Breakthrough in automated tissue dissection for molecular pathology lab

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Analysis of tumour tissue for molecular biomarkers is a labour-intensive and time-consuming process, requiring precision at each of the many steps involved. This article discusses how recently launched technology is helping to improve sample selection and tissue dissection.

Globally, the incidence of cancer continues to rise. In part this can be attributed to lifestyle choices, pollution and an aging population. But greater precision and reproducibility in diagnostic testing has also seen more cancers being detected at an earlier stage – leading to improved outcomes for patients.

Advances in sequencing technology have led to better understanding of molecular mechanisms, leading to the expansion of targeted, personalized therapies [1–3]. These rely on the presence of molecular biomarkers in the patient's tumour tissue and they need to be detected with high sensitivity and specificity. This is pivotal when faced with advanced stage cancers where surgery is no longer an option, and targeted therapies are often the last line of defence.

Significantly for the pathologist, as next-generation sequencing (NGS) costs have reduced considerably, sequencing techniques have become more accessible for routine clinical applications.

Addressing gap in precision oncology

Further, the growth of artificial Intelligence (AI) is also playing an increasingly important role in improving disease management. Already integral to digital pathology, AI offers the promise of automated diagnostic algorithms for interpreting results, both in histopathology with image analysis and bioinformatics for molecular pathology.

AI-based algorithms could also help in the difficult task of ROI selection by the pathologist. However, the benefit of such a move has been potentially undermined by a missing link in the initial stages of the cancer diagnostic pathway.

While other processes have been fully automated and integrated into a lab's workflow, technological developments in the area of sample selection and dissection from the histopathology slide have lagged behind. No workable system has been deemed suitable for use in a hospital laboratory. The lack of innovation has seen them remain time-consuming, manual processes, relying on subjective analysis and prone to 'misestimation' and sub-optimal results [4]. The consequence had been to limit the quality, traceability and reliability of precision medicine [5]. Further, any realistic involvement of AI in the selection of ROI only makes sense if the dissection process itself is first automated. Novel technology that would address this missing link and enable all these initial processes (including sample scraping) to be automated has been long overdue.

Automating the entire process

This was mooted two years ago by CLI in an article exploring the call for an automated tissue dissector with built-in robotic scraper that was able to digitize the sample selection process and integrate it into routine clinical practice.

This has now become a reality with the launch this year in both Europe and the USA (research use only) of pioneering new technology for the histopathologist. Significantly, the solution automates and integrates the entire process - something no other system can offer. The histopathologist will have results that are more reproducible and precise than manual dissection. This is particularly important when handling either minute specimens or samples with very small tumour areas.

Research laboratories already use technologies like robotic laser capture microdissection. However, this is a relatively slow process, requires a great deal of expertise and cannot realistically be integrated into a hospital lab's routine workflow. Further, such a high level (<1 μm) of spatial resolution is unnecessary.

Compact footprint

An alternative automated tissue dissection solution already exists for larger industrial labs that require precision, alongside speed and higher throughput [Tissector High Throughput (HT) system, Xyall BV]. This combines automated tissue dissection technology with an accuracy of <0.1 mm, an 1800-slide capacity and a throughput of 80 dissection slides per hour. Significantly, the same company has just launched the first tabletop version (Tissector TableTop, Xyall BV), with a footprint that is compact enough to integrate into existing laboratory workspace, and able to run uninterrupted for more than two hours.

The system can hold 72 slides, with 12 case slots, dissecting 30 slides an hour with an accuracy of ≤0.1 mm. Cases can be

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Figure 1. The manual process of slide marking and dissection is automated by the Tissector TableTop

>> added without interrupting the operation and the number of dissection slides per case can be chosen freely. Quality reports are generated for each case, containing pre- and post-dissection images and quality metrics for full traceability.

Pathologists can make a selection of the region of interest by assessing digital whole slide images of the sample. The annotations of the regions of interest are then interactively created and automatically transferred to dissection slide images.

The new solution combines the required degree of precision, quality and reproducibility in a platform that offers full traceability so that steps can be digitally captured, stored and reassessed at any stage. The pathologist will no longer need to be on site to annotate the ROI, this can now be done remotely from wherever they are working. Another benefit is to provide speedy access to a specific dissection result. This is especially useful when the findings are being reconsidered or reviewed by a Molecular Tumour Board.

Integration into routine lab workflow

While molecular workflow remains a batch-driven system, any new automation solution still has to improve overall lab efficiency. Further, the decision to switch clinical practice to any automated system requires confidence that results will be robust and reproducible.

But any new solution cannot just offer improved quality. It must address the challenges set by higher performance targets, over-stretched resources and limited staff availability. Importantly, it should offer the potential to reduce long-term cost in terms of time, resources and workflow efficiency. Hands-on time must therefore significantly be reduced and less experienced staff should be able to safely use it.

Once the process is set in motion, staff must be able to walk away and direct their attention to other, added-value tasks. Further, it should be flexible enough to integrate into the existing routine workflow.

