Histo-Logic in the Beginning...

Brent Riley
Managing Editor

It was sometime in 1969 when Lee Luna, Chairman of the Department of Histopathology at the Armed Forces Institute of Pathology, began thinking about a newsletter for Histotechnologists. He had already thought long and hard about creating a national society and realized that publishing a newsletter was an important part of reaching his goal. Luna knew the vital role that good communication must play in advancing the histotechnology profession.

"Back then, there was no intercommunication anywhere for Histotechnologists," Luna said. "There was no vehicle for disseminating information throughout the country to the histotechnology community."

So Luna began to generate ideas. For the next 2 years, he discussed his idea with other Histotechnologists, developed objectives, created the style, and planned the logistics of publication. And by early 1970 he was ready.

"Creating a newsletter would do two things," Luna explained. "First, it would be a technical information vehicle for the field of histotechnology. Second, it would serve as the foundation for a journal of histotechnology." The first objective was right on target. The second would have been, but it turned out to be unnecessary, as *The Journal of Histotechnology* was later developed by Antonio (Tony) Villanueva, Ph.D.

"Some people thought we should simply contribute our material to already-established journals," Luna recalled. "But the purpose of *Histo-Logic* was not to replace a journal or compete with a journal. It was primarily to provide technical information to the working Histotechnologist. It was not intended for the publishing of lengthy research papers that are usually of interest to a limited percentage of readers. I wanted to publish information that would be of interest to at least 80 percent of our readers. And because *Histo-Logic* is primarily comprised of bench-type technical information, most people are interested in everything we publish."

Luna knew that the publication he envisioned would require a sponsor, so he approached Bill Williams, who was the Eastern Region Manager for Lab-Tek Products Division of Miles Laboratories, Inc. Williams recognized the value of the idea and, without hesitation, took the idea to Bob Myers, Marketing Manager at Lab-Tek.

Myers was impressed by Williams' enthusiasm. He recognized the potential and knew that newsletters had been very successful with other product lines at the company. But he wanted to meet Luna and discuss the project in more detail.

A short time later, Myers and Williams were sitting in Luna's AFIP office in Washington discussing the newsletter. "I was very impressed with the sincerity and absolute dedication that Lee exuded," Myers recalled. "He really sold me."

"When we went to see Lee," Myers continued, "I was expecting to meet a very flamboyant individual. What I found was a quiet, sincere, and very professional man. My only remaining concern was whether we could get enough articles to make the newsletter worthwhile. But Lee already had files full of papers."

According to Luna, the key to the success of any newsletter is being able to produce enough material. He believes that the editor of such a publication must have the ability to fill in when there is not sufficient information for an issue.

For some time, Luna had been encouraging fellow Histotechnologists to write papers on various tips and methodologies. He kept them all on file, knowing that someday he would publish them. Some of the papers had been read at one of the AFIP meetings, but most had never been published.

Before *Histo-Logic*, there was no exclusive outlet for histology papers. Such papers were sometimes published in other professional journals such as *Stain Technology* or those sponsored by the ASMT, ASCP, or CAP.

Luna was also prepared with a mailing list. He had been compiling names and addresses for several months and had about 5,000 names for the first issue. Lab-Tek sales representatives also accumulated names from their customer lists. After the first issue, the mailing list grew quickly as a form was inserted in the publication asking readers to send the names of friends and colleagues.

At the time, *Histo-Logic* was a quarterly publication. Luna served as the editor and many of the details of publishing were handled by Myers' secretary, Shirley Winters. Luna would submit the articles to Winters, who would create a layout, then coordinate printing and distribution.

Today, *Histo-Logic* is read by an estimated 25,000 Histotechnologists, Pathologists, and other healthcare professionals across the United States and in at least 44 foreign countries. Using *Histo-Logic* as a model, many countries have created their own histotechnology newsletters. Many articles are translated and published in Japanese, German, and other languages.

Twenty years ago, Lee Luna had a vision. Through his enthusiastic efforts, that vision spread throughout the histotechnology community. He was confident that *Histo-Logic* would succeed. But even Luna is surprised at the high level of success that *Histo-Logic* has ultimately achieved.

Histo-Logic: What Makes It Happen?

Brent Riley
Managing Editor

You might think that publishing an issue of *Histo-Logic* isn't all that difficult. After all, it's just a matter of writing a few articles, printing them up, and mailing them out. Right? Well, not exactly.

The truth is, there's a lot more to it than that. From start to finish, hundreds of hours are invested in every issue of *Histo-Logic*. At least 30 people are involved in its publication and distribution. And planning for a single issue might begin as much as 6 months before it appears in your mailbox.

From the beginning, *Histo-Logic* has been sponsored by Miles Inc. The process begins with Lee Luna, the editor of *Histo-Logic* since its inception in 1971. The following three major *Histo-Logic* personnel are all employees of Miles Inc., Diagnostics Division. Brent Riley, Senior Marketing Manager for Cellular Diagnostics, serves as Managing Editor. John Emgenbroich, Communications Manager, and Alison Buckley, Project Planner, are both in Marketing Communications.

Luna is responsible for the scientific articles published in *Histo-Logic*. That means being constantly on the lookout for ideas — new tips and techniques that would be of value to the average Histotechnologist. Because Histotechnologists are an innovative group, they are always seeking solutions to problems, trying new approaches, and attempting to inject more accuracy and efficiency into routine tasks.

Through his lectures, travel, and communications with Histotechnologists around the world, Luna typically becomes aware of new ideas and techniques immediately. If he were a newspaper reporter you might say he has a "nose for news." Luna then contacts the innovative Histotechnologist and asks him or her to write a brief article about the idea. According to Luna, most people are honored to receive such a request and are very willing to contribute. While most are initiated in this way, Luna also receives many unsolicited articles.

Many of the scientific articles in *Histo-Logic* are authored by Luna himself. As a devoted Histotechnologist, he has developed many tips and techniques to simplify and improve various procedures.

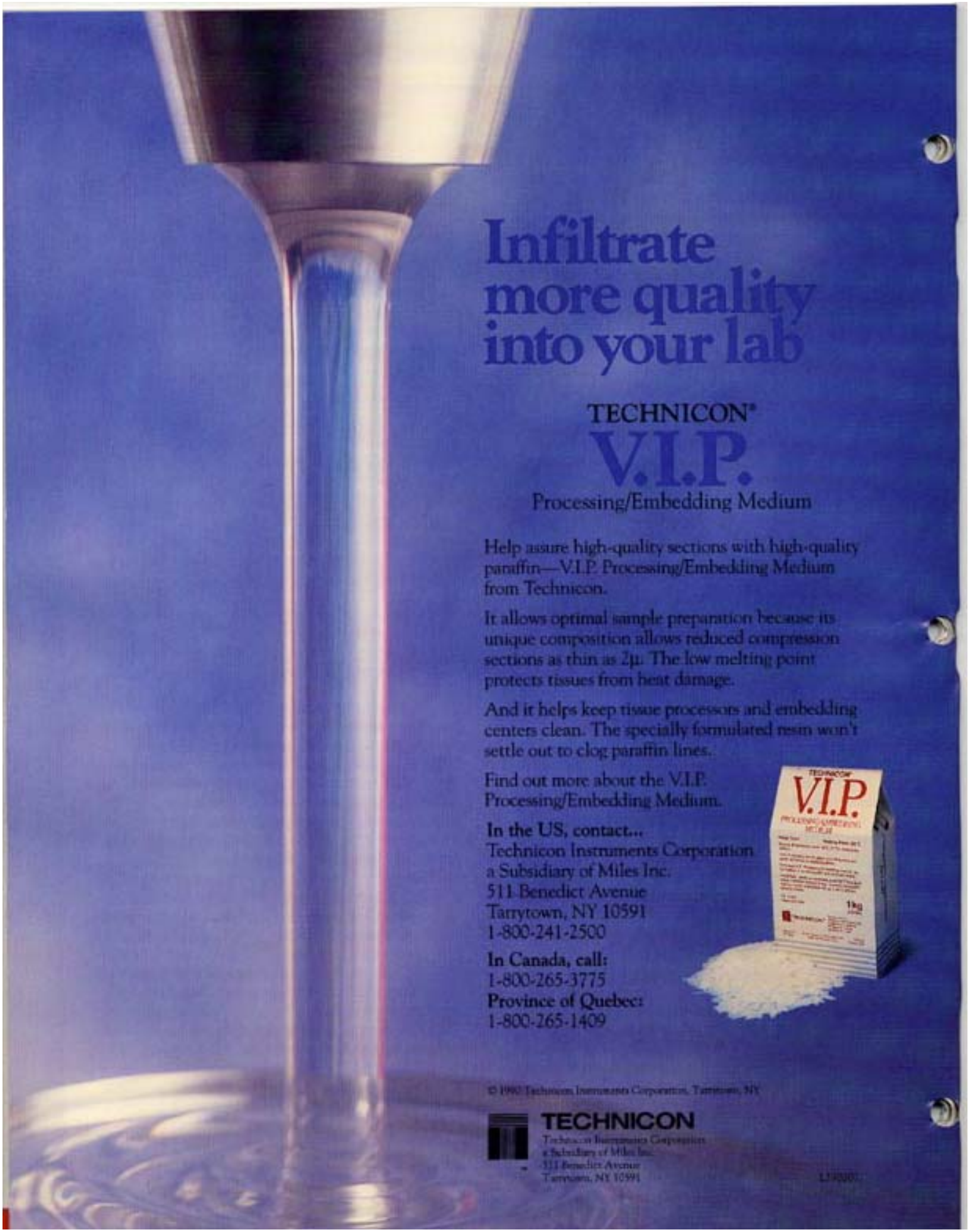
Preparing the scientific material for *Histo-Logic* has become somewhat of a family affair for Luna. No less than five other members of the Luna family are involved. One is Roberta Mosedale, executive secretary for the NSH, who is also Luna's sister-in-law. Mosedale is responsible for responding to the authors of any articles submitted for publication. When an unsolicited article is received, she sends a letter of thanks. She also confirms with the author that the material is original and previously unpublished.

