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Histopathology Solvent Recycling. *Hawkes, Jack, Kumar MSA. Tufts Cummings School of Veterinary Medicine. USA*

In 2007, after we set up our plastination program, we realized that we weren't using our solvent recycler to its full capacity. Like many plastination programs we are part of a medical school, in our case a veterinary school, which is associated with a hospital. We worked out an agreement with that hospital's histopathology unit that would benefit both them and us.
Methods: We report our experience over the last four and a half years recycling alcohol and ProPar (a xylene substitute), and the lessons we have learned from that experience. We discuss the financial arrangement we agreed upon, and how that benefits all parties concerned. We briefly review the process of histopathology, and discuss the chemical and possible legal constraints on a recycling program. Finally, we emphasize the importance of a quality control program, and share some of the tools we have found to maintain the quality of our recycled solvents.
Results: To date we have recycled approximately 1500 gallons of solvents for the histopathology section; this has more than paid for our distillation unit.
Conclusion: A recycling program can be a good source of income for a plastination program, and is environmentally sound.

Combination of casting with E20 and consecutive plastination with S10 is a suitable technique for studying the vascularization of the kidney. *Dall, Annette M, Department of Neurobiological science, University of Southern Denmark, Denmark.*

The giraffe is an interesting animal, among other things, because it has an extremely high blood pressure and the vascularization of the kidneys is of special interest. Through a collaboration with the local zoo, where incidentally a giraffe was necropsied, we got the possibility to study the structure of the kidneys using our facilities for plastination, which are commonly utilized for human specimens used for teaching at the medical faculty.
Methods: Fresh kidneys were cast by injection of red epoxy-resin (Biodur E20) in the renal artery – according to Gunther von Hagens' "Heidelberg Plastination Folder." Afterwards one kidney was macerated using NaOH, while the other was immersion-fixed with 4% paraformaldehyde and thereafter plastinated (Biodur S10). Before curing, this kidney was cut in half by a coronal section using a Virchow brain sectioning knife.
Results: The anatomical division of the organ into cortical and medullary parts was extremely conspicuous using the combination of casting and plastination. The renal arterial structure from the renal artery to the interlobular arteries was visualized and examination at higher magnification showed the interlobular arteries associated with glomeruli. These structures were also visible in the macerated kidney.
Conclusion: The combination of casting and plastination sustains the relations between the vascular system and the surrounding parenchyma, which is not possible using the methods separately. The described combination of methods will also be suitable for kidneys from other species showing variations.

Whale Shark with Silicone Technique by Dalian Hoffen. *Gao, Hai-Bin[1], Sui, Hong-Jin[2]. [1]Dalian Hoffen Bio-Technique, [2]Dalian Medical University.Plastinated*

A 5.5 meter-long whale shark preserved by Dalian Hoffen using the silicone plastination technique is the largest plastinated fish in the world. The specimen remains with both its original shape and its internal structures.
Methods: A whale shark was found stranded and died in the Bohai gulf in June, 2009. After local fishermen found it and it was verified by fishery sector, Dalian Hoffen was entrusted to preserve it with the plastination technique. After injecting 750 liters of 10%

formalin and perfusing it with 300 liters of dye the specimen was immersed in 10% formalin for 6 months. The specimen was dissected to display muscles and organs, and then bleached using 5% hydrogen peroxide until it was a uniform color. The process of dissection and bleaching took one month. Due to the large size of the whale shark, it was cut into 3 parts. Cantilever cranes and tanks were specially designed and used for its dehydration and forced impregnation. The 3 whale sections were precooled at 5℃, then dehydrated in cold acetone for 3 months, and then degreased in acetone baths at room temperature for 3 months. The whale shark was impregnated in a cold vacuum for 2 months by forced impregnation. After impregnation was completed, the whale shark's 3 parts were reassembled through a reinforced steel framework fitted into its body. It was modeled to resemble the swimming posture above the surface of sea. The process of modeling and anatomically repairing took 2 months, followed by curing with gas and heat for 1 month.

Results: The process of the whale shark plastination took 18 months. The flexibility of its skin, muscles and organs tissues after plastination was easily differentiated. The whale shark with vividly swimming posture clearly showed its dorsal and ventral structures.

Conclusion: Dalian Hoffen preserved a dry, odorless, resilient, and durable whale shark specimen used not only for science popularization but also anatomical learning.

Micro-CT and sheet plastination assessment of the cadaver cricoarytenoid joint cavity. *Chen Shenghuo [1], Ren Jun, Wang Huaqiao[2], Xu Wen, Zhang Ming[3,4]. [1]Xinjiang Medical University. [2]Sun Yat-Sen University. [3]Capital Medical University, China, [4]University of Otago, New Zealand.*

The shape and movements of the cricoarytenoid joint (CAJ) remain subject of dispute and controversy. They are difficult to study in vivo, although in recent years a number of studies have presented tomographic observations in cadaveric and living subjects. However, the CAJ cavity has not been precisely described to date. The aim of this study was to identify the morphological features of the CAJ.

Methods: Twenty-five cadavers (nine females and 16 males; age range, 67-95 years) were used for gross anatomy, micro-CT arthrography, histology and E12 sheet plastination examinations. The cadavers were donated for the purposes of teaching and research under the Human Tissues Act, New Zealand.

Results: This study demonstrated that the dimension of the CAJ cavity was much larger than that of the articular surfaces, particularly at the medial and posterior aspects of the joint. The CAJ was lined by a wide joint capsule. The structural arrangement was mainly due to shear forces and only to a lesser extent to compressive forces based on the different strain acting in the CAJ. The size of the direct contact area of the opposing articular surfaces varied significantly between the sides of the same subject and among individuals.

Conclusion: Micro-CT arthrography and E12 sheet plastination are novel techniques for the direct visualization of the distinct features and structural analysis of the CAJ of the human cadaver, and to correlate the appearance of the CAJ cavity to its fibrous capsule, allowing investigators to visualize and to analyze mathematically the trajectory of the laryngeal cartilages more correctly.

Principles of epoxy plastination technique (E12). *Sora, Mircea-Constantin. Center for Anatomy and Cell Biology, Medical University of Vienna, Austria.*

The E12 plastination process is a well-established preservation technique used for demonstration in teaching and also in research (von Hagens et al., 1987).

Methods: *Slicing*: For E12 plastination we usually use fresh tissue which has been frozen at –80°C for one week, and cut with an average thickness between 3 and 5mm. The slices were stored at -25°C overnight. *Dehydration and Degreasing*: The acetone used for dehydration was cooled at –25°C. Each slice was placed between soft plastic grids in order to allow better circulation of the dehydration fluid. The acetone was changed once after 3 days at a concentration of 96%, by using technical quality acetone. The final concentration of the dehydration bath was 99%. When dehydration finished the freezer was disconnected. The temperature increased and after one day room temperature (+15°C) was reached. The acetone was changed with room temperature methylene-chloride for degreasing. Degreasing finished after 7 days.

Impregnation: Impregnation was performed at +5°C using the following epoxy (E12) mixture: E12/E1/AE10 (95:26:10 pbw) (von Hagens, 1985). The slices were submerged in the E12 mixture and placed in a vacuum chamber, directly out of the methylene chloride bath. Pressure was continuously reduced over the next two days down to 2 mm Hg. *Casting and Curing:* The slices were cast between two sheets of tempered glass and a flexible gasket was used as a spacer (4 mm). The following E12 mixture was used for casting: E12/E1/AE30 (95:26:5). The slices were placed between glass plates, sealed and the flat chambers were filled with casting mixture. Then they were placed for one hour in a vacuum chamber at 3 mmHg to remove small air bubbles present in the resin. Large bubbles were removed afterwards manually. After bubble removal, the flat chambers were placed horizontally inclined at 15° for one day. After one more day the flat chambers containing the slices were placed in an oven at 45°C for 4 days.

Results: The transparency and color of the slices were perfect and shrinkage was not evident. The finished E12 slices were semi-transparent, easy to orientate and offered a lot of anatomical details down to the submicroscopic level. The transparent loose areolar and adipose tissues contrasted perfectly with the muscle tissues and all epithelial parenchyma.

Conclusion: The E12 technique was and still is the preferred method for producing transparent body slices. Transparent body or organ slices are used for teaching and research purposes, because they allow studying the topography of all body structures in a non-collapsed and non-dislocated state. In addition, the specimens are useful in advanced professional training programs.

Knowledge, awareness and applicability of plastination technology for anatomical teaching and studies in Nigeria: Opinion of teachers of anatomy in medical institutions. *Azu, Onyemaechi Okpara,[1] Peter, AniekanImo.,[2] Aquaisua, NyongAquaisua.,[2] Ekandem, GabrielJohn.,[2] Naidu, Jesse.,[1] [1]Discipline of Clinical Anatomy, Nelson R Mandela School of Medicine, South Africa. [2]Department of Anatomy, Faculty of Basic Medical Sciences, University of Uyo, Nigeria.*

Plastination as a technique of tissue preservation was developed by Dr. Gunther von Hagens in 1977. It is a process of preservation of anatomical specimens by a delicate method of forced impregnation with curable polymers like silicone, epoxy or polyester resins with vast applications in medical fields of study. This technique preserves every part of the body and tissues for educational purposes. This article examines the knowledge and awareness of this invention amongst lecturers of anatomy in medical colleges as well as the possible applications of the technique.

