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THE JOURNAL OF PLASTINATION

ADDS PJ
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The Journal of Plastination is the official publication of the International Society for Plastination. The journal was first published in 1987 as 'The Journal of the International Society for Plastination"; the shorter title was adopted in 2009. The journal aims to provide a medium for the publication of scientific papers dealing with all aspects of plastination and preservation of biological specimens. The Journal of Plastination, which is published twice-yearly, includes papers on a wide variety of topics, including technical articles on the process of plastination in all its forms, original research using plastinated specimens, or research into novel techniques or applications of plastination, and historical articles on significant figures or techniques from the past. Submissions on the following topics are invited: Original Research – Plastination, Original Research – Preservation, Education, Case Reports, Technical Brief Notes, Review - by invitation only, Legacy - Institutions and People, and Correspondence. In fact, the great majority of published articles are Original Research and Technical Brief Notes. Recent papers have described the applications of plastination in education and research into human as well as animal anatomy, plants, the preservation of archaeological specimens, and the long-term preservation of the bodies of famous people, such as Lenin.
The Journal of Plastination has a global readership. In the last two years, submissions have been received from authors in Bangladesh, Brazil, China, Czech Republic, Namibia, Poland, Russia, Spain, United Kingdom, and the USA. The journal is currently listed in SCOPUS, and an application for inclusion in the Web of Science in under consideration.

DIGITIZATION OF PLASTINATED ANATOMICAL SPECIMENS TO INCREASE AVAILABILITY OF CADAVER-QUALITY LEARNING MATERIAL

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It is becoming increasingly difficult to source fresh cadavers for veterinary anatomy training. Plastination units, whilst growing in popularity are still quite rare. Computer-generated anatomy content is becoming more common, however, many models which are created using medical imaging data such as CT or MRI lack 'realness' and reconstructing particularly small structures such as nerves can be a challenge. Here, we describe a way to increase the availability of high-quality plastinated specimens which are able to demonstrate small structures at a real-life / cadaver level of fidelity, through digitization. This increases availability of cadaver-quality material for teaching. For this project, the specimen used was a previously plastinated canine hemisection of the thorax and abdomen from the University of Murcia Anatomy and Plastination unit. A digital camera (Nikon D5300 with Nikon nikkor 60 mm lens) was used to take approximately 150 photographs from all angles of the specimen using the camera RAW format. These photographs were then processed using Adobe Bridge, after which they were run though the photogrammetry software RealityCapture to generate a

3D model and color texture. This process was done on a Windows 10 laptop with dedicated GPU & 64GB of RAM. Once reconstructed, artefacts from the 3D model were eliminated using the digital sculpting tool Zbrush. A cleaned 3D model and texture sets containing fine surface topology and color information were then exported from Zbrush and taken into Substance Painter - a 3D painting program, which allowed us to add back more realistic colors for the anatomy. For actual teaching usage, the model was placed within the Unity game engine to provide real-time interactivity of the important anatomical landmarks. Additionally, as well as providing real 3D interaction with anatomy, static images could then be generated of the colorized specimen and used within other teaching tools such as flashcards & multiple-choice questions. This content is provided free for veterinary students and academics globally at www.vin.com, and for all others via www.ivalalearn.com as a paid resource.

PLASTINATION OF A CALF HEAD PREPARED WITH ETHANOL-BASED FIXATIVE SOLUTION

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The recommended fixative solution for plastination is formalin. Nevertheless, it is a very toxic reagent. It has been now proven to be carcinogenic for humans, which is why one should avoid its use, or use it as little as possible. A soft embalming solution based on a mixture of ethanol, propylene glycol and benzalkonium chloride was developed in the Laboratory of Animal Anatomy, Faculty of Agrarian Sciences, University of Antioquia. This fixative solution is used for the preservation of all the cadavers for prosections and dissections. The aim of this study was to evaluate the plastination process and the quality of the plastinate produced using this ethanol-based fixative. A calf head from a cadaver was preserved with the solution and dissected to show the superficial (one side) and deep (other side) nerves and vessels. It was dehydrated with isopropyl alcohol, then submerged in three baths (one week each) of acetone, impregnated in S10 mixed with S3, and cured with S6 (Biodur™). The final product preserved tissue elasticity and good appearance. Other organs, like hollow viscera (e.g., intestine) and parenchymal viscera (e.g., liver) preserved with this fixative solution should be plastinated for evaluating the results of the plastination process in other types of tissues. All the animals came from ethical sources by a body donation program, with the approval of the Institutional Committee for Animals Care and Use (CICUA).