Mosedale submits the articles to Luna, who decides their appropriateness and edits them. She also types the articles that are authored and dictated by Luna.

After editing, Luna turns over the articles to his daughter-in-law, Nancy Luna, who enters them into a computer. She determines when there is enough material for a complete issue of *Histo-Logic*. The material is then reviewed and re-edited by both Luna and Mosedale.

When the articles are ready to be published, Luna turns them over to his daughter, Alita, who sends them to a

(continued on page 269)



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Chicago advertising agency that prepares the material for printing. Meanwhile, Luna's son, Darryl, creates diagrams and helps take color photographs that support the articles.

As Managing Editor, Riley is responsible for the non-technical news and feature articles that appear in *Histo-Logic*. Riley writes some of the articles himself, but many are written by Thom Gill, an independent medical writer from South Bend, Indiana. Gill has been writing for *Histo-Logic* for the past 6 years. He also writes for several other healthcare companies and is a member of the NSH.

For each issue, Riley, Gill, and Marketing Communications at Miles discuss potential articles. They look for ideas that are of professional or human interest. For example, *Histo-Logic* typically includes feature articles on NSH award winners, NSH committees, the national symposium/convention, management advice, and various issues faced by the histotechnology profession.

Riley or Gill then research the articles, looking up historical information and interviewing people who are involved in the particular subject matter. These articles are combined with the scientific articles to make up an issue.

All the articles are proofread and typeset. A comprehensive layout (comp) is then created that serves as a mock-up of how the final printed issue will appear. This comp is sent to Lee Luna, Brent Riley, Marketing Communications, Roberta Mosedale, and Thom Gill, who proofread it and may suggest changes. The suggested changes are made and a second comp is sent to the same group of people. When the final changes are made, camera-ready keyline boards are prepared and sent to a printer.

Over 17,000 copies are printed for United States distribution. First, mailing labels are applied. The copies are then collated and delivered to the post office. All of this happens under the direction of Emgenbroich and Buckley.

As you can see, publishing *Histo-Logic* is not a simple task. It takes time, skill, experience, hard work, and dedication from a lot of people. But anyone who sees the success of *Histo-Logic* and the enthusiasm of its readers must agree that it is well worth the effort.

Editor's Note: The following are portions of letters that transpired among the originators of *Histo-Logic* in 1971.

Your plans for a histology newsletter exceeded my expectations. The desire on your part to disseminate the vast store of useful information you have is admirable and Lab-Tek is pleased we can help. We have a good deal of work ahead of us, but if desire can accomplish the task we will mail the first issue July 1.

March 10, 1971 — Robert F. Myers

We are extremely pleased with the progress of *Histo-Logic*. I will be looking forward to receiving the copy for the first two issues. Charlie Kalt has received your invitation to exhibit at the next AFIP Seminar and you will be hearing from John and Bill. We look forward to exhibiting as well as sponsoring a social hour prior to the banquet. I understand there is a dedication ceremony May 21.

May 3, 1971 — Robert F. Myers

The manuscript and mailing list arrived right on schedule. We will do all in our power to have the proof ready for you prior to the ASMT meeting. The enlarged mailing list causes no problem and is greatly appreciated. We will see that questions related to printed material or submission of proposed manuscripts will be sent to you with your box number and that mailing list additions will be sent to you.

May 18, 1971 — Robert F. Myers

Your manuscript was received August 17. Thank you for being prompt. I have read the manuscript and can see we have an excellent second issue. Every attempt will be made to include the comments you submitted for the Editor's Corner. We also have received quite a few. To say the least, it looks like we have a hit on our hands.

August 23, 1971 — Robert F. Myers

Histo-Logic, Vol. 1, No. 2, is in the final stages of production. We appreciate the additional names for the mailing list. I have attached copies of the comments we received relative to the first issue. I am sure that these will continue to be received. We will not be able to include more than three or four quotes in the Editor's Corner; we have simply run out of room. As you can see by the attached layout, it is completely filled.

September 24, 1971 — Robert F. Myers

The Golden Forceps Award: An Important Part of *Histo-Logic*

Brent Riley
Managing Editor

Every year, scientific articles from authors throughout the country and around the world are submitted for publication in *Histo-Logic*. Most of them are eventually published. But of those that are published, only one is chosen to receive the prestigious Golden Forceps Award.

The Golden Forceps Award is presented annually to the author of the best article published in *Histo-Logic* between June 1 and May 31. Choosing the Golden Forceps Award winner is the responsibility of the NSH Awards Committee. When judging, the committee considers the originality, clarity, content, and scientific contribution to the field of histotechnology.

The NSH Awards Committee is organized to eliminate bias in the selection process. While the chairman is known to the NSH membership, the rest of the committee members are not. They are appointed by the chairman, but their names remain confidential.

The award consists of an all-expense paid trip to the Annual NSH Symposium/Convention, as well as a plaque. It was first presented in 1973. While the purpose of the Golden Forceps Award has essentially remained constant over the years, other aspects of the award have not. For the first eight years, the award was given to an author published in *Histo-Logic*. In 1981, and for the next five years, the award was presented for articles published in *The Journal of Histotechnology*.

In 1987, a new award, the Diamond Cover Award, was established for the *Journal*. From that point on, the Golden Forceps Award was again reserved for *Histo-Logic*.

The first winner of the Golden Forceps Award was Elaine Boyd. Her winning article, published in a 1973 issue, was titled, "An Evaluation of Problems in Lymph Node Preparation."