Methods: The survey was carried out amongst 85 lecturers who participated at the 10th Annual Conference of the Society of Clinical and Experimental Anatomists of Nigeria, Enugu, March 2011.

Results: It was found that 50% and 23.75% of respondents had their masters and doctorate degrees in Anatomy respectively. Less than 8% utilized plastination as a tool for teaching as against 40% (plastic models), 36.25% (cadavers) and 15% (pathology pots). Conventional methods such as fixation by immersion (15%) and embalming (52.5%) with formaldehyde were commonly used for long term preservation of tissues in their various institutions. These methods were found to be less costly (25%), easy to use (56.25%) and the only method available (12.25%) even though they posed some health hazards (96%). The study found that 6.25% of the respondents did not know anything about plastination while 93.75% were aware of it.

Conclusion: The advocacy of preservation of tissues by plastination has been gradual in developed countries. All the respondents recommended the use of plastinates in medical schools in Nigeria. However, the extent to which it may impact the developing countries appears to depend on cost effectiveness and feasibility of implementation, as well as training of personnel.

Exploring the effectiveness of combining plastinated specimens with online learning modules in enhancing anatomy education. *Raoof, Ameed. The University of Michigan Medical School, United States*

The purpose of this study is to explore the effectiveness of guided modules that include diagrams, photos of prosected and plastinated specimens, and clinical vignettes to teach anatomy to medical students. Furthermore, it is to interpret which areas of existing

resources are most useful and how to maximize their benefit in order to best facilitate learning within the medical school.

Methods: This study measured how new styles of teaching clinical anatomy translated into effective learning in students. Data were obtained from two groups of medical students. Each group completed identical pre- and post- test exams covering both clinical and practical anatomy of the brachial plexus. The control group was the existing anatomy webpage as a study reference. Each student had a maximum of 30 minutes for each test and three hours to study. Each group also answered pre- and post-questionnaires asking them various questions about how effective each learning method was and demographic information.

Results: Our result was a pilot study. Our main goal was to increase the difference between post-test and pre-test scores for the test versus the control groups. The results were not statistically significant. However, if our numbers stayed consistent and our sample size was 82 students rather than 37, this result would be significant. With a class size of 170, it was not improbable that this study would prove to have significant results when applied to an entire class of medical students. Additionally, students who used the module to study rated themselves more comfortable with the material than the control group and more confident in their ability to do well on the post-test. Further, students who rated their confidence level higher had significantly better post-test scores.

Conclusion: Building upon these studies and tailoring them to the University of Michigan will be an extremely helpful tool as our anatomy program evolves to use more online instruction resources to supplement both lecture and dissection time. Additionally, there has not been extensive research done to show how an online module with a corresponding painted plastinated cadaver would improve learning. We hypothesize that mixing online-based education with actual specimens will further enhance the learning process, as students will be able to translate knowledge gained from online modules directly to a specimen in the laboratory. Furthermore, most studies of this nature do not focus on clinical correlates and seldom deal with neurology such as the brachial plexus.

3D Multidetector CT Reconstructions of a Heart and A Diencephalon and Brain Stem Plastinated by Biodur S10 Standard Technique. *Cerqueira, Esem.,[1] Baptista, Carlos Augusto.[2] [1]Department of the Heart Institute, University of São Paulo, Brazil, [2]University of Toledo, United States.*

Comparison between plastinated specimens and Computed Tomography (CT) and Magnetic Resonance (MR) images are used in anatomy and clinical practice. However, the direct examination of specimens, by CT, may be performed to evaluate its internal and external structures, and to ascertain whether the integrity of the structures can be of value in gross anatomy teaching.

Methods: A Toshiba Aquilion 64-multdetector CT scan, at Radiology Department of the Heart Institute – University of São Paulo - USP/Brazil, was used to evaluate plastinated specimens of a heart, a diencephalon and a brain stem. The specimens were plastinated in 1986 in The Department of Anatomy of the Institute of Biomedical Science - ICB/USP/Brazil, using Biodur S10 standard technique. Several images were generated from the scanned specimens. Cross-section slices of 0.5-mm slice thickness and 0.5-mm reconstruction interval were used. 3D images were reconstructed through MIP (Maximum Intensity Projection) and VR (Volume Rendering) techniques using Aquarius Net Viewer Workstation TeraRecon. The rate of CT attenuation coefficient (UH) of the images was measured and compared with images obtained from myocardium and white/grey matter of a living individual.

Results: The anatomical aspect of the heart, diencephalon and brain stem of the plastinated specimens were preserved. The internal structures of the heart, such as cardiac valves, ridges and bridges (trabeculae carneae), fibrous threads (chordae tendineae) and papillary muscles were remarkably preserved. The internal structures of the third ventricle and external aspect of the midbrain were also preserved after twenty-two years of plastination. The 3D reconstructions of anatomical structures of these specimens showed great detail and high spatial resolution. The radiological imaging showed increased attenuation rates, compared with the myocardium of the living specimen. When images of the plastinated grey and white matter were compared with the images of the living brain it showed reduced attenuation but less than the attenuation values of any kind of

calcification. CT images were not clear enough to recognize the layers of the myocardium wall, or the grey/white matter of the nervous tissue of both specimens.
Conclusion: CT scanning is an excellent method for assessing plastinated specimens, especially to reveal and evaluate either inner or outer surfaces, but not to differentiate their wall structures because the silicone impregnation altered the CT attenuation rates of specimens.

The strategy for the three dimensional reconstruction of anatomical structures by using plastinated cross-sections. *Matusz, Petru[1], Sora Mircea-Constantin[2], Wengert Georg[2]. [1]Anatomical Department, University of Medicine and Pharmacy "Victor Babes" Timisoara, Romania. [2]Center for Anatomy and Cell Biology, Medical University of Vienna, Austria.*

Computerized reconstruction of anatomical structures is becoming very useful for developing anatomical teaching modules and animations. Although databases exist consisting of serial sections derived from frozen cadaver material, plastination represents an alternative method for developing anatomical data useful for computerized reconstruction. The purpose of this study was to describe a method for developing a computerized model of different anatomical specimens by using plastinated slices.
Methods: Several anatomical specimens (ankle, lumbar spine, skull, shoulder joint) were used for this study. A tissue block containing the desired region was removed from the cadaver, then dehydrated, degreased and finally impregnated with the mixture E12/ E6/ E600. Using a band saw the E12 block was cut into 1 mm slices. Once scanned, these images of the plastinated slices were loaded into WinSURF and traced. After all contours were traced, the reconstruction was rendered and visualized.
Results: The generated 3D models displayed a morphology corresponding qualitatively to the actual cadaver specimen. The quality of the reconstructed images appeared distinct, especially the spatial positions and complex relationships of contiguous structures. Soft tissue features were easily seen when displayed with the bones positioned in the background. All reconstructed structures were displayed in groups or as a whole and interactively rotated in 3D space.
Conclusion: Plastination provides a useful alternative for generating anatomical databases. The reconstructed model can be used for residency education, testing an unusual surgery, and for the development of new surgical approaches.

The Use of Plastination in Community Outreach & Leadership Development for Medical Students. *Coulter James [1] Goodmurphy Craig W.[2], Gellman Jon[1]. [1]Eastern Virginia Medical School. [2]Pathology & Anatomy, Eastern Virginia Medical School, United States.*

A 4-week elective on Community Outreach and Leadership was developed at Eastern Virginia Medical School to help meet the institutional goals of being more community-centered. The course was intended to provide 4th year medical students with a basic sciences elective that would build their knowledge of leadership method and logistical consideration, empowering them to become community-minded leaders. This project was designed to engage 6th to 12th grade students in public schools in Hampton Roads, Virginia, using prepared kits of plastinated human specimens. The purpose of the kits was to provide visual and tactile learning aids that addressed the impact of 5 lifestyle choices on both related disease and US health care costs.
Methods: Over 150 middle and high school students were presented with human specimens prepared using room temperature plastination that included stomachs, hearts, pancreas, brains, cranial vaults and lungs. They were used as launching points for discussions about obesity, heart disease, diabetes, alcohol and drug consumption, preventable head trauma and smoking. After the discussion and presentation, students were given a 5 point Likert scale survey to measure their satisfaction, perceived knowledge acquisition and willingness to enter into a promissory contract to change their habits in an effort to mitigate the effects of these notable diseases and lifestyle choices. Videos of the presentations are being produced to place on the http://www.anatomyguy.com website for wider distribution beyond the Hampton Roads area. The organizing medical students have been asked to keep records of the organizational logistics, and a brief journal of their impression of each presentation with both student and teacher/adult responses to the presentation. Data on the

globalization of the project is being tracked on Google Analytics.

