Preservation of Goat Kidneys Using Low Concentration Formalin for Human Anatomy Learning

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ABSTRACT

Background: Exposure to formaldehyde from a preserved cadaver can cause several health problems in students, lecturers, and laboratory technicians working in medical anatomy laboratories. These conditions include respiratory tract irritation and cancer due to acute and chronic exposure, respectively. Lowering the frequency of contact with formaldehyde from preserved organs helps to ensure the safety of students, lecturers, and laboratory technicians. Therefore, this study aims to determine the effectiveness of low concentration formalin as a preservative solution for goat kidneys. The organ was selected due to its anatomical structure similarity to humans.

Method: Goat kidneys with 50-60 ml of volume were collected, washed, immersed in preservative solution, and stored for two months. The samples were then divided into four groups, which were preserved with different formalin concentrations, namely 30%, 20%, 10%, and 5%. The preservation parameters, namely organ structure and integrity, color, springiness, odor, and mold growth were qualitatively described, while the kidney volume was quantitatively measured.

Results: After the samples were preserved for two months, they were observed, and all groups showed similar characteristics. The kidneys preserved with 5% or 10% formalin showed equal volume before and after two months of preservation.

Conclusion: Low concentration formalin has the same effectiveness in preserving goat kidneys as the high concentration. It also has the potential to be applied in human anatomy learning.

Keywords: cadaver; formaldehyde; goats; kidney

INTRODUCTION

A Cadaver is a preserved dead human body, which is often used for anatomy learning in the fields of medicine¹. From the renaissance period until the 21st century, embalmed cadavers were used for dissection and learning, and have not been fully replaced with the interactive multimedia method until now²–⁵. Compared to artificial anatomical figures, they show a keen precision of anatomical structure and complexity, which portray the anatomy of living humans. Moreover, touching and palpating preserved cadavers by medical students provide a better three-dimensional picture of the body. Students also agreed that learning with this method helps them to understand anatomy completely⁵–⁷. Previous studies revealed that preserved cadavers using typical embalming formulas, such as formalin show a different color, consistency, and organ volume compared to living humans⁸–⁹.

Formalin is a solution containing approximately 40% formaldehyde, which can easily evaporate at room temperature, hence, its exposure to the eyes and respiratory tract is difficult to avoid¹⁰,¹¹. Acute exposure to formaldehyde irritates the eyes and respiratory
tract, which causes sore eyes, throat inflammation, or breathlessness\textsuperscript{12\textendash}15. Its odor also made students uncomfortable while learning anatomy from the preserved cadaver\textsuperscript{12,16}. Several studies also showed that formaldehyde is potentially carcinogenic in chronic exposure in humans and animals, and can cause nasopharyngeal cancer, sinonasal cancer, and leukemia\textsuperscript{10,11,15}.

Formalin is still used for preserving cadavers because it is inexpensive, widely available, and can excellently preserve the detailed structure, which makes the preservative suitable for dissection and anatomy learning\textsuperscript{2,17}. However, its formaldehyde content is toxic, harmful to the human body, and has an unpleasant odor\textsuperscript{11,12}. This makes it important to lower the exposure to formaldehyde in anatomy learning to keep students safe and create a comfortable environment.

Several preservation techniques were developed to eliminate or reduce exposure by using non-formalin-based preservative solutions or simply reducing the concentration\textsuperscript{18}. Low concentration formalin as a component of mixed preservative solution showed satisfactory results with supple and moist preserved organs as well as light colorization, which makes the organs easy to distinguish\textsuperscript{19\textendash}21. It also showed a reduction of unpleasant odor and a good preservation result\textsuperscript{22}. There are currently no studies on the use of pure formalin without other reagents for organ preservation and a comparison of effectiveness among different concentrations. Goat kidneys were selected for this study because they have smaller dimensions as well as similar anatomical structures to humans, namely bean shape, smooth surface, reddish brown color, covered by capsule, and surrounded with perirenal fat\textsuperscript{23\textendash}25. Therefore, this study aims to compare the effectiveness of various concentrations of formalin in preserving goat kidneys.

**METHOD**

**Study Design**

This experimental study was conducted using a qualitative descriptive design, where all parameters were qualitatively described by the observers. A quasi-experimental pre-test and post-test design were adopted to observe the change in kidney volume.