Here is a complete list of Golden Forceps Award winners:

1973 - Elaine Boyd, "An Evaluation of Problems in Lymph Node Preparation," *Histo-Logic*
1974 - Therese Ansman, "The Vacuum Infiltrator in Routine Fixation," *Histo-Logic*
1975 - Louis Chang, Ph.D., "A Silver Technique for the Study of Cellular Injuries," *Histo-Logic*
1976 - Edna Prophet, "Technique for Processing Eye Specimens," *Histo-Logic*
1977 - Livia Molnar, presented for numerous articles published in *Histo-Logic*
1978 - Anna Marie England, "Gram-Positive and Gram-Negative Bacteria Control," *Histo-Logic*
1979 - Cathy Adler, "Gelatin-Chrome Alum: A Better Section Adhesive," *Histo-Logic*
1980 - Charles Churukian, presented for numerous articles published in *Histo-Logic*
1981 - Ada Feldman, "The Chemistry of Fixation & Staining: Basic Concepts," *The Journal of Histotechnology*
1982 - Kathleen Spencer, "Micro-Organisms: The Perfect Control," *The Journal of Histotechnology*
1983 - Ernestene Sims, "Whole Organ Processing with the Tissue-Tek Vacuum Infiltration Processor (V.I.P.)," *The Journal of Histotechnology*
1984 - Paul Duray, M.D., "A Simplified Azo Dye Method for the Demonstration of Acid Phosphatase in Paraffin-Embedded Tissue," *The Journal of Histotechnology*
1985 - F. S. Waldrop; Holde Puchtler, M.D.; Susan N. Meloan, "Fluorescent Thiazole Stains for Amyloid Without Differentiation," *The Journal of Histotechnology*
1986 - L. K. Pugliese & C. Anderson, "A Method for the Determination of the Relative Distribution and Quantity of Mineral in Bone Sections," *The Journal of Histotechnology*
1987 - Mabel Pierce, "Connective Tissue Staining in Water Soluble Methacrylate," *Histo-Logic*
1988 - Cheryl H. Crowder, "Helpful Hints for Microwave Oven Use: More Than Just Staining," *Histo-Logic*
1989 - Terri Staples, "Methods for Staining *Campylobacter Pylori*," *Histo-Logic*
1990 - Leonard Noble, "A Simplified and Reliable Technique for the Demonstration of *Pneumocystis carinii* in Cytoprep Smears," *Histo-Logic*

After 18 years, the Golden Forceps Award will continue to be a measure of excellence, a reward for outstanding work, and an enduring honor.

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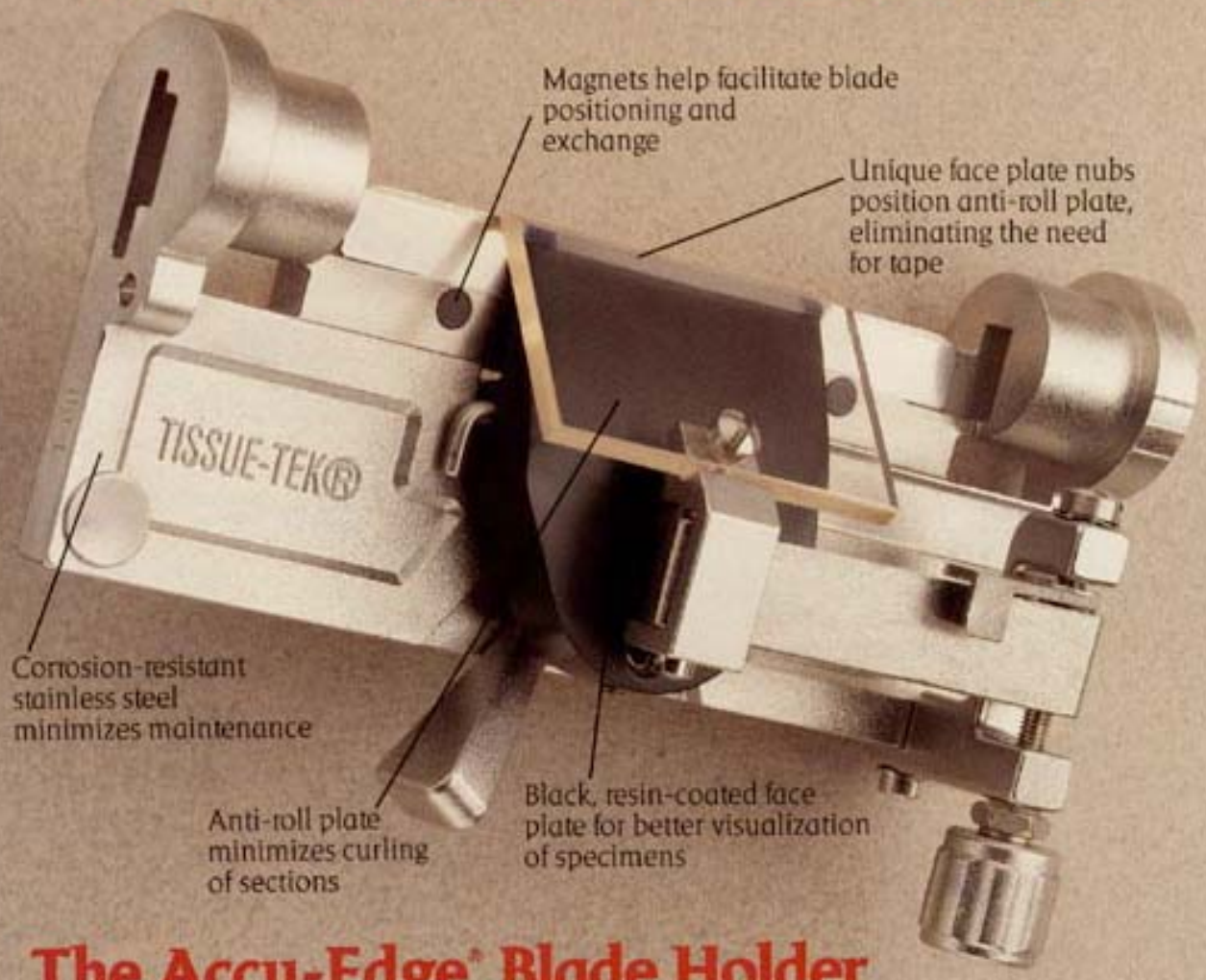

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Make It Better...Again

Brent Riley
Managing Editor

Some people we meet in life are never satisfied with simple answers or easy achievements. For many years now, Lee Luna has been one of those people.

Lee's philosophy of improvement has been the major factor in the success of *Histo-Logic* — its transformation from a four-page, single-color publication to a twenty-page, four-color newsletter, complete with four-color photomicrographs.

Even before the first issue was in the mail, Lee's plan for the first wave of improvements was already taking shape. In the very next issue of *Histo-Logic*, he enhanced the design to improve readability. Over the next few years, Lee's efforts focused on maintaining a consistent publication schedule while he continued to improve the scientific quality of the articles that appeared.

In 1976, Lee suggested the first of many paper stock changes for better aesthetics and suggested that a comic illustration be included. It is recalled that Lee grinned and stated, "Reading *Histo-Logic* should be fun as well as educational." Lee's attention then shifted to boosting circulation, both in the United States and overseas. His efforts resulted in the first Japanese-language issue in 1979, followed by the first German-language issue in 1981.

Lee's commitment to improvement was established at our first meeting in 1983 when I became involved with *Histo-Logic*. Improvements from two to four colors; from eight to twelve to sixteen to twenty pages; from expanded scientific articles to professional and human-interest articles — all are due to Lee's personal philosophy of improvement... making it better, again.

As you read and review the original issue of *Histo-Logic* that appears in the following pages, I hope you take a moment to compare current issues and appreciate the many design and editorial improvements that are an extension of Lee Luna's philosophy of and commitment to constant improvement.

Thank you, Lee Luna, and Happy 20th Birthday, *Histo-Logic*.

Reflections and Comments

Following are some current reflections and comments relating to the articles that appeared in the original issue of *Histo-Logic* (July 1971) — a reproduction of which is reprinted in its entirety for your interest and review.

Method for Destaining Aldehyde Fuchsin

Comment: 1991

This article introduced a destaining method that is still very useful today because aldehyde fuchsin continues to be used extensively in demonstrating Hepatitis B surface antigen, beta cells in the pancreas, elastic fibers, etc.

Zenker's Fixation and Related Problems

Comment: 1991

This article is perhaps more pertinent today than it was in 1971. Zenker's fixative was becoming obsolete until the advent of immunochemical staining procedures. Today, Zenker's is used extensively to preserve antigens, so the problems associated with its use are still of great concern.

Restoration of Nuclear Basophilic Properties

Comment: 1991

This article introduced a method developed by Lee Luna for restoring nuclear basophilic properties. It is still an important procedure today because, even with modern techniques, we have not resolved the problem of destroying nuclear properties.

A Modified One Hour Giemsa

Comment: 1991

This technique was developed by Elbert Gaffney. It remains a valid technique today, particularly for bacteria. It is commonly used for the demonstration of *helicobacter pylori*.

