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Experimental Forensic and Bioanthropological Aspects of Soft Tissue Taphonomy: 1. Factors Influencing Postmortem Tissue Desiccation Rate

REFERENCE: Aturaliya S, Lukasewycz A. Experimental forensic and bioanthropological aspects of soft tissue taphonomy: 1. Factors influencing postmortem tissue desiccation rate. J Forensic Sci 1999;44(5):893–896.

ABSTRACT: Euthanized rats' carcasses were exposed in an environmental chamber to multiple variables including: (1) position, (2) enveloping clothing, and (3) soil interment in an effort to determine the individual variables' effect on postmortem rate of body and visceral organ water loss. Results indicated that body water loss was enhanced by a horizontal position versus vertical, probably because of wider spread of bacteria- and enzyme-laden abdominal fluid secondary to diaphragm digestion with consequent greater tissue digestion and liquefaction. Clothing also accelerated the desiccation rate. Desiccation was about equally as effective by soil interment as by air exposure, though simulating windy conditions by tripling the air flow rate resulted in much more rapid desiccation in the air-exposed specimen. These studies suggest that the single most important factor influencing postmortem body water loss rate is the environment at the skin surface that acts to enhance or impair water removal from the skin surface and thus influences the water concentration gradient between the skin and underlying deeper tissues.

KEYWORDS: forensic science, water loss, body position, clothing effect, mummification, taphonomy

In 1940, a 12-page article by I.A. Efremov identified a little-studied area of paleontology and called it taphonomy (Greek *taphos:* grave and *nomos:* ordinance or law), the study of postmortem mechanisms that transform the state of body tissues (1). With this modest tract he launched a subdiscipline of his scientific field that is still expanding today. While Efremov concentrated primarily on the mechanisms and agents that transformed organic into fossilized tissue, later workers subdivided the process into distinct stages, some of which deal only with the earlier phases. Most of the taphonomy literature, however, deals with skeletal remains.

Soft tissue alterations beginning immediately following death and occurring during the subsequent month are of greater forensic and bioanthropological interest than those involving fossilization. While a limited number of experimental reports focus on this period, observations made on postmortem soft tissue changes during this earlier interval rely heavily on the uncontrolled circumstances presented in medicolegal cases (2–8). In such a milieu, multiple destructive agencies (temperature, humidity, enzymes of lysosomal, bacterial and insect sources, predator tissue ingestion et al.) are operating simultaneously and commonly frustrate efforts to attribute an observed alteration to the effect of a specific mechanism.

Our laboratory is involved in both forensic pathology as well as the reconstruction of human disease patterns in ancient populations. Most of our study material is derived principally from the dissection of archaeological human soft tissue remains that have become mummified spontaneously as a result of environmental influences. We soon recognized that neither our clinical hospital autopsy nor our forensic experience had prepared us to understand many of the tissue alterations we encountered. Hence we have initiated an experimental study series designed to control environmental conditions of soft tissue exposure sufficiently to permit isolation of the effects of studied variables during the first month or two following death. We anticipate that our findings will have equal applications to forensic and bioanthropological interests, particularly those relating to the rate of tissue disintegration. In this initial article we describe the model system in which animal bodies or their isolated tissue can be exposed to varying but controlled conditions, and report our initial results with variables of interest to forensic scientists, bioanthropologists and paleopathologists studying preserved soft tissues.

Determination of Water Content of Living and Mummified Soft Tissues

Because the postmortem enzymatic decay process needs an aqueous medium, understanding of that process requires knowledge of tissue water content. Tissue samples of 0.5–2.5 g from heart, lungs, liver, spleen, kidney, and muscle from sacrificed rats were weighed in open small Petri dishes that were then placed in an incubator at 100°C for 48 h, transferred to a desiccating chamber to cool to room temperature and weighed again. Animals in this study were sacrificed between 8:00 and 10:00 a.m.; food and water were continuously available to the animals until removal from the cages for sacrifice. Results of 12 samples (Table 1) indicate a water content varying from 75 to 80% (absolute dry weight of 20 to 25%) in living tissues, with a mean of 77.4%. Similar results were achieved by 42 days of sample exposure in desiccating chambers. Small deviations from the mean value seem to reflect varying fractions of fibrous or other nonepithelial structures in the different tissues.

