ORIGINAL ARTICLE

VACUUM ASSISTED PLASTINATION USING MELAMYNE

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ABSTRACT

Introduction: This study is an attempt to perform plastination using indigenous method which is cheap, executable and does not require extremes of cold temperature and technical instruments. The effectiveness of the procedure was judged by calculating shrinkage post plastination, external appearance, polymer cost and consumption, extra equipment cost, and student's feedback post teaching with plastinates.

Methodology: The present study was conducted from June 2016 to December 2016 in Department of anatomy, GMC Kota Rajasthan. The cadaveric specimens procured were fixed in 10% formalin. The measurements were taken by scale and digital vernier caliper. The steps of plastination were same as the original technique i.e. Fixation, Dehydration, Degreasing, Forced Impregnation in vacuum and Curing (hardening). Simple technique using indeginous instruments were used to apply the vacuum fitted with the timer for 20 minutes Final measurements were taken of all the specimens and then ANOVA between the groups in medcalc software was used to compare the shrinkage.

Results: Dry, odourless, portable and aesthetically pleasing plastinates were obtained. The dimensions were much reduced after each step except after vacuum assisted impregnation there was slight increase in the dimensions though it did not reach the initial value.

Conclusion: Feedback from students regarding plastinates revealed that they were easy to handle but less flexible and difficulty in visualizing deeper structures but good for cross-sectional anatomy as the tissue got fixed and displacement of structures was minimal. These plastinates are excellent adjunct for teaching as they have nullified exposure to the toxic fumes of formalin.

Keywords: Plastination, intermittent vacuum assisted impregnation, melamyne, acetonometers

INTRODUCTION

Plastination is the process of permanently preserving tissue in a natural state by replacing the body fluids (i.e. fat and water) with synthetic materials. The S 10 technique is the standard technique in plastination which gives opaque, more or less flexible, and natural looking specimens.[1] But the standard technique is quite costly as it requires extremes of temperature and technical instruments. Thus cost effective and feasible plastination technique is an ongoing challenging aspect for anatomist all over medical colleges in India due to lack of funds and limited infrastructure. In 1998, Daniel Corcoran, Dow Corning Corporation, proposed a different silicone impregnation mixture, and the room temperature plastination technique was developed [2,3,4,5,6]. This study is an attempt to perform plastination using indigenous method which is cheap, executable and does not require extremes of cold temperature and technical instruments. The effectiveness of the procedure was judged by calculating shrinkage post plastination, external appearance, polymer cost and consumption, extra equipment cost and student's feedback post teaching with plastinates.

METHODOLOGY

The present study was conducted from June 2016 to December 2016 in Department of anatomy, GMC Kota Rajasthan. The cadaveric specimens procured were fixed in 10% formalin. The measurements were taken by scale and digital vernier caliper. Prosection of arm, forearm, neck at C6, brain with internal capsule ,paranasal sinuses coronal section and floor of fourth ventricle and lateral ventricle were used. Consumables—Acetone, xylene, hardener, paint brush, acetone, glass jars , vacuum chamber, acetonometer /alcoholmeters, mortuary chamber.

Dehydration—The specimens were then transferred to acetone specific gravity -0.8 which was measured by acetonometer /alcoholmeters (fig no. 1). Specimens were subjected to 3 changes of acetone. Each

change comprised of 7days treatment. Last change was reused for other specimen till the specific gravity decreased to 0.6. Measurements were taken after last change.

Degreasing: Dehydrated specimens were then subjected to 3 changes of xylene, each change of 7 days. Xylene acts as the volatile intermediary with the acid curing polymer and also a degreasing agent for lipid rich specimens.[10] Measurements were taken after last change.

Impregnation—A intermittent vacuum assisted impregnation was done for the specimen which comprised of solution of melamyne and xylene mixed in proportion 1:1 (Fig 2) A vacuum of 7 mm of Hg was applied till the bubbling ceased (as average of 10 days for the specimen.) Timer was set for twenty minutes with a pause of five minutes. The specimens were taken out of vacuum chamber and they were wiped off from extra polymer. Again all the measurements were taken.