Results: At the time of the abstract submission data from the surveys is still being gathered and tabulated. Anecdotal responses from teachers and students were overwhelmingly positive with several requests for project expansion. The medical students involved reported that the experience was positive and helped them to feel both pride and a sense of community that other medical educational experiences have not offered them. They have recommended that the course in community leadership become a formalized opportunity.

Conclusion: The success of the interaction with the public school students, teachers and medical students will be formally put forward for Curriculum Committee assessment and approval for the 2013-14 electives calendar. The Galen Club, at EVMS is going to be continuing the project in the fall of 2012 and spring of 2013.

Silicone Plastination Trials at Room Temperature on Non Dysmorphic Pediatric and Perinatal Hearts Obtained at Autopsy for Medical Education. *Daniel, Richard L., Li, Xianming, Kelly David R., Daniel. Richard. Children's of Alabama Hospital, United States.*

Nondysmorphic perinatal and pediatric heart specimens from autopsies were plastinated with DowTM PR-14 silicone as trials prior to implementing a plan to plastinate selected hearts with congenital malformations for medical education. Most of the hearts had been stored in 10% formalin for two to ten years. Some hearts had been embalmed at the time of autopsy using a conventional embalming fluid.

Methods: Nondysmorphic formalin-fixed hearts were rinsed from 24 to 48 hours in tap water. Some hearts were submitted to ethanol baths prior to acetone. The previously opened hearts were posed prior to dehydration and impregnation of the polymer. Acetone at room temperature was used for dehydration and as the intermediary solvent. The dehydration progress was measured with a hydrometer. Following dehydration the hearts were submerged in DowTM PR-14 silicone polymer (premixed with cross linker and chain extender) and contained in polycarbonate vacuum chambers. The hearts were left to equilibrate in the polymer for 24 hours after which vacuum pressure was gradually applied over a 2 day period using a 1-1/2 horse power electric pump with onboard pressure gauges. The peak pressure utilized was approximately 20 inches Hg. Vacuum pressure was applied at intervals of 6 hours per 24 hour period until infrequent or no bubbles were observed. The impregnation time varied due to differences in the size of the hearts. The hearts were removed to drain for 3 to 4 days and intermittently wiped of excess polymer. Next a topical application of the curing or hardening catalyst Ct32 was applied with a brush and the hearts were carefully sealed in a plastic wrap for 24 hours. Finally, the hearts were suspended in sealed containers with petri dishes filled with the curing catalyst in an attempt to gas cure the specimens. The hearts were observed at 24 hour intervals until dry at which point the process was deemed complete.

Results: Well-fixed previously unembalmed hearts with shorter fixation intervals (<2 years) that had been submitted to ethanol baths rendered very good finished specimens. Older heart specimens particularly those housed in an inadequate volume of fixative and those not bathed in ethanol had color variations and many had white precipitate.

Conclusion: The quality of these plastinated nondysmorphic heart specimens seems to depend on initial specimen integrity, shorter fixation intervals with adequate fixative volume, thorough rinsing with water, and utilization of ethanol baths prior to acetone dehydration.

Plastinated bodies exhibitions – A survey study in 500 young individuals. *Raikos Athanasios[1], Paraskevas George[2], Tzika Maria[2], Kordali Panagiota[2], Natsis Konstantinos[2]. [1]Department of Anatomy, Faculty of Health Sciences & Medicine, Bond University, Gold Coast, Australia. [2]Department of Anatomy, Medical School, Aristotle University of Thessaloniki, Greece.*

Plastinated bodies exhibitions are amazingly popular and attract millions of spectators worldwide. The aim of the study was to investigate the opinion of young individuals about such exhibitions and expose any positive or negative comments and worries on the issue.

Methods: For the purpose of the investigation, an anonymous survey study was conducted on 500 randomly selected young individuals aged 18-35 years

old. The questions concentrated on the public opinion about shows exhibiting plastinated human bodies, specimens, and organs while additional questions were provided about body donation strictly for scientific use.
Results: Some participants in the study (4.5%) had already visited a plastinated bodies exhibition, while 51% intended to visit one in the future. Moral issues were expressed by 46.3% of the sample with the way such shows were conducted, and 21.8% of the sample raised religious/philosophical concerns, while 28% stressed the potential for psychological/mental health disturbances. The majority of the participants in the study (73.4%) agreed with body donation strictly for educational use.
Conclusion: As demonstrated in our results, a considerable number of participants in the study raised various moral worries about the attendance of plastinated human bodies' exhibitions. On the other hand, the frenzied rhythm of knowledge spreading and the ease of access through the Internet allow no more close doors and barriers to the general audience. However, normal and pathological anatomy education of the general public requires special management and should be handled by specialists in a series of steps and according to the cohort's age in order to secure the physical and mental health of the public.

Chinese Visible Human Project and its Application.
Zhang Shao-Xiang. The Third Military Medical University, China.

Research on the digital visible human is of great significance and application value. The first digital image data sets of complete humans (male and female) were made in the United States in 1995. To promote worldwide application-oriented Visual Human Project (VHP), more visible human data sets representative of different populations of the world are in demand.
Methods: The team in the Third Military Medical University worked on the Chinese Visible Human Project from 1999 until the present.
Results: The Chinese Visible Human (CVH) male (created in Oct. 2002) and female (created in Feb. 2003) project achieved greater integrity of images, easier blood vessel identification, and were free of organic lesions. The most noteworthy technical advance of CVH Project was construction of a low-temperature laboratory, which contributed to prevention of small structures (including teeth, concha nasalis, and articular cartilage) from falling off the milling surface. Thus, better integrity of images was ensured. So far, we have achieved acquisition of five CVH data sets and their volume visualization on PC. 3D reconstruction of some organs and structures has been finished. The work of segmentation of a complete data set is also under way.
Conclusion: Although there is still a distance to go to make the visible human meet the application-oriented needs in various fields, we are taking our first steps toward future application by acquiring new data sets, performing segmentation and setting up a platform of computer-assisted medicine.

Three-dimensional reconstruction of the deep epaxial muscles of the chick using Biodur E12.
Adds, Philip. St George's School of Medicine, University of London, United Kingdom

The developmental processes leading to the formation and individuation of the deep intrinsic postvertebral muscles have been little studied and are not well understood. The detailed anatomy of these muscles is complex and descriptions in the literature often differ between authors. As part of an on-going investigation into the development of the deep epaxial musculature and its corresponding tendons of attachment, a 3-D reconstruction of these muscles has been undertaken from sectioned day-old chicks.
Methods: Day-old chicks of Gallus domesticus were used for this study. Two sets of chicks were prepared for sectioning: formalin-fixed and unfixed. The chicks were first dehydrated in acetone at -30°C then embedded under vacuum in Biodur E12 epoxy resin, using E6 hardener and E600 accelerator. The chicks were removed from the vacuum and dissected to remove the head, wings and legs. The skin was removed, and the body was cut into sections (neck/thorax/abdomen). The sections were placed in rectangular moulds and covered in epoxy resin mixture before degassing and curing. Sequential thin sections (0.5 mm) were cut using a slow-speed diamond circular saw (Buehler IsoMet Precision Sectioning saw) with a 100 mm diameter blade. After each section was cut, the exposed surface of the block was photographed with a Nikon D60 Digital SLR with a

Sigma 105 mm f/2.8 EX DG Macro lens. Three-dimensional reconstruction of the serial sections was carried out using 'Reconstruct', a free editor for serial section microscopy.
Results: The individual muscles and their tendons of attachment could be differentiated, enabling the muscles and tendons to be delineated by drawing around them on the computer screen. It was then possible to create the 3-D image of the musculature demonstrating the morphology of the muscles.
Conclusion: Embedding in Biodur E12 epoxy resin facilitated the sectioning and analysis of serial sections of the thorax of the day-old chick. Using 'Reconstruct' software a 3-D image of the musculature can be built up allowing the image to be rotated to study the detailed anatomy of the postvertebral muscles and tendons.

State of preservation and extraction of DNA in silicone plastinated specimens. *Yu, Shengbo, Fu Yuanshan, Gong Jin, Chi Yanyan, Zhang Jianfei, Zheng Nan, Sui, Hong-Jin. Dalian Medical University, China*

The technique of plastination has developed rapidly in recent years. It can preserve macro-biological specimens almost forever. Silicone plastination technology is the most widespread method to be applied in this field. But the state of preservation of hereditary substances in the plastinated sample was unknown after the process. The objective of this study was to investigate the state of preservation of the hereditary substance and explore the extraction of genomic DNA from specimens plastinated with silicone ploymer.
Methods: The specimens investigated were livers of Wistar rats which were divided into experimental and control groups. Experimental samples were treated through the standard plastination process including formalin fixation, dehydration, degreasing, forced impregnation and hardening. The genomic DNA extraction kit was provided by Takara. Direct and pretreatment methods were applied in the extraction process. Paired observation was used to compare fresh samples with plastinated ones by agarose gel electrophoresis.
Results: Genomic DNA was not obtained in the plastinated specimens by the direct DNA extraction method. Although a clear smear was observed by agarose gel electrophoresis compared with fresh samples. If pre-treated with sodium salt solution, a small amount of genomic DNA remained which could be extracted from plastinated samples.
Conclusion: Silicone plastination technology may preserve the general morphology, while genomic DNA may also be kept to some extent. Sodium salt solution pretreatment is the key step which guarantees successful DNA extraction and quality from silicone-plastinated specimens.

