

PITFALLS IN AUTOPSY TISSUE SAMPLING

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ABSTRACT

Objective: To determine the pre-analytic factor which lead to autolysis in tissue sample thus hindering the establishment of histo-pathological cause of death in some autopsy tissue samples.

Methods: Data collected from April, 2017 to September, 2018 (18 mo) at Department of Pathology, Dr. S. N. Medical College, Jodhpur.

Results: The study, conducted over 18 mo from April 2017 to September 2018 at the Department of Pathology, Dr. S. N. Medical College, Jodhpur, analyzed 637 autopsy cases. These cases were predominantly from urban areas (70.17%), with a lesser number from rural regions (29.83%). A total of 698 tissue samples were received in various containers, with the majority (95.71%) preserved in formalin. The examination revealed that 38.4% of the samples were not autolysed, 36.1% were partially autolysed, and 25.5% were completely autolysed. The findings highlight a significant impact of the pre-analytic phase on tissue sample quality, notably the interval between death and autopsy, which was less than 24 h in 86.19% of cases, and the time from autopsy to histopathological examination, which ranged up to 1044 d. These factors were pivotal in the preservation status of the tissues, with formalin proving to be the most effective fixative in preventing autolysis, thus facilitating the accurate establishment of histopathological causes of death.

Conclusion: If autopsy specimens are submitted for histopathological examination with proper protocol of transfer to fixative solution and transportation of tissue, this will help in determining the cause of death in majority of the autopsy specimens, therefore avoiding false negative results.

Keywords: Autopsy sampling, Histopathology examination, Autolysis

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INTRODUCTION

Medico-legal autopsy examinations represent a pivotal facet of forensic science, aimed primarily at elucidating the underlying cause of death. These meticulous investigations are initiated upon the directive of legal authorities in response to a wide range of circumstances surrounding the deceased [1]. These include deaths that are shrouded in suspicion, occur suddenly and without apparent explanation, present obscure or enigmatic features, exhibit unnatural characteristics, or are embroiled in litigious and criminal connotations. The overarching objective of medico-legal autopsies is to unearth vital information that can be wielded for legal purposes, ultimately facilitating the course of justice [2].

In the intricate realm of post-mortem examinations, one pivotal tool frequently called into action is histopathology. This technique entails the microscopic examination of tissue samples procured during the autopsy, allowing for a detailed scrutiny of cellular and structural changes [3]. The histopathology examination becomes particularly relevant when the autopsy surgeon detects morbid anatomical alterations within the deceased's tissues. These alterations raise suspicions that they may have played a significant role in the cessation of vital functions, thus warranting a closer and more specialized analysis [4].

Histopathology serves as an invaluable adjunct to the autopsy process, enabling pathologists to discern subtle but crucial details that might otherwise escape notice. By delving into the microscopic realm, it becomes possible to unravel the intricate pathology underlying the cause of death [5]. This, in turn, enhances the accuracy and comprehensiveness of the autopsy findings, adding a layer of clarity and precision to the medico-legal examination. Consequently, histopathology plays an indispensable role in the quest for justice, as it provides a vital bridge between the macroscopic and microscopic aspects of post-mortem investigation, ensuring a more comprehensive understanding of the circumstances surrounding an individual's demise [6].

MATERIALS AND METHODS

Methods

During the period of 18 mo from April, 2017 to September, 2018, a retrospective study was conducted at Department of Pathology, Dr. S. N. Medical College, Jodhpur. 637 cases of autopsy specimens who died of various causes like medical illness, poisoning, drowning, road/railway accidents, burns, etc was done. Sections from the various organs were submitted for processing and then stained with Hematoxyline and Eosin and microscopic examination was done.

All the Post mortem cases received during the period of 18 mo from April, 2017 to September, 2018, were taken in this cross-sectional study conducted at Department of Pathology, Dr. S. N. Medical College, Jodhpur.

After following the proper protocol of receiving the autopsy specimen, the reported data was collected from the department of Pathology and analysed.

