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TECHNICAL NOTE

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A Fast and Safe Non-Bleaching Method for Forensic Skeletal Preparation

ABSTRACT: Over the last three decades, forensic anthropologists increasingly have consulted on fleshed human remains cases in which the examination of skeletal elements is critical in answering questions of identification and the circumstances of death. This was certainly the case at the Human Identification Laboratory in Tucson, Arizona. As the caseload increased, it became clear that a method for defleshing human remains was needed in order to expeditiously expose the osseous surfaces for analysis, yet at the same time, preserving the evidentiary nature of the material. As a result, a fast, safe and economical method for defleshing human remains and producing high quality, degreased skeletal elements was developed. This non-bleaching cooking method utilizes chemicals that are easily obtained and inexpensive standard household ingredients that can be purchased at most grocery stores.

KEYWORDS: forensic science, forensic anthropology, skeletal preparation

Forensic anthropologists and forensic pathologists are frequently faced with human remains cases in which the examination of skeletal elements is critical in answering questions of identification and the circumstances of death. In many of these cases, the remains are fleshed, which necessitates the employment of skeletal preparation methods to remove the soft tissues in order to expose the osseous surfaces for analysis. Forensic cases normally require that these methods expeditiously remove soft tissue while preserving the evidentiary nature of the skeletal elements.

A fast, safe, and inexpensive non-bleaching method for removing tissue to recover osseous remains from a forensic context was developed at the Human Identification Laboratory when it existed at the University of Arizona. This three-stage cooking procedure to remove soft tissue and degrease bone evolved over a period of 30 years. This method produces high-quality skeletal specimens for analysis, documentation, and curation. In addition, it is effective on a wide range of cases, including fresh, decomposing, mummified, skeletonized and formalin-fixed human remains. With regard to the overall time it takes for fleshed remains to become a fully degreased and stabilized specimen, this may represent the most efficient method of skeletal preparation.

The evolution of this tissue removal and degreasing procedure at the Human Identification Laboratory was not documented. However, a number of cooking procedures were tried and rejected over the years because of harsh chemicals in the rendering processes, such as chlorine bleaches, various other oxidizers, and even a proteolytic enzyme (papain) derived from the papaya plant. Most such chemicals were rejected simply because their effect on bone was deleterious and continued to react following of the cooking process. Specifically, bleaching agents (e.g., bleach and hydrogen peroxide) degrade bone by consuming calcium, and should not be part of any method that processes human remains from a forensic context. Another major drawback of oxidizers such as 30% H_2O_2 (hydrogen peroxide) is the endangerment of workers handling the chemicals, their potential to support combustion in case of a fire, and their difficulty to store. These drawbacks, plus their much greater expense, were significant in the decision to forego the use of such chemicals.

Materials and Methods

The Human Identification Laboratory gradually settled on a fairly simple procedure for removing soft tissue to recover osseous remains. The chemicals used are easily obtained and inexpensive standard household ingredients that can be purchased at most grocery stores. This eliminates the need to purchase more expensive A. R. Grade (Analytical Reagent) chemicals from a commercial vendor.

The "tried and true formula" for defleshing is as follows: 1. Water to submerge the specimens; 2. Powdered detergent (e.g., Alconox®, Tide™, Cheer™), approximately 20 cc per 2 L water; 3. A powdered sodium carbonate (e.g., Arm and Hammer Washing Soda™), approximately 20 cc per 2 L water. 4. For degreasing the bone use: Liquid household or sudsy ammonia, approximately 150 mL per 2 L water.

The specimens to be cooked are placed in the water-detergent-carbonate solution over low heat such that the solution never reaches the rolling boil, but is nevertheless at or just below a low simmer. During this stage, the enzyme-active ingredients in the so-

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lution break down the soft tissues. The specimen may have to be cooked in several sequential changes of the solution to remove the soft tissues and most of the fats, whether dealing with postcranial or cranial remains. Decomposing and mummified remains are more quickly rendered than is a fresh specimen. Far more difficult to reduce are embalmed remains where the fats have undergone chemical changes and have permeated the bone itself.

After each detergent/carbonate cooking episode, the specimens must be thoroughly rinsed in running water (If a mouth with teeth is being rendered, it is imperative that the worker slowly adds tepid water to the cooking pot when changing solutions so that the dental enamel does not fracture). At this point in the process, the adhering tissues have begun to soften and must be manually removed. The more tissue removed prior to cooking means there is less to remove during the cooking process. It should be noted that no amount of cooking or brushing will remove articular cartilage. It has been our experience that the best way to remove cartilage is to carefully scrape it off with a blunt scraping tool (e.g., Scoopula®, Fisher Scientific). It is important to bear in mind that great care must be taken not to alter the bone when mechanically removing tissue with sharp implements. It is recommended that the novice practice on fresh non-human bones (both heavy cortex limb bones and thin cortex vertebral bodies) before attempting to render evidentiary material. A local butcher shop would be a quick and easy source for such materials.

Following the last detergent/carbonate cooking, the specimen should be thoroughly rinsed and placed into the ammonia and water solution for continued low cooking to facilitate degreasing. The fats will pool on top of the water during this last stage of cooking and should be skimmed off before giving the specimen its final rinse.

Any remaining soft tissues still adhering to the bone will have expanded during the ammonia stage, and can easily be removed with a medium stiff bristle brush. This degreasing stage is critical especially for skeletal remains that will be curated for long periods of time. It is recommended that several degreasing cookings be undertaken.

