

Effect of Embalming Fluid on Histological Appearance of Organs From Embalmed West African Dwarf Goat Cadavers

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Abstract

The effects of a low-formalin embalming fluid on the histology of some organs from West African dwarf goat cadavers were investigated. This study was designed to ascertain the veracity of the claim believed by many histopathologists that the use of specimens from embalmed cadavers is good enough for investigative research and forensic medicine, especially in determining the cause of death at autopsy. The visceral organs examined include kidney, lung, spleen, and liver. The quality of slides from embalmed cadavers was graded using these criteria: general



Transverse section of liver from embalmed cadaver showing close to normal general organ microscopic architecture. Note the hepatocytes in the hepatic parenchyma (HP), central vein (CV), and hepatic capsule (HC). H&E, 100X.

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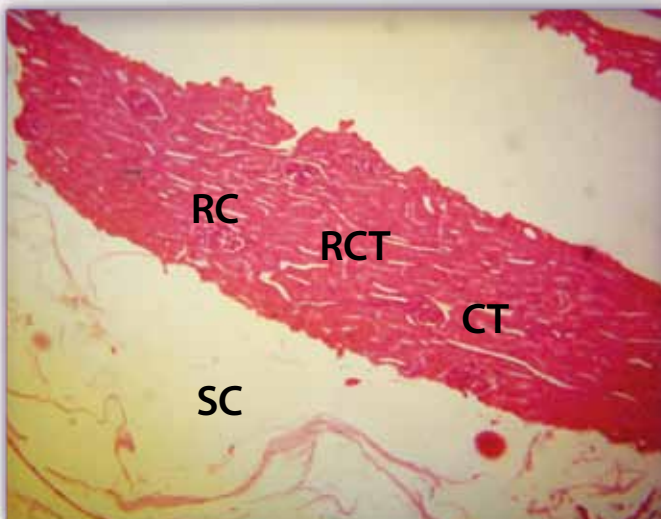


Fig. 1. Transverse section of kidney from embalmed cadaver showing renal cortex (RCT) containing renal corpuscle (RC) and convoluted tubules (CT). Note the separation of the capsule from the cortex (SC). H&E, 100X.

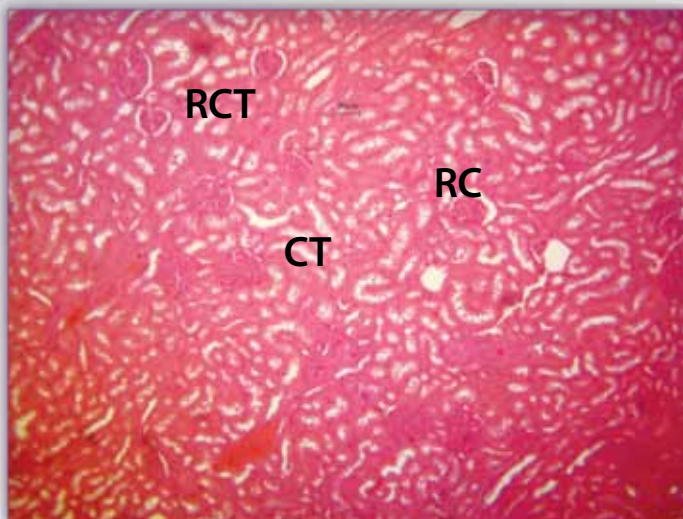


Fig. 2. Transverse section of kidney from fresh sample showing renal cortex (RCT) containing renal corpuscle (RC) and convoluted tubules (CT). H&E, 100X.

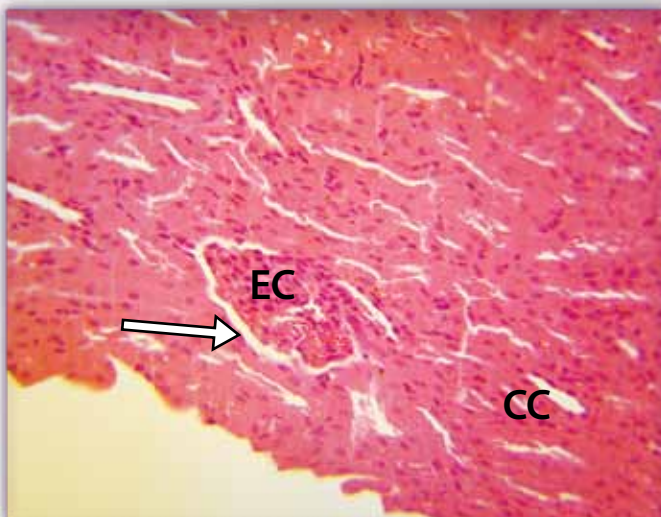


Fig. 3. Transverse section of kidney from embalmed cadaver showing simple squamous cells of Bowman's capsule (arrow), endothelial cells of the glomerulus (EC), and the simple cuboidal cells of the convoluted tubules (CC). Note the darkly stained nuclei. H&E, 400X.

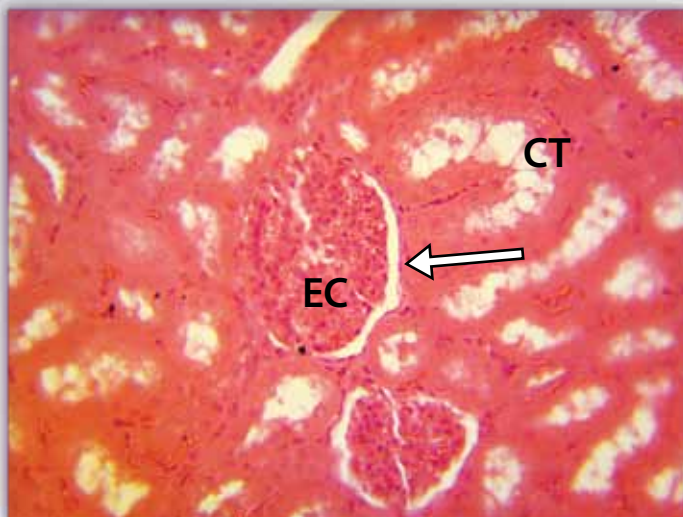


Fig. 4. Transverse section of kidney from fresh sample showing simple squamous cells of Bowman's capsule (arrow), endothelial cells of the glomerulus (EC), and the simple cuboidal cells of the convoluted tubules (CT). H&E, 400X.

organ microscopic architecture, cell morphology, and state of the epithelium. They were graded on a scale of 1 to 3, where 1 refers to a high degree of cell distortion, 2 is used to grade moderately good sections, and 3 is used for sections that are very close to normal. The slides from embalmed cadavers were seen to be moderately good, scoring 2 on the scale described above. Liver presented the least tissue distortion in this study, which may be attributed to its high degree of vascularization that allows for better perfusion by embalming fluid, resulting in proper fixation. This study suggests that tissue sections from embalmed cadavers with at least 4% formalin embalming fluid may be adequately fixed for histopathology.

