

# BRISBANE INTERNATIONAL MEETING

## ABSTRACTS / POSTER PRESENTATIONS

### PLASTINATION - A NEW TECHNIQUE FOR THE PRESERVATION OF VISCERA & TISSUE FOR TEACHING AND RESEARCH PURPOSES

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Plastination is a process for impregnating a biological specimen with a curable polymer, using the difference in vapor pressure between the infiltrating polymer and a volatile intermediate solvent, to propel the impregnation.

#### Objectives

1. To do long term, preservation of bodies, and body tissues and organs for teaching, demonstration and research purposes without repetitive use of fresh tissue or sacrificing live animals.
2. To minimize the shortage of cadavers/organs/human tissues (in the presence of and increasing the number of teaching institutes) for teaching and research purposes.
3. To develop this speciality area of professional activity in order to encourage other institutions to adopt plastination preservation methods and to invite individuals to learn and practice plastination as a career in science.
4. To demonstrate that it is safer, more economical and better method of preservation of body/organs/tissues for medical education. With the development of the technique, the repetitive use of gloves in handling the specimens can be avoided. Museum glass jars are not required. Funds required for buying formaldehyde and other preservative chemicals can be reduced. It also ensures the safety of the workers.
5. To build the library of clean and odorless teaching and research specimens.
6. To use plastinated sections as a guide for computerised axial tomography (CAT) scans, MRI and ultrasound techniques and will provide a new three dimensional perspective of normal anatomy and its variations.

In the fall of 1992 I was asked to initiate the development of a Plastination Laboratory to prepare anatomic teaching specimens for undergraduate and postgraduate medical students. After three and one half years of effort, funds have been allocated to start the Plastination Laboratory at all India Institute of Medical Sciences, New Delhi, India.

### A SHARK HANDSAW BLADE SIGNIFICANTLY ENHANCES THE QUALITY OF CUTS MADE FOR PLASTINATED SPECIMENS

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The popularity for use of band saws in preparing plastinated specimens has increased significantly over the past few years. This use has brought forth many modifications that have facilitated the production of even higher quality specimens.

In this study a Shark Band\* blade was evaluated and found to be superior over more commonly used (10 to 12 teeth per inch) blades. Trials were performed upon frozen tissues in preparation for S10 plastination, E12 sheet plastination, as well as for slicing cured S10 plastinated whole brains. The advantage of a Shark Band blade is that by a reduction to only 3 teeth per inch the creation of, and subsequently contamination of specimens by, sawdust is significantly reduced. The minimal set and thickness (14 gauge/.014 inches) of the teeth and blade similarly promotes a smooth and very fine cut. The effects of this blade can be even further enhanced through an increase in the blades cutting speed.

Unfortunately the same design that facilitates the quality of the cut also greatly reduces the blade life. When cutting dense bone or enamel the blade teeth are quickly dulled and taken out of set. This necessitates the replacement of the band after only a few cuts.

### PREPARATION AND PLASTINATION OF THE CEREBRAL DURA MATER WITH THE SKULL BASE

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The aim of this work was to provide a plastinated specimen of the cerebral dura mater and its main partitions (falx cerebri, tentorium and falx cerebelli). The skull was sawed one centimeter above the junction of its base and its vault, the latter being broken up and then removed in order to expose the convexity of the dura mater without damaging it. The eye balls and the accessory organs of the eye were also removed and the sheath of the optic nerve was carefully dissected. One side of the dura mater was opened, the brain was broken up and removed and the specimen was dehydrated and impregnated according to the standard S10 technique. For the gas curing the whole dural cavity was filled with absorbent paper in order to restore the normal shape of the cerebral dura mater. Finally the opening was enlarged to show the falx cerebri with its inner free margin and the incisura superior sagittal sinus to be seen. This plastinated specimen will help students to understand the topography of the dura mater and the transtentorial herniations.

### E 12 TECHNIQUE: AN AID TO STUDY SINUSES OF PSITTACINE BIRDS

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Infraorbital sinus infections are commonly encountered in clinical practice in companion birds. They form the bulk of upper respiratory tract infections (URI) and require medical and/or surgical management for the avian patient. The infraorbital sinus has extensive connections to structures of the head and its dorsal drainage into the nasal cavity makes it difficult to treat these infections. A more complete understanding of the anatomy than is described in the fowl would enhance the clinical diagnosis and management of URI in companion avian species, particularly psittacine birds. For comparison and as an aid in determining the location and extent of the sinuses of the parrot and

macaw, computed tomography (CT) images of cadaver heads were completed at 2mm scans. The scan lines were marked of the skin to allow sawing in a similar plane. Subsequently, the sinuses of the imaged heads were injected via a 20 gauge needle inserted into the infraorbital sinus with a mixture of colored epoxy (E20 and El mixture) in a ration of 2:1. The epoxy was allowed to harden over night and then the specimens were frozen (-25C) for sawing. A shark band blade was used to saw 2mm thick sections which corresponded to the CT scans. The sections were placed on grids (covered with screen) and submerged in cold acetone for saw dust removal and dehydration (freeze substitution). Two changes of 100% acetone were used. After 24 hours at room temperature, the filled flat chambers were placed in a 45C oven overnight. The next day the chambers were removed from the oven, allowed to cool, and dismantled. Individual slices were sawed apart. The slices were photographed and compared to the CT images and the sinuses delimited.

### **ENDOANAL MAGNETIC RESONANCE IMAGING OF THE ANAL SPHINCTER COMPLEX: CORRELATION WITH SHEET PLASTINATED SLICES AND HISTOLOGY**

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Introduction: The purpose of this presentation is to correlate in vivo endoanal MRI findings of the anal sphincter to the cross-sectional anatomy and histology.

Materials and Methods: Fourteen patients with rectal tumors were examined with a ridged endoanal MR coil before undergoing abdomino-perineal resection. In addition, twelve cadavers were used to obtain sheet plastinated slices. The imaging findings were correlated with histology and anatomy of the twelve cadavers.

Results: The data of the eight patients, eleven rectal preparations and ten cadavers, could be compared. In these cases, there was an excellent correlation between endoanal MRI and the sheet plastinated slices and histology. With endoanal MRI, all muscle layers of the anal canal wall, comprised of the sphincter ani internus, conjoint longitudinal muscle, the sphincter ani externus and the puborectalis muscle were clearly visible. Also the levator ani muscle and ligamentous attachments were well depicted, the perianal anatomical spaces, containing multiple septae, were clearly visible.

Discussion: Endoanal MRI is excellent in depicting the anal sphincter complex and the findings show a good correlation with the sheet plastinated slices and histology.

### **PLASTINATED CANINE GASTROINTESTINAL TRACTS USED TO FACILITATE TEACHING OF ENDOSCOPIC TECHNIQUE AND ANATOMY**

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Plastinated specimens, when prepared with a design for endoscopic use, can serve as a practical model for teaching. Intact alimentary canals were excised from fresh canine cadav-

ers. Cannulas in excess of the intended endoscope size, were placed in restrictive openings (pyloric/cardiac sphincters and ileocolic orifice). These cannulas allow ingesta to be removed and maintain adequate diameters for scoping. After flushing out the gastrointestinal contents, specimens were formaldehyde fixed overnight in dilated anatomical conformation. Prior to S10/S3 impregnation, fixative was flushed from the specimens and they were dehydrated in acetone. After impregnation, a slow cure (elongation of S3 molecules at room temperature) was initiated for approximately 1 week with the GI tracts maintained in a dilated conformation by a positive pressure air flow. When polymer seepage was minimal, small quantities of S6 (second curing agent for crosslinking of polymer molecules) were volatilized into the air flow providing a final cure of the specimen. Vapors from the S6 were further contained around the specimen by enclosing them in plastic bags. The completed specimens retain their dilated anatomical conformation, and may be used to teach both endoscopic technique and gastrointestinal anatomy.

