HISTO-LOGIC®

No reader should utilize materials and/or undertake procedures discussed in HISTO-LOGIC articles unless the reader, by reason of education, training and experience, has a complete understanding of the chemical and physical properties of all materials to be utilized and the function of each material by which any procedure is accomplished.

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SPECIAL NOTICE

If anyone in your laboratory is not receiving **Histo-Logic**, please submit their names to Lab-Tek, Naperville, Illinois, to ensure receipt of our special Tenth Anniversary issue, to be mailed soon.

Possible Causes of the Strong Bond Between Floating Debris and Tissue Section

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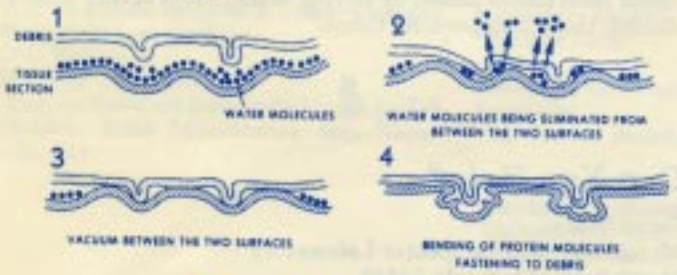
Ed. Note: This information is in response to an editorial on the above subject which appeared in **Histo-Logic**, Vol. IX, No. 1, January 1979.

The tight bond between debris and tissue sections observed after staining may be a submicroscopic mechanical occurrence of suction and clasping. Large protein molecules may have a fairly flexible dome shape.[1] The loosely bound water molecules surrounding the protein molecules[2] may be removed by mechanisms of water movement (e.g., absorption) when in contact with another surface (Fig. 2). A vacuum between the two surfaces forms, resulting in the two surfaces being held tightly next to each other (Fig. 3).

The bending and folding of protein molecules in response to surface bound water molecules are discussed by several authors.[4] The protein molecules may mechanically lock or fasten to the molecules of the other surface by folding and clasping it as the water is removed between them (Fig. 4).

Either one of these events or a combination of both, or the addition of physiochemical forces may also be responsible for the strong bonding of debris and tissue sections. As stressed by Luna[3] further investigation should be made to understand this problem.



BONDING OF FLOATING DEBRIS TO TISSUE SECTIONS

References:

1. Lehninger, A.L.: *Biochemistry*, Second Edition, Worth Publishers Inc., New York, 1976.
2. Kuntz, I.D., and Kauzmann, W.: Hydration of Protein and Polypeptides. Adv. Protein Chem., 28:279, 1974.
3. Luna, L.G.: An Intriguing Question in Search of an Answer. Histo-Logic, Vol. IX, p. 125, 1979.
4. Wolfenden, R.V., Aullis, P.M., and Southgate, C.C.F.: Water, Protein, Folding, and the Genetic Code. Science, 206:575, 1979.

A Method For Keeping Paraffin Blocks Cold Prior to Microtomy

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There are numerous ways of cooling paraffin blocks prior to cutting. Ice and icewater are quite messy, while commercial freezing products are expensive.

My microtome is set-up to the left of a Tissue-Tek II Tissue Embedding Center.* After embedding (60-80 blocks) I remove the embedding molds and place the blocks face down on the cold plate in the order in which they are to be sectioned. This keeps the blocks cold and makes it very easy to section paraffin blocks without the use of other cooling or soaking procedures.

The cold plate on this system will hold approximately 60 blocks. Those blocks which cannot be contained on the cold plate are placed in the drawer just below the cold plate. As space becomes available the blocks from the drawer are transferred to the cold plate.

This procedure works very well in our laboratory and hopefully it will prove helpful to fellow histotechnologists.

*Lab-Tek, Naperville, IL 60540.

Preparation of Cell Blocks Containing No Visible Button

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The consistency of embedded cell blocks often varies. Some are firm and are easily processed with other surgical specimens, while others are loosely formed or not visible at all. These are often impossible to process. The following procedure facilitates handling and processing of all types of cell blocks. This procedure may be used alone or in conjunction with membrane filtration techniques.

Solutions:

Citrated Plasma (Ci-Trol*)
Add 1.0 ml of distilled water to vial, restopper and invert gently to mix.