In certain cases, the forensic scientist will need to retain the prepared skeletal specimen for evidentiary purposes. In that event, it is recommended that efforts be made to stabilize the bone to prevent future deterioration. The final stage in this method utilizes a solution of Vinac (available from AirProducts and Chemicals Inc., Allentown, PA) and methanol to impregnate the bone. In a chemical resistant tank (e.g., Nalgene), 210 g of Vinac to each gallon of methanol is the recommended solution. The bones are soaked in this solution for several minutes, removed and placed on a mesh screen to dry.

A Survey of Defleshing Methods

There are four categories of skeletal preparation methods employed for the purposes of forensic evaluation and/or museum curation: cooking, water maceration (cold and warm), chemical maceration, and carrion insects. Within each of these categories are numerous methodological combinations that result in an exponential number of skeletal preparation techniques.

Cooking has been successfully used in both museum preparation and forensic contexts. Hangay (1) states that cooking human remains is the quickest way to remove the flesh from the bones, although cautions that overcooking can ruin the specimen.

Good results have been reported by cooking fleshed remains in enzyme-active laundry detergents (2–4). Repeated cooking with certain enzyme-active laundry detergent dilutions has been employed for effective removal of formalin-fixed tissues (2). Another method utilizes a dilution of sodium perborate and water that is allowed to cook for several hours before the mechanical removal of tissues (5). This is generally accomplished by repeatedly cooking the materials until the tissue is completely removed.

Water maceration is the slowest and least aggressive method for processing human remains. Due to the time constraints placed on the forensic anthropologist or forensic pathologist, water maceration is the least ideal method of skeletal preparation when doing casework. If the purpose of processing human remains is research based, however, the slow decomposition that occurs during water maceration improves the chances that bone details will be preserved. Water maceration in an enclosed container allows the remains to decompose at warmer temperatures, thereby increasing natural bacterial processes (1,6–9). The temperature of the water can be increased to no more than 37°C(1).

Chemical maceration methods can produce analyzable remains within a few hours, although there is increased risk to both the skeletal evidence and the forensic scientist. To prevent bone damage, the chemical concentrations and the duration of maceration must be closely monitored. In addition, a well-ventilated area is necessary due to the noxious nature of many of the chemicals (1). Hangay (1) summarizes various other chemical maceration techniques, including an ammonium and sodium hypochlorate combination, sodium perborate, potassium hydroxide, sodium hydroxide (10), trypsin, pepsin and papain. The antiformin technique, proposed by Snyder et al. (11), may be the fastest chemical maceration technique. With this technique, formalin-fixed cadaveric specimens and human remains from forensic contexts can be defleshed within an hour. However, great care must be taken while employing the antiformin technique or the bone will begin to disintegrate. Even so, there is a tendency for flaking of cortical surfaces due to the use of bleaching agents. Stephens (12) proposes a chemical maceration method using bleach and sodium hydroxide for the rapid removal of tissues from the bone that may take from one to a few days for complete tissue removal, dependent on the degree of decomposition. As discussed above, great care must be taken when using bleach on bone.

Many museum specimens are prepared using carrion insect larvae, usually for small and medium sized specimens. There are numerous techniques for the processing of skeletons using dermestids (9,13–15), while others recommend the use of mealworms (16). Dermestid cleaning requires initial skinning and defleshing of the remains, after which the remaining flesh is allowed to dry for several hours. The remains are then placed on a shelf in a humidity and temperature controlled instrument and covered with cotton batting or some other material. After the specimen is cleaned, it is soaked and rinsed to eliminate exuviae, frass, and living dermestids. The advantage of this method is that it results in pristine and durable skeletal material. The disadvantages are that the bone retains odors and fatty acids, it takes much more time, and you need to keep the larvae alive during periods of inactivity.

Degreasing Methods

If the specimen is to be retained for evidentiary or research purposes, further degreasing of the bone is recommended after defleshing. Chemicals such as acetone (12), trichloroethylene, hydrogen peroxide and a variety of dishwashing products have been used to degrease bone by cooking or macerating in dilutions of these compounds (5). Bleaching agents have also been used to degrease and whiten bone (11). Again, the problem with using bleaching agents is their deleterious effect on bone as it continues to react even after processing. Furthermore, although bone whitening is im-

portant in a museum context, it is not a goal of the forensic scientist. Therefore, it is our recommendation that the best method for degreasing is cooking the skeletal specimen in a solution of water and household ammonia.

Summary

Choosing the appropriate skeletal preparation method depends on the preparator's goal, the amount of time available, the condition of the specimen, and the type of facilities. The goal in most forensic cases is to quickly deflesh human remains and at the same time preserve the evidentiary nature of the skeletal elements. The cooking method proposed in this technical note provides an expeditious procedure whereby fully defleshed and degreased bone can be rendered within a day or two. This method can be used on remains of varying postmortem conditions, ranging from skeletonized to fresh bodies. The major strength of this method is the safety of the ingredients employed, both to the forensic scientist and to the skeletal specimen. First, there are no volatile compounds that produce noxious fumes. Second, the chemicals in our method are non-bleaching, and therefore do not consume calcium. Finally, this fast and safe method produces durable, high-quality skeletal specimens for analysis, documentation, and curation.

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