Histopathological evaluation of biomedical devices using a plastic embedding technique. *Medlej, Bahaa, Slob Viviane., Loeb Emmanuel., Hershkovitz. Tel-Aviv University, Israel.*

The objective of this study was to present a method that allows histological examination of soft tissues containing solid medical devices, for the purpose of histopathological and morphometric evaluation.
Methods: The targeted tissues were fixed in 10% formaldehyde for 48 hours (time varies according to sample thickness), and then dehydrated in increasing concentrations of ethanol (40%-100%) and in xylene (3-5 days). Samples were then placed in methyl methacrylate 99% (Sigma-Aldrich, Germany) for 3-5 days. Subsequently, samples were embedded in a mixture of methyl methacrylate 99%, polyethylene glycol 400 and benzoyl peroxide powder (Sigma-Aldrich Germany); and placed in a vacuum at room temperature. Sections (minimum thickness 500µm) were cut using an Isomet low-speed saw (Buehler, Germany) equipped with a diamond wafering blade (Buehler, Germany). The specimens were then glued to plastic slides and ground to a thickness of 6-50µm by a Phoenix/Beta Grinder/Polisher (Buehler, Germany), using metallographic grinding discs of decreasing coarsening (320-1200 Grit) and Metadi Supreme polycrystalline diamond suspension (1-6µm) (Buehler, Germany). Prior to staining, the specimens were immersed in formic acid for 1-5 minutes.
Results: Histological slides, with different types of medical devices implanted in blood vessels, bones, joints and skin tissue, were demonstrated. In addition to the standard histological observations, such as necrosis, inflammation and fibrosis, this technique provides reliable data on size and shape of anatomical

structures, such as blood vessel diameter, tissue proliferation adjacent to the implant, etc.
Conclusion: The plastic embedding method offers the investigator the opportunity to examine the pathological changes and tissue reactions to an implanted medical device, which cannot be adequately examined using the paraffin embedding technique.

Morphological aspects of renal arterial supply and the small intestine of *Procyon cancrivorus*. *Kleber, Fernando Pereira. Federal University of Goias, Brazil.*

The use of polymers to study the architecture of organs and tissues is becoming feasible because of low cost and ease of handling. The aim of this study was to describe the arterial supply of the small intestine and kidney of the raccoon (*Procyon cancrivorus*) using injection of acrylic.
Methods: Animals killed by accident on highways were collected. The aorta was cannulated and injected with a mixture of autopolymerized acrylic and red pigment. After hardening the specimen was fixed in formaldehyde solution (10%). The dissections were documented using a digital camera (Sony Cyber-shot camera, 8.1 megapixels).
Results: The cranial mesenteric artery supplied the jejunum and ileum on the mesentery. Near the mesenteric border, anastomoses formed vascular arcades and vasa recta supplied the mesenteric side. The artery for the ileum arose as the final branch of the cranial mesenteric artery. In the kidney, the renal arteries were always single, bifurcating into dorsal and ventral segments, and subdivided into several segments
Conclusion: The technique of injecting polymers is of low cost and easy implementation, generating material of excellent quality, enabling the identification of all mesenteric and renal vasculature. The technique of plastination, which will be used in the next study, will provide more organic life parts to be handled for study and demonstration, as well as those that are kept on permanent display.

Mysterious Life Museum - a unique museum with plastinated specimens. *Sui, Hong-Jin[1], Gao Hai-Bin[2]. [1]Dalian Medical University, [2]Dalian Hoffen Bio-Technique, China.*

The museum is a social classroom for improving people's scientific knowledge and cultural level.
Methods: Traditional biological specimens in the museum include stuffed specimens, skeleton specimens, bottled specimens, etc. Plastinated biological specimens can show not only the original shape and appearance but also the internal structure.
Results: The Mysterious Life Museum is the first museum showing plastinated biological specimens, set up in Dalian, China in April, 2012. This museum contains 2,000 plastinated specimens of ocean animals, vertebrate animals and human bodies. The ocean animal hall reveals the mystery of blue ocean life. It shows different kinds of fishes as well as marine mammals. The key point of this hall is to introduce the differences between the fishes and marine mammals. The vertebrate animal hall reveals the mysterious kingdom of vertebrates. Through comparative anatomy, the characteristics of homologous, analogous and vestigial organs show the great variability and uniformity of vertebrates, and the evolution of the life will be understood further.
Conclusion: The hall of human body shows the mysterious body world. The exhibits lead people into the internal world of the human body, and allow visitors to understand the importance of health, and the danger of bad living habits.

P40 Sheet plastination: An emerging tool in medical teaching and imaging studies. *Dhingra, Renu [1], Pandey Jayashrip[1], Seith Ashu[2], Lalwani Sanjeev[3], Kumar Ranik[1]. [1]Department of Anatomy, [2]Department of Radiology, [3]Department of Forensic Medicine, All India Institute of Medical Sciences, New Delhi, India.*

Knowledge of cross sectional anatomy is a pre-requisite for accurate interpretation of CT and MRI images of the brain. Sheet plastination gives a better understanding of cross sectional anatomy, it augments the diagnostic and clinical acumen of radiologists and physicians thus enhancing the care and and monitoring of patients. Plastination is an excellent tool for tissue preservation. In our laboratory at AIIMS, we standardized the protocol for plastination of brain slices

using the P40 technique as it offers better gray and white matter differentiation and has better instructional value.

Methods: The brains were procured from autopsy cases being done in the Forensic department and cadavers donated to the Anatomy department at AIIMS. Coronal and horizontal 4 mm slices of each fixed brain were dehydrated in acetone at -25°C. Forced impregnation with P40 and A4 mixture (100:1) was carried out at room temperature as well as at -25°C. The brain slices were slipped into glass chambers filled with immersion mixture and then exposed to UVA light in the UV curing chamber. Measurements (length and breadth) were obtained before dehydration and after curing and shrinkage percentage was calculated.

Results: Room temperature impregnation was faster than cold impregnation. It is also cheaper in the Indian scenario because of the electricity cost. The mean % shrinkage in length was more in specimens impregnated at room temperature (7.99± 2.15) as compared to specimens impregnated at -25°C (6.12± 1.32). Similarly the mean % shrinkage in breadth was more in specimens impregnated at room temperature (8.90 ± 2.37) as compared to specimens impregnated at -25°C (7.20± 1.46). The brain slices exhibited a clear visual contrast between gray and white matter and beautifully highlighted blood vessels. The gray and white matter differentiation was much more distinct with P40 impregnated brain slices (both room temperature and at -25°C impregnation) as compared to S10 slices. The structures like caudolenticular bridges, anterior commissure, external capsule and claustrum were better appreciated in P40 sections than S10 brain sections.

Conclusion: P40 plastinated brain slices can be ideal for teaching and examination purposes. A thorough knowledge of brain sections can help in correct interpretation of CT and MRI images thus enhancing the diagnostic acumen of clinicians. They require minimal aftercare and can be used repeatedly without deterioration.

The neurovascular bundle of the knee: an anatomic study using plastinated cross sections. *Wengert, Georg[1], Sora Mircea-Constantin[1], Matusz, Petru[2]. [1]Center for Anatomy and Cell Biology, Medical University of Vienna, Austria. [2]Anatomical Department, University of Medicine and Pharmacy "Victor Babes" Timisoara, Romania.*

The aim of this study was to evaluate the topography of the saphenous nerve branches and the popliteal neurovascular bundle of the knee. The anatomic relationships of these structures at different levels of the knee were studied as an aid in planning minimal invasive surgery. A thorough knowledge of the local anatomy is a prerequisite prior to attempting posterior arthroscopy, in regard of the saphenous nerve, popliteal artery or the sciatic nerve and its branches.

Methods: A sectional anatomical study, using plastinated transparent knee cross sections, was performed on twelve intact right male cadaver lower limbs. The measurements were performed at the level of the medial epicondyle, at the joint line level and at the level of the tibial attachment of the posterior cruciate ligament (PCL).

Results: The popliteal artery is predicted to be 8.66 ± 2.17 mm dorsal and the sartorial branch of the saphenous nerve 4.27 ± 0.05 mm posterior to the joint capsule at the level of the medial epicondyle. At the joint level, the popliteal artery is 7.86 ± 2.26 mm away from the posterior cruciate ligament and the sartorial branch of the saphenous nerve is predicted at 2.41 ± 0.12 mm posterior to the joint capsule. At the level of the tibial attachment of the PCL the popliteal artery to PCL distance is 5.93 ± 3.61 mm and the sartorial branch of the saphenous nerve is situated 4.36 ± 0.43 mm posterior to the joint capsule.