Activated Thromboplastin†
Calcium Chloride, 0.02 m‡
Thoroughly mix equal amounts of activated thromboplastin and calcium chloride, 0.02 m.

*Ci-Trol, Dade Products B4224-10
†Activated Thromboplastin, Dade B4210-2
‡Calcium Chloride 0.02 m, Dade B4211-2

Procedure:

1. Centrifuge specimen for 10 minutes at 1000-1500 R.P.M.
2. Decant as much supernatant as possible. Leave drop in tip of centrifuge tube if no button is visible.

3. Depending on size of button, add 1-3 drops of freshly prepared Ci-Trol. Agitate gently.
4. Add twice the number of drops (i.e., 2-6 depending on button size) of freshly prepared thromboplastin-calcium chloride.
5. Agitate gently while adding thromboplastin in order to trap entire button in clot.
6. Let stand for 1 minute then add 10% formalin. (Invert test tube gently to insure that artificial clot is completely immersed in formalin.)
7. After fixation, place clot in labeled cassette and process in the conventional manner.

Note:
Clot may be wrapped in tissue paper to prevent loss of specimen. Centrifuge tubes must be rinsed several times in distilled water before use.

The Preparation of Multiple Slides From a Single Block

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Histotechnologists are sometimes required to cut 200 to 300 slides from a single block, or to prepare a serial set of slides. How to prepare a large number of slides from a single block can present a problem to the inexperienced technician, particularly when the block has been cut before and not much material remains. The same is true of preparing a serial set without losing a single section. This paper was prepared to relieve the anxiety neurosis of the technician facing either problem. Both problems can be handled in the same manner.

Materials Required:
1. Any type of pasteboard box, with lid, of sufficient size to hold ribbons of 25 to 30 sections in length, such as a suit or shirt box. I use a fiberboard box 10" to 12" used to file blocks. A large towel-covered tray may be used if the task is to be completed that day and if there is no danger of a draft disturbing the ribbons. Even the small draft caused by a person passing close by is enough to move the ribbons. A box is preferred since the entire block may be cut at once and the ribbons will be protected by the box until the time when they are mounted. The first section and the accession number can be indicated by simply writing in the box.
2. Any good quality single-edge razor blade.
3. 3" x 2" slide. A regular 3" x 1" slide may be used if the block is narrow enough to leave an edge of glass on both sides of the ribbon.
4. Dropper bottle containing 40% alcohol.
5. Fine-pointed forceps and teasing needle.

Method:
A ribbon of any length to fit the box is cut and the ribbon is transferred to the box. It is recommended that forceps be used in the transfer instead of a camel hair brush since the weight of the ribbon will be greater than usual, and there will be no drop of water from the brush to stick the ribbon to the box, resulting in a lost section. For a serial section, the corners of the block should not be trimmed to facilitate ease of separating the sections since the sections may come apart too easily and the order of cutting may be lost.

When the desired number of sections are made, the ribbons are cut with the razor blade into lengths to fit on a slide. A segment of ribbon is then placed on the large glass slide and 40% alcohol is dropped on the slide to float the ribbon. The slide is then tilted sideways to drain the excess alcohol and one end of the slide is slowly lowered into the water bath. The ribbon is guided — or even stretched — from the slide to the water bath using the forceps or needle.

Individual sections may be separated from the ribbon by the use of the slide on which the section is to be placed and a teasing needle. The slide is immersed in the water with the edge at the junction of two sections. The slide is then tilted so the water drains from beneath the desired section, and the needle drawn across the **edge** of the slide (Fig. 1). The slide and section are then separated from the rest of the ribbon and the section oriented on the slide in the desired position. This is repeated until the desired number of slides are obtained.

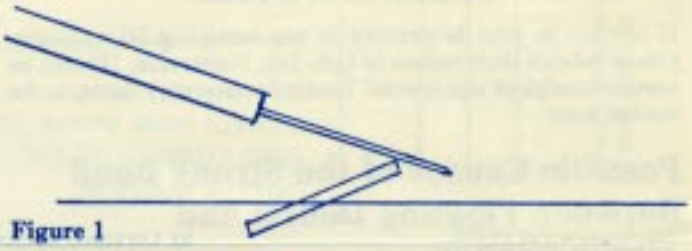

Figure 1

A Box of Salt

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How many times have we, as histologists, performed a silver stain and somehow managed to allow our hands to come in contact with the silver solution. The result is black metallic silver deposits that defy acid alcohol, clorox, sandpaper . . . you name it, the stain remains. Of course this always happens just at a time when black spotty hands are the last thing in the world we want.