Introduction

The objective of embalming is to achieve perfusion of fixative throughout all parts of the body. Immersion and direct injection into tissue cannot achieve this satisfactorily, so the injection of embalming fluid through the circulatory system is done for best results.^{1,2} The fluid used for embalming animals is designed to fix and preserve the tissue, preventing deterioration. It should also render the cadaver suitable for dissection, and prevent fungal and bacterial growth during the period of dissection.^{1,2} It is also important that tissue from embalmed cadavers be suitable for histological sectioning and observation.³⁻⁵

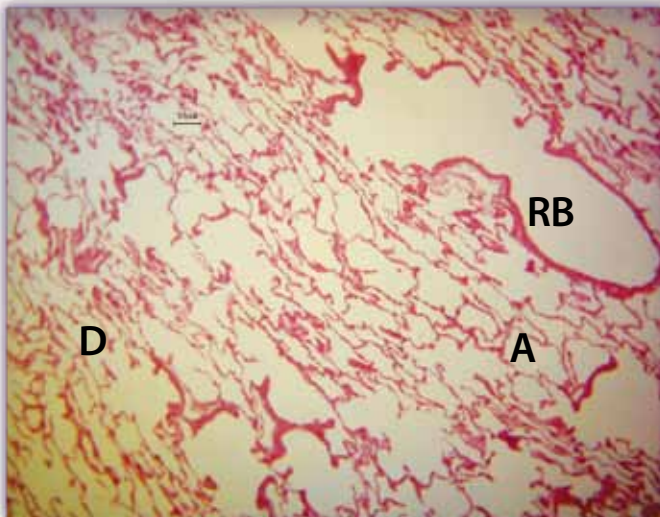


Fig. 5. Transverse section of lung tissue from embalmed cadaver showing respiratory bronchiole (RB), alveolar duct (D), and alveolar sac (A). Note the distorted general organ morphology. H&E, 100X.

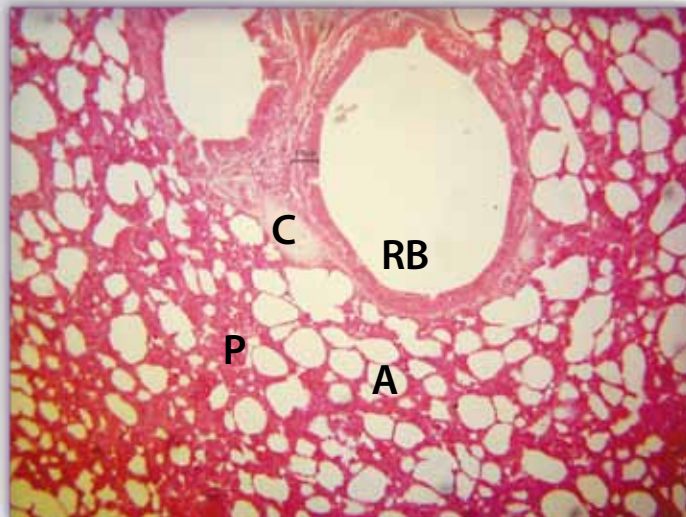


Fig. 6. Transverse section of lung tissue from fresh sample showing respiratory bronchiole (RB), pneumocytes (P), cartilage (C), and alveolar sac (A). Note the abundant pneumocytes in the organ parenchyma. H&E, 100X.

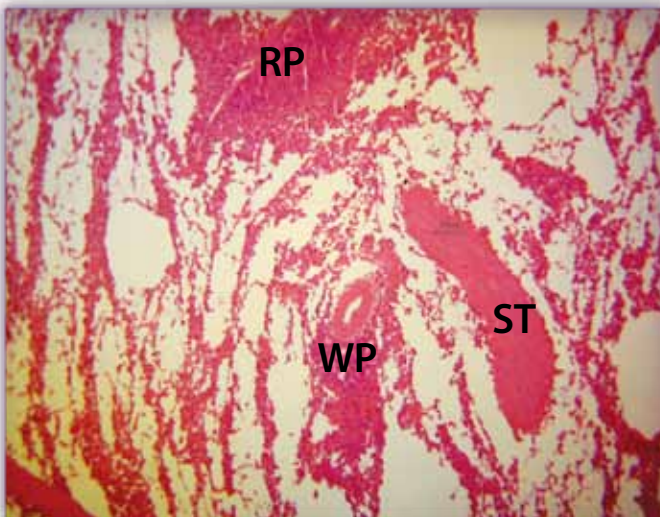


Fig. 7. Transverse section of spleen from embalmed cadaver showing organ microscopic architecture distortion. Red pulp (RP), white pulp (WP), and splenic trabeculae (ST). H&E, 100X.

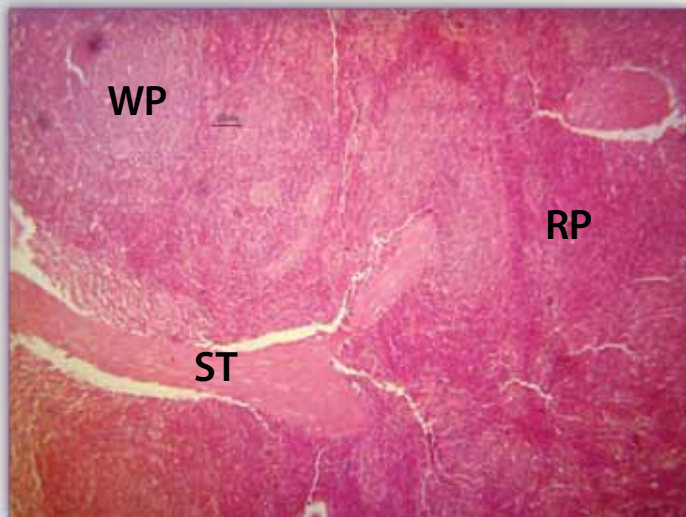


Fig. 8. Transverse section of spleen from fresh sample showing normal organ microscopic architecture. Red pulp (RP), white pulp (WP), and splenic trabeculae (ST). H&E, 100X.

A comparative study on the histological quality of specimens from cadavers embalmed with different embalming fluids was undertaken by Nicholson et al in 2005.⁵ A report by Kalanjati et al on the use of a low-formalin solution (5%-7.5% formaldehyde) for an effective, efficient, and safer embalming process was published in 2012.² In Nigeria, the use of cadavers for forensic histopathology is a common practice, but there is a dearth of information in published literature on the suitability of these autopsy samples for histopathology, hence the need for this study. The aim of this work is to compare the histological appearance of samples processed from 4% formalin-embalmed animal cadavers with samples processed from freshly slaughtered animals fixed in 10% formalin. The information obtained from this study will fill the knowledge gap and help in ascertaining the extent of suitability of such cadaver samples for histopathology.

