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Review Article

Plastination - An Innovative Preservative Technique in Anatomy

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Abstract

Preserving the cadavers from ongoing natural processes of decomposition and putrefaction have always been the focus of medical professionals especially anatomists, in order to use them for future studies and research. Embalming is the most commonly used procedure to prevent decomposition/putrefaction of cadavers. However, there are certain limitations/disadvantages ascribed to traditional "Embalming" procedures. Among these disadvantages, the most obvious ones are the repulsiveness of the students due to the smell and the formation of irritant fumes from the embalming fluid. Use of formaldehyde and methanol is also not considered environment friendly. In contrast, plastination technique has several advantages like making cadaver easy to handle, easy storage, being cheaper, odorless presentation and presents more anatomical details. Customization of the technique has been done for particular organs and according to the environmental conditions but still the basic techniques considered as pioneers for plastination are the Silicone (S10) and Sheet Plastination (P40). Though plastination has been established as an excellent preservation method but the ethical issues are still debatable. The moral dilemma has been under discussion since the development of this procedure. The major reason for this ethical and moral discussion is the social and cultural restraints towards the depiction of such exhibitions. In this article, a comprehensive detail history, methodology, advantages/limitations and a brief discussion on the ethical and moral dilemma have been presented.

Introduction

Keeping in view the potential of living tissues to undergo putrefaction and decomposition after death, anatomists were forced to develop the procedures to stop or at least significantly slow down the process

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of decomposition [1]. This process of decomposition is a consequence of the presence of microbial organisms like bacteria, fungi or protozoa [2]. The history of preservation of bodies goes back 2150±500 BC when mummification was the most demanded art in the ancient Egypt. Time passed by and with the assistance of some great scientists like Leonardo da Vinci (1452-1519), Robert Boyle (1663), Jan Swammerdam (1672) and other notable contributors, the technique of preservation was evolved [3]. Among these scientists, Robert Boyle tried to change the trend of studying dried up viscera by preserving a snake in wine spirit [4]. This was probably the first scientific attempt towards the preservation of tissues for anatomical study [5].

Embalming refers to the preservation of entire cadaver to prevent the desiccation of the body for future research/study purpose [6]. Embalming was first attempted by Jan Swammerdam who injected wax and turpentine in a dead body in order to solidify it internally and prevent the decomposition. In 1863, Wilhelm von Hoffman invented the gas "Formic Aldehyde" which was later on termed as Formaldehyde. It wasn't until 1893 when formalin (40% aqueous solution of formaldehyde gas) was first used by Ferdinand Julius Cohn to preserve a cadaver [3]. Formalin is quite effective in slowing down the process of decomposition. Almost a century passed and formalin along with phenol and glycerin was used to embalm the cadavers world-wide [7]. In 1979, a German anatomist, Von Hagen presented a new procedure "Plastination", a term of his own making for the preservation of cadavers [8].

Plastination

It is a procedure through which living specimen can be processed and preserved in their near-natural state. In this procedure, the lipids and the water present in the cadaver is replaced with synthetic components like polyester or silicone etc., and then after the hardening of these materials, we obtain a natural looking and durable anatomical specimen [9]. These specimens can be specified organs or entire organisms (if required) [10]. The most striking feature of this technique over conventional formalin based preservation is the anatomical detail and ease of handling of bodies after the procedure [11].

While classifying the plastination procedures, we can use the type of end-product as the reference point. Specimen can either be preserved as a whole in 3-D matter or slices of the processed specimen can be made for illustration. These two procedures are termed as silicone (S10) and sheet plastination (P40), respectively. A wide number of resins have been tested over the past 30 years for their potential use in the plastination procedure. Resins that are commonly used for the plastination are Silicone, Epoxy resin and Polyester. Now-a-days different variants of these chemicals are used by different laboratories and museums each having their own advantages and disadvantages.

Silicone Plastination (P10)

Of all the techniques, this is the most versatile one [12]. Silicon Plastination models are depicted in figures 1-3. The chemicals required for this procedure are acetone, methylene chloride, silicone

polymer, catalyst to prepare silicone molecules, chain extenders and cross linkers for silicone molecules [13].



