



## EFFICACY OF CHLORINE-ENHANCED BURIAL VERSUS CONVENTIONAL BURIAL FOR BONE CLEANING: A CONTROLLED COMPARATIVE STUDY

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### ABSTRACT

Bone cleaning methods must balance efficacy with bone integrity preservation, yet comparative data on traditional burial techniques remains limited. This study compared conventional burial (9 months) with chlorine-enhanced burial (6 months) at J. S. Ayurveda Mahavidyalaya, Nadiad, examining soft tissue influence on preservation outcomes. Forty cadaveric bone specimens were used: 20 for conventional burial and 20 for chlorine-enhanced burial, each method including 10 bones with soft tissue attachments and 10 without. Conventional burial used ash paste post-treatment; chlorine-enhanced burial incorporated cow dung medium and Ayurvedic herbal decoction containing Aristaka, Ingudi, Mudga, and Shrivati. Soil analysis using EDTA extraction and atomic absorption spectrophotometry measured micronutrients. Results demonstrated that soft tissue attachments were essential for preservation in both methods, while bones without tissue showed severe degradation. Chlorine-enhanced burial with soft tissue proved optimal, accelerating tissue removal by 50% (3 versus 5-6 months) while maintaining structural integrity. The Ayurvedic herbal decoction combines natural saponins Aristaka (*Sapindus mukorossi* Gaertn.), Ingudi (*Balanites aegyptiaca* (L.) Delile), Mudga (*Vigna radiata* (L.) R.Wilczek), and Shrivati (*Mussaenda frondosa* L.), as described in Bhavaprakasha Nighantu to effectively remove chemical residues, neutralize chlorine deposits, dissolve organic matter, restore natural bone whiteness and luster, and provide antimicrobial protection while maintaining structural integrity.

**KEYWORDS :** Bone Cleaning, Burial Methods, Soft Tissue Preservation, Chlorine Enhancement, EDTA Soil Analysis

### INTRODUCTION

Bone cleaning methods must balance cleaning efficacy with preservation of skeletal integrity, and while traditional burial techniques offer cost-effective alternatives to modern maceration methods, comparative data on their efficacy remains limited.<sup>1, 2</sup> This study compared conventional burial with chlorine-enhanced burial techniques for bone cleaning across forty cadaveric bone specimens from J S Ayurveda Mahavidyalaya Nadiad, stratified by soft tissue presence (n = 20 each) and burial method. Method 1 (conventional burial, 9 months) used ash paste post-treatment, while Method 2 (chlorine-enhanced burial, 6 months) incorporated pre-burial chlorine application, cow dung medium, and post-treatment with traditional Ayurvedic herbal decoction containing Aristaka (*Sapindus mukorossi* Gaertn.), Ingudi (*Balanites aegyptiaca* (L.) Delile), Mudga (*Vigna radiata* (L.) R.Wilczek), and Shrivati (*Mussaenda frondosa* L.), as described in Bhavaprakasha Nighantu.<sup>3,4,5</sup>

The herbal decoction's mechanism of action derives from multiple phytochemical properties: *Sapindus mukorossi* contains triterpenoid saponins that act as natural surfactants, emulsifying lipid residues and facilitating removal of adhered soil particles without causing surface erosion;<sup>4</sup> *Balanites aegyptiaca* provides antimicrobial saponins and flavonoids that prevent microbial colonization while enhancing surface smoothness;<sup>5</sup> *Vigna radiata* contributes proteolytic and cleansing activity helpful in the removal of residual soft tissue; and *Mussaenda frondosa* offers glycosides and mild chelating compounds that assist in whitening and reducing discoloration.

