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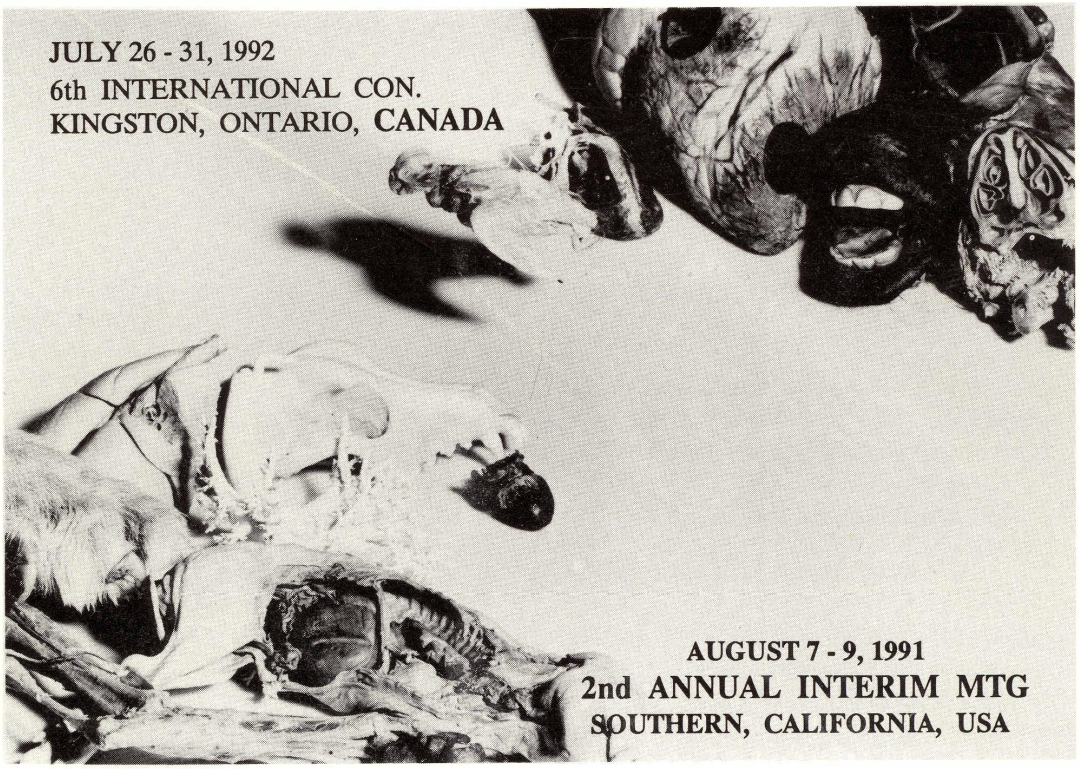
INTERNATIONAL SOCIETY for PLASTINATION



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What a GREAT place and meeting, **HEIDELBERG**. The 5th Conference for Plastination was truly a success in every way. Plans are in process to make the 6th International Conference even better. Approval has been granted by Queen's University in beautiful **Kingston, Ontario, CANADA** for the Conference to convene at the University. The date is set for **July 26 - 31, 1992**. Please place these dates on your calendar and begin preparation to participate. For a keep in touch and update sessions, the **2nd Annual Interim Meeting of the International Society for Plastination** will be held in **Southern California** at **Chaffey College**. The dates will be **August 7 - 9, 1991**, Wednesday through Friday. This promises to be another great event. We are starting a question/answer column and a column for tips. If you have a question which you can not get an answer to or if you have questions recently answered that you think is beneficial to others please send it to the Journal. Any time savers or tips would be welcome. We hope this will be a mechanism to distribute small items of interest. We appreciate those of you who were able to send your manuscripts on disc. It has saved considerable hours of typing and retyping. I would like to encourage you to continue this form of submission on disc. Please let us know how the Journal can better serve you. To try and serve you better, it was proposed and passed at the 5th International Meeting to establish an editorial board. The appointments to the board are nearing completion. A partial listing of appointees appears in the preliminary pages of this issue. We encourage you to consider color figures for your manuscripts. We do ask you to help with publication costs. The approximate fees are listed in the "Instructions to Author" section. Help your association: encourage your library to subscribe to the Journal.

CHAFFEY COLLEGE *NEWS RELEASE !!*

2ND INTERIM PLASTINATION MEETING

The International Society for Plastination announces plans for the 2nd Interim Plastination Meeting. Scheduled for **August 7, 8, 9, 1991**, the meeting will take place at **Chaffey College** in **Rancho Cucamonga, California**. A series of lectures and hands-on lab demonstrations will be featured. The basic S/10 technique as well as advanced techniques will be presented. The meeting will benefit both novice and expert alike. Chaffey College is located about 40 miles east of Los Angeles and about 60 miles from Palm Springs. The College is ten miles from Ontario International Airport and is convenient to hotels, restaurants and shopping. Special room rates will be available for meeting guests. A shuttle will be provided to and from hotels for attendees. Continued on page 32!

THE 5TH INTERNATIONAL CONFERENCE ON PLASTINATION

Heidelberg, Germany, July 22-27, 1990

It hardly seems possible! - - After all that planning and anticipation, the Heidelberg conference is history. But most would agree that it's the kind of history that will be remembered for a long time.

Attendance was outstanding. Slightly over 200 attendants from 42 countries were officially registered. It had the feeling of a large meeting with the personal warmth of a smaller gathering of friends. And the program was equally impressive with 40 speakers, 20 demonstrations and an extensive, post-conference tour that included Munich, Vienna and Rome. We are all indebted to Andrea Whalley, Gunther von Hagens and their hard-working staff for a job well-done. Thanks also to the tour hosts, Alfred Riepertinger, Martin Lischka and Maurizio Ripani.

The Pathologisches Institut of the University of Heidelberg proved to be an ideal meeting site. The auditorium was exactly the right size, comfortable and well-equipped. Spacious, sunlit corridors provided a generous amount of room for displays and casual conversation. And, equally important, it was accessible, from almost anywhere in Heidelberg by public transportation. Our thanks to the administration of the Pathologisches Institut for the opportunity to use this wonderful facility.

And who can forget the Riverboat Party? Plastinators seem to have a better-than-average ability to generate merriment. There was dining, dancing and plenty of conversation among people who might never have met one another were it not for a mutual interest in plastination. To crown an evening of gaiety, suitable awards were presented to the "Funniest Plastinators in the World."

Highlights of the Conference were too numerous to describe in detail, but mention must be made of the dramatic unveiling of the plastinated man -- a whole body done in silicone. That's going to be a "hard act to follow" but we all have two years to prepare.

The next **(6th International)** conference is planned for **Kingston, Ontario, Canada**, which sounds like another winner. The exact dates **(between the 20th and the end of July of 1992)** will be announced in the near future. Mark your calendars now so you can't possibly forget.

Write if you are interested in joining the International Society for Plastination. If I don't hear from you before, I'll see you at the 6th International Conference.

By: Harmon Bickley

MINUTES of the meeting of the
International Society for Plastination

Heidelberg, Germany
Thursday, July 26, 1990.

The meeting was called to order at 8:30 AM.

The director presented a report on the status of the society:

A. 72 members are on record for the Jan-Dec, 1990 period.
B. The operating account has a balance of 3,862 American dollars.
C. The principal source of income is membership dues.
D. The principal expenses are the printing of the journal, the mailing of journals, the providing of sample copies of the journal to those interested in beginning plastination and the purchase of stationery.

A motion to change the frequency of the International Conference from bi-annual to an annual occurrence was defeated. International conferences (and society meetings) will continue on a bi-annual schedule.

A motion to divide the conference into two concurrent programs, one for beginners and one for advanced plastinators, was defeated. The conference format will remain as now practiced.

A motion to request that speakers provide an abstract of the important points of each talk in advance and that such abstracts be distributed (either by publication in the journal or as a handout) was approved unanimously. Future conferences will incorporate this feature.

There was a consensus that fifteen-minute presentations were practicable if there was the need to include a large number of speakers, however some means, such as a bell or light, should be used to keep the speakers on schedule.

After considerable discussion, the site for the next (6th) International Conference on Plastination was established as Kingston, Ontario, Canada. Blake Gubbins will present this as a proposal to the administration of Queen's University and the membership will be advised of the outcome when a decision has been made.

It was decided to authorize the appointment of an editorial board for the Journal.

The meeting was adjourned at 9:30.

Respectfully submitted,
Harmon Bickley,
Executive Director

POST CONFERENCE TOUR:

Following the conference, an international group of travelers set out from Heidelberg on Saturday, July 28th, by rail for a week tour of Munich, Vienna, and Rome. South Africa, Canada, Iceland, Japan, Mexico, Honduras, and the USA were represented. At Munich, we were greeted by Alfred and Ralf and escorted to the Einhorn Hotel near the station. The balance of the afternoon was spent in individual pursuits in and around the Bavarian capital. My preference was the Deutsches Museum and in the evening to imbibe some of the famous Munich refreshment.

Our tour of the plastination facilities at the pathology institute began on Sunday morning. We were appropriately impressed by the facilities, but even more outstanding was the manner in which the specimens were displayed. The walls of a hallway, on the lower level, were given over to glass fronted cabinets to accommodate their varied collections. Many of the specimens were color injected or stained and pleasing to look at even if one had no interest in specimens for medical education or research. The color conservation advantages of Kaiserling's solution were demonstrated and the safety and environment protecting features that have been incorporated into their facility were explained. After refreshments and considerable "shop talk", we adjourned to a nearby restaurant for a typical Bavarian meal. A bus tour of the city topped off our stay in Munich.

Monday morning, we entrained for Vienna and spent most of the day rolling eastward through the highly productive agricultural area north of the Alps. We were promptly welcomed by C. Stradal and settled in our nearby accommodations. After a briefing of the events planned for Tuesday, we were allowed to go our individual ways. Prime among our concerns was any place that had air conditioning. Most of Europe was suffering a spate of heat and humidity that would have seemed extreme even for a resident of Bangkok or St. Louis, Missouri. (These conditions persisted for the remainder of the trip and it is the opinion of the writer that any group that can travel under these conditions and continue to speak to each other in a civil manner deserves the "von Hagen's Medal" for heroic action in the name of plastination.) A tour of the *Institute of the History of Medicine* at the University of Vienna was the high point of our stay in Austria. The extensive display of antique Italian medical models was a revelation. The collection is over 200 years old. Not only the wax models themselves but also the individual inlaid cabinets in which they rested were works of art. The colors in the models have been preserved through these many years. Some exaggeration of the general lymphatic system was noticed and a wholly fictional display of an extensive lymphatic system of the brain gives one a jolt at first viewing. At the time of their manufacture, it was assumed that lymphatics were present in that area also. Many of the problems of birth were depicted, as

Magnetic resonance imaging *gif* plastinated specimens: Study of tissue characteristics and evaluation of the hardening process of the S10 technique. C.A.C. Baptista, R.W. Henry,[1] G. Williams* R. Brinker,[2] Medical College of Ohio, Toledo, OH, [1]The University of Tennessee, Knoxville, TN; [2]Medical College of Ohio, Toledo, OH, USA.

Magnetic resonance (MR) imaging, a new, flexible tool for clinical diagnosis, provides diagnostic quality images. While the DEC microVAX workstation software has been developed to analyze and to study MR images. To document that the MR images obtained from the GE signa MR scanner reproduce the anatomy of the scanned organ accurately, plastinated specimens seemed ideal for this purpose. Both pre and post-plastination organs were scanned. The pre-plastinated, scanned organs were hearts perfused with a mixture of gelatine and para-magnetic contrast (gadopentetate dimeglumine). The post-plastinated scanned S10 specimens were lungs and brains which were scanned at known intervals after exposure to the curing agent (S3). All specimens were scanned in a GE Signa 1.5 Tesla MR Scanner and the images stored in a Vax 3600 computer. Pre-plastinated specimens scanned nicely. Post-plastinated specimens yielded good images prior to curing and up to 10 days of exposure to gas cure. From this point, as curing proceeded, the quality and quantitative value of the MR images declined proportionately. Conclusions of our investigation include: (1) The scanning of phantoms, for example, cadaver hearts, verified that MR reproduced the anatomical detail of the scanned organ; (2) Plastinated organs were useful for interpreting MR images and thin sections of plastinated organs maintain their shape and form; (3) Since, the MR images are generated from "vibration" of hydrogen protons, it was better to scan the organs before plastination: (A) Quality of the MR image obtained from plastinated specimens had an indirect relationship to the time of hardening of the specimen. (B) Imaging of plastinated specimens should be limited to the beginning of the hardening process, that is, between the withdrawal of the specimens from the impregnation chamber and up to 10 days of hardening.

Plastination as an additional option in anatomy museum preparation. Russell Barnett, Anatomy Museum, University of Otago, New Zealand.

Plastination was introduced to the Department of Anatomy at the University of Otago in such a way as to demonstrate to our teaching staff the potential this material has for a medical curriculum. We are currently involved with only the S10 and PEM27 plastination techniques but as the realization of the advantages of the technique increases, so to is the demand for plastinated specimens and a need for expansion of or facilities. Although we are involved with various other preservation techniques, which some believe for security and ethical reasons still have their place, I believe the use of plastinated specimens will prove to be far more beneficial in the long term.

Preparation of holograms from plastinated specimens. Harmon Bickley, Mercer University School of Medicine, Macon, GA, USA.

Because of their striking three-dimensional quality, it is reasonable to expect that holograms would make excellent teaching media for medical subjects such as anatomy and pathology. The preparation of holograms from fresh or fixed human soft tissue, however, is almost impossible (without an extremely expensive pulse laser) because of the long photographic exposure time required and the unusual vulnerability of the process to even the slightest movement. Quite in contrast, plastinated specimens, which are comparatively rigid and practically free of water, make ideal subjects for holography. Holograms have been prepared (by Dr. Bert Myers of the Veterans Administration Medical Center, New Orleans, Louisiana, USA) using plastinated specimens provided by Mercer University School of Medicine. Although they represent the first efforts in a project of this kind, these holograms have proven useful as teaching media. With further development in holographic technology, master holograms of even higher quality could be prepared and copies could be shared among all medical teaching institutions.

Plastination of old formalin fixed specimens. Mario Cannes, P. Fuda, L Padella, A. Rosate, Universita Degli Di Torinto, Torino, ITALY.

This paper offers an alternative for the study of gross anatomy for Medical Schools experiencing a scarcity of organs for various reasons. The objective of our work was to use old formalin fixed specimens as a source of material for plastination. This resulted from a decreased organ and body supply in Torino and much of Italy, even with an organized Organ Donation Program. Fortunately at the University of Torino Medical School, we had a large collection of formalin-fixed specimens, preserved from previous years anatomical dissections. The standard S10 procedure was used, so the really different aspect was represented by the specimen source. These specimens had not been subjected to perfusion and washing as indicated in the plastination manual nor to injection of coloring substances. They were fixed in alcohol, followed by formalin at not well-defined concentrations, and held at room temperature. Before dehydration in -25 °C acetone, excess fixative was washed out. Forced impregnation of Biodur S10/S3 was conducted in the vacuum chamber at -25'C, followed by standard procuring and curing with minor modifications. The structural integrity of these specimens seems similar to fresh plastinated organs.

We have not examined the microscopic characteristics of these plastinated specimens. This will be the next step, as we consider possible implications in the field of the paleopathology. We suggest using this approach to supplement Medical School teaching sets, where there is a scarcity of organ donors, or as in our case, the presence of a very restrictive law.

The technique and use of plastinated specimens in teaching and research: Gross anatomical sections of the head and neck. **Margaret Cooper,** St. Louis University School of Medicine, St. Louis, MO, USA.

With the advent of CT and MRI technology, the need for understanding sectional anatomy has become extremely important. This is especially true in regard to sectional anatomy of the head and neck. The plastination technique is advantageous in providing sections for study purposes. Head and neck specimens were scanned by CT or MRI prior to sectioning and marked, using the laser beam as a guide, during the scanning process. The specimen was frozen and 4-8mm slices were prepared on a Hobart band saw, using the markings as guides. Following cleaning and rinsing in water, the sections, which should be kept in order, were dehydrated in a series of graded alcohols and 100% acetone (which is an intermediary solvent and completes the dehydration process) and placed in Biodur S10/S3 mix in a vacuum chamber at a pressure of 10-15 mm Hg for a week. All of the preceding were done at room temperature. Following impregnation, the specimens were allowed to drain for a week, prior to gas cure exposure. Upon removal from the gas cure, the specimens were labelled with a predetermined code and put back into the cure. After a short period of being exposed to ambient air, the specimens were ready for use. Students compare the plastinated sections with the CT or MRI films and therefore, quickly learn the anatomical features of the head and neck sections. The advantage of using plastinated head and neck sections is their repeated use by many students without any deterioration. The keys to good sections are cutting a frozen specimen with a sharp blade, cleaning the section thoroughly, proper plastination technique, and labelling the sections for identification purposes.

Plastination in adversity. **Edward V. Crabill,** University of Pittsburgh, Pittsburgh, PA, USA.

For their laboratory experience, gross anatomy students at our school study museum specimens, models, and prosections prepared by faculty and graduate students. Because the prosections are used extensively each year by approximately 75 students, delicate structures, especially in the Head and Neck preparations, had a relatively short life. Decreasing availability of dissectors (and cadavers) hindered our efforts to replenish the supply of prosections. Therefore, the relative permanence and durability that plastination provides seemed ideally suited to our needs. Unfortunately, establishing a plastination laboratory coincided with the start of extensive renovations of the Dental School building, and space for laboratories, existing or new, was at a premium. After a few unsuccessful attempts to work in shared quarters, a more suitable laboratory, which also serves as a departmental store room, was made available in October, 1987. Although there have been constant problems with the effects of the renovation project upon health, safety and cleanliness, the laboratory is now marginally satisfactory. All specimens plastinated to date are from routinely embalmed cadavers, some of which have been in use for 15-20 years. Irreparably damaged structures were trimmed away, salvageable structures were repaired, and deeper regions and structures were dissected. Only a few relatively new dissections of superficial structures of the head and neck have been prepared. The plastination procedure is routine with few exceptions. After impregnation with S10, dissected structures are thoroughly drained and carefully positioned and anchored prior to the gas cure. To provide for a slow, but steady curing process, an interval timer is used to control the gas-cure pump. To date, 35 head and neck dissections, 8 kidneys, 17 hearts, and 3 joint preparations have been prepared and used in our teaching labs with great success.

Application of plastination in meat inspection pathology. **Boris G. de Gritz,** College of Veterinary Medicine, Helsinki, FINLAND.

Since the early eighties, the total number of animals slaughtered in Finnish abattoirs has decreased from over 3 million units in 1981 (excluding poultry) to less than 3 million units in 1984 and the trend is still decreasing. Because of the decline in the number of animals slaughtered and the improvements in the veterinary service, the number of detected disease conditions has reduced. Various animal diseases, e.g. tuberculosis, have not been recorded in recent years. The above mentioned circumstances necessitate the collection of samples of representative pathological conditions from inspected carcasses and viscera for preservation. Additionally, the obvious documentary value of a museum display of such specimens is of educational importance. A comprehensive selection of plastinated specimens of each disease condition, which is important for meat inspection, will be useful in teaching meat inspection pathology to veterinary students.

Plastination at Baylor College pf Medicine. **Gregory Duncan,** Baylor College of Medicine, Houston, TX, USA. In setting up a plastination lab, the first need is space. Without available space, the equipment and specimen preparation had to be integrated into existing laboratory space. Most equipment was obtained from university and departmental storage rooms. The limited space and

budget allowed only for the establishment of plastination at room temperature. A recent source of chemicals has been the Environmental Safety Office, who are called upon to dispose of methylene chloride, acetone, and ethanol. Obtaining polymer and dehydration chemicals are our continuing costs. Second hand equipment has some repair and refurbishing costs that are indefinite but must be expected. A cost of operation that was overlooked was that of dry ice used in the acetone condensing bath. Results with room temperature S10 have been good and shrinkage has been at an acceptable level using acetone as the dehydrant and volatile medium. Methylene chloride use has been discontinued. Utilization of plastinated specimens has been restricted due to loss of specimens and intermixing of plastinated specimens with wet specimens in the anatomy labs. Therefore, specimens must be monitored in some fashion to prevent this occurrence. Specimens are now on display in the anatomy museum. Future applications include plastination of all wet museum specimens and lab prosections that are easily and frequently damaged, such as pelvic diaphragm, lateral pelvic wall, and posterior pharynx. Plastination specimens may be repaired by gluing.

Plastination jn bone histology. **F. Eitel,** R. Seibold, Chirurgische Klinik Poliklinik Ludwig-Maximilians Universitat, Miinchen, WEST GERMANY.

A decisive factor influencing fracture healing is microcirculation. As a result of methodological difficulties in determining microcirculation in bone tissue, there are numerous methods for representation of this variable (Table 1).

TABLE 1

Methods for representation of the microcirculation in bone tissue:

INVESTIGATION OF VASCULARIZATION (direct functional variable) Measurement of the venous outflow Clearance techniques (^Sr, ^{125}J)
Erythrocyte labelling with radioisotopes
Microsphere technique Dilution methods
Intramedullary pressure measurements
Vital microscopy
INVESTIGATION OF VASCULARITY (indirect functional parameter)
Storage of radioisotopes in bone tissue Capillary-occluding microsphere techniques Microangiography for representation of the vascular pattern (with India ink, soot dispersion, barium sulfate, plastics or dyes)
Histomorphological investigation techniques (fluorescent microscopy of labelled bone)

Each of these methods has its specific advantages and disadvantages. The method chosen must be appropriate to the problem to be investigated, i.e. in the one case flow studies or in another case representation of indirect functional parameters (Table 2) are indicated.

TABLE 2

Vascularity is determined as an isomorphic parameter of the microcirculation. Definition of parameters which can be represented by microangiography Vascularity (Pattern of vascular distribution): Density indicates: the degree of tissue supply (number of vessel filled by contrast medium per unit bone volume).

Direction of branching reflects: the direction of flow (number of vessel branches open to periostea! or endosteal surfaces or in the longitudinal axis of bone per unit bone volume).

Vascular network indicates: the capacity to compensate interruptions of the vascular bed, e.g. after trauma or after surgery (number of anastomoses between anatomically separated vessel areas per unit bone volume).

In respect to the investigation of fracture healing, microangiographic studies are well established. Direct gross and microscopic visualization of the vascular pattern in bone was first described by Splalteholz (1911). This technique involves injection of the specific vascular network with India ink, demineralization of the whole bone, dehydration and clearing of the cortex with a mixture of methylsalicylate and benzylbenzoate. The following disadvantages of this technique are apparent: 1) length of time (6 weeks) for demineralization of long bones, 2) toxic and odorous nature of methylsalicylate, 3) bone must be examined in the fluid methylsalicylate which does not maintain the original form of the bone for an extended period, and 4) because the bone is not embedded in a solid material, standardized bone sections are not obtainable. This study addresses improvements in microangiographic techniques. Plastination techniques originally developed for macroscopic sections (von Hagens, 1981) can be modified to prepare microscopic sections (Eitel et al., 1986). Particularly good penetration of the histological section occurred during preparation when standard techniques for freeze substitution, defatting and forced impregnation with resin were employed. Various polymers and polymer mixtures were used and their results compared, to determine the advantages and disadvantages of each polymer-mix. Biodur E50, Biodur E7, Biodur E700 and flexibilisator AE15 were used for plastination of bone sections. Sectioning techniques and the use of histological stains have been described for both calcified and decalcified bone. The modified Spaltehdz technique was used for decalcification of primarily plastinated specimens. These decalcified specimens under went a second plastination procedure (according to sheet plastination). Utilizing these techniques, plane parallel standardized sections were

produced for morphometric examination of the vascular structure of the bone and soft tissue and the filling quality of various contrast media were assessed. Variables influencing the degree of filling and the visualization of vessels were determined (Table 3) and compared to a reference method (intravital microscopy of pancreatic vascularization).