### **COMPARISON OF FOUR CLASSES OF ANATOMICAL LAYERS OF PLASTINATION SECTIONS WITH FOUR ANATOMICAL LAYERS OF COMPUTER IMAGES**

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Several demonstrations of each class of body tissue layers have been photographed from E12, S10 and P35 plastinated human sections and computer images (MRI and Ultrasonography). These photographs are labelled to reflect four classes of tissue layers which include somatic, extra visceral (visceral), intravisceral luminal, and intravisceral nonluminal classes of layers. The four classes of layers are based upon embryonic layers and location.

The somatic layers correspond with layers derived from somatopleure in the developing embryo. The intravisceral layers include the internal organs positioned in deliberate sequence and derived from splanchnopleure of the embryo. Intravisceral luminal layers are organs with a prominent lumen or canal which courses through the organ. Intra visceral nonluminal layers include organs without a prominent lumen.

### **OUR OWN PLASTINATION LABORATORY**

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With the acute problem of getting new cadavers in most countries, there is the need to plastinate available cadavers. However, the cost in setting up such a laboratory may not be within reach of many departments.

Our lab was set up in 1991 using a room 1400 sq. feet in size, adjoining the campus workshop. The laboratory is divided into various areas without partitions. The plastination tank, curing chamber and separator were made locally at a cost of US\$1,700. Three items of equipment were ordered from Germany, namely vacuumometer, vacuum pump and acetometer. The total cost was about US\$2,000.

Personnel from two neighboring universities have visited our laboratory to enable them to set up their own laboratories. The department's next project is to undertake sheet plastination. In the meantime, the laboratory is the pride of the medical school.

## **PREPARATION OF THE CORONARY ARTERIES FOR PLASTINATION**

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The technique of preservation of anatomical specimens using the method of Dr. Gunther vonHagens provides excellent demonstrations for teaching and further education. Of particular interest in the heart is its vascular supply, which we can display in plastinated specimens injected with red Biodur E20.

The heart should be removed from the thoracic cavity taking care to divide the vessels some distance from the heart. Then the heart is rinsed with cold tap water and all vessels are blocked with cork stoppers. The stoppers for the aorta and aortic valve and the pulmonary valve are in as almost completely closed position.

Injection of the coronary arteries with 49.5 gram Biodur E20 red requires only slight pressure to fill the coronary vessels. For the dilatation-fixation 15L of 20% formalin solution are infused into the ventricle from a height of 1.5 meter. A week of immersion-fixation in 5% formalin solution at a temperature of +5C follows.

The sub endocardial fatty tissue and all fascia are carefully dissected away with scalpel, scissors and forceps. Subsequently the specimen is bleached in a 2% hydrogen peroxide solution with 5% formalin added to achieve the desired color. After washing for about 1 hour the auricles are tamponaded with polyethylene-foils and the specimens are frozen overnight in a plastic bag. Dehydration lasts for 3 weeks at -25C in the deep-freezer. During this time the acetone is changed twice.

Forced impregnation with Biodur S10 plus S3 hardener takes place for 3 weeks at 25C in the deep-freezer. After removal from the vacuum kettle and draining overnight in the freezer, the specimen drains for further day at room temperature. Then the specimen is placed into a plastic container for gas hardening with Biodur S6. This takes 2 weeks. Finally, a comprehensible injection specimen of the heart is available in a most life-like condition for instruction and research in medicine.

## **COMPUTERISED LABELLING: A NEW METHOD FOR S10 PLASTINATED SPECIMENS**

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The correct labelling of anatomical specimens not only allows the specimen to be readily identified, but also makes salient information regarding the deceased easily accessible. Well prosected specimens, with emphasis on the relevant morphological structures, are aesthetically pleasing. In teaching and research it is essential that the structure being studied is clearly visible. In order to avoid large surface labelling, and appropriate method which wouldn't obscure important parts of the specimen was sought.

Initially, several factors were of concern; cost, accessibility (within the specimen), resistance to chemicals (in particular formaldehyde), size and strength. The Trovan Transponder System meets all these requirements. The transponder chip is approximately 10X3mm in diameter and can be easily inserted into an appropriate area of the specimen. A suitable applicator can be purchased or made at minimal cost. This ensures deep penetration into the hardened silicone matrix (S10), as well as the security of the chip. In cases of minimal skin coverage, a hole of approximately 3.5mm diameter is drilled into bone. The chip is placed within the hole which is then sealed using a suitable adhesive. The Trovan Minireader unit is battery

powered. It has digital readout capabilities as well as a serial port for direct input to a computer. At the University of Melbourne data on the chip corresponds to details of the specimen and cadaver from which the tissue was taken, and therefore includes information such as age and cause of death. Of the forty S10 plastinated specimens labelled in this fashion, only a few of the original injection sites are visible. All of the specimens have their own electronic serial number which can be sensed by the minireader at varying ranges. Significantly, the high presentability of the plastinated specimens remains unobscured by this labelling technique.

## **AN IDEAL CURING CABINET FOR S10 PLASTINATION**

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Curing of specimens is an integral part of the S10 process. On completion of this final stage the specimens are touch dry and resilient for everyday use. The fast curing variation of the technique requires a chamber of sufficient size to house the specimens. It needs to be relatively air tight, easily accessible, adjustable and conducive to the curing environment. This presentation details the development of a custom made curing cabinet at the University of Melbourne which has significantly enhanced our ability to produce specimens of a consistently high quality.

Because of the high turnover in prosected "wet" specimens the Departmental anatomical technologies laboratory is involved in the production of as many S10 plastinated specimens as possible. This necessitated the design of a curing cabinet capable of accommodating a large number and variety of specimen sizes. Because specimens at the curing stage need to be constantly monitored the chamber also needs to be transparent. Perspex satisfies all of these requirements, it is relatively inexpensive and is easy to work with. It may be glued, moulded, bent or drilled making it very suitable to our needs. A variety of additional requirements can be accommodated, such as variation in size of the cabinet to suit fume cupboard, or the connection of an extraction system. By assembling the cabinet with a removable front panel the specimens can be readily manicured and the concentration of volatilized S6 varied. Further modifications may be made, such as adapting the shelving to suit the specimens, by gluing several small pieces of perspex to the inside walls of the unit.

## **PLASTINATION IN CLINICAL MEDICINE**

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The changing trends in medical education warrants the availability of clinical material for training of doctors at venues beyond the confines of the medical school. The present training of medical doctors uses patients in the clinical context where patients often endure physical indignities and stress when being subjected to examination by students, registrars and even consultants when gaining experience in various clinical procedures. Furthermore, with newer clinical procedures evolving recently, and with tremendous advances in minimally invasive surgery the

need has arisen for patients not to be used repeatedly in the training of medical students and registrars in clinical procedures. Furthermore, ethical issues which were previously largely ignored by clinicians in third world settings are now enforced in the transforming South Africa. For these reasons, extended applications of plastinations are now more needed than ever.

