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Effects of Temperature on the Flexibility of Cow's Skeletal Muscle on Silicone (S10) Plastination

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Abstract

Context: *Among many factors responsible for producing a good plastinate, temperature is one of the most important factors. The present study had been designed for making a suitable method of plastination of skeletal muscle in our country.*

Materials and Methods: *This experimental study was carried out in Bangabandhu Sheikh Mujib Medical University (BSMMU), Dhaka, Bangladesh during the period of March, 2018 to February, 2019. Twenty (20) pieces of skeletal muscle were collected from approximately one-year-old slaughtered cow (*Bos indicus*) of government authorized slaughter house of Dhaka, Bangladesh which were designated as the 'Cold Temperature group' and the same numbers of pieces of skeletal muscle as the 'Room Temperature group'. Then their percentage of changes in flexibility after every stages of plastination were measured and compared.*

Results: *The percentage of changes in flexibility was non-significantly larger at cold temperature than at room temperature in fixation, dehydration and gas-curing stage. After gas-curing, the overall (from fresh stage to the end of plastination) the changes in flexibility is larger in Cold Temperature group than in Room Temperature group.*

Conclusions: *In the present study, the percentage of change in flexibility of skeletal muscle shows better outcome at room temperature than at cold temperature, but this findings was statistically non significant. So, further researches with larger samples are recommended to reach a definitive conclusion.*

Key words: *Temperature, skeletal muscle, change in flexibility, silicone (S10) method.*

Introduction

Study of cadavers and biological specimens have no alternatives in learning anatomy. If these specimens are plastinated, then they become long lasting, dry, easily handleable and maintenance free.[1] The health hazards can be reduced much by this way. The number of medical college is increasing day by day, but the supply of cadavers, viscera and other specimens are not adequate. In Bangladesh, body donation is not well practiced because there are some religious and social stigma. With increasing emphasis on the use of prosections to aid teaching of gross anatomy and limited availability of cadavers and funds, plastination is a unique method of preserving the specimens. Although plastination is a simple process, the results are often not as satisfactory as expected. Various factors have been found to contribute to the determination of the quality of plastinates. Among them temperature is one of the most important factors. Bangladesh is a tropical country and temperature ranges from 14°C to 30°C.[2] It is said that cold temperature (-18°C to -21°C) plastination method produces good quality specimens. But it can be possible in room temperature (20°C to 28 °C) also. If a good quality specimen can be produced by room temperature method the expenditure will be reduced more times. Few researches[3,4,5] have been done in Bangladesh, to show the effects of temperature on the procedural times and on the

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gross morphology of selected pig and goat organs in silicone (S10) plastination. However, these studies could not conclude a consistent result regarding temperature difference in which the plastination process takes place. So, a series of researches may be needed to acquire a consistent result.

Materials and Methods

The specimens were collected from pectoral muscle of chest of the cow (*Bos indicus*). Twenty (20) pieces of skeletal muscle were designated as the 'Cold Temperature group' and the same numbers of muscle as the 'Room Temperature group'. The skeletal muscle was fixed with 10% formalin. Then rinsed with water, pre-cooling was done only in Cold Temperature group. Afterwards, the specimens of Cold Temperature group was dehydrated with cold temperature acetone at -19 ^{0}C to - 23^{0}C and the specimens of Room Temperature group was dehydrated with room temperature acetone at 20 ^{0}C to 28 ^{0}C separately. Forced impregnation of the specimen of two groups were done with silicone S10 and S3 at both temperatures separately. Lastly, gas-curing of the specimens of two groups were done with S6 at room temperature.

An older light microscope was modified by attaching a digital slide caliper, which has a fixed and a movable part, with it to form a locally made flexibility machine (Figure 1) for measuring the flexibility of skeletal muscle. The objective lenses of the microscope were removed and a circular, metallic plate was attached at that place. The fixed part of the digital slide caliper was attached to the stage of the microscope and the movable part was moved with the circular metallic plate.

The specimen was placed on the stage of machine and was touched by the circular metallic plate by moving the adjustment knob. When the circular metallic plate just touched the upper surface of the specimen the value of the digital measuring display was made zero (0). Then the specimen was compressed by moving the stage upward with the movement of the adjustment knob. The value of the change in flexibility of the specimen in millimeter would be yield directly on the display in millimeters (Figure 2).

Change of flexibility was measured from each piece (sampling unit) of skeletal muscle. At fresh stage and after every stage of plastination the percentage of change in flexibility was calculated. Overall percentage of change in flexibility was also measured. For the comparison of percentage of change in flexibility, the values of the individual sample units were expressed as means and medians (as there were non-normal distributions as well) for the two groups. Hypothesis testing was done for the differences between the two groups using Mann-Whitney U test with the help of software Statistical Package for Social Sciences (SPSS) Version 20.

Ethical clearance

The research was dealt with animal materials (skeletal muscle of the cow) collected from dead animals in government authorized slaughterhouses which were sacrificed by cutting of both carotid arteries and both jugular veins by maintaining international animal sacrificing protocol.[6] Permission was taken for carrying out the research from the Institutional Review Board (IRB) of BSMMU.

Result

After fixation, dehydration and gas-curing stage the percentage of changes in flexibility of skeletal muscle were non-significantly larger at cold temperature than at room temperature. But after forced impregnation stage the percentage of change in flexibility of skeletal muscle was non-significantly smaller at cold temperature than at room temperature. The percentage of change in flexibility is larger means the specimen is less flexible. In this study, flexibility were larger in Cold Temperature group than in Room Temperature group. That means the plastinates of the Cold Temperature group were less flexible than that of Room Temperature group.

