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Preparation of Wet Specimens in Anatomy Museum: A Practical Approach to Overcome Difficulties

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Abstract

Introduction: Museum is an essential part of Anatomy department in a medical college. Museums are in part historical, representing the premium work of that department which gives complete information and knowledge of the human body in its four walls. The aim of present study is to procure, fix, prepare and mount the specimens in consistent manner and compatible with the standards for a good Anatomy museum.

Methods: Perspex jar, embalmed cadavers and soft parts were used to prepare museum specimens. The procured specimens were fixed and mounted using Kaiserling solution.

Results: The mounted specimens were classified based on the regional classification. A total number of 160 specimens were mounted.

Conclusion: The study concludes with explanation of challenges and ways to overcome difficulties in a step by step manner extensively. The major problems for poor quality specimen and reduced visibility can be solved by temporary mounting the specimen before permanent mount.

Keywords: Anatomy museum, Perspex jars, wet specimens, Kaiserling solution.

Introduction

Human Anatomy is a branch of science that can be studied mainly by cadaveric dissections, wet and dry specimens prepared and preserved from passed on

human beings [1]. Museum is an essential part of Anatomy department in a medical college. Museums are in part historical, representing the pioneer work which gives complete information and knowledge of the human body in its four walls [2].

The development of more modern computers, sophisticated training tools, high-quality cross-sectional imaging, radiologic and living anatomy are currently available in the classroom, but it can't replace the reality and liveliness of real anatomy museum [3]. Wet specimens either in the form of organs or different levels of body sections which gives the whole picture to understand the human body, the organs or the body sections [4][5]. Moreover, the specimens are mostly seen in the glass or in the plastic jars in anatomy museum [6]. Meticulous and clear dissection determines the quality of the specimen [7]. The museum specimens must create awareness into the detailed aspects of study for better diagnosis and medical knowledge. When preparing an anatomical/pathological Museum either for educational purpose or general public visiting museum, it requires an enhanced quality display. Kaiserling's method for museum specimens remains the classical technique for preparation and it involves fixation of the specimen, colour restoration, preservation of the fixed specimen and presentation in glass, acrylic jars or transparent plastic (Perspex) jars filled with Kaiserling solution [7], [8]. The magnificence of the jar specimens depends on how finely it is dissected, properly prepared, mounted in a self-explanatory manner [9]. Only review papers are seen in the literature to prepare the museum jar specimens, but none of the studies have been showed the complete basic ideas to prepare museum jar specimens and its difficulties. Therefore, the present study was initiated as an effort to solve the reasonable problems step by step in preparing and mounting the specimen. A well-organised anatomy museum is aimed to be an ideal demonstration for undergraduate and postgraduate self-education and research. Hence, at present it is mandatory for every medical college to have a museum to get approval [10]. Therefore, our aim of the present study is to procure, fix, prepare and mount the specimens in consistent and compatible standards for Anatomy museum, by overcoming the practical difficulties for the purpose of academic and research, in a holistic way and scientific approach.

Materials and methods

i. **Cadavers**

Five well embalmed cadavers and organs were procured from Symbiosis Medical College for Women and Symbiosis University hospital Pune respectively, used for the preparation of the specimens over the period of 3 months.

ii. **Perspex jars**

A total number of 160 Perspex jars in different sizes and specifications with centre plates were used to mount the specimens.

S. No.	Jar Specifications	Quantity
1	8Hx8Lx6W	10
2	4Hx4Lx3W	10
3	6Hx6Lx3W	10
4	10Hx10Lx6W	90
5	8Hx10Lx8W	20
6	10Hx14Lx6W	20

Table 1: Shows Jar Specifications and Quantity

iii. **Chemicals**

The following chemicals were used for fixing the specimen, restoration of colour and mounting, based on the widely used modified method of Kaiserling solution

Kaiserling solution – I (Fixing solution/fluid)

Formalin 40% - 400ml

Potassium nitrate - 3gm

Potassium acetate	-	60gm
Water	-	2000ml

Kaiserling solution – II (Colour restoration)

Ethyl alcohol 80%	-	based on the size of the specimen

Kaiserling solution – III (Original Kaiserling solution)

Glycerine	-	500ml
Arsenious acid 1%	-	200ml
Potassium acetate	-	250gm
Thymol	-	2.5gm

Pulvertaft - Kaiserling mounting fluid

Glycerine	-	300ml
Sodium acetate 10%	-	100gm
10% formalin	-	1000ml
Camphor	-	5gm
Thymol	-	2.5gm

Suturing needles and thread

Dissection instruments

Drilling machine

Meat cutting machine

Stationary materials (markers, ruler)

Methods

Step – 1 skilful dissection

Specimens were procured from a well embalmed cadaver and soft parts. As preliminary procedure the tissue or the organ procured from the body was trimmed of all extraneous tissue, dissected and sectioned. Under expert guidance, fine and skilful dissection was performed. The edges of the specimens were trimmed finely with sharp instruments. Figure 1 shows a well dissected elbow joint specimen.