TABLE 3

Variables which influence the degree of filling and the visualability of the vessels: CONTRAST MEDIUM
- Nature of the contrast medium (particle size, adhesivity)
- Viscosity of the contrast medium

NATURE OF APPLICATION
- Vessel contents (preflushing to remove blood)
- Infusion pressure and duration
- Time of injection (intra vitam/post mortem)

FUNCTIONAL STATE OF THE VESSEL REGION TO BE VISUALIZED
- Width of the vascular lumen (tonus, vasomotion)
- Flow volume/unit of time

PREPARATION TECHNIQUE
- Fixative (degree of shrinkage)
- Means of decalcification
- Section thickness

Barium sulfate accurately filled the arterial system to a vessel diameter of 0.05 mm. Epoxy resin (Biodur E20, MEK, Biodur E2) filled capillaries to a mean diameter of 0.011 mm, but only 12% of the capillaries visualized by intravital microscopy demonstrated a constant filling by epoxy resin. Improved microangiography techniques were demonstrated following plastination and using the modified Spalteholz method. Plastination appears to be useful for the examination of microscopic specimens. Plastinated sections allow different methods of evaluation (light microscopy, fluorescent microscopy, microradiography and microangiographic studies) on the same specimen. The disadvantages of the original Spalteholz method are overcome by the described modifications.

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Applications of plastination Jn embryoloaical and histoloqical research. Helga Fritsch, Medical University of LQbeck, Lubeck, WEST GERMANY.

Research into topographical relationships between human fetuses and adults requires a method that provides: 1) Preserved topographical relations between various tissue components; 2) An insight into the architecture of the connective tissue; and 3) Preparations of fetuses comparable to preparations of adults. Since neither the commonly used paraffin method nor the celloidine technique meet these requirements, we developed a new histological method based on the plastination technique [1]. In order to study the topography of the connective tissue in the human pelvis and to investigate its developmental changes, pelvis of fetuses impregnated with the epoxy resin E12 were serially cut with a diamond-wire saw [2]. The sections (300 - 600//Jm) were mounted on microscopic slides and the free surface was smoothed and polished to remove the traces of the diamond wire [3]. The sections were stained with methylene-blue/azure II and counterstained with basic fuchsin [4]. This staining technique yielded excellent results (Fig. 1, pg. 7) and conserved tissue. Topographical relations were preserved, allowing the study of developmental changes of: The pelvic connective tissue surrounding the rectum [1, 5], the topography of the pelvic autonomic nerves [6], and the ligamentum teres. Three-dimensional computer assisted reconstructions of certain structures were performed on serial sections. This technique is suitable in other fields of basic clinical research such as dermatological studies concerning the nail organ [7].

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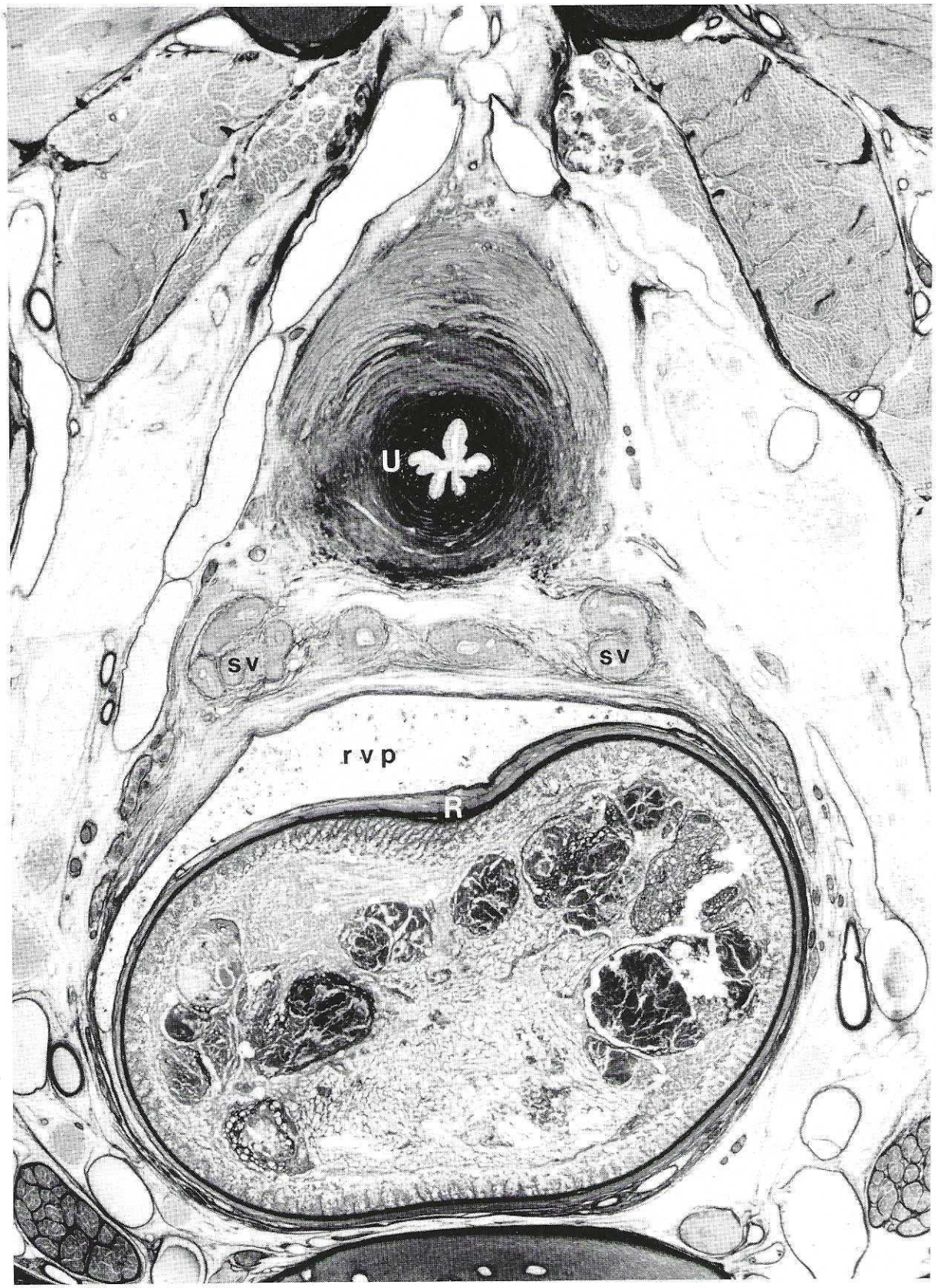
Pre-natal cervical lordotic curvature. M. R. Haffajee, University of Durban-Westville, Durban, SOUTH AFRICA. In the preparation of plastinated specimens of foetuses ranging in age from three to six months, it was noted that a cervical curvature was present after utilizing both the S10 and PEM techniques. Previously, it was accepted knowledge that a pre-natal, cervical-lordotic curvature only occurred late in the intra-uterine life and was accentuated when head elevation begins at about three months, postnatally. This discovery, in plastinated specimens, led us to question whether its existence was a result of plastination or whether it existed in the fresh state. Grey's Anatomy (7th Edition) confirms the presence of two primary curvatures, namely, the thoracic embryonic axis concave anteriorly, as well as, the sacral curvature concave anteriorly. Bagnall (1977) verified the existence of a cervical curve radiographically in foetuses as young as 91/2 weeks of age when ossification centres could be identified. Our



H. Fritsch: Figure 1 - Transverse section (600/μm) through the pelvis of a male fetus (191 mm C-R length) X 14.
U = urethra, sv = seminal vesicle, rvp = rectovesical pouch, R = rectum.

investigation using a series of fresh abortuses, measured the cervical and thoracic curvatures pre and post plastination (S10 technique). The curvatures were measured as radii of matched circumferences of the curvatures. Thus the smaller the radius, the greater was the curvature. The results verified the existence of a pre-natal cervical lordotic curvature, generally a greater curvature than the thoracic curvature. This cervical lordotic curve was more or less constant about a mean, even as the foetus' age increased. Following plastination, the cervical lordosis actually increased but disproportionately with age. As the foetus aged, the thoracic curve tended to increase towards a relatively fixed curvature, but less than the cervical curve. These changes in the thoracic curve may result from its relative immobility as compared to the cervical curve.

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Dehydration of specimens for plastination. Robert W. Henry, College of Veterinary Medicine, The University of Tennessee, Knoxville, TN, USA.

Usually specimens to be plastinated are moist. Therefore, it is necessary to remove the tissue water (dehydrate) prior to forced impregnation or plastination. During dehydration, specimen fluid (water), as well as, some fat is replaced with an organic solvent. The solvent must be miscible with water to be a dehydrating agent and may be of various chemical structure (alcohols or ketones). For plastination either alcohol or cold acetone may be used. Methylene chloride (chlorinated hydrocarbons) is not a dehydrating agent. An inherent problem of dehydration is shrinkage which may be minimized by: 1) starting dehydration in a lower % of ethanol or 2) using cold acetone (freeze substitution). In freeze substitution, the ice in the specimen is replaced by the acetone (dehydrating liquid). When using either ethanol or cold acetone, it is important to use an adequate volume of dehydrating liquid. A suggested ratio is: 1 volume of tissue to 10 volumes of dehydrating fluid. The concentration of the dehydration fluid must be monitored at prescribed intervals (weekly). After the water content has remained similar for two or three days, the specimen is moved to a new dehydrating solution. ETHANOL: Room temperature specimens are started out in a low % of room temperature ethanol (50%), allowed to equilibrate and later placed in ascending concentrations of ethanol, ie: 60%, 70%, 80%, 90%, 100%. Advantages: Carried out at room temperature; therefore, less deep freezer space is necessary. Specimens can be stored in 70% ethanol. Specimens from embalmed tissues, containing standard embalming fluids, are cleansed of the polyvalent alcohols (glycerin or ethylene glycol) or phenols. Specimens are defatted. Disadvantages: shrinkage; an intermediary solvent is necessary; the specific gravity of ethanol and alcohol are similar 0.79. Cold ACETONE (-25 °C): For the past few years, this has proven to be the BEST METHOD of dehydration. Acetone dehydration must be carried out in the cold and not at room temperature. Room temperature acetone will cause excess shrinkage. Advantages: Acetone serves as the intermediary solvent; Minimal shrinkage; Superior specimens; and Shorter dehydration time. Disadvantage: must be done in a deep freezer.

Preparing and plastinating veterinary anatomy and pathology specimens. Robert W. Henry, College of Veterinary Medicine, The University of Tennessee, Knoxville, TN, USA.

Veterinary specimens offer an array of opportunities and challenges to the plastinator. Specimens range from extremely small to immense. Equipment should be versatile to accommodate a variety of needs. For consistently good results, hollow organs should be fixed, dehydrated and cured in the dilated state. Gravity or air may be incorporated in the dilation process. Hollow organs may be cut to allow cleaning of fecal material from the organ and then closed with suture material prior to dilation. In particular, the anatomical position of the organ or specimen should be considered and appropriately positioned during fixation, otherwise, the final product will be distorted. Whole body slices are fantastic aids for study of sectional anatomy, CT scans or MR images. Fibrous tissue or skin covered specimens must be perforated and polymer injected to minimize shrinkage. Polymer is easily removed from hairy specimens prior to curing. Reptiles, opened on the ventral midline may be plastinated. Dried lungs, which are anatomically preserved, may be forced-air-impregnated with a S10/S3-xylene mix. Fixation and dehydration vats should be similar in size to your plastination chamber. Therefore, if the specimen fits into the dehydration vat, it will fit into the plastination chamber. Thickened polymer may be thinned with xylene (3-50%) and used for impregnation of lungs or gastrointestinal organs. Impregnate slowly. It takes time to produce beautiful specimens.

Room-temperature "forced air" impregnation pf dried lungs with SlO/S3-xvlene mix. Robert W. Henry, College of Veterinary Medicine, The University of Tennessee, Knoxville, TN, USA.

Air-dried lungs were successfully impregnated with SlO/S3-xylene mix at room temperature by using a laboratory air source to propel the polymer from the air ways into the parenchyma of the lung tissue and to the lung surface. After thorough drying, six mixtures of S10/S3 and xylene, varying from 15% to 40%, were used to impregnate lungs, using forced air rather than

vacuum. The trachea was cannulated and the lungs positioned so that one-half of the polymer-xylene mix was poured into the right tracheobronchial tree and one half into the left portion of the tracheobronchial tree. High velocity laboratory air was utilized, for four to seven days, to force the polymer mix from the air ways into the lung parenchyma. During the first 15 minutes of impregnation, the lungs were turned in all directions to assure that polymer reached all areas of the lungs. Thereafter, the lungs were inverted daily. The trachea and surface of the lungs were coated daily with polymer mix. After one week of air impregnation, the lungs were cured by bubbling air through 86 and into the trachea for 15 minutes a day for 3 days with a plastic bag surrounding the lung preparation to serve as curing chamber. Air impregnated lungs maintain their anatomical relationships. High percent xylene lungs are more spongy than lower percent xylene lungs. Conversely lower percent xylene lungs are firmer.

Plastination pf granular hvdroxvlapatite and attached tissue. GUnther Hotz, H. Gilde, R. Mannl, T. Honer, Department of Maxillo-Facial Surgery, Universitat Heidelberg, Heidelberg, WEST GERMANY.

A plastination and sawing-grinding technique for undecalcified histological sections of bone, which contain implants of granular hydroxylapatite with soft and hard tissue is presented. The clear and thin sections contained minimal artifacts, especially along the tissue-implant interface. Both the hard and soft tissues adjacent to the granular implant was preserved. The sections were beneficial for observation of cell morphology and for obtaining morphological data.

Fixation solutions for color preservation in plastination. Half Kessler, Institut Fur Pathologic, München, GERMANY.

Color plays an important role when presenting many pathological findings. Most solutions used for color retention in fixation, which use formalin-salt mixtures and an alcohol bath for color development, can be used for silicone plastination. Two of these methods are presented: Method after KAISERLING: Fixation in a solution of: 15 g potassium nitrate, 30 g potassium acetate, 200 ml concentrated Formalin, in 1000 ml distilled water. Fixation is completed when the specimen is grey-brown. After fixation, color development is in 80 - 95% alcohol for less than 12 hours and then start dehydration; or store the specimen in a solution of: 200 g potassium acetate, 400 ml glycerine, in 1000 ml distilled water. When ready for dehydration, rinse in water for 24 hours. Method after JORES: Fixation in a solution of: 50 g synthetic salt of Carlsbad, 50 g chloral hydrate, 50 ml concentrated Formalin, in 1000 ml distilled water for 7 to 14 days. Rinse in water for 24 hours and start dehydration; or store the specimen in a solution of: 300 g potassium acetate, 600 ml glycerine, in 1000 ml distilled water. When ready for dehydration, rinse in water for 24 hours. If possible, dehydrate and defat via freeze-substitution with acetone.

Foetal anatomy using the PEM and S10 techniques. G. Mathura, University of Durban-Westville, SOUTH AFRICA.

Freshly obtained foetuses were plastinated using either the standard PEM or S10 technique. After plastination, a suitable container was selected which would allow 15-25 mm of resin to surround the foetus, as resin is used as an embedding media to facilitate maneuverability and cutting of thin slices. Catalyzed resin (15 mm thick) was poured into the container and allowed to harden to form a base upon which the specimen is to lie. The foetus was centered on the hardened resin and more resin was poured to cover one quarter of the foetus. When this had hardened, a similar amount of resin was poured covering half the foetus. Finally, the entire foetus was covered by 15 mm of resin. It is important to pour the resin in stages, since the heat generated during the setting period of a thick layer may damage the foetus. When the resin had set and cooled, it was removed from the container and either sagittal, horizontal or coronal sections were made. A band saw with a sharp, fine-toothed blade was used to cut off the excess resin and the foetus was carefully cut in 3-5 mm slices. Resin dust settling on the foetus was gently vacuumed off, the slice was shaped and sanded as desired, and painted with a warm mixture of resin.

FORCED IMPREGNATION. P.P.C. Nel, Department of Anatomy and Cell Morphology, University of the Orange Free State, Bloemfontein, South Africa.

Forced impregnation is the central most step in plastination. The art of plastination is the ability to match the quantity of the solvent with the type of tissue and specimen size and at the same time other variations, such as the vapor pressure, and still avoid shrinkage. Preparation of material for forced impregnation can be divided into three main steps: Fixation and dehydration, Preparation of the polymer mixture, and Introduction of the specimen into the polymer mixture. For the proper polymer mix, combine the proper quantity of hardener and polymer (irrespective of the technique used) and stir thoroughly. Place the mixture under vacuum to draw out the air that was stirred into the polymer by mixing. The specimen must be submerged into the polymer mix directly and quickly from the volatile intermedium. If the specimen surface dries, the finished product may show white spots which are small points of shrinkage. Specimens soaked in acetone tend to float while those soaked in methylene chloride sink. The specimen should not be forced against the bottom of the vacuum chamber or float out of the polymer as the polymer mixture must surround the specimen for uniform

impregnation. Forced impregnation may be divided it into three steps: Specimen placed into the polymer mix, Intermedium "boiled out" of the specimen, and Polymer drawn into the specimen. The volatile intermedium has a high vapor pressure and a low boiling point (acetone: +56"C and methylene-chloride: +40^{8}C). The polymer mix, in contrast, has a low vapor pressure and a high boiling point. The intermedium is extracted out of the specimen which causes a pressure difference between the specimen and the impregnation bath. This pressure difference allows the polymer to be drawn into the tissue. The extraction of the volatile intermedium (boiling) must be slow enough to allow sufficient time for the polymer to enter the specimen. Boiling is monitored by observing bubble formation on the surface of the polymer mix. If boiling is too fast, the polymer does not have enough time to flow to all parts of the specimen and the structural framework of these areas may collapse and the specimen will shrink. Boiling is controlled by a air inlet valve and pump speed. Impregnation time is dependent both on polymer viscosity and specimen thickness.

Developing § plastination laboratory for § new medical curriculum. Marita L. Nelson, John A. Burns School of Medicine, University of Hawaii, Honolulu, HAWAII, USA.

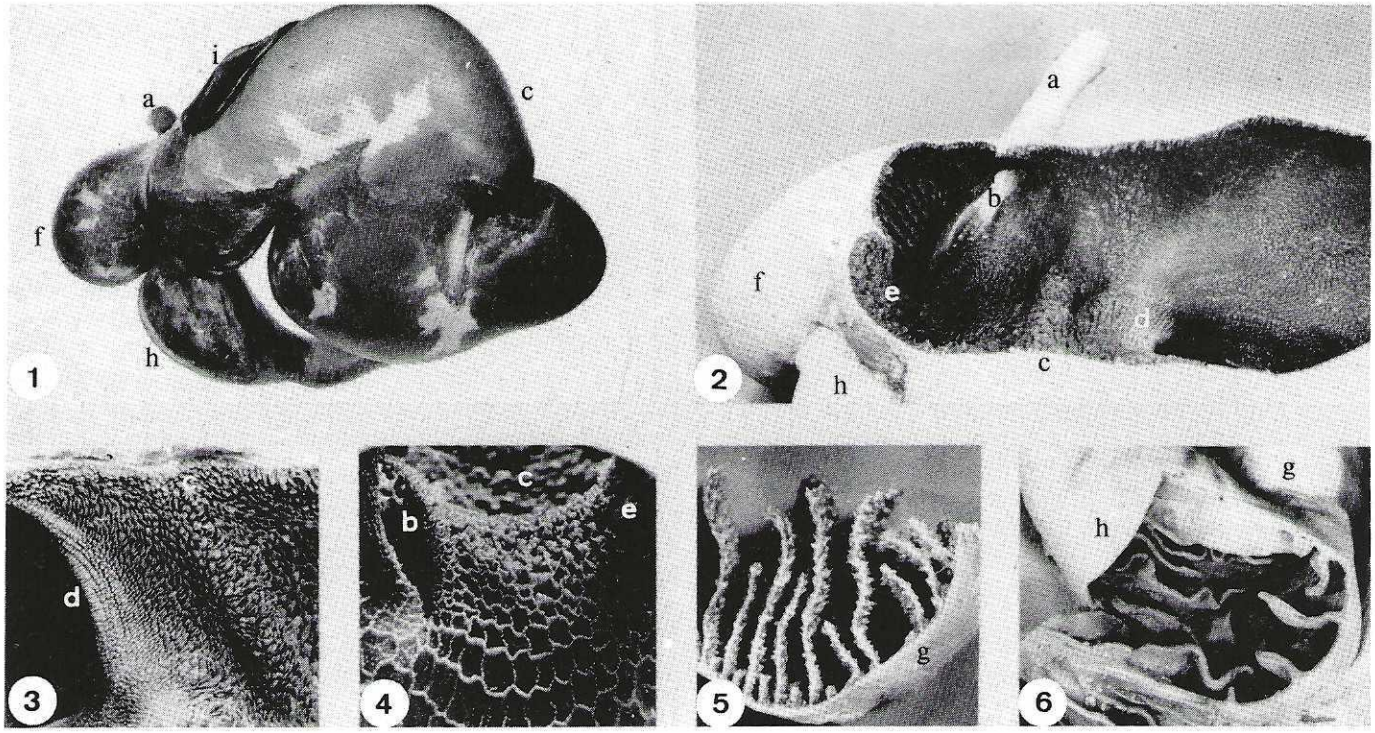
When recent curricular revision, to a problem-based learning format, eliminated all formal lecture and laboratory courses, we had to find other ways of teaching human gross anatomy to medical students. Our three choices were: cadaver demonstrations by advanced medical students on request, short elective regional dissection courses, and independent study of prosected cadaver specimens. Plastination was the method chosen to preserve prosections for long-term use. A plastination laboratory was developed and specimens were processed at room temperature to increase the speed of preparation and to minimize cost. Our specimens were processed completely in one month and shrank a maximum of 17%. Twenty-four specimens have been prepared including a series of sagittal sections through the head and neck. The latter are valuable in helping students interpret CT scans and magnetic resonance images. A collection of plastinated specimens are being prepared for use in a multipurpose learning resources facility, which we anticipate will enhance student learning in anatomy.

On the production of plastinated specimens for teaching veterinary morphology. Monique Nicaise, P. Simoens, H. Lauwers, Faculty of Veterinary Medicine, State University Ghent, GHENT, BELGIUM.

In order to obtain demonstration specimens for teaching veterinary gross anatomy, plastination by means of the standard 310-technique was started in our Institute two years ago. Some aspects of the technical procedure were experimentally tested and modified: Perfect fixation can be obtained by intravascular flushing with saline and subsequent perfusion with a fixative. However, this perfusion technique should not be performed for plastination of hearts and muscles, because haemoglobin provides good natural pigmentation which supplements that of myoglobin. JORES-solution, in which Carlsbathsalt is replaced by anti-oxidants, is preferred to the original Jores-fixation-solution and to the classic formalin-solution. Silicone polymer penetrates dense-connective-tissue capsules of organs very poorly. Therefore, these capsules should be removed or perforated to enhance impregnation and prevent shrinkage of the organs. In animals smaller than medium-sized dogs, impregnation of the thick wall of the left ventricle of the heart may be incomplete, with impregnation from the outer surface only. This is caused by air-bubbles which are trapped inside the ventricular lumen. Bubbles expand when vacuum increases and penetrate the cardiac wall, thus preventing impregnation by silicone. Plastinated specimens proved to be very useful for teaching veterinary anatomy (comparative, descriptive, developmental and clinical). This is illustrated by the plastinated stomach-complex of a goat (see Figure), which is superior to formalin-fixed-dried models or to commercially available models. The plastinated specimen is most similar to the fresh organ and will not collapse upon incision. Other examples are plastinated hearts, brains, joints and nematodes which were useful for demonstrations in anatomopathology, neuroanatomy, arthroscopy and parasitology, respectively. Formula of the modified JORES-solution, obtained by our experiments: Distilled water 10 L, Chloral hydrate 500 g, Formalin 800 ml, Glycerol 1 L, Ascorbic acid 40 g, and Sodium sulfate 20 g.