Results demonstrated that soft tissue attachments play an essential role in protecting bones during burial-based cleaning by buffering environmental fluctuations and limiting direct soil-bone interaction. Conventional burial with soft tissue (Method 1) showed better preservation, whereas burial without soft tissue resulted in increased brittleness, cracking,

and discoloration. Chlorine-enhanced burial with soft tissue attachment (Method 2) reduced processing time while maintaining structural integrity, supported by the synergistic cleansing actions of the herbal formulation. In contrast, Method 2 without soft tissue attachment produced surface burns, white deposits, and fragility due to direct chlorine exposure. Overall, Method 2A with soft tissue attachment demonstrated optimal efficiency and preservation when combined with Ayurvedic post-treatment cleansing.

### Objectives

1. To compare the cleaning efficacy of conventional burial with chlorine-enhanced burial methods
2. To assess the impact of soft tissue attachments on bone preservation and structural integrity
3. To evaluate discoloration patterns and surface quality across different bone types
4. To document the restorative effects of Ayurvedic herbal decoctions in post-burial bone treatment

### MATERIALS AND METHODS

#### Study Design

Comparative experimental study conducted at J S Ayurveda College Nadiad campus over 6-9 months.

#### Materials

##### Bone Specimens:

- 40 bone specimens of varying types (long bones, flat bones) obtained from cadaveric sources
- 20 specimens allocated to Method 1: 10 with soft tissue attachments and 10 without soft tissue attachments
- 20 specimens allocated to Method 2: 10 with soft tissue attachments and 10 without soft tissue attachments

##### Burial Infrastructure:

- Two separate burial pits: 5×5×5 feet dimensions
- Natural soil environment
- Controlled depth layering system

**Treatment Materials:**

**Method 1:**

- Ash pastes for post-burial treatment
- Hot water for initial washing

**Method 2:**

- Chlorine pastes for pre-burial application
- Cow dung (10 kg) as burial medium enhancement
- Herbal decoction containing:
  - Aristaka fruits (*Sapindus mukorossi*) - natural saponin source
  - Ingudi fruits (*Balanites aegyptica*)
  - Mudga (*Vigna radiata*)
  - Shrivati flowers and leaves (*Mussaenda frondosa*)

**Methodology**

**Method 1: Conventional Burying (9 months)**

**1. Day 1 - Burial Phase:**

- Bone specimens (with and without attachments) placed in pit
- Arranged in separate layers with soil separation
- Pit covered and left undisturbed

**2. Month 9 - Retrieval Phase:**

- Bones excavated from burial site
- Washed thoroughly with hot water
- Thin layer of ash paste applied uniformly
- Specimens kept for 24 hours with ash treatment

**3. Post-Treatment:**

- Final washing with tap water
- Documentation of condition

**Method 2: Chlorine-Enhanced Burying (6 months)**

**1. Day 1 - Pre-treatment and Burial:**

- Thin chlorine paste layer applied to all specimens
- Bones placed in pit with cow dung (10 kg) integrated in layers
- Soil layering maintained for separation

**2. Month 6 - Retrieval Phase:**

- Bones collected and washed with hot water
- Immersed in herbal decoction for 24 hours
- Specimens rubbed with Aristaka rind
- Final washing with tap water

**Data Collection**

**Parameters Monitored:**

- Tissue removal completion rate
- Bone structural integrity
- Surface discoloration patterns
- Insect activity and biological cleaning
- Surface texture and condition
- Odor emission (qualitative assessment)
- Signs of chemical degradation

**Documentation:**

- Photographic evidence at Day 1, mid-point, and completion
- Comparative analysis of bone types
- Soil composition testing

**RESULTS**

**Method 1: Conventional Burying Method (9 months)**

**Bones WITH Soft Tissue Attachments (Group A)**

**Decomposition Timeline:**

- Complete tissue removal: 5-6 months
- Natural biological cleaning aided by insect activity (ants, beetles)
- Gradual breakdown without chemical intervention

**Physical Characteristics:**

- **Structural Integrity:** Well-preserved, bones remained intact
- **Surface Condition:** Smooth with slight staining
- **Discoloration:** Mild yellow-brown staining pattern
- **Odor:** Strong during first 3 months, significantly diminished thereafter