In the Faculty of Medicine at the University of Natal, Plastination Laboratory has been specifically focused to provide this need. The combination of plastinated specimens in the skills laboratory for the training of undergraduate and postgraduate studented is been fully exploited with this potential in mind.

Plastinated specimens are used at undergraduate levels to teach medical students not only basic human anatomy and embryology as well as neuroanatomy but also clinical examination techniques such as nasal, aural, oral, rectal and vaginal examinations obstetrics as well as gynecological cases. In the teaching of dermatology, plastinated biopsies of skin segments could be used for demonstration rather than using slides or video tapes. This obviates the use of patients in the training of students.

On a different level, medical and surgical procedures such as inserting intercostal drains, pericardiocentesis, cutdowns, biopsies of various organs and bone marrow aspirations could be attempted and perfected by medical personnel in training. While initially, normal procedures could be performed using plastinated specimens, the technique could be expanded to include pathological specimens such as those used in the study of surgical pathology. In this regard, plastinated specimens of benign prostatic hypertrophy, malignancies of the breast and congenital deformities could be prepared.

On the medical front, procedures such as sigmoidoscopy, colonoscopy and gastroscopy could be accomplished on plastinated torsos. Complicated procedures such as endoscopic retrograde pancreatico-cholangiogram could be performed in a plastinated specimen. On the highly specialized side, angioplasty of the coronary arteries could be practised of plastinated specimens. In reconstructive surgery, injected arteries in plastinated specimens could be used by surgeons to devise myocutaneous flaps in complicated reconstruction of traumatized cases which are prevalent in South Africa where violence is endemic in major cities and townships.

With the emergence of minimally invasive surgery, expertly prepared plastinated specimens could be used in laparoscopic surgery to acquaint the operator with basic anatomy as visualized through the laparoscope, albeit the aspect of hemorrhage would be nonexistent in these "test patients", but the techniques of laparoscopic surgery could be mastered without any inconvenience and morbidity to patients.

In conclusion the use of silicone impregnated plastinated specimens has great potential and could be of tremendous value in the training of medical and Paramedical personnel in the 21st Century.

## **A NEW BUILDING IS PROPOSED TO RESOLVE FIRE CODE INFRACTIONS AT ISU**

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The production of plastinated and skeletal teaching specimens for veterinary anatomy requires large volumes of acetone to accommodate the parts of the carcasses of large animals. However, the uniform building code limits the use or dispensing of flammable liquids to 10 gal per 100 square feet of floor space. Using quantities in excess of this limit requires space classified as H-2 occupancy. In this paper, the requirements of an H-2 occupant room are explained, and a strategy of obtaining such a facility is explored.

According to the Uniform Building Code (UBC) of the USA, and H-2 occupancy room needs to have the following features :

1. Electric wiring, class 1 division II NEC (National Electric Code)
2. Ventilation to maintain air/fuel mixture below OSHA TLV levels
3. Back up power
4. Two exits, one of which leads to the outside
5. Two hour fire separation with 1 1/2 hour fire door
6. Sprinkler system
7. Spill containment
8. Blow out panel

Electric wiring: All electric wiring in as H-2 occupancy room must conform with class 1 division II NEC. This means that motors of standard freezers as well as motors of "explosion proof" freezers are not allowed in this room. The so-called explosion proof freezers provide an explosion proof interior while still allowing air and fumes to enter the motor from the exterior. A solution to this problem is to relocate the motor into a adjacent room and connect the cooling coils to the freezer through the wall. The expense of an explosion proof freezer is unnecessary. Vacuum pumps need to be relocated as well. Obviously, it would be a code violation to operate any non explosion proof power equipment such as band saws in this area. Ventilation: The ventilation system will be of the exhaust type exchanging the air at a 1:1 ratio 6 times per hour. In addition, the plans call for local exhaust systems at the perimeters of the freezers and degreasing tank. We will evaluate whether an exhaust duct at the back of the freezers is sufficient to remove fumes form the freezer box.

Back up power: Back up power, provided by an emergency generator, will ensure that even during power failures the air exchange will continue and the acetone in the freezers will remain cold.

Second exit: The Uniform Building Code requires a second exit if the distance of the remotest corner of the room to the door exceeds a specifies distance. Both exits need to be equipped with panic hardware.

Fire separation: The Uniform Building Code requires that the walls be able to withstand 2 hours of fire and the doors 1 1/2 hours to ensure that a fire would not spread to adjacent rooms before the building has been evacuated.

Sprinkler System: The sprinkler system for the planned plastination room will be able to deliver 4000 gal in 20 min. Some of the sprinkler heads will be pointed directly at the freezers and the degreasing tank.

Spill containment: The spill containment system must accommodate the volume of liquids that are present in the room (200 gal of acetone) as well as the volume of water that is discharged by the sprinkler system in the event of a fire. That means 4000 gal of water in 20 min. plus approximately 200 gal of acetone which must be initially contained and drained to a secondary containment. The initial containment of 4200 gal could be accomplished by providing a 4" liquid tight curb along the perimeter for the room. This will necessitate a ramped sill at the entrance of the room. A potential spill must then be drained to a "safe location". The preliminary design of the facility will place the floor drains for the secondary spill containment in front of the emergency exit eliminating the need of a second ramp in the room. A safe location for the spilled liquids could be an underground tank. However, this would require a double wall construction with leak detection equipment making this a very expensive solution. The alternative is an open pit, which not only accommodates the design flow of the sprinkler water, but also the volume of the largest container in this room, plus the 24 hour rainfall as recorded from the worst storm of the last 25 years. This would be accomplished by constructing a pond with a plastic liner using a feeder line of 12" diameter and providing a drainage valve to empty the pond after regular rainfalls. A fence surrounding the pond has been suggested to limit access.

Blow out panel: One outside wall or the roof of an H-2 occupancy room must be designed to vent the intial blow of an

explosion while the other walls must withstand the remaining pressure. This is required to limit the damage to the building in case of an explosion. The new building would be constructed out of precast concrete panels and the roof would be equipped with a blowout panel similar to a skylight.

Obviously, it cannot be cheap to construct such a facility, the preliminary estimate is based of \$200.00 per square foot or 2 million dollars for the facility. Several Departments at ISU are under pressure to obtain an H occupancy room for their operation. Sharing a facility is perceived much more cost efficient than attempting to achieve compliance with the code at each location. The Department of Veterinary Anatomy needs a facility to handle large quantities of acetone. The Veterinary Diagnostic laboratory needs a facility to handle ethanol, methanol and agricultural herbicides and pesticides. The laboratory for greyhound racing chemistry needs room for ethanol. And finally the Department of Environmental Health and Safety needs a facility to handle chemical waste created throughout the university. E, H&S currently is using a facility outside the contiguous campus grounds which necessitates a tremendous amount of paperwork with the Iowa Department of Transportation to permit the tracking of chemical wastes from the various campus locations to the existing facility. It would be in their interest if the money would be spent in an on-campus facility, rather than upgrading their existing-off campus steel building.

It is our hope that this alliance of interest will help find funding for a new facility.

1. Paul Rietz, Rietz Engineering Consultants, Ames, Iowa: College of Veterinary Medicine Rooms 1304, 1304 A & 1304B Fire Safety Improvements Feasibility Study, September 1994, Page 1.
2. Gerhard Penz, University of Toronto, Banting Institute, Department of Pathology, Telephone conversation.