Since many of the human mummified tissue samples in our files have achieved spontaneous mummification in arid environments, we determined the water content of lung, liver, and muscle tissue from seven such human bodies that had been excavated and dissected 1000 to 3000 years after their burial (9). Table 2 indicates these tissue samples had virtually reached absolute dry weight conditions.

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Received 7 Aug. 1999; and in revised form 1 Dec. 1998, 22 Jan. 1999; accepted 26 Jan. 1999.

TABLE 1—*Water content of living rat tissues.*

Tissue*	% Dry Wt. of Living Tissue	% Water Content of Living Tissue	N†
Lung	20.9 (0.85‡)	79.1 (0.85)	3
Liver	27.3 (0.68)	72.7 (0.68)	3
Muscle	24.9 (1.90)	75.2 (1.90)	2
Heart	20.9	79.1	1
Spleen	21.2	78.8	1
Kidney	20.0	80.0	1
Thymus	20.6	77.2	1

*Tissues from sacrificed Wistar rats.
†N = number of specimens examined.
‡( ) = one standard deviation.

TABLE 2—*Water content of human spontaneously mummified tissue.*

Tissue*	% of Original Water† Content Present in Mummified Tissue	Number Studied
Lung	0.82 (0.37)	7
Liver	0.53 (0.45)	7
Muscle	2.39 (2.40)	7

*Spontaneously mummified human tissues were dissected shortly after excavation. Bodies were adults of Cabuza (c. A.D. 1000) and Chinchorro (c. 1000 B.C.) archaeological culture groups from the coastal area of the Atacama desert near Arica in northern Chile (9).

†The weight of residual water (A) in the mummified human mummy organ tissue sample was determined by the difference in the sample weight at time of excavation and that following complete desiccation of the sample in an oven. The formula used to calculate the percent of the original water content (W) of the sample (at the moment of death) still remaining in the sample at the time of dissection is:

$$W = \frac{A}{B(c/d)}$$ where:

A = (weight of sample at time of dissection) – (weight of sample after complete desiccation).

B = absolute (completely desiccated) dry weight of organ sample.

c = water fraction of living tissue of organ type sampled (from Table 1).

d = absolute dry weight fraction of living tissue of organ type sampled (from Table 1).

Parentheses indicate one standard deviation value.

TABLE 3—*Mice whole body weight loss comparisons of environment (air/sand) and clothing (nude/clothed) status.*

Body Environment*	NC†	Body Weight in Grams: Day 0	Day 69	Loss (Grams)	Loss (%)
Air	N	23.2	9.6	13.6	58.6
Air	C	24.4	7.0	17.4	71.3
Sand	N	23.0	9.0	14.0	60.7
Sand	C	24.9	8.2	16.7	67.1

*Air = mice bodies exposed horizontally on bed of wire mesh 5 cm above sand surface of chamber; sand = mice bodies interred horizontally in sand at depth of 5 cm in same environmental chamber; exposure time 69 days at 22°C at airflow rate of 785 cc/minute:
†N = nude, C = clothed.

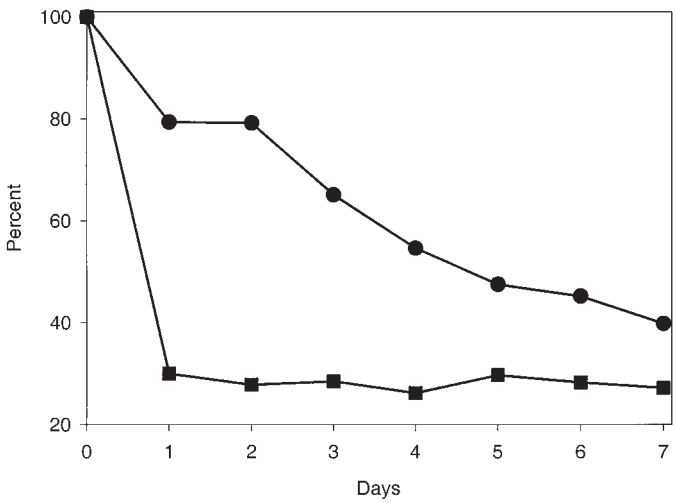

FIG. 1—*Rate of desiccation of bare liver tissue specimen in air and in sand.*

NOTE: Bare liver tissue samples were suspended in air (squares) and in sand (circles) at 22°C at a flow rate of 2021 mL/min; y-axis represents the percent of original weight on indicated days.