Step – 2 Groundwork for fixation

The specimens were kept around the volume of 15 to 20 times of Kaiserling fixative solution – I in a storage tray with a lid for adequate and proper fixation. The specimen was stored in the fixative solution for minimum 24 hours to few weeks based on the size and component of the specimen. A well dissected heart specimen and a section of brain tissue fixed with Kaiserling fixing solution was shown in figure 2 and 3 respectively.

Step - 3 Restoration of colour

The fixed specimen was transferred to a jar containing Kaiserling solution – II until the colour is fully restored. The specimen which were floating was lightly covered with surgical gauze, and the tray was closed to prevent evaporation. The specimen can be kept around two to eight hours based on the characteristics and size of the tissue. Figure 4 shows a kidney specimen with restored colour in Kaiserling colour restoring solution.

Step - 4 Temporary mount

The specimens were transferred to the original Kaiserling solution – III for temporary mounting in a suitably sized jar. Specimens thus sealed in individual temporary mounts can be safely stored until time is available to complete the permanent fixation.

Step - 5 Final Mounting of specimen

Specimens are trimmed to the anticipated size and shape so that it perfectly fits into the Perspex jar. All surplus and non-representative tissues were removed by careful dissection. Regular cuts were made to keep the specimen in anatomical position. The specimen kept laid flat on dissection table under sufficient light source, for better orientation and proper positioning before final mounting. The specimens were measured, allowing a half an inch clearance at the bottom to ensure a label to be fitted without hindering part of the specimen. The centre plate was thoroughly washed in a detergent, and dried with a gauze piece. Now the specimen was arranged in the required position of the centre plate, and markings were done using temporary

marker. Holes were drilled on the appropriate position using hand drill machine. Number of stitches was done based on the weight and consistency of the specimen. The centre acrylic plate with attached specimen was kept into the Perspex jar and ensured that the centre plate in proper position to prevent the movement of the stitched specimen. After positioning the centre plate with stitched tissue inside the jar, it is gently filled with Kaiserling mounting fluid on the side of the walls. With a spatula the trapped Air-bubbles in the centre plate and between the specimen were released and the jar was hermetically sealed. Figure 5 shows a permanently mounted cerebral folds specimen with Kaiserling permanent mounting solution.

Mounting bone and joint sections

After removal of soft tissues from the surface of the bones, it was dried in the incubator and it was washed and bleached with hydrogen peroxide. Then the bones were sectioned using the band saw meat cutting machine and then mounted. Upper half of the femur with soft tissue cleaned and bleached was sectioned as shown in figure 6. Figure 7 shows the section of knee joint using a band saw machine.

Bone decalcification and mounting

Soft tissues were excised from the bone for better penetration of decalcifying solution into the tissue. The specimen was washed with tap water and in a decalcification solution made by the mixing concentrated HCL 250 ml with 5 litres of water. On the 3rd day 250 ml of concentrated HCL is added to same solution to hasten the reaction. On 6th day quality of decalcification was checked to see if the bone can be bent. 250 ml HCL was again added to further facilitate decalcification process. On the 9th day the decalcification was again checked. At this stage it was able to bend spongy bone but not the compact bone. So again added 250 ml of concentrated HCL was added. On the 12th day the compact bone also gained the ability to bend. Then the specimen was removed from solution and rinsed in running tap water for at least ten minutes, and then specimen of decalcified bone was fixed in an acrylic jar for storage and display as shown in figure 8.

Labelling and Presentation

The specimens in the Perspex jars was clearly labelled in a descriptive manner with printed sheets as shown in figure 5; examination specimens were numbered with a numerical system of labelling as shown in figure 9. Catalogue was designed based on the arrangement for better understanding and referencing the specimen mounted.