#### **CORROSION CASTS OF SPLENIC ARTERY AND VEIN AS A MODEL FOR THE STUDY OF VASCULAR ANATOMY OF SPLEEN**

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Morphological segmentation of the human spleen was described on the basis of presence of fibrous septae (Kyber 1870). Vascular segmentations of the spleen on the basis of the splenic vein was reported by Dreyer et al 1952 in their studies on diagnostic splenic venography. Vascular segments to the spleen were also demonstrated by corrosion casts of the splenic artery and its branches. (Gupto et al 1976). By radio opaque techniques, the human splenic segment was found to have an independent hilar branch of splenic artery and a tributary of the splenic vein (Braithwaite et al 1956). This technique cannot be used routinely for the study of vascular anatomy to spleen. The present study was therefore conducted on human spleens to demonstrate the various vascular segments and their interconnections on the basis of the distribution of both the splenic artery and vein using corrosion casts.

Forty fresh adult human spleens were obtained from AIIMS mortuary within 24 hours of death. The splenic artery and vein were dissected out and cannulae were passed into artery and vein. The spleen was washed with tap water to remove blood. With the help of syringe, red solution of cellulose acetate butyrate (CAB) dissolved in acetone was injected in the artery and blue CAB was injected in the splenic vein. Injected specimens were transferred to 10% formal saline for 24 hours for butyrate to harden. The specimen were then transferred to a jar of concentrated hydrochloric acid until corrosion of soft tissues was complete. The resulting cast was then cleaned and examined for the splenic artery and its branches and the splenic vein and its tributaries.

The vascular segmentation of spleen was determined on the basis of the distribution of both splenic artery and vein. Variations in the vascular segments of the spleen and their interconnections were seen when both artery and vein were taken into consideration.

#### **SHEET PLASTINATION OF THE VASCULARITY OF THE LUNATE BONE**

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Introduction: The aetiology of lunate necrosis (Kienbocks disease) remains unclear. Pressure measurements show that the preferred working position of the wrist-dorsiflexion-leads to significantly increased intraosseous pressure in the lunate. The phenomenon can be explained by an impairment of the venous drainage of the lunate in dorsiflexion.

Material and Method: 12 fresh cadaver forearms were injected on the arterial and venous side by EI2 technique followed by sheet plastination. 6 arms were fixed in neutral position and 6 were fixed in dorsiflexed position.

Results: the lunates showed an abundant arterial blood supply in wrist neutral position, the arterial supply of the lunates was accentuated on the palmar aspect. In dorsiflexion the arterial supply of the lunate was accentuated on the dorsal aspect. The stretched palmar wrist capsule compressed the palmar vessels, but there was no difference of the rich intraosseous blood supply in the lunates between neutral and dorsiflexed position. The venous drainage of lunates was maintained by dorsal and palmar plexas. In wrist dorsiflexion the venous drainage of the lunates was impaired by the dorsal capsule wedged between the distal radius and the capitate. The palmar capsule was stretched in dorsiflexion and compressed the palmar vessels. Intraosseous veins were congested.

Discussion: Venous congestion in dorsiflexion of the wrist was demonstrated by sheet plastination. Pressure characteristics and sheet plastination showed concordant results. We conclude that there is increased risk for lunates to be damaged by venous congestion when the wrist is dorsiflexed.

#### **LOW COST PLASTINATION TECHNIQUES FOR DEVELOPING COUNTRIES**

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The use of plastination is increasing, due to its many advantages. Developing and the underdeveloped countries are yet to catch up because of the high cost.

Some modifications have been made to the procedure to reduce the cost and save time. The specimens so plastinated are better for museum display, demonstrations and discussion, compared to the specimens in jars.

Fresh specimens were selected and cleaned in running water; preserved in 10% formalin for 48 hours; dehydrated using 70-80-90-Absolute alcohol changes spending 3-4 days in each; cleared with Benzene for 3-4 days; transferred to the resin; after 48 hours, exposed to hardener for 2 hours and then air dried. The dissection was done to display the part of interest and then mounted on wood/acrylic base using wires. Colors were added to enhance contrast.

Advantages are a minimal use of materials and the procedure is done at room temperature and pressure. This results in greatly reduced cost.

## **PREPARING AND USING S10 AND P35 CLEARED RABBIT FETUSES TO DETERMINE SOME OSSIFICATION CENTERS**

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**Introduction:** The determination of some ossification centers of rabbit fetus is part of a study performed by the author using various animals. It is of great significance for academic and medico-legal studies. Similar work on the goat was done in 1992, by the use of clearing, Alizarin staining, and normal radiological procedures. The present study on the rabbit is a trial of plastination in our research.

**Material and Methods:** Pregnant rabbits were killed and their fetuses were cleared and stained with Alizarin red. Some specimens were plastinated, using either S40 or P35 (S10 P35).

**Results:** The presence of ossification centers in different parts of the skeleton in relation to different fetal age was recorded.

**Conclusion:** The results obtained are of great interest and importance as a means of aging the embryos through determining the state of the ossification centers.

## **THE USE OF CORROSION CAST TECHNIQUE AS MODEL TO STUDY THE PATTERNS OF SEGMENTAL ANATOMY OF LIVER**

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**Introduction:** Advances in hepatic surgery and the trend to conservative hepatic surgery prompted the study the segmental anatomy of the liver in greater detail. Compared with gross dissection, histological and radiological techniques, the corrosion cast preparation appears to give a better display of intrahepatic anatomical relationship. The corrosion cast may be of immense help in localizing and understanding various pathological processes and in planning the consequent surgical approach. Segmental anatomy of the liver has been studied by Glisson (1954), Goldsmith and Woodburne (1957), Gupta et al (1977) and Ralph (1989). Because of the conflicting accounts in the literature it has been decided to make an exhaustive study of the anatomical segmentation of the liver.

**Aims:** This study aimed to formulate a practical classification of the various segments of the liver particularly on the basis of intrahepatic course of the portal vein, hepatic artery and bile duct by using corrosion cast preparation.

**Material and Methods:** Liver specimens in block (all males) were randomly selected from post mortem examinations. Cadaver showing intraabdominal pathology, previous surgical exploration of the abdomen or abdominal trauma were excluded from the study. All the samples were injected with different colored cellulose acetate butyrate (CAB) granules dissolved in acetone to exhibit the intrahepatic patterns of hepatic artery (red) portal vein (blue) and bile duct ramification (green). Casts were prepared according to the method described by Tompsett (1970).

**Results:** Each cast was studied separately for its segmental pattern. Hepatic segmentation was done on the basis of intrahepatic distribution of the portal vein, hepatic duct and the hepatic artery. The study showed the marked variation in the sizes of the segmentation.

## **LOW COST PRESSURE INJECTOR**

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There are a number of situations when plastinated specimens lose some of their natural contours during processing. This may be due to inadequate fixation, loss of fat during the dehydration and defatting phases or failure of polymer to penetrate intact skin and dense fascial layers. Some of these problems can be rectified by injection of polymer into the specimen before or

after impregnation. To inject viscid polymer requires substantial pressures (up to 10,000 p.s.i.) and the equipment is correspondingly costly.

Sometimes quite sophisticated equipment is relatively cheap if it is mass produced. Our laboratory does not use custom made stainless steel vacuum chambers costing \$2,000 but buys instead similar sized vacuum chambers which are parts of milking machines and thus made in significant numbers. They cost \$500.