The most important item to prevent this problem is a common ordinary box of table salt. Anytime silver nitrate is used (after handling the beaker, coplin jar, or whenever contact is made with the silver), I wet my hands with tap water, pour about a rounded teaspoonful of salt on my hands and rub the solution thoroughly over the hands, rinse and dry. A second application can be used to make certain all areas have been reached.

The salt treatment must be done soon after silver contact in order to prevent silver-stained hands. This ounce of prevention has worked well for me. Consequently I have never been embarrassed by having black, silver spotty hands during those inopportune times.

Can You Help?

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We have heard warnings about formaldehyde and hydrochloric acid in combination forming bis-chloromethyl ether (bis-CME), a carcinogenic substance. Is there an allowed level, and if so, how does this equate to volume utilized by any particular laboratory? Do we as histotechnologists have to be concerned using B_5 fixative? What alternative "safe" fixative can we use that would give a similar result to B_5?

Please forward a copy of your reply to the Editor.

Visual and Audio Aids

This is part three of a four-part article listing various educational aids applicable to the field of histotechnology. Part one, "Text Books," and part two, "Journals and Publications," appeared in the April and July 1980 issues of **Histo-Logic.**

The Essential Laboratory
Skills Series
Audio-Tape with carousel tray, instructor's manual and student workbook.
Unit 12: Histologic Technique I
Unit 13: Histologic Technique II
RMI Learning Systems, Inc.
14 Story Street
Cambridge, MA 02138

Color Filmstrips to Supplement Histology
Thomas S. Leeson & C. Roland Leeson
Film strip I, 50 frames
Film strip II, 50 frames
W. B. Saunders Company
West Washington Square
Philadelphia, PA 19105

28 Grams of Prevention
16 mm movie film
See or write your:
Fisher Scientific Company Representative

Safety in the Clinical Laboratory
Audio Tape
Richard J. Henry, M.D.
Bio-Science Laboratories
Main Laboratory
7600 Tyrone Ave.
Van Nuys, CA 91405

Legionnaires Disease
Audio Tape X 2
American Society for Medical Technology
5555 West Loop South, Suite 200
Bellaire, TX 77401

Tech Sample Program - Histotechnology
Package (6 exercises)
Catalogue #30-1-179
American Society of Clinical Pathologists
P.O. Box 12073
Chicago, IL 60612

Theory and Application of Tissue Embedding in the Histopathology Laboratory
Tissue-Tek II Embedding Film
16 mm movie film
M.D. Anderson Hospital & Tumor Institute
Medical Communications
6723 Bertner
Houston, TX 77025
(Also available on loan basis from Lab-Tek Division, Miles Laboratories, Inc., Naperville, Ill.)

Self-study courses are available from the University of Texas Health Science Center at San Antonio. They include audio cassette tapes with slides and printed study guides when provided. Courses include the following:

Method of Processing, Sectioning and Staining & Application of Mineralized Sections of Bone
by Antonio R. Villanueva

Self Assessment of Fungi - Part I - Part II
by Gerre Wells

An Evaluation of Elastic Tissue Staining
by Coranelle Lambert

A Modified Knife Sharpening Technique for Microtome Knife Sharpeners
by Lyn Richardson

Histotechnology and the Forensic Pathologist
by Frederick Jordan, M.D.

Bone Marrow: How, Why and the Results
by Dale Van Wormer, M.D.

Self Assessment of Special Stains - Part I -Part II
by Dezna Sheehan
(No study guide or slides; designed to be used with Ms. Sheehan's book.)

Cost Accounting - Part I - Part II
by Helen Futch

Carbohydrate Histochemistry
by Tom Palmer, Ph.D.