- Overall quality: Excellent preservation with minimal degradation

**Bones WITHOUT Soft Tissue Attachments (Group B)**

**Physical Characteristics:**

- **Structural Integrity:** Fragile and brittle, increased breakage risk
- **Surface Condition:** Rough, flaky texture with visible cracks in multiple areas
- **Discoloration:** Severe blackening with persistent soil residue
- **Insect Activity:** Minimal (primarily soil-based effects only)
- **Environmental Damage:** Significant susceptibility to black spot discoloration

**Bone Type-Specific Observations:**

**Flat Bones (Scapula, Hip Bones):**

- More prone to physical breakage due to structural composition
- Mild discoloration when detached from soft tissue
- Relatively better preservation than long bones

**Long Bones:**

- Ends deteriorated within 6 months
- Faster degradation rate compared to flat bones
- Greater vulnerability to environmental factors

**Method 2: Chlorine-Enhanced Burying (6 months)**

**Group A: Bones WITH Soft Tissue (Chlorine-treated)**

**Decomposition Timeline:**

- Complete tissue breakdown: 3 months (50% faster than Method 1)
- Enhanced dissolution in soil environment

**Physical Characteristics:**

- **Ease of Tissue Removal:** High - tissue partially dissolved
- **Surface Reaction:** Mild bleaching effect, moderate chalkiness
- **Structural Integrity:** Intact with some moisture density loss
- **Odor Emission:** Low (chlorine masked decomposition smell)
- **Chemical Effects:** Controlled, minimal damage

**Group B: Bones WITHOUT Soft Tissue (Chlorine-treated)**

**Physical Characteristics:**

- **Surface Reaction:** Severe surface burns evident
- **Chemical Deposits:** White powdery deposits covering surfaces
- **Structural Integrity:** Increased fragility with signs of erosion
- **Degradation:** More extensive than Group A
- **Surface Quality:** Compromised due to direct chlorine exposure

**Herbal Decoction Effects (Method 2 Post-Treatment)**

The traditional decoction demonstrated multiple beneficial properties:

**Cleaning Action:**

- Effective removal of residual soil and organic matter
- Natural saponins from Aristaka provided gentle cleansing
- Reduced chemical residue from chlorine treatment

**Decolorization:**

- Neutralization of discoloration pigments
- Restoration of natural bone colour
- Reduction in white powdery deposits

**Surface Restoration:**

- Improved overall appearance
- Enhanced surface smoothness
- Better aesthetic outcome

Parameter	Method 1A (With Tissue)	Method 1 B (Without Tissue)	Method 2A (With Tissue)	Method 2B (Without Tissue)
Duration	9 months	9 months	6 months	6 months
Tissue Removal	5-6 months, natural	N/A	3 months, accelerated	N/A

Integrity	Excellent	Poor (brittle)	Good (slight moisture loss)	Com-promised (fragile)
Dis-coloration	Mild yellow-brown	Severe blackening	Mild bleaching	Severe (white deposits)
Surface Quality	Smooth	Rough, cracked	Moderate chalkiness	Burns, erosion
Odor	Strong (3 mo), then reduced	None	Low (masked)	None

**DISCUSSION**

**1. Critical Role of Soft Tissue Attachments**

The presence of soft tissue attachments emerged as the most significant factor in bone preservation across both methods. Soft tissue provides:

- Protective barrier: Shields bone surface from direct soil contact and chemical exposure
- Moisture regulation: Maintains hydration balance, preventing excessive drying and cracking
- Biological cleaning advantage: Attracts decomposer organisms that clean bones naturally without damaging structure
- Chemical buffer: Reduces direct impact of chlorine and soil chemicals on bone matrix

**2. Environmental Factors in Bone Degradation**

Bones without soft tissue showed heightened vulnerability to:

- Soil minerals: Direct contact caused black spot discoloration and staining
- Moisture fluctuation: Led to surface cracking and flaking
- Microbial action: Without organic tissue, microbial activity had minimal beneficial cleaning effect
- Chemical exposure: Particularly severe in chlorine method

