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PAPER

ANTHROPOLOGY

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Testing the Use of Pigs as Human Proxies in Decomposition Studies*

ABSTRACT: Pigs are a common human analogue in taphonomic study, yet data comparing the trajectory of decomposition between the two groups are lacking. This study compared decomposition rate and gross tissue change in 17 pigs and 22 human remains placed in the Forensic Investigation Research Station in western Colorado between 2012 and 2015. Accumulated degree days (ADD) were used to assess the number of thermal units required to reach a given total body score (TBS) (1) which was used as the measure of decomposition. A comparison of slopes in linear mixed effects model indicated that decomposition rates significantly differed between human donors and pig remains $\chi^2_{(1)} = 5.662$, $p = 0.017$. Neither the pig nor the human trajectory compared well to the TBS model. Thus, (i) pigs are not an adequate proxy for human decomposition studies, and (ii) in the semiarid environment of western Colorado, there is a need to develop a regional decomposition model.

KEYWORDS: forensic science, taphonomy, decomposition, animal models, human proxies

Interest in forensic taphonomy has increased steadily over the last forty years. The interaction between human remains and the postdeposition environment may alter or obscure features necessary for effective analysis of postmortem interval (PMI), trauma, pathology, and the biological profile, affecting the analytical conclusions and the quality of information gained. Facilities with the capacity to study human remains increased from one facility in 1981 to seven facilities worldwide in 2017. Due to anatomical and physiological similarities to humans, pigs (*Sus scrofa*) are a standard proxy for those endeavoring to study taphonomy outside of these facilities. Similarities between pigs and humans frequently cited as the basis for equivalence include: internal anatomy, diet, endogenous microbiome, fat-to-muscle ratio, tissue density, structure, density, and distribution of body hair, monogastric digestive system, and omnivorous diet (2). Due largely to cost and ability to amass large sample sizes, smaller animals, such as rabbits, are also frequently used (3). However, aside from identifying general overlap in anatomy and physiology, the efficacy of the use of pigs as human analogues remains largely untested.

The origin of pigs as human proxy can primarily be traced to entomological studies where the use of animal proxies for the comparison of insect succession throughout decomposition is well tested. Rodriguez and Bass (4) compared available insect succession data collected from canine cadavers (5) to that observed on human cadavers and found significant overlap. It appears that the majority of insect species (99.67%) show no preference between pigs or humans (6,7). However, caution is warranted when using entomological studies to validate the use of pig carrion in decomposition studies. While the entomologist and anthropologist both seek to understand PMI, it is incorrect to assume that the disciplines are studying the same underlying physiological phenomenon. The entomologist is concerned with insect development, progression, and succession guided by the sum of the decomposition event (e.g., the release of chemical constituents that guide chemotaxis, moisture content and related patterns of oviposition, and visual and mechanosensory cues that govern insect movement). Conversely, the anthropologist is concerned with the sum of stages of gross morphological change, which may correlate with various stages of decomposition (e.g., rate, chronology, and presentation of tissue dissolution under various circumstances). Therefore, the biological features that make pigs a viable research tool in entomology cannot be assumed to translate to taphonomic investigation.

Early observational studies on decomposition used a variety of species, including cows, seabirds, and sea mammals (8). Payne (9) studied dogs, cats, squirrels, rabbits, chickens, birds, and pigs, concluding that relatively large animals of uniform size were best for his studies (10). The results of these studies were rarely compatible and consistently resulted in opposing criteria and sequences used to describe the trajectory of decomposition. Micozzi (10) suggested that the incompatibility was due to the wide variety of animals used and the conditions under which the studies were carried out. Stokes et al. (11) evaluated potential proxies for use in soil decomposition studies. A suite of physiochemical soil characteristics was compared among and between tissue types using small samples of skeletal muscle tissue from humans, pork, beef, and lamb. Different species exhibited similar patterns of change, but none of the proxies were an ideal predictor of human skeletal muscle tissue for soil decomposition studies. Wang et al. (12) conducted a comparative analysis of insect

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succession on human remains, pig carrion comprising two size categories, and rabbit carrion. They conclude that body size and decomposition rate share an inverse relationship, and insect species diversity and complexity are more complicated on larger remains. Dipteran development rate was consistent across all size and species categories, although a subset of forensically significant Calliphoridae species were unable to complete first-generation development on rabbit carrion. Dautartis et al. (13) observed a similar correlation between body size and trajectory of decomposition. Five each of pigs, humans, and rabbits were used to study gross morphological changes and insect activity across a 5-month interval. The conclusion was that the pattern of decomposition differed between the three groups, and they were not likely to be interchangeable for decomposition research. However, the continuing challenge in using human remains for research study is sample size. While these studies present intriguing patterns, there is still a need for research conducted within human samples, using larger sample sizes, and in differing environments. Additionally, longitudinal studies are warranted in dry areas where desiccation occurs and bodies take much longer than 5 months to skeletonize. The goal of this research was to further test whether, and to what degree, pigs provide a useful proxy for human decomposition.