Materials and Methods

Tissue samples used in this study were obtained from West African dwarf goat (WADG) cadavers in the veterinary gross anatomy laboratory of Michael Okpara University of Agriculture, Umudike, Nigeria, and from apparently healthy freshly slaughtered WADGs from the goat farm at the same institution. The cadavers were embalmed with fluids containing formalin 4%, alcohol 33%, phenol 2.5%, glycerol 2.5%, and tap water 58%. The embalming fluid was infused into the cadaver through the common carotid artery. Slices of samples from the slaughtered goats were fixed in 10% formalin. The organs used for the study include kidney, lung, spleen, and liver.

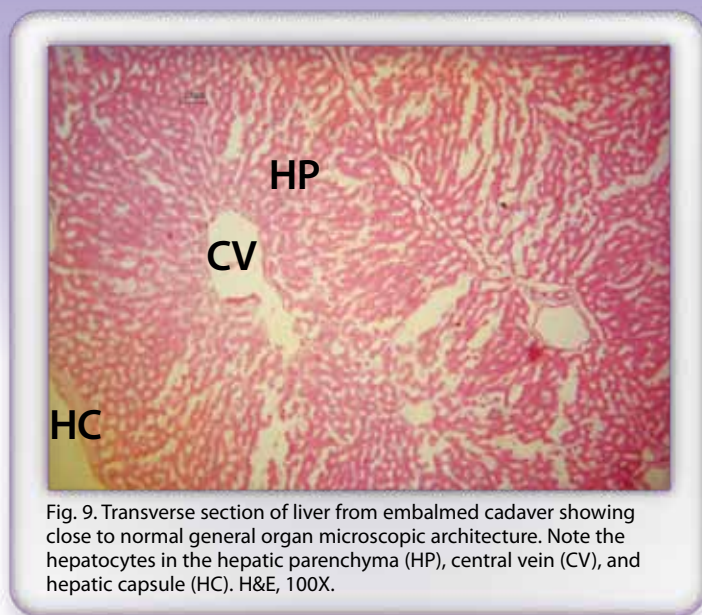


Fig. 9. Transverse section of liver from embalmed cadaver showing close to normal general organ microscopic architecture. Note the hepatocytes in the hepatic parenchyma (HP), central vein (CV), and hepatic capsule (HC). H&E, 100X.

Slices from these organs were dehydrated in graded alcohol, cleared in xylene, and embedded in paraffin wax. They were sectioned with a rotary microtome to 5 microns. Sections were routinely stained with hematoxylin and eosin (H&E).⁶ The stained sections were examined with an Olympus microscope and photographed with a Moticam camera attached to the microscope.

The qualities of the stained sections from embalmed cadavers were graded using these criteria: general organ microscopic architecture, cell morphology, and state of the epithelium. They were graded on a scale of 1 to 3, where 1 refers to a high degree of distortion, 2 is used to grade moderately good sections, and 3 is used for sections that are very close to normal.

Results

Kidney: Tissue sections from embalmed cadaver presented distorted microscopic architecture (Fig. 1) as seen in the separation of the capsule and the collapsed appearance of the convoluted tubules, in contrast to sections from fresh tissue (Fig. 2). At higher magnification, the nuclear morphology and epithelial covering of the convoluted tubules appeared moderately good (Fig. 3) when compared to the section from fresh tissue (Fig. 4), hence a grading of 2.

Lung: The architecture at low magnification for tissue sections from embalmed cadaver presented general organ morphology that appeared to be distorted, as demonstrated by irregular expanded open spaces (Fig. 5), unlike the sections from fresh tissue (Fig. 6). However, at higher magnification (not shown), the cellular morphology appeared normal as did the epithelial lining of the respiratory bronchioles, hence a grading of 2.

Spleen: General organ microscopic architecture of tissue sections from embalmed cadaver appears to be distorted (Fig. 7) as demonstrated by open spaces in the organ parenchyma, unlike the normal appearance of the section from fresh tissue (Fig. 8). However, cellular morphology of the cells of the white and red pulp, as well as the covering capsule and vascular endothelium, were normal, hence a grading of 2.

Liver: General organ microscopic architecture of tissue sections from embalmed cadaver appears to be moderately distorted (Fig. 9), but the cell morphology of the hepatocytes and Kupffer cells was normal. The central vein and portal triad were normal. The covering capsule and vasculature, especially the sinusoids, were normal, hence a grading of 3.

Discussion

Phenol was added to the embalming fluid because of its fungicidal and bactericidal properties. The alcohol serves as a fixative with a faster rate of penetration and better preservation of the nucleic acids while the glycerol helps the cadaver retain its natural color and also make it pliable during dissection.^{3,5}

In this study, the loss of normal tissue architecture could be attributed to the inability of the 4% formalin embalming fluid to properly fix the connective tissue stroma supporting the cells. This may be due to its inadequate protein cross-linking in the connective tissue fibers by the low formalin content.⁶ Therefore, if the diagnosis is to be based strictly on cellular morphology, then samples from cadavers embalmed with fluids containing 4% formalin can be considered reliable.⁷ But if the tissue architecture is to be the determining factor, a slightly higher concentration of formalin—not more than 20% formalin—should be used in the preparation of the embalming fluid.^{2,5} The higher-quality preservation of the liver seen in cadaver liver samples may be attributed to the liver's increased vascularization, since the vasculature was used as the route for the infusion of the embalming fluid.⁸

Conclusion

Results of tissue samples collected from 4% formalin-embalmed cadavers can be used reliably for histopathologic investigation if cellular morphology is the major criteria for diagnosis. This finding will encourage laboratory staff of morbid anatomy units to keep using a low percentage of formalin in preparing embalming fluids. When considering the health hazards associated with formaldehyde exposure, this will help increase safety for laboratory personnel who use formalin to fix and preserve cadavers for diagnosis and forensic investigation.

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Stereo-orientation of Mohs Surgery Frozen Section Specimens: A Technical Perspective

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Abstract

Maintaining specimen orientation is of the utmost importance to Mohs surgeons and technologists who prepare specimens for cryotomy. Many techniques are employed to achieve correct tissue orientation, including placing nicks into the tissue sample or using marking dyes with a clockface orientation. Stereo-orientation of tissue sections offers useful advantages for maintaining the surgical sample's orientation—from start to finish. This report offers detailed instructions for implementing this technique. Although there may be some reservations about or difficulties in making the transition to stereo-orientation, the series of illustrations presented in this paper will aid the reader in understanding how stereo-orientation is achieved in order to make the transition easier.

Introduction

Mohs micrographic surgery is a highly specialized treatment for the total surgical eradication of skin cancer. This technique was developed to conserve normal tissue while removing the malignancy, and is solely dependent on the microscopic examination of each surgical layer until clear margins are achieved. Proper orientation of Mohs specimens is vital to the successful surgical removal of the skin cancer.