Conclusion: Based on our anatomical data, a posteromedial portal placed at the level of the medial epicondyle seems to be safe, effective and reproducible. At this level the popliteal artery is situated widely lateral to the medial epicondyle and the distance between the saphenous nerve branches and the articular capsule is greater than at the other levels. Therefore, a portal placement here would be advantageous and safer. Anatomical characteristics should be kept in mind when knee surgery is performed, thereby reducing the risk of injury to the saphenous nerve and neurovascular bundle, and offering easy access to the posterior compartment of the knee.

Sectional anatomy of plastinated knee joint: A boon for anatomists, radiologists and orthopedists. *Jain, N.[1], S. Lalwani.[2], R. Dhingra.[1]*
[1]Department of Anatomy, [2]Department of Forensic Medicine, All India Institute of Medical Sciences, India.

The increasing use of cross sectional imaging techniques for clinical diagnosis makes it mandatory to understand anatomy in all dimensions. Since in humans the knee joint supports nearly the whole weight of the body, it is most vulnerable both to acute injury and the development of osteoarthritis. Plastinated slices of the knee region constitute excellent teaching material in cross-sectional anatomy, a field of ever-increasing importance, and correlate well with radiographic images. Exact knowledge of the topographical anatomy is not only a pre-requisite but also facilitates accurate clinical diagnosis in imaging techniques like MRI, CT and sonography. The present study was undertaken to plastinate the knee region for studying sectional anatomy of the knee joint.

Methods: Knee regions were collected from the Department of Forensic Medicine at AIIMS. They were washed, cleaned, and fixed in 5-8% formalin. The joint was filled with 120-250 ml of fixative. The knee region was plastinated using the standard S-10 silicone technique with some modifications. The specimens were frozen to -40°C before sectioning. Plastinated knee specimens were sliced in coronal (1cm) and sagittal planes (1 cm) with a band saw to demonstrate the internal features and to understand the relationship of various muscles, extra- and intra-articular ligaments, blood vessels and nerves. These slices were then compared with MRI images of the same.

Results: In the mid-coronal slice of the knee region the medial and lateral femoral condyles, medial and lateral menisci, medial and lateral collateral ligaments, posterior cruciate ligament and the lateral head of gastrocnemius were observed. In the mid-sagittal section, the patella, lateral condyles, popliteus, gastrocnemius, lateral head, plantaris, patellar ligament and both the horns of the lateral meniscus, infrapatellar fat pad and suprapatellar bursa could be seen. All the structures correspond exactly to the MRI images.

Conclusion: Plastinated coronal and sagittal sections of the knee are ideal for studying the sectional anatomy of the knee not only because of their instructional value but also their durability, ease of handling, transportation to the operation theatre and provide a ready reference material at the work place for clinicians.

A new method of brain plastination. *Esfandiary, Ebrahim [1], Asadi Mohammad Hossein[2], Rabiei Abbas Ali[1], Setayesh Mehr Mohsen[1], Taghipour Monir[1], Bahadoran Hossein[2]. [1]Department of Anatomic Sciences, Isfahan University of Medical Sciences, Isfahan, Iran. [2]School of Medicine, Baghiatallah University of Medical Sciences, Tehran, Iran.*

Plastination is a unique technique for preservation of biological specimens used for teaching medical students. The standard protocol of flexible sheet plastination includes fixation, slicing, dehydration, forced impregnation, casting and curing which when done by P87 flexible unsaturated polyester resin, gave heavy, gross fragile and bubbling plastinated sheets. Through this project, we omitted the casting stage and saved polyester resin. Then, we compared fragility, weight and bubbling of the new sheets with those of sheets provided by the standard method.

Methods: This study was carried out on three human brains. Initially according to the conventional methods they were fixed in 10% formaldehyde, and cut into sagittal, coronal and horizontal thin sections of 3mm thickness, with a meat slicer. They were then dehydrated in cold acetone (-25°C) and immersed in P89 unsaturated polyester resin at 25°C. Finally the specimens were taken out from the vacuum chamber and exposed to room temperature. When both surfaces of the specimens became dry, they were taken to P89 polyester resin again. This step was repeated 10 times.

Results: The weight of plastinated sheets with P89 was about one tenth of the weight of plastinated sheets with P87. Some degree of gross fragility was observed in the standard method, while it was not seen in P89 flexible sheets. P89 plastinated slices showed excellent differentiation between white and gray matter of the brain fixed for 2 years, while the fresh brains which were fixed for 12 weeks showed less differentiation.

Conclusion: In conclusion, our study showed that the P89 technique is a quick and less expensive method of producing sheet plastinated specimens suitable in teaching neuroanatomy.

Staining of brain slices and room temperature impregnation with Biodur S10/S3. *Mooncey, Mumtaz, Mandeep Gill Sagoo. St George's Hospital Medical School, London, United Kingdom.*

Stained and plastinated brain slices have the potential to be used as an effective tool in the teaching of neuroanatomy. These specimens are easy to handle, long-lasting and a good alternative to plastic models. Different staining methods can be used to differentiate between the grey and white matter, and this study compares the effects of the following methods on the brain slices: Mulligan's, Tannic acid, Le Masurier and Roberts's methods.

Methods: The standard procedure for plastination using Biodur S10/S3 silicone involves low-temperature dehydration in a volatile solvent, followed by forced impregnation under a vacuum at -15° C. However, due to the health and safety concerns and high expenses implicated by such conditions, this study uses low-temperature dehydration and room temperature impregnation instead. Previous studies at St. George's have shown that this adapted protocol has the potential to produce results which are comparable, if not equal, to the results from the standard procedure, with the additional advantages of lower costs and simplicity of set-up. This project investigates the reduction in size of unstained brain dissection specimens and stained brain slices, after each stage of the staining and plastination process (after staining/before dehydration, after impregnation and after curing). In this study, some of the brain specimens were sliced and comparisons were drawn between the effects which the four different staining methods had on them. The process of plastination involves four different stages; fixation, dehydration, impregnation and curing. In this case, the dehydration step was performed at -30° C, over three weeks, using acetone of increasing concentrations over this period. Next, the vacuum impregnation with S10/S3 and curing with S6 steps were carried out at room temperature. Measurements of the slices were taken at each stage of the process, in order to monitor the level of shrinkage.

Results: It was found that the color produced by the staining methods was maintained, following completion of the plastination process. Although in some cases there was a small degree of shrinkage of the specimens, this was found to be of an acceptable level for teaching purposes.

Conclusion: The stain which was found to be most effective was the Roberts's method, as it provided the clearest differentiation between the grey and white matter. This study further extends the potential applications of a cost-effective room temperature plastination (impregnation) method.

Considerations during S10 Plastination of a Gabon Viper (*Bitis gabonica*). *Arredondo Jorge[1], Dom_nguez Guillermo[1], Jackniuk Mariela[1], L López-Albors Octavio[2], Hernandez Wendy[1], Gonz_lez Patricia[1], Aja Guardiola Santiago[3], Ayala María-Dolores[4], Sarri_ Ricardo[2], Latorre, Rafael [2]. [1]Department of Anatomy and Physiology, Autonomous University of the State of Mexico, Mexico. [2]Department of Anatomy and Comparative Pathologic Anatomy, Murcia University, Spain. [3]Department of Morphology, National Autonomous University of Mexico, Mexico.*

The Gabon viper (*Bitis gabonica*) is highly venomous species from sub-Saharan Africa. Its venom is deadly as it is hemotoxic with the capability to destroy the blood and lymphatic vessels causing severe hemorrhage. During its plastination process it is mandatory to handle this specimen carefully to avoid accidental inoculation of the venom in the operator, since it is not certain if the venom of this viper remains active after plastination. The main purpose of this work is to describe a method to minimize the risk of damage by the toxins of the venom's glands of a Gabon viper during S10 plastination method.

Methods: The full cadaver of the viper was longitudinally aligned and mounted over a wooden board and firmly attached to it and then frozen at -25°C for 72 hours. A block section containing the head was sectioned and separated from the rest of the body and submerged in a solution of 1% phenol with 37% formaldehyde for one week. The baths were replaced weekly for three weeks. The head was rinsed with tap water and then placed in cold acetone baths to dehydrate it. The rest of the body was frozen at -25°C for one week and then sectioned 1cm thick in the

transverse plane. The head and the rest of the body were plastinated by the cold temperature S10 method.
Results: The specimen showed great anatomical details. The topography of the organs was fully represented on the plastinated slices. The full attention and careful preparation of the specimen during plastination avoided any potential harm to the laboratory personnel, and permitted the use of the rest of the S10 polymer contained in the vacuum chamber in another plastination process, without risk for the operator.
Conclusion: This method provides a good alternative to handling venomous vipers during plastination and can also reduce the loss of S10 Biodur polymer due to contamination with poisonous toxins, however, more specific studies on the activity of the venom after plastination are needed.

