

Tissue-Tek SmartSection® The First Random Access, Fully Automated Robotic Microtome System That Provides Safe, Consistent Section Quality, Designed For 24 hr./7 Days Per Week Production

Introduction

Sectioning is the most important and challenging technical skill of Histotechnologists. Producing quality slides with consistent section thickness without tears, wrinkles and folds is critical for Pathologists to make a diagnosis and for image analysis to quantify. Traditionally, microtomy has been a manual process that can result in ergonomic and health challenges including Repetitive Motion Disorders (RMD).

Tissue-Tek SmartSection[®] (Sakura Finetek, Torrance, CA) is a robotic microtome system that was developed to fully automate sectioning of formalin-fixed paraffin -embedded (FFPE) blocks and produce mounted sections onto printed slides

Histotechnologists can



— Continuously load up to 20 blocks in 5 magazines (100 blocks)

– Continuously unload up to 18 baskets with up to 20 slides (360 slides)

Program the method of sorting and drying slides

The data presented establishes the SmartSection as a valuable tool for research laboratories to reproducibly cut high quality serial sections and repeatedly place them in the same location on the slide.

Materials and Methods

Specimen Information

— Specimen 1 - Human breast cancer; HER-2 positive, ER/PR negative

— Specimen 2 - Animal kidney (Macaca mulatta)

Two types of specimens were procured, fixed, grossed, tissue processed, and embedded. Slides were created using SmartSection and were H&E-stained using Tissue-Tek Prisma[®] (Ref.1) (Sakura Finetek, Torrance, CA), then coverslipped using Tissue-Tek Film[®] Coverslipper (Sakura Finetek, Torrance, CA). The human tissue slides were further IHC stained with antibodies against ER, PR, HER-2, Ki-67 and Pan-Keratin. Slides were live reviewed and scanned using the VisionTek® Digital Microscope (Sakura Finetek, Torrance, CA) and NanoZoomer 2.0 RS Digital Slide Scanner (Hamamatsu, Japan) then evaluated by scientists and image analysis software Definiens XD and Definiens Image Miner (Definiens AG, Munich, Germany).

For this study, SmartSection was programmed to:

- Face each block to expose the tissue
- Label slides, Matsunami Platinum Pro (Osaka, Japan)
- Serial section blocks at 4µm with Accu-Edge[®] Low Profile Blades (Sakura Finetek, Torrance, CA)
- Mount one section per slide in user-defined location
- Sort slides into Prisma baskets by staining protocal in the output area

Table 1 describes the sequential steps for general operation of the SmartSection.

Step	Action	Step	Action	Step	Action
1	Load blocks into magazines	7	Facing is performed and paraffin debris is vacuumed away	13	Section is moved to the transfer roller in the circulating water bath
2	Load magazines into input area	8	Stager moves the block to utilize a new section of the blade	14	An image of the section is captured on the transfer roller and logged
3	Robot picks up block and transfer it to staging area	9	Moisture is applied to rehydrated the block	15	Section is mounted onto the slide
4	Camera captures an image of the block label and block surface	10	Slide is labeled with specimen number and/or 2D barcode	16	An image of the section is captured and QC inspection is performed
5	Block is auto aligned in the stager, using sensors	11	One section at a time is cut based on the saved sectioning program	17	Accepted slides are placed on a heated stretching plate to remove wrinkles
6	Moisture is applied to rehydrated the block	12	Section is transferred to the wet conveyer belt	18	Slide is placed and sorted by stain application in the slide storage area

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Patient Safety Aspects

To prevent transcription or slide/block mix-up errors, SmartSection uses Lean principles such as single-piece flow and on-board slide printing. To avoid "floater" tissue cross-contamination typical of the manual microtomy process (Ref.2), a vacuum removed paraffin debris from the block and blade surfaces and a circulating filtered water bath captured floating paraffin debris. Additionally, blades were automatically changed between blocks to prevent DNA/RNA cross-contamination of specimens. QC checks were performed at the block and slide level, at staging, transfer roller, and stretching plate.

User Safety Aspects

To protect Histotechnologists, SmartSection was designed with the following safety features:

- Hands-free sectioning; eliminating RMD
- Automated blade exchange; operators never touch the blades, instead blade cartridges are loaded onto the instrument and used blades are automatically deposited into an on-board sharps container

Results

Human Breast Cancer

Presented below are 10 H&E-stained serial sections showing consistent placement of sections on the slides and even staining on a macroscopic level (Figure 2).



Figure 2

SmartSection performs QC on every slide based on user-defined acceptance criteria for wrinkles, tears, folds and number of tissues. Figure 3 shows 10 serial sections at 4um with co-localized area of interest, with microscopically equal H&E-staining.



Figure 3

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SmartSection can continually serial section tissue at 4um to produce consistent sections for IHC staining. Figure 4 shows a 200X microscopic representation of 6 serial sections with the same morphology of one co-location. Test results obtained from SmartSection slides correlate with the specimen information; ER/PR negative, HER-2 amplified and Ki-67 presence, resulting in the ability of SmartSection to fully automate sectioning.



Figure 4

Image Analysis

Animal kidney (Macaca mulatta)

The 10 H&E-stained kidney sections were scanned at 200X, then analyzed using the Definiens Tissue Phenomics Technology for data analysis. Figure 5 shows image analysis results: Tissue (red) is separated from the background, and nuclei (orange) are segmented and classified in all 10 sections. Whole slide results (left) and a selected area of interest in each of the slides (right).



Figure 5

As a parameter for measurement of section consistency, the area of the nuclei was used in the ratio of Σ (area of nuclei) / (area of section); the ratio average was 16% over 10 sections with a precision of +/-10% (95% confidence interval).

Continuous Innovation For Pathology

Conclusions

SmartSection provides the following benefits for the histology laboratory:

- Fully automates sectioning
- Consistently produces high quality sections
- Precisely mounts tissue in the desired location on slides
- Provides safety features to prevent cross-contamination and labeling errors
- Prevents molecular cross contamination, by replacing blades automatically between blocks
- Sorts slides by staining application into designated slide baskets
- Labels slide with legible text and readable 2D barcodes
- Performs QC inspection at block, section and slide levels
- Easy to operate: input magazines, and blank slides; replace blade cartridge; remove slide baskets
- Eliminates operators risk of developing RMD
- Provides reproducible, predictable performance

References

Ref. 1

H&E for specimen 1: Protocol for modified Harris H&E for specimen 2: Protocol for modified Mayer

Ref. 2

Extraneous Tissue - A Potential Source for Diagnostic Error in Surgical Pathology L.J. Layfield MD, B.L. Witt MD, K.G. Metzger HTL(ASCP), and G.M. Anderson HT(ASCP); Am J Clin Pathol 2011; 136: 767-772 Molecular Approaches to Identification of Tissue Contamination in Surgical Pathology Sections M.J. Worsham, S.R. Wolman, and R.J. Zarbo MD DMD; Journal of Molecular Diagnostics, Vol. 3, No. 1, February 2001 Extraneous Tissue in Surgical Pathology - A College of American Pathologist Q-Probes Study of 275 Laboratories G.N Gephardt MD, R.J. Zarbo MD DMD; Arch Pathol Lab Med - Vol. 120, November 1996

Acknowledgments

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