We therefore sought a source of mass produced pressure injection equipment capable of producing 10,000 p.s.i. and soon located it in the form of a hand powered grease gun used to inject lubricating grease into bearings.

It has been modified. Instead of a cartridge of grease, a cut down 50 ml disposable syringe filled with polymer can be loaded into the grease chamber. An adaptor screws into the existing grease outlay and has a male Lure taper fitting as the new outlet. It incorporates a screwed collar to retain the needle and prevent it from becoming a projectile.

While it has limited capacity and would be tiresome to use of large volumes needed to be injected it has the outstanding advantage of having cost \$35. There are other forms of greasing equipment some of which is fed from 25 liter drums and which may be powered by compressed air. This equipment may also prove suitable for modification to enable it to inject polymer.

## **THE DESIGN OF LIFTING GEAR FOR A PLASTINATION LABORATORY**

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With increasing suspicion about the harmful effects of formaldehyde one of the advantages of plastinated specimens is the improved safety to staff and students which accrues from not using formalin preserved specimens. However, plastination suffers from a considerable safety handicap in that it usually utilizes acetone (di-methylketone) which is colorless flammable and can form explosive mixtures with air. This property dominates the design of a plastination laboratory with particular emphasis being paid to the elimination of sources of ignition. Spark-proof light fittings, explosion proof ventilation fans and the removal of freezer compressors to an adjacent room are common precautions.

Many plastination laboratories have had to utilize existing buildings and the Brisbane laboratory is no exception. It has a standard single doorway. In order to accommodate the largest possible freezers these were designed to just pass through the doorway without their lids which are fully detachable. In order to accommodate the largest possible freezers were designed to just second design feature was to incorporate extra insulation to ensure that -25 Celsius could be readily maintained during a Queensland summer when ambient temperatures sometimes approach 40 degrees Celsius. In consequence the freezer lids are bulky and heavy and not easily manhandled by a single operator and it was not before a back injury was sustained.

Also the movement of drums of acetone or polymer into or out of the freezer is a task that can only be accomplished if they are almost empty.

There was an obvious solution; some form of lifting gear. However this had to operate safely in the presence of flammable vapor. To meet this requirement it proved necessary to consider a number of factors which included considerations of the nature of acetone, the circumstances in which explosive vapors were used and finally the design feature of the lifting gear and operational regulations for its use.

## **DEVELOPMENT OF AN ANATOMICAL TECHNOLOGIES LABORATORY**

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The University of Melbourne, Australia

This presentation documents the procedures followed in establishing an anatomical technologies laboratory for the preparation of plastinated specimens in the Department of Anatomy and Cell Biology at the University of Melbourne. Funding for the laboratory was obtained through a Faculty of Medicine teaching development and innovation grant, supplemented by the Department. A space was found in the basement to the building, next to the embalming room and consisted of a "tank room" which was formerly used for the storage of cadavers. This was converted into two suitable work areas, one a preparation room and the other a spark-proof area housing the freezer with an isolated vacuum pump and refrigeration unit.

Part of the funds obtained from the Faculty have been used to employ a technical officer who has dedicated responsibility to the facility and its day-to-day operation.

The laboratory has been fully operational for over a year and a large number of plastinated specimens have been prepared during that time. We are in the process of developing 'sheet plastination' and have a project under-way with radiologists from the Royal Melbourne Hospital correlating cross-sectional plastinated specimens with CT-scans and magnetic resonance images.

It is intended to direct most of our work towards the Department's teaching requirements, however we also intend using the laboratory to train technicians in anatomical techniques and encourage honors and higher degree students in the area.

## **PLASTINATION AS A CLINICALLY BASED TEACHING AID AT THE UNIVERSITY OF AUCKLAND**

Peter Cook  
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University of Auckland, New Zealand

As a concerted move toward closer integration of the clinical and pre-clinical aspects of the undergraduate medical curriculum at the University of Auckland, the Department of Anatomy has established a formal link with the Department of Radiology resulting in a structured program of clinically based teaching of gross anatomy to second and third year medical students.

As sophisticated diagnostic techniques and methods of treatment have become common place, our teaching program has been tailored to accommodate a greater degree of case based learning within the undergraduate course.

Dissecting room demonstration is provided by Radiology, Pathology, Ophthalmology and Surgical registrars with a number of clinical procedures, pathological observations and diagnostic methods employed during the routine dissection of the cadaver.

Through use of a number of plastination techniques, the learning process is enhanced and aided on a number of levels. The E12 epoxy method for producing MRI and CT based serial sectioned cadaver specimens has allowed an accurate and highly detailed orientation of the planes of the body and provides the student with a clearer understanding of anatomical structure and pathological anomalies as seen with modern imaging techniques.

## **ULTRASTRUCTURE OF THE TRACHEOBRONCHIAL EPITHELIUM OF THE NORMAL AND FORMALDEHYDE-EXPOSED GUINEA PIG**

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Formaldehyde has long been used as a preservative of cadavers in most of anatomy departments all over the world.

This work was designed to study the ultrastructural changes of the tracheobronchial epithelium of guinea pig after formaldehyde exposure.

Forty adult male guinea pigs divided into four groups were exposed to 0.5 part per thousand formaldehyde for 4 hours per day. The period of exposure was 3 days, one week, two weeks and four weeks for the first, second, third and fourth group respectively. A fifth group was examined without exposure as a control.

Many changes were observed in the tracheobronchial epithelium according to the period of exposure. These changes included increased mucous secretion, epithelial desquamation, erosion, ulceration thickening of the basement membrane and proliferative changes in the form of basal cell hyperplasia, squamous metaplasia, dysplasia and micropapillomatosis. The ciliated cells showed numerous changes including vesiculation of cytoplasm, dilation of rough endoplasmic reticulum, chromatin margination and many ciliary changes. The ciliary changes were in the form of loss of cilia, swollen cilia, truncated cilia, intracellular axonemes, disoriented basal bodies, disorganized cilia in the form of \*8+2) and (9+0), and vesiculation and rupture of the ciliary membrane. Three forms of ciliated cell loss were unidentified, the first was desquamation of intact cells, the second was rupture of the apical cell membrane and release of its organelles into the lumen and the third was ciliocytophthoria.

The goblet cells underwent massive secretion of their contents and the mode of secretion was changed from merocrine to apocrine. Later on, goblet cell hyperplasia occurred but many cells contained only few abnormal secretory granules, new cells were observed after formaldehyde exposure and also were the tunnel cells which were considered to be degenerating cells. In the subucosa, many undifferentiated cells were observed after two and four weeks formaldehyde exposure. The presence of these cells raises the possibility of fibrosis and permanent airway obstruction as sequel of long term exposure.

The similarity between the lesions produced by formaldehyde and the early lesions occurring during carcinogenesis raises the possibility of carcinogenic effect of formaldehyde. Assessment of formaldehyde carcinogenicity, teratogenicity and its effect on different parts of the body deserves further studies which are planned to be done in our department in the future. From our results, we recommend stopping, or at least minimizing the use of formaldehyde and employing other safer methods e.g. plastination.

## **A REPORT ON PRODUCTION OF THE FIRST TRANSPARENT HUMAN BODIES IN IRAN**

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In 1991, after taking part in the workshop held in Heidelberg, we started our activities in the field of plastination. In 1994, during the 7th International Conference of Plastination, we announced our success in starting plastination using existing equipment in our country. Now this year we are honoured to declare the production of the first transparent human bodies in Iran.