Curing and hardening –Specimens were shifted into a sealed chamber with diluted sulfuric acid in a beaker and calcium chloride as hygroscopic element in other beaker. The whole chamber was then transferred into freezer (mortuary chamber) with temperature of 10°C for 1 month. After drying the whole chamber was kept in UV light at room temperature. Final measurements were taken of all the specimens and then ANOVA in medcalc software was used to compare the shrinkage. Shrinkage was calculated as Final reading(of each step)/ Initial Reading X100.

RESULTS

Dry, odourless, aesthetically pleasing plastinates were obtained which were lighter (Fig 3-8)

Table 1: Comparision of specimens using ANOVA within subjects

Specimens	Sum of Squares	DF	Mean Square	F	P
PNS	2.342	4	0.586	2.83	0.052
4th Ventricle	0.782	4	0.195	24.82	0.004
Internal Capsule	4.686	4	1.172	7.95	0.035
Lateral Ventricle	3.16	4	0.8	8.2	0.035
Anterior horn of Lateral Ventricle	3.181	4	0.795	8.05	0.034
Root of Neck	1.063	4	0.266	5.51	0.02
T.S. of Hand (Wrist)	0.808	4	0.202	1.67	0.316
T.S. of Proximal Forearm	1.09	4	0.273	1.29	0.405
T.S. of Distal Forearm	0.947	4	0.237	2.46	0.202

DF – degrees of freedom, PNS- paranasal sinuses T.S- transverse section

Table 2: Specimens showing shrinkage in Percentage

Specimens	Initial	After Dehydration	After Degreasing	After Impregnation	After Plastination
PNS	100	93.02±5.22	91.83±4.74	96.23±10.60	86.55±7.67
4th Ventricle	100	91.19±3.34	85.77±5.93	87.52±4.85	85.50±9.26
Internal Capsule	100	90.64±5.94	86.60±2.50	89.54±2.38	87.42±3.70
Lateral Ventricle	100	90.67±1.18	84.16±7.22	90.93±3.51	88.14±4.21
Anterior Horn	100	91.56±1.58	88.44±2.80	89.65±1.85	81.33±9.31
Root Of Neck	100	94.67±3.09	93.03±3.09	95.35±4.09	92.10±2.63
Wrist	100	95.88±3.53	90.89±5.06	97.52±9.84	90.41±0.33
Proximal Forearm	100	91.33±1.69	89.88±1.63	100.45±7.14	90.02±13.40
Distal Forearm	100	89.27±0.54	87.48±0.63	94.28±7.85	83.73±12.66

Table 3: External appearance of specimens after each step

Specimens	Formalin fixed specimens	Post dehydration	Post degreasing	Post impregnation	Final plastinates
PNS	Brown	Dark brown	Black	Brown	Light brown
4th Ventricle	Grey	Dark brown	Black	brown	white
Internal Capsule	Grey	Dark brown	Black	brown	white
Lateral Ventricle	Grey	Dark brown	Black	brown	white
Anterior Horn of Lateral Ventricle	Grey	Dark brown	Black	brown	white
Root of Neck	Brown	Dark brown	Dark brown	Brown	Light Brown
T.S. of Hand(Wrist)	Brown	Dark brown	Dark brown	Brown	Light Brown
T.S. of Proximal Forearm	Brown	Dark brown	Dark brown	Brown	Light Brown
T.S. of Distal Forearm	Brown	Dark brown	Dark brown	Brown	Light Brown