**Sample**

Goat kidneys were used for organ preservation to substitute for human kidneys. The inner part of the organ has a detailed architecture with distinguished renal pyramids, and the alteration caused by preservation or putrefaction will be noticeable. Moreover, goat kidneys showed a similar configuration compared to that of humans and had an acceptable size for macroscopic observation\textsuperscript{24\textendash}26. Samples with a volume of 50-60 ml were collected from a slaughterhouse and processed using preservation procedure in the Anatomy Laboratory of Faculty of Medicine, Jenderal Soedirman University, within eight hours after the animals were slaughtered.

**Preservation Procedure**

Goat kidneys were cleansed by striping from the outer lipid layer and capsule, followed by washing with saline solution, namely 0.9\% NaCl. Their volume was then measured with a measuring cylinder in milliliter (ml) by observing the water displacement before and after they were added\textsuperscript{27,28}. The kidneys were then sliced at Brodell’s line to visualize their inner part, followed by immersion in formalin with concentrations of 30\%, 20\%, 10\%, or 5\% formaldehyde within closed glass containers at room temperature, and stored for two months.
Observation

After two months of preservation, the kidneys were observed for their change of structure, integrity, color, volume, springiness, odor, mold growth, and presence of organ fragments in the preservative solution. Except for volume, these parameters were described qualitatively. The structure and integrity were defined by their shape and wholeness as well as the clear differentiation of the internal structure. The color was defined based on the coloration of the inner and outer part when it was reddish brown as in fresh kidneys or it changed to pale brown as in the preserved variant. Springiness was described by the palpation of the preserved organ compared to the fresh form. The fresh kidneys were springy and tender, while the preserved form can be hard and stiff. The odor was defined as the smell of the preserved sample, namely putrefied or formalin odor, and the strength was defined by observers as strong, mild, or no odor. Mold was described by the presence of patches on the kidney surface, which can be grey, green, or white. Change in the organ volume was defined by difference in volume before and after two months of preservation.

All parameters were observed by two observers without blinding and documented by a digital camera. The classification of the parameters is presented in Table 2.

Ethical Clearance

This study's protocol and ethical clearance were approved by The Medical Research Ethics Commission of The Faculty of Medicine, Jenderal Soedirman University, with reference number 86/KEPK/X/2016.

RESULTS

The goat kidney showed a similar structure to humans, but it was smaller. The outer part of the fresh organ exhibited a reddish-brown color, enveloped by a transparent capsule, and covered with perirenal fat. After incision at the Brodell’s line, the inner part displayed structures called cortex in the outer layer and medulla, including pyramids, in the inner layer, which were similar to human kidneys, as shown in Figure 1.

![Figure 1. Structure of a fresh goat kidney (a) outer part and (b) inner part incised at the Brodell’s line.](image1)

Two months of preservation using 30%, 20%, 10%, and 5% formalin showed decent and comparable results. It was observed...
that they preserved the anatomical structure of the organs without putrefaction in all groups. There were also no fragments in all formalin solutions, which indicates that the integrity was well preserved, but all groups showed a pale brown color for the preserved kidneys, as shown in Figure 2.

Furthermore, the 5% and 10% formalin groups showed no reduction in kidney size after two months preservation, while the volume in the 20% and 30% groups was reduced by 10 ml, as shown in Figure 3. The samples preserved with 5% and 10% formalin had a lesser unpleasant odor compared to the 20% or 30% group. The results showed that the springiness of the preserved kidneys in all groups was reduced when observed through the palpation technique, but the 5% and 10% exhibited better springiness compared to the 20% or 30% formalin. A summary of changes in the parameter of preserved kidneys in all groups is presented in Table 1.

Table 1. Change in characteristic and structure of goat kidney after preservation with formalin of 30%, 20%, 10%, and 5% concentration for two months.

| Preservation Solution | Anatomy Structure | Integrity | Color       | Volume* | Springiness and Hard and Stiff | Odor                      | Mold                      |
|-----------------------|-------------------|-----------|-------------|---------|-------------------------------|---------------------------|---------------------------|
| Formalin 30%          | Well preserved    | Well preserved | Pale brown | Reduced | Hard and stiff                | Strong formalin odor, no putrefied odor | None                      |
| Formalin 20%          | Well preserved    | Well preserved | Pale brown | Reduced | Hard and stiff                | Strong formalin odor, no putrefied odor | None                      |
| Formalin 10%          | Well preserved    | Well preserved | Pale brown | Equal   | Springy and tender            | Mild formalin odor, no putrefied odor | None                      |
| Formalin 5%           | Well preserved    | Well preserved | Pale brown | Equal   | Springy and tender            | Mild formalin odor, no putrefied odor | None                      |

Note: Classification of parameters is described in Table 2.

