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COMPARISON OF VARIOUS MACERATION TECHNIQUES FOR COLLECTION OF HUMAN BONES IN ANATOMY DEPARTMENT

Shahabuddin[1], Anwar Ul Haq[2], Faiza Nadeem[3], Mahvish Javed[1], Iftikhar Ahmad[2], Naheed Siddiqui[2] , Urooj Khushbakht[2]

[1]Department of Anatomy, Khyber Girls Medical College, Peshawar
[2]Departments of Forensic Medicine, Khyber Girls Medical College, Peshawar
[3]Department of Forensic Medicine and Toxicology, Khyber Medical College, Peshawar

ABSTRACT

Objectives: *To compare different maceration processes in terms of efficacy, efficiency, and destructiveness.*

Materials and Methods: *Four techniques were used to clean the bones, following dissection the samples were buried for a period of two years in the department's burial area. Following the excavation, the bones were cleaned; the dismembered portions were manually de-fleshed. The bones were released from the joints.. In the first technique, the bones were immersed in detergent for 24 hours following excavation. The second approach involved dipping bones into a liquid lime solution. In the third technique hydrogen peroxide was used to chemically cure the bones. In the fourth technique 30% hydrogen peroxide and baking soda were combined to create a paste. Applying this, bones were coated for 24 hours. All the four techniques were compared.*

Results: *The procedure utilizing 30% H2O2 was the best in terms of maceration duration, ease of soft tissue removal, and successful procurement with little damage. The new technique, which involved application of paste was made of baking soda and H2O2 was effective and efficient for smaller specimens. It didn't take very long and with almost no damage to the bones.*

Conclusion: *Excellent results were obtained with maceration using 30% H2O2. The newly developed approach worked well for small specimens and save time. The removal of soft tissue was simple and caused no or little bone damage.*

Key words: *Maceration, Bones, Embalmed, Hydrogen peroxide, Baking soda*

INTRODUCTION

Osteology teaching is a vital component of the anatomy curriculum. There is almost no replacement for using real human tissue for teaching students about the human body, despite the fact that tools like textbooks, computer programs, and plastic models can be helpful. When it comes to understanding the insertion of soft tissues and the course of neurovascular systems, as well as teaching three-dimensional osteology, human bones are unmatched[1].

Correspondence:
Dr. Anwar- Ul- Haq
Associate Professor
Forensic Medicine Department, Khyber Girls Medical College, Peshawar - Pakistan
Email: dranwarulhaq23@gmail.com

Software, pictures, or models cannot accurately represent the typical differences in human anatomy. Even if they are essential as documentation, photos cannot substitute touching and feeling a bone. Even if animal remains are easily accessible, a comparative study is impossible without access to human skeletal materials. There has recently been a shortage of bones that may be used for education. For students studying medicine, Forensic medicine, physiotherapy, dentistry, and associated fields, the demand for dried human bones is skyrocketing. There is a limited availability. Therefore, when the supply decreases, a solution must be developed to match the demand. One option could be to recover the skeleton from the dissected corpses. A current option comes from

unused supplies of human skeleton material from the preserved tissue[2].

Maceration is the process of removing all soft tissue and retrieving the bones. There are several maceration methods described in the literature. The goal of all maceration methods is to separate the soft tissue from the bones. In general, multiple stages are necessary including the thorough removal of all soft tissue and procedures to clean and then whiten the bone. First, soft tissue is physically removed. To allow the bone to macerate at a more manageable rate, this involves peeling, gutting, and typically disarticulation. These methods may generally be categorized as boiling, employing chemicals, burying, or utilizing dermestid beetles[3].

It is challenging to remove bones from formalin-fixed cadavers. There are no official guidelines for maceration. The conventional approaches are laborious and time-consuming. The use of fixed specimens in research is feasible. It requires precision to create dry skeletons from preserved tissue without altering the bone since they are difficult to macerate.

As a result, a research was designed to compare the efficacy, efficiency, and destructiveness of the various ways to collect bones. The aim of this study is to apply a variety of techniques to determine which one is best in terms of time commitment, accessibility, and complete bone harvesting from preserved cadavers.