Effect of Clothing on Rate of Body Desiccation

To evaluate the effect of clothing on the rate of water loss from an intact animal body two adult mice were sacrificed (CO_2), shaved and exposed in a horizontal position in an environmental chamber of 1334 mL capacity at a flow rate of 785 mL/min of 22°C dry air. One of these mice was exposed in the nude state, the other wrapped in multiple layers of cotton strips two centimeters wide that enveloped the individual extremities and finally the entire body to a thickness of approximately 1 cm. After 69 days the nude mouse had lost 58.6% of its original body weight while the "clothed" mouse lost 71.3%. A repeat study with larger animals (rats) had similar results (51.2 versus 65.3% of whole body weight loss as well as similar proportions for individual organs) (Table 3).

Simultaneously two similarly treated mice were buried horizontally in sterile sand at the bottom of this same chamber at a depth of 5 cm below the chamber's sand surface. Of these two, the nude mouse lost 60.9% of its original body weight while the "clothed" mouse lost 67.1%.

Finally, the effect of simulated windy conditions (by tripling the flow rate to 2021 mL/min) and of specimen size and surface area was evaluated in a separate experiment but under the same conditions as above, though only for a period of seven days. Two pieces of liver tissue measuring $3 \times 2 \times 2$ cm were weighed. One of these was suspended in the air above the sand chamber and the second in sand at a depth of 5 cm. Resulting daily weight losses are displayed in Fig. 1. The greater efficiency of desiccation in rapid air flow under these conditions is clear and discussed further under the Discussion section below.

Effect of Position on Pattern of Organ Conservation

Both modern human forensic bodies as well as ancient interments present themselves to the examiner in a variety of positions. This study was carried out to determine the effect, if any, on the two extremes of position: horizontal and vertical.

Pilot Study

Two rats were sacrificed (CO_2) and exposed in an environmental chamber to a flow of dry air at 22°C at a flow rate of 2021 mL/ min. One of these was mounted in a vertical and the other in a horizon-

tal position. Only daily gross observations were carried out over a period of 55 days after which the viscera were dissected. The following items of interest were noted: The soft tissues of the body became distended by focal areas of bacterial fermentation in both rats. Bloating occurred principally in the dependent areas: over the lower half of the ventral body and pudendum in the vertical rat but most prominently over the entire ventral and flank areas of the horizontal rat. Fermentation sites did not involve the peritoneal or thoracic cavities in the earlier stages of exposure. Between seven to ten days the skin over the distended areas broke down, leaked abundant quantities of black liquid, collapsed and ceased draining by the end of the second week. After three weeks tissue degeneration extended from the abdominal wall into the peritoneal (but not pleural) cavity. At autopsy after 55 days of exposure the diaphragm on both sides were intact in the vertical rat but had become digested bilaterally in the horizontal rat. The thoracic organs were visibly better preserved in the vertical than in the horizontal rat.

Quantitative Determination of Effect of Position on Organ Preservation during Desiccation

Ten rats were sacrificed (CO_2), five of which were mounted on a screen mesh in a horizontal and five in a vertical position in an environmental chamber under conditions the same as those described above for the pilot study. One vertically-suspended and one horizontally-suspended rat were removed from the chamber on days 3, 7, 14, 22, and 33, and dissected. The day 0 wet weight of the organs was calculated from previous studies in which mean organ weights were found to represent the following fractions of total rat body weight: heart 0.35%, lungs 0.62%, liver 5.18%, spleen 0.22%, and kidneys 1.00%.