This report discusses a technique first identified by Barbara Beck, HT(ASCP), a former Mohs histotechnologist, while working in Tallahassee, Florida, with Mohs surgeon Armand Cognetta Jr, MD, who quickly appreciated the value of the section-mounting technique Beck used in her work. As a result, Dr. Cognetta began to request that all of his slides be oriented in this manner; this is where the stereo-orientation technique was conceived.¹

Laboratories use a number of different strategies when orienting tissue for Mohs micrographic surgery. In my own work at the same Mohs lab for 15 years, orienting and embedding tissue specimens became so routine that I never considered changing my technique. However, when I changed jobs, I was suddenly thrust into learning how to orient specimens using the stereo-orientation technique.

Before this technique was introduced, it was not uncommon for a tissue section to be placed onto a microscope slide randomly without consideration to its placement. Stereo-orientation requires mounting the frozen section specimens onto the microscope slide in such a way that the tissue placement precisely matches the orientation of the Mohs surgical map and the surgical wound (Fig. 1).² When the Mohs surgeon views the slide through the microscope with the label to the left, the specimen is oriented exactly as it is on the surgical map. This strategy for specimen orientation can offer significant advantages for the accurate tracking of areas of malignancy.

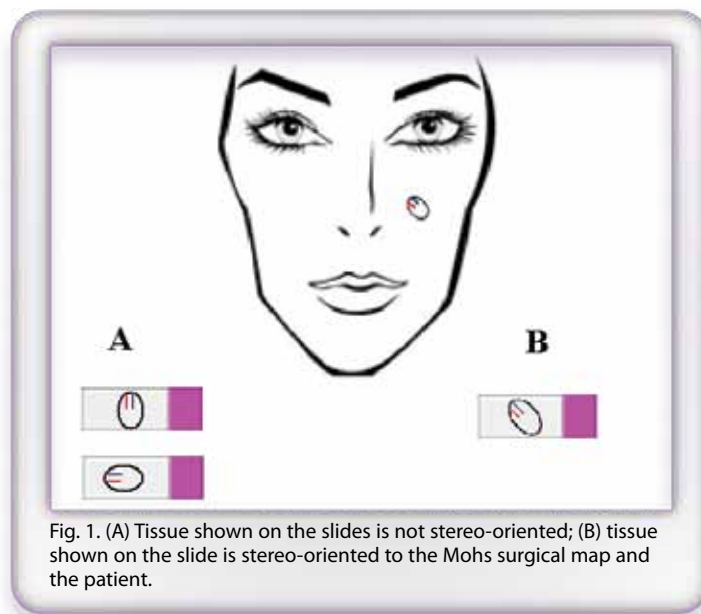


Fig. 1. (A) Tissue shown on the slides is not stereo-oriented; (B) tissue shown on the slide is stereo-oriented to the Mohs surgical map and the patient.

Materials and Methods

The Mohs surgeon delivers the specimen and map to the laboratory where the tissue is sectioned by the technologist. The sample is compared to the map to ensure accurate orientation and marking. The surgeon makes a nick in the specimen; this is where the marking dyes are applied. The map will reflect this color coding. To further relax the tissue, a nick can be placed opposite the original slit. Figure 2 demonstrates how the specimen is color coded and illustrates how its orientation is identical to the map and the surgical wound.

The tissue is embedded using Tissue-Tek® O.C.T.™ Compound (Sakura Finetek USA, Inc., Torrance, CA) and the slide embedding technique.³ Once the block is ready for sectioning, it is mounted in the block holder of the cryostat with the marked slit facing the blade. When the technician places the section onto the slide, the label should be to the right (Fig. 3); however, when the slide is viewed under the microscope, the slide should be rotated 180 degrees so that the label is to the left. This is to account for the optical inversion produced by the microscope. Now what the Mohs surgeon sees is an exact replica of the map and the excised Mohs tissue (Fig. 4).

If the specimen is bisected, both superior edges are dyed identically with one color, and the inferior edges are dyed identically to each other but with a different color. Each half of the bisected specimen is embedded separately and labeled as blocks

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Fig. 2. The tissue sample is arranged to match the surgeon's map. In this photo, red and blue inks are applied to maintain sample orientation. The illustration drawn onto the microscope slide is to demonstrate for the reader how the tissue section should be mounted onto the glass.

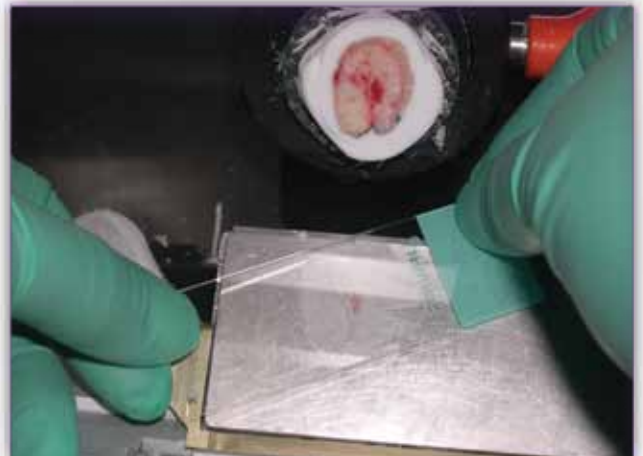


Fig. 3. The section is mounted onto the glass slide with the slide label to the right.

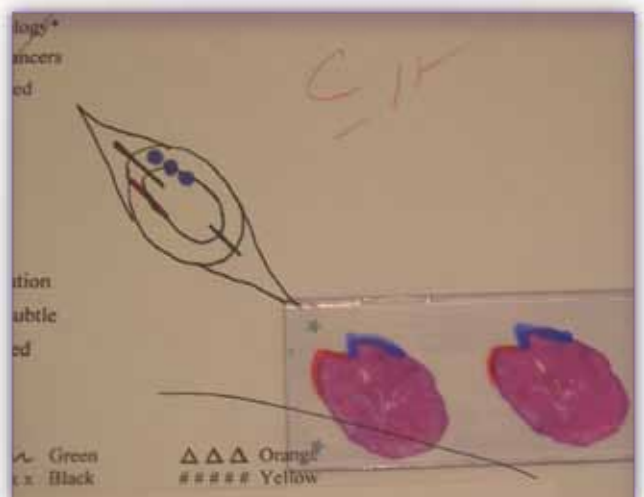


Fig. 4. Stained sections with apparent color-coding inks are mounted with stereo-orientation, which directly matches the specimen map drawn by the Mohs surgeon.