PLASTINATION APPLIED TO THE CONSERVATION OF CULTURAL HERITAGE: STUDY OF ITS RELIABILITY ON WATERLOGGED IVORY

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The archaeological excavation of the Phoenician wreck of the Bajo de la Campana, San Javier, Murcia, Spain, was carried out between 2007 and 2011, under a collaboration agreement between the Institute of Nautical Archeology of the Texas A & M University and the Ministry of Culture of Spain, through the National Museum of Underwater Archeology of Cartagena. After successive systematic excavation campaigns, archaeologists

have documented and recovered an extraordinary Phoenician cargo, dated between the 7th-6th centuries BC. Among the raw materials it carried were a magnificent set of 53 elephant tusks and elephant tusk fragments, some of them inscribed. The uniqueness of this archaeological finding is that it is one of the few known examples of Phoenician navigation in the Mediterranean Sea, and the first time that the maritime trade of ivory has been documented as a raw material on the Spanish coast. We selected the Biodur® S15 plastination technique at room temperature as an alternative to traditional impregnation and other consolidation methods that have been ineffective due to the extreme density of ivory. This procedure has been applied to a small representation of elephant tusks (three tusk fragment sections, a tusk fragment and three complete tusks. The fragments were dehydrated with acetone, and subsequently impregnated with a mixture of polymer and catalyst, Biodur® S15 and 1% S3. After impregnation, they were exposed to the crosslinker Biodur® S6, in the curing phase. The time required varied proportionally for different pieces, depending on its size. All the plastinated pieces have been periodically weighed since their processing with the Biodur S15 technique at room temperature. The weight loss in all cases ranged between 2.2% and 11.4%, far from the 35.1% to 57% recorded for the untreated fragments used as control. In the first years after plastination, the weight loss was significant, but it has become less over time; we have even registered slight increases (tusk fragment sections and tusk fragment). The results obtained were satisfactory, both in dimensional stability and in visual aspects, essential for the study and exhibition of these pieces that are part of our cultural heritage.

EXPERIENCES USING RED MUSCLE PIGMENT (RMP) ON ANIMAL SPECIMENS SUITABLE FOR PLASTINATION, WET PRESERVATION AND DRYING OF LUNGS

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Early in the development of anatomical techniques, pigments have played an important role to stain products that can then be injected into vessels or ducts. For plastination, the Biodur AC10® stain was developed to add color to specimens. A commercial Red Muscle Pigment (RMP) has been tested to stain cadavers. Animal skeletal muscle and fresh lungs were used to determine the best concentrations to use for staining. Nevertheless, the product can be used directly on the specimens; our results showed that 50% of the pigment created a good skeletal muscle color; 15 to 10% dilution is good to stain smooth muscles or mucosa; 10 to 5% is a good dilution to stain fresh lungs when they are still moist and ready to be dried using an air pump. The pigment can be diluted using tap water, and also works really well in other tissues like heart, placenta, gills, brain and skin (mammals & mollusks). The RMP is acetone resistant, making this product suitable to be used in the plastination process. The product is easy to be manipulated, and the concentration can be modified according to the visual taste of the anatomist. The best way to stain anatomical structures is painting using a brush. One disadvantage is that stain spread to adjacent tissues in wet specimens. Commercial hypochlorite can be used to clear the surfaces/tissues that were accidentally pigmented. Another issue could be that after a year you may need to repaint wet specimens that have been manipulated for numerous students if you want to bring the bright color back.

VALIDATION OF AN S10 PROTOCOL FOR REPTILE FULL BODY PLASTINATION

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Human activity and climate change have contributed to the loss of species. Live animal collections have been established to protect endangered wild animals. When wild animals die in captivity, it represents a great loss for its species, and best use of those bodies must be made. Collections for further research can be made by body conservation. Plastination has great advantages compared with other methods but has the disadvantage of specimen shrinkage. In 1986, the Heidelberg Plastination Folder was published; it describes several plastination protocols in different species, but reptiles are not included. Fifteen indeterminate-time-frozen reptile bodies (3 snakes, 7 lizards and 5 turtles) underwent a plastination protocol. Bodies were thawed for 24 h at 5° C, and 12 h at room temperature. Fixation: once unfrozen, bodies were formalin fixed for 48 h. Immersion, injection and infiltration of formalin was carried out, and needles were inserted in the skin of non-visible structures for better penetration. Once fixed, specimens were rinsed with running water and then remained for 24 h at 5° C. Dehydration I**:** 3 x 1-week long -25° C pure acetone baths were made. On the last bath, acetone concentrations were measured every third day. Dehydration was considered complete when 2 consecutive measurements were the same (98.5%). Dehydration II: specimens were immersed for 5 days at room temperature in a 98% acetone bath for defatting. Impregnation: needles were inserted in the skin of non-visible structures. Specimens were immersed in a Biodur™ S10+S3 silicone mixture for 3 days at -25° C with no vacuum, then standard forced impregnation was performed. Specimens remained in the impregnation bath for 2 weeks before opening the chamber to release the vacuum. Precuring: the specimens were left for 48 h at 5° C to drain excess silicone; specimens were then positioned. Curing: at room temperature with Biodur™ S6, 4-10 days, specimens were then wrapped with plastic film. Body measurements were performed before and after plastination to assess shrinkage. Student's t-test was performed on *SAS*™ software with no significant differences (P>0.05), other than in "curve width of the shell" of a turtle specimen. In conclusion, 15 reptiles were successfully plastinated, validating that the modifications to the standard protocol were adequate, with no significant shrinkage.