Table 4 lists the "wet" weights of the body and organs on days 0 and 33. While these results suggest clear evidence for greater weight loss in bodies desiccating in a horizontal position, it is apparent that the final organ weight was a combination of its absolute dry weight plus an unmeasured amount of tissue water; i.e., it was not possible to determine how much of the weight loss was due to simple tissue desiccation and how much to enzyme-digested tissue destruction. Hence the above study was repeated with only two rats (one vertical and one horizontal) and the exposure lengthened to 75 days. After dissection and recording of organ "wet" weights, the hearts and livers were transferred to a desiccation chamber and carried there to dry weights. This latter value was then compared with the dry weight on day 0 (obtained by multiplying the calculated day 0 organ "wet" weight by .209 for heart and .273 for liver, as had been determined in the tissue water content study noted above). Results are listed in Table 5. Comparative results (vertical/horizontal) indicate less structural tissue loss of both heart and liver tissue in the vertical position. This is particularly dramatic for the heart. The spleen and kidneys had completely digested away during the prolonged exposure of the second study.

TABLE 4—*Effect of body position on weight loss (33 days).*

	Percent Weight Loss (Day 33)	
	Vertical	Horizontal
Heart	60.8	86.8
Lungs	62.0	77.4
Liver	55.0	70.6
Spleen	91.9	90.4
Kidney	72.3	86.2
Thoracic organs	61.6	80.8
Abdominal organs	59.0	73.7
Whole Body	45.4	58.2

NOTE: Both whole body and individual organs lost more weight in horizontal than in vertical positions.

TABLE 5—*Effect of position on wet and absolute dry weight loss of air-exposed rats, day 60.*

	Vertical	Horizontal
% total body weight loss	62.5	63.8
% wet weight loss, heart	73.9	78.4
% absolute dry weight loss, heart	10.3	40.4
% wet weight loss, liver	75.9	85.9
% absolute dry weight loss, liver	31.9	51.6

NOTE: Results indicate that horizontal rats lost more structural heart and liver tissue than did vertical rats over a 60 day period. Values obtained from measured values plus heart (0.35) and liver (5.18) organ "wet" weights as percent of total body weight; also heart (20.9) and liver (27.3) organ absolute dry weights expressed as percent of "wet" weights.

Discussion

Water removed by rapid desiccation such as in an oven is principally free or unbound water. Prolongation of the desiccation process may be accompanied by a change in tissue composition producing a change in its "water activity" (i.e., the intensity of the tissue's affinity for bound water) and resulting in tissue loss of additional water (10). About 77% of the fresh weight of rat visceral tissues was composed of water. This value is 11% greater than the total body water content (66%) of a rat measured by desiccation as reported in Spector (p. 340) (11). However, the total body weight includes largely water-free fat, while our measured visceral specimens are essentially fat-free. While we cannot exclude the effect of a possible increase in the water activity of our tissues during the desiccation process, the 11% greater fat-free value we found in our study would appear to be within the range expected in fat-free, cellular visceral tissue. Thus their "absolute dry weight" (ADW) was about 23%. The studied specimens from spontaneously desiccated human mummies buried in the groundwater-free soil of northern Chile's Atacama desert for intervals up to 3000 years approached these ADW values containing only traces of water.

Not all bodies excavated from such sites, however, reveal well-preserved soft tissues. Varying mortuary practices include the presence of clothing, vertical flexed, or extended horizontal position as well as placement of the corpse into an empty space within an often rock-lined cist or simple interment directly into soil. Evaluation of the extent of weight (presumably water) loss changes introduced by these variables revealed that all had significant effects.

A clothed body lost 21.7% more weight than a nude body under otherwise similar circumstances. In an intact animal, moisture from the body's interior (thoracic and abdominal cavities or soft tissues deep to the surface) must traverse all tissues lying between such deeper positions and the skin surface. Thus initially the skin and more superficial tissues lose part of their water content, establishing a gradient of tissue water concentration that will direct the subsequent movement of water from the deeper areas toward the skin. The greater this gradient, the more rapid the water movement. The nature of the skin surface becomes critical, since conditions permitting the accumulation of water there will decrease the gradient

and slow the water flow. Alternatively, the flow will be accelerated by conditions that accelerate the removal of water as it emerges on the skin surface. Absorption of moisture at the skin surface by the clothing and subsequent evaporation into the ambient air flow is the most probable explanation for the increased rate of desiccation in the clothed animals of the study. If so, then it is of further interest that a similar comparison in which the animals were interred in sand instead of exposed to ambient air flow produced similar results. In view of the common assumption that the porosity of desert sand conducts moisture away from a buried body's surface, it must be concluded that at least the type of clothing employed in this study was more efficient for such moisture removal than was the sand alone.