An insight into amateur plastination or plastination at home. Stephen M. Probst, Universitat Heidelberg, Heidelberg, WEST GERMANY.

It seems quite difficult to be engaged in plastination at home, since plastination is based upon the physical properties of polymers under defined physical conditions. Since vacuum pumps, manometers, and desiccator jars do not belong to the normal houseware of an average German family, I needed some persuasion to convince my parents that my luck depended on the purchase of some technical equipment for use in my own little plastination laboratory. The most important of these have been made available to me and with some improvising, I was able to produce my first specimens. This shows that even with primitive means successful plastination can be carried out, which might encourage the beginners in the well equipped professional labs. Formaldehyde fixation was carried out in tight sealed plastic or stainless steel containers. Somewhat more complicated was the dehydration process. I prefer freeze substitution, as acetone is used in smaller amounts and the cost is not much higher than fo



M. Nicaise: Plastinated stomachs of goats: 1 - Left lateral view. 2 - Dorsal view after removal of the dorsal ruminal wall. 3 - Ruminal papillae. 4 - Reticular ridges and cells. 5 - Omasal laminae. 6 - Abomasal folds. Esophagus (a), Reticular groove (b), Rumen (c), Cranial pillar of rumen (d), Ruminoreticular fold (e), Reticulum (f), Omasum (g), Abomasum (h), Spleen (i).

premium gas. Alcohol dehydration consumes more time and materials and is therefore more expensive. A further disadvantage is the greater danger of shrinkage. The only unpleasant effect of acetone is that it is flammable, but with some safety precautions acetone can be handled safely. Acetone was used in acetone proof containers inside a household deep freezer at -25°C. For safety reasons, the inside light was removed. My acetone supply was stored in our garden house, so that never more than 4 liters of acetone were kept in the freezer. Because my mother was strictly against it, acetone was not recovered via distillation, but rather discarded by burning small portions in a coffee tin. For impregnation, a pump with a displacement of 1.5 m^3/h was sufficient for a small vacuum tank. Desiccator jars were used as vacuum chambers. Taping the glass bell jar will prevent glass splinters in case of implosion. Plastic pails containing the impregnation bath were placed within the desiccator jar. For larger specimens, two jars were set one upon the other, thus doubling the volume of the container. It is desirable to impregnate in a freezer, but because small specimens are impregnated in a short time, cooling is not crucial. Nevertheless, in winter time, I used snow cooling outdoors. A pail containing a salt-ice mixture or bottles of frozen glycerine may be used for cooling. The cost of the appliances used for the Forced Impregnation was $100 for the manometer, $200 for a desiccator jar, $300 for a rotary vacuum pump, and $30 for tubings and valves. The total cost was $630. Many other items were found in the kitchen, as well as in the garden house which made them quite inexpensive to use. The last step of the S10/S3 technique is gas curing which was carried out in sealed containers in which the specimen lay on a wire mesh while the S6 gas cure covered the container base.

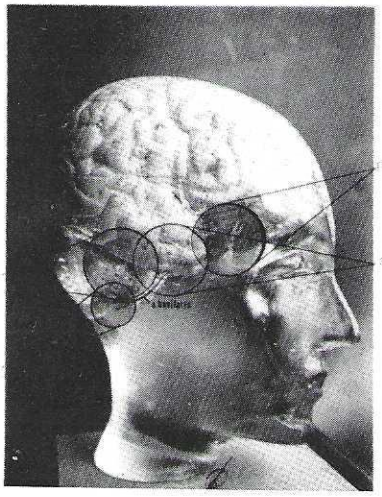
The use of plastinated specimens in teaching veterinary gross anatomy: substitution or supplementation, **P. T. Purinton**, College of Veterinary Medicine, University of Georgia, Athens, GA, USA.

The dissection laboratory is a good problem-solving environment. With a textbook, dissection guide, a few dissection instruments and a cadaver, the student must decipher the spatial relationships of the structures of the animal in question, attach the appropriate names to the structures, and extrapolate functions. Prosected, plastinated specimens are of great value in orienting the student to difficult dissections, showing views and approaches different from the dissection, and for review during that course or at a later time. Thus plastinated specimens are an important supplement to dissection in the study of gross anatomy. The University of Georgia, College of Veterinary Medicine is currently revising the curriculum to decrease lecture hours, decrease rote memorization, increase problem solving, and increase student responsibility for education. With this revision,

the current two courses in gross anatomy (Anatomy of the Dog and Cat, Anatomy of Ungulates and Birds) will be replaced by a course in Principles of Anatomy and a species-specific anatomy course of the student's choice. The principle course will be non-species specific and will have minimal dissection by the student. The laboratory portion will be taught principally with prosected plastinated specimens. The species specific course will be chosen from a menu of courses including anatomy of the dog and cat, anatomy of the horse, anatomy of ruminants and pig, and anatomy of exotic animals and birds. Species specific anatomy courses will be in depth dissection courses with strong clinical correlation. Plastinated specimens will be used to supplement the dissection. The proposed curriculum will reduce the number of embalmed specimens used for dissection thus allowing the shifting of funds to plastination. A significant increase in the number and use of plastinated specimens is anticipated.

The use of plastinated head specimens in planning microsurgical approaches to the skull and brain base, **K. D. M. Resch**, A. Pernecsky, Klinikum der Universität Mainz, Mainz, WEST GERMANY.

The introduction of the microtechnique to neurosurgery called for a new paradigma of the clinical anatomy of the head: the key-hole anatomy through the microscope had to be established. To transfer the idea of sophisticated, anatomical key-holes of the brain base to anatomists and surgeons, it was necessary to introduce three-dimensional plastinated head specimens with which to demonstrate the clinical anatomy. For approach analysis, approach design, and planning of an operation, plastination was useful in neuropathological anatomy and clinical cases. Four head specimens with numerous approaches and four clinical cases (1 carotid aneurysm and 3 giant basilaris aneurysms) were presented to demonstrate the principles of this method. In the future, plastinated head specimens will be helpful in working out the standards of Endoscopic Neuroanatomy.



Thin sheet specimens behind photoglass, **Alfred Riepertinger**, Stadtkrankenhaus München-Schwabing, München, WEST GERMANY.

Thin sheet specimens behind photoglass is a new way of presentation of plastinated material. The characteristic feature of this method is that in a small area a relatively high quantity of pathological findings, be it in a particular organ or in different preparations, can be displayed. A further advantage is that the BIODUR S10 technique is used. The time for the special manufacture of thin sheet specimens is between forced impregnation and gas curing. In select cases (depending on the organ) it is possible to prepare the thin sheet after gas curing. FIXATION - The fresh specimens taken during autopsy were cut in slices of 1.0 - 3.0 cm and put into a color-preserving solution after Kaiserling. One surface of the specimen was cut either during or after fixation. In order to obtain a clean cut, a special rotation-cutting machine was used. After seven days of immersion-fixation and color development, the preparation was removed from the solution and dehydrated. DEHYDRATION was achieved using cold acetone which was changed five times until a residual water content of less than 1% was reached and no yellow coloration was noted. FORCED IMPREGNATION was in BIODUR S10 for a period of 14 days at -25°C. After pressure equalization, the specimens were immediately taken out of the vacuum chamber. PREPARATION OF THIN SECTIONS: After the impregnated specimens had drained for two days at room temperature on a slanting support padded with an air-cushioned, plastic foil and were only slightly sticky, 1-2 mm slices were cut from the 1.0 - 3.0 cm organ slices with a rotation-cutting machine. These thin slices were placed on a BIODUR S2-pasted glass plate and covered with another glass plate. Approximately 6 kg of weight was used to press the thin section between the glass plate for about 10 minutes. The BIODUR S2 rapidly hardens the back of the thin specimens, otherwise the preparations would be distorted. GAS CURING of the slices was for 48 hours and S6 volatilization was hastened using an aquarium pump. Before gas curing, the surface was swabbed with S10 if necessary. INSTALLATION AND FINISHING - A suitable glass pane and background was selected for the size and number of specimens. It is advisable to seal panes along their edges to prevent dust from collecting. Arrange the slices onto the base plate and glue them in place with a thin layer of silicone paste. Clear labelling completes this new way of presenting plastinated specimens.

Teaching in macroscopic pathology. Value of plastination using the silicone S10 technique, **Joseph Rüschoff**, K. H. Plate, C. Thomas, Philipps University of Marburg, Marburg, WEST GERMANY.

Education in macroscopic pathology deals with two obviously contradictory facts. First, in West Germany

the autopsy rate has dropped dramatically during the last 15 years and thus only select cases can be used for teaching macroscopic pathology. On the other hand, imaging techniques have been markedly improved and are of central importance for the clinical diagnosis. The prerequisite of correct image interpretations, however, is a profound knowledge about the typical macroscopic changes in the diseased organ. Therefore, we started plastination in 1985 using the silicone S10 impregnation technique. Three hundred organs and organ sections were plastinated. In general pathology, seminars were performed by video presentation. Systematic pathology was taught with special regards to differential diagnosis. Methodologically, the use of a modified Kaiserling's technique (adding sugar to the original formula and restricting alcohol incubation to an eight hour maximum) yielded good color preservation which was superior to formalin fixation alone. Histologically, structural detail was well preserved as was shown by a two-step silicone dissolving technique.

1 Week Fixation: *modified Kaiserling's*
200ml formalin (40%) 1000ml Aqua dist.
T5g K nitrate
30g K acet. acid
100g Sugar

Color Restitution: *4-8 hours*
95% alcohol

Plastinated specimens in medical anatomical teaching. **T. M. Scott,** Memorial University of Newfoundland, St. Johns, Newfoundland, CANADA.

We have used plastinated specimens in teaching anatomy to medical undergraduates for four years. We have a double pump two-kettle system which has allowed us to produce two hundred human specimens ranging in size from small single organs to head, neck and upper thorax. We have restricted production to the S10 technique and are confident that we can plastinate any reasonable specimen within the size limitations of our system. Our plastinated specimens are used in gross anatomy, systems anatomy, neuroanatomy and embryology courses. The gross anatomy course runs for seven weeks using prosected specimens in a small group teaching setting. The systems anatomy is taught within multidisciplinary courses on the body systems. Neuroanatomy is taught within a course on neuroscience, and embryology is taught as a lecture course with a small laboratory component. We have limited the production of plastinated specimens to specific areas. For gross anatomy, we have produced a range of joint specimens (knee, hip, elbow, shoulder), a variety of organs (heart, kidney, liver, stomach), and "short" limbs including hands and feet. Our neuroanatomy collection includes several spinal columns dissected to show the spinal cord and spinal nerves. In teaching neuroanatomy, our goal is to supply each student with a plastinated set of specimens composed of a whole brain, a half brain, and a brain stem with cerebellum. Although the production of plastinated specimens is primarily directed towards undergraduate medical teaching, our materials are frequently used in teaching pharmacy and nursing students, as well as public displays.

Time and money savers in SlO-techniaue. Acetone distillation. Michael D. Smith, Department of Anatomy and Neurobiology, Colorado State University, Fort Collins, CO, USA.

Several time or money saving items applicable to plastinators were discussed: constructing an S10 impregnation tank using a common household pressure cooker; use of Plexiglass, aquarium pumps, and aquariums to make S6 gas cure chambers; injection and plastination of canine brains using red latex; use of disposable cardiac catheters for heart injection; rapid separation of canine heart and lungs for dilation of the heart; sources of fetuses and other animal specimens for plastination; explanation of the features and use of commercial freeze driers as plastination machines; distillation of waste acetone produced via freeze substitution using a commercial programmable distiller; elimination of paraformaldehyde precipitate; cooler venting as a means of eliminating formaldehyde fumes associated with anatomy laboratories.

Staining pf thin brain slices combined with subsequent plastination. Norbert Ulfig, Zentrum der Morphologic, J. W. Goethe-Universitat, Frankfurt, WEST GERMANY.

The configuration of subcortical structures in the human brain is often difficult to recognize in unstained sections. Subdivisions of nuclear greys are difficult to delineate, in particular, for the inexperienced student. Moreover, wet brain tissue is easily destroyed. Therefore, it appears desirable to combine the advantages of plastination with those of staining. Staining of 1-4 mm thick frozen sections (or thicker sections made with the aid of a macrotome) with astra-blue or aldehyde-fuchsin provides a sharp contrast between white and grey matter. Aldehyde fuchsin demonstrates the architectonics in the adult human brain in detail. It stains the intra neuronally stored lipofuscin granules and varying amounts of lipofuscin pigment are recognizable (pigment architectonics) in the subcortical structure. For standard S10/S3 plastination, the stained sections are placed between perforated plates to obtain sufficiently flat sections. The standard S10/S3 plastination procedure is used for impregnation. The items necessary for this technique are chemicals, buckets and vacuum chamber. Therefore, this technique can be utilized in any laboratory without much

expenditure or experience. These plastinated stained brain sections are durable, convenient to handle, and can be used in teaching neuroanatomy and pathology.

GAS CURING. **Andreas Weiglein,** Institut fur Anatomie, Universitat Graz, Graz, AUSTRIA.

After impregnation, polymer curing has to take place to keep the polymer inside the specimen. For the S10 process, **Biodur Gas Cure (S6)** is used. There are two curing procedures: In one case, exposure to gas cure (S6) commences after a long precuring interval; this is called **slow curing.** In the other case, immediate exposure to gas cure is followed by a short final curing interval; this is called **fast curing.** Both slow and fast curing utilize Biodur S6. Gas curing is carried out at room temperature in a closed chamber containing a bowl with Biodur Gas Cure (S6) and a bowl with a desiccant (calcium chloride) (to absorb water condensation which may cause white silicate salt precipitations on the specimens surface). S6 evaporates at room temperature and builds up a gaseous atmosphere inside the chamber. To accelerate vaporization and circulation of the gas, a small membrane pump (used for aquariums) may be used. During **slow curing,** allowing the SlO/S3-reaction mixture to thicken in the specimen before exposure to S6 is called precuring. To avoid shrinkage, precuring begins at room temperature. If time permits, it is best to use room temperature only. To shorten precuring, 40 or 50 °C temperature may be used as follows: 12 weeks at room temperature, 6 weeks at room temperature and 2 weeks at 40 *C, 4 weeks at room temperature and 1 week at 50 °C. Precuring is finished when the specimen is notably tacky. Then the specimen is put into the gas chamber for 2 to 4 days until it is dry to the touch. For **fast curing,** the specimen is initially placed in the gas chamber with a high concentration of S6 until no polymer oozes out of the specimen. For final curing, the preparation is put into a tightly closed receptacle (e.g., a plastic bag) for 2 to 3 weeks to allow the center of the specimen to cure.

ROOM-TEMPERATURE "FORCED AIR" IMPREGNATION OF DRIED LUNGS WITH S10/S3-XYLENE MIX.

Robert W. Henry, Jessica Butler

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The University of Tennessee, College of Veterinary Medicine
P.O. Box 1071, Knoxville, TN 37901-1071

INTRODUCTION

Preparation of beautiful and anatomically correct whole lung preparations is difficult using the standard S10 forced vacuum impregnation technique. Lungs which have been saturated with solvent and later with polymer are difficult to cure, such that their proper anatomical shape is retained. If lungs in the fresh state are dried in their proper anatomical configuration, they can be plastinated using a modified forced air impregnation method. Instead of submerging the lungs in polymer and using vacuum for impregnation, a polymer-xylene mixture is poured into the tracheo-bronchial tree and air is used to impregnate the lung tissue thus leaving the alveoli empty.

MATERIALS AND METHODS

Fresh lungs were collected from necropsy and from a slaughter house. The heart, mediastinal tissue and fat were carefully separated from the lungs and tracheobronchial tree. A cannula or tubing of the appropriate diameter was ligated in the trachea. A water source was attached to the cannula and the lungs gently inflated to near capacity. After filling, the water source was removed and the water and blood allowed to flow

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MATERIALS AND METHODS

Fresh lungs were collected from necropsy and from a slaughter house. The heart, mediastinal tissue and fat were carefully separated from the lungs and tracheobronchial tree. A cannula or tubing of the appropriate diameter was ligated in the trachea. A water source was attached to the cannula and the lungs gently inflated to near capacity. After filling, the water source was removed and the water and blood allowed to flow

out of the trachea. This flushing procedure was repeated 8 to 10 times, which cleared much of the blood from the lungs. The lungs were covered with moist toweling to prevent drying. Finally, the lungs were connected to the water source for over night flushing. Flow was adjusted so that the lungs remained inflated as blood/water mixture flowed out of the pulmonary vessels and parenchyma. The next morning, provided the blood had been flushed sufficiently, the water source was removed and the water allowed to drain from the lungs via the trachea. After the majority of the water was drained, the lungs were placed on their dorsal (posterior) surface and the cannula was attached to the laboratory air compressor. The air flow was increased until the lungs were inflated and remained inflated to near capacity. This flow was maintained until the lungs were thoroughly dried. To assure that the lungs remained inflated, air-flow was monitored at one-half hour intervals the first day.

Larger specimens (pig, cow and horse) were fixed by injecting 1-5 ml of 100% formaldehyde via a syringe and needle through the cannula wall, thus allowing the air to carry the formaldehyde throughout the lung. Smaller specimens (feline and canine) often were not fixed. To prevent the lungs from sticking to the counter as they dried, as soon as specimens could be picked up and retain their shape, they were suspended by the trachea. Air was allowed to flow through the lungs continually until the specimen was completely dry. Drying time was dependent on specimen size (cats:1-2 days, dogs: 2-3 days, pigs and sheep: 3-6 days, cows and horses: 5-15 days). The lungs felt very light when completely dry.

After drying was complete, a polymer-xylene mixture, 5-10 times the weight of the dried lungs, was poured into the trachea. Six variations of polymer-xylene mixture were utilized in canine lungs of similar size. The percentage of xylene ranged from 15 to 40% at 5% increments. Mixtures of 30 and 40% were used in two each of feline, porcine, bovine, and ovine lungs. The lung was positioned so that as one-half of the polymer was poured into the trachea it flowed into the bronchi of the right side. The lung was then turned so that the other one-half of the polymer, when poured, flowed into the bronchi of the left side.

Immediately, after the polymer mixture was poured into the trachea, the air source was secured to the cannula in the trachea and air flow was started. Air flow velocity was not measured. However, nearly full flow from the laboratory air supply was used. During the first fifteen minutes of air impregnation, the lung was turned in many directions to allow maximal availability of polymer to all areas of the lungs. Within a few minutes, small beads of polymer mix were observed on the surface of the lung. Maximal air flow was continued for four to ten days and the lungs were inverted daily to assure maximal air impregnation, emptying of alveoli of polymer, and removal of xylene. Twice a day, the surface of the lungs and trachea was coated with a

fig 23

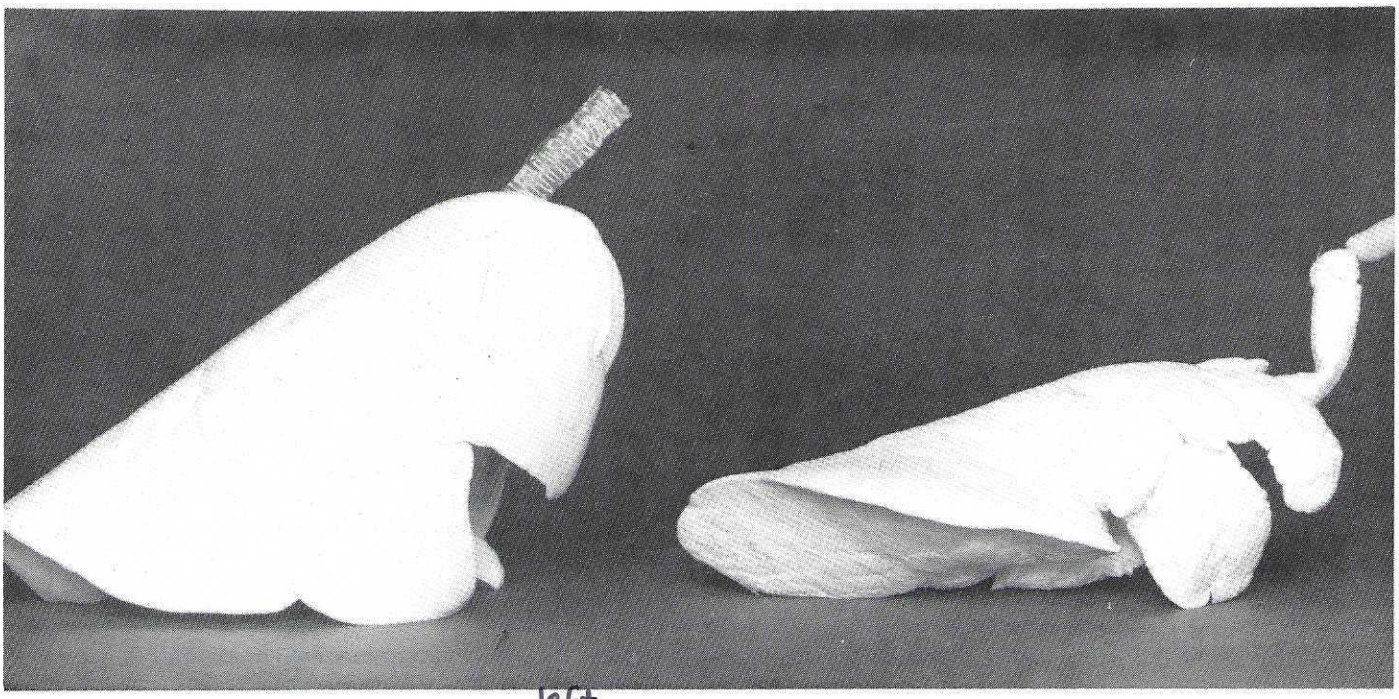

Figure 1: Right lateral view of canine lungs (right), "forced air" impregnated using 30% xylene/70% polymer-mix and porcine lungs (left), vacuum impregnated using 30% xylene/70% polymer-mix. Note anatomical relationships are more precise in the "forced air" impregnated lungs. However, the trachea of the vacuum impregnated lung is flexible and more durable.

S10/S3 mixture, to assure a durable outer layer and to allow wicking of polymer into the lung parenchyma as the xylene evaporated from inside the lung. After 4 to 10 days of impregnation, the air source was removed and the lungs were coated with polymer and inverted for 24 hour periods, every day for one week.

At the end of this period, curing was commenced by bubbling air from the laboratory air supply through S6 and into the trachea for 15 minutes a day for three days. A plastic bag was placed around the lung preparation to serve as a gas curing chamber. The lungs were coated with polymer as needed and left bagged until cured.

RESULTS:

All six percentages of S10/S3 polymer-xylene mix produced satisfactory specimens. Lungs impregnated with lower percentages of xylene were firmer and probably made better student specimens as they appeared to be more durable. Higher percentage xylene impregnated lungs were more flexible and spongy in texture. Hence, they may not be as durable, especially for student use. Canine and feline lungs were more flexible after impregnation than were porcine, ovine and bovine lungs. In all specimens, anatomical relationships were maintained (Fig. 1). Some specimens experienced areas of blow-outs during air drying and hence had areas which were not totally inflated. These lungs were not so beautiful, but still useful for teaching. Since air impregnation does not infiltrate dense tissues, the surface of the trachea must be coated as no polymer penetrates the dried cartilage. Hence, the trachea is hard and not flexible, whereas, vacuum impregnated lungs yield a flexible trachea (Fig. 2). All fat must be removed prior to air drying or the trachea will be greasy. Air impregnated lungs may not be as durable as vacuum impregnated lungs. However, the preservation of anatomical relationships is superior to vacuum impregnated lungs.