Decalcified Bone Plastination by the new UP89 resin. *Rabiei, Abbas Ali., Esfandiary, Ebrahim., Setayesh Mehr, Mohsen., Shamosi, Atefeh., Mardani, Mohammad., Dashti, Gholam Reza. Isfahan Medical University, Iran*

Plastination specimens have a unique position as teaching aids. In plastination, water and adipose tissue are replaced with an intermediate solvent which is replaced with suitable polymer. The specimens obtained are odorless, stable and non-fragile. The scope of this study was to preserve the whole structural details of bone and make it not fragile, durable and very light.
Methods: Sheep bone specimens (femur, hip and scapula) were fixed in 5% formaldehyde for 20 to 30 days according to the bulk of the samples. They were decalcified with 5% nitric acid for different times and then washed under tap water. They were dehydrated in –25°C acetone and defatted in 30°C acetone. Finally they were placed in a vacuum chamber for forced impregnation. Acetone present in the tissue was replaced with a new flexible unsaturated polyester resin (UP89). After this process, curing was carried out via heat and UV light exposure.
Results: Plastinated bones prepared by this method were found to be lighter than their primary samples. They were not fragile, and were durable and odorless. All parts of the bones, e.g. groove, tubercle, cartilage, ligament, were preserved.
Conclusion: We propose this new method of decalcification of bones before plastination for human cadavers because the method prepared bones of excellent quality for teaching.

Double staining method for fetal specimens to enhance the educational value of plastinates. *Miklosova, M.[1], Miklosova, Z.[3], Korpova, M.[1], Kalatova, B.[1], Dankovcik, R.[2], Dudas, M.[1]. [1]Department of Biology and Ecology, Safarik University, [2]Department of Gynecology and Obstetrics, Safarik University and Pasteur University Hospital, Slovak Republic, 3Department of Pharmacology, Masaryk University, Czech Republic.*

The improved double-staining technique to demonstrate cartilage and bone can now be used in teratological and toxicological studies as well as in morphological studies of large specimens. Moreover, it can be used to process museum skeleton specimens preserved in liquids. The principle used here is the affinity of alizarin red S to bind with the calcium of bones, while alcian blue reacts with acid mucopolysacharides in cartilage. The end product of this technique is a specimen in which the ossified parts of bone are stained red and cartilages are stained blue. This staining allows us to observe primary ossification centers, transition of cartilage to bone, and related abnormalities.
Methods: We examined large fetuses and neonates. The improved procedure involves rapid, complete skinning of fresh eviscerated specimens. The specimens are incubated with Alcian Blue 8 GS and Alizarin Red S. The cartilage stains blue in color and bones are red, both embedded in the transparent (cleared) muscle. Before examination or storage, the double stained specimens must be cleared in graded concentration arrays of glycerin. The specimens can be stored in pure glycerin.
Results: Differentiation of colors was very good, but specimens were wet and handling was difficult. Fresh, formaldehyde-fixed or alcohol-fixed specimens from various species worked well.
Conclusion: Although fragile and not ideal for mounting, this type of specimen is excellent for detailed scientific skeletal studies in 3D, and may also be used in multiple settings in university teaching curricula. **Supported by the grant GEMIN-2007/65-UPJS-02 from the Slovak Ministry of Health (M.D.).*

Plastination of specimens using new flexible unsaturated polyester resin (UP 89). *Setayesh Mehr Mohsen, Ebrahim Esfandiari, Abas Ali Rabie, Gholam Reza Dashti. Isfahan University, Iran.*

Specimens perserved using the new flexible unsaturated polyester resin (UP89) were synthesized in the plastination laboratory of the Anatomy department in Isfahan University of Medical Sciences. Plastination was first introduced by Gunther Von Hagens in 1978 in Germany. Plastinated specimens have a unique position as teaching aids for easy storage and handling by students. In this study we applied the UP89 technique to examine the new synthesized resin instead of the silicone resin in the conventional (S-10) technique.

Methods: This study was carried out by using suitable raw materials of modified unsaturated polyester resin (UP89). The synthesized resin was found to be transparent and flexible enough to be used in plastination. Animal specimens such as heart, liver, femur of sheep, fish, lizard and human specimens such as heart were utilized. Initially according to the conventional method, they were fixed in 5% formaldehyde, then dehydrated in cold acetone (-25 C°) and defatted in warm acetone (+25 C°). In the forced impregnation stage, 1% peroxide (MEKP) was added to the resin, and then the specimens were placed in the resin within the vacuum chamber. Forced impregnation was performed at a low temperature (10 C°) in order to avoid the gelation of the resin. Curing of the specimens was carried out in the UV light and heat cabinet. After a period of two months, appropriate curing was obtained and the specimens were found to be dry and odorless.

Results: The specimens obtained by UP89 technique were found to show acceptable appearance for teaching anatomy and as museum specimens. The specimens were odorless, dry and flexible compared to the specimens prepared by the S-10 technique. The cost was observed to be lower than that of specimens prepared by the S-10 technique. The synthesized resin was found to be much more appropriate and economical for plastination of specimens.

Conclusion: The synthesized resin UP89 provides an excellent opportunity to preserve and study the different tissues of human and animal specimens. The unsaturated polyester resin (UP89) can be an appropriate material in place of silicone resin for S-10 technique.

Medial aspect of the canine elbow joint: Anatomical study by plastinated sections. *Latorre, Rafael [1], Villamonte Aquilino[2], Soler Marta[2], Sarria Ricardo[1], Gil Francisco[1], Agut Amalia[2]. [1]Department of Anatomy and Compared Pathological Anatomy, [2]Department of Medicine and Surgery, University of Murcia, Spain.*

Excessive local pressures and rotational forces transmitted through the canine elbow joint have been proposed as a possible cause of medial coronoid process disease. The purpose of this study is to assess the relationship of the ligamentous and muscular structures of this region.

Methods: Six cadavers of adult German shepherd cross-breed dogs were used in this study. Synovial and vascular injections were performed in the elbow joint. These joints were destined either for dissection or frozen to obtain sagittal or dorsal cryosections to assess the relationship between myotendinous structures. Sections were then plastinated using the E-12 plastination method.

Results: The tendon of insertion of the biceps brachii muscle divided into two branches: a main cranial branch that inserted on the radial tuberosity and a caudal branch that inserted with an extended fan appearance on the ulna next to the medial coronoid process. The brachialis muscle had a partly fleshy insertion on the cranial branch of the tendon of the biceps brachii muscle and was finally inserted on the ulna in an open fan shape along with the insertion of the biceps brachii muscle. A fibrous sheath formed by the tendon of insertion of the biceps brachii muscle and the reinforcement of the oblique ligament was observed. This leads to the formation of the biceps brachii–brachialis muscles complex whose main point of insertion is the radial tuberosity where it inserts along with the cranial branch of the oblique ligament and the cranial branch of the medial collateral ligament; furthermore the annular ligament covers the radial head attaching from the medial coronoid process to the lateral coronoid process; these features make the myotendinous structures responsible for elbow joint stability.

Conclusion: The insertion of the brachialis muscle on the ulna is mainly related with the medial coronoid process. The section of this tendon could be considered a subject of study as a treatment to diminish the traction and rotational forces during the medial coronoid disease.

Morphological changes of the caudal vena cava under laparoscopic conditions. A study by MRI and plastinated sections. *Latorre, Rafael[1], P_rraga Ester[1], López-Albors Octavio[1], Sarri_ Ricardo[1], Ramirez Gregorio[1], S_nchez-Margallo Francisco[2]. [1]Department of Anatomy and Compared Pathological Anatomy, University of Murcia, [2]Jesús Usón Minimally Invasive Surgery Center, Cáceres, Spain.*

Pneumoperitoneum and patient positioning during laparoscopic surgical procedures cause hemodynamic and anatomical changes in several abdominal organs. Hemodynamic changes in the abdominal portion of the caudal cava vein (CCV) have been described in a pig model, but how the vein morphology and size are affected is unknown. The objective of this study is to assess morphological and morphometrical changes in the CCV of the pig caused by pneumoperitoneum and reverse Trendelenburg position by *in vivo* magnetic resonance imaging (MRI).

Methods: Six pigs were scanned using magnetic resonance imaging under four situations: S1- control (no pneumoperitoneum), S2- control with reverse Trendelenburg position, S3- pneumoperitoneum (14 mmHg). Euthanasia was done after S3 examination and the whole cadavers (pneumoperitoneum maintained) were frozen at -20 °C. Then, transversal body blocks including the abdominal cavity were obtained and frozen at -70 °C. Serial sections of 2-3mm thickness were obtained with a high speed band saw and plastinated by epoxy impregnation according to the E12 protocol. MRI and plastinated body sections were used to evaluate the topography, morphology and cross-sectional area of the CCV.

Results: MRI images and plastinated anatomical sections allowed the differentiation of two morphologically different portions in the CCV, a pre-hepatic portion from the caudal origin of the vein to the liver (T15 vertebra) with flat and irregular morphology, and a hepatic portion, where the vein is surrounded by this organ until it reaches the caval foramen of the diaphragm (T14-T11). Whole-body plastinated sections were used to assist the interpretation of these images and to establish accurate anatomical correspondence with the MRI images. The reverse Trendelenburg position caused an increase in the lumen, affecting mainly the pre-hepatic portion, while pneumoperitoneum caused a decrease in the total vascular lumen, exerting a greater effect on the hepatic portion.

Conclusion: The use of MRI and anatomical plastinated sections demonstrated that both the reverse Trendelenburg position and pneumoperitoneum significantly affected the morphology of the CCV of pigs.