COLLABORATIVE ONLINE ANATOMY EDUCATION USING HEAD AND NECK PLASTINATES

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Collaborative online anatomy education has become increasingly utilized due to a trend toward student-centered independent learning, as well as the ongoing Covid pandemic limiting in-person group activities. Anatomy education is heavily reliant on visuo-tactile experience, and presents a challenge during online instruction. In an effort to provide an effective experience, the Head and Neck block in Gross Anatomy (Fall Semester, 2020) was presented as an online activity that included plastinated anatomical dissection models

posted on the *Sketchfab* platform. The purpose of this study was to assess the usefulness of plastinated specimens as part of a workflow enabling extended reality (XR) presentations within the context of online gross anatomy education. A photogrammetry workflow was developed to digitize the dissected specimens that were posted to the *sketchfab.com* platform and presented via a university-based website hosting service (*xrcore.jabsom.hawaii.edu*). A comparison of the perceived usefulness of the actual dissections, dissection models, artistic models, and segmentations models was conducted using student surveys (n=79). Plastination models received an average of 144 views. When compared to all learning resources, actual dissections were most preferred (34.1%). However, *Sketchfab* dissection models were considered most/more preferred (54.3%) compared to other assets, suggesting a broader preference as a learning resource. These results suggest that plastinates provide useful learning resources for online gross anatomy learning.

PLASTINATION OF THE SPERM WHALE

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This study was approved by the National Forestry and Grassland Administration of China. Plastination has become the gold standard for the preservation of anatomical specimens by replacing tissue fluid with a curable polymer. The longevity of plastinates is advantageous for the preservation of biological tissue, but especially for rare or unique specimens of inherent scientific and educational interest. Such was the case with the world's first plastination of sperm whales. In 2016, two adult male sperm whales were stranded on the beach of Yangkou Port in Nantong City, Jiangsu Province, China. The local government planned to preserve two sperm whales by making specimens, one of which was entrusted to Dalian Hongfeng Biological Co., Ltd., the first sperm whale to be plastinated and preserved in world history. The resulting plastinate of this large marine mammal shows the mutual adaptability of its internal structure, living environment, and living habits. The plastination process also provides a new method for studying the anatomy of large animals in the future. The plastinated sperm whale promises to be an enduring asset of tremendous scientific, educational, and artistic value.

THE HOFFEN P45 TECHNIQUE AND ITS APPLICATIONS IN CLINICAL ANATOMY

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The P45 plastination technique can be applied in order to clearly show the internal structure of specimens. Because of its extraordinary properties, the accurate anatomical structure of the specimen can be seen on a large scale. In recent years, using the P45 technique, we found that several of the deep suboccipital muscles, which span from the cranial-most vertebrae to the caudal surface of the skull, also have direct attachments on to the dura. It is now named the myodural bridge. It provides a new direction for the study of cerebrospinal fluid circulation. Also, in the field of plastic surgery, we have determined that the retro-orbicularis oculi fat is composed of fascial adipose tissue, by investigating thirty-eight Chinese hemifacial P45 specimens. Its relationship with the surrounding tissues was also studied. We were therefore able to improve the

intraoperative management of patients who underwent upper eyelid surgery in the plastic surgery department of the First Affiliated Hospital of Dalian Medical University in 2019. Furthermore, formalin-fixed specimens of 27 adult knee joints which were normal in imaging examination, were selected for plastination with P45. We have discovered that the bone cortex at the attachment site of the cruciate ligament was slightly thicker than the surrounding cortex, and that the bone trabeculae deep to the cortex were thick and dense. The trabecular bone at the site of the femoral attachment was mainly radially enhanced, while the tibial attachment was mainly enhanced in the direction of, and vertical to, the ligament. In addition, the popliteus muscle was connected to the PCL, ACL, lateral meniscus, and posterior meniscofemoral ligament via the dense connective tissues near its tendon-muscle transition. Finally, a cluster of dense, thick, longitudinal bone trabeculae in the posterior lateral part of the tibial plateau was found, which terminated in the posterior lateral cortex of the fibula and tibial shaft, to form an arch beam structure, with the fibula acting as the mechanical fulcrum of the arch beam. Thus, it provides further accurate anatomical information of the knee joint and provides a new vision for the clinical diagnosis and treatment of knee joint diseases, especially knee osteoarthritis. This study was approved by the Ethics Committee of Dalian Medical University.

A RARE OPPORTUNITY TO PLASTINATE AND EXAMINE THE HEART OF THE ENDANGERED SOUTHERN RESIDENT KILLER WHALE (*ORCINUS ORCA*)

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The importance of high quality and durable anatomical specimens is well-known to academic institutions, including schools of human and veterinary medicine. However, such specimens are also important to museum communities which endeavor to engage the public and communicate specific natural history content. The killer whale (*Orcinus orca*) is the second largest toothed whale species, with male killer whales exceeding 5 metric tons. There are few well-preserved coronary specimens representing different whale species, and while hearts of the *Odonticeti* are the most commonly described, little is known of killer whale coronary anatomy. The death in 2016 of an endangered, adult male southern resident killer whale (SRKW) afforded a rare opportunity to plastinate an intact killer whale heart, using the standard S10-S3, cold impregnation, plastination technique. We present the process and technical challenges of plastinating such a large, hollow organ, as well as subsequent examinations. Results yielded a specimen of exceptional anatomical detail, amenable to highly resolved endoscopic and computerized tomography evaluation. Museum exhibited in a comparative context, the plastinated SRKW heart assures enduring value as an object for education, and a specimen of scientific interest.
Ethics Statement: The whale heart specimen was collected postmortem during necropsy and transported under permit with the Departments of Fisheries and Oceans (DFO) Canada, a Canadian letter of possession, as well as endangered species transport permits for both ports of exit and entry (CITES export permit 16CA02387/CWHQ, CITES import permit ES-LA-00010/161).