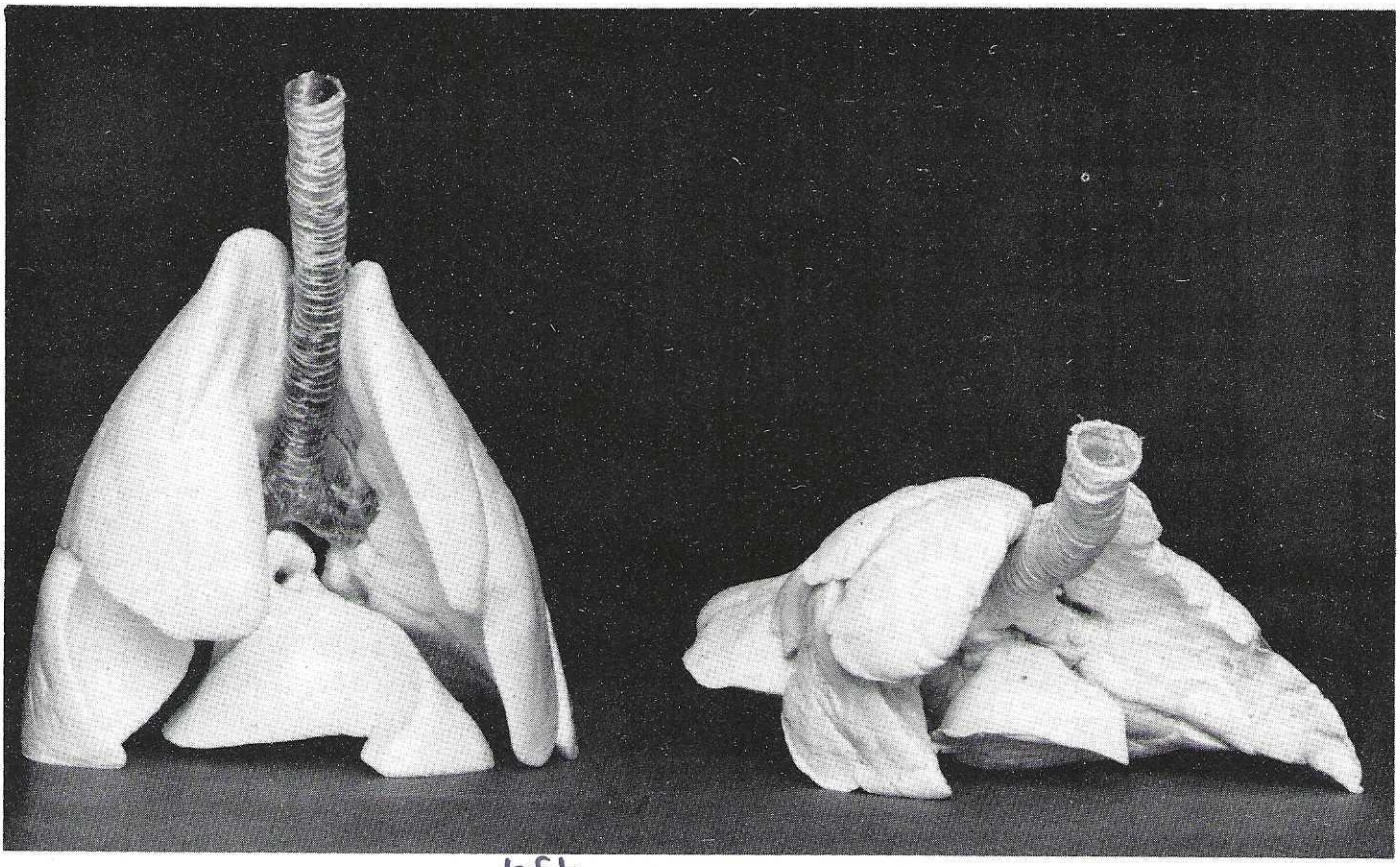

Figure 2: Cranial view of canine lungs (~~right~~ left), "forced air" impregnated using 30% xylene/70% polymer-mix and porcine lungs (~~left~~), vacuum impregnated using 30% xylene/70% polymer-mix. Anatomical relationships are not retained, as well ~~as~~, in the vacuum impregnated lungs.

SECTIONAL ANATOMY: STANDARDIZED METHODOLOGY

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Visiting Associate Professor, University of Illinois at Chicago.

This paper proposes a standardized method for the teaching of sectional anatomy to correlate with computer imaging modality scans. The standardized method of sectional anatomy instruction has been previously discussed and illustrated (Lane, 1988, 1989, 1990 a,b, 1991) and sometimes referred to as the Lane method (Stuart, 1990). The first part of the methodology is to identify, regardless of plane, the area of the body from which the section is derived. For example, in axial sections, the distance from Reid's baseline (Infraorbital meatal line) (in the head), and vertebral levels (neck and trunk), can indicate the origin of the section. In sagittal sections, the distance of the section from the median plane is a convenient system. In this case, a need exists to indicate whether the section is from the right side or left side of the body. For frontal/coronal sections, the distance anterior or posterior to the midaxillary line is a suitable system for identifying the origin of a section. Orientation lines, also known as a grid, are needed in both cadaver and *in vivo* sections to further substantiate the derivation or origin of a section (Fig 1.)

It is important to keep in mind the surface orientation for each plane. For example, in a transverse/axial section, the inferior surface is observed to correlate with the standards of imaging modalities such as magnetic resonance imaging (MRI), computed tomography (CT) scan, and ultrasound. In frontal/coronal sections, the imaging modalities display the anterior surface of the sections. Thus, this custom is used in sectional anatomy. In the computerized imaging modalities, the sagittal section is viewed from the right side. Therefore, this custom is used in sectional anatomy. In some instances the sagittal plane sections are viewed from the leftside.

Regardless of plane, the structures of each section would be classified into four anatomical units or categories. Figures 2, 3, 4, and 5 show examples of each anatomical unit using MRI axial (transverse) clinical slices of man. Note that a thrombus in the vena cava and a hypemephroma of the right kidney are depicted. Figures 6, 7,8 and 9 show examples of each anatomical unit using sheet plastinated human sections. The four anatomical units and an explanation of structures included in each unit follows:

Musculoskeletal unit - a group of bony features and muscular structures located in a section.

Neurovascular unit - a group of nerve structures and vessels as well as dural sinuses located in a section.

Visceral unit - a group of internal organs located in a section.

Enclosing unit - the membranes, spaces, fossae, and other special features seen within a section.

Another aspect of this standard method of instruction for sectional anatomy includes the relationship of adjacent structures which may differ depending upon the plane in which a structure is seen. In transverse sections, anterior/posterior relations, as well as, lateral/medial relations are considered for each structure noted. For example, the internal jugular vein, in an axial section of the neck, is compared in location to a structure anterior to it, posterior to it, lateral to it, and medial to it.

In frontal/coronal sections, superior/inferior relationships, as well as, medial/lateral relationships are compared for each structure under consideration. In a coronal section at the midaxillary line, the arch of the aorta may serve as an example. In a section at this location, the trachea is usually seen superior to the arch of the aorta while the ascending aorta is inferior to it. The superior vena cava is seen lateral (right) to the arch, while the pulmonary trunk is seen lateral and left of the arch.

In sagittal sections superior/inferior relationships and anterior/posterior relationships are used in locating any structure. The trachea is used as an example here. The thyroid gland is seen anterior to the trachea and bodies of thoracic vertebrae are seen posterior to the trachea. The cricokJ cartilage of the larynx is observed superior to the trachea in a mid-sagittal section, and the left primary bronchus may be seen at the inferior end of the trachea. The standard information about each structure varies. However, all structures should be described as seen in any particular section regardless of plane. In a mid-sagittal section the trachea appears as an elongated (superior to inferior), half cylinder shaped structure, whereas in a transverse section, it appears as a round structure. Another piece of information, which is standard regardless of plane, is the location of the structure.. The location of organs and structures can be determined by measurement using some assigned reference point. In some preliminary studies of transverse (axial) sections of the cadaver (Table 1), the

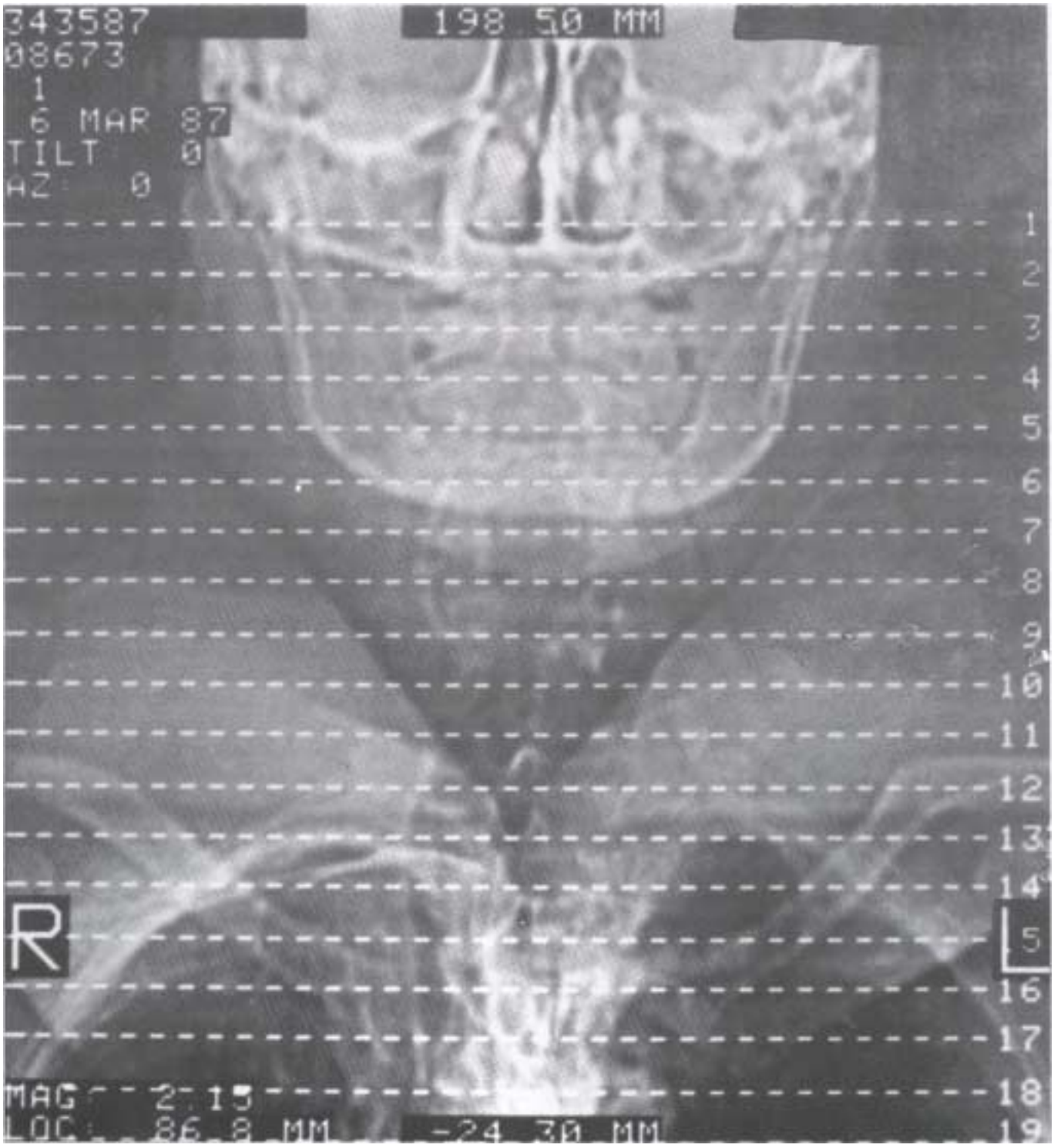


Figure 1. Computed tomography scan, showing the orientation lines for reference slices.

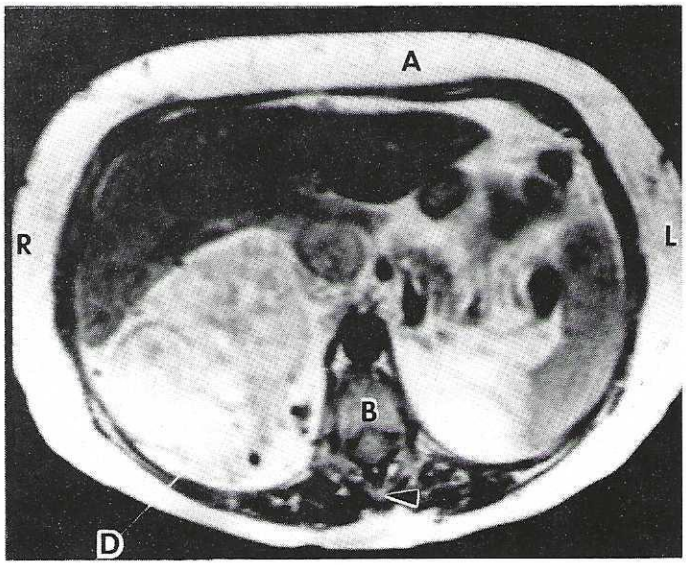


Figure 2. Magnetic resonance image. Transverse (axial) section of the abdomen which highlights the MUSCULOSKELETAL UNIT. A - Rectus abdominis muscle, Vertebra (B - Body, arrowhead - spinous process), D - Hypernephroma.

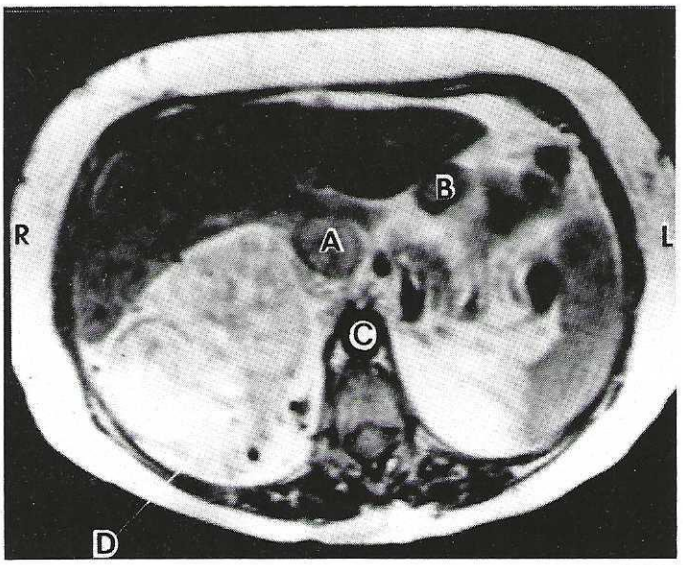


Figure 3. Magnetic resonance image. Transverse (axial) section of the abdomen which highlights the NEUROVASCULAR UNIT. A - Inferior vena cava, B - Hepatic portal vein, C - Descending abdominal aorta, D - Hypernephroma.

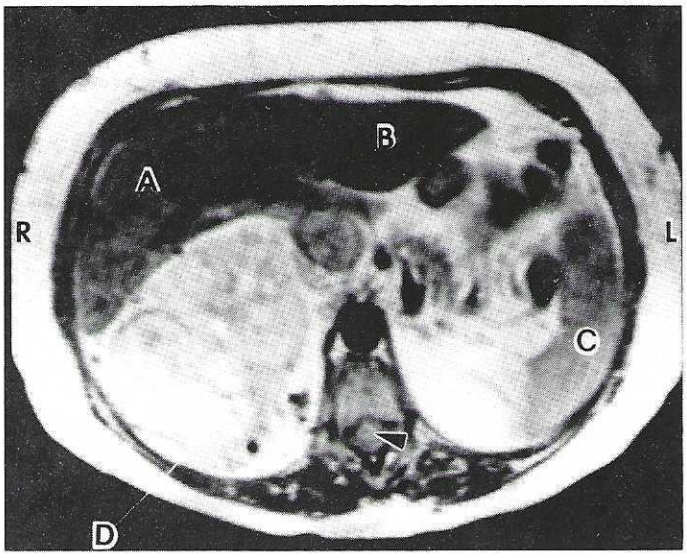


Figure 4. Magnetic resonance image. Transverse (axial) section of the abdomen which highlights the VISCERAL UNIT. Liver (A - Right lobe, B - Left lobe), C - Spleen, arrowhead - Spinal cord.

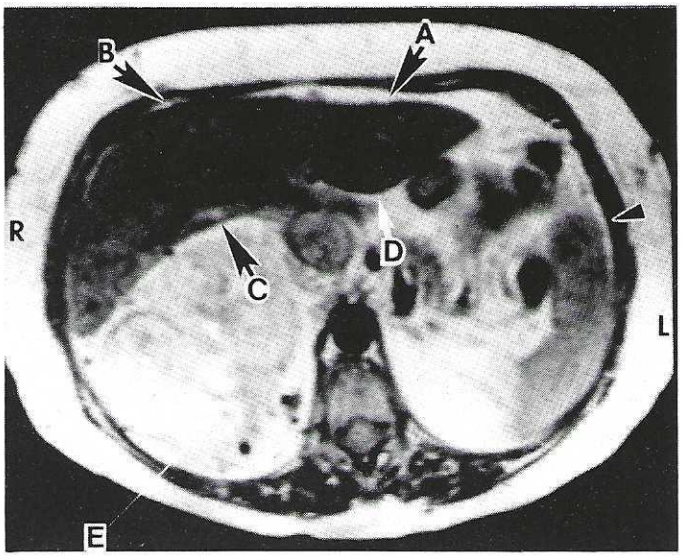
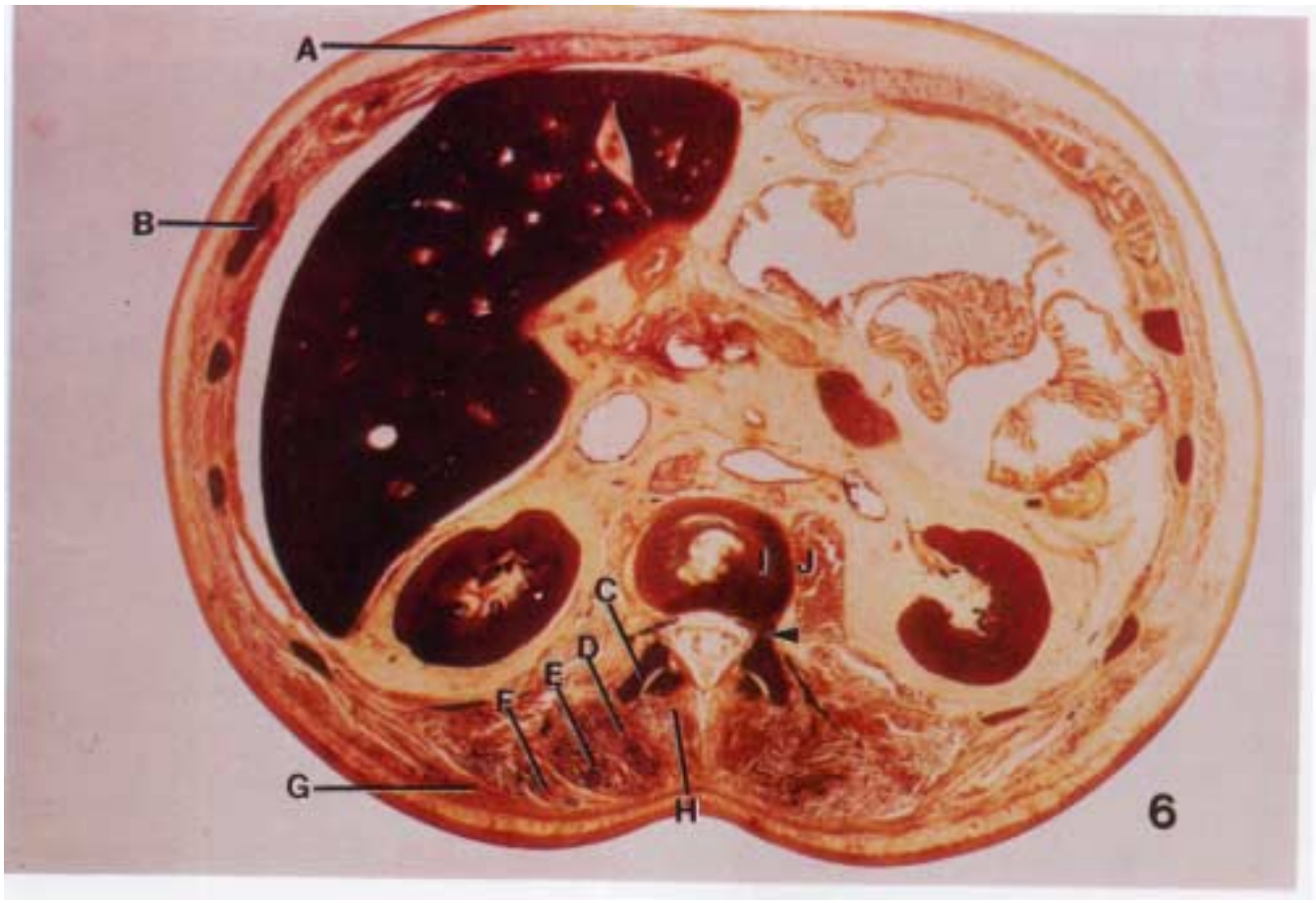