These models require the fluid deep inside the intact body to traverse a series of other tissues of varying density before it can emerge at the skin surface. The importance of maintaining a large gradient of water concentration can be demonstrated in a more simple model. Figure 1 reveals the rate of desiccation occurring in a specimen of liver tissue suspended in a flow of air and another buried in sand, all at 22°C. In this simplified model the effectiveness of maintaining a high gradient by rapid evaporation from the sample surface (by suspending it in a rapid air flow) is made clear. This air-exposed tissue approached absolute dryness in 24 h (92.9% of original water content removed) while its sand-buried equivalent did not reach the same point even after seven days (80.0% of original water content removed). While this over-simplified model's absolute quantitative values can not be compared directly with those of the much more complex situation of the intact body, it does dramatically verify the key feature: the rate of removal of moisture in contact with the specimen surface to maintain a water concentration gradient. This is probably the single most critical determinant of postmortem soft tissue mummification in either mortuary or forensic science contexts. This concept also indicates the skin is the last of a mummified body's tissues to desiccate, accounting for its frequent absence in such bodies except in body areas of little underlying soft tissue intervening between bone and skin. In circumstances of the opposite extreme excessively high temperatures dry and harden the skin so rapidly after death that it becomes impermeable to moisture transfer. In such conditions the fluids trapped within the body provide the milieu for progressive visceral liquefaction (3).

The effect of position is worthy of note. The initial qualitative study indicated that the location of fermentation foci in subcutaneous and muscular tissues were predominantly in the dependent areas and did not include the abdominal cavity until later. Horizontal positions also commonly resulted in destruction of the diaphragms, providing the bacteria and enzyme-laden abdominal fluids access to the thoracic cavity and its organs. Both the entire body and the individual visceral organs lost more weight in horizontally-positioned animals than in those placed in a vertical position. A focus on "absolute dry weight" (the tissue structure), however, demonstrated that vertically-positioned animal carcasses retained more of the original structure and in a more desiccated state than did those of the horizontally-placed animals. Thus the increased weight loss in horizontal rats was enhanced by greater autodigestion and liquefaction of the organs' tissue structure.

In contrast to human skin, that of rats contains no suderiferous pores. Extrapolation to humans of the absolute rate of transdermal water transfer found in our studies would need to accommodate that feature (i.e., transfer rates for human skin would probably be higher). However, since most studies in this report are comparative, it seems improbable that this feature would alter our qualitative conclusions regarding the direction of the tested variable's impact.

Under the conditions of this study, findings that are of forensic or bioanthropological interest include the following: 1. Water content of visceral and other soft tissues was established. 2. A model for studying the taphonomic effects of individual variables was created, tested and found functional. 3. The postmortem body position is a significant contributor to soft tissue taphonomic changes. Compared to a vertical position, the supine position results in greater water loss but also more tissue destruction with liquefaction and consequently poorer organ survival. A possible explanation of these findings may be retention of the diaphragm's integrity in the vertical position. The locations of foci of bacterial fermentation with subcutaneous and muscle liquefaction and gas formation were found to correlate with dependent areas of the positioned body. 4. Clothing substantially enhances body water loss, both in air-exposed and in interred whole animal bodies. 5. Desiccation in sand-interred, intact whole animal bodies was similar to that in air-exposed. 6. Among the features studied in this report, our findings are consistent with the hypothesis that factors contributing to the maintenance of a strong gradient of decreasing tissue water concentration (from the body's interior to the skin surface) by continuous, effective removal of water from the skin surface is the most important determinant in the enhancement of spontaneous soft tissue mummification.

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