RESULTS

In this cross-sectional study conducted at Department of Pathology, Dr. S. N. Medical College, Jodhpur, during a time period of 1 ½ year from April 2017 to September, 2018 a total of 637 cases were studied. Out of the 637 autopsies studied in this study, 447 (70.17%) was conducted in urban area and 190 (29.83%) was conducted in rural area. The ratio of conducting autopsy at Urban: rural region is 2.35: 1. 698 jars were received for 637 cases. The table 1 is showing the number of containers received.

Out of the total 698 samples received 268 (38.4%) were not autolysed, 252 (36.1%) were partially autolysed and 178 (25.5%) were completely autolysed while performing the microscopic examination. The fig. 1 is showing the gross and microscopic examination of the samples received.

Table 1: Number of containers received for the cases under study

Containers	Number of cases	Percent
1 JAR	593	93.09 %
2 JAR	25	3.93 %
3 JAR	12	1.88 %
4 JAR	4	0.63 %
5 JAR	3	0.47 %
Total	637	100 %

For histo-pathological Examination 668 (95.71%) jars having formalin, 29 (4.15%) jars having normal saline and 01 (0.14%) jar having spirit was sent as the solution for transportation of the tissues.

**Fig. 1: Showing the gross and microscopic examination of the samples received**

Time lag between Death and Conducting Post Mortem was <24 h in 549 (86.19%) cases, 24-48 h in 56 (8.79%) cases and >48 h in 32 (5.02%) cases. The maximum time lag between Death and Conducting post-mortem was 54 d.

Time lag between Conducting Post Mortem and receiving of specimen for Histo-Pathological Examination was from 6 d to 1044 d (2 y, 314 d)

DISCUSSION

The foundation of all good histological preparations is adequate and complete fixation. When the organ is removed from the body cavity, it should be thoroughly washed of blood using cool water and placed in fixative immediately [7]. The ideal ratio of fixative-to-volume of tissue when submitting sections for histological analysis is 15-20 to 1. The tissues should not be pressed against each other or the bottom or walls of the container. Refrigerated fixative may be used to slow down autolysis. Formalin is the most common fixative. It is inexpensive, easy to make, penetrates readily and there is no loss of cytologic detail [8]. Specimen color fades on long storage and with very prolonged storage, formalin acidifies to formic acid which precipitates out as black to brown formalin-hematin pigment that needs to be distinguished from bacteria and from anthracotic and dust pigment in macrophages are its disadvantages. 10% buffered formalin solution contains 100 ml of 40% formaldehyde, 900 ml of water, 4 g sodium phosphate, monobasic and 6.5 g sodium phosphate, dibasic (anhydrous). Other fixatives which can be used are Glutaraldehyde, Absolute ethanol, Carnoy's fixative and Bouin's fixative [9].

Transportation: The sample in chain of custody should be transported at the right place by the right person to the right person in the right manner. The inner package must be made of leak-proof glass or plastic containers, preferably with the lid taped in place. Sufficient volume of absorbent material must be placed around the primary receptacle. The primary receptacle and absorbent wrap are then placed in a second leak-proof container [10].

We went through the literature and couldn't find much work done and published by any other author so we were unable to compare

our data results with other authors except for the study by Divya Sharma *et al.* which showed that the postmortem examination was conducted within 24 h in 66.2% (n=57) and a small proportion of cases (n=29) showed time since death of more than 24 h. Our study showed the Time lag between Death and Conducting Post Mortem was <24 h in 549 (86.19%) cases, 24-48 h in 56 (8.79%) cases and >48 h in 32 (5.02%) cases, which was acceptable [11, 12].

CONCLUSION

If autopsy specimens are submitted by trained forensic personnel for histopathological examination with proper protocol of transfer to fixative solution and transportation of tissue, this will help in determining the cause of death in majority of the autopsy specimens, therefore avoiding inconclusive results. With the combined systematic approach of trained Autopsy Surgeon and Forensic Pathologist, we can provide justice to the departed soul.

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AUTHORS CONTRIBUTIONS

All authors have contributed equally

CONFLICT OF INTERESTS

Declared none

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