Figure 5. Magnetic resonance image. Transverse (axial) section of the abdomen which highlights the ENCLOSING UNIT. Subphrenic space (A - Left, B - Right), Subhepatic space (C - Right, D - Left), arrowhead - Peritoneal cavity.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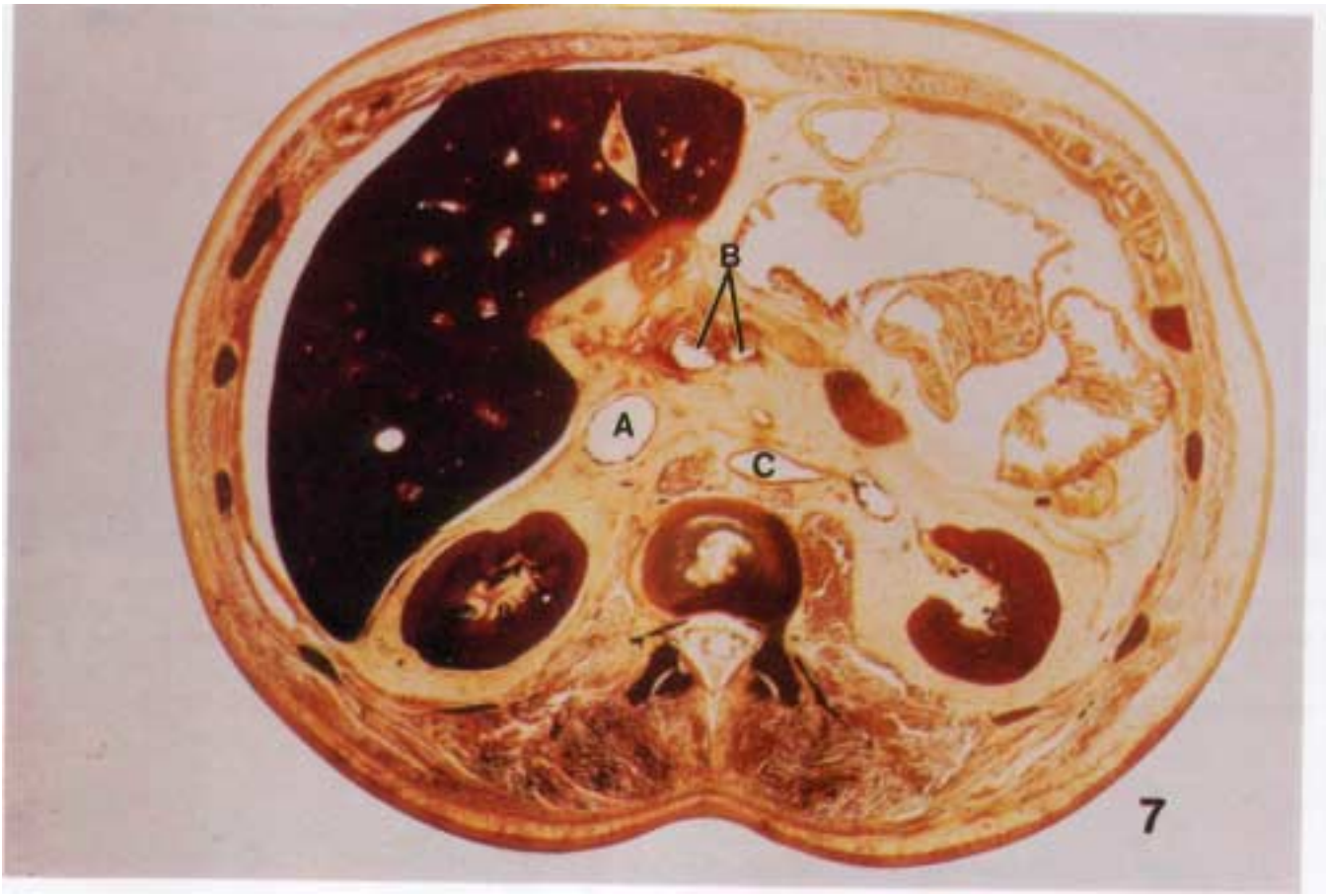
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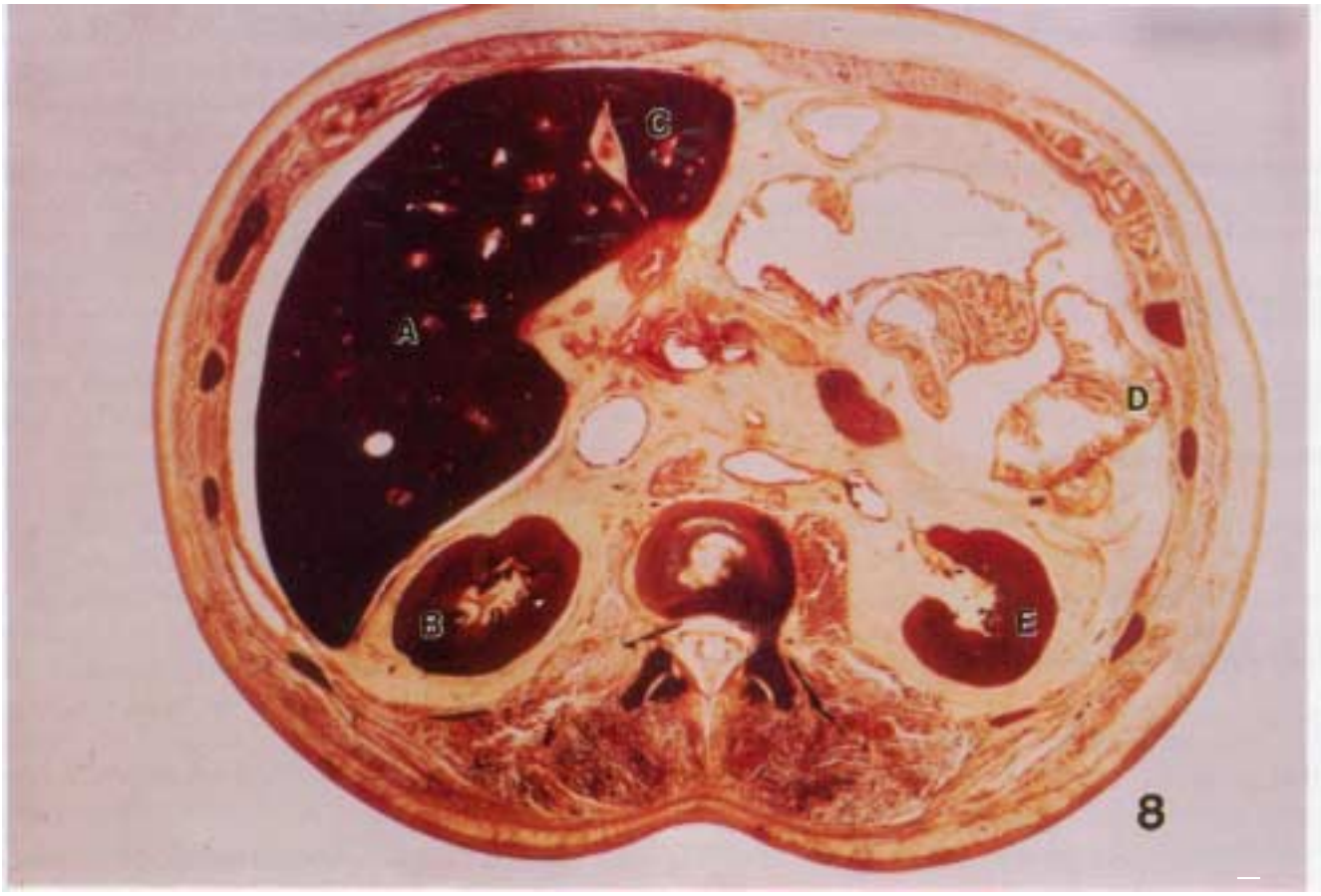
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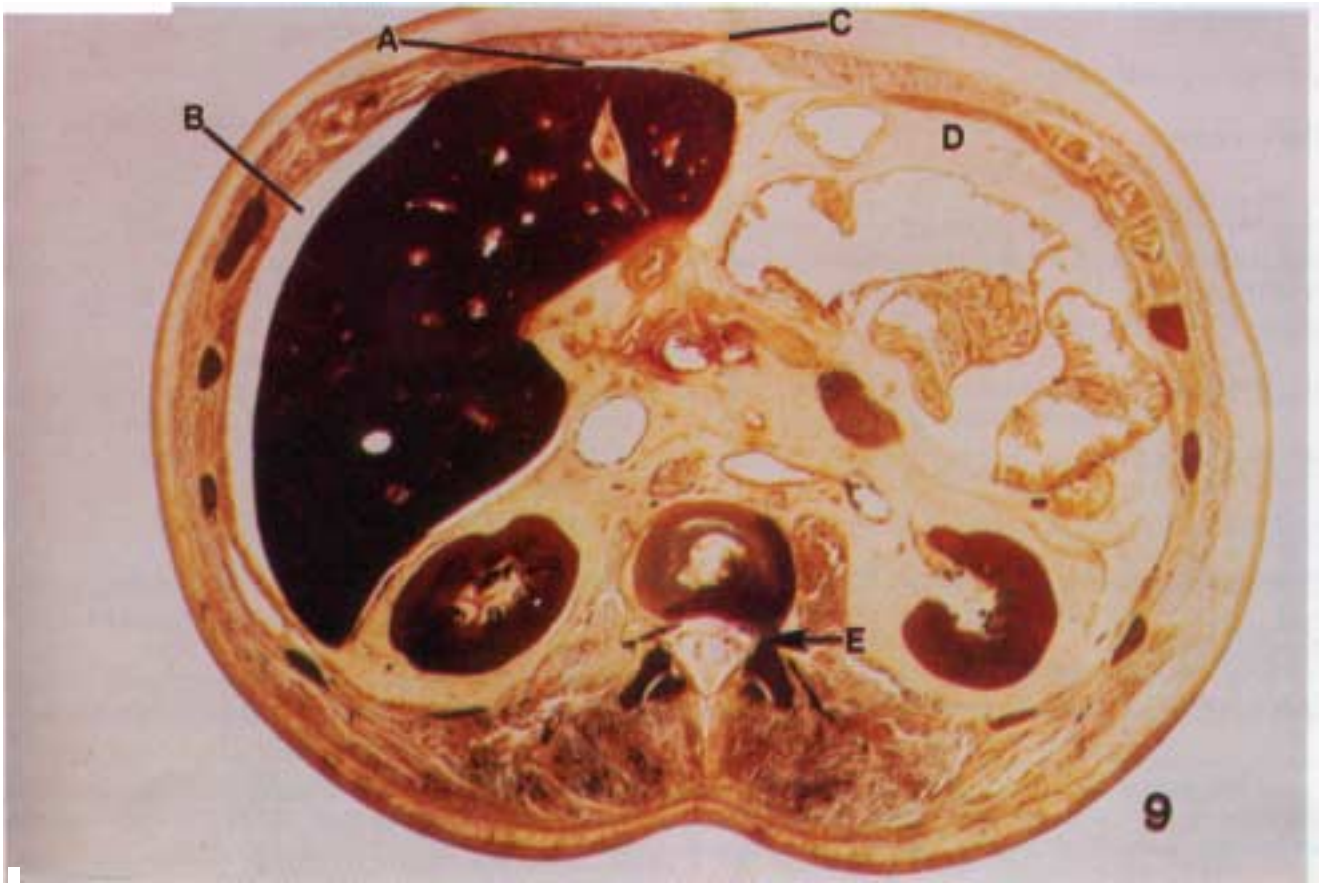
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Table 1 : Distance, clock, and angle position of structures in head transverse (axial) cadaver sections.

		Level	Distance	Angle 1	Angle 2	Clock
Internal occipital protuberance	—	8 cm +	65	90	0	6:00
Internal occipital protuberance	—	6 cm +	72	90	0	6:00
Superior sagittal sinus (anterior portion)	—	8 cm +	75	90	0	12:00
Superior sagittal sinus (posterior portion)	—	8 cm +	70	90	0	6:00
Internal jugular vein (R+)	—	1 cm +	40	42.5	46.5	7:30
Internal jugular vein (Rt)	—	on line	37.5	32.2	57.8	8:00
Sternocleidomastoid (Rt.)	—	1 cm +	87.5	41.1	48.9	7:30
Sternocleidomastoid (Rt.)	—	on line	83	42.0	48.0	7:30
Temporalis (Rt)	---	1 cm +	46	30.7	59.3	10:00
Temporalis (Rt)	—	on line	68	35.5	54.5	10:00
Trapezius (Lt)	—	1 cm +	99	75.9	14.1	5:30
Trapezius (Lt)	—	on line	97.5	77	13	5:30
Semispinalis capitis (Rt)	—	1 cm +	83	68.1	21.9	6:50
Semispinalis capitis (Rt)	—	on line	84	72.2	17.8	6:30
Vomer bone	—	1 cm +	13	90	0	12:00
Nasal septum	—	1 cm +	72	90	0	12:00
Spinal cord	—	1 cm +	53	90	0	6:00
Vitrous body of the left eye	—	1 cm +	74	58.3	31.7	1:00
Nasopharynx	—	1 cm +	6	90	0	12:00
Maxillary sinus (Rt)	—	1 cm +	43	59.4	30.6	1:00
Sphenoid sinus	—	on line	18	90	0	12:00
Condyle of mandible (Lt)	—	on line	54.5	0	90	9:00
Temporal lobe (Lt)	—	on line	46	40.7	49.3	1:30
Medulla oblongta	—	on line	35	90	0	6:00

Level: "on line" = Reid's baseline, "+" = above Reid's baseline; Distance = Millimeters from median-midaxillary junction; Angle 1 = Angle with midaxillary and median-midaxillary junction (degrees); Angle 2 = Angle with median and median-midaxillary junction (degrees); Clock = clock face position.

junction of median and, midaxillary lines were used as the reference point. In addition, organs were located on the section using the clock face analogy. That is, on a cross section of the head, the frontal crest was assigned the 12:00 position and the internal occipital protuberance the 6:00 position.

In addition to location and description of a structure, other information gathered depends upon the kind of structure under consideration. In structures of the musculoskeletal unit, histological class of bone (cancellous or compact) and origin and insertion of muscles should be specified. In the neurovascular unit, origin, distribution, and course of arteries should be noted. Veins and lymphatic vessels should be described in a similar manner, but instead of distribution, termination is more appropriate. The origin, + distribution, and course of each nerve seen in the section should be noted. For structures included in the visceral unit, size (as seen in the section), course, and major functional role should be discussed. However, for the structures of the enclosing unit, description and location are adequate. For example, the interpleural space is seen in thoracic sections as an empty circular space between the visceral and parietal pleurae.

ACKNOWLEDGEMENTS: Bruker Medical Imaging, Inc., Lisle, Illinois, provided the MRI scans. W. B. Saunders Co. published "Modern Sectional Anatomy" (Lane and Sharfaei) which is based on this methodology.

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LEGENDS for color plates, previous pages 19 and 20:

Figure 6. Sheet plastinated. Transverse (axial) section of abdomen which highlights MUSCULOSKELETAL UNIT structures. A - Rectus abdominis, B - Rib, C - Transverse process of a lumbar vertebra, D - Spinalis, E - Longissimus, F - Ilio-costalis, G - Latissimus dorsi, H - Multifidus, I - Body of lumbar vertebra, J - Psoas major, Arrowhead - Pedicle.

Figure 7. Sheet plastinated. Transverse (axial) section of abdomen which highlights NEUROVASCULAR UNIT structures. A - Inferior Vena Cava, B - Superior mesenteric vein and artery, C - Descending abdominal aorta.

Figure 8. Sheet plastinated. Transverse (axial) section of abdomen which highlights the VISCERAL UNIT structures. A - Right lobe of liver, B - Right kidney, C - Left lobe of liver, D - Large bowel (colon), E - Left Kidney.

Figure 9. Sheet plastinated. Transverse (axial) section of abdomen which highlights the ENCLOSING UNIT structures. A - Left subphrenic space, *B* - Right subphrenic space, C - Linea alba, D - Peritoneal cavity area, E - Dura mater.

DESIGN OF A PLASTINATION LABORATORY

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INTRODUCTION

In the fall of 1988 I was asked to initiate the development of a plastination laboratory for the preparation of pathologic teaching specimens for medical students. This paper, the result of this experience, describes how this project was accomplished, and also considers some of the changes that would be made if it had to be done again. The laboratory was designed for utilizing the S10 technique only.

DESIGN OF THE PLASTINATION LABORATORY

The major equipment required for a plastination laboratory is: a freezer for dehydration and impregnation, a vacuum system to remove the acetone and allow the plastic to enter the tissue, and a chamber in which to cure the specimens. Each can present hazards which must be addressed.

A major concern in designing a plastination laboratory is the control of the fire/explosion hazard. The main solvent, acetone, is used in large volumes, and may be very dangerous if not handled properly. Acetone has a flash point of -18 * C. Cooling the acetone in a freezer to -25 * C reduces this potential hazard.

The three ingredients necessary for fire are: Fuel (the acetone), Oxygen (from the air), and Energy (sparks, flame, etc.). If one of the three is absent, then fire will not occur. Since acetone is recommended in the S-10 procedure, and air is all around us, the best alternative was to remove all sources of energy. This premise guided our design. We were fortunate in having a room available which already contained a fume-hood, although extensive modifications had to be made (Fig. 1). Originally there was a large sink unit at bench level in the hood with cupboards underneath. These, along with all the water, steam and gas lines; drains; and electrical outlets were removed. Much of the plumbing, including needle valves from the gas and steam lines, were salvaged and useful when the vacuum lines were connected. A fluorescent light, inside the top of the hood and above a sheet of glass, was sealed off from the chamber using silicone caulking compound. The remainder of the cavity was also sealed so as to create a relatively air-tight box, with no potential internal sources of heat or sparks. The lower area, which had originally been cupboards, was prepared for closure with %" plywood once the freezer was installed.

SELECTION AND INSTALLATION OF FREEZER

If flammable fluids are stored in a refrigerator or freezer, these appliances should be "explosion-proof in the sense of having all potential ignition sources located outside the cabinet. However, if the vapors can escape from the freezer and sink to floor-level (acetone vapor being heavier than air), then the potential for fire or explosion continues to exist if the vapors come in contact with other sources of heat or flame which are inherent in most laboratories. Therefore, unless the whole laboratory is made explosion-proof, an explosion-proof freezer does not necessarily solve the explosion problem. In fact, a modified commercial freezer, under the conditions outlined below, is safer than the more expensive "explosion-proof type.

After reviewing various quotes, a 13 cu. ft. freezer, 23"W x 51 "L x 36"H, with a remote compressor, was installed for $700.00. At the time of installation, care was taken to ensure that the thermostat was also relocated outside the fume-hood, and that all other electrical lines were disconnected (eg. warning lights). One final safety feature was to connect a ground wire to the hinges of the freezer to reduce the possibility of static build-up. A hole (V diam.) was drilled through the wall of the freezer, using care not to damage any cooling pipes hidden in the wall of the freezer. A tygon vacuum line was fed through the hole and the hole around the tubing was sealed with silicone caulking compound. It is important to modify the freezer before it is placed in the fume-hood and connected to the compressor, after which it is difficult to move.

PUMP AND VACUUM LINES

Two inherent problems associated with the use of a vacuum pump are: 1) the motor is a source of ignition and 2) the exhaust contains acetone vapor.

Therefore, the vacuum pump and freezer compressor were located four feet from the fume-hood (Fig. 2). The vacuum pump (Fig. 3) was taken from an out-dated Hitachi electron microscope which was no longer in use. The vacuum and exhaust lines were located underneath the counter as they run from the pump to the hood. Note that the exhaust from the pump ends up in the fume-hood, where the vapors are safely vented. The vacuum control unit is mounted inside the hood on the right side, about 18 inches above the freezer (Fig. 4). The vacuum lines were made from %" copper water pipe, with

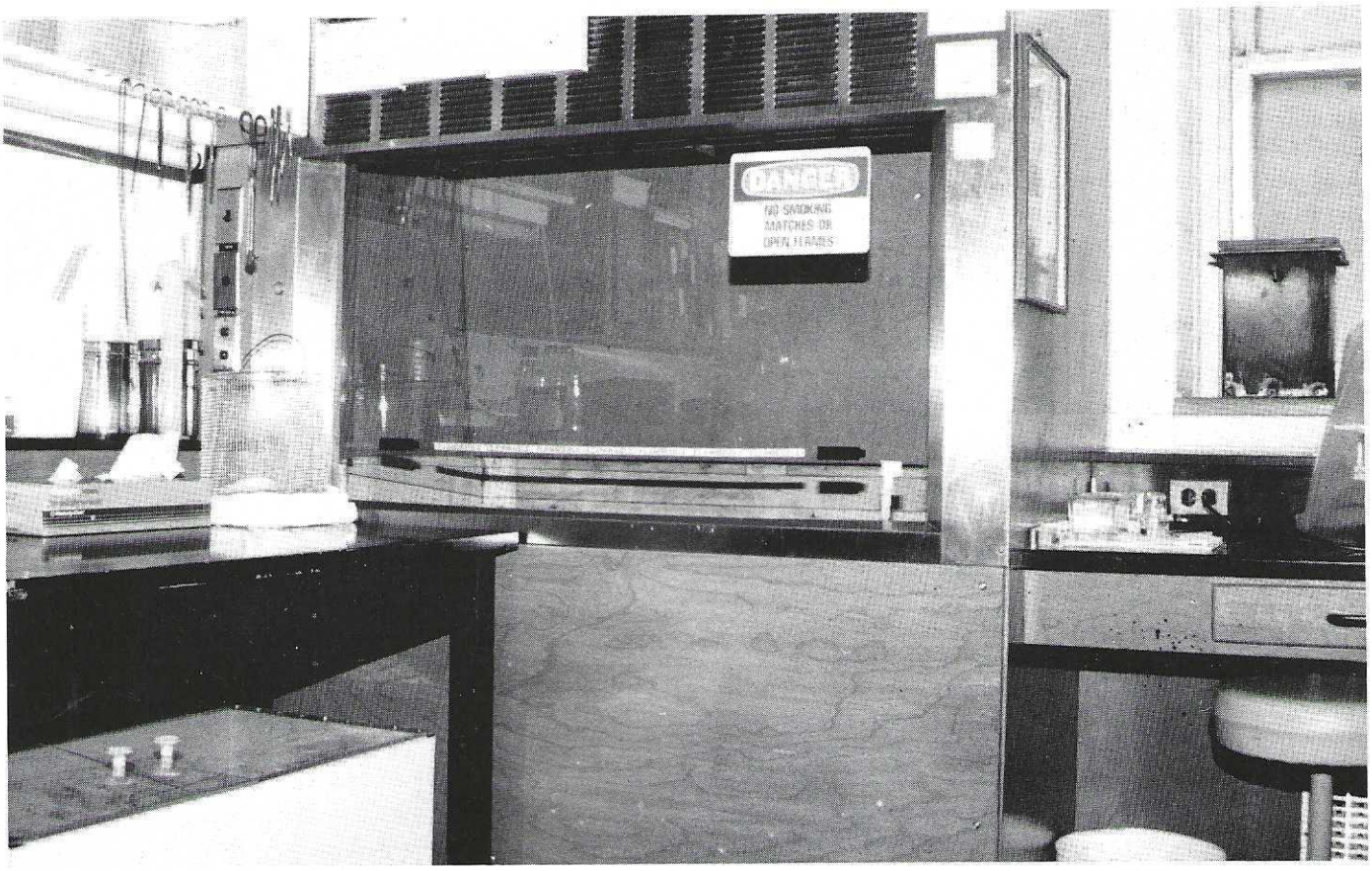
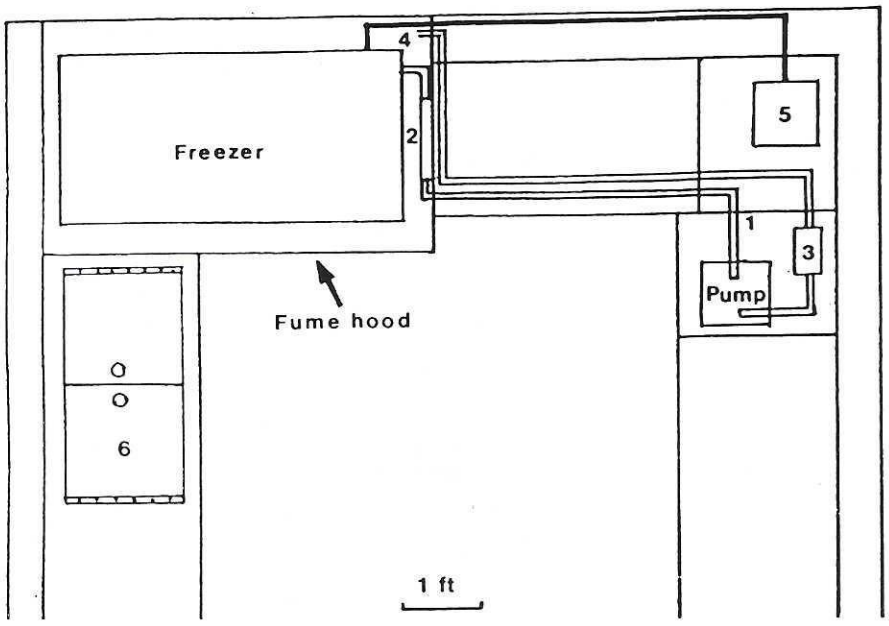


Figure 1. Rear of laboratory showing fume-hood (center) and gas cure box (lower left, under the counter).



1) Vacuum line
2) Vacuum controls
3) Exhaust trap
4) Exhaust vent
5) Freezer compressor
6) Gas cure box

Figure 2. Schematic of rear area of laboratory (See Fig. 1).