Substitutes for Ethyl Alcohol in Histologic Techniques

Comment: 1991

The study discussed in this article demonstrated the practicality of substituting other types of alcohol for ethyl alcohol. Although a number of new dehydrating agents have been introduced since 1971, the substitution of isopropyl alcohol for ethyl alcohol is still a common practice because it is less expensive, and because it is not a controlled chemical.

HISTO-LOGIC™

Lee G. Luna, D. Lit., H.T. (ASCP), Editor — A Technical Bulletin for Histotechnology — Vol. I No. 1 – July, 1971

HISTO-LOGIC is a publication devoted exclusively to the dissemination of information pertinent to the field of histotechnology. We are fortunate indeed that Lee G. Luna has agreed to edit this new publication. His background, experience, and deep dedication to the profession — and its problems — will bring to the reader a great deal of useful, relevant material.

Since histotechnology is a relatively new art and science, it is felt that there are many messages to be delivered, many mutual problems to be discussed, and in general, a need to establish standards for the profession through a communications "clearing house."

Hopefully HISTO-LOGIC can serve, at least in part, to help fill this void by providing a continuing professional histology-oriented newsletter.

Charles J. Kalt
President, Lab-Tek Products
Div. Miles Laboratories, Inc.

Letter of Intent

The birth of histologic technique dates back to 1664 when Robert Hooke cut sections of cork with his pen knife and observed them microscopically. In 1670, Leeuwenhoek made sections from a writing feather, a bovine optic nerve, and the centers of dried flowers by using his hand-sharpened shaving razor. These historic events were the beginning of what has become, in recent years, an exciting laboratory discipline. Many historical developments could be mentioned; let it suffice to say that the greatest advances in this laboratory field have been made in the 20th Century and particularly during the last two decades. Among the notables of this period were Doctors Frank Mallory, George Gomori and R. D. Lillie. In addition to microscopic investigations of pathological conditions, these dedicated individuals also conducted experiments in histological procedures and staining technologies. Although many accomplishments are evident as the result of the tremendous work conducted by these individuals, there has never been an established system of intercommunication within the field of histological technique.

Two important steps forward in the dissemination of information in the field of histopathology took place in 1965 and 1966. In 1965 a seminar on histopathological technique was sponsored by the North Florida Pathologist Association. This significant program was conceived and promoted by Mrs. Barbara Spillan, Chief Histopathology Technician at the U.S. Naval Hospital, Jacksonville, Florida. To my knowledge this was the first seminar ever presented in this country devoted exclusively to this particular laboratory discipline. In the fall of 1966 the Armed Forces Institute of Pathology conducted its first "Symposium on Histopathologic Technique." This symposium has become an annual event and is designed to: (1) Disseminate information of new technical developments; (2) Stimulate interest for the establishment of seminars, symposiums and/or workshops in various sections of the country; (3) Make histotechnologists aware of the many interesting and stimulating opportunities available in their field. These goals have been achieved beyond expectation. However, the means of reaching all histotechnologists for a more thorough exchange of information is still deficient.

To a great extent HISTO-LOGIC Newsletter will fulfill this gap. It will also serve the needs of those individuals who are unable to attend the more than 15 seminars, workshops and symposiums conducted annually in this country.

The purpose of this newsletter is:

1. **Interchange of Technical Information.** To be successful we need your contribution of technical information of a practical nature.
2. **Establishment of Tissue Control Centers.** To successfully accomplish this, it is hoped that hospital laboratories will make known when ample control material is available for dissemination to other laboratories.
3. **Technical Inquiry and Reply Service.** Questions concerning histopathologic methodology and procedures are welcomed. Replies will be published. Those inquiries which cannot be answered readily will also be published in order to obtain reader's opinions and conclusions.
4. **Announce Meetings Related to Histotechnology.** Salient information regarding meetings will be published. Requirements for such an announcement will be nature of the meeting, society, location, date and any additional pertinent information.
5. **Provide Information.** Information the editor feels will enhance the knowledge of the histotechnologist and the field of histopathology will be published.

The success or failure of this newsletter is dependent upon the cooperation of **all** technicians and technologists. Your help is solicited by subscribing, but more importantly by contributing your technical suggestions, modifications and/or developments.

Lee G. Luna, D. Lit., H.T. (ASCP), Editor

Method for Destaining Aldehyde Fuchsin

Aldehyde fuchsin is used for demonstration of elastic fibers, Paget cells, mucosaccharides, pancreatic cells, etc. Because of its wide use it is desirable to have a simple method of removal from previously stained tissue sections on which one desires to perform a different stain.

Potassium Permanganate Solution

Potassium permanganate	0.25 gm
Distilled water	100.00 ml

Sodium Bisulfite Solution

Sodium bisulfite	5.0 gm
Distilled water	100.0 ml

Procedure

1. Deparaffinize and hydrate to distilled water.
2. Place in potassium permanganate solution for 1 minute.
3. Rinse in distilled water.
4. Place in sodium bisulfite solution until section is clear (usually several minutes).
5. Wash in running tap water for 5 minutes.
6. Stain as desired.

Zenker's Fixation and Related Problems

Zenker's fixative has been used for many years for bone marrow, surgical and biopsy specimens. Its primary value lies in its chemical effect on tissue which results in better preservation and differentiation of cell and cell particles. This is due in part to the chemical action of the following ingredients on specific cell structures:

Mercuric chloride, facilitates staining with most dyes, making the colors more brilliant.

Potassium dichromate, fixes cytoplasm without precipitation, producing a homogeneous cytoplasm which reacts well with eosin-type dyes.

Acetic acid, gives life-like preservation to nuclei, penetrates rapidly and swells cell constituents making them more visible microscopically.

These results are only possible if fixation time is controlled. This is best controlled if one understands a hypothesis developed regarding tissue changes during fixation: Tissues undergo three distinct changes during Zenker's fixation. During the first stage, nuclei shrink considerably. Microscopically the nuclear chromatin appears as one dense mass. This normally takes place during the first 2 to 3 hours. This is followed by swelling of the nuclei to approximately their original shape and size. Microscopically the nuclear particles are very distinct as are all cell products. This desirable effect takes place after the first three hours. The third change relates to staining qualities, particularly hematoxylin and eosin preparations. Tissues overexposed to Zenker's fixative will not exhibit the usual hematoxylin staining characteristics. Overexposure (8 hours or longer) produces an acidic staining appearance. Chromatin material cannot be differentiated chromatically and in general nuclei are not distinct. Staining qualities can be restored by placing the deparaffinized slides in a 10% aqueous solution of sodium bicarbonate overnight, followed by routine hematoxylin and eosin staining. Additional problems related to Zenker's fixative are listed below.

Mercuric Chloride Crystals

Mercuric chloride is an ingredient of Zenker's fixative and deposits in tissue as so-called "Zenker crystals." In the classic form, these crystals are spheroid with an irregular periphery. The crystals have also been observed in needle forms and are often polymorphic. Because of the deposition of uncharacteristic forms, they can be confused with other crystalline material. The following method will differentiate mercuric chloride (Zenker) from other crystals which may be present in tissue.

Staining Method for Mercuric Chloride Crystals

20% Ammonium Hydroxide Solution

Ammonium hydroxide, 28%	20.0 ml
Distilled water	80.0 ml

0.2% Light Green Solution

Light green, SF yellowish	0.2 gm
Distilled water	100.0 ml

(Add several crystals of thymol as a preservative.)

Staining Procedure

1. Deparaffinize and hydrate to distilled water.
2. Twenty percent (20%) ammonium hydroxide solution for 30 seconds.
3. Rinse in distilled water (5 dips).
4. Counterstain in light green solution for 30 seconds.
5. Dehydrate in two changes of 95% alcohol, absolute alcohol, and clear in xylene.
6. Coverslip.

Results

Mercuric chloride (Zenker) crystals	Black
Background	Light green

Remarks

The mercuric chloride crystal color reaction will fade after several weeks. Sections cannot be retreated with the ammonium hydroxide solution if the original reaction is light.

Basic information concerning the reaction was obtained from: Nebergall, W. H. and Schmidt, F. C.: "General Chemistry," D. C. Heath and Company, Boston, Massachusetts, 580, 1959.

Erythrocytic Crystallization

Zenker's fixative will often crystallize erythrocytes. Microscopically the RBC's will exhibit a transparent or a light translucent appearance and possess excellent polarization properties.