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WHY DOES THE ACETATE FOIL STICK TO CURED EPOXY SANDWICHED SLICES? RECOMENDATIONS TO MINIMIZE THE PROBLEM

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An occasional problem in epoxy plastination during the curing by the so-called sandwich method is the difficulty or impossibility of separating the acetate foil from the cured specimen. In those cases, slices lose transparency and have a reduced potential for detailed anatomical interpretation. No previous experiments have related this problem with either the age of the chemicals or the age of the acetate foil. In this work, 120 epoxy impregnated slices from dog thoracic limbs were cast using 4 different combinations of the E12 resin plus E1 hardener mixture, both in fresh (1 year after purchase) or old (4 years) conditions. Those groups were named as E12freshE1fresh, E12freshE1old, E12oldE1fresh and E12oldE1old. Then, the 30 slices of each group were sandwiched with 5 different types of acetate foil (6 slices per group). AcB1 (Biodur® 1 year old), AcB2 (Biodur® 2 years), AcB3 (Biodur® 3 years), AcB4 (Biodur® 4 years) and Ac3M (3M® 1 year old). After impregnation and curing for 3 days at 40 ºC, the acetate's resistance to removal from the slices was rated as 1 (maximum resistance, do no separate at all), 2 (partly separated), 3 (mostly separated), 4 (no resistance, complete removal). The results showed a direct influence of the age of the E12 resin and the age of the acetate on the ability to remove the acetate after curing. Thus, specimens cast with E12 old resin and with acetates from groups AcB2, AcB3 and AcB4 showed significantly lower rates of success while separating the acetates from specimen. On the other hand, no effect of the age of the E1 was observed on the results. Hence, to minimize the problem, the use of fresh E12 resin and new acetates is recommended.

MICROCHIP TAGS, A USEFUL TOOL TO FACILITATE THE TRACEABILITY OF S10, P40 AND E12 SAMPLES

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A current problem in plastination labs is related to the difficulty of maintaining the traceability of the specimens during the plastination process. For this purpose, different strategies such as labels or magnetic tags are commonly used. Since specimens are most of the time immersed in different liquids (fixative, acetone, polymer) and processed in batches containing a variable number of specimens from different background and characteristics, the use of magnetic tags might be particularly useful. In this work, we aimed to validate a particular type of magnetic tag (common pet microchip) as an efficient way to maintain the traceability of the specimens during and after the silicone (S10), polyester (P40) and epoxy (E12) plastination techniques. With regards to the silicone technique, 10 cow whole brains were tagged at the level of the medulla oblongata during fixation and kept in place throughout the plastination process. Systematic reading of tags was done at the end of each plastination step. Tags were also incorporated into polyester-filled flat chambers, including 10 cow brain sections, and to the curing E12-E1 mixture of 10 dogs stifle sandwiched sections. It is remarkable that the reading of the tags was satisfactory at all checking steps and for the three

plastination techniques. The tags implanted in the medulla oblongata proved to be functional while the specimens were immersed in 10% formalin, as well as during all the dehydration steps, impregnation with S10-S3 mixture, and curing with S6 vapor. Likewise, the reading was correct after the curing of the polyester flat chambers and the epoxy sandwiches. Furthermore, tags were stable and functional a long time after the end of plastination. It is concluded that the type of tag used in this study is a valid and helpful strategy to maintain the traceability of the specimens, not only during plastination but also after it, which is important when evaluating the long-term quality of specimens.

AESTHETIC ANALYSIS OF PAINT IN PLASTINATION

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Aesthetics is a branch of philosophy that analyses from the standpoint of sensation and feelings, the "artistic or not" production of human sensitivity, and therefore, art is not restricted to the ability to provoke these aesthetic responses. In line with the concept of art-anatomy, there is an overlapping relationship from the Renaissance times to contemporary, between body, art, anatomy, death and, consequently, the exposition of the corpse. The latter, if plastinated, offers the spectator a feeling that not only permeates the teaching-learning of anatomy, but also offers an aesthetic appeal, by which, color becomes a fundamental element of presentation. Considering the aspects and the importance of the aesthetic appearance of the anatomical plastinated specimens presented in exhibitions, museums, or educational institution, as well as the paucity of information about materials and painting methods of plastinated specimens in the scientific literature, we aim to perform an artistic and aesthetic analysis about these materials and their application to a satisfactory result in the final appearance of the plastinated human specimen. It is noted that the use of industrial paints, or inadequate materials, sometimes does not correspond to the approximated natural color, acting as shades that highlight the differences of tissues in their anatomical constitution, however, moving away from a more natural or realistic aesthetic presentation. To provide a basis for further analysis, an ink was formulated, using natural inorganic pigments, that could be easily applied to the silicone. The results showed good ink adhesion and integration to the room-temperature impregnated human specimen, applied before and after curing. This study concludes that the color and mode of presentation of the anatomical plastinated specimen has become an improving device for the aesthetic appreciation and for the diffusion of Art-Anatomy and Anatomical Science education. All documentation for receipt of unclaimed or donated bodies is regulated, according to Federal Law No. 8,501 (November 30, 1992), that authorizes the use of human bodies for teaching and research purposes.