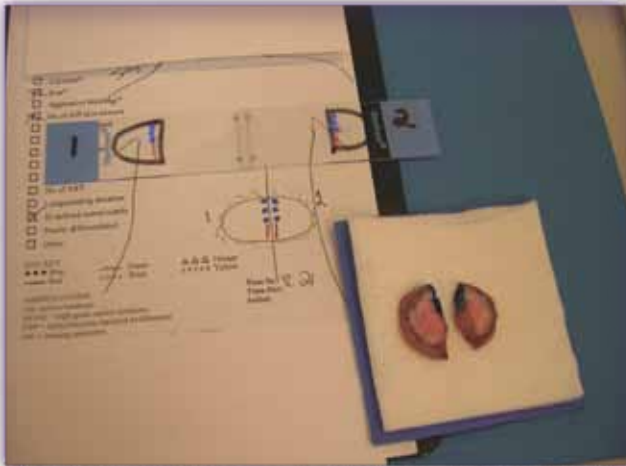


Fig. 5. The sample is bisected, mounted, and color coded as indicated on the map.

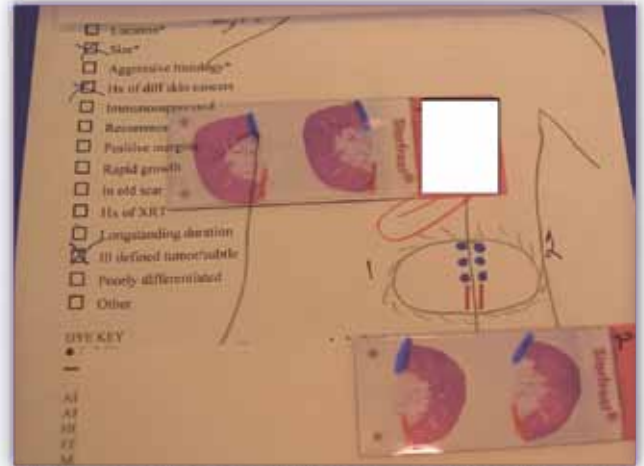


Fig. 7. H&E stained slides that were stereo-oriented and labeled as specimens 1 and 2.

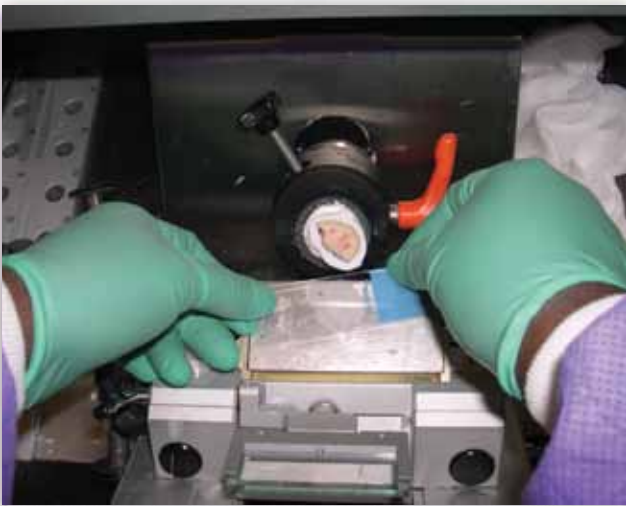


Fig. 6. Specimen is labeled as 2 on the map shown in Fig. 5.

1 and 2. Figure 5 illustrates how the specimens are color coded and oriented in such a way that they replicate the map of the wound. Once the blocks are ready for sectioning, they are placed in the cryostat block holder with the marked superior edge of the specimen facing the blade. The sections are picked up with the slide label to the right and are oriented to be identical to the surgical map and the surgical wound (Fig. 6).

Once the slide is rotated 180 degrees and viewed under the microscope (with the label on the left), it will be an exact replica of the Mohs map and the surgical wound (Fig. 7).

Discussion

The premise behind this technique is that, when reading slides, the Mohs surgeon can be confident that the tissue mounted on the slide is an identical representation of the specimen map (as if the specimen were superimposed on the map) so that the location of residual tumor can be identified and tracked accurately. Adopting and consistently applying this strategy for mounting tissue sections onto glass slides can reduce any possibility of error while minimizing mental fatigue. It may even save time. In instances when the specimen is too large to fit onto a standard glass slide for stereo-orientation, the technologist should consult with the surgeon before proceeding.

Conclusion

Stereo-orientation is a novel technique for mounting tissue sections that offers important benefits to the Mohs surgeon for accurately tracking residual tumor foci. There are many ways to orient tissue specimens, each with its advantages and disadvantages. Stereo-orientation is one such method that may be unfamiliar to many Mohs surgeons and technologists. This article is meant to familiarize others with this technique and to allow them the opportunity to adapt it to their own practice.

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Third-hand Immobilization in Gross Sectioning of Pathology Specimens

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Abstract

This technical note proposes the “third-hand” principle for additional immobilization during gross sectioning of tissue specimens. It consists of pressing the specimen to be cut against a vertical surface (referred to here as the “third hand”). This method is especially beneficial for calcified specimens (bones and tumors) and soft tissue samples that require thin, uniform gross sections to be achieved. The third-hand immobilization principle is the underlying foundation for the design of the Biopsy Uniform Section Device, a concept developed by this author. Although the development of the device is still a work in progress, the testing of prototypes confirms the advantage of third-hand immobilization in achieving uniform and perpendicular sections of pathology specimens.

Introduction

There is no question that gross sectioning of surgical pathology specimens requires a firm surface beneath the specimen. The use of a cutting board surface (similar to that used in food preparation environments) is quite commonplace in the surgical pathology laboratory. The degree of firmness or pliability of the specimen dictates the need for such immobilization. In many cases, a skilled grossing technologist can achieve reasonably satisfactory sections freehand without much difficulty. However, some specimens require additional support from special techniques, devices, gadgets, and improvised at-hand materials. This technical note presents the third-hand principle for additional immobilization during gross sectioning of pathology specimens. This technique consists of pressing the area to be cut against a vertical surface (the “third hand”). The “first hand” holds and moves the specimen while the “second hand” operates the cutting instrument. The third-hand immobilization method is most beneficial when cutting calcified specimens and soft tissues that can otherwise pose significant challenges in achieving optimum sections.

Calcified Specimens

An acceptable section of femoral head with osteoarthritis can be easily achieved by using either a mechanical saw or handsaw because the relative firmness of the specimen is fairly consistent throughout. However, technical challenges can arise when sampling calcified tumors, fragile complicated bones, small fragments of bone, and samples requiring serial sectioning at the grossing table. In my experience, perfectly acceptable results can be achieved with a hacksaw when working with these specimens. There are certain particularities in sawing technique, but the main issue is preventing the specimen’s movement due to a snag between the saw and the calcified tissue. The employment of the third-hand principle for additional immobilization offers significant advantages when working with such samples.

It is important to find an appropriate vertical surface to press the bone specimen against during sawing. Depending on the size of the specimen, the vertical surface can be different in form, but must be more or less flat. A wooden holding tray, like the one used in the Davidson Marking System® (Bradley Products, Inc, Bloomington, MN), can be useful as an immobilization support gadget for some specimens. This wooden stand with round bottle holders and multiple different-shaped notches can be modified to accommodate many configurations of bone specimens. The pegs are very useful, especially if some of them are flattened so they can be utilized as the third hand during sectioning (Fig. 1).