Mucosaccharide Alteration

Buffered neutral formalin does not produce any gross alterations affecting the staining properties of hyaluronic acid. Zenker's fixative has an adverse effect on hyaluronic acid and cannot be demonstrated by staining with colloidal iron type preparations. It is not known if other mucosubstances are similarly affected.

Nucleoli Chromatic Alteration

Frequently, nucleoli stain blue after Zenker's fixation. This is in contrast to other fixatives where nucleoli normally stain red after well differentiated hematoxylin and eosin staining.

These remarks are not intended to suggest that Zenker is a poor fixative. But the above information should, if applied, prevent many existing problems related to Zenker's fixation.

Restoration of Nuclear Basophilic Properties

One of the most important aspects of histologic technique is proper staining. Properly fixed, washed, decalcified, and processed specimens pose no staining problems. Frequently, however, one encounters specimens overexposed to unbuffered formalin, Zenker's, Bouin's and/or a decalcifying fluid which subsequently prevents nuclei from staining adequately. In these instances, one of the following methods can be employed to restore the nuclear staining properties of most specimens:

Zenker's Fixative

1. Place deparaffinized sections into 10% aqueous sodium bicarbonate for 6 to 8 hours.
2. Wash slides in running water for 10 minutes.
3. Stain with hematoxylin and eosin.

Unbuffered Formalin and/or Decalcification

For specimens overexposed to unbuffered formalin and/or decalcifying agents, the following method has proven the most useful.

1. Stain deparaffinized sections in Weigert's hematoxylin for 30 minutes.
2. Rinse sections in running tap water.

NOTE: If staining results are too dark, decolorize in 1% acid alcohol (1 ml HCL, 99 ml 80% alcohol). Wash slides in running tap water for 15 minutes.

3. Counterstain with eosin.

Understand that some overly decalcified bone specimens will not stain regardless of the restoration method used.

Weigert's Iron Hematoxylin Solutions

Solution A

Hematoxylin crystals	1.0 gm
Alcohol, 95%	q.s. to 100.0 ml

Solution B

Ferric chloride, 29% aqueous	4.0 ml
Distilled water	95.0 ml
Hydrochloric acid, concentrated	1.0 ml

Working Solution

Mix equal parts of Solution A and B and stir vigorously.

Bouin's Fixative

1. Place slides in 5% aqueous lithium carbonate solution for 16 hours.
2. Wash for 30 minutes in running tap water.
3. Stain with hematoxylin and eosin.

The best remedy is prevention. Poor nuclear staining properties can be avoided by prompt removal of specimens from the fixative and /or decalcifying fluid soon after complete fixation or decalcification has been achieved.

A Modified One Hour Giemsa

Elbert Gaffney, Washington, D.C.

The following modification has proven to be a simple, consistent, and reproducible stain for all types of tissue. The staining results are good and seldom vary regardless of the tissue being stained or the technician performing the procedure.

Fixation

Zenker's solution or 10% buffered neutral formalin.

Solutions

Azure II Eosin Solution

Place a few sterile glass beads in a 500 ml amber bottle.

Azure II eosin (Harleco)	1.3 gm
Glycerin	80.0 ml

Incubate at 56°-60° for 2 hours. Mix and cool.

Add: Methanol	170.0 ml
Acetone	170.0 ml

May-Greenwald Stain Solution

Combine in one liter Erlenmeyer flask.

May-Greenwald stain (Harleco)	0.15 gm
Methanol	290.00 ml
Acetone	290.00 ml

Combine Azure II eosin and May-Greenwald solutions. This stock solution can be used immediately but improves with aging.

Acetic Water Solution

Glacial acetic acid	0.01 ml
Distilled water	1000.00 ml

Working Giemsa Solution

Giemsa solution (stock)	10.0 ml
Acetic water	50.0 ml

Staining Procedure

1. Deparaffinize and hydrate to distilled water.
2. If Zenker fixed, remove mercuric chloride crystals with iodine and sodium thiosulfate (Hypo) in the usual manner.
3. Stain in Giemsa working solution for one hour.
4. Dehydrate in absolute alcohol, three changes.
5. Clear in xylene, three changes.
6. Coverslip.

Results

Nuclei	Blue
Erythrocytes	Red
Mast cell granules	Purple
Cytoplasm	Pink

Substitutes for Ethyl Alcohol in Histologic Techniques

Thomas C. Allen, B.S., Washington, D.C.

A study was conducted to determine where methanol and isopropyl may be used as substitutes for ethyl alcohol in histological techniques.

Specimens of skin and liver were obtained from an autopsy (human) and macrosectioned into blocks approximately 4 millimeters thick.

Fixation

To determine the comparative effects of isopropyl, methanol and ethyl on fresh tissue, specimens were placed in the following for 48 hours. (Also provided are the overall section results.)

1. 10% buffered neutral formalin — Good sections produced.
2. Isopropyl alcohol — Severe shrinkage, especially on the peripheral margins of the tissue.
3. Ethyl alcohol — Moderate shrinkage.
4. Methanol — Slight shrinkage.
5. Formol-alcohol — Good sections produced.

40% formalin	10 ml
Alcohol	90 ml
Glacial acetic acid	5 ml

Comment

Isopropyl and ethyl alcohol should not be used for routine fixation of tissue specimens. Methanol should be used when the preservation of glycogen is desired. Formol-alcohol can be used to expedite fixation especially of biopsy and surgical type tissue specimens.

Tissue Processing

No noticeable tissue differences could be detected among the three alcohols. On the basis of this, isopropyl alcohol and methanol can be used in place of ethyl for tissue dehydration when either xylene or chloroform is used as a clearing agent.

Special Stains

Isopropyl alcohol and methanol were used in the following special stains in the steps calling for ethyl alcohol: Fontana-Masson, Manuel's method for reticulum, Mallory's iron, Masson's trichrome, and periodic acid Schiff.

No significant adverse effects were noted in the above stains except in the Masson trichrome. The aniline blue and biebrich scarlet-acid fuchsin demonstrated collagen and muscle rather light, and therefore methanol and isopropyl should not be used for this technique.

This brief study proved that it is possible to utilize isopropyl alcohol and methanol as substitutes for ethyl in many procedures.

Additional methods and procedures should be attempted.

HISTO-LOGIC™
A Technical Bulletin for Histotechnology Vol. 1 No. 1
LTM 21-721 Printed in the U.S.A.

Editor's Corner

7th Annual Symposium on Histopathologic Techniques
4-8 October 1971

Armed Forces Institute of Pathology, Washington, D.C.

	Morning	Afternoon	Evening
Mon., 4 Oct.	Special Stains Workshop		-----
Tues., 5 Oct.	Tissue Identification Workshop		Exhibits 7-9 P.M.
Wed., 6 Oct.	Lectures/Exhibits	Lectures	Cryotomy & Microtomy
Thurs., 7 Oct.	Lectures/Exhibits	Optional	Cocktails & Banquet
Fri., 8 Oct.	Lectures	Lectures	-----

All activities will be held at the Sheraton Motor Inn, Silver Spring, Maryland.

For information write: Director, Armed Forces Institute of Pathology, Wash., D.C. 20305 ATTN: Lee G. Luna, Rm. 254.

ASCP Conducts "Good Histology Workshop"

The American Society of Clinical Pathologists will present a "Good Histology Workshop," 28 October 1971. The workshop will be in two 3½ hour sessions and will cover histochemistry, stain technology, and related histopathological procedures.

For information write: American Society of Clinical Pathologists, 1300 West Harrison St., Chicago, Illinois 60612.

California Histopathology Technologists' Installation Banquet

The California Society of Histopathology Technologists' Installation Banquet will be held 23 July 1971. The banquet speaker will be Mr. Lee G. Luna, Chief, Histopathology Laboratories, Armed Forces Institute of Pathology, Washington, D.C. For information write: Miss G. Wilma Cline, President, C.S.H.T., 227 DeAnza, San Gabriel, California 91776.

The editor wishes to solicit information, questions, and articles relating to histotechnology. Submit these to: Lee G. Luna, Editor, Histo-Logic, Box 552, Hyattsville, Maryland 20782. Articles, photographs, etc., will not be returned unless requested in writing when they are submitted.