Additionally, we are working on plastination of the dissected whole body. A more detailed description will be given during the lecture.

## **A SIMPLE AND ECONOMICAL METHOD IN DISTILLATION OF ACETONE FOR A PLASTINATION LAB**

Ebrahim Esfandiari, Mahmoud Sheibanifar  
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Because large amount of acetone are used in a plastination laboratory, it is necessary to reuse it by distillation. However, a distillation unit is very expensive so we designed a distillation unit by existing equipment in our laboratory. A detailed description will be given in the paper.



## **CORRELATING SHEET PLASTINATED SLICES, COMPUTED TOMOGRAPHY IMAGES AND MAGNETIC RESONANCE IMAGES OF THE PELVIC GIRDLE: TEACHING TOOL**

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i Department of Anatomy and 2 Department of  
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**Introduction:** In the Dutch medical curriculum there is only a limited amount of time reserved for practical anatomy lessons. As a result the time spent in the dissection rooms has decreased steadily. In order to compensate for this problem we use pre-dissected specimen for educational purposes.

Most of our pre-dissected specimens are conserved using formaldehyde fixation or silicone impregnation. Recently the epoxy resin impregnation technique was introduced in our departments. With this technique we have produced anatomical slices of many different body structures which allowed us to study their topography in an anatomical correct state. These slices can also be used for teaching medical students.

**Materials & Methods:** The pelvis of a female cadaver was used, the bowels were removed before scanning and preparing the pelvis. Using a Computer Tomograph (CT) (somatom plus, Siemens, Germany) 2mm thick coronal coupes were obtained (120 kV, 210 mAs). For this purpose we mounted the pelvis on a wooden board as described by Entius et al. With manual reposition we obtained good coronal positioning. Using the reference crosses, as projected by the light beams of the CT, we marked this coronal position with a permanent marker. The CT data were used to obtain a 3-D reconstruction of the pelvis. Thereafter the pelvis was stored in a freezer (-20C) for about 7 days. The deep-frozen pelvis was sawed into 36 slices of approximately 2mm, using a handsaw (AEW 600, 5 teeth inch-1). The slices obtained were kept between plastic netting to keep them separated and allow more easy penetration of the acetone. Plastination was performed using an epoxy resin (E12, Biodur™) as described by von Hagem.

**Magnetic Resonance Imaging (MRI)** (Gyrosan T5, Phillips Medical Systems, Best, the Netherlands) of a preserved pelvis, using formaldehyde, is impossible due to soft-tissue changes, especially in the musculature. Therefore we had to compare the coronal CT images and plastinated slices with coronal MRI of the pelvis of a healthy female volunteer.

**Results:** We obtained 36 anatomical slices of the female pelvis. The most ventral part of the pelvis could not be sectioned because of technical problems (the risk of hand trauma to the handsaw operator was too high). We found a good overall correlation between the CT images and the anatomical plastinated slices. Correlation with selected MRI slices was also high.

The plastinated slices in combination with CT and MRI, were used for teaching and examination purposes. It was found that the use of 3-D images strongly enhanced the comprehension of 3-D anatomical structures.

**Discussion:** The combined use of plastination and radiological techniques as we have described will allow students, radiology residents, and anatomists to gain a better insight in 3-D aspects and the mutual relation between the various anatomical structures of the pelvis region. In order to gain insight in normal variations of the anatomy of the female pelvis, plastinated slices of different bodies could be useful.

The major problems with this technique is deviation from the coronal plane during sawing. This is due to the inhomogeneity of the pelvis and is very difficult to prevent.

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## **THE CURRENT PLASTINATION INDEX**

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Since the introduction of plastination some seventeen years ago, the developments of this process and its applications led to an increasing number of publications and communications in several languages all around the world. The aim of the Current Plastination Index (CPI) is to provide a data bank to those who decide to learn plastination, as well as to experience plastinators who want to search the literature about plastination. The CPI will be revised at regular intervals as a result to the information that you pass on to us. The participation of every plastinator is therefore necessary in order to compile and classify (according to language, journal, key-words) the world literature on plastination.

## **PLASTINATION IN ORTHOPEDIC SCIENCE FIRST RESULTS OF AN ORTHOPEDIC MANAGED PLASTINATION LABORATORY**

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We started to use the plastination method in our own laboratory one year ago, in the Department of Orthopedic Surgery at the University of Heidelberg. We have been interested in the following questions:

1. Is there a relationship between the intra-osseous vascularization of the patella and degenerative changes in the hyaline cartilage?
2. Rupture of the Achilles Tendon. Is it a problem of the vascularization?
3. The vascularization of the Anterior cruciate ligament. Where does its blood supply come from?
4. Is it possible to demonstrate arterial and venous vessels in bone with different colored plastic material?
5. The vascularization of the lunate bone in osteochondritis. Studies during extension and flexion of the carpus of man.
6. An experimental study in sheep. Treatment of osteonecrosis of the hip joint by drilling. Demonstrated by plastination.
7. Case report. Degenerative changes in a human meniscus demonstrated by plastination

As a result, we think that plastination is a very good method to investigate problems in orthopedic surgery. In all the above mentioned cases it has not been possible to use other methods in our experiments.

## **RECLAMATION OF ACETONE IN PLASTINATION LABORATORIES: A SIMPLE AND INEXPENSIVE METHOD**

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The cost of acetone used for the dehydration step in the plastination process has always been considered an important factor in the cost of plastination. Discarding used acetone also presents a problem because it must be treated as a toxic waste.

We have developed a method in three steps that permits us to re-use our acetone. The first step simply consists of storing the contaminated acetone in the freezer and separating by filtration the frozen water and fat that has congealed. The second step is vacuum distillation of the acetone and can be carried out with the freezer vacuum pump that is already found in any plastination



laboratory, with just a few additions. It produces 95% -97% pure acetone. The last step uses a desiccant to remove the residual water from the distilled acetone and brings the purity to 99.5%.

We used the above process for some time and now have modified it by adding two distillation coils and two collection reservoirs in series. Each coil was made from 2.5mm to 6mm copper tubing. Acetone is warmed to a temperature of 40-5°C and vacuum is applied and adjusted to maintain a moderate boil. The vaporized acetone passes into the distillation coils which are inside a deep freezer.

With these methods, we have reduced to a minimum the amount of acetone to be purchased and completely eliminated the cost of discarding used acetone.

As an additional reevaporation process, we direct the exhaust line from our vacuum pump during impregnation into the freezer and collect the vaporized acetone by condensing it into a jug inside the deep freezer.

### **THE USE OF PLASTINATED EQUINE HEAD SECTIONS OR THE GODFATHER PART V**

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As part of a study investigating ethmoid haematomas, a rare disease in equines, the ethmoid region was examined using conventional radiology (CR), computed tomography (CT) and magnetic resonance imaging (MRI). An atlas of correlative imaging of the normal equine head was produced. Photographs of transverse sections of 6 normal heads were matched with the corresponding CT and MRI images. Selected sections were useful for referencing when identifying anatomical structures during the completion of the atlas. In addition, sections from the head of a horse diagnosed with an ethmoid haematoma were plastinated, providing a permanent record of the destructive nature of this disease.

### **SPECIMEN PREPARATION FOR SILICONE PLASTINATION**

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Specimen preparation is a very important step of the silicone plastination process, especially when using fresh tissue. If you plastinate specimens that have been poorly or inadequately prepared, the final product will be inferior no matter how well the specimen is impregnated with silicone.