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PLASTINATION OF MARINE INVERTEBRATES: A PROTOCOL WITH ROOM TEMPERATURE AND LOW VISCOSITY SILICONE

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The invertebrate group is the most diverse in the *Animalia* kingdom; it is estimated that about 90% of all animal species are part of this paraphyletic group. Although widely known and used in vertebrate conservation, no research has been found in the literature that apply the plastination technique exclusively to taxa of marine invertebrates. Thus, the development of a protocol adapted for these specimens could bring a gain in the use of this technique for these animals, especially as this group presents individuals with the most diverse shapes and morphological characteristics, ensuring a minor tissue retraction and morphological alterations. A total of 94 specimens from the phylum Mollusca, Crustacea, Echinodermata, Annelida, Cnidaria, Porifera and Chordata were selected. The animals were initially anesthetized and then euthanized with magnesium chloride previously diluted in sea water (75 g/L), and then plastinated, following the four steps: fixation in formalin (10%), dehydration in acetone at low temperatures (-25° C), forced impregnation with low viscosity silicone at room temperature, and hardening. Changes in the protocol sought to reverse the high degree of tissue retraction in the soft-bodied specimens used in this work, such as mollusks. Results showed that the plastinates retained the properties of a plastinated specimen, such as absence of toxicity, lack of odor, and a dry appearance, showing little alteration in their initial body shape. However, some specimens require a greater care in handling because these ones suffered a fragilization in their appendages, mainly arthropods. Regarding color, most specimens presented changes following fixation and dehydration, requiring less time in these steps. The plastination of these animals constituted the largest collection of plastinated invertebrate specimens in Brazil, showing different application possibilities to be used in diverse areas, such as practical classes, exhibitions, and in the constitution of a biological collection, expanding the horizons of plastination beyond the anatomy of vertebrates, and ensuring the dissemination of the technique. Although only a qualitative analysis, the excellent aesthetic results obtained in this study proved that plastination of marine invertebrates, including small animals, and those with a higher percentage of water, is feasible and achievable.

FIRST SLICED AND PLASTINATED FULL HUMAN BODY IN LATIN AMERICA: A PIONEERING EFFORT OF THE LIFE SCIENCES MUSEUM

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The Life Sciences Museum of the Federal University of Espírito Santo (MCV-UFES) has in its collection replicas of human and animal skeletons of the highest quality, as well as real specimens preserved by different techniques, mainly plastination. In order to expand the MCV collection, the goal of this work was to plastinate a sectioned full human body – a huge, bold and pioneering plastination work in Latin America. For this purpose, a male human body was used, 1.65 m in height and aged between 60-65-years-old. The following method was used: prior fixation in 10% formalin, freezing at -25° C, polyurethane packing, cutting with a bandsaw into axial sections 13-15 mm thick, dehydration at cold temperature (-25° C) with four acetone baths (95, 95, 100 and 100%), impregnation with polydimethylsiloxane (PDMS) silicone and 8% DBTL catalyst at low

temperature (-25° C) and maximum vacuum of 8 mm Hg lasting 26 days, and cured with TES crosslinker vapor for 2 days. The 190 cross-sections, numbered skull-caudally, were made in the horizontal plane, except for the hands (one sectioned in coronal and the other in sagittal planes), and one of the feet that was sagittally sectioned. The complete set of horizontal slices was organized in two groups: 1 - even slices, designated to be used in practical anatomy classes for 11 courses in the health and biomedical sciences at UFES; 2 - odd slices, used to assemble the full body specimen, similar to a tomographic display. This set of slices, comprising 80 sections in 37 skull-caudal planes was mounted with a spacing of approximately 10 cm gaps between the slices, resulting in a 4-metre-long man. Since its inception, the specimen remains on display for visitors from schools, the academic community, and the general public. The display of this unique specimen allows for a better understanding of the human body and tomographic perspective. This work follows the unclaimed and body donation regulations, according to The Federal Law No. 8,501 (November 30, 1992), that authorizes its use for teaching and research purposes. **Grant Support**: PROEX-UFES

CONTRIBUTIONS OF THE LABORATORY OF PLASTINATION AND ANATOMICAL TECHNIQUES, UNIVERSIDAD DE LA FRONTERA (CHILE), IN RESEARCH AND POSTGRADUATE TRAINING ON PLASTINATION TECHNIQUES