MATERIALS AND METHODS

It is an observational study carried out in Anatomy department (KGMC) Peshawar; since the number of cadavers in the dissection hall Anatomy department is limited so all available dissected parts from four dead bodies, upper limbs, lower limbs, hip region, shoulder region, and vertebral regions were used for the study. After receiving approval from the institutional ethical committee (No 1195) the study was conducted. The study used dissected human cadavers from February 2020 to February 2022 that were provided by the anatomy department of a reputable medical college. Handling tissues and chemicals was done with all due care. Four distinct approaches were used and compared. Following dissection for the first three procedures, the bodies were buried for a period of two years in the department's burial area. After that the cadavers were excavated. The bones were washed.

In the first technique, the bones were immersed in detergent for 24 hours following excavation. The remaining soft tissue was carefully removed with a brush. The bones were clean and dried. The second approach involved dipping bones into a liquid lime solution for 24 hours. They were then cleaned and dried. This approach involved dipping bones into a liquid lime solution.

In the third technique, the bones were immersed in detergent for 24 hours following excavation. Then hydrogen peroxide was used to chemically cure the bones. In 15 L of water, 2 liters of 30% H2O2 was dissolved. The bones were covered for 24 hours after being dipped. After that, they were rinsed and sun-dried.

The fourth technique included de-fleshing the dissected portions. They weren't laid to rest. Using 100 g of baking soda and 30 g of H2O2, a paste was made. The bones were covered for 24 hours after this paste was applied. They were cleaned and dried the next day. The time needed for maceration, the simplicity of removing soft tissue, and the extent of bone tissue procurement or damage were all taken into consideration when comparing the procedures. The following observations were recorded.

In the first method where detergent was used on already buried bones for period of two years detergent made it simple to remove soft tissue and clean it. Bones had a dark hue. Soft tissue at the ends had to be manually removed since it could not be detached itself.

Bones were not damaged in any way and bone colour was darker.

In the second method quick lime was used in which more than two years are needed for maceration. It was ineffective to remove soft tissue with ease of access. The bones had a layer of chalk on them (Figure 1). The remaining tendons and ligaments were not softened. The tissue was left in place. Bones were not damaged in any way. Soft tissue could not be removed. The bones were left with a chalky deposit on them.

In the third method hydrogen peroxide was used in which maceration period was over two years. It was simple to remove soft tissue and clean it. The tone of the bones was lighter. Figures 2 and 3 show

that just a few of the bones required brushing to appear clean. No bone damage was observed; bones with lighter tones have a pleasing appearance to the touch.

In the fourth method baking soda (NaHCO3) and H2O2 in which there was no burial. The entire process took less than a week. The cadaver was buried and the bones were excavated in the previous three techniques, which typically took two years. Soft tissue was easily accessible and much of it falls off with little effort. Using a scalpel, some tissue had to be manually removed. No bone damage was ob-

Fig 1: Tibia bone showing chalky deposits after treatment with quick lime

Fig 2: Bones removed after 2 years of burial

Fig 3: Sacrum after treatment with H_2O^2

Fig 4: Bones treated with paste of baking soda and H_2O^2

Table 1: Showing various methods used in maceration with Advantages and Disadvantages

Method Type	Technique	Advantages	Disadvantages
Method 1	Burial and use of Detergent	Fairly Inexpensive Requires Minimal Training	Time Consuming Requires Manual Extraction
Method 2	Burial and use of Quick Lime	None	Time consuming Removal Of soft Tissue impossible Could not break ligaments, tendons Chalky deposit on Bones
Method 3	Burial and use of H_2O^2	Fairly Inexpensive Requires Minimal Training Effective Cleaning Visually appealing Lighter colored Bones No damage to Bones	Time Consuming
Method 4	Use Of Hydrogen peroxide and Baking Soda Paste with no Burial	Lot of Bones can be Done together Process takes less than a week Fairly Inexpensive Effective Cleaning Visually appealing Bones No damage to Bones	Requires Extensive knowledge

served and the bones were clean and lighter in tone, as shown in Figure 4.