Minimal fixation may help assure a more natural looking specimen. Care must be taken to keep the specimen in its normal anatomical position. Loose portions of the specimen may be held in position with suture. Intravascular injection of colored silicone, gelatin, latex or epoxy may be used to highlight vessels. Hollow organs need to be flushed, cleaned, dilated and then fixed in a dilated position. Dilation of hollow organs will increase the flexibility of that organ by overcoming rigor and hence a thinner wall. Intestinal specimens may be opened to remove ingested matter, sutured closed and then dilated. Ostia with strong sphincters must be held open. Appropriate sized cannulas or tubing may be used. All cut vessels of heart preparations must be closed by ligatures or inverted corks ligated in place, except for one vena cava and one pulmonary vein. These will have tubing ligated in place and used for dilating each side of the heart. The atrioventricular valves of the heart can be accentuated by holding them away from the chamber wall and semilunar valves aided by packing with cotton or other materials. Joint capsules, distended with 20% formalin, aid in studying the internal anatomy of the joint.

Plan the theme of the specimen and limit the focus of the specimen to fewer items especially musculoskeletal preparations. Holes drilled into the marrow cavity of long bones in less noticeable areas enhance defatting and prevent sticky/greasy specimens years later. Old faded long-term fixed brains may be reju-

venated and made more useful by sectioning or prosecting and staining with astra blue, aldehydefuchsin or Darrow red to highlight the gray matter. Specimens may be plastinated following histochemical studies for long term review or study. A pink color may be added to the surface of the specimen using Biodur stain in the last acetone bath.

Beautiful specimens do not just happen.

### **CONSTRUCTION AND SERVICE OF A DATABASE OF PLASTINATED SPECIMENS**

Yayoi Masuda Kitamura, Yutaka Yoshida,  
Kazushi Minagawa and Jinghua Zhang Medical  
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About 200 plastinated specimens were made at the Medical Museum, University of Tokyo between 1991 and 1995. All of the specimens were registered in a databased system on a personal computer (Macintosh, Apple Co.) with commercial software FileMaker (Clarion Co.) and HyperCard. This database is used not only to catalogue the specimens but also to show the specimens to those who cannot come to the museum. Besides the database, information about the characteristics of specimens and methods of making specimens are edited with HyperCard for beginners. A part of the database was modified and demonstrated at two public exhibitions on plastinated specimens in 1995 at Tokyo. The database is also distributed on the Internet (<http://7202.230.170.207>).

### **MAGNETIC RESONANCE IMAGING (MRI) VERSUS THIN PLASTINATED CADAVERIC SECTIONS AS AN ANATOMIC LEARNING TOOL**

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The advent of new medical imaging technologies, such as magnetic resonance imaging (MRI), computed tomography (CT) and ultrasonography demand that cross-sectional anatomy be studied. Hence, anatomy students of all disciplines, as well as physicians and medical technologists, must understand the three-dimensional structural relationships of the body from a perspective different to that observed during dissection of a cadaver or conventional radiography.

It is believed that the relatively new technique of plastination, developed by Gunther von Hagens (1979, *Anat. Rec.*, 194:247) can serve as a highly effective anatomical teaching tool. Thin body slices used for educational purposes are well suited to plastination using various epoxy resins.

One half and two whole human cadaveric heads were scanned with an MR imager. Coronal, sagittal and transverse planes of imaging were recorded. After the heads had been frozen using dry ice they were sliced, with sections being cut to a thickness of approximately 6mm in the desired plane. These were then subjected to sheet plastination using either the PEM 11 or E12 technique. Thus, the intrinsic three-dimensional anatomical detail is preserved in specimens that are dry, odorless, durable and are pleasant to handle.

Measurements of certain anatomical structures will be taken on both the plastinated specimen and its corresponding MRI to determine the degree of correlation between the two modalities. The efficacy of plastinated sections as teaching tools will be evaluated by testing two groups of students studying head and neck anatomy. The existing anatomical knowledge of each group will be pre-tested using a correspondingly labelled MRI and plastinated specimen. Individual scores will be recorded, however student anonymity will be maintained.

Each group will then be given 30 minutes to revise the topographical arrangement of approximately 20 labelled features. One group will use MRI's and the other plastinated specimens. After this "study" period, students will again be presented with correlated plastinated sections and MRI's and be required to

identify the same labelled structures. Students will also be asked to complete a questionnaire asking for their personal preference on methods of learning anatomy. The pre and post test scores will be statistically analyzed to determine the educational value of the two distinct anatomical media.

### **PROBLEM ENCOUNTERED IN E12 SHEET PLASTINATION TECHNIQUE**

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Introduction: With the standard technique of E12 Sheet Plastination I have found separation of glass plate difficult to impossible.

Method: Fresh aborted fetus was played on a cork board and left in deep freeze at -25C for 3-4 days. On the 4th day using a fine-toothed saw, the fetus was sliced sagittally through the center. Thin slices (34mm) were made of each sagittal section, cleaned of sawdust and placed on a gauze for dehydration. Dehydration commenced with cold acetone in deep freeze. After 2-3 weeks dehydration was complete.

The specimen was now immersed in E12 solution and placed in a vacuum chamber for impregnation. Impregnation solution was made as follows: E12 100mls/EI Hardener 28 p.b.w./AE1020 mis.

After 24 hours these slices were placed in fresh E12 solution. The next day these slices were cast between glass plates (flat chamber method) using E12 casting solution. The specimen looked excellent. Casting Solution E12 100mls/EI Hardner 28 p.b.w./AeIO 20mls/AE30 5 p.b.w.

They were allowed to dry for 3-4 days and then placed in an incubator at 50C.

Results: The problem arose when attempts to separate the plates became difficult to impossible because the solution remained "sticky". Any forced separation resulted in ruining the specimen. This became worrisome because having gone through the whole plastination process, the specimen was destroyed at this late stage.

Consideration: Is it the shelf life that has no effect? The chemicals were 18 months old. However, E12 + EI Hardener were kept at +18C and AE10 and AE30 were kept at 4C. Previously, I have had no problems with this technique. If this is a problem of chemical storage:

1. What is the shelf-life of the chemicals?
2. Where should one store these chemicals - In the deep freezer?

### **APPLICATION OF PLASTINATION TO PRIMATE LOCOMOTOR ANALYSES**

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We have attempted to apply the double method for plastination to analyse the structural-functional relationships of macaque extensor digitorum et hallucis brevis muscle (EDB and EHB), the single intrinsic muscle of the forsum of the foot.

The muscle samples were dissected, fixed in 10% formalin and embedded in 10% gelatin. Using a Sartorius microtome, the specimens were sliced with section thicknesses ranging for several millimeters to 30mm perpendicularly (5X20mm sheets) or horizontally (15X25mm Sheets) to the longitudinal axis of the muscle bellies. Some 30mm thick sections were stained with Sudan Black B. The other sliced specimens were dehydrated in increasing grades of ethanol and plastinated with epoxy resin (Epon/DDSA+MNA=0.35/0.65, MNA/ DDSA=1/1). The thick specimens (3-5mm) were re-sliced at 3-20mm sheets after being incubated at 60C and were then stained with toluidine blue (40C).