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Table I

Comparison between the two temperature groups regarding the percentage of changes in flexibility of skeletal muscle in different stages of plastination

Stage of plastination	Percentage of change in flexibility Mean ± SD Median ± (25 th and 75 th percentile)		Probability (p) and significance		95% confidence interval of dfference	
	Room temperature	Cold temperature			Room temperature	Cold temperature
Fixation	24.25 ± 14.74	34.19 ± 15.71	0.079	NS	17.35 to 31.15	26.84 to 41.54
	23.59 (12.34, 38.08)	35.69 (18.30, 44.17)				
Dehydration	29.99 ± 14.11	30.14 ± 14.79	0.766	NS	22.69 to 35.90	23.21 to 37.06
	27.15 (15.46, 45.55)	28.81 (19.27, 38.34)				
Forced impregnation	45 ± 18.09	43.68 ± 17.82	0.665	NS	36.53 to 53.46	35.34 to 52.02
	51.77 (33.08, 58.73)	47.13 (30.64, 57.56)				
Gas-curing	28.75 ± 14.01	32.19 ± 15.73	0.534	NS	22.19 to 35.30	24.43 to 39.55
	25 (16.37, 44.32)	33 (17.76, 45.08)				

n (Number of specimens in each group): 20; S:Significant ($p \leq 0.05$); NS: Non-significant

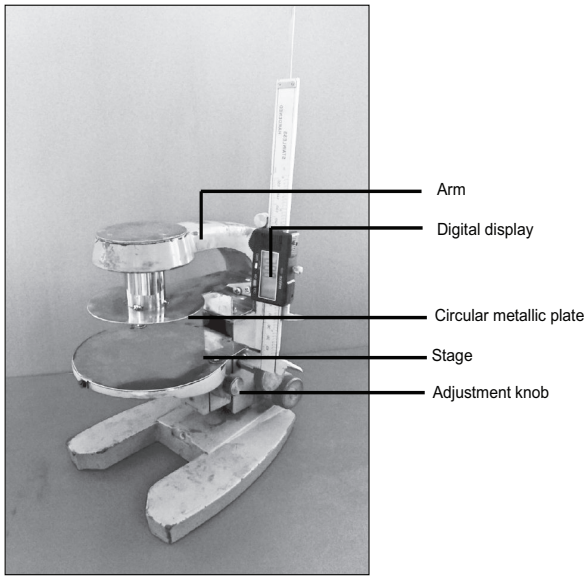

Fig.-1: *The specially constructed flexibility measuring instrument.*

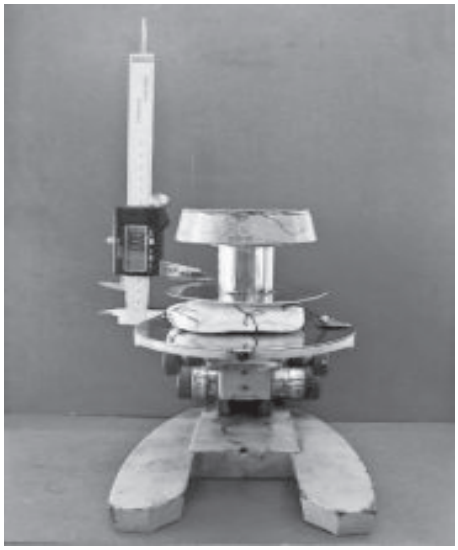

Fig .-2: *Procedure for measuring the flexibility of the specimen.*

Discussion

In the present study, after fixation stage, the mean percentage change in flexibility of skeletal muscle was found larger at cold temperature than at room temperature. Akter[7] found similar result for goat heart. However, Ahmed[4] found opposite result for pig heart where she found change in flexibility was smaller at cold temperature. Ameko et al[8] worked with cow heart where minimal shrinkage (0.93%) occurred at room temperature, although they did not study at cold temperature. According to Henry, Janick and Henry[9], more flexible specimen can be yielded by shortening of fixation time using low concentration of formalin. So, the mean percentage

of change in flexibility of skeletal muscle did not show any consistent result regarding temperature.

In the present study, after dehydration stage, the mean percentage of change in flexibility of skeletal muscle was found larger at cold temperature than at room temperature. However, Akter[7], Paul[3] and Ahmed[4] found opposite result for goat heart, goat tongue and pig heart respectively where they found change in flexibility were smaller at cold temperature and Paul proved her finding. So, it may be said that, the mean percentage of change in flexibility of skeletal muscle during dehydration stage showed inconsistent result regarding temperature.

In the present study, after impregnation stage, the mean percentage of change in flexibility of skeletal muscle was found smaller at cold temperature than at room temperature. Akter[7] and Paul[3] also found similar result for goat heart and goat tongue respectively. However, Ahmed[4] found opposite result for pig heart where she found the change in flexibility was larger at cold temperature. So, it may be said that, the mean percentage of change in flexibility of skeletal muscle during forced impregnation showed an inconsistent result regarding temperature.

In the present study, after gas-curing stage, the mean percentage of change in flexibility of skeletal muscle was found larger at cold temperature than at room temperature. Akter[7] and Ahmed[4] also found similar result for goat heart and pig heart. However, Paul[5] found opposite result for whole and sectioned goat tongue where the change in flexibility was found smaller at cold temperature. According to Tianzhong, Jingren and Kerming[10], low percentage of hardener produces more flexible specimen. Moreover, Mendez et al[11] stated that, longer exposure to gas-curing agent produces less flexible specimen. Thus, it may be said that, the mean percentage of change in flexibility of skeletal muscle during gas-curing showed inconsistent result regarding temperature.

Conclusion

In the present study, the percentage of change in flexibility of skeletal muscle shows better outcome at room temperature than at cold temperature. But the difference between the two groups was not significant. So, further researches with larger sample are recommended to reach a definitive conclusion.

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