Figure 1: Brain models preserved through plastination showing various sides (Source: von Hagen Plastination [8]).



Figure 2: The silicon plasination models of blood vessels of heart and lungs used for human body (Source: von Hagen Plastination [8]).



Figure 3: Anatomical plastination models of camels showing inside out ((Source: von Hagen Plastination [8]).

Procedure

Dissection and fixation of specimen: Fresh or embalmed specimen can be used. If more flexibility is required, then it is preferable to use fresh specimen [14]. Embalming fluid must be removed before dehydration if preserved sample is to be processed.

Dehydration: Water in the specimen is substituted with acetone at -25°C. Usually, acetone is changed up to three times. Ethanol can also be used but is not very preferable in routine procedures [15].

Defatting: In this step, lipid is removed by transferring the specimen from -25°C to room temperature. As soon as the lipid starts turning opaque, procedure is stopped [16].

Forced impregnation: The basic principle is to replace the volatile solvent (acetone etc.) with the silicone reaction mixture. Vacuum is required so that the viscous mixture can come in equilibrium with the dehydration agent. Forced impregnation ensures that the specimen will not be shrunk [17]. Although studies indicate that shrinkage of tissues actually takes place even with forced impregnation. But with the use of proper vacuum pressure, this shrinkage can be minimized [18].

Hardening: Curing or hardening is obtained by exposure of specimen to the curing gas, heat or ultra-violet light in an enclosed chamber. This leads to chain extension and cross linkage of the polymer [19]. After drying of the specimen surface, it is placed in an air tight bag which ensures the internal hardening of polymer [20].

Sheet Plastination (P40)

In this technique, 1-3 mm semi-transparent slices of tissue are obtained after the removal of tissue fluid and partial substitution of tissue fat with curable epoxy resin (Figure 4). Although this technique requires more preciseness but the advantage is that the preserved samples can be examined at microscopic as well as microscopic levels [21]. The basic chemicals used are fixatives (if required), acetone, epoxy resin, epoxy hardeners and epoxy plasticizers [22]. The procedure of sheet plastination is as follows:



Figure 4: Sheet (slice) plastinates are 1.5 mm thin slices of real bodies, impregnated with resins and subsequently hardened. Sheet plastinates illustrate anatomical structures, in color, either transparent or translucent. The method of plastination facilitates the production and preservation of thin, transparent body slices of unparalleled transparency, vibrant colors, and size (Source: von Hagen Plastination [8]).

Fixation

Specimen can be fixed using formalin to prevent bio-hazards but it is not obligatory.

Cold dehydration

This is the most important step in this technique. Specimen must be frozen at -70°C to -75°C. Body is placed at this temperature for 7-10 days to assure complete freezing. After thorough freezing, slic-

ing is done through butcher saw band. Cleaning of saw dust is essential to prevent artefacts. Then the slices are submerged in acetone at -25°C for dehydration [23].

Defatting

The dehydration bath has final acetone concentration of 98.5% for production of high quality transparent slices. Specimen having higher lipid content must be dehydrated with a stronger dehydration agent like methyl chloride [24].

Forced impregnation

In this step, solvent from the cellular and interstitial places is substituted with epoxy impregnation mixture in the presence of vacuum. At 5°C, complete impregnation takes about 36-48 hours while at room temperature, it takes almost 32 hours [15].

Hardening

This step can be performed with two techniques: In flat chamber method, slices are put in the flat chamber made of glass. After proper placement, epoxy resin is poured in the chamber. All the air bubbles must be removed before closing the chamber. Specimen is first kept at 15°C for 24-48 hours and then placed in oven at 45°C for 4 days. After removal from the chamber and cooling down, sheets are cut to desired size [25]. Sandwich method is faster than the former one. In this method, impregnated slices are floated over a pool of de-aerated casting mixture kept on the foiled sheet. Then the casting mixture is spooned on the slices and covered with another foil using the spatula. Pressure is applied to remove the air bubbles and glass sheet is placed on the top to ensure the stability of pressure. The sandwich block is kept at room temperature for one day and then in oven for 4 days at 45°C. After removal from oven, sheets are cut to desired size [26].