These specimens showed several characteristics of good

preservation and cutting quality which enabled the depiction of structural and functional relationships. Muscle fiber types (type 1 and type II) were differentiated in sections stained with toluidine blue or Sudan Black B. There was sharp contrast between muscle fibers and myelinated nerves.

Using this method in the serial sections, muscle spindles can be reconstructed in three dimensions. Therefore, it is possible to calculate precise innervation ratios of EDB and EHB muscles by using the number of muscle spindles, muscle fiber population and myelinated thick nerves in given areas. In addition, the three dimensional distribution pattern of the muscle fibers arising from the lateral and dorsal side of the calcaneus can be traced until they divide into four bellies which extend to the four medial toes. We have found that the plastinated specimens obtained in the present study are superior to those of previous methods.

### **PREPARATION OF THE CORONARY ARTERIES FOR PLASTINATION**

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The technique of preservation of anatomical specimens using the method of Dr. Gunther von Hagens provides excellent demonstrations for teaching and further education. Of particular interest in the heart is its vascular supply, which we can display

in plastinated specimens injected with red Biodur E20.

The heart should be removed from the thoracic cavity taking care to divide the vessels some distance from the heart. Then the heart is rinsed with cold tap water and all vessels are blocked with cork stoppers. The stoppers for the aorta and the pulmonary trunk contain tubing, so that during the dilatation-fixation the aortic valve and the pulmonary valve are in an almost completely closed position.

Injection of the coronary arteries with 49.5 gram Biodur E20 red requires only slight pressure to fill the coronary vessels.

For the dilatation-fixation 15 L of 20% formalin solution are infused into the ventricle from a height of 1.5 meter. A week of immersion-fixation in 5% formalin solution at a temperature of +5C follows.

The sub endocardial fatty tissue and all fascia are carefully dissected away with scalped, scissors and forceps. Subsequently, the specimen is bleached in a 2% hydrogen peroxide solution with 5% formalin added to achieve the desired color. After washing for about 1 hour, the auricles are tamponaded with polyethylene-foils and the specimens are frozen overnight in a plastic bag. Dehydration lasts for 3 weeks at -25C in the deep freezer. During this time the acetone is changed twice.

Forced impregnation with Biodur S10 plus S3 hardener takes place for 3 weeks at -25C in the deep freezer. After removal from the vacuum kettle and draining overnight in the freezer, the specimen drains for one further day at room temperature. Then the specimen is placed into a plastic container for gas hardening with Biodur S6. This takes 2 weeks. Finally, a comprehensible injection and research in medicine.

### **LOW COST PLASTINATION TECHNIQUES FOR DEVELOPING COUNTRIES**

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The use of plastination is increasing, due to its many advantages. Developing and the underdeveloped countries are yet to catch up because of the high cost.

Some modifications have been made to the procedure to reduce the cost and save time. The specimens so plastinated are better for museum display, demonstrations and discussion, compared to the specimens in jars.

Fresh specimens were selected and cleaned in running

water; preserved in 10% formalin for 48 hours; dehydrated using 70-80-90-Absolute alcohol changes spending 3-4 days in each; cleared with Benzene for 3-4 days; transferred to the resin; after 48 hours, exposed to hardener for 2 hours and then air dried. The dissection was done to display the part of interest and then mounted on wood/acrylic base using wires. Colors were added to enhance contrast.

Advantages are a minimal use of materials and the procedure is done at room temperature and pressure. This results in greatly reduced cost.

### **STUDENT PARTICIPATION IN PLASTINATION AS A LEARNING EXERCISE IN A SCIENCE DEGREE EMBRYOLOGY UNIT**

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Students enrolled in their third year of a Bachelor of Science course at Curtin University used plastination techniques to preserve their dissolved specimens as part of the practical component of the embryology module of the Human Structure and Development unit coordinated by GMF at Curtin University. The experience and expertise of GW in S10 plastination technique was utilized. Students attended an information session on the methodology of plastination, which related well to their experience and knowledge of histological techniques using resin embedding. They then examined fetal piglet specimens, measured and weighed them to determine their various ages, and subsequently decided which aspects of development they wished to portray in their plastination specimens.

Dissection work was carried by each student, and the resulting specimens were dehydrated and plastinated over several weeks. Progress was monitored by the students with respect to dehydration, shrinkage and color retention. After 8 weeks, the plastinated specimens were examined and their component parts identified. The specimens were photographed with a digital camera and the resulting images imported into a hypercard stack representing the work of the class. The images were labelled and linked to information about their age and sectioning, and the plastinated specimens retained for use in the next years class.

As a result of participation in the plastination process, students were engaged in the technical aspects of their work and this increased their feelings of responsibility to the resulting dissection. Students gained insight into fixation and embedding techniques combined with image digitisation and multimedia presentation of graphical and text information.

Results of student evaluation of the plastination component of the unit, and examples of their work will be presented with this paper.

### **INVESTIGATIONS OF LYMPHATIC VESSELS WITH PLASTINATION TECHNIQUE: FIRST EXPERIENCE**

Alia M. Zaidman

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The lymphatic system remains one of the less well investigated systems. At the same time a number of pathologic processes are associated with the changes of lymph outflow. Demonstrations of the lymphatic vessels for teaching are also rather limited because of the difficulties in the preparation of permanent specimens. Plastination technique can solve these problems.

Lymphatic vessels were studied in rats and dogs. Intra-organ and surface vessels were detected by injection of either a mixture of indian ink with 10% gelatin or latex. This was followed by formalin fixation, dehydration and impregnation with Alizain stain. Epoxy resins were used for plastination. Currently attempts are being made to visualize lymphatic and blood vessels using plastination.

### **A POTENTIAL BEST STEREOGRAPHIC PRESENTATION OF THE PLASTINATED ANATOMICAL SPECIMEN PART 1 DEMONSTRATION OF REAL AND STEREOGRAPHIC IMAGES OF THE PANCREAS AND DUODENUM COMPLEX**

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In the 7th Congress at Graz, Dr. Yoshida, one of our authors, presented a Poster on the brain cortex and medulla to distinguish cortex in the process of making the plastinated brain slices. Hereafter, he made many sort of plastinated specimens and felt the volume of the plastinated specimen was decreased compared to the classical alcohol-solution immersed specimen mainly due to no use of the glass-ware. However, the dimensionality was not changed, although the silicon plastinated specimen hold such fabulous qualities as dryness, odourlessness, durability and tangibility, potentiality of literally being palpated and grasped. Space occupying characteristics is not improved through the plastination. In general, space occupying lesion is well known as a cancer. Only one of the disadvantages of the plastination is this space occupying characteristics. There is a cancer in the plastination technique.

Just at the very time, Mr. Iwahara of JVC proposed a classical stereographical technique of photography, the volumegraph the principally of which will be explained later in the second part by Dr. Koike. I think the method can be suffice for the deficit of the plastination, that is to say, the volumegraph cure the cancer of plastination. Surely, the plastination technique has marvellous qualities as a preserving method, compared to the alcohol immersion system but no dimensionality is lost.

The reduction of the dimensionality is attained by the sheet film stereographic presentation; The 3-dimension of the specimen in the glassware is converted into 2-dimension of stereographic sheet film of the volumegraph by Iwahara (JVC) without loss of stereographic images of the specimens. Furthermore, the volumegraph shows the same level of the color representation as that of the plastinated specimens as far as the naked eyesight is concerned. The volumegraph can be copied easily and less expensively and imported without completing a Quarantine Declaration.