RESULT

Dissected parts from four dead bodies were studied. Table 1 shows the advantages and disadvantages of various techniques. The 30% H2O2 approach was the best in terms of maceration duration, soft tissue removal simplicity, and successful procurement with little to no bone damage. The chemicals are freely accessible and reasonably priced. The novel technique of applying a paste of baking soda and H2O2 was particularly successful for smaller specimens. The time needed was really brief. No interment was needed. Thus, we may draw the conclusion that the hydrogen peroxide procedure can be effectively employed to obtain whole skeletons from dissected bodies.

DISCUSSION

The main aim of the current study was to compare different maceration processes in terms of efficacy, efficiency, and destructiveness, in order to determine the best way to extract human bones from an embalmed cadaver. Our study shows hydrogen peroxide procedure can be effectively employed to obtain whole skeletons from dissected bodies. Over the past few decades, there has been a sharp rise in the demand for real bone specimens among professionals and students working in both established fields and emerging sciences. For the purposes of teaching human anatomy, Philip Tobias stressed the value of using human skeletons. This includes teaching anatomy to students of dentistry, medicine, occupational therapy, physiotherapy, pharmacy and nursing, as well as, primatology, general morphology, Forensic medicine and comparative anatomy[4]. The number of new disciplines that depend on, this educational content has rapidly increased and expanded. Fields, like DNA study, forensic sciences, and molecular biology, are worth mentioning[2].

The department had to experiment with several maceration techniques to accommodate the growing demand for bones. Given the viability, costs, and other considerations, four techniques were selected for the current investigation. Even though boiling of bones is an easy and time-tested method, it was not recommended because of the unpleasant smells and environmental risks. This drawback prevents it from being taken anywhere.

In our study the 30% H2O2 approach was the best in terms of maceration duration, soft tissue removal simplicity, and successful procurement with little to no damage. It showed many bones can be processed together; the process is inexpensive and takes less than a week. It is also associated with effective cleaning and yields visually appealing bones.

No damage to Bones. The chemicals are freely accessible and reasonably priced. Multiple bones can be grouped up at once. The entire technique may be completed by one skilled individual. Previous investigations came to the same conclusion about this method's effectiveness[5]. The bones were kept buried for two years since burial is required. Burying the bones close to the surface in the topsoil will shorten the time needed for burial[6].

In our study we concluded that the hydrogen peroxide and baking soda procedure can be effectively employed to obtain whole skeletons from dissected bodies and H2O2 was particularly successful for smaller specimens. In their method of skeleton preparation, Snyder, Burdi, and Gaul used a quick-acting substance they called anti-formin that was made by mixing bleaching powder and sodium carbonate[6]. Triaca used different maceration techniques on burnt remains[7]. Modern research warns about using bleaching chemicals, due to its harmful qualities[8-11].

It has been demonstrated that using bleaching powder to prepare skeletons causes bone structure to become unstable even after the solution has been removed[7,9,10]. Researchers have noted that although sodium hypochlorite is excellent in removing fat and lightening specimens, the resulting bone will be fragile and may crumble to dust. In the literature Hydrogen peroxide has been recommended as a substitute for sodium hypochlorite[9,12]. The work done by Soni supports our findings[13] whereas Horák , described different light sources for degreasing and bleaching the bones[14].

Proteinaceous materials can be softened with sodium bicarbonate. Sodium bicarbonate consistently came out on top in tests evaluating which chemical was the fastest and simplest to de-flesh[2-8]. In order to achieve quick and efficient cleaning, hydrogen peroxide and sodium bicarbonate were combined. Help from a qualified individual is necessary to de-

flesh and disarticulate the bones using this approach.

The reagents are easily available. It goes by quickly. No burial is necessary. Applying this technique to the axial skeleton may be challenging. However, for little dissected specimens, it can be the best approach.

CONCLUSION

AFor best results, 30% H2O2 must be used during maceration. The newly developed method works well for tiny specimens and saves time. The removal of soft tissue was simple and caused no or little harm to the bones. For little dissected specimens, it can be the best approach. These simple procedures will make it easier for medical colleges to obtain bones from preserved tissues.

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