PNS- Paranasal sinuses, T.S.- Transverse section

Figure no.1 Alcoholmeters /Acetonometer

Figure 2 Vacuum chamber with timer

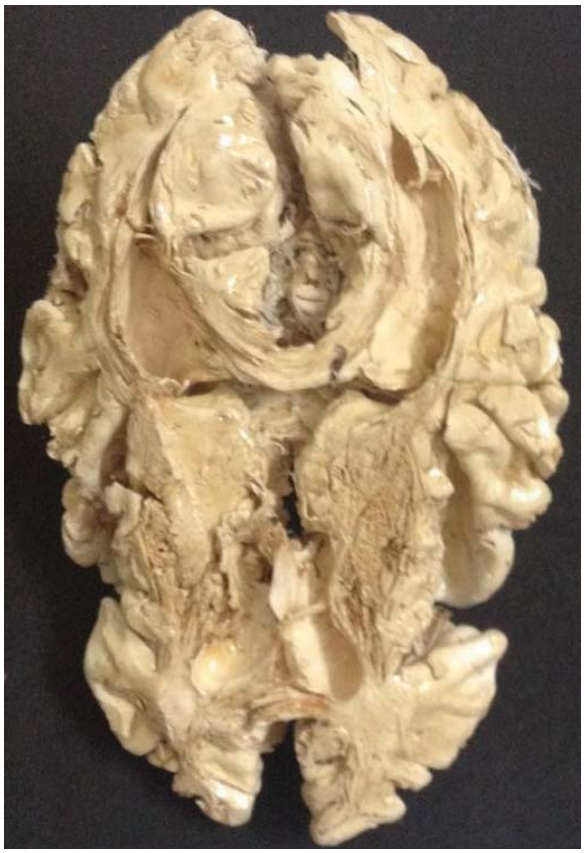

Figure .3 Transverse section of brain showing internal capsule and basal ganglia

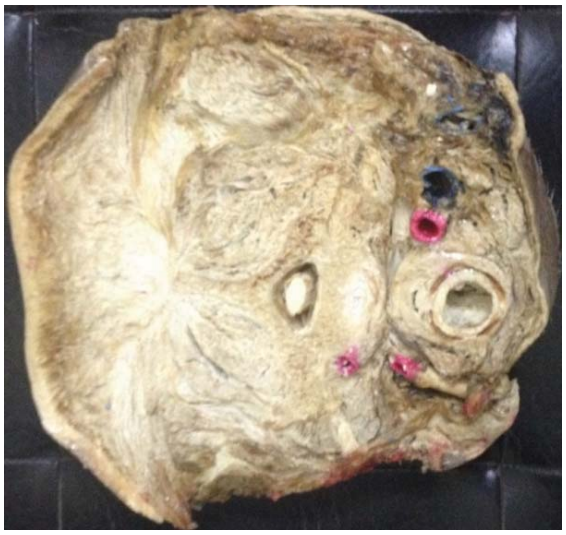

Fig 4 Transverse section of neck at C6 level

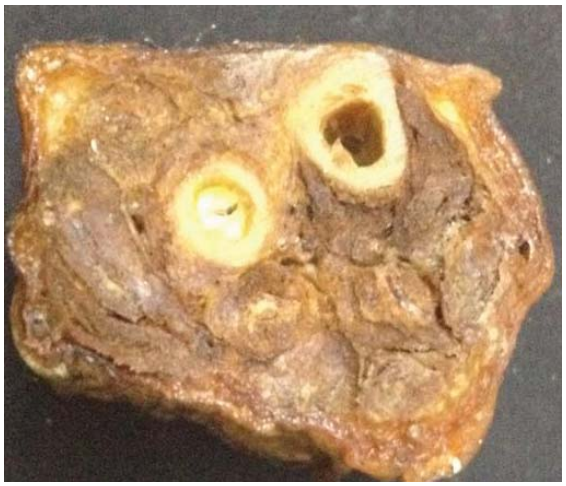

Fig 5.Specimen of proximal section of forearm

Fig. 6 Specimen showing lateral ventricle of brain

Figure 7: Specimen of paranasal sinus

The dimensions were much reduced after each step except after vacuum assisted impregnation there was slight increase in the dimensions though it did not reach the initial value. Shrinkage though was present but was not so remarkable as it varied from 10%-17%(fig. no. 3-7). The change was significant amongst the specimens except in the case of transverse sections of extremities as depicted by p value in table no.1. Decolourisation was maximal in case of brain prosections as compared to extremities. (Table no.3).