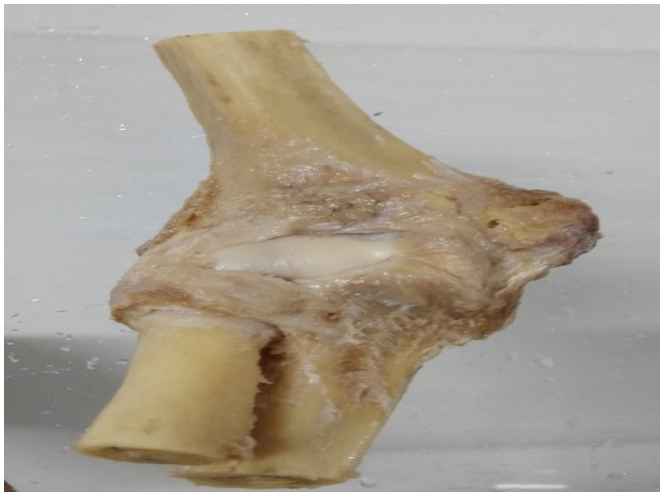

Fig. 1: finely dissected elbow joint

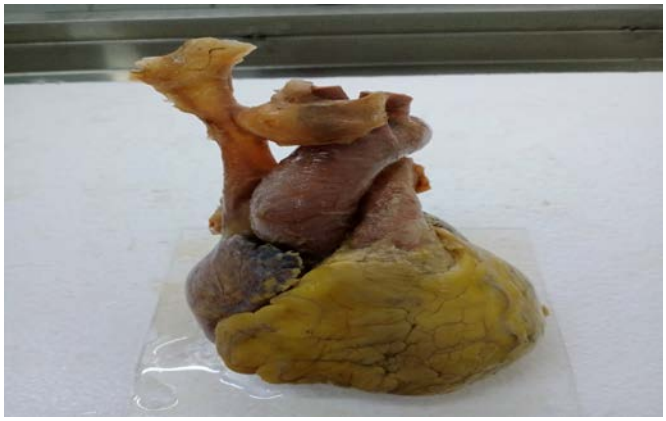

Fig. 2: Heart specimen supported with padding and fixed

Fig. 3: Brain specimen after fixation

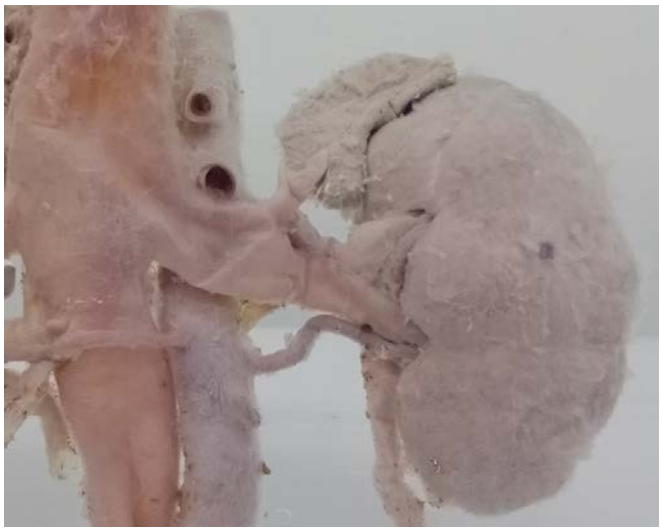

Fig. 4: After fixation colour is maintained (colour restoration)

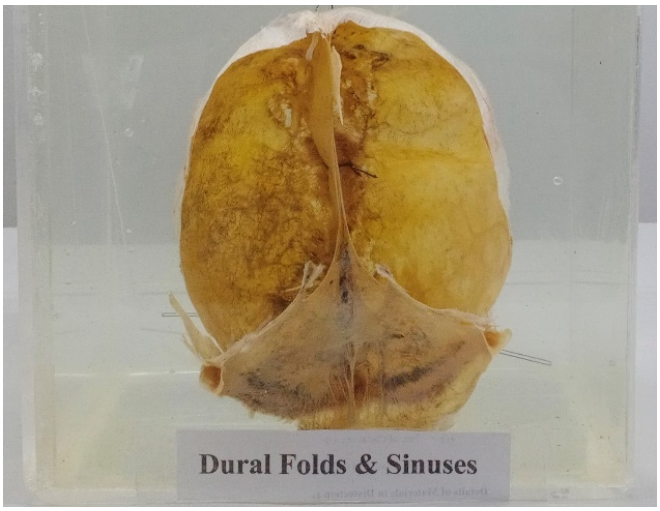

Fig. 5: Permanent specimen mounted with labelling for observational purpose

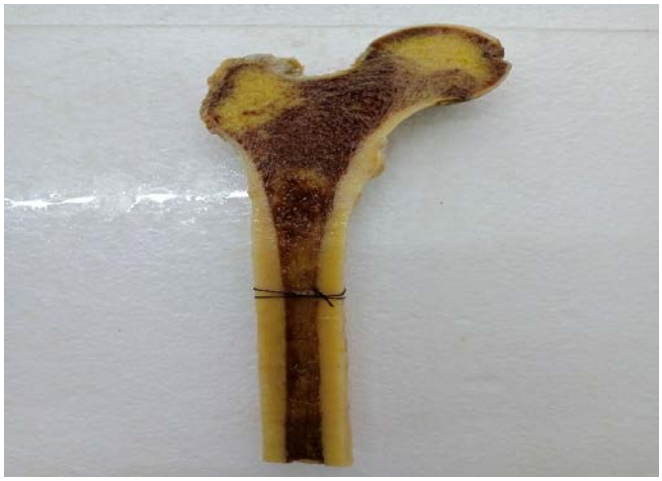

Fig. 6: section of femur before mounting

Fig. 7:Section of knee joint before mounting

Fig. 8: Femur decalcified and curved

Fig. 9: specimen labelled in numerical system for examination purpose

Fig. 10: Shows a poorly dissected and mounted specimen

Fig. 11a: shows tissue tags and poor visibility after two weeks of temporary mounting

