due to slowness and lags in the robotics.[1] The Aperio system from Leica Biosystems described in this report uses Whole Slide Imaging (WSI) to produce a complete digital representation of the tissue. The images produced by WSI are stored on a local server, which can be accessed by remote users who have been granted the required access level. The images are viewed at the remote site on a computer screen, with the remote user having complete control of selection of fields across the entire slide. In a comparison of WSI with conventional microscopic interpretations of frozen sections of ovary, Fallon et al concluded that WSI is accurate, reproducible and reliable for remote reporting.[2]

Frozen section is a well-established procedure for the intra-operative reporting of urgent biopsies. These cases are treated urgently with the aim of producing stained sections as quickly as possible so that a report may be relayed to the surgeon while the patient is still in theatre. In neuropathology, it is also common for fresh smears to be produced instead of, or as well as, frozen sections. In some centres, 'touch' preparations of fresh neurosurgical specimens have also been used.[1] Fresh neurosurgical tissue, including some types of tumour, is usually soft and amenable to the production of smears quickly and easily, providing, in most cases, relatively flat layers of cells. Some specimens however, are more fibrous, for example, Schwannomas, and might be less suitable for smear preparations.[3] Despite this, it was also reported that the thickness of smears does not impose an impediment to diagnosis, as many thinner areas of the slide will be available for study.[3] Promptly fixed and stained smears usually provide excellent cellular and nuclear detail that is probably superior to that of frozen sections, although the latter have the advantage of retaining the architectural detail of the tissue. Also, although frozen sections are usually

suitable for a wide range of neurosurgical tissues, they are more time-consuming to prepare compared with smears. In our department, both frozen sections and smears are usually prepared to give a 'best of both worlds' protocol, but this may vary according to several factors including the clinical details, and the type and quantity of specimen received. Also for consideration is the availability of follow-on formalin-fixed tissue, which is used to prepare higher quality formalin-fixed, paraffin-embedded sections at a later time.

Implementing the Aperio CS2 system at Aberdeen Royal Infirmary

The loss of the neuropathology post from the pathology department at Aberdeen Royal Infirmary resulted in the suspension of the intra-operative reporting of neurosurgical biopsies. The Aperio CS2 system was installed in early 2014, with the primary aim of re-establishing this important service. In conjunction with this, the neuropathology department at the Western General Hospital in Edinburgh agreed to include the remote reporting of urgent neurosurgical biopsies as part of their overall referral service. This article describes our experience of the Aperio system in its first year of use, during which time approximately 70 urgent neurosurgical cases have been reported.

The Aperio system consists of:

- Aperio CS2 scanner with five slide holder – with 20x objective, also capable of 40x magnification by way of a 'doubler' mechanism
- PC with Windows and Scanner software and eSlide Manager program
- Server unit for storage of scanned images, accessible by designated remote users
- Light source unit (Techniquip 21DC) – for the scanner microscope.

The equipment listed above is compact and fits easily on a bench less than two metres in length.

No special equipment is required at the remote site because images are accessed and viewed via a standard PC.

Software

- ScanScope – this provides a virtual image of the slide holder and loaded slides allowing the user to control the scanning process. Each loaded slide is colour-coded on the screen to signify its current status, for example, slides that are coloured green have been successfully scanned
- eSlide Manager – this includes an administration module, which allows the setting up of users, including local administrators and users at remote sites, with logins, passwords and access levels. Another module in eSlide Manager displays the scanned images by way of a list of 'thumbnail' images which can be selected for viewing, deletion, copying, etc
- ImageScope – this software program is used for viewing and manipulation of the scanned images

Comprehensive training on the Aperio system, including hands-on experience of the scanner and software, was delivered by Leica Biosystems to a small group of Biomedical Scientists (BMS). A BMS Aperio duty rota was produced to cover the urgent neurosurgical service. It was found that the entire intra-operative laboratory operation including sampling, preparation of slides, staining and scanning could be carried out by a BMS working alone, although for a more streamlined, efficient service, a group of two BMS might be preferable.

Communications

The intra-operative reporting is carried out by one of a team of neuropathologists on a rota basis.

The efficient operation of the service is reliant on good communications between the neurosurgical staff, pathology laboratory staff and the neuropathologists at the remote site. In most cases, an e-mail message from the neurosurgeon gives at least 24 hours notice of a frozen section request, although it is often difficult to accurately estimate the time of its arrival in the department on the day.