DISCUSSION

Preserving prosections has been a persistent aspiration for anatomist all over India. The standard plastination process consists of four sequential steps viz. Fixation, Dehydration, Forced Impregnation in vacuum and curing (hardening). [7, 8, 9]. The steps of plastination were same as the original technique i.e. fixation, dehydration, impregnation and curing. This study instigated possibility of plastination partially at room temperature, which was also tried as pink city technique[10].The modification was introduction of intermittent vacuum which reduced the shrinkage of specimens to a greater extent and also drying in a cold environment of 10 °C which was achieved by keeping the specimen in a mortuary chamber with calcium chloride . The introduction of cold temperature and calcium chloride (since it is hygroscopic) speeded up the drying process and reduced the shrinkage.

Dehydration was achieved by using acetone as it can be used as dehydration as well as intermediary agent [12]. Also it can be recycled especially of last change which can be used as first change for new specimen till its specific gravity reaches 0.6.Dehydration was carried out at room temperature in the department .Next step was clearing or degreasing for which xylene was used which dissolves all the fat. Incomplete impregnation have been tried earlier by diluting the polymer with the xylene to enhance the pliability of the specimen as proportionately less polymer is drawn by the specimen.[13] The major shortcoming of this step was remarkable color change of the specimen specially of brain due to which we had to discard a specimen of cerebellum as the nuclei could not be differentiated after this degreasing.

This is a ground breaking trial of preserving specimen by use of inexpensive and indigenous chemicals like, acetone, xylene and melamyne. A few anatomists have earlier tried at room temperature[11] . We prompted the use of melamyne for plastination as its curing temperature is below 100°C which made this step of impregnation feasible at room temperature and also catalyst or gas curing agents were not required. [14] Melamyne is readily soluble in xylene so the mixture obtained had the desirable viscosity and transparency required for plastination.[13] Xylene acts as intermediary volatile solvent.

The vacuum impregnation was done till bubbling ceased. It was fitted with timer for 20 minutes for applying intermittent impregnation. This reduced shrinkage as clearly depicted by the dimensions taken after each step as the polymer replaced acetone which preserved life like state. Intermittent vacuum assisted impregnation was applied instead of continuous sudden and rapid impregnation as it avoids compression of specimen. Curing is often done by gas, heat or light. In this step we utilized H_2SO_4 solution and for drying and hardening CaCl2 as its highly hygroscopic and the whole procedure was further speeded up by cold temperature of 10 degrees centigrade which was achieved by using mortuary chamber.

The major shortcoming of our procedure is the time required which is 1968 hours as compared to standard procedure which is 101.7 hrs[16] but it is cost effective.

Alsopercentage shrinkage coincided with that of other room temperature technique which was 10-17% in the present study as compared to 12-16% and much lower than standard procedure which is about 20-30%.[16] Decolourisation of the specimen was also minimal as compared to that of ameko et al.[17]

CONCLUSION

Specimens obtained were durable and portable .In short specimens more as a model with real look was

obtained. Feedback from students regarding plastinates revealed that they were easy to handle but less flexible and difficulty in visualizing deeper structures but good for cross-sectional anatomy as the tissue got fixed and displacement of structures was absent . Further the cost of plastination due to use of melamyne is nominal and also vacuum was applied by using locally assorted instruments which did not required any expertise to handle. This procedure minimized dependence of cadavers for teaching anatomy. These plastinates are excellent adjunct for teaching as they have nullified exposure to the toxic fumes of formalin.

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