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Plastination was created by Gunther von Hagens in 1977, in Heidelberg, Germany. Since its creation, its implementation has spread mainly in Europe, Asia, and the United States, reaching, our region of South America by the end of the 20th century. Since 2014, we have made innovative contributions from the point of view of developing new plastination technique protocols, especially a new plastination technique at room temperature, with the development of the concepts of active-passive forced impregnation, as well as a new way to combine the impregnation mix at room temperature. Likewise, new sheet plastination techniques with epoxy resin were developed, using local resins, and modifications to the classic protocols developed by von Hagens. On this same topic, the first review on the E12 plastination technique was developed. Articles have also been published for the first time, in which plastination is applied in experimental morphology protocols, in animals in which diseases were generated, and these were identified through sheet plastination, developing from this the concept of microplastination. Also, a very important part of the research lines of our laboratory is the analysis of DNA in plastinated samples, both animal and human, managing to verify for the first time the presence of whole and intact DNA in plastinated samples. In addition, we carry out plastination of a diverse range of specimens, both human and animal, with the aim of developing an Anatomical Museum in our institution. As well as this, all the experience shared through scientific publications is used to provide advanced courses and diploma courses for undergraduate and postgraduate students, seeking to further disseminate plastination and anatomical techniques, for use in teaching and research. In this work, we show the Laboratory of Plastination and Anatomical Techniques, of Universidad de La Frontera, Chile, from the point of view of the creation of the laboratory, its start-up, and the scientific productivity derived from it, as well as postgraduate training activities. All the investigations carried out in plastination, both in human and animal bodies, have the approval of the Scientific Ethics Committee of Universidad de La Frontera, Temuco, Chile.

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"FOREST INHABITANTS": EXHIBITION OF PLASTINATED WILD ANIMALS FROM THE ATLANTIC FOREST IN THE LIFE SCIENCES MUSEUM

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The Atlantic Forest is one of the most threatened biomes on the planet, being classified among the 34 global biodiversity hotspots. The federal highways that cross these regions are the main cause impacting the survival of the wild animals. There is no report in the literature on the use of plastination to preserve collections of Atlantic Forest wild animals. The aim of this work was to plastinate the deceased animals, enabling environmental education, the development of the exhibition "Forest Inhabitants", and the creation of a virtual teacher training named "Know to Preserve", developed at the Life Sciences Museum (LSM) of the Federal University of Espírito Santo (UFES) and conceived for environmental education. For this work, carcasses of animals that had been run over were used, which were collected on a federal highway that crosses the "Sooretama Biological Reserve" and the "Vale Natural Reserve", in the Espírito Santo State, Brazil. The animal carcasses used were: *Cerdocyon thous, Tapirus terrestre, Leopardus wiedii, Procyon cancrivorus, Bradypus variegatus, Nasua nasua, Cuniculus paca, Picumnus cirratus, Salvator merianae, Porphyrio martinicus, Eupsittula aurea, Micrurus corallinus* and *Boa constrictor.* The plastination was divided into four stages: fixation in 10% formalin, dehydration with acetone at room temperature, forced impregnation with silicone at room and low temperatures, depending on the type of fur or plumage, and curing. After fixation, some specimens were dissected for different educational purposes. After plastination, the animals were placed on display at the LSM, being arranged according to the classes represented: amphibians, reptiles, birds, and mammals. The animals became more durable, lighter, and easier to move. Their anatomical characteristics have not undergone major changes, and the coat or plumage has remained very natural. This material will remain exhibited to the public, free of charge, and is available for research and education, and to compose biological collections. This unique collection of wild animals of the Atlantic Forest was made possible by the pioneering efforts of the LSM and the Plastination Laboratory of UFES for plastinating the collection. The use of the wild animals was authorized by the ethics committee of the institution. ***Grant Support***: PROEX-UFES

IMPLEMENTATION OF THE ISO 9001 QUALITY CONTROL SYSTEM IN A PLASTINATION LABORATORY

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The identification of specimens and their traceability are essential parts of the production process in a plastination laboratory. Also, during all steps of plastination, different sources of variability related to the handling of the specimens and equipment make it difficult to obtain regular standardized products of optimum quality. The implementation of an ISO 9001 quality management system (QMS), according to the International Organization for Standardization (ISO), is a powerful tool to provide a series of benefits in order

to increase the performance of internal procedures. This is made possible by an internal control of the equipment that is part of the routine in a plastination laboratory, as well as the implementation of standard operating procedures (SOPs) at all activity levels. An example is the possibility of keeping a meticulous control of the pressure inside the vacuum chambers where the impregnation takes place. However, it is common for doubts to arise as to whether the value shown by the digital manometer during the vacuum process is reliable. Therefore, it is not enough just to check that the equipment indicates the values we expect to obtain. Being able to verify that the pressure inside the vacuum chamber is adequate is only possible if we have previously calibrated standard equipment, one of the premises of ISO 9001 QMS. This would be the only way to be sure that the process is on the right track. Moreover, the fact that all the procedures are controlled by SOPs allows the early identification of errors derived from each of the processes. This translates into greater control of unsatisfactory and non-conformities derived from daily work, and what is more important: being able to detect them, investigate and delve into the possible cause, and seek effective solutions over time.