Fig. 1. A small bone specimen can be pressed against a flattened wooden peg of a Davidson Marking System holding tray, which serves as the third hand, offering additional immobilization when cutting.

In the case of a fragile bone or calcified tumor, it is reasonable to fashion a hard-pressed packing carton into an immobilization support using this at-hand material (Fig. 2). In this situation, the wall serves as the third hand, preventing the fragile bone’s surface from crashing, while allowing the cutting instruments, such as a saw, to pass through the carton, maintaining the integrity of the specimen and the section.

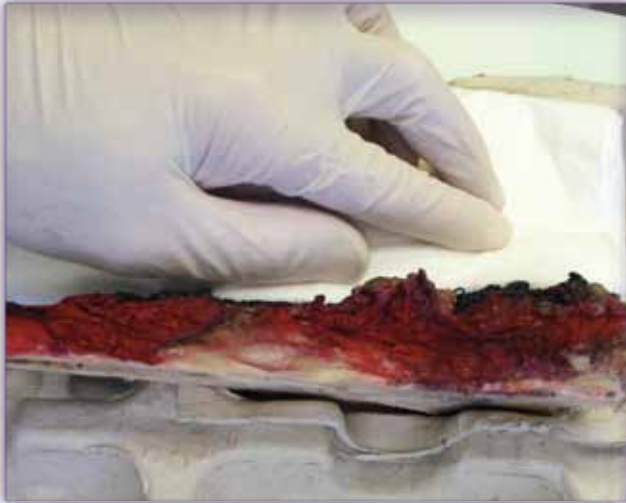


Fig. 2. A femur bone with sarcoma is pressed against the border of a hard-pressed carton tray for a longitudinal section.

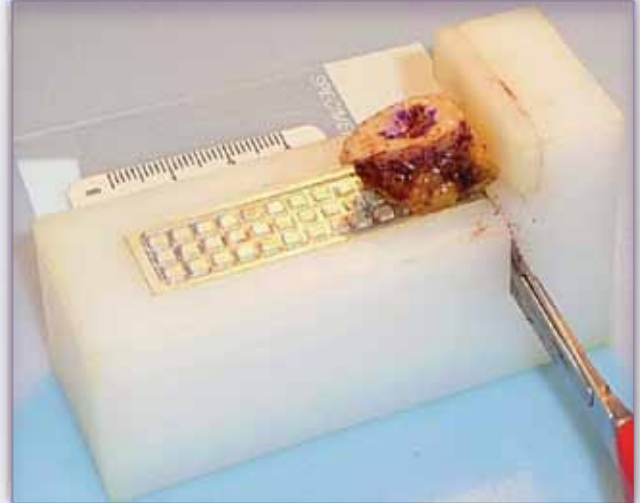


Fig. 4. Sectioning of a skin ellipse using the Biopsy Uniform Section Device.

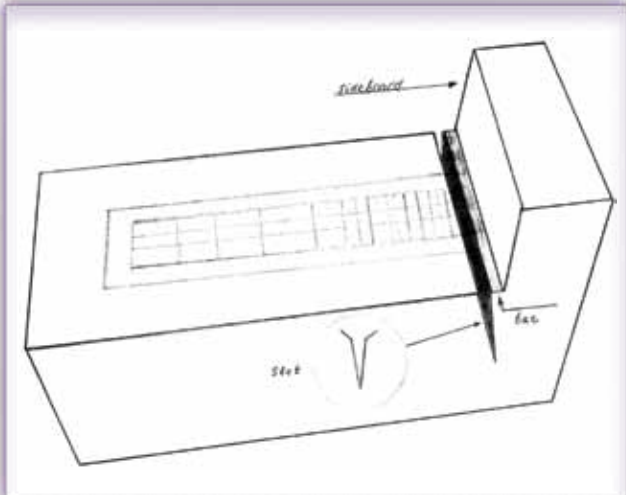


Fig. 3. A sketch of the Biopsy Uniform Section Device, designed by the author.

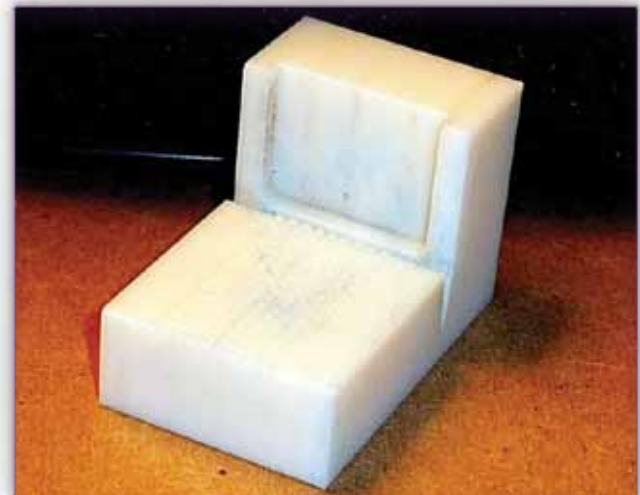


Fig. 5. A prototype of an immobilization device with a vertical sideboard niche.

Biopsy Specimens

Achieving uniform thin sections (slices) of pathology samples has always been one of the primary goals in preparing specimens for histotechnology, but today it is an absolute necessity when processing tissues using microwave-accelerated processing technology because the success of the protocol is dependent on the size of the sample. While implementing the Sakura Tissue-Tek® Xpress® (Sakura Finetek USA, Inc., Torrance, CA) microwave processing instrument, the University of Miami/Jackson Memorial Hospital developed a grossing board with a slotted metal plate and a knife/blade track to achieve uniform thin sections.¹ Sliding the knife/blade in the track along the surface of the cutting board

produces sections with uniform (1.5 mm) thickness. Sakura Accu-Edge® Grossing Tools include a specially designed grossing board to accommodate different size specimens by using "adjustable wells that ensure to an exact thickness."² Actually, the bottom of the well serves as a third hand of sorts for the immobilization of the tissue sample in order to achieve uniform sections.