To receive your own personal copy of HISTO-LOGIC, or to have an associate added to the mailing list, write: Lab-Tek Products, Division Miles Laboratories, Inc., 39 E. Burlington, Westmont, Illinois 60559.

Letters Received as a Result of the First Mailing of *Histo-Logic*, July 1971

May I personally add a note of appreciation to you at Lab-Tek and to Mr. Luna. A newsletter in this form has been clearly needed by all of us for a long time. Well done!
— Elizabeth M. Mayle, HT(ASCP), University of Rochester, School of Medicine & Dentistry, Rochester, New York

I have just received a copy of No. 1, Volume III — January 1973 of *Histo-Logic*. The technical and practical information that *Histo-Logic* contains is very useful to our work at the Pan American Health Organization. I would, therefore, appreciate receiving a complete set of previous issues, and would be interested to know whether this publication is available in Spanish.
— Humberto Torloni, M.D., Pan American Health Organization, Washington, D.C.

I was recently assigned as supervisor of histopathology in our hospital and came across a copy of *Histo-Logic*, and I must say I am impressed that you are sponsoring a fine bulletin for us "block-choppers." When I read the name of the editor, I was doubly impressed. I was a student under the tutelage of Mr. Luna when I was stationed at the AFIP. You could not have made a wiser choice. This bulletin is a terrific way of updating procedures and exchanging techniques.
— Dominic Rotondo, USAF Regional Hospital, Maxwell, Alabama

Note of Apology

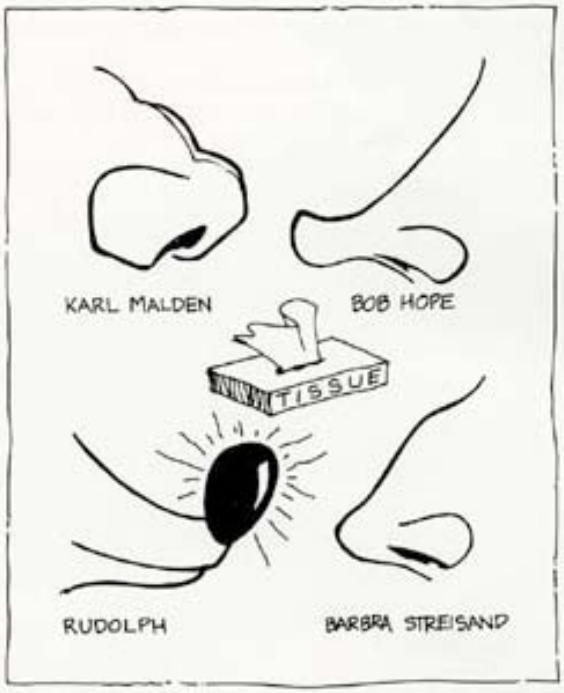
Over the last three years, many of my colleagues have sent copies of slides and numerous questions relative to the field of histotechnology. While I have responded to many of these inquiries, I have been concerned about those that have gone unanswered. My apologies are extended to anyone who has not received a reply. Please understand this has not been from lack of concern for your technical problems, but rather due to illness.

— Lee G. Luna

Note of Thanks

This is to thank the worldwide histotechnology community for their concern during my illness over the last three years. During this period I have received hundreds of cards and letters with wonderful notes of encouragement. I know of no one who, in their final comments, did not say they were praying for me. Your warm, uplifting messages, telephone calls, plants and beautiful flowers have been very graciously received. As a young boy, my mother instilled in me the philosophy that I should never be afraid to embrace any member of the human race. Through your expressions of love, this embrace has been returned one hundred fold. Thank you for your concern and affection. They have been a sustaining power over the past few years.

—Lee G. Luna



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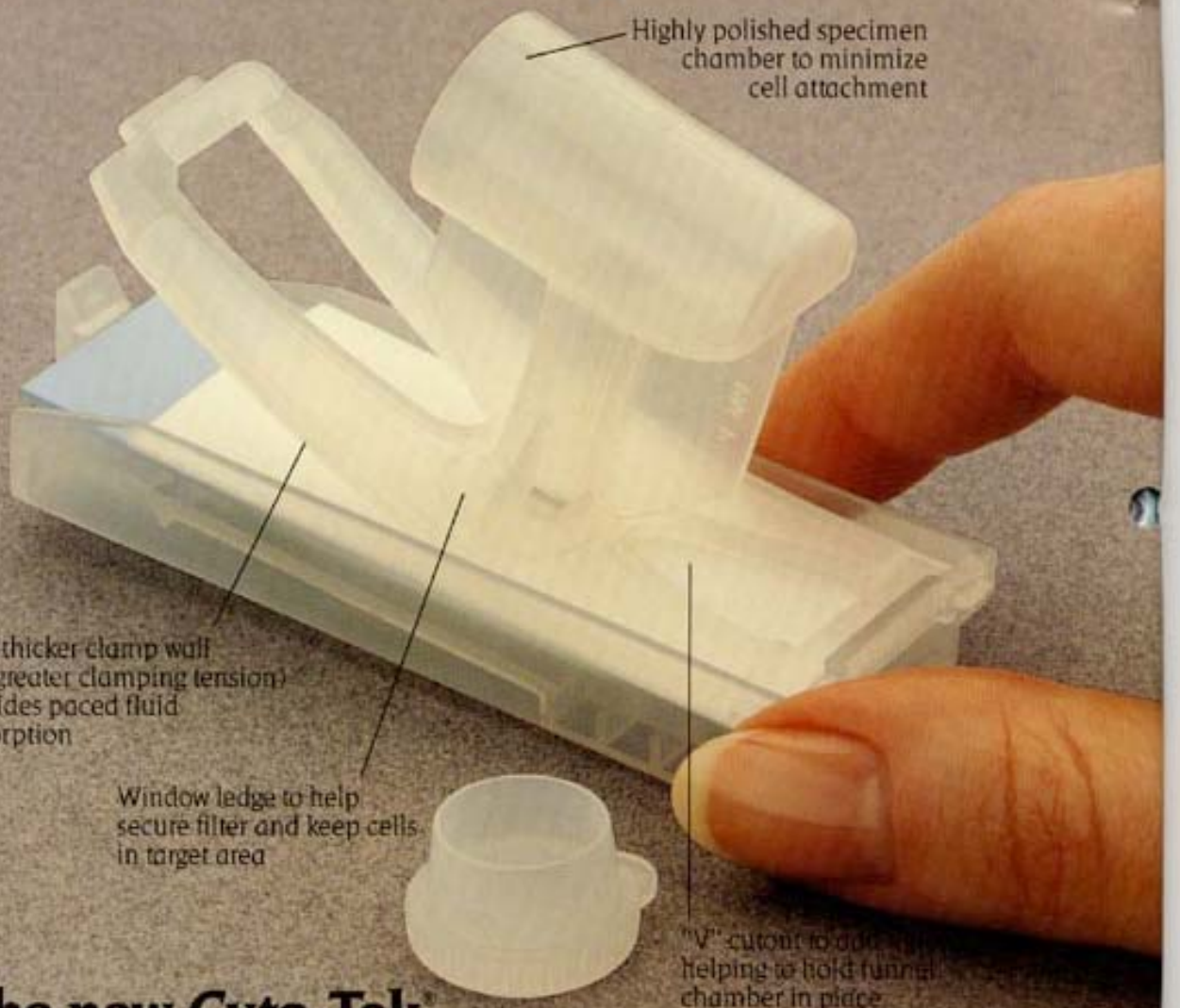

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1991 Kudos

• It has been a very rewarding experience to have been involved with the production of *Histo-Logic* since its inception in 1971. Working with editor Lee Luna, in fulfilling a dream of bringing histotechnology to the attention of the healthcare system, we have witnessed fruition of a profession that is now recognized worldwide. Through publication of *Histo-Logic*, the first link in a chain uniting the histology community was formed.

It is inconceivable that just 20 years ago there were no communiqués for histology, there were few programs, only two state societies, no schools of histotechnology, national or international societies. With the development of *Histo-Logic*, the process of change became inevitable. Those in the profession began to feel a sense of camaraderie in knowing there were others across the nation with whom they could communicate. *Histo-Logic* gave them an opportunity to learn new procedures, ask questions, share knowledge, and begin the evolutionary process of joining together in becoming one voice.