**3. Method Comparison**

**Method 1 (Conventional Burying):**

- Advantages: Natural process, minimal chemical damage, excellent structural preservation with attachments
- Disadvantages: Longer duration (9 months), strong odour in early stages, requires biological cleaning period
- Best for: When preservation quality is priority over speed

**Method 2 (Chlorine-Enhanced):**

- Advantages: Faster tissue removal (3 months), reduced odour, shorter overall timeline
- Disadvantages: Risk of chemical damage, requires careful application, herbal post-treatment essential
- Best for: When time constraints exist and post-treatment is feasible

**4. Herbal Decoction Efficacy**

The traditional Ayurvedic decoction formulation demonstrated remarkable restorative properties:

- Sapindus mukorossi (Aristaka): Natural saponins provided gentle but effective cleaning action
- Synergistic effect: Combined herbs addressed multiple cleaning challenges simultaneously
- Chemical neutralization: Reduced chlorine residue and restored bone appearance
- Traditional wisdom validation: Classical formulations proved scientifically relevant

**Bone Type Vulnerability**

**Flat Bones (Scapula, Pelvis):**

- Structural fragility makes them breakage-prone
- Thinner cortical bone more susceptible to environmental damage
- Better preserved with soft tissue protection

**Long Bones (Femur, Tibia, Humerus):**

- Ends deteriorate faster than shafts
- Cancellous bone at epiphyses more vulnerable
- Require longer burial period for complete cleaning

**Limitations**

1. Seasonal variation: Study conducted in specific climatic

conditions at JS Ayurveda College

2. Soil composition: Results specific to campus soil characteristics
3. Bone type diversity: Limited representation of all skeletal elements
4. Controlled environment: May not reflect all natural burial conditions

**CONCLUSIONS**

1. **Soft Tissue Protection:** Soft tissue attachments provide superior bone preservation, contrary to conventional assumptions, by offering protective buffering against environmental and chemical damage.
2. **Method Performance:**
  - o **Method 1A** (conventional with tissue): Excellent preservation, minimal discoloration, 5-6 months tissue removal
  - o **Method 1B** (conventional without tissue): Severe brittleness, cracking, and blackening despite 9-month duration
  - o **Method 2A** (chlorine-enhanced with tissue): Optimal—50% faster cleaning (3 months), intact structure, enhanced aesthetics with herbal treatment
  - o **Method 2B** (chlorine-enhanced without tissue): Severe burns, fragility, and surface erosion
3. **Method Selection:** Method 1 for maximum preservation quality (academic/museum use); Method 2 (chlorine-enhanced with tissue) for time-sensitive applications with superior outcomes (forensic contexts).
4. **Chemical Caution:** Chlorine accelerates cleaning but requires soft tissue protection and herbal post-treatment to prevent structural damage.
5. **Traditional Validation:** Ayurvedic herbal decoction effectively enhanced luster, whiteness, neutralized discoloration, and removed chemical residues, validating traditional knowledge.
6. **Environmental Impact:** Soil composition, moisture, and microbial activity significantly influence outcomes, requiring site-specific optimization.

**Recommendation:** Method 2 A (chlorine-enhanced burial with soft tissue and herbal post-treatment) is optimal, combining efficiency (6 versus 9 months), superior preservation, and enhanced aesthetics.

**Images:**





### Declaration of Patient Consent

The authors certify that all bone specimens used in this study were obtained from cadaveric sources through voluntary body donation programs at JS Ayurveda College, Nadiad, with appropriate consent and legal documentation. Institutional Ethics Committee approval was obtained. All bones were handled in accordance with the Anatomy Act and relevant regulations, maintaining dignity and confidentiality of donors. The authors acknowledge the noble contribution of body donors and their families to medical education and research.

### Conflicts of Interest

There are no conflicts of interest.

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