Plastination versus glycerol preservation method in Alizarin red - Alcian blue double staining. *Esfandiary, Ebrahim, Setayesh, Mehr Mohsen, Rabie, Abas Ali, Sayed, Hanaei Mahsa. Isfahan University of Medical Sciences, Isfahan, Iran.*

In this study, the new technique of plastination has replaced the glycerol preservation method, after fetal and tiny samples of skeletal tissues were stained by Alcian blue- Alizarin red staining.

Methods: Rats of four different age groups were chosen and were skinned after death. The specimens were double-stained with Alizarin red & Alcian blue, and then cleared in 1% KOH. Four groups of rats: day 1, day 3, day 12, and mature rats were chosen for the experiment. One rat of each group was selected to be preserved in glycerol, and one of each group was selected to be preserved by plastination.

Results: The quality of staining had an inverse relationship with the age of the rats. The younger the rats, the brighter the staining. The brightness of the plastinated samples also had an inverse relationship with the age of rats. Therefore, the young rats' skeletal tissues were stained clearly and brightly, both in the glycerol and plastination preservation methods. Older rat tissues were dull in both method of preservation.

Conclusion: This study showed that plastination is a good preservation method for fetal and young skeletal tissues, stained by Alizarin red- Alcian blue combined technique. The, plastinated samples were dry, odorless and durable.

The elbow joint of the dog three-dimensionally reconstructed. *Arredondo Jorge[1], López-Albors Octavio[2], Sora Mircea-Constantine[3], Lozanoff Scott[4], Ayala María-Dolores[2], Sarri_ Ricardo[2], Becerril Sigrid[5], Victoria Mauro[5], Latorre, Rafael[2]. [1]Department of Anatomy and Physiology, Autonomous University of the State of Mexico, Mexico. [2]Department of Anatomy and Comparative Pathologic Anatomy, Murcia University, Spain. [3]The Medical University of Vienna, Austria. [4]Department of Anatomy, Biochemistry and Physiology, John A. Burns School of Medicine, Hawai, U.S.A. [5]Academic Research Group in Animal Medicine and Surgery, Autonomous University of the State of Mexico, Mexico.*

The veterinary community is currently discussing how to improve the therapeutic options and understanding of the pathophysiological origins of elbow disorders in the dog. The debate indicates the high level of complexity of this multi-segment joint and it is clear that intensive research is required, not only in the clinical aspects but also on the anatomy and biomechanics of the joint. One way to solve the scientific challenge given by this joint could be to combine *in vitro* and *in vivo* research methods, thereby permitting the design of anatomically realistic two and three-dimensional models. Such methods can provide significant insights into how the neuromuscular and musculoskeletal systems interact to produce movement.

Methods: One elbow joint of a dog was used in this study. The whole forelimb was removed from the cadaver and the axillary artery injected with epoxy resin. It was then frozen at -30°C for 48 hours and a block containing the elbow joint removed. The block was plastinated by epoxy impregnation E12-E1-E600 (Biodur) and then cut into 0.4-0.6 mm thick slices with a contact point diamond band saw (Exakt). The plastinated slices were scanned and the images uploaded into the WinSURF 3D reconstruction software.

Results: The thin plastinated slices provided good anatomical details of the elbow joint. In the 3D model bony structures were particularly well reproduced. Subtraction of specific structures was possible; so all the elements in the model could be displayed in groups or as a whole, as well as rotated in the simulated 3D space. This facility increased the understanding of the anatomy of the elbow joint of the dog and may be useful in assessing surgical or clinical problems in this complex joint.

Conclusion: The deep study of the elbow joint in the dog will be of great relevance for the veterinary clinicians, however, it will be necessary to combine several fields of scientific research and to involve clinicians, biomechanics, physical therapists, anatomists and physiologists. The 3D anatomical model of the elbow joint of the dog obtained from ultrathin plastinated sections is a reliable tool for the study of this joint and could be considered as a useful addition to experimental studies in the investigation of physiopathological and therapeutics options.

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Abdominal cavity of the mouse. A study by ultrathin plastinated slices. *Latorre, Rafael [1], Sora Mircea Constantin[2], López-Albors Octavio[1], Ayala M. Dolores[1], Vazquez Jose María[1]. [1]Department of Anatomy and Compared Pathological Anatomy, University of Murcia, Spain. [2]Plastination Laboratory, Institute of Anatomy, University of Vienna, Austria.*

The purpose of this study was to increase the anatomical knowledge of the normal topography within the abdominal cavity of the adult mouse by means of ultrathin plastinated sections.

Methods: A mouse abdominal cavity was used in this study. After freezing the specimen, a block containing the abdominal cavity was sectioned and plastinated by epoxy impregnation with E12-E1-E600 (Biodur). The epoxy block was then cut in transversel 1 mm thick slices with a contact point diamond blade saw. Both surfaces of the plastinated slices were scanned into a computer using an EPSON GT-10000+ Color Image Scanner. The obtained images were uploaded into 3-D reconstruction software WinSURF (http://www.surfdriver.com) and traced in the monitor manually with a graphic table. The structures used in the reconstruction were the kidneys, spleen and caudal vena cava.

Results: The ultrathin slices provided good anatomical details of the abdominal structures at the macroscopic and submacroscopic levels. The 3D model was useful to understand the spatial relationship between different organs. All reconstructed structures can be displayed in groups or as a whole and could be rotated in 3D space.

Conclusion: Ultrathin plastinated sections provided excellent anatomical details of the different organs of the abdominal cavity in the mouse. 3D modelling is a reliable tool for the study of specific organs and related structures. These results may help in further research and in the diagnosis of experimentally induced pathologies as well as correlation with diagnostic imaging like CT or MRI.

Can Sucrose Prevent Shrinkage in Silicone Brain Plastination? *Baptista, Carlos A. C.,. Parsai, Shireen, University of Toledo, College of Medicine, United States.*

One of the most significant disadvantages of silicone plastination of brain tissue is shrinkage. Sucrose has been used as a cryoprotectant prior to processing in many neurosciences techniques. Our previous experiments included evaluating the efficacy of sucrose treatment in protection of brain tissue during silicone plastination techniques. The results were deemed inconclusive due to the number of variables present in the experimental protocol. The purpose of this study was to reevaluate sucrose treatment as a means of preventing shrinkage in a more controlled environment. In addition to sucrose, we tested DMSO (dimethyl sulfoxide) as a solvent media.

Methods: The experimental groups were as follows: control (no treatment), sucrose treatment, and DMSO treatment. Care was taken to normalize all known variables to only produce variance in initial treatment prior to dehydration. Four brains, fixed in formalin, were sectioned coronaly using a deli slicer producing 1 cm thick slices (samples). Each slice was divided into right and left hemispheres. The right hemispheres were used as control groups and the left hemispheres were used as experimental groups. Therefore each experimental sample could be compared to an equivalent control sample from the same region of the brain. Two different brains were used in each experimental group. The experimental groups were immersed in 10% sucrose and DMSO, respectively, at 5 degrees Celsius overnight prior to dehydration. The control group was immersed in distilled water at 5 degrees overnight prior to dehydration. The surface area of the specimens were measured after each step of plastination using image analysis software.

Results: To determine the correlation between treatment and control, the average percent shrinkage was calculated for each experimental group. Findings are summarized as follows: control group 38.5% shrinkage, sucrose treatment group 31.9% shrinkage, DMSO treatment group 39.4%. The sucrose treatment resulted in the least overall shrinkage. Other notable findings include greatest shrinkage occurring during the curing step. Statistical Analysis: for brain 1 and brain 2, the difference between the DMSO - treated brain and the control was not statistically significant. For brain 3, there was a statistically significant difference between the sucrose-treated brain and the control as determined by one-way ANOVA ($F(1,10) = 7.947$, $P = 0.018$). Finally, for brain 4, there was a statistically significant difference between the sucrose-treated brain and the control as determined by one-way ANOVA ($F(1,22)=11.463$, $P = 0.003$).

Conclusion: Treating brain tissue with 10% sucrose prior to plastination is effective in decreasing shrinkage. DMSO treatment did not prove to be effective in preventing shrinkage.

Comparing properties of specimens before and after chemical treatment for plastination: volume. *Kim, Sang-Hyun, Hong Byung-Ouk, Lee U-Young, Lee Mi-Sun, Han Seung-Ho. The Catholic University of Korea. Seoul, Korea.*

Plastinated specimens have some advantages: elasticity and slight movement, but there are no trials to assess the mechanical properties of the specimens. The aim of this research was to compare the volume between pre- and post-plastinated specimens using a three dimension coordinate measuring machine (VIVID9i).

Methods: Brain, lung, liver, heart and kidney were extracted from one embalmed male cadaver. The specimens were produced by the common S-10 method. The volume of each prepared organ was measured before and after plastination.

Results: Compared to the pre-plastinated specimens, the volume of post-plastinated specimens decreased 19% in the liver, 17% in the brain, 14% in the kidney, 19% in the heart and 22% in the lung.