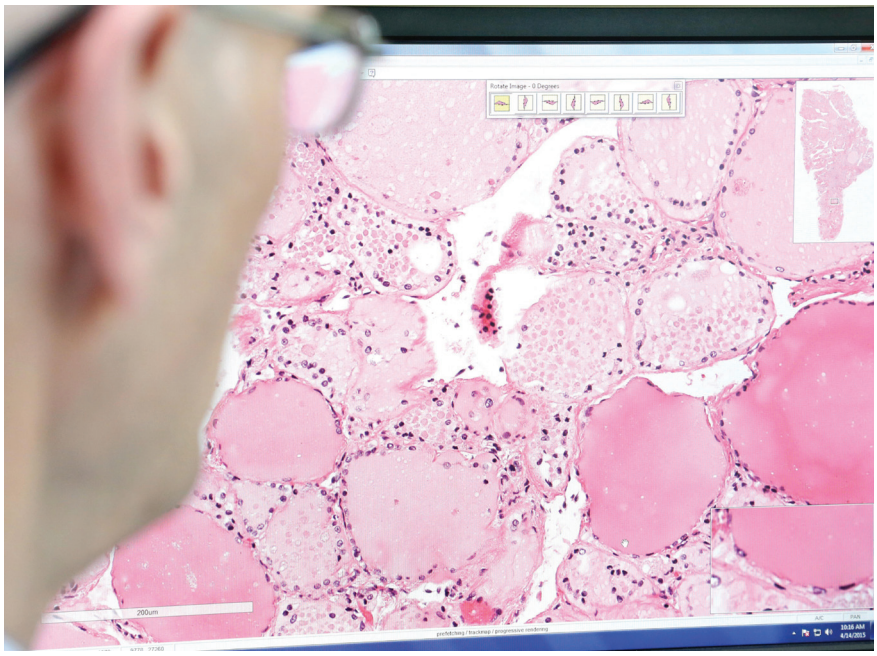
During the intra-operative procedure, telephone contact between the department and the remote neuropathologist is not continuous, but is made at appropriate stages, for example, when the specimen has newly arrived or when the first slide has been scanned and is available to access remotely. Likewise, the neuropathologist contacts the neurosurgeon with the findings and also lets the laboratory know when the procedure is concluded.

Laboratory intra-operative procedure

Specimen sampling and laboratory procedures are carried out following the guidelines and advice of the neuropathology team in Edinburgh.

On arrival of the urgent specimen, the scanner facility of the photocopier is used to send a copy of the accompanying request form, which includes the laboratory case number and clinical details.

Fresh samples can be very variable in size and consistency. The BMS is responsible for sampling of the tissue although it is important for the laboratory to refer any queries on any aspect of the case to the neuropathologist on duty. If tissue is sparse, it might be decided to produce smears only. In other cases, the neuropathologist may prefer to examine a frozen section in the first instance. Our usual routine is to produce two smears and one frozen section. One of the smears is stained by toluidine blue while the other smear and the frozen section are stained by haematoxylin and eosin (H&E). In order to provide good quality cytological staining, it is important that the smears and frozen sections are fixed instantaneously in 3% acetic alcohol fixative for at least 30 seconds. Pre-printed, bar-coded labels, displaying the laboratory case number and patient's surname, are attached to each stained slide. The label, which forms an integral part of the scanned slide, is turned 90° clockwise by the scanner software so that it appears the right way up on the 'thumbnail' of the scanned image. For the sake of urgency, the toluidine blue smear is usually prepared first of all, so that at least one scanned slide is available for examination at the referral centre as quickly as possible. The frozen section, which takes more time to prepare than the smears, can be cut and stained while the first slide or slides are being scanned. It is vital to ensure than there is no excess mountant on the surface of the slide as this can make contact with the objective lens of the scanner, which would need to be cleared of contamination before



resuming successful scanning.

For our purposes, the scanner objective is set at 20x magnification, although there is the option of 40x magnification by deploying the scanner's doubler mechanism. Obviously, the time required to scan each slide is dependent on the magnification and the area of stained tissue to be scanned – with the 20x objective in place, a large smear covering most of the slide may take perhaps four or five minutes to complete, whereas a small stereotactic frozen section, typically only a few mm in size, can be scanned in less than 30 seconds.

When the scanner has detected the area of tissue to be scanned, this area is indicated on screen by a green box. The scanner also sets focus points, which create a three-dimensional map of the tissue. The value of each focus point is based on the perceived depth of the tissue surface at that location. During scanning, the objective is adjusted automatically to take account of the set focus point values across the slide.

For most slides, it is possible to allow the scanner to detect the tissue and scan the slides automatically, although there is also the option to use the 'snapshot' facility, which allows greater user interaction, for example, the size of the green box around the detected tissue may be altered so as to pinpoint particular areas of interest or to preclude other areas. By reducing the size of the green box, the scanning time can be reduced greatly. Also, set focus points may be removed, or new focus points added to the detected tissue as required.

Conclusions

During its first year of use, the Aperio system has been found to be easy to use, providing excellent image quality, and requiring minimal calibration. For urgent intra-operative cases, we have found that it is possible to have at least one scanned slide ready to view within 15 minutes of receipt of the specimen. There was only one failure of the intra-operative procedure in the first year of use, but this was due to a network problem at the remote site rather than a hardware or software problem.

In the majority of cases, the automatic scanning mode is ideal, especially when scanning frozen sections, which provide a relatively flat surface and a clear outline on the glass. It is possible to load up to five slides at a time for automatic scanning, allowing the operator to attend to other duties.

The option of using the snapshot facility gives extra flexibility and the opportunity for greater user interaction in streamlining the scanning process. This is especially true when dealing with more difficult smears of firmer tissues, which may give uneven spreads with a less distinct outline. The snapshot also allows the user to be completely selective when choosing areas of interest, and the resultant smaller detection box can greatly reduce the scanning time; this is especially important when dealing with urgent samples.

Allowing for the extra time required for the scanning process, the reporting turnaround time should be only marginally greater than that of a conventional in-house, intra-operative case using a microscope.

For the neuropathologist, WSI gives complete control of the entire scanned slide, similar to conventional microscopy, but without the need for focussing, and allowing quick and easy selection and examination of fields.

Remote reporting obviates the inconvenience, expense and time required for a neuropathologist to travel to other centres. With good communications and a little pre-planning, the referring laboratory can ensure that the whole procedure has minimal impact on the reporting neuropathologist's normal daily working commitments.

From our laboratory's perspective, the introduction of scanning for remote reporting has been a necessary new technology, which has restored an important service for the benefit of patient healthcare. ✦

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