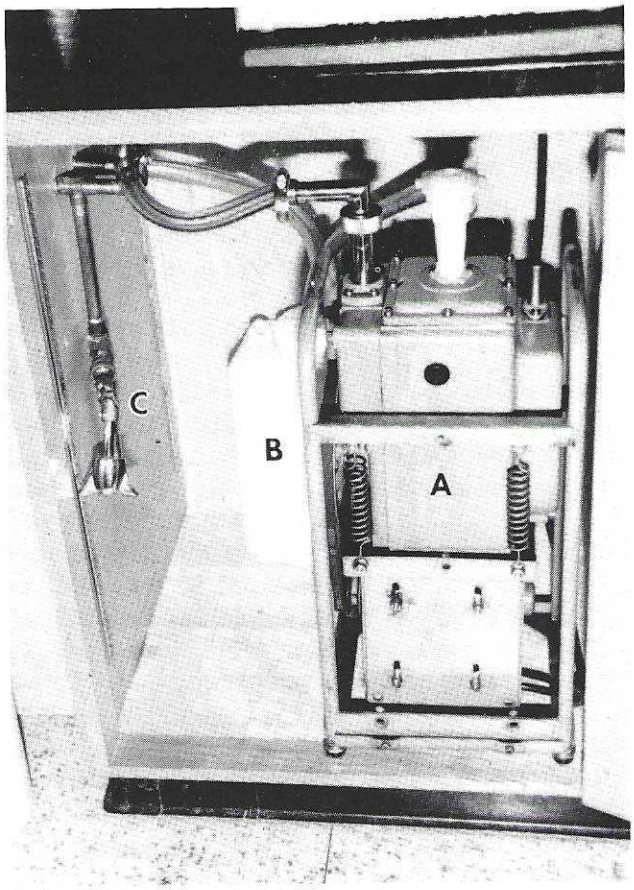


Figure 3. Vacuum pump loacated underneath the counter. A - pump, B - Oil trap for exhaust, C - Drain in Vacuum line.

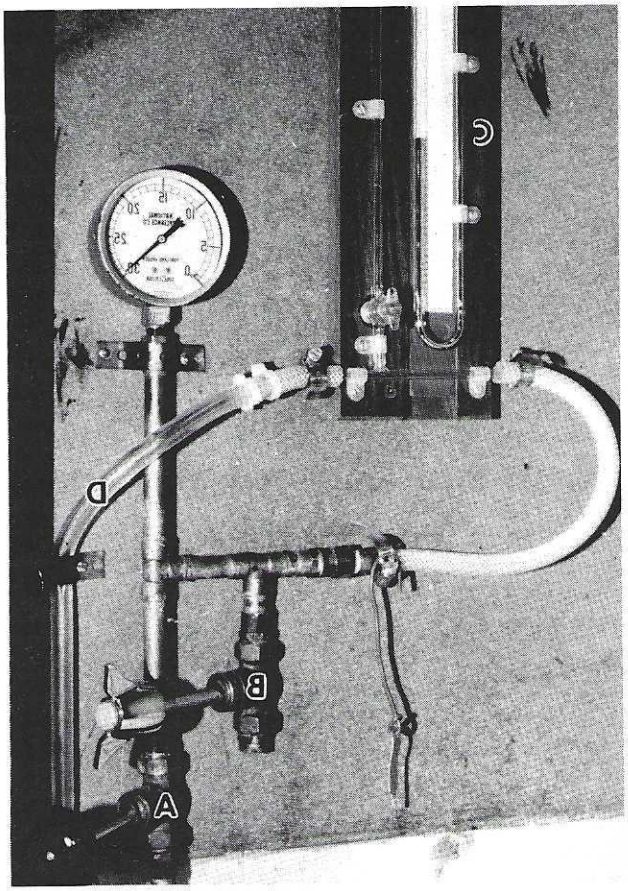


Figure 4. Vacuum control unit. A - Shut-off valve, B - Vacuum control valve, C - Bennert manometer, D - Tubing to freezer.

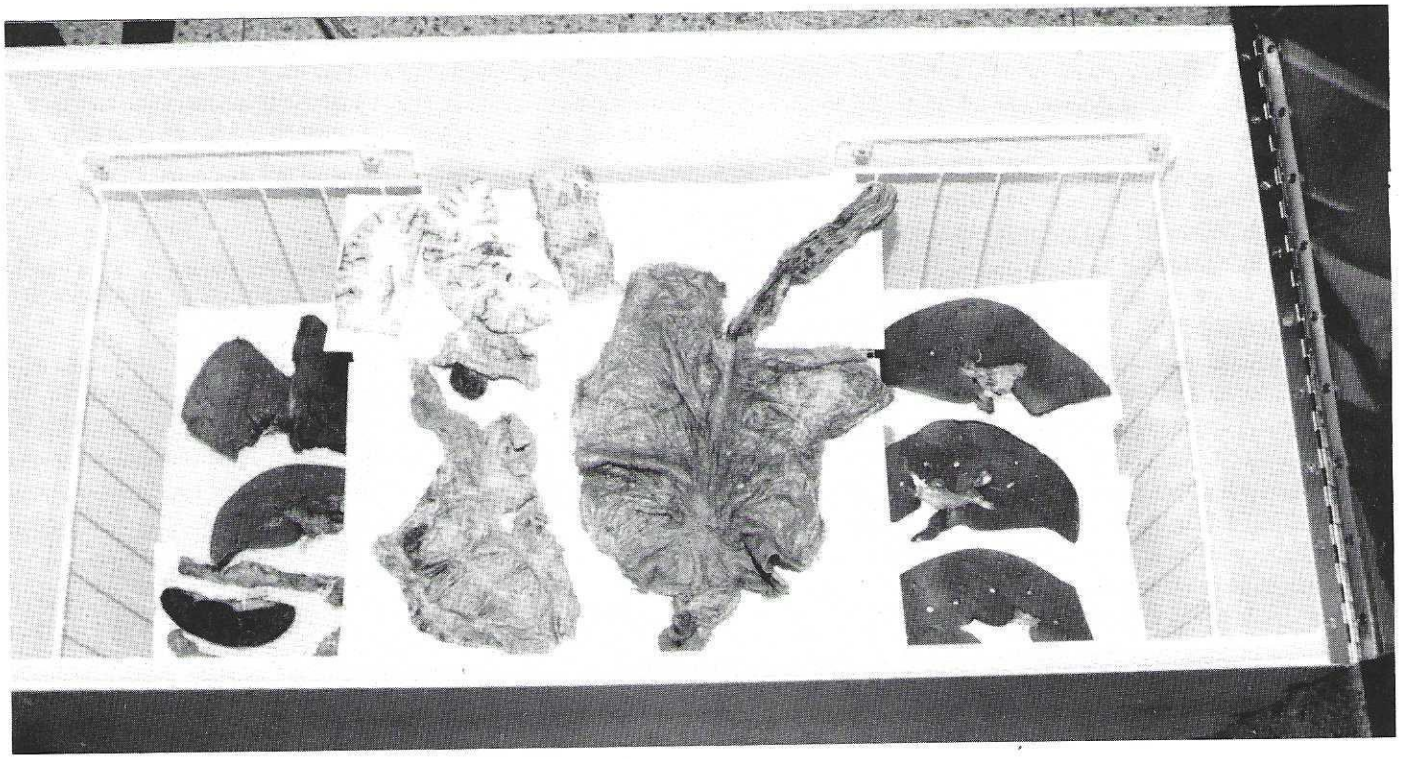


Figure 5. Gas cure box with assorted specimens. Note hinged lid (arrows) on each end.

soldered joints (Rumph, 1987). Using the salvaged pipe, joints and valves from the old fixtures, the cost of the vacuum control unit was minor (approximately $50.00).

The Bennett manometer, an additional but essential expense, was connected with tygon pressure tubing, as was the final connection to the freezer.

GAS CURE CHAMBER

The primary safety consideration with the gas cure chamber, is the ability to disperse the fumes that escape when the lid is opened to manicure the specimens. Initially, we considered using an old incubator, laid on its back. This proved to be awkward and utilized a great deal of space. A box 36"L X 18"W X 24"H was constructed from arborite coated chip-board (the same as a kitchen counter-top) and the top was closed with a hinged, transparent, V. thick acrylic plastic top (Fig. 5). The box was relatively air-tight and was mounted on castors for easy movement. Cost of materials and labor was $350.00. The plastic-coated wire baskets that had come with the freezer were modified and used to make shelves for specimens in the gas cure box. When curing specimens, the box was stored under the counter. When manicuring specimens or opening the box for any other reason, it can be moved out to the front of the fume-hood. By opening the glass front of the fume-hood about 6 inches, a draught is created which is sufficient to draw up any gas from the opened curing box. We have found this to be sufficient to keep the rest of the room free from fumes.

SUMMARY

This design has been in use for one year and has for the most part been entirely satisfactory. We use 12 liter (9" diameter x 11") stainless steel containers for dehydration, and for impregnation one of these cylinders is placed in a Heidelberg kettle, thus reducing the amount of polymer needed to cover the specimens. Specimen baskets, made from V galvanized wire mesh, are used to transfer specimens. These baskets are simply made from a piece of mesh 28" x 9" and an additional piece 9.5" square. First form a cylinder from the large piece, 8.5" diameter and 9" tall; then place the square piece over one end, cut off the excess around the edges, and solder all joints, using a propane torch. Total cost for three baskets was $20.00. Depending on size, approximately 20 specimens can be prepared for plastination at a time. Results have been quite acceptable and we are now concentrating on colour preservation and flexibility.

The one flaw is the location of the vacuum pump in the cupboard under the counter. The pump is noisy and generates considerable heat. Ideally it should have been located outside the laboratory, in an isolated area that could be cooled. Apart from this, the room is quite adequate for the volume of specimens being plastinated, and the air quality is such that the other end of the room is used as a full-time office. Absolutely no acetone fumes can be detected in the room at any time, and the gas cure system, if handled properly, results in only a temporary, mild irritation while actually working in the box.

SOME ADVICE ON HOW TO START:

1) Write to Dr. von Hagens in Heidelberg and order a copy of the Heidelberg Plastination Folder (von Hagens). Enclose a money order for $15.00 U.S. (or current price).

2) While waiting for this to arrive, consider the following points: a) Are the necessary funds available to establish the laboratory, and if so, how much is required? We spent about $5000.00 to establish our laboratory. This will vary according to the availability of used equipment in your department.

b) What space is available for the lab and does it contain a fume-hood or other type of ventilation system?

c) What size of specimens will you be plastinating? Before buying your dehydration containers, be sure they will be big enough.

d) Will you be starting with the basic S10 technique, or are you going to do epoxy slices?

3) Using the addresses given at the back of this Journal, contact a convenient lab and try to arrange a short visit. This should provide you with the basic idea of what is going on. Discuss any problems that they might have had, or continue to have, and plan on overcoming them in your own lab. Do not expect to learn all the details of plastination on this first visit; rather, try to get an overall picture. Later, other visits will provide an opportunity to learn more of the practical details.

ACKNOWLEDGEMENTS: I would like to thank Dr. Robert Kisilevsky, Professor and Head of the Department of Pathology, for his interest and support in establishing this laboratory. My thanks also to Gerhard Penz for allowing me to visit his laboratory and introducing me to the basics of plastination. Finally, thanks to Uoyd Kennedy for taking the photographs for this paper.

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PLASTINATION OF WHOLE ANIMAL PREPARATIONS FOLLOWING HISTOCHEMISTRY: IN SITU LOCALIZATION OF THE ENZYME ACETYLCHOLINESTERASE

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SUMMARY

A method for whole animal acetylcholinesterase (AChE) histochemistry was combined with standard S10 plastination. Resultant whole or partial animal specimens prepared in this manner were superior to whole animal specimens processed for AChE histochemistry and stored in formalin. The results compared favorably with those attained with isolated organ or tissue whole mounts while preserving and enhancing existing anatomical relationships. The technique was found to be useful in studying peripheral autonomic innervation by AChE histochemistry, in teaching specific anatomic details to laboratory personnel, and was suggested to be of potential use with other histochemical or immunonistochemical techniques in a variety of small animals, organs, or tissues.

INTRODUCTION

A variety of methodologic innovations have been devised to examine the distribution of autonomic nerves in organs and tissues of human and laboratory animals. In order to investigate patterns and properties of peripheral autonomic innervation, diverse neuro-histochemical techniques including enzymatic and immunohistochemical methods (Koelle and Friedenwald, 1949; Costa et al., 1980) have been applied to whole mounts of organs or organ laminae (Baljet and Drukker, 1975; Costa et al., 1980; Papka et al., 1981; Papka et al., 1985) or to paraffin or cryostat sections of organs (El-Badawi and Schenk, 1967; Papka et al., 1985; Schultzberg et al., 1979). Organ or tissue whole mounts are particularly useful for *in situ* demonstration of ganglia, ganglionated nerve plexuses, nerve trunks, and nerve fibers. Since anatomical relationships are preserved, these specimens are valuable for studying the density and distribution of nerve fibers and neuroeffector relationships.

We have developed a technique to produce permanent animal mounts by standard S10 plastination (Bickley et al., 1981; Bickley, 1980; von Hagens, 1979a; von Hagens et al., 1987; von Hagens, 1979b) of whole or party animal specimens following *in situ* histochemistry; in particular, localization of the enzyme acetylcholinesterase (AChE). Specimens prepared in this manner can be used as a reference during regional dissections of other animals to obtain tissue blocks for research purposes that contain specific anatomic structures of interest. They can also be utilized to teach laboratory personnel about important anatomic relationships as they relate to specific research projects. Plastinated whole or partial animal preparations are superior to organ or tissue whole mounts because additional anatomic relationships are maintained and they are permanent.

MATERIALS AND METHODS

ANIMAL PERFUSION, SPECIMEN PREPARATION AND AChE HISTOCHEMISTRY

An adult female Sprague-Dawley rat was anesthetized with intraperitoneal sodium pentobarbital (50 mg/kg) and fixed *in toto* by transcardial perfusion with normal saline (300 ml of 0.9% NaCI in water) followed by a fixative solution of 4% paraformaldehyde in 0.1 M phosphate buffer adjusted to a pH of 7.1. The total time of fixation was 4 hours. After fixation, regional abdominal and pelvic dissection was performed to expose the organs (intestinal tract, uterus, cervix, vagina) of interest to this laboratory. The urinary bladder and urethra were removed in order to reveal the ventral surfaces of the uterine cervix and vagina. Since we were primarily interested in the organs of the female genital tract, the body of the rat was transected at approximately the level of the kidneys and the rostral portion discarded. The caudal portion was washed thoroughly with 0.2 M acetate buffer (pH 5.5),and then preincubated in acetate buffer containing 10 M ISO-OMPA to inhibit nonspecific chdinesterase activity. The caudal body segment was incubated at ambient temperature and lighting for 16 hours in a medium containing acetylthiocholine iodide as substrate and 10^{-4} M ISO-OMPA. The medium was prepared In the following manner: acetylthiocholine iodide (500 mg) was dissolved in 20 ml of distilled water, and then 35 ml of a 1.0 M copper sulfate solution was added. The resultant solution was agitated thoroughly and centrifuged at 2000 rpm for 20 minutes. To 50 ml of dear supernatant, 310 mg of glycine was added, the pH was adjusted to 5.5 with 1.0 M sodium acetate, and the total solution volume was increased to 250 ml with distilled water. Next,

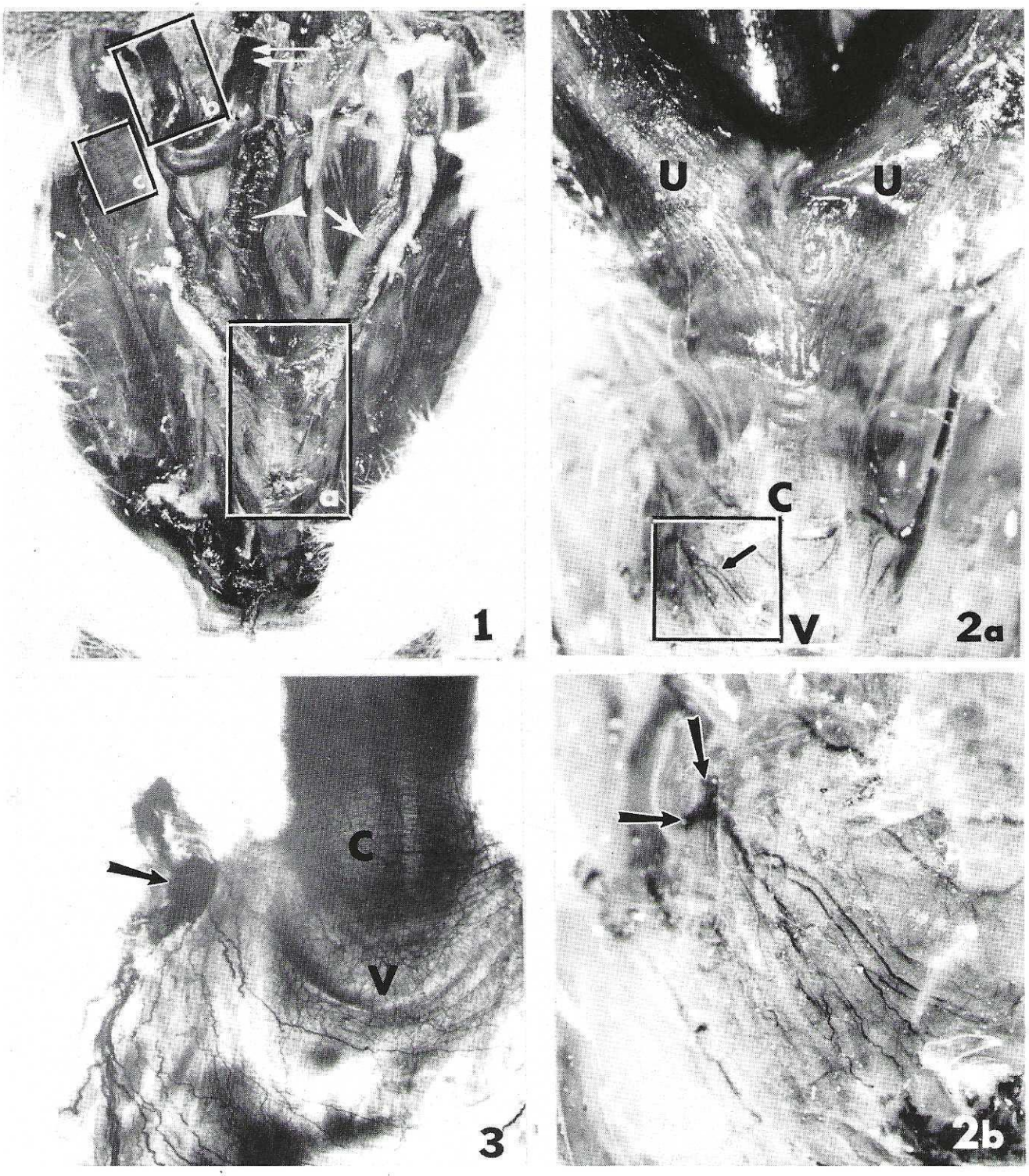


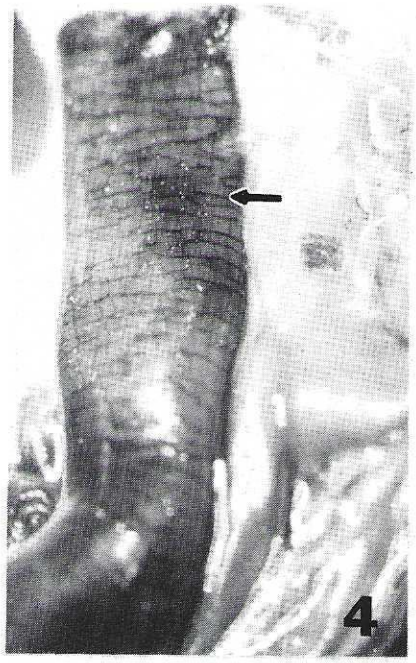
PLATE 1

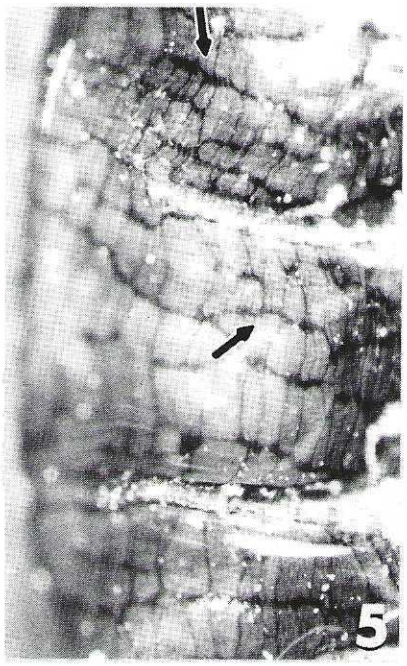
Figure 1. Low power macrophotograph of the pelvic and lower abdominal contents of a female rat processed *in situ* for AChE histochemistry. Caudal is toward the bottom of the photograph. Major landmarks include the colon (arrowhead), uterine horn (arrow), and small intestine (double arrow). *Box a* outlines the uterine cervix and rostral vagina and is enlarged in Fig. 2. *Box b* outlines part of the gastrointestinal tract and is enlarged in Fig. 4. *Box c* outlines part of the lateral body wall and is enlarged in Fig. 7.

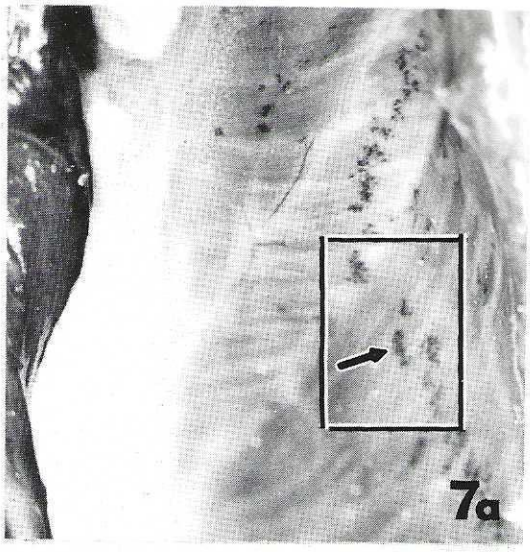
Figure 2a. Enlargement of *box a* of Fig. 1. AChE-positive nerves (arrow) are evident as they emerge from the paracervical ganglion and course across the cervix-vagina junction. Uterine horns (U), Cervix (C), Vagina (V). X 8.6.

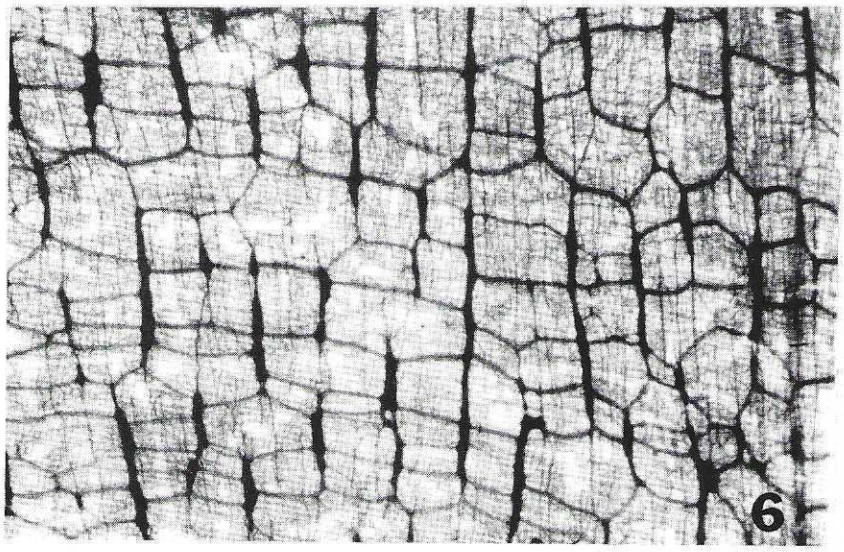
Figure 2b. Enlargement of the area in the *box* of Fig. 2a. Note the AChE-positive nerves emanating from the paracervical ganglion (arrows). X 13.5.

Figure 3. Isolated whole-mount preparation of the uterine cervix (C) - vagina (V) junction comparable to the area illustrated in Fig. 2. AChE-positive nerves stem from the paracervical ganglion (arrow) and form plexuses associated with the cervix and vagina similar in pattern to those observed in whole body preparations. X 13.5.









