Continuous Throughput Rapid Tissue Processing Revolutionizes Histopathology Workflow

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Continuous throughput rapid tissue processing (CT-RTP) has the potential to introduce dramatic changes into the practice of modern histopathology, a field which has been relatively unchanged for many years. These changes can be grouped into 3 major categories: 1) where we work, 2) how we work, and 3) when we work.

The implementation of CT-RTP allows for rapid turn around of tissue biopsies, providing a standard hematoxylin and eosin stained slide, while preserving various macromolecules such as mRNA and proteins of interest for development of target directed therapies.

Despite initial resistance to these changes, we have successfully implemented CT-RTP by recognizing the necessary changes in our work patterns, including location, re-education in gross tissue sectioning techniques, fixation, and schedule adjustments in both technical and professional staffing.

Where We Work

Traditionally, pathologists have worked in offices located within a centralized pathology department separate from the histopathology laboratory, and when needed, would go to a small dedicated area near the operating room where a freezing microtome would be located. The need for rapid turnaround for transplantation patient biopsies, as well as for outpatients waiting in clinic reception areas for biopsy results, and in addition, the necessity for interacting directly with surgeons in immediate tissue

**Image 1** (A) Section of colonic carcinoma produced using the continuous throughput processing method, compared with (B) similar section processed by conventional methods (Hematoxylin and eosin stain, original magnification 200x).
procurement, all suggested the need for a tissue laboratory located
adjacent to the surgical suites. We have opened such a laboratory,
which also facilitates communications between surgeons and
pathologists. This laboratory contains materials and instruments
needed for frozen section examinations, as well as all the elements
of a routine histopathology laboratory (tissue processor, embed-
ding station, microtomes, staining, and coverslipping equipment);
special stains and other procedures are performed at another site.

Rapid fixation in a molecular friendly fixative has been shown
to protect macromolecules. A molecular friendly tissue fixative was
developed and has been shown to be useful in recovery of macro-
molecules such as long mRNA from paraffin embedded blocks,
obliterating the need for rapid freezing and frozen tissue storage.3

Prompt fixation is necessary for the most optimal recovery of
macromolecules. The on-site pathologist performs frozen sections
and also is available to go into the operating room to select tissue
for immediate fixation in molecular friendly fixative. Preliminary
work underway in our laboratory indicated that this same univer-
sal molecular fixative is also useful as a fixative for frozen section
slides, eliminating the formol-alcohol currently used in most
frozen section procedures.4

How We Work

Traditional histopathological techniques have evolved over
long periods, typically on an empirical basis. Current practices
include routine overnight fixation for large specimens, followed
by dissection and preparation of tissue slices up to a maximum of
about 3 mm in thickness. Smaller specimens may be dissected the
same day followed by overnight processing, and very small biop-
sies can be rush processed the same day they are received using
abbreviated protocols available on modern tissue processors. For
the most part, however, tissues are routinely fixed and processed
overnight, followed by manual embedding, sectioning, staining,
and coverslipping.

The use of 10% neutral buffered formalin as the fixative of
choice is a time-honored tradition in histopathology. Formalin is
well-recognized as an irritant and is also considered toxic, and re-
quirements for efficient ventilation and fume concentration moni-
toring add to the expense and complexity of the laboratory. The
development and use of a universal molecular fixative together with
the elimination of xylene and formalin from the tissue processor
precludes the need for atmospheric monitoring of these chemicals.

Rapid tissue processing in our laboratory requires thinner
tissue sections than those typically prepared by pathologists for
routine processing. For this reason, new dissecting boards have
been developed, consisting of a metal plate mounted on dissecting
board, with shallow wells or chambers in which partially trimmed
tissues are placed (Image 3). A special guide allows the insertion
of a blade, which when moved along the chamber results in a sec-
ton of uniform 1.5 mm thickness. A few ancillary tools to aid in
dissection have also been developed. These ideal thin sections are
then processed using the microwave based method.

When We Work

In traditional histopathology practice, there is an expecta-
tion of at least a 1 day (overnight) wait for pathologic diagnosis.
The schedules of pathologists, histotechnologists, and ancillary
personnel have evolved to accommodate this delay, but with CT-
RTP the vast majority of cases can be interpreted and reported
on the same day as the surgical procedure. This is especially use-
ful for small biopsies, including those from transplantation
cases5 in which immediate adjustments in therapy may follow, as
well as for clinic patients who may be waiting in a holding or
reception area. An example would be our breast disease center at
JMH/UM where diagnosis is provided within 3 hours of receipt
of the specimen. This rapid turnaround time allows for discussion and scheduling of follow-up visits before the patient leaves the facility. Since the inception of CT-RTP, we have dramatically reduced the turnaround time for surgical pathology specimens (Figure 1).

In most traditional pathology laboratories using overnight processing, pathologists typically receive the slides for interpretation early in the day, and technical staff having reported to work hours earlier to embed, section, and stain materials prepared the previous afternoon. Reports are generally available to physicians by the afternoon. CT-RTP dramatically changes this traditional work flow, since cases from the previous day have, for the most part, already been reported. Thus, histotechnologists typically begin working later during the day, and using a staggered arrival time, staff the laboratory throughout the day as cases arrive from the surgical suites. The process precludes “batching” of specimens and slides, as there is a continuous output of the final products (blocks and slides) throughout the day. Likewise the pathologists typically have few cases to review in the early morning hours, but as the day progresses, more cases become available for immediate interpretation, as shown in Figure 2. Since some cases inevitably arrive later in the afternoon, it follows that a pathologist needs to be available to interpret these “late” cases. Figure 3 illustrates the work patterns for histotechnologists prior to and after the inception of CT-RTP. The resultant schedule is more “family-friendly” and avoids the need for extremely early arrival times for technical staff. Pathologists cover the “late” cases on a rotational basis, thus assuring the same-day availability of results and improved patient management. An added advantage is that since cases are signed out late Friday afternoon, there is no need for Saturday morning histotechnologist and pathologist staffing.

All of the above-described changes in the pathology laboratory have been evolving as CT-RTP has been instituted. While the more immediate behavioral changes, including preparation of thin (1.5mm) sections and introduction of molecular friendly fixative, are more readily achieved, changes in work location, and also in the schedules of histotechnologists and pathologists, can be more difficult to implement. By our nature, we prefer to stay with what has served us well in the past, yet, as we enter the 21st century we know that we simply cannot continue business as usual. The evolution of modern histopathology practices must move forward as molecular analyses and target-directed therapies augment our reliance on traditional histopathologic diagnoses. CT-RTP has set the stage for this radical departure from reliance on morphology alone to a synthesis of histopathology, immunohistochemistry, and molecular analysis.

Figure 1. Turnaround times (TAT) at Jackson Memorial Hospital before (1996) and after (2003) the inception of CT-RTP.

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<tr>
<th>TAT Surgical Specimens</th>
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<tr>
<td>JMH</td>
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<td>1996 (23349 Cases)</td>
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<td>2003 (30124 Cases)</td>
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Figure 2. Graphic representation of pathologist sign-out activities with conventional overnight tissue processing and CT-RTP.

Figure 3. Schedule for histotechnologists before and after the inception of CT-RTP. In general the bulk of work is done during the middle of the work day as cases are received from surgeries.