The Biopsy Uniform Section Device implements the principle of third-hand immobilization while using a different technological strategy to achieve the desired outcome. The core of the design is its 3 features: (1) a specific horizontal sidebar measurement (2 mm, 3 mm, and 4 mm) for achieving a specific thickness of the

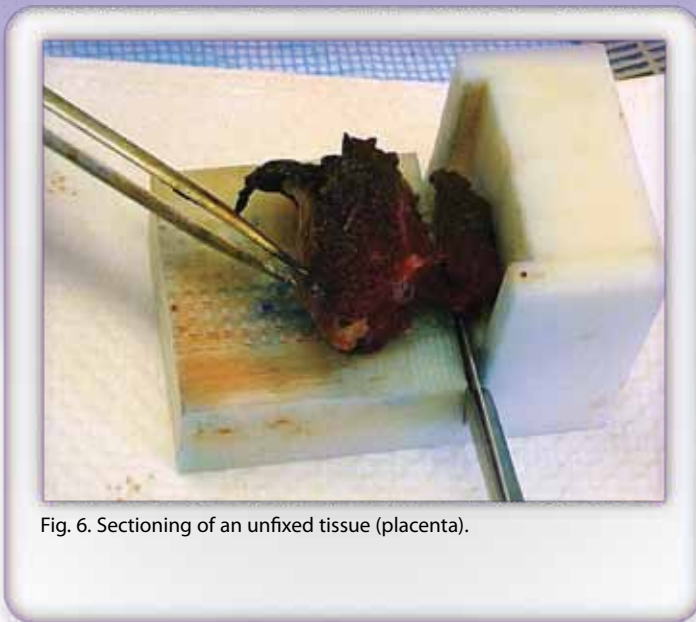


Fig. 6. Sectioning of an unfixed tissue (placenta).

section, (2) a vertical sideboard, and (3) a V-notched slot for the cutting blade. The sideboard provides immobilization for cutting the end of the tissue, ultimately functioning as the third hand (Fig. 3).

Figure 4 shows the principle of using the Biopsy Uniform Section Device. A skin ellipse is placed on the platform and moved toward the slot and the vertical sideboard using forceps. When the skin sample reaches the vertical sideboard, the part of the sample on the horizontal sidebar is the desired thickness of the section.

By pressing the specimen against the vertical sideboard, secure immobilization is achieved for a definitive uniform perpendicular cut. The slice is removed with forceps after each section has been made and placed into a processing cassette oriented as it is intended to be embedded. Milestone Medical has improved the design of the vertical sideboard by making a niche that provides additional immobilization for the section (Fig. 5). This is especially important in the case of large samples.

The Biopsy Uniform Section Device is designed for small specimens, but the vertical sideboard can be adjusted to accommodate larger specimens. However, as shown in Fig. 6, it is difficult to obtain uniform serial sections when the specimen is not adequately fixed. Third-hand immobilization is not sufficient in this situation.

Discussion

The principle of precise cutting against a sideboard is used in a variety of applications, such as the cutting of lumber, for example. This principle is also used in grocery stores for the uniform slicing of cheese and ham. Although the cutting tools differ from each other in their specific uses, the immobilization platform is a common element that contributes to a precise cut. However, the goal in surgical pathology is to obtain a diagnostically valuable section. This often requires additional devices and materials in the preparation of optimum specimens, including some that can be used in the practical application of the third-hand immobilization principle.

Variants of gadgets and at-hand improvised materials that might serve as additional third-hand immobilization aids are endless.

The aforementioned contrivances reflect personal experience, which is always limited.

Numerous experiments I have undertaken in search of optimal third-hand support materials have led me to conclude that hard-pressed packing cartons appear to be optimal for the type of support needed for bone sampling.³ They provide the necessary firmness, but not too much. The relative roughness of the carton's surface gently grasps the specimen's inevitably uneven surface. The saw can cut through the specimen and carton without crashing, which can occur at the end of the cut. With a collection of disposable immobilizing materials, it would be appropriate to point out that every calcified specimen can be cut without preliminary decalcification, including bones from facial-maxillary surgery and calcified tumor, providing valuable sections for evaluating surgical margins or to answer other diagnostic questions.

The holding trays from the Davidson Marking System are permanent fixtures in every surgical pathology laboratory.⁴ The configuration provides endless options for third-hand immobilization of small bone or calcified tissue. Saw-friendly wood makes it possible to cut a fragile or small fragment without fear of a crash toward the end of sawing. Even some excisional biopsies can be cut in this way, although a special gadget like the Biopsy Uniform Section Device would be a better option.

The Biopsy Uniform Section Device not only provides sections of predetermined thickness, depending on the horizontal bar's size and the size of the vertical sideboard, but its third-hand immobilization capability facilitates a strictly perpendicular gross section that is very important in dermatopathology, gastroenterology, and gynecology samples, as well as in the evaluation of surgical resection margins.

The development of the Biopsy Uniform Section Device is still a work in progress. Much work remains before a final design will be ready.

At this time, there have been no reports of studies with comparable devices, perhaps because sectioning strategies differ. The main, and perhaps most important, variable is in the ability to visualize the sample to be sectioned while it is immobilized, which is a large advantage and the primary focus of the concepts proposed in this report. A blind, indiscriminate cut is not acceptable in excisional biopsy slicing.

The desire for one universal tool that can be used to achieve thin, uniform gross sections for all types of pathology specimens is unrealistic. The diversity of surgical pathology specimens will make a variety of devices necessary to achieve the desired outcome in each instance. The principle of third-hand immobilization lays the foundation for the design of such devices.

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The Effectiveness of Even Ice Surface on Microtomy of Paraffin Blocks

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Abstract

Our high-volume histology lab requires that a histotechnologist be able to cut 50 paraffin blocks per hour, an increase from the previous requirement of 30 blocks per hour. In order to ensure consistent, high-quality outcomes, the histotechnologist must develop a routine that reduces the variables that can slow output or diminish quality since there is no time for adjustments during cutting.

In our laboratory, we established a consistent output of slides using a methodology that makes blocks easy to cut with quick output and fewer cutting artifacts. The technique consists of chilling the blocks on an even, flat surface of ice. The even cooling of the blocks allows for the temperature and soaking of the blocks to be uniform, which helps achieve sections that are free of chatter and folds.

Introduction

Chatter, which is the term for artifact that results from microscopic vibration during microtomy, can be observed in tissue sections when viewed with the microscope (Fig. 1). Thompson and Luna attribute this artifact primarily to overdehydration or removal of protein-bound water, which can occur when tissues are overexposed to dehydrating solutions during tissue processing.¹ Soaking paraffin blocks prior to sectioning can restore the necessary moisture to tissues, which facilitates sectioning (Fig. 2). Microscopic chatter can also be caused by a dull blade or an incorrect blade angle (knife tilt), which causes the section to be scraped rather than sliced. Chatter can also occur by cutting too rapidly.²

The fact is, in today's laboratories, the mass production of slides and rapid turnaround time expectations have raised the bar for production from technicians. The use of a flat, even ice surface for the cooling of blocks allows more blocks to be sectioned per minute. Even though rapid cutting is required to achieve desired output, the uniform ice surface reduces artifacts that can result from rapid sectioning and ensures that section quality is acceptable.

Materials and Methods

Achieving an even ice surface for block cooling is easy to accomplish by placing clean water on a tray and keeping the tray level in the freezer until frozen. The goal is to obtain a surface of ice similar to the ice of an ice skating rink. The ice should be flat and even throughout the entire surface area without bumps or cracks (Fig. 3).