Compare this to what is still taking place in today's histopathology lab. Current practice is for the experienced histopathologist to have to mark the ROIs by hand on hematoxylin and eosin (H&E)-stained slides, using subjective analysis to determine the boundaries of the tumour area. This manual process becomes increasingly difficult with smaller regions of interest.

Removing error-prone, manual process

One of the current workflow bottlenecks is at the point of manually transferring the annotated area from the reference slide to unstained dissection slides, matching these by visual judgement. The dissection operation itself is often done with a scalpel, and the lab technician has to copy the annotation as accurately as possible. The cellular composition of the sample and therefore the reliability of the test results are once again based on this subjective, manual process.

The new technology will replace these processes, automatically collecting tissue material, operating continuously, with the risk of cross-contamination minimized by the automatic disposal of the scraping head. There is no need for liquids ensuring easier compliance with all molecular sample prep protocols.

Dangers of manual tissue dissection

Several studies have highlighted the limitations of manual tissue dissection. A 2022 study published in *Modern Pathology* highlighted deficiencies in the way 'misestimation may cause tissue waste and increased laboratory costs' [6]. The same study also indicated the benefit of combining automated tissue dissection with AI for accurately determining the parameters of ROIs whatever the clinical analysis.

The study noted the particular challenges pathologists face to achieve minimum DNA input requirements for NGS. Without any automation solution, they currently have to visually estimate the dissection areas and slide count decisions, while taking care

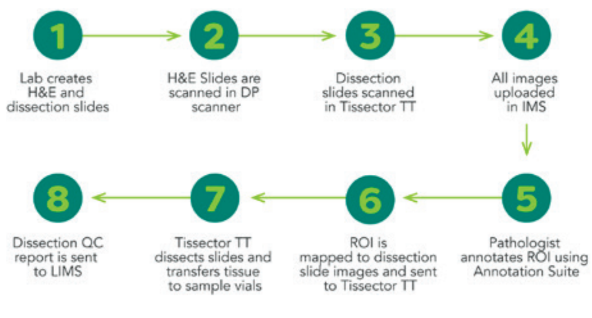

Figure 2. Flow chart showing automated tissue dissection within workflow
H&E, hematoxylin and eosin; DP, digital pathology; IMS, image management system; LIMS, laboratory information system; QC, quality control; ROI, region of interest; TT, TableTop.

not to recommend excessive dissection. Tissue stewardship guidelines help them to protect tumour tissue in case it is needed for further molecular tests – but they still have to rely on subjective interpretation.

The study further comments that 'using manual dissection techniques is difficult, and thus, there is an increasing need to optimize tissue extraction procedures as NGS becomes more relevant in clinical practice'. An algorithm in combination with an automated system like Tissector TableTop enables the dissection to preserve more tissue and avoid excessive dissection. An obvious development from this would be the addition of software that would flag up to the pathologist if an ROI needed further review.

Benefit of digitally guided dissection

Even before automated tissue dissection was on the horizon, the 2014 paper in *Modern Pathology* from scientists at the University of Utrecht Medical Centre crystalized the current challenge faced by pathologists in its title 'The estimation of tumour cell percentage for molecular testing by pathologists is not accurate'. The study aimed to determine the reliability of estimated tumour cell percentages using lung tissue samples.

Forty-seven H&E-stained slides were evaluated by nine pathologists, using categories of 0–5%, 6–10%, 11–20%, 21–30%, and so on, until 91–100%. The percentage of tumour cells was counted manually. On average, the range between the lowest and the highest estimate per sample was 6.3 categories. In 33% of estimates, the deviation from the gold standard was at least three categories.

The evidence for automation

Further anticipating a role for AI in the selection and characterization of regions of interest, the study noted the consequence of 20% of tumour cells being considered the lower limit for detecting a mutation. In this case, samples with an insufficient tumour cell percentage (<20%) would have been estimated to contain enough tumour cells in 27/72 (38%) observations, possibly causing false negative results and leading to 'misinterpretation of the test results'.

Evidence of the diagnostic benefits of automated tissue dissection can also be seen in a US Cancer Genetics study. Here, the authors performed and compared digitally guided dissection with traditional manual dissection in a series of pancreatic adenocarcinoma specimens to compare the effectiveness of both methods [7]. The researchers found that the KRAS mutant allele fraction and estimated neoplastic cell fraction were significantly higher in samples obtained from digitally guided dissection. In 7 out of 32 (22%) of the samples, a detectable mutation was found only with the digitally guided dissection.

Boosting patient outcomes

The quality of the sample and the precision of its dissection impacts many areas of precision diagnostics – from the clinical researcher needing to stratify patient cohorts in clinical trials, to the histopathologist determining ROIs. Automating tissue dissection as part of a digital workflow setting will be a significant building block for precision oncology.

A platform like the Tissector TableTop easily integrates into a lab's routine workflow, offering full traceability, quality control, documentation and consistency. It automates the tissue sample scraping by combining the imaging, and annotation transfer with the physical dissection of the selected areas – something that no other desktop system can achieve.

Delivering greater precision and reproducibility to routine workflow increases overall efficiency and turnaround time, while making better use of scarce resources and reducing hands-on time. Further, it would boost the quality of molecular diagnostic analysis while improving patient outcomes.

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