My personal involvement with *Histo-Logic* began as a favor, helping an overextended editor complete secretarial tasks. This resulted in a myriad of responsibilities that continually grew as *Histo-Logic* became a reality. Together we were responsible for preparing articles, typing, editing, rewriting, formatting, paste-up, and layout. Lee and I would work together at least one evening every week and sometimes met on weekends to discuss the progress of completing an issue. In the early years, maintaining the address list also fell within my jurisdiction. One horrendous task I shall never forget was initiating the mailing list for the first *Histo-Logic*. This was accomplished by using the U.S. Hospital Directory and manually tabulating the name and address of hospitals throughout the United States with an in-service histology lab. Abstracting this information was the only way to guarantee every hospital laboratory received the first technical newsletter for their profession. Yes, computerization is a phenomenal advancement to this system!

As evidenced today, these arduous tasks have been more than successful. By producing that first link, the chain now reaches around the world with a society in nearly every state, four international societies, accredited schools of histotechnology, technical and state newsletters, training programs, workshops, lectures, teleconferences and, of course, the National Society for Histotechnology.

I am grateful for the opportunity of sharing in the birth of these activities. I am even more grateful for the tutelage, support, and constant encouragement of the editor, Lee Luna. Our working together has always been an enlightening and enjoyable pleasure. Special thanks also to Miles Inc. for their 20th year of production. With a mailing list reaching around the globe, *Histo-Logic* continues to be received without charge.

Twenty years of revolutionary progress! Through the visionary efforts of Lee Luna, his hard work, knowledge of the profession, solicitation of support and belief in its reality, *Histo-Logic* was born in 1971. I am honored to be associated with its production and to have contributed in a small measure to its success. Happy Birthday, *Histo-Logic!*
— Roberta Mosedale, Executive Secretary, National Society for Histotechnology, Lanham, Maryland

• *Histo-Logic* came on the scene 20 years ago when there was a dearth of scientific publication in our field. Although we now have *The Journal of Histotechnology*, *Histo-Logic* continues to provide "bullets" of meaningful information that I am delighted to receive.
— Diane Burica, HT/HTL (ASCP), MS/MA, Lutheran General Hospital, Park Ridge, Illinois

• In the 1960s the only vehicle for journal publication of a staining method (or, more likely, an improvement thereof) was *Stain Technology*. Well, most of us histologists with something to say were intimidated by the thought of submitting a manuscript (I know I was) because we believed our information didn't meet all the requirements of such an established journal. Lee Luna and the people at Miles, like Brent Riley, were all too cognizant of this dilemma, and, to their credit, they went ahead and established *Histo-Logic*. It opened the door into the world of the publishing process for many of us.

I look back with fond memories and pride to my first publication in *Histo-Logic* — a combined trichrome-spirochete stain. This experience provided the impetus for me to go on to publish future articles, not only in *Histo-Logic* but in established journals. I'm sure other members of NSH had the same experience. Perhaps more importantly, *Histo-Logic*'s success encouraged Dr. Antonio

(Tony) Villanueva to establish a journal under the aegis of NSH. *The Journal of Histotechnology* is now recognized outside the community of Histotechnologists and is representative of what professional Histotechnologists can do.

As the present editor of *The Journal of Histotechnology*, I believe that *Histo-Logic* complements us in providing a platform for the dissemination of useful information, and many of our members submit articles to both publications.

Congratulations to Lee, Brent, and Tony for giving us our voice.
— Jules Elias, Ph.D., Editor, *The Journal of Histotechnology*, Stonybrook, NY

• Since its inception 20 years ago, *Histo-Logic* has served an important function in the dissemination of a wealth of information to many thousands of persons engaged in the art of histotechnology. It has been my privilege to have contributed several articles to the publication. Many thanks to Lee Luna for an excellent job as Editor, and to Miles Inc., Diagnostics Division, for their generous continuing support for publication of *Histo-Logic*. Their efforts and commitment have benefited many.
— Charles J. Churukian, B.A., HT/HTL(ASCP), University of Rochester Medical Center, Rochester, New York

• Looking back 20 years ago, when the first issue of *Histo-Logic* was published, one realizes that the world has taken a "quantum leap" in acquiring and disseminating knowledge in all disciplines. There is such a proliferation of knowledge, even with the sophisticated communication systems, it is difficult to stay abreast of the most current literature in any given field.

Fortunately, for the histology technician, *Histo-Logic* is there. It has grown from a newsletter of a few articles relative to the laboratory to a world-renowned source of information for the technician. It now serves as a networking tool. It keeps technicians informed of what is going on in other laboratories, what new procedures have been developed, what new technology and research exists, what the NSH and state societies are doing, and it addresses a myriad of other concerns of the technician.

I was fortunate to have contributed to the first issue of *Histo-Logic*. This publication, being the first of its kind, has had a significant impact in histotechnology. Whether it is addressing issues of legislation, education, technology, laboratory procedure, or some other issue, *Histo-Logic* fosters the recognition of the histology technician as a true professional in the medical laboratory field. Twenty years later it continues to be the major source of news and information for the histology technician.
—Thomas C. Allen, B.S., HT(ASCP), AFIP, Washington, D.C.

• For many years I have advocated the dissemination and sharing of histotechnological information and in a manner that speaks to practical needs and concerns of the bench laboratorian. Since its inception, I believe that *Histo-Logic* has served as an invaluable resource to this end.

I was privileged to publish an article in the first edition of *Histo-Logic* and look forward to publishing in future editions.
— Elbert Gaffney, MT/HT (ASCP), AFIP, Washington, D.C.

• Congratulations to Lee, Brent, and Miles Inc., as well as the many authors who have contributed to the continuous flow of highly informative articles for the past 20 years. *Histo-Logic* has been and continues to be an invaluable source of education, serving the field of histotechnology.
— Ken Urban, HT(ASCP), Surgipath Medical Industries, Inc., Grayslake, Illinois

• I recall my feelings of elation when the first issue of *Histo-Logic* was published. It was exciting to, finally, have a nationwide mode of communication for histology professionals! *Histo-Logic* played a significant role in the eventual formation of the National Society for Histotechnology and has continued to support our Society in many ways. Every issue has been read and enjoyed! Thank you for your immeasurable contributions to the field of histotechnology. Happy 20th Anniversary!
— Marilyn Gamble, HT/HTL (ASCP), President, National Society for Histotechnology, North Hollywood, California

• In 1969, when I entered the Histotechnology profession, *Stain Technology* and *The Journal of Histochemistry and Cytochemistry* were the main publications I was encouraged to read. Although the articles appearing in these journals were excellent in content, they did not answer a basic need of the practicing Histotechnologist. A vehicle did not exist where Histotechnologists could share ideas and procedures with their peers in a published format. In 1971, *Histo-Logic* became that much-needed publication.

Ever since the first issue was being planned, Histotechnologists have been able to share their procedural hints, modifications of existing stains, newly developed procedures, and safety tips. I personally value the information contained in *Histo-Logic* and am honored to have been published there.

Mr. Luna and Miles Inc., Diagnostics Division, started a tradition when they printed the first issue of *Histo-Logic*. I for one hope that the tradition continues.
—Leonard W. Noble, Jr., HT/HTL (ASCP), North Carolina Baptist Hospitals, Inc., Winston-Salem, North Carolina

• Congratulations, Lee and Miles Inc., on the 20th-year anniversary of *Histo-Logic*. It certainly has been an important educational tool for the profession of histotechnology.

Due to your modesty I'm sure there are not too many people who are aware that it was your vision of the AFIP Symposium developing into a national society for histotechnology, and that *Histo-Logic* would ultimately grow into or serve as impetus for a national journal for our profession. It has done that, as well as continue to be an important communiqué. Without your foresight, hard work, and dedication, I wonder where our profession would be today.

Again, congratulations on reaching yet another high point in your career.
—Don Hammer, HTL (ASCP), University of Washington Medical Center, Seattle, Washington

International Kudos

• The chairman, committee, and members of the Histotechnology Group of N.S.W., Australia, wish to congratulate *Histo-Logic* and its Scientific Editor on 20 years of continuous service to Histotechnology both in the United States of America and all other areas of the Globe for the invaluable information the pages have presented over this period.