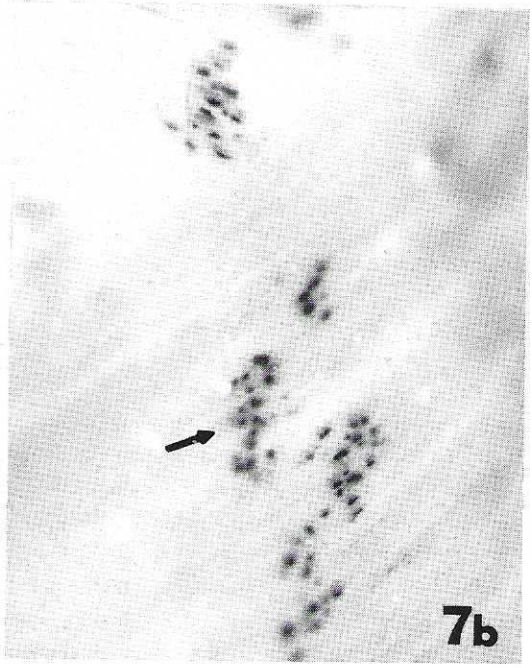


PLATE 2

Figure 4. Low magnification of intestine (*box b*, Fig. 1). Even though some parts of the gut are slightly out-of-focus, it is possible to appreciate the arrangement and density of the AChE-reactive myenteric plexus (arrow) around the wall. X 6.8.

Figure 5. Higher magnification of the intestinal wall showing the arrangement of the AChE-positive nerves forming the myenteric plexus (arrow). X 9.2.

Figure 6. Whole-mount preparation of the myenteric plexus of the gut, histochemically stained, demonstrating the AChE-positive ganglionated nerve plexus. X 10.8.

Figure 7a. Low magnification of the body wall musculature (*box c*, Fig. 1). Note numerous AChE-positive motor endplates (arrow) aligned at the muscle midpoints. X 9.2.

Figure 7b. Higher magnification (*boxed area*, Fig. 7a) demonstrating detail of the motor endplates. X 23.4.

the reaction product was developed by placing the specimen in a solution of ammonium sulfide [1 part ammonium sulfide (light) :22 parts distilled water] for one minute. The specimen was washed thoroughly in 0.2 M acetate buffer (pH 5.5), drained, blotted dry, and frozen in an ultracold freezer (-70 °C) for six hours pending plastination.

To compare the histochemical reaction product localization for AChE of this technique to that of isolated whole mount preparations of organs/structures, specific tissues were prepared as whole mounts. Rats were killed by an overdose of sodium pentobarbital (50 mg/kg, i.p.) followed by exsanguination. Fresh tissues (i.e. intestine and uterine cervix/vagina) were dissected in phosphate-buffered saline (PBS). The wall of the intestine was opened, gently stretched and pinned to balsa wood and fixed for 14-16 h in cold picric acid-2% formaldehyde in 0.1 M phosphate buffer, pH 7.1, whereas, the cervix/vagina preparation was fixed intact. These specimens were dehydrated in ethanol, cleared in xylene, and rehydrated in PBS. At this point, the different layers of the intestinal wall were separated into the outer longitudinal smooth muscle, the inner circular smooth muscle, the submucosa, and the mucosa. In these intestinal preparations, the myenteric plexus of nerves remains adherent to the outer muscle layer which produces a thin preparation facilitating the observation of nerves and ganglia. The cervix/vagina preparation required only the removal of excess fatty tissue. Acetylcholinesterase histochemistry was performed on both whole mount preparations (intestine and cervix/vagina) as described above.

S10 PLASTINATION

The caudal segment of the rat was dehydrated via freeze-substitution in -25 "C acetone for 6 weeks using 3 changes of acetone at 3, 2, and 1 week intervals. The specimen was submerged in a standard mixture of Biodur S10/S3 (von Hagens, 1985). After a 24 hour period of ambient pressure, vacuum was applied initially at 160 Torr and decreased by approximately 3 to 5 Torr daily over the next 4 weeks until the pressure reached 10 Torr. Subsequently, the pressure was decreased by 1 Torr each day until the pressure reached 4 Torr. At this time the system was completely closed with no additional air ingress. This setting was maintained for an additional 5 days until all bubble activity at the specimen surface had ceased.

Following forced impregnation and restoration of ambient pressure, excess polymer was drained from the specimen at -25 °C. For a period of 24 days, the specimen was removed daily from the freezer and placed in a closed gas cure chamber at room temperature. The chamber contained volatilized S6 gas and the specimen was exposed to this gas for increasing intervals. For the first 5 days, the specimen was exposed to 86 for 15 minute intervals (6 to 10 times per day) and then returned to the freezer for at least 30 minutes between gas exposure periods. Excess polymer was wiped from the surfaces of the specimen at least once during the cure period and always before returning the specimen to the freezer. On successive days, the length of exposure time was increased by approximately 5-10 minutes each day while the number of exposure periods per day was decreased. Finally the specimen was allowed to remain continuously in the gas cure chamber for 5 additional full days. The plastinated specimen was photographed with a Wild photomacroscope; whereas, photographs of the tissue whole mounts were taken on a Leitz Orthomat microscope.

RESULTS

The normal anatomic relationships of structures within the lower abdominal and pelvic cavities of the rat were maintained following AChE histochemistry and S10 plastination (Fig. 1). With the aid of a dissecting microscope, the dark histochemical reaction product resulting from the AChE activity of nerve fibers, ganglia, ganglionated nerve plexuses, and skeletal muscle motor end plates was readily visualized in the plastinated specimen (Figs. 2, 4, 5, 7). Selected areas from the plastinated specimen (Figs. 2, 5) were comparable in appearance and patterns of enzyme localization to corresponding areas from isolated whole mounts (Figs. 3, 6, respectively).

Low power microscopy of the uterus, cervix, and vagina (Figs. 1, 2a) demonstrated the presence of AChE-positive nerve fibers. Their density and distribution as they radiated out from the paracervical ganglion toward the junction of the cervix with the vagina was evident (Figs. 2a, 2b).

In intestine (Fig. 4) at low magnification, the expected AChE-reactivity of the ganglionated myenteric plexus (Fig. 5) was observed. The net-like appearance of the plexus was similar to that achieved with AChE histochemistry of isolated whole mount preparations (Fig. 6). Motor end plates appeared as discrete structures aligned along the muscle motor point (Figs. 7a, 7b) in the skeletal muscle of the lateral body wall.

DISCUSSION AND CONCLUSIONS

Whole animal preparations offer advantages over whole mounts of organs or organ laminae, since the latter are limited to examination of single organs or to portions of a single organ. Whole animal preparations demonstrate and preserve anatomic relationships that exist between organs and functionally related structures. A major problem with the current methods for preparing whole mounts is that most organs must be separated into laminae or physically disrupted in some manner to gain the resolution needed for microscopy. In addition, they must be stored in fixative (usually formalin) and therefore have a limited useful life. The disadvantages

inherent to fixed specimens (unpleasant odor; irritation to eyes, skin and mucous membranes; necessity to maintain a moist environment; and discoloration and deterioration with prolonged storage) make this method of preservation and storage less than optimal.

Use of standard 810 technique to plastinate whole mounts stained for acetylcholinesterase (AChE), results in specimens superior to those stored in formalin that compare favorably with isolated organ or tissue whole mounts in which AChE activity has been demonstrated. Plastinated whole animal specimens are particularly valuable for demonstrating to laboratory personnel the proper tissue blocks to remove from experimental animals for preparing whole mounts of organs or organ laminae or for freezing and cryostat sectioning. We suggest that this method could be used for other types of histochemical, and possibly immunohistochemical procedures in a variety of small animals, organs, and tissues.

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CHAFFEY COLLEGE *NEWS* ***RELEASE !!***

Many of Southern California's most popular attractions, including **Disneyland, Knotts Berry Farm, Universal Studios, Hollywood, The San Diego Zoo, Sea World,** and **beach resorts** are **a** short distance from the college. A group trip to Disneyland is tentatively planned for August 10th. Space is limited to the first 100 applicants. Registration fees include the 3-day meeting, a journal covering all lectures, demonstrations and abstracts, lunches and refreshments. Discounts will be given to attendees who register prior to June 30, 1990. The cost for the 3-day conference is $100.00 for I.S.P. members and $115.00 for non-members. After June 30th, the costs are $125.00 for I.S.P. members and $140.00 for non-members. Students may attend at the special rate of $15.00 per day or $40.00 for the entire meeting. Anyone currently working on new developments in Plastination is invited to speak on their work. For information contact: Jim Johnson, Plastination Technician, Chaffey College, 5885 Haven Ave., Rancho Cucamonga, CA 91701 (714) 941 - 2355.

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STAINING OF HUMAN FETAL AND ADULT BRAIN SLICES COMBINED WITH SUBSEQUENT PLASTINATION

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INTRODUCTION

Teaching morphological aspects in neuroscience is accompanied by two main problems. 1. Wet brain tissue is inconvenient to handle and is easily destroyed when demonstrated a number of times. 2. Macroscopic demonstration of subcortical nuclei is difficult in unstained sections. In particular, subdivisions of nuclear greys are difficult to delineate. These two problems are accentuated when demonstrating human fetal brain tissue. These problems can be prevented by the methods described in this paper which combine the advantages of staining of brain tissue sections with those of plastination (Ulfig and Wuttke, 1990). An additional procedure, prior to staining, is necessary for the human fetal brain tissue because of its high vulnerability. Fetal brain tissue should be embedded in a suitable medium before cutting and staining, otherwise these sections will not withstand the staining procedure.

MATERIALS AND METHODS

ADULT HUMAN BRAIN TISSUE

Fixation: Fix human brain material by immersion in a 4% aqueous formaldehyde solution for several months.

Cutting: Rinse in running tap water overnight. Cut sections (1 to 4 mm) with the aid of a freezing microtome. [Warm a plane block of aluminum to about 35 ° C. Place this block to the surface of the frozen tissue for a few seconds (the time required is determined after a few trials). Sections should be cut at the junction of frozen and thawed tissue. Sections may also be cut with the aid of a macrotome or a knife (0.5 to 2 cm].

Oxidation: Rinse sections in running tap-water and oxidize in freshly prepared performic acid [100 ml perhydrol (30% H_2O_2) to 900 ml 100% formic acid] for one hour. Sections must be kept in motion during oxidation. Handle the performic acid solution with great care under a chemical hood. Following one hour of oxidation, wash sections under tap-water for at least one hour and considerably longer when thick macrotome sections are used. The previous color of the sections should have returned prior to staining.

Staining: Stain sections with either of two stains: 1) Astra blue [dissolve 0.1 g astra blue (Merck) in 1000 ml distilled water and add 1 ml HCI (37%)] for two days. Keep sections in motion, or 2) After rinsing in 70% ethanol for 15 minutes, stain with aldehvdefuchsin for two days. Keep sections in motion. Prepare aldehydefuchsin stock solution by dissolving 0.5 g pararosanilin (Chroma) in 100 ml 70% ethanol and then add 1 ml of 25% HCI and 1 ml 100% crotonaldehyde. Shake briefly and let react for one week. Use this stock solution for one week only. Prepare the staining solution by adding 6 ml stock solution, 100 ml distilled water, 50 ml 100% formic acid, and 5 ml performic acid (prepared as for oxidation) to 400 ml of 96% ethanol and filter.

Dehydrate: Sections for 12 hours in each ethanol solution (70, 80 and 96%), and then for at least 24 hours in 100% ethanol. In the 96 and 100% ethanol solutions, place the sections between filter paper and perforated porcelain plates.

Plastination procedure: Transfer the sections, covered by filter paper and between perforated plates, to 100% acetone for one day (1 - 4 mm sections) or three days (0.5 - 2 cm sections). Place the sections, between perforated porcelain plates, into the S10/S3 polymer mix in the vacuum chamber for three days (1 - 4 mm sections) or seven days (0.5 - 2 cm). Remove sections and drain excess silicone. For curing, transfer the sections to a curing chamber with a large volume of S6. During the first few hours of curing, excess silicone must be regularly wiped off to avoid superficial precipitations. After the surface is cured, transfer sections to an air-tight container, which has calcium chloride in it to remove the moisture. The sections are ready after two weeks.

FETAL HUMAN BRAIN TISSUE

Fixation: Fix by immersion in a 4% aqueous formaldehyde solution for several weeks.

Dehydrate: in a graded series of ethanol. After dehydration in 100% alcohol, place into a refrigerated mixture (1:1) of 100% diethyl-ether and absolute ethanol.

Embedding: with Cedukol (Merck, Darmstadt), produces optimal results. Soak in 2%, 4%, and 8% solutions of celloidin. Finally, concentrate to a 16% solution of celloidin. Harden in 70% ethanol (Romeis, 1989).

Cutting: Cut at 0.5 - 5 mm with the aid of a freezing microtome. The block must always be wetted by 70% alcohol and sections must be stored in 70% alcohol. Excess celloidin should be cut away.

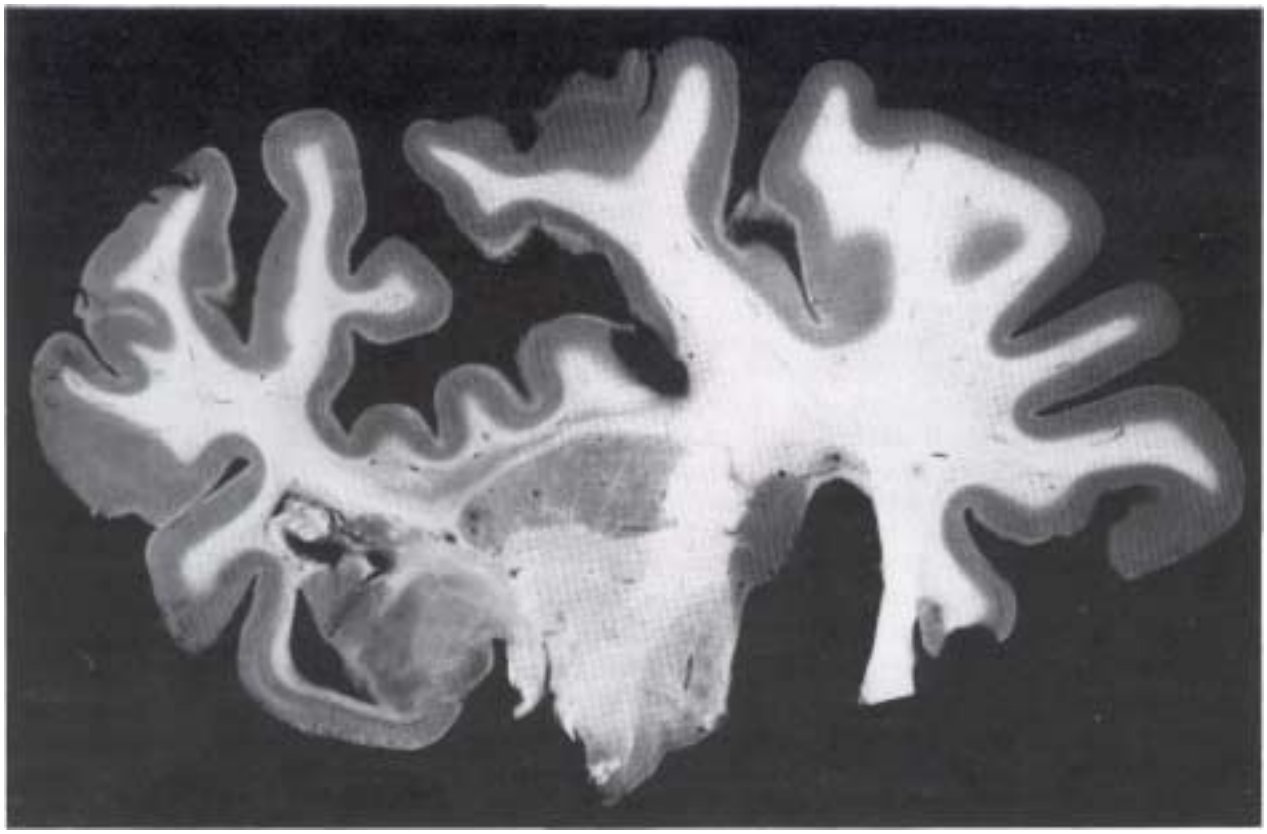

Figure 1. Coronal frozen section (2 mm) stained with astra blue and subsequently plastinated.

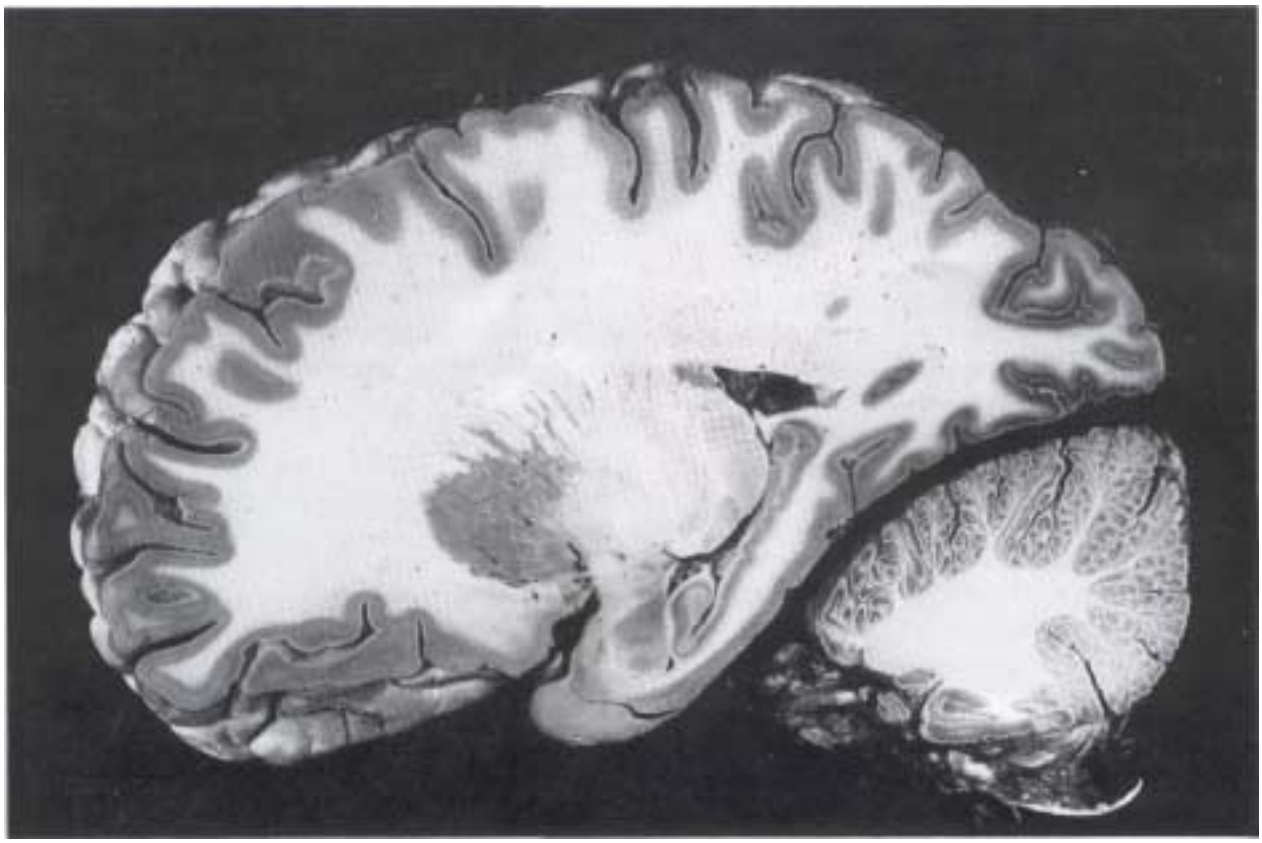

Figure 2. Sagittal macrotome section (1 cm) stained with astra blue and subsequently plastinated.

Staining: Rinse the sections in distilled water for 10 minutes before the staining procedure. Stain section with Darrow-red free-floating (Powers et al., 1960; Powers and Clark, 1963 slightly modified). Prepare a stock solution: dissolve 0.1 g Darrow-red (Aldrich Chemical Company, Inc., USA) in 4.8 ml acetic acid and 395.2 ml distilled water. Stir and boil gently for 10 minutes. Cool to room temperature and filter. Use 4 parts of stock solution and 1 part 0.2 molar sodium acetate solution to prepare the staining solution which should be used the same day. Stain the sections for 12 hours. Keep sections in motion while staining.

Dehydration: Place sections into 70% ethanol for 20 minutes and observe carefully. If sections lose color, transfer them to 96% ethanol immediately. Place sections between filter paper and perforated porcelain plates to obtain flat sections. Leave sections in 96% ethanol for one hour and transfer into 100% propylalcohol for 60 minutes (between plates). Do not use 100% ethanol. Then dehydrate sections free-floating in 100% propylalcohol for 20 minutes. If sections are stained too intensively, keep sections in 100% propylalcohol until the desired intensity is achieved.

Plastination procedure: The same procedure as described above.

RESULTS AND DISCUSSION

ADULT HUMAN BRAIN TISSUE

Both frozen sections, as well as, thicker macrotome sections may be used for these staining techniques. A series of the thinner frozen sections provides more detail of nuclear configurations. Adult brain sections, stained with astra blue, provide a sharp contrast between grey and white matter, thus facilitating the demonstration of the macroscopic morphology of the brain (Figs. 1, 2). The contrast between white and grey matter is more pronounced than achieved by the staining method described by Sincke (1926). Staining with astra blue is preferred to Mulligan's method (1931) as astra blue stains the whole block and does not fade (Braak, 1978b).

Aldehydefuchsin demonstrates the distribution of lipofuscin granules in nerve cells of the adult brain. Varied lipofuscin content is visualized by different staining intensities (Figs. 3, 4). In this manner, a topographical delineation and the internal organization of nuclei can be studied by this pigmento-architectonic approach. (Braak, 1978a; 1984). The preparation of the aldehydefuchsin staining solution is complicated. Therefore, this staining procedure is much more complex than with astra blue. Commercially prepared aldehydefuchsin is not recommended since it may stain the white matter. Furthermore, it tends to produce artificial spots of intense staining in thick sections. For these same reasons, crotonaldehyde is used instead of paraldehyde, and pararosanilin is employed instead of fuchsin (Braak, 1978a). To obtain satisfactory results, only adult brains of more than 40 years of age should be used for the staining with aldehydefuchsin.

FETAL HUMAN BRAIN TISSUE

Embedding fetal brain tissue in celloidin provides 0.2 - 2 mm thick sections. Other sectioning techniques (gelatin-embedded, polyethylene-glykol-embedded, or frozen tissues) are not applicable for fetal brain tissue because these sections do no withstand the staining procedure. Celloidin-embedding results in sections which shrink minimally and provide excellent preservation of vulnerable structures, such as in the septal region. These quality sections seemingly justify the relatively high expenditure of time which is required for the celloidin embedding procedure.

The varied quantity of Nissl-substance within the nuclear greys are highlighted by Darrow-red, which is, to our experience, not possible by means of other Nissl stains in sections of these thickness. Darrow-red can easily be removed from the background of celloidin-embedded material (Powers and Clark, 1960; 1963). A gapless series of fetal brains can be cut frontally, horizontally or sagittally and stained. Hence, the architectonic organization of fetal brains at various developmental stages can be followed throughout developmental changes of the nuclear configurations (Ulfig, in press).

The standard S10/S3 plastination procedure is used for impregnation of fetal, as well as, adult brain sections (vonHagens, 1985). Dehydration of celloidin-embedded fetal brain tissue in 100% acetone removes the celloidin from the sections. Complete impregnation leads to flexible and opaque preparations with high tensile strength. Without much expenditure or experience this method can be established in any laboratory. Moreover, it should be stressed that material which has been stored for several years and which has lost its natural contrast between white and grey matter can be used.