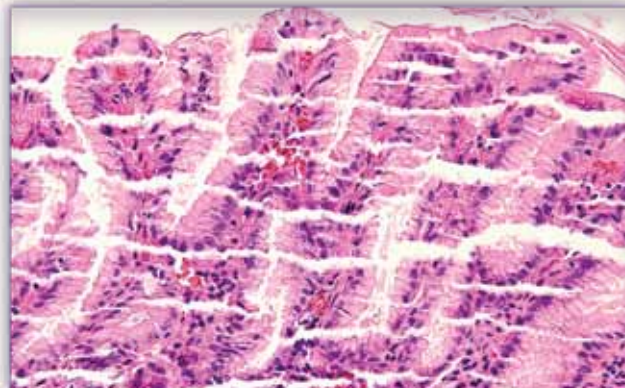


Fig. 1. Chatter in section of gastric antrum. H&E, 200X.

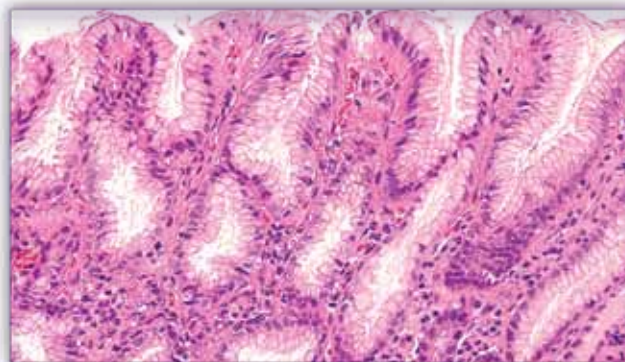


Fig. 2. Uniform cooling and soaking of paraffin blocks yield a section of gastric antrum that is free from artifacts. H&E, 200X.



Fig. 3. An even ice surface allows for uniform cooling of paraffin blocks.



Fig. 4. A rough or uneven ice surface provides inconsistent cooling of blocks, leading to sectioning artifacts.

A flat ice surface can also be achieved by scraping the ice. The extra time needed to prepare the ice properly is worth the effort when your cutting output achieves both the desired quality and quantity. In our lab, we use large blue thermal trays that allow the ice to remain frozen longer (Fig. 3).

We process our gastrointestinal (GI) biopsies with a 2-hour processing protocol on a conventional closed-system processor. GI tissue is prone to overdehydrating if longer processing times are used. If overdehydration is a problem, this can be overcome by soaking the faced paraffin block in water directly over ice, which is imperative in order to achieve good sectioning outcomes. This is easier to accomplish when water is added onto a flat, even ice surface before placing your block face down on the surface with or without a paper towel. If the ice surface is not level, you can use wet paper towel directly in contact with the ice surface to compensate.

Blocks are soaked for about 20 minutes to allow sufficient moisture to reenter the blocks to soften the tissues. You may need to determine the best time for your tissues through trial and error. Each laboratory should optimize soaking times based on the tissue types and processing schedules you use. Soaking time can vary between different tissue types. Exercise caution not to oversoak your blocks, which can cause other artifacts that are undesirable at cutting. With optimal soaking time, artifact-free sections can be achieved more rapidly during sectioning.

Results

The chatter artifact is often invisible to the eye while sectioning tissue on a microtome. More dramatic artifacts, like the "Venetian blind" effect, can be evident when sections are placed onto the water bath surface. Chatter identified in microscopic sections is often the result of tissue that has not been well soaked; it may also occur when sectioning (cutting) with the microtome is done too rapidly.

On an uneven ice surface, the blocks cool at uneven temperatures due to bumps on the ice and uneven exposure to ice surface (Fig. 4). Well-soaked blocks on a smooth, even ice surface will chill at even temperatures and produce ribbons of good quality that reduce chatter and can be cut with ease.

References

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Acknowledgment

The author would like to acknowledge Dr. Gerald P. Bailey, MD, PhD, for his collaboration in preparing the micrographics for this article.

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Speaker: Damien Matusiak
Leica Biosystems
Buffalo Grove, IL
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- 26 **NSH Teleconference 1:00 pm Eastern Time**
Title: **Identifying Histology Look-Alikes**
Speaker: Amy Aulthouse, PhD
Ohio Northern University
Ada, OH
Phone: (443) 535-4060 or register online at www.nsh.org

JULY

- 19 **University of Texas Health Sciences Ctr/San Antonio**
Teleconference 12:00 pm Central Time (800) 982-8868
Title: **Green Histology**
Speaker: Lawrence Patton
Leica Biosystems
Buffalo Grove, IL
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- 24 **NSH Teleconference 1:00 pm Eastern Time**
Title: **How Did This Slide Contamination Happen?**
Speaker: Violet Swazer
Detroit Medical Center
Detroit, MI
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AUGUST

- 16 **University of Texas Health Sciences Ctr/San Antonio**
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Speaker: Damien Matusiak
Leica Biosystems
Buffalo Grove, IL
-
- 28 **NSH Teleconference 1:00 pm Eastern Time**
Title: **What Should a Good H&E Look Like?**
Speaker: Robert L. Lott
Skin Pathology Associates
Birmingham, AL
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SEPTEMBER

- 18 **NSH Teleconference 1:00 pm Eastern Time**
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Rocklin, CA
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- 20 **University of Texas Health Sciences Ctr/San Antonio**
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Title: **Histology Process Improvement: Tissue Processing From Conventional Overnight to Continuous**
Speaker: William DeSalvo
Collaborative Advantage Consulting
Tempe, AZ
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- 20-25 **National Society for Histotechnology Symposium/Convention**
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Providence, RI
Contact: Aubrey Wanner
Phone: (443) 535-4060 or register online at www.nsh.org
Fax: (443) 535-4055
Email: aubrey@nsh.org

OCTOBER

- 18 **University of Texas Health Sciences Ctr/San Antonio**
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Title: **Safety in the Histology Laboratory**
Speaker: Terri L. Braud
Holy Redeemer Hospital
Rockledge, PA
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- 23 **NSH Teleconference 1:00 pm Eastern Time**
Title: **Building Effective Teams**
Speaker: Louis Anderson
Johns Hopkins Medical Institutions
Baltimore, MD
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NOVEMBER

- 15 **University of Texas Health Sciences Ctr/San Antonio**
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Title: **Role of Pathologist in Colorectal Cancer Diagnosis and Management**
Speaker: Prashant A. Jani, MD, FCAP, FRCPC
Thunder Bay Regional Health Sciences Centre
Thunder Bay, Ontario, Canada
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- 20 **NSH Teleconference 1:00 pm Eastern Time**
Title: **An Introduction to Commonly Used Immunohistochemical (IHC) Stains in Dermatopathology**
Speaker: Alison Uzeblo, MD
William Beaumont Hospital
Royal Oak, MI
Phone: (443) 535-4060 or register online at www.nsh.org

DECEMBER

- 18 **NSH Teleconference 1:00 pm Eastern Time**
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