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Effect of Temperature on the Gross Shrinkage of Pig Hearts by Using Silicone (S10) Plastination

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Abstract

Introduction: *Plastination is a modern method of body preservation. The plastinated specimens are dry, odourless, highly durable, non-toxic, non-infectious. As various factors may contribute to the determination of the quality of the plastinates, this research is designed to find out the influence of one factor such as temperature on the plastinates.*

Objectives: *The study was done to compare the percentage of gross shrinkages by volume of pig hearts in different stages of silicone (S10) plastination at cold temperature with those at room temperature.*

Materials and Methods:*. This experimental study was carried out in the Department of Anatomy, Bangabandhu Sheikh Mujib Medical University, Dhaka on twenty-four pig hearts. The percentages of gross shrinkages by volume were calculated for different stages of plastination at cold temperature and at room temperature.*

Results: *The overall mean percentage of gross shrinkage by volume of the pig hearts was significantly greater (38.01±18.78) in cold temperature group than in room temperature group (26.50±12.63).*

Conclusions: *The present study showed the quality of plastinates is better in room temperature than in cold temperature. Thus, room temperature procedure is suitable for our country and also cost-effective which fulfill the ever-increasing demands of user-friendly anatomical tools for the teachers and learners.*

Key words: *Silicone plastination, shrinkage, acetone, dehydration, forced impregnation, cold temperature*

Introduction

Plastination is a method for long-term preservation of biological specimens. In plastination, fluids and soluble fat in biological tissues are extracted to stop decomposition and are replaced by plastic polymers, such as silicone, epoxy or polyester resins which finally results in natural looking, dry, odourless, highly durable specimens. Several authors claimed that the plastinated specimens are non-toxic.[1,2] Use of plastinated specimens reduces the irritation and harmful effects of preservatives or embalming fluids such as formalin, alcohol.[3] They can be stored in simple plastic bags, require little storage and no maintenance.[4] This modern and unique method of preservation of biological specimens is invented by Dr. Gunther von Hagens in 1977 at the Department of Anatomy of Heidelberg University in Germany. During dealing with plastination in research, the most common complication is 'shrinkage'. The common reasons for this defect depend on different factors which include temperature, dehydration medium, tissue quality, duration of fixation, dehydration, forced impregnation and curing and also using of old bad formalin fixed specimens. The aim of this study is to compare the percentage of gross shrinkages by volume of pig hearts in different stages of silicone (S10) plastination at cold temperature with those at room temperature.

Materials and Methods

A total of twenty-four hearts from approximately one-year old pigs were collected from two government authorised slaughter houses of Dhaka. Twelve pig hearts were taken for each temperature group (room temperature group and cold temperature group). Out of the twelve hearts, nine

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were kept whole and three were sectioned longitudinally into two pieces. Both a whole or sectioned pig hearts was considered as individual sampling unit. Each of these units was numbered with a tag. Volume of each sampling unit was measured. At the fresh stage and after every stage of plastination the percentages of gross shrinkage were calculated. Overall shrinkages by volume were also measured (i.e., from the fresh stage to the end of plastination). The volumes of the specimens at the fresh stage and after every stage of plastination were measured by fluid displacement method. At first, the container was put on a steady table near its edge and was filled with water until water escaped through the tube. The water was allowed to escape spontaneously to the last drop. Then an individual sample unit (a whole organ or sectioned organ) was put into the container. The displaced water that escaped through the plastic tube was collected and measured in a graduated cylinder (Figure 1). The volume of this collected water was considered as the volume of the individual sample unit and was recorded in milliliters.

Fig.-1: *The escaping volume of water being collected in a measuring cylinder after putting the whole or piece of an organ into the water-filled container.*

At first fixation was done. The procedures were same for both temperature groups. All the pig hearts (whole and sectioned) were placed inside a plastic bucket containing 10% formalin for 5 days.[5] After fixation, the pig hearts were rinsed in running tap water for overnight to remove excess formalin. Then a plastic container was filled with acetone for dehydration. The amount of acetone was five times more than the volume of all the pig hearts. The pig hearts were kept in acetone (Figure 2).

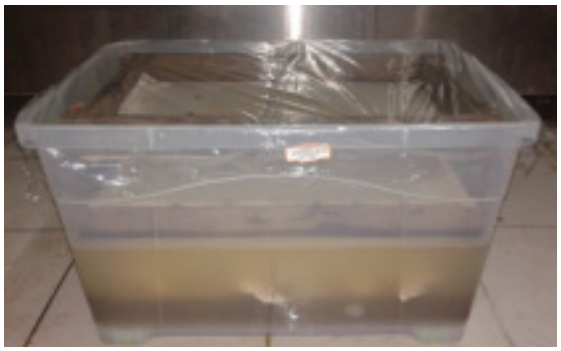

Fig-2: *Keeping the whole and sectioned organs in the plastic container for dehydration*