I would also like to thank Lee Luna for his kind words about this group in the Jan/Feb 1991 edition of *Histo-Logic* and the reproduction of the cover of our quarterly newsletter.
— Bill Sinai, Chairman, Histotechnology Group of N.S.W., Australia

• The publication of *Histo-Logic* has given to Histotechnologists a better understanding of this diverse discipline. Mainly through the efforts of Lee Luna, a close bond was formed in Australia with *Histo-Logic*, which is distributed by Bayer to most Histopathology Laboratories.

To Lee Luna I send the deepest praise for his devotion and encouragement in the formative years of our own Histotechnology Group. Lee visited Australia on several occasions to spread the word relating to the principles of Histotechnology from which we all greatly benefited.

Congratulations on 20 years of logical histotechnology and thanks to those who have contributed.
— Bruce Munro, University of Sydney, Australia

• Brent, congratulations to you and Lee Luna on the occasion of the 20th Anniversary of *Histo-Logic*.

On behalf of my New Zealand colleagues we wish you all the best and continue to look forward to many more publications supporting HISTOLOGY.
— G.E. Bongiovanni, New Zealand

• Congratulations on the celebration of the 20th anniversary of the publication of *Histo-Logic!* It is indeed an honor as editor and translator of the Japanese version of *Histo-Logic* since 1973 to tell you how much *Histo-Logic* has taught Japanese histologists and assisted in developing technical expertise in histology. May I extend sincere appreciation to Miles Inc. and Mr. Lee Luna for their great contribution to histotechnology on behalf of all the Japanese readers. I hope that you will continue to supply news and useful scientific information to Japanese readers in pathology and histology.
— A. Hirayama, M.D., Translator, Japanese version of *Histo-Logic*, Japan

(continued on page 278)

• On behalf of the Editor, Managing Editor, Editorial Committee, and the readers of "Tissue Talk," I wish to convey our sincere wishes and congratulations to the Editor and Editorial Committee of *Histo-Logic* on the occasion of their attaining 20 years of scientific publication.

The merit of such a publication has exemplified its worth in providing a communication vehicle between all persons involved in some related area of histotechnology.

Its dissemination to many countries throughout the world has been a practical avenue of interlaboratory communication with contributions being published from numerous countries.

Special mention must be made of the Editor, Mr. Lee Luna, whose vision to produce such a publication was made possible by his enthusiasm and personal commitment to advancing the professional acceptance of histotechnology.

Congratulations may also go to Miles Inc.—U.S.A., for their continued support of the profession that has been identified by the real commitment they made 20 years ago to support the establishment of *Histo-Logic* and continued to expand upon by aiding its dissemination throughout the world.
— Keith Cole, Editor, "Tissue Talk," Australia

• We thank you for your continuous labor of helping the Pathologists and Histotechnologists of all the Spanish laboratories.

Your comments and articles about new techniques from all the hospital specialties and universities have been very useful for us.

We hope that the trajectory will follow as beneficial as at the moment.
—Rafael de Solis, Spain

The above photograph demonstrates the evolution of *Histo-Logic*. On the left is the most recent issue of *Histo-Logic* and on the right the original July/August 1971 issue. The significant change with current issues is that color photographs are now used to emphasize the value of a stain procedure or entity being demonstrated.

Just Another Fun-Time Reminder

The Miles Party!...
at the 1991 NSH Symposium Convention

All dressed up and no where to go?

Histotechnologists from around the world are invited to attend the Miles STAR NITE CAST PARTY. This is the 17th Annual NSH Symposium Convention which will be held in Orlando, Florida, from September 28 to October 4, 1991.

It's a gala event, destined to make the glitterati Hollywood press writhe with envy, squirm with delight, and report with words of wrath any and all goings on.

(continued on page 280)

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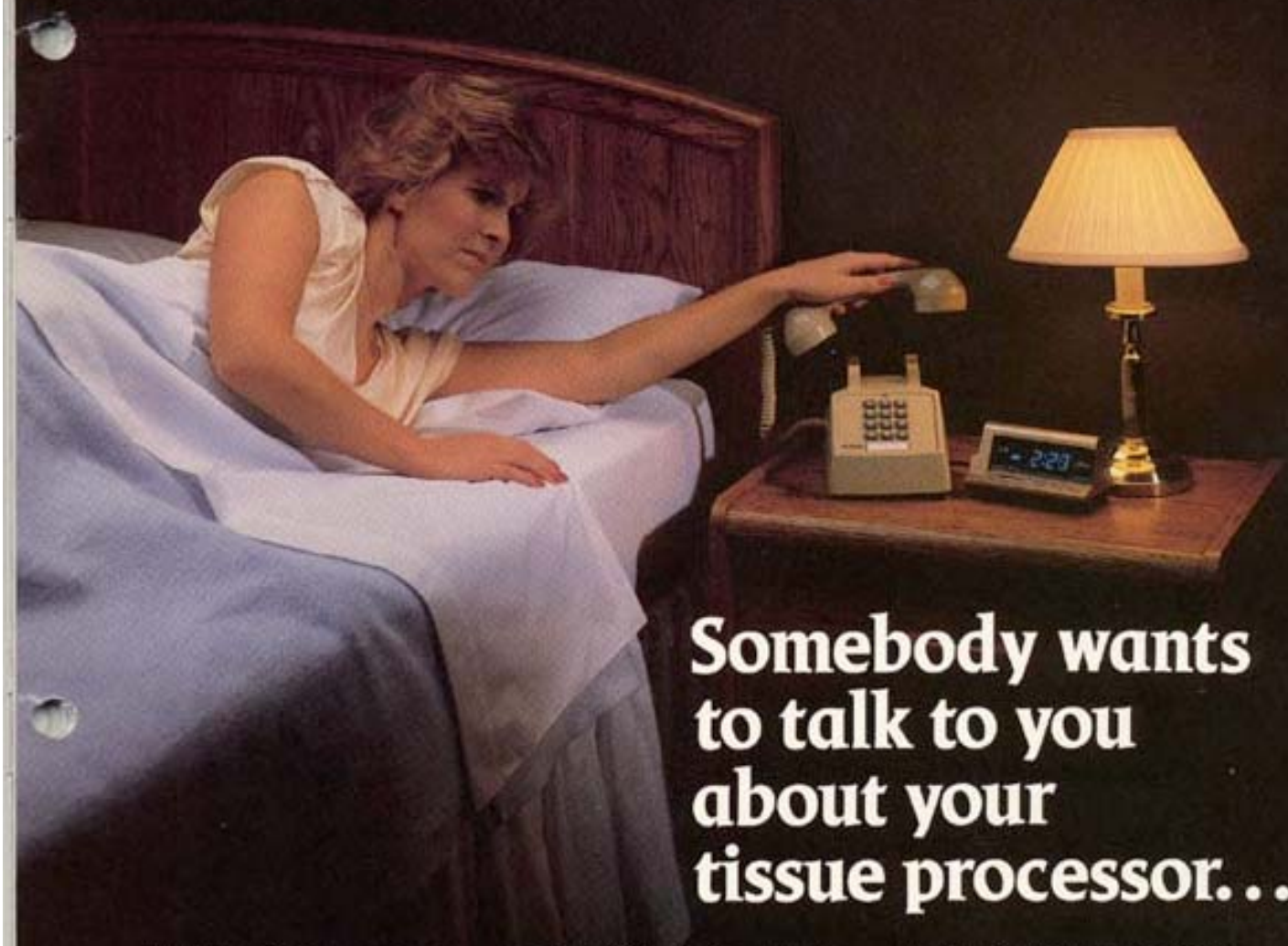

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The gala Hollywood Ball follows immediately. And refreshments aplenty will abound.

So "cast about a bit" for just the right costume. See you there, movie fans...and

"Here's looking at you, kid."

To receive your own copy of *Histo-Logic*® or to have someone added to the mailing list, submit home address to: Miles Inc., Diagnostics Division, P.O. Box 70, Elkhart, IN 46515.

The editor wishes to solicit information, questions, and articles relating to histotechnology. Submit these to: Lee G. Luna, *Histo-Logic* Editor, 7605-F, Airpark Rd., Gaithersburg, MD 20879. Articles, photographs, etc., will not be returned unless requested in writing when they are submitted.

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