*Kidney volume before preservation and after two months preservation as shown in Figure 3.

Table 2. Classification of preserved goat kidney parameters.

| Parameters       | Classification     | Explanation                                                                 |
|------------------|--------------------|-----------------------------------------------------------------------------|
| Anatomy structure| Well preserved     | Normal kidney shape (bean-shaped) and clear differentiation of structures    |
| Poorly preserved | Change of kidney shape or unclear differentiation of structures             |
| Integrity        | Well preserved     | Intact kidney without fragmented structures                                  |
| Poorly preserved | Brittle kidney with fragmented structures                                  |
| Color            | Reddish brown      | A similar color to that of fresh goat kidney                                 |
| Pale brown       | Pale brown color appearance                                             |
| Volume           | Equal              | No reduction of kidney volume before and after two months preservation       |
| Reduced          | Reduction of kidney volume before and after two months preservation        |
| Springiness      | Springy and tender  | Springiness and tenderness, which was similar to the fresh kidney           |
| Hard and stiff   | Clear reduction of springiness and tenderness of the kidney was observed to be hard and stiff through palpation |
| Formalin Odor    | No odor            | No specific formalin odor perceived                                          |
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| Parameters       | Classification | Explanation                                      |
|------------------|----------------|--------------------------------------------------|
| Mild odor        | Mild and tolerable formalin odor perceived by the observers |
| Strong odor      | Strong and intolerable formalin odor perceived by the observers |
| Putrefied Odor   | No odor         | No specific putrefied odor perceived               |
|                  | Mild odor       | Mild and intolerable putrefied odor judged by observers |
|                  | Strong odor     | Strong putrefied odor judged by observers          |
| Mold             | None            | No patches on the kidney surface                  |
|                  | Presence        | Presence of patches on the kidney surface         |

| Parameters | Concentration | Before | After |
|------------|---------------|--------|-------|
|            | 30%           | 70     | 50    |
|            | 20%           | 60     | 50    |
|            | 10%           | 50     | 50    |
|            | 5%            | 40     | 40    |

Figure 3. Changes in goat kidney volume before and after preservation with formalin concentrations of 30%, 20%, 10%, and 5% for two months.

DISCUSSION

The use of preserved human bodies as a tool for learning anatomy or surgery has not been replaced by interactive multimedia. Apart from the three-dimensional advantage, it also provides a detailed structure of the human organs, thereby giving complete visualization for the students to study.4,7,9 Anatomy laboratories often use formalin solution without dilution, which contains approximately 40% formaldehyde. However, formaldehyde is harmful to the human body, and it causes respiratory tract irritation and nasopharyngeal cancer in acute and chronic exposures, respectively.10,11,15

This study showed that low formalin concentrations of 5% and 10% showed a comparable preservation result to the 30%. There was also no alteration in kidney volume before and after immersion in the solution for two months, and its springiness was only slightly reduced. Shrinkage after preservation with high concentration formalin was caused by the dehydration process, which involves the transfer of water from the organ to the preservative due to the difference in osmotic pressure between them.9,29 However, all groups experienced decolorization to pale brown color instead of reddish-brown. This color change was caused by the oxidation of globin protein within the capillary vessel in the kidney.9 A lower concentration of formalin can reduce the exposure of formaldehyde in the human body, thereby making it safer to be applied in anatomy learning compared to the higher concentration.

The limitation of this study is that it only examined the preservation of kidneys, but did not explore other organs. Various organs or a whole cadaver must be tested to define the effectiveness of the preservation solutions. The kidneys were also preserved using the immersion method, which can have a different infiltration capacity from the standard method using intravenous injection.2,30 This study cannot conclude on the effectiveness of low concentration formalin for long-term preservation because the kidneys were preserved for only two months. Apart from the volume, other parameters were described qualitatively based on the observers’ interpretation instead of an objective
measurement. Furthermore, this study only focused on the macrostructure of the kidneys, while the microstructure was not observed. Further studies need to be carried out to clear these issues.

CONCLUSION

Low concentration formalin can preserve goat kidneys as effectively as the high concentration variant, hence, it can potentially be applied in human anatomy learning. This study emphasized on the benefit of low concentration preservation solutions to reduce the harmful effect of formaldehyde exposure on students, lecturers, and laboratory technicians.

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Conflicts Of Interest

The authors declared that there is no competing interest.

Author Contributions

SW designed the study construct and wrote the manuscript. SW and KM carried out the study procedures. SW, KM, MM, and FA reviewed the manuscript.

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