ENSURING EFFECTIVE ANATOMY TEACHING DURING THE PANDEMIC: USE OF PLASTINATED SPECIMENS AS A RESOURCE

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An array of new measures has been introduced to cope with the challenges imposed by the Covid-19 pandemic. Such challenges included shifting from face-to-face lectures to online sessions, and shortened dissection time. New measures included, among other things, the use of plastinated specimens as a resource, and online 3D learning modules. A survey administered to assess students' satisfaction with the new measures showed a general agreement that the measures have been useful in enhancing understanding and facilitating a better performance. New measures introduced to the first-year medical gross anatomy course to cope with the pandemic restrictions included: weekly online self-assessment questions and image identification questions; *Zoom*-based lectures with 3-4 polling questions; focused lab dissection sessions using complete personal protective equipment (PPE); use of prosected and plastinated specimens; peer presentation/evaluation; and weekly faculty-driven online reviews of clinical anatomy questions and discussion. The plastinated specimens were prepared locally using the room-temperature technique. Neurovascular pathways were painted to facilitate pattern recognition and better understanding. Students' performance in the online self-assessment modules was closely monitored on *Canvas* and compared with their grades in the weekly quizzes. An anonymous survey was administered to assess students' satisfaction. The overall anatomy grades average was 84% (±9.7), 3% higher than last year's; 82% of the students agreed that the new teaching strategies helped them understand anatomical concepts better, 77% appreciated the use of clinical relevance, improving performance in 72% of the class. The higher average grade may be attributed to the wider range of measures introduced and the intense monitoring and follow up of students' performance. The *Canvas*-based self-assessment modules proved to be effective in stimulating students' engagement and comprehension. Plastinated specimens made an excellent resource to students when lab dissection time was shortened. The new measures have been effective in ensuring better performance and satisfaction despite the curricular challenges faced during the pandemic. Ethics Statement: All plastinated specimens were obtained from a legitimate donation program.

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EXPLOSION PREVENTION IN PLASTINATION

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In this presentation, an overview of the basic information about explosion prevention related to plastination work is given. The presentation is structured in four sections: Occurrence of an Explosion Hazard, Flammable Liquids, The Proper Equipment, and Working Safely. Explosion hazard develops when a mixture of air and vapor of a flammable liquid forms, and a potential source of ignition gets in contact with this mixture. Furthermore, the temperature must be approximately equal to or above the liquid's flash point. There are a number of different potential ignition sources, including electrical appliances, static charge, or hot surfaces. Characteristics of some selected flammable liquids are introduced. Acetone which is frequently used for dehydration and defatting of specimens has a low flash point of ca. -20° C (-4° F). Therefore, while working with acetone explosion protection must always be observed. Besides the flash point the upper and lower explosion limits of the liquid indicating the minimum and maximum vapor concentration in air allowing an explosion are of importance. Electrical devices classified explosion-proof, like fan motors or conveyor pumps, are available in three categories, depending on the level of protection. Some examples of explosion-proof pieces of equipment typically used in plastination are given. A safe work environment is based on carrying out risk assessments and determining areas exposed to explosion hazards. In addition, organizational measures like periodic instruction of the staff involved in plastination decreases the risk of human error which might lead to dangerous situations.

ATLAS OF HUMAN BODY CROSS-SECTIONS PLASTINATED WITH E12 TECHNIQUE

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The great Russian surgeon Professor Nicolai Pirogov was the first to propose cutting sections of a frozen human body with a bandsaw for studying anatomy. He published the first Atlas of frozen body cross-sections and created the new discipline in medical education of Topographical Anatomy. However, only large anatomical structures could be seen on Pirogov's sections. Due to the lack of photography at that time, it was necessary to draw the specimens within a few days, otherwise the anatomical material would quickly dry out and decompose. The invention of epoxy plastination by Gunther von Hagens permitted the visualization of small anatomical structures in transparent sections. Von Hagens, in collaboration with Professors H. Ross, L.J. Romrell and K. Tiedemann, published the first Atlas of plastinated sections "Farbatlas der Schnittanatomie" (Color Atlas of Sectional Anatomy) in 1991. The Atlas included 127 photographs of epoxy plastinated human body cross-sections. The development of microsurgery in the third millennium required more detailed knowledge of small anatomical structures in various areas of the human body. Advances in digital technology have made it possible to scan sheet plastinated slices at high resolution. Therefore, since 2015, we started creating a new Atlas of human body cross-sections using epoxy plastination. It was produced from more than 1000 sections, 2-5 mm thick, of 8 bodies, donated to the University according to the law of the Russian Federation. We used the standard E12 technique for dehydrating, degreasing, impregnating of epoxy resin, and casting. Resulting specimens were scanned on an office scanner with a resolution from 600-1200 pixels

per inch. The Atlas contains 160 color scanned images of transparent sections of all areas of the body in the horizontal, frontal, and sagittal planes. All anatomical structures are well visualized in the presented images, with a magnification of up to 20x. The images contain official names of the anatomical structures in Latin. All drawings are supplemented with diagrams showing the level of the section in each body area. The Atlas can be used as an illustrated textbook for the study of topographical Human Anatomy for students, anatomists, surgeons, and radiologists.

ACETONE RECYCLING: DO MOLECULAR SIEVES CAUSE BREAKDOWN TO ACETALDEHYDE?