The Chemistry of Hematoxylin and Eosin Staining - Part I - Part II
by Jules Elias

Formalin Pigment
by Nita Searcy

Laboratory Quality Control Data
by Sue Judge

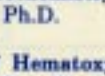
Histologic Techniques in Ophthalmic Pathology
by Carmen Berlanga

Please forward requests to:
Continuing Education Services
University of Texas Health Science Center
7703 Floyd Curl Drive
San Antonio, TX 78284.

The Art & Science of Histotechnology - A Visit to the Laboratory
Career Awareness Film
16 mm movie film or ¾" video cassette
Film was released October 1979, by Lab-Tek Division, Miles Laboratories, Inc., Naperville, Illinois. It was part of a Career Awareness Program conducted at the National Society for Histotechnology's Annual Symposium/Convention. It focuses on the working responsibilities of the histotechnologist and his important role as a member of the diagnostic or investigative team. The primary goal of the film is to show prospective trainee candidates the workings of a histotechnology laboratory. The film is to be used as a visual aid when conducting career awareness programs.

Also, a booklet entitled *The Art & Science of Histotechnology — A Career to Consider* has been developed for persons in a counseling role or schools having a strong vocational interest in histotechnology.

At the present time we are in the transition of switching the source of these materials to the NSH Office. However, until that is completed, materials can be secured from both sources as follows:
Histotechnology Materials
Corporate Training Department
Miles Laboratories, Inc.
1127 Myrtle Street
Elkhart, IN 46515
or
National Society for Histotechnology
P.O. Box 36
Lanham, MD 20601



The following films are available in three different formats: 16 mm color; Sony U-matic videotape cassette; and Super 8 mm in Fairchild cartridge. Rental films available in 16 mm only.

Cytopreparation With Micro Slides

Cytopreparation With Membrane Filters

Fixation in Diagnostic Cytology

Papanicolaou Stain: Principles

Papanicolaou Stain: Materials and Methods

Coverslipping in Diagnostic Cytology

Sputum Specimens: Collection and Preparation

Bronchoscopy Specimens: Collection and Preparation

Contact:
Health and Education Resources, Inc.
4733 Bethesda Ave.
Suite 735
Bethesda, MD 20014.

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Practical Stain-Technology Workshop March 8-13, 1981

Presented By
Center for Histotechnology Training

This five day extensive workshop will afford the registrants the opportunity to utilize 19 special stains demonstrating more than 25 pathologic entities.

Some of the entities being stained are: Gram positive and Gram negative bacteria; Hepatic B antigen (HBAg); fungi; calcium; acidic and sulfated mucosaccharides; elastic fibers; tubercle bacilli; nucleic acids - RNA and DNA; amyloid; cell granules from the neuroendocrine system; copper; connective tissue; reticulum; mucin; spirochetes; Legionnaires disease bacilli; and melanin. Also, the phosphotungstic acid hematoxylin (PTAH), and a hematoxylin and eosin procedure will be performed.

In addition to the practical special staining aspects, lectures will be presented daily on chemistry of staining and staining mechanisms. Lectures on tissue identification will also be conducted daily.

Workshop Objectives:
Upon completion of this course, the registrant will be able to: (1) efficiently and effectively perform 20 special stains; (2) understand the mechanism and chemistry of staining reactions; (3) gain a working knowledge of general and specific pitfalls regarding special stains; (4) gain necessary expertise to determine the quality of stained slides; (5) acquire extensive knowledge for determining shelf-life of staining solutions and solution pitfalls; (6) in addition to the above, registrant will learn how to identify various tissue structures. The latter subject is most important since it allows one to identify the staining qualities of a given structure or entity.

For further information, contact: Registrar; Center for Histotechnology Training; P.O. Box 2453; Rockville, MD 20852.

To receive your own personal copy of HISTO-LOGIC, or to have an associate added to the mailing list, submit home address to: Lab-Tek Division, Miles Laboratories, Inc., 30W475 North Aurora Rd., Naperville, Illinois 60540.
Printed in U.S.A.

The editor wishes to solicit information, questions, and articles relating to histotechnology. Submit these to: Lee G. Luna, Editor, Histo-Logic, P.O. Box 36, Lanham, Maryland 20801. Articles, photographs, etc., will not be returned unless requested in writing when they are submitted.