For the cold temperature group, precooling of the whole and sectioned pig hearts were done in a freezer for 12 hours. Then they were dehydrated in a series of 83%, 95% and 100% acetone at -19 to -23^{0}c in freezer. For the room temperature group, the whole and sectioned pig hearts were dehydrated in a series of 83%, 95% and two changes in 100% acetone at room temperature. After that impregnation procedure was done. For the cold temperature group, the impregnation procedure was done at -18^0 to -23^{0}c in a deep freezer and for the room temperature group, the procedure was done at 20-28^{0}c. The organs were kept in the plastination kettle in reaction-mixture S10 / S3 for overnight to allow excess acetone to escape and to equilibrate with the reaction-mixture of silicone naturally. Then vacuum was applied and pressure was slowly lowered to 5 cm of Hg. The rate of pressure decrease was monitored by observing bubbles formation on the reaction-mixture of silicone surface (Figure 3). Impregnation was judged completed when the bubbles production ceased for 12 hours.[6]

Fig- 3: *Bubbles formation on the surface of the reaction mixture S10 / S3.*

Fig- 4: *Gas-curing chamber.*

For gas-curing, the organs were placed in the curing chamber on absorbent papers (Figure 4). The cross linker BIODUR® S6 (20 cc) and some amount of the desiccant ($CaCl_2$) were put in the curing chamber. Then the chamber was closed and S6 was helped to vapourise using a small ventilator fan for twenty minutes two times a day. Every day the surfaces of whole and sectioned organs were manicured and were exposed to gas-curing agent (S6) until the surface remained dry.

Results

After fixation, no shrinkage by volume occurred either at room temperature or at cold temperature. Rather, there was an increase in volume and the difference between two temperature groups reached to statistically significant level. After dehydration, the mean percentage of gross shrinkages by volume was significantly smaller at cold temperature than at room temperature. But after forced impregnation and gas-curing**,** it was significantly greater at cold temperature than at room temperature.

The overall mean percentage of gross shrinkage by volume (from fresh stage to the end of plastination) of the pig hearts was significantly greater (38.01±18.78) in cold temperature group than in room temperature group (26.50±12.63) (Table I).

Table-I

Comparison of percentage of gross shrinkage of the pig hearts by volume in the different stages of plastination at cold temperature with that of room temperature

Stage of plastination	No. of organs (for each group)		No. of pieces (for each group)	Percentage of gross shrinkage by volume Mean ± SD Median (25^{th} and 75^{th} percentile)		P value
				Room temperature	Cold temperature	
Fixation	Whole	9	9	-8.27 ± 3.91	-3.90 ± 2.55	
	Sectioned	3	6	-7.52 (-11.05, - 5.90)	-3.22 (-5.26, -1.76)	0.00
Dehydration	Whole	9	9	17.71 ± 9.71	9.04 ± 4.32	
	Sectioned	3	6	18.75 (10.55,24.08)	8.62 (5.26, 12.82)	0.01
Forced impregnation	Whole	9	9	6.09 ± 3.85	17.49 ± 10.33	
	Sectioned	3	6	4.59 (2.78, 7.69)	13.10 (11.84, 25.53)	0.00
Curing	Whole	9	9	3.50 ± 1.73	16.09 ± 13.52	
	Sectioned	3	6	3.41 (2.40, 5.00)	9.38 (7.00, 26.00)	0.00
Overall shrinkage (from fresh stage to the end of plastination	Whole	9	9	26.50±12.63	38.01±18.38	
	Sectioned	3	6	21.42 (17.45,37.50)	30.54 (24.73,52.56)	0.04

Level of significance P ≤0.05

Discussion

Akhter[7] mentioned in her study that there was no shrinkage by volume of the pig kidney occurred on fixation stage either at cold temperature or at room temperature. Rather there was an increase in volume. But this did not reach any significant level. In this study, the pig hearts exhibited the similar outcome. But the difference between the two temperature groups was statistically significant. Brown et al did a similar type of study using kidney, liver, testicle and heart.[8] On dehydration stage, they observed that the average percentage of shrinkages was 14.5% at cold temperature and 20.2% at room temperature. Akhter[7] dehydrated pig kidney and found that the mean percentages of gross shrinkages by volume were significantly smaller at cold temperature than at room temperature. That finding matches with that of the present study. On forced impregnation stage, the mean percentage of gross shrinkage by volume was significantly greater in the pig heart at cold temperature than at room temperature. Akhter[7] did a similar type of study using pig kidney. She found that it was smaller in the pig kidney at cold temperature than at room temperature and did not reach any significant level. The mean percentages of gross shrinkage by volume in this study were significantly greater at cold temperature than at room temperature on gas-curing stage. This finding matches with Akhter.[7] The overall shrinkage of the pig hearts was less in the room temperature method than at the cold temperature method.

Conclusion

In this study, it was found that room temperature plastination method produces less shrinkage than cold temperature plastination method. Plastination offers new alternatives for the study of Anatomy. But it is quite difficult to prepare well plastinated specimens. Therefore, a series of research dealing with one or more issues at a time would be required. With suitable plastination method developed, good quality and cost effective plastinates may be obtained that can be used as effective teaching-learning tools in Bangladeshi condition. Such research would not only be useful in the field of Anatomy, but would also prove beneficial in other specialties such as Pathology, Forensic medicine, Surgery and Veterinary Medicine.

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