Fig 11b: surplus and non-representative tissues removed before permanent mounting

Results

S. No	Specimens	Number
	Abdomen and Pelvis	60
2	Upper limb	16
3	Lower limb	45
4	Thorax	16
5	Head neck and face	21
6	Embryology	02
Total		160

Table 2: Number of Specimens Mounted In The Perspex Jars

The specimens were mounted according to respective sized perspective jars. Based on regional classification the mounted specimens were tabulated as shown in table 2.

Discussion

This section explains the procedure of specimen mounting, the problems and difficulties faced in each step from the stage of tissue procurement, fixation, colour restoration and mounting the specimen. The clarification and knowledge, regarding the specimen mounting were gained from the literature as well as our

own practical experience during the entire process of this extensive work.

The mounted specimens should provoke the interest regarding how fascia, muscles, bones, joints, nerves, vessels, organs function together [11], [12]. Washing the specimen with tap water, increases the haemolysis and it is one of the most common causes for poor quality specimen [13] [14] [15]. It is essential to select a fresh specimen for dissection or a well embalmed specimen must be procured [16] [17]. After washing in saline, the specimen should not be kept in saline for too long (less than two hours) [18] [19] [20]. Figure 10 shows an improperly trimmed specimen with unwanted tissue which spoils the quality of the visibility.

The fixative should be kept minimum 15 times the volume of the specimen [16] [21]. While tissue fixation is a chemical process, certain time must be endorsed for the process to complete. “Under-fixation” and "over fixation" are the detrimental factors which may leads to poor results which are overcome by fixing the specimen for minimum 24 hours to few weeks based on the size and component of the specimen [15] [22]. In the present study penetration was enhanced by spot injection with a syringe and needle in some tissues, such as whole upper and lower limbs. To fix and also to maintain the natural shape of the heart specimen, all the atrial and ventricular cavities were padded and the major vessels such as aorta, superior and inferior vena cava and the pulmonary vessels were padded with cotton wool as shown in figure 1. The spleen was fixed by injecting the main vessels, and ligatured under slight tension. Placenta specimen was fixed by injecting the fluid through umbilical vessels. In the present study the specimens were kept for minimum of 24 hours in fixative solution for proper fixation [23] [24] [25].

The present study implemented the technique of Ryter A [15] by placing the specimen in Kaiserling solution – II intended for Colour restoration for optimal period of 1 hour to 4hrs based on the size of the specimen [26] [27] [28]. But specimen was not kept too long in alcohol as it causes the irreversible colour fading [29] [36]. For colour restoration of floating specimens, were gently covered with surgical gauze, and the lid was closed to prevent evaporation [17] [34] [35]. Pulvertaft described that this method of mounting the specimen helps to withstand the colour for 25 years [17].

Temporary mounting was done to remove the tissue tags loose particles, and other soluble pigments from the specimen for around 1 to 2 weeks [36]. The advantages of temporary mounting before permanent mounting was shown in figure 11. Perspex jars have proved to be slightly flexible, quite acceptable with the solutions in use [17]. The jar was filled with Kaiserling mounting solution, thymol and camphor also added to prevent the growth of the moulds and the air bubbles were eliminated. Davidson's AFA, Gordon, N Brenner described the disadvantage of stitching the specimen with nylon thread on centre plates as it causes damage to tissue due to weight of specimen’s over period of time [37]. In the current study stitches and ties were made with suture thread to mount the specimen. Bone decalcification process was done based on the preparation method from the studies of [30] [31] [32] [33].

Conclusion

The present study has made an attempt to procure, fix and mount 160 specimens for the educational purpose in the Anatomy department museum. The study concludes with explanation of challenges and how to overcome difficulties in a step by step manner. The study suggests major problems of poor quality specimen and reduced visibility, could be solved by

temporary mounting of specimen before permanent mounting and moving into the museum.

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Author Profile: Dr. Raju Bokan, MBBS MD Anatomy, 9 years of Teaching and Research experiences in different Medical Colleges. Currently working on developing the Anatomy Museum under heads of Osteology, Radiology and Wet Specimens and other body preservations methods.

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