ACKNOWLEDGEMENTS: The author wishes to thank C.H. Medina for his excellent help.

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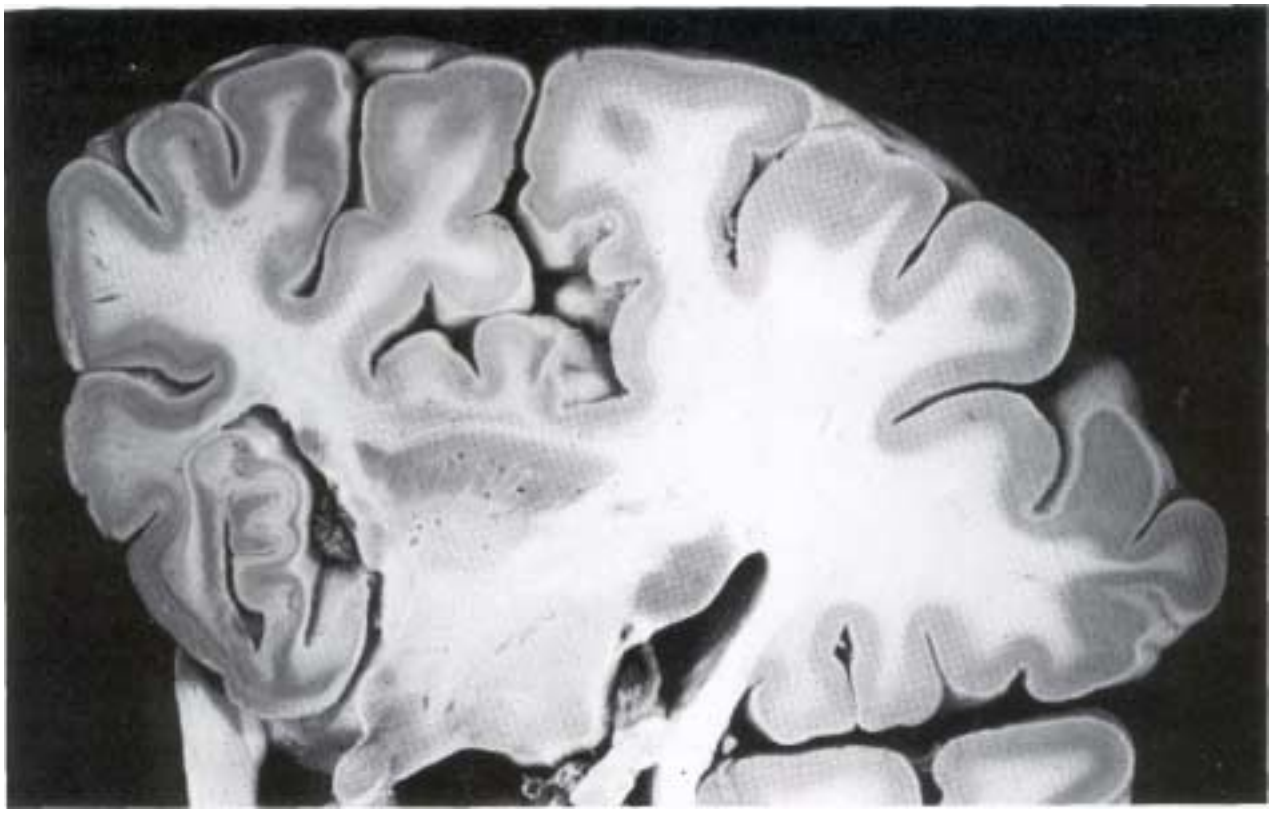

Figure 3. Coronal macrotome section (1 cm) stained with aldehydefuchsin and subsequently plastinated.

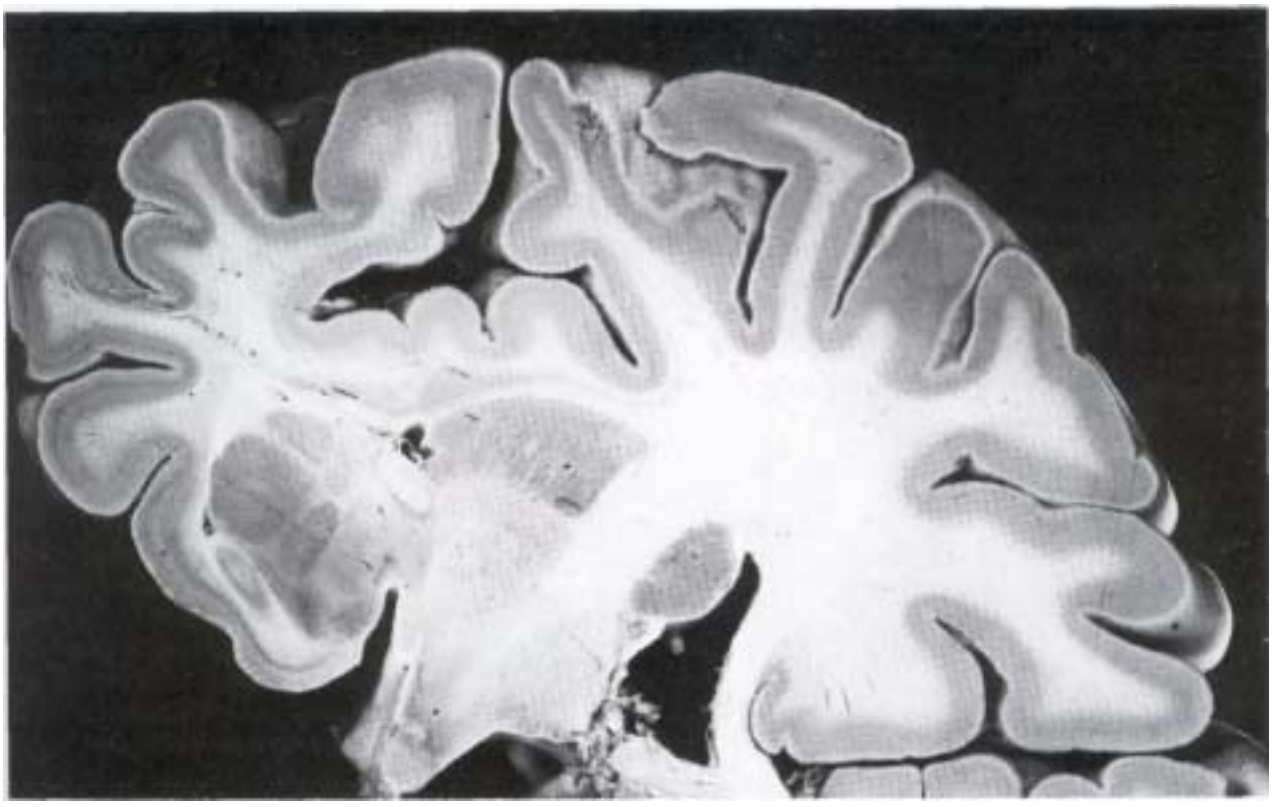

Figure 4. Coronal macrotome section (1 cm) stained with aldehydefuchsin and subsequently plastinated.

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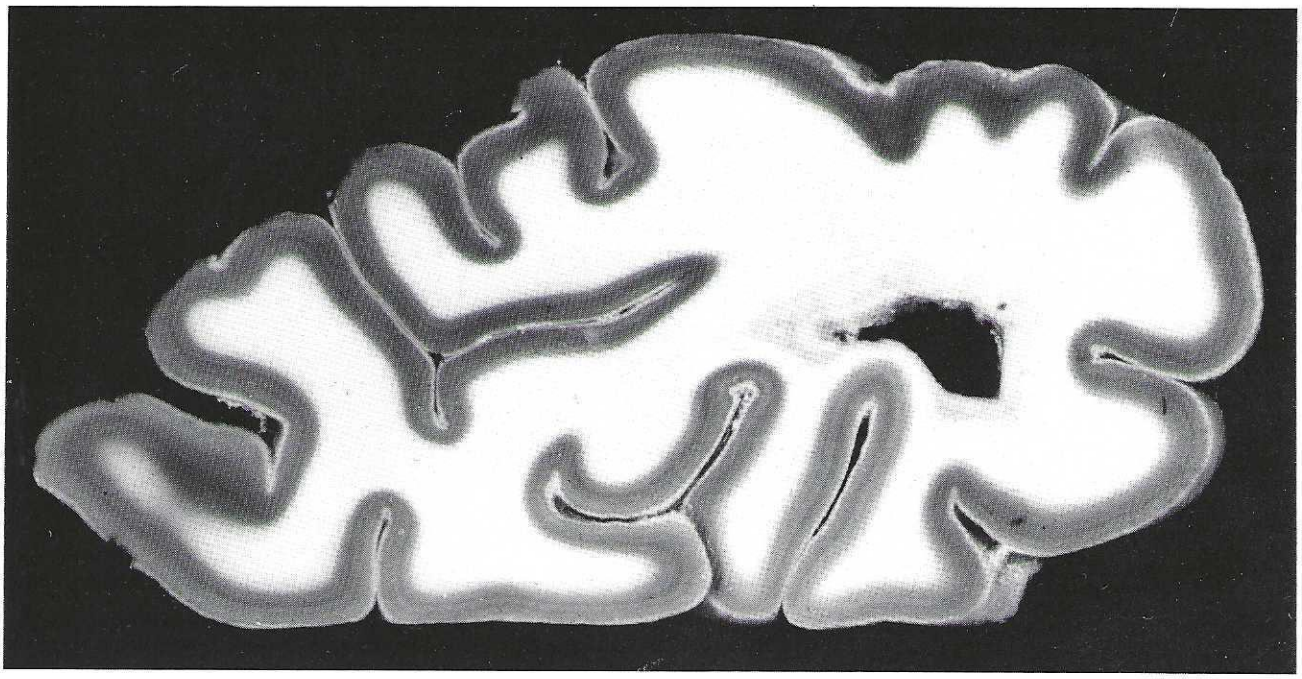

Figure 5. Fetal coronal section (embedded in celloidin, 2 mm) stained with Darrow red and subsequently plastinated.

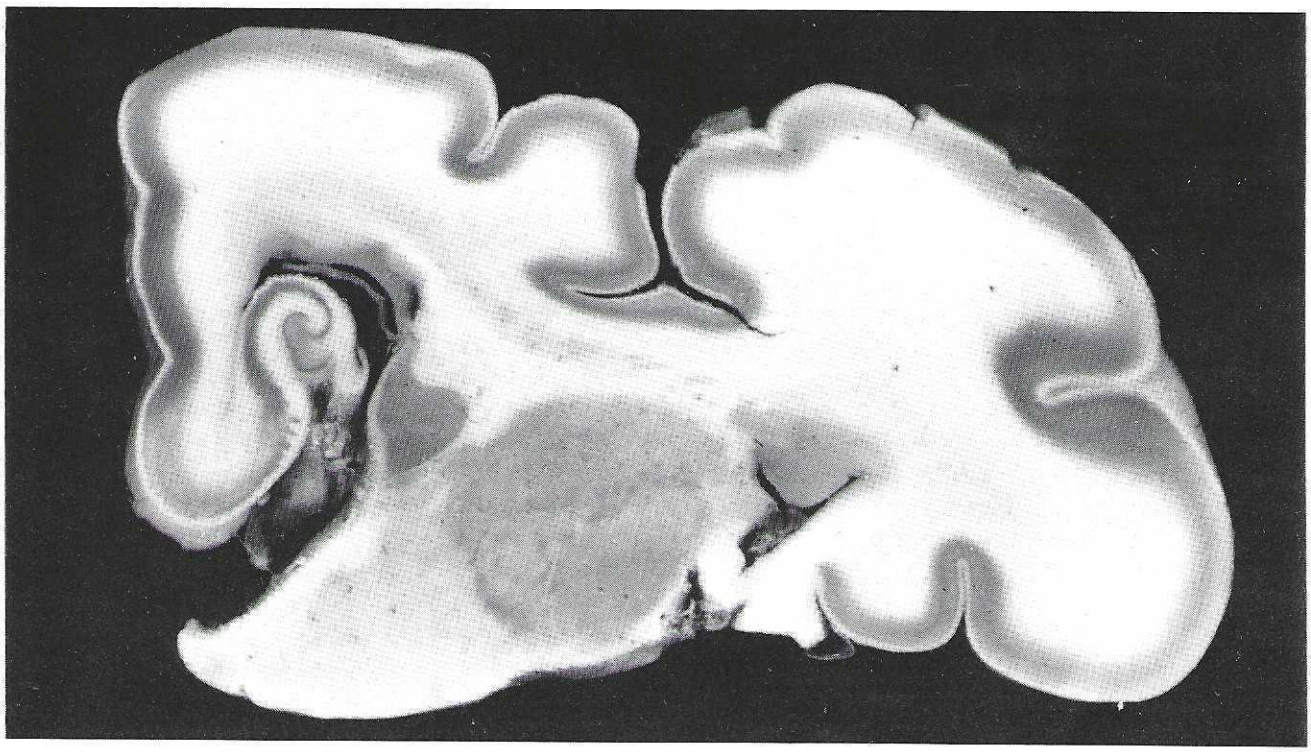

Figure 6. Fetal coronal section (embedded in celloidin, 2 mm) stained with Darrow red and subsequently plastinated.

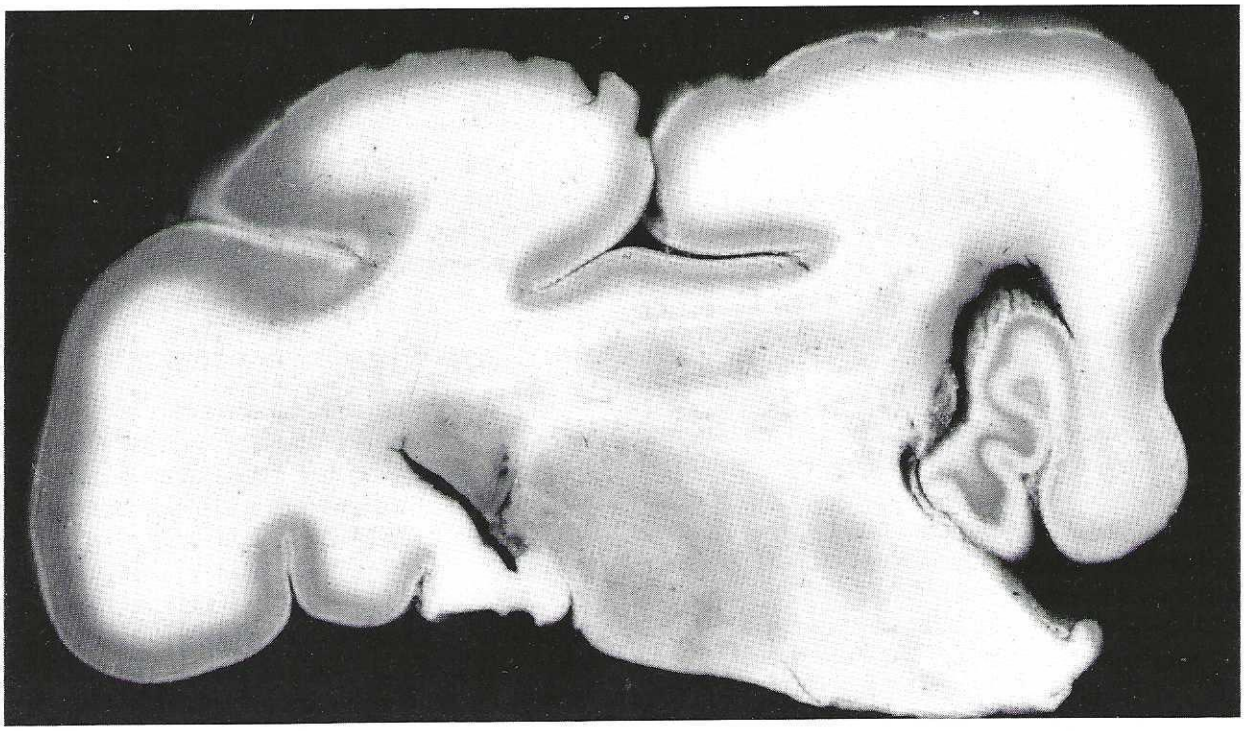

Figure 7. Fetal coronal section (embedded in celloidin, 2 mm) stained with Darrow red and subsequently plastinated.

USE OF PLASTINATED TISSUE IN THE CONSTRUCTION OF HOLOGRAMS

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INTRODUCTION

Holography is a vast new field of wave-interference technology discovered by Denis Gabor and first reported in 1948 (Collier, 1971). Despite its profound significance, holography has been described as a "solution in search of a problem" because it has fulfilled only a small part of its potential. This is due to a number of limitations, among which is that the production of holograms requires considerable knowledge, skill and equipment.

A hologram is a pattern of wave-interference microimages of an object, encoded onto a photographic plate with completely coherent (laser) light. Illumination of this plate with coherent light of the same wavelength produces a three-dimensional image of this object. This is a true three-dimensional image, not an optical simulation. Because of this, holography would seem to offer boundless promise, however its practical applications have been limited by the uncompromising severity of its technical demands. At present there are only about 100 working holographers in the United States and its contemporary use is confined almost exclusively to optical elements (holographic lenses), security-enhancing devices (on credit cards) and the creation of novel forms of art.

HOLOGRAPHIC IMAGES AS MEDICAL TEACHING MEDIA

Because of their striking three-dimensional quality, holograms would make excellent teaching media for the medical subjects such as anatomy and pathology. Notonly would the student benefit from the third dimension, but the hologram, unlike the specimen, can be reproduced, permitting the sharing of a single image among many institutions. Making (and sharing) holograms of human tissue, however, presents a unique set of problems.

For example, in constructing a hologram it is essential that the object remain absolutely motionless during a rather long photographic exposure period. (The only alternative to this is the use of an extremely expensive pulse-laser). Movement through a distance corresponding to % of the wave length of the light being used will cause blurring. Thus, even the slight dimensional change associated with evaporation of tissue water is intolerable.

HOLOGRAPHY OF PLASTINATED TISSUE SPECIMENS

It occurred to the authors that, due to enhanced rigidity, and because natural water has been replaced with polymer, plastinated specimens would make ideal holographic objects. We had established a laboratory for experimenting with the preparation of holograms from tissue and had access to a collection of plastinated specimens. Thus, the construction of holograms of plastinated tissue was undertaken with the intent of using them as an adjunct to pathology instruction in the Mercer medical curriculum. The results have been promising.

Different types of holograms were prepared in an effort to find which would be most useful. Given our present technical capability and teaching requirements, the image-plane hologram has proven most appropriate. This type can be illuminated with a simple, hand-held halogen lamb and easily transported to the classroom.

Also, it became apparent that the hologram is best used as a substitute for specimens that actually depehdon three dimensions for their information content. Holographic images of flat tissue surfaces offered little improvement over photographs but those of specimens with depth were considerably better. Our favorite hologram to date is one of a segment of colon, showing hereditary podyposis.

SPECULATION ABOUT THE FUTURE OF HOLOGRAPHY IN HEALTH SCIENCE EDUCATION

The use of holography in health science education has been extremely limited (Lungershausen, 1983). Most of the reason for this has to do with technical limitations and cost. If holography is to be useful as a medium for medical-school instructing, not only must we be able to create high-quality master images but accurate hologram copies must be mass-produced. This would reduce the cost sharply and would even allow their incorporation into textbooks.

At present, two methods hold most promise for doing this: 1) The embossed hologram (heat-shaped onto plastic) and 2) photopolymer hologram (contact-printed onto photosensitive plastic). The former process has not, as yet been able to yield holograms of suitable quality (this is the type used on credit cards). The latter is rapidly approaching a degree of sophistication that should prove useful. The senior author has recently begun negotiations to be able to experiment with photopolymer material in the preparation of holograms from plastinated tissue specimens.

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obstetrics was a principal duty of physicians at the time the models were made. The artistic detail, to achieve a life-like appearance, was noted in a female model, with beautiful blonde hair and a string of pearls around her neck, as she reclined on a tufted and fringed satin pad. At the plastination facility in Vienna, the variations in equipment used and the types of specimens needed for their curriculum were viewed. Herr Stradal is an accomplished plastinator and had several illuminating comments and tips to pass on to us.

We departed from Vienna Wednesday evening after having had time for a look around that old capital of Austro-Hungarian Empire and arrived in Rome mid-morning Thursday. As most Europeans take a holiday during August, the travel facilities were operating at capacity and most cities were devoid of permanent residents except those operating tourist-related enterprises. Nourished and rested, Dr. Filadora, with great flair and graciousness, conducted our group to the University for a meeting with the department head and for a look at what is being accomplished in their plastination laboratory. We were interested to note that the Italians forego deep freezers and accomplish plastination at room temperature. The resulting specimen is adequate and they pay particular attention to displaying the completed specimens against contrasting colors and textures for maximum effect. Their efforts in electronically enhanced medical education were demonstrated by the staff. The remainder of the time in Italy was at the individual's leisure before returning to either Heidelberg for the second workshop or to a point of embarkation for returning home.

In conversation with the other tour participants, it was agreed that the tour gave us insights into different methods and equipment options, as well as, new uses for the specimens that we are creating in our plastination labs at home. It was reassuring to see that what we are doing deserves the efforts and funds that we expend to enhance education and research at our facilities.

Our thanks to Herr Hillebrands for organizing this illuminating journey.

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POST CONGRESS WORKSHOP: Sheet Plastination.

This workshop started on Monday after the post conference tour. During the week of the post conference tour, a basic workshop was conducted on the S10 technique. Several of the participants from the basic workshop enrolled for the second workshop as well. The first day of instruction compared: the durability, ease of preparation, optical properties, equipment considerations, and preparation time of the E12, PEM, S10 and P35 techniques. (See chart that follows). We were surprised to learn that only a few laboratories throughout the world utilize sheet techniques, since sheets have such broad, interdisciplinary applications. The samples of sheet plastination that we were given during the congress, have met with great interest not only in my immediate area of anatomy but also among the microanatomy and pathology faculty at our school. Since it appears that we will all may be moving toward some variation of problem-based learning, sheet specimens will have many applications.

The 25 participants had many opportunities for "hands-on" instruction during the week: cutting and scraping of the frozen specimens, packing for dehydration, assembling of flat chambers, and marveling over the finished products. I doubt that I will ever forget being with the entire group on the roof of the anatomy institute: two stories above the street, specimens bubbling in methylene chloride, Gunther in his bare feet (so that he won't ruin his new shoes), everyone trying to avoid the fumes, and amateur photographers backing away from the container so that they could get that one perfect picture - "and they were backing toward the edge of the roof!!!" It was not a scene for the faint-of-heart! Imagine 12-15 people attempting to fill flat chambers with polymer, the result: spreading far more over the table tops and floor than ever reached the interior of the chambers. This brought memories of an ill advised taffy pull for a pack of cub scouts in which I had participated some 40 years ago. In short, the workshop instructors and assistants deserve great kudos for their efforts in working with that "inept gaggle of good intentioned students".

I strongly urge you to review the article "The current potential of plastination" by von Hagens et al. in the 1987 Anatomy and Embryology 175:411 - 421. It has many beautiful color photographs and is more thorough in its explanation of techniques than the editor will allow me to write. Detailed instructions for sheet plastination using the various polymers is soon to be published by von Hagens. - - - **By: Tim Barnes**

Among the notes given to us during the workshop, the following chart aids in selection of the most appropriate process for plastinated slices:

Slice Process Selection Chart

Wanted Properties	Polymer Techniques	Prep for Curing	Mode of Curing	Thickness of Slices
Elastic, opaque	S-10	draining	gas	solid organs 5mm or 1.5cm body slices
Firm, opaque (brains)	P35	flat chamber	light heat	4 - 8 mm
Firm opaque (other)	PEM	draining	room temp or heat	1.5 cm
Firm, transparent	E12	sandwich or flat chamber	heat	2.5-5 cm