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Plastination is a process that is used to preserve biological tissue. In the plastination process, one of the major steps involves dehydrating the tissue with acetone. During this process, large amounts of acetone are used. In order to reduce lab costs, acetone is routinely recycled. This process uses a commercial acetone recycler to remove the majority of impurities that are present in acetone after the dehydration process. Once recycled, the acetone is exposed to molecular sieves to remove water from the acetone. Molecular sieves work by binding to the smaller molecules (water) as the acetone molecules are exposed to them. When exposed to the sieves, the recycled acetone can reach a purity level approaching 100%. The use of molecular sieves in acetone recycling is thought to change the chemical makeup of acetone to acetaldehyde. Acetaldehyde is more reactive than acetone because it has less steric hindrance (physical presence of a larger molecule that slows down or prevents reactions) since the methyl group is larger than the hydrogen. Acetaldehyde has a lower melting point (-120° C) and boiling point (20° C) than acetone (MP -95° C and BP 56° C). Because of this phenomenon, it has been recommended that molecular sieves should not be used to recycle acetone. To observe the veracity of this assertion, we utilized NMR (nuclear magnetic resonance) to analyze the recycled acetone for the presence of acetaldehyde. The samples used included pure (new) acetone, used acetone before recycling, used acetone after recycling, and used acetone after molecular sieves. From the preliminary and subsequent NMR spectra studies, there were no aldehyde groups present in any of the sample groups. Based on our NMR results, the use of molecular sieves does not appear to alter the chemical makeup of the acetone during the recycling and purification process. Since the acetone is not affected by this process, we conclude that the use of molecular sieves is an effective and cost-saving method of acetone recycling for its use in the plastination process. Ethics: no cadaveric tissues or student subjects were used in this study.

SOCIAL MEDIA IN ANATOMY INSTRUCTION: WHAT, HOW MUCH, TO WHOM?

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Modern-day anatomy educators should keep themselves up to date with the learning style of their students; otherwise, they face the risk of failing in the interaction with them. For this reason, they need to make consistent teaching interventions. Today's students are already using social media in their learning experiences. The use of educational social media accounts successfully increases the engagement, motivation,

and examination self-confidence of students in anatomy education. This suggests that students are willing to become members of a community of practice using a social media network. On the other hand, having their educators' presence on social media, they perceive to be in their private personal space, and this may contribute to student interaction, collaboration, and participation. Social media not only changed the way that individuals interact with each other, but also changed the way we shop, eat, communicate, and even learn. Being so popular and powerful, it is like a double-edged sword with its considerable benefits and many challenges. In regards of anatomy, social media enable anatomists all over the world to engage, interact, and form new collaborations in a virtual community that otherwise would not have been possible. The students as well as members of the public see the content posted on an anatomy education social media account. Posting appropriate content is one of the challenges raised by the use of social media in anatomy. Human cadaveric material is frequently shared on social media and there is divided opinion among anatomists on whether or not such content is appropriate.

ULTRASOUND OBSERVATION OF THE LATERAL FEMORAL CUTANEOUS NERVE AT THE ANTERIOR SUPERIOR ILIAC SPINE AREA

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Meralgia paresthetica is characterized by symptoms of pain, numbness, or itching at the anterolateral thigh, commonly caused by mechanical entrapment of the lateral femoral cutaneous nerve (LFCN). The mechanical entrapment site is believed to be at the point where the nerve exits the pelvis around the anterior superior iliac spine (ASIS). The aim of this study was to reveal the fascial configuration around the LFCN, particularly its sonographic features, at the ASIS area. Six cadavers (2 females, 6 males; age range 46-87 years) were used for plastination (2 transverse and 4 sagittal sets of slices) and confocal microscopy. The morphological sonographic features of the LFCN and its surrounding structures were evaluated in 34 healthy volunteers. The results showed that (1) the LFCN exited the pelvis via a tendinous canal within the internal oblique-iliac fascia septum and then ran in an adipose compartment between the sartorius and iliolata ligaments inferior to the ASIS; (2) there were 3-4 iliolata ligaments and the LFCN pierced the medial 2-3 ligaments and then ran in a longitudinal ligamental canal bordered by the iliolata ligaments; (3) under ultrasound, the LFCN, ASIS, iliolata ligaments, and ligamental canal were easily detectable. This study, thus, concluded that ultrasound scanning from the upper thigh to the pelvis is recommended for localizing the LFCN. This study was approved by the Human Research Ethics Committee at the University of Otago (H18/027) and Anhui Medical University (20190532).

EPOXY SHEET PLASTINATION AND MORPHOLOGICAL RESEARCH

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The basic principle of epoxy sheet plastination is to replace water and lipids of a biological specimen with curable epoxy resin. The size of the plastinated specimen can be up to the whole body but the plastinated

structures can be visualized at a cellular or even subcellular level. Of morphological research techniques, epoxy sheet plastination technology has two unique features: (1) examination of the interface *in situ* between hard and soft tissues without decalcification and dissection, and (2) macro-microscopic examination on the same slice. It can combine with various pre-treatments (e.g., coloured vascular casting) and post-treatments (e.g., various histochemistry staining, confocal microscopy). In this lecture, two examples of its applications in clinical anatomy research will be presented. One is to demonstrate how we reveal the structural mechanism underlying the new finding that intracranial and intraorbital subarachnoid spaces are not continuous in the optic canal. Another is the establishment of a 3-dimensional somatotopic map of the trigeminal ganglion, which is essential for neurosurgeons to guide the precise positioning of the needle tip during percutaneous trigeminal rhizotomy for patients with trigeminal neuralgia. These two examples will strongly indicate that epoxy sheet plastination is a revolutionary powerful morphological research tool. All the studies presented in the lecture were approved by the Human Research Ethics Committee in University of Otago.