Conclusion: Volume change is known as a general problem in the S-10 plastination method. This study showed the volume change in several internal organs. Comparing the volume change at each step of the plastination process using 3-dimentional scan is our

future study goal and it could be helpful to reveal in what step of the S10-plastination the volume change occurs.

Plastination and staining of brain slices using two different dehydration methods. *Asadi Mohammad Hossein, Bahadoran Hossein, Azami Abolfazl. Department of Anatomy, Baqiatollah Medical Science University, Iran.*

Unstained formalin-fixed whole brain specimens and brain slices do not give satisfactory results in teaching neuroanatomy. In addition, difficulties in obtaining human brains for dissection have increased the demand for more durable brain specimens which are obtained by plastination. The purpose of this study was to compare two different dehydration methods for brain slice preparation after staining with a modified staining method, followed by the S10 plastination technique.
Methods: Brain slices were fixed and stained using the Mulligan method. Plastination was performed after two different dehydration methods: stepwise using lower concentrations of acetone in room temperature and the standard method. Fading of color and shrinkage of the specimens were measured and compared in both methods.
Results: The stained plastinated brain slices were dry, odorless and durable. Staining of samples gave a clear visual contrast between grey and white matter in brain slices. Stepwise dehydration in room temperature caused no significant loss of color in stained slices compared to the standard method. Shrinkage in the stepwise method was higher than in the freeze substitution method (P value<0.05).
Conclusion: Considering the low cost of stepwise dehydration with used acetone and without needing a freezer, the stepwise method could be used for educational purposes in gross anatomy courses.

Cleaning the Crystallization off the Plastinated Brain Slices. *Alshehry, Murad [1], Ba Abbad Radwan[1], Alobaysi Maher[1], ABAbutaleb Nada[2], Alhamdan Nasser[1]. [1]King Fahad Medical City - Faculty of Medicine. [2]King Fahad Medical City - Hospital Laboratory Microbiology, Saudi Arabia.*

The objective of this study is to share the treatment method of cleaning crystallization from silicone plastinated slices.
Methods: Start with manual brushing of crystallization on plastinated brain slices under running cold tap water. Afterwards, dip the plastinated brain slices in a solution of an industrial surface disinfectant commonly used in Hospitals (Virkon). Furthermore, we dry the plastinated brain slices in a well ventilated area. Afterwards, the brain slices are tested for any microbiological contamination that could damage the plastinate in the future.
Results: This treatment had good results, as none of the brain slices was damaged by the solution or any microbiological growth. However, there were a few left-overs of crystals in some fissures that were colored with the solution's pink color. This minor discoloration could be the only reported disadvantage.
Conclusion: This new method of treatment could disinfect and remove any crystallization or suspected microbiological contamination on damaged brain slices.

Preparation of blood vessel configuration from lung specimens using corrosion casting method with unsaturated polyester resin (UP87) – glass fibers composite. *Setayesh Mehr Mohsen, Rabie Abas Ali, Esfandiari Ebrahim, Dashti Gholam Reza, Karubi Arezu. Department of Anatomic Sciences, Isfahan University of Medical Sciences, Isfahan, Iran.*

The first to apply this classic injection technique in the reproductive arena was John Hunter (1754). As the science was developed and industrial polymers were produced, it was August Schummer about 70 years ago who, for the first time, applied this type of polymer as corrosion casting. The purpose of this study is to prepare vessel configuration using unsaturated polyester resin (UP87) – glass fibers composite synthesized in the plastination laboratory of Esfahan University of Medical Sciences.
Methods: Fresh sheeps' lungs were obtained and the ridge of the trachea, pulmonary veins and pulmonary artery were fastened by a plastic tube. At the beginning, 4% formalin solution was injected into the cavities by a syringe and then the samples was immersed into 4% fixative for about 2 days. The fixative was drained from the cavities and an

appropriate amount of injection mixture consisting of: 100 PBW (part by weight) UP87 resin, 1 PBW glass fibers (diameter: 5-10um, length: 5mm), pigment, 2 PBW catalyst, 0.4 PBW accelerator, was injected into the cavities. The samples remained in the oven for about 12 hours at 40ºC temperature, till the resin was cured. Finally the samples were kept in a concentrated acid bath till the lung tissue was completely removed.

Results: The prepared specimen obtained was found to show the configuration of tracheal tube, bronchioles, pulmonary veins and pulmonary arteries which were manifested very clearly.

Conclusion: Since the available specimens from UP87 resin were brittle and not durable enough, this study was carried out to improve the weak point of the technique. The specimens obtained by (UP87) – glass fibers composite were found to show acceptable appearance and durability for teaching anatomy and museum quality specimens. Therefore (UP87) – glass fibers was found to possess the necessary quality for preparing vessel configuration and corrosion casting method.

Application plastinated specimens for teaching pathology. *Starchik, Dmitry, International Morphological Centre, Saint Petersburg, Russia*

Plastinated specimens have been an integral part of general anatomy courses at medical schools in Russia for more than a decade. However, we still do not see extensive use of plastinates in teaching pathology.

Methods: The International Morphological Centre in Saint Petersburg, Russia, has produced more than 300 plastinated specimens to be used in pathology courses. Pathological material sampling was carried out in the anatomy departments of St. Petersburg hospitals in compliance with the requirements of a pathology department's curriculum, covering all essential pathological bulk specimens. The specimen selection process included taking photographs of a specimen, and tissue and organ fragment sampling for histological study. The selected specimen was dissected again and marked, then it was fixed in formaldehyde solutions with increasing concentration for 2 to 8 weeks. Fixation completed and histological study results obtained, each specimen was then used for producing both a three-dimensional silicone plastinate and transparent plastinated slices. The silicone impregnation technique at room temperature was applied to produce three-dimensional plastinates, and the conventional flat chamber E12 method was used to make transparent plastinated slices.

Results: It has been found that the most efficient way to demonstrate pathological changes in organs is to present three-dimensional plastinates along with transparent plastinated slices. Some pathological states, e.g. recent myocardial infarction or chronic venous congestion, cannot be clearly seen in silicone plastinates after formaldehyde fixation, and thus transparent plastinated slices are the only way to demonstrate such phenomena. Silicone plastination should be applied to produce this type of specimen only after Kaiserling fixation, which preserves the natural color of organs and tissues. Transparent plastinated slices can efficiently be used when studied in transmitted light of a binocular loupe with low magnifying power.

Conclusion: Plastinated specimens have proved to be a highly efficient teaching resource in pathologic anatomy courses, and they undoubtedly have a number of advantages when compared with conventional specimens, because students showed a better understanding of pathological processes. Integrating plastinated specimens in a pathological anatomy curriculum gives more variety in teaching and empowers research techniques.

Application of silicone impregnated brain and brain sections in modern medicine. *Kumar, Rani[1], Pandey, Jayashrip[1], Seith Ashu[2], Lalwani Sanjeev[3], Dhingra, Renu [1]. [1]Department of Anatomy, [2]Department of Radiology, [3]Department of Forensic Medicine, All India Institue of Medical Sciences, New Delhi , India.*

Teaching of neuroanatomy to medical students, neurologist and neurosurgeons requires an in-depth knowledge of the brain and its sections. Even for correct clinical diagnosis of neurological diseases, the right interpretation of CT and MRI by a radiologist is a must. The formalin-embalmed brains and brain slices get easily damaged by repeated handling and the procurement of brain tissue at autopsy involves ethical issues. Therefore charts, models and pictures are being used for teaching purposes, but they do not give a 3D picture. Thus, plastinated specimens of brain and

its sections can serve as the only answer which provides safe, durable, odorless and permanent specimens which can be reconstructed, and which also help in interpreting radiological plates. These specimens also give a clear gross and external view of the structure of the brains and brain slices and can be used for teaching, research and diagnostic purposes.

Methods: A total of 8 human brain specimens used for the study were collected from the Department of Forensic Medicine AIIMS and from bodies donated to the Anatomy Department for teaching and research purposes. The brains were fixed in 10% formalin and plastinated with the standard S-10 technique. Before plastination, coronal and horizontal sections were cut and each section was subjected to various changes of dehydration at -25°C and impregnation both at room temperature as well as -25°C.

Results: The percentage of shrinkage was greater at room temperature (12.39%±1.26 for length and 12.0%±1.49 for breadth) while it was less at -25°C (8.65%±0.89 in length and 8.37%±0.59 in breadth) in both coronal sections and in horizontal sections. Gross observations were made on brain slices to evaluate the level of satisfaction with plastination. The gray and white matter were well-differentiated and the morphology was well-preserved. The internal structure of the sections was distinct and easily identifiable. The nuclei and tracts in the sections were also well-differentiated. Even the blood vessels and *tela choroidea* were recognizable. The S-10 plastinated brain sections showed both external and internal morphology very clearly and looked similar to fresh specimens, although these changes were more distinctly appreciated in specimens impregnated at -25°C than those which were impregnated at room temperature.

Conclusion: These plastinated brains and their coronal and transverse sections can thus be used by neurosurgeons for understanding the normal anatomy and also by radiologists while analyzing CT and MRI images.