Advantages over formalin-based preservation

- Storage of the preserved samples is easier as we can store the plastinated samples even in a shopping bag with appropriate labeling [27].
- 2. This procedure offers completely preserved specimen without any toxic fumes or foul smell [28].
- 3. It is relatively cheaper than the conventional formalin based method in a longer run [29].
- 4. It is sometimes difficult for some students to study due to the smell of formalin, this in turn leads to impairment of interest of the student [30].
- 5. Specimen can be preserved for up-to 40 years if plastination is performed which is almost 10 times more than that of conventional method [31].
- 6. Plastination offers relatively more detailed features as all the structures are fully preserved in their near-natural state [30].
- 7. With the help of sheet plastination, it becomes easier to study the topographical anatomy in detail [32].
- 8. We can preserve the parasites present in the flesh such as larvae in the putrid flesh can be preserved for demonstration [33].
- 9. Fragile tissue sample such as intra-cerebral hematoma can be preserved perfectly and made durable for the future use [34].

Limitations of Plastination

Although plastination is a good technique for the preservation of specimen but, like any other procedure, it also has certain limitations. Few of these cons are that plastinated specimen are relatively inflexible (due to the presence of silicone in the tissue) so it becomes troublesome to reflect the specimen and demonstrate the deeper anatomical features. This inflexibility is the major problem that the scientists have to face while using plastinated specimen for clinical practices such as ultrasonography and endoscopy [35]. Thus for the clinical practices, plastinated specimen are not ideal for use. Similarly, after the completion of plastination, tissue becomes so much hard that further dissection is difficult so, the final dissection has to be done before the procedure. In addition, the process is quite time consuming and sensitive technique so it requires skilled manpower [19]. Plastination laboratory development also requires a huge amount of investment which also poses major bottleneck while developing the laboratory [36].

Chemicals used in the plastination procedure pose a health hazard if they are not properly handled. Acetone used in different variants of plastination techniques can cause respiratory and dermal irritation in case of short term exposure in low concentration (250ppm-1000ppm). High concentration (>12000ppm) can cause more severe symptoms such as vomiting and unconsciousness [37]. Long term exposure to acetone in mice with concentration >19000ppm has shown to produce reversible decrease in the absolute brain weight of cadaver [38]. Like most of the organic solvents, acetone also causes visual impairment and loss of sensitivity if exposed to for a long time [39]. Hydroxyl-terminated polydimethy isiloxane and Ethyl silicate used for the silicone impregnation technique has been reported to cause allergic reactions and dermatitis respectively in case of long term exposure [40,41]. Dibutyltin dilaurate used for sheet plastination not only causes allergic reaction but also can cause asthma if vapors are inhaled by the operator [42].

Another major limitation regarding the plastination protocol is the exposure to pathogens especially during the early processing of samples [43]. This risk factor can be reduced either by the use of protective equipment such as gloves, masks and aprons etc., or by the fixation of specimen in formalin prior to the further processing as formalin effectively neutralizes most of the pathogenic organisms [44].

Moral and Ethical Dilemma

Although there are well developed policies regarding the donation of human bodies after death but still there are a number of questions regarding the questionnaire for the consent form [45]. According to a survey, individuals were found more inclined to give consent for whole body plastination instead of the plastination for few body parts only [36]. The reason may be the resentment of the public towards the words "autopsy or dissection". Similarly, few cultures are more prone towards getting offensive against these body displays in exhibitions [46].

Conclusion

Unlike most of the innovations that faced huge criticism in the beginning, plastination was appreciated right from the start. Although it is a much better preservation technique than the conservative methods but room for improvement is still there. Despite the ethical questions being raised, the number of donors, willing to undergo this process,

gives a good idea of how this procedure has fascinated the public and this fascination can be used to educate the society for betterment.

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