Materials and Methods

Observations were made at the Forensic Investigation Research Station (FIRS), Colorado Mesa University, located on the western slope of Colorado. The FIRS performs ongoing research using human donors to study the trajectory of taphonomic change. The FIRS is situated at an elevation of 1457 m AMSL. The environment is classified as mid-latitude steppe/semiarid cool in the Köppen–Geiger climate classification system and nets approximately 20–25 cm of precipitation annually. Five years of on-site data collection yields an average high of 33.33°C with daily highs exceeding 41.1°C and an average low of −3.88°C, with daily lows exceeding −25.27°C. The western slope is a relatively sunny area, receiving approximately 70% of available sunshine. The FIRS is in the area of Utaline-Neiman-Lazear association of soils. These are well-drained loam soils formed in materials weathered from basalt and sandstone (14). The county soil map shows that the facility is in an area of Billings silty clay loam (14). The natural vegetation is primarily four-winged saltbush (*Atriplex canescens*), rabbitbrush (*Ericameria nauseosa*), galleta (*Pleuraphis jamesii*), and Indian rice grass (*Achnatherum hymenoides*).

This study compared the pattern of decomposition in 17 pig carrion and 22 human remains. Animal and human remains were placed in the facility between September 2012 and December 2015. The pigs were systematically placed so that the first 12 were placed one pig a month from September 2012 to August 2013. The remaining five pigs were placed on the same day as our second through sixth human bodies. The human remains were placed when they were received as part of FIRS donation program, resulting in placement during all four seasons. The numbers of human remains placed each month were: January=3; February=3; March=2; May=2; July=2; August=2; September=2; October=2; November=2; December=2. Although a convenience sample, the sample represents all seasons.

Hourly environmental data were collected from an Onset HOBO remote-logging weather station placed within the research facility (Onset; Bourne, MA). Accumulated degree days (ADD) were used as an indice of PMI and calculated using a base temperature of 0°C following Vass et al. (15) and Megyesi et al. (1). Decomposition was scored at regular intervals following Megyesi et al.'s (1) total body score (TBS) system. The ADD was calculated for each date that a TBS score was collected, resulting in 2627 ADD-TBS pairs for the human subjects and 1100 ADD-TBS pairs for the pig subjects.

Euthanized pigs were delivered to FIRS for placement within less than four hours of death; the supplier euthanized the pigs with a gunshot wound to the head. Body sizes ranged between 25 and 64 kg at the time of death, with a median of 35 kg. Both male and female pigs were included in this study. A single supplier was used, and the pigs were fed similar diets, lived in a similar environment, and were in generally good health at the time of euthanization. None of the pigs were refrigerated. The initial TBS for all pig subjects was three.

The human sample was amassed under the auspices of the FIRS human donation program. Cause of death within the human sample included pathogeneses summarily categorized as "natural" (e.g., heart attack), diagnosed chronopathology (e.g., cancer, lupus), and trauma (blunt force following a fall). Because the thoracoabdominal incision attendant to autopsy approximates dramatic penetrating trauma, and the trajectory of decomposition in autopsied versus nonautopsied remains has not yet been assessed at FIRS, autopsied remains were excluded from this study. Intragroup rate and pattern of decomposition were also considered. Placement of human remains varied from the day of death with no refrigeration prior to placement to 53 days postmortem with refrigeration in the interim. If date of death was unknown, donors presenting a TBS >7 (fresh to early decomposition on the Megyesi scale) were excluded from this study. Pig and human remains were photographed and scored at regular intervals consistent with FIRS TBS protocols (16). Under the data collection protocol, each body was scored daily until a TBS of 24 was reached. To allow for visible changes to accrue (i.e., score-able changes), remains with TBS ≥24 were scored weekly for humans and monthly for pigs. This protocol was adhered to as weather and staffing allowed. All personnel performing data collection were trained and tested in FIRS data collection protocol prior to entering the facility, and all training scores were assessed blindly by two full-time staff members to ensure that data quality and accuracy were maintained, and inter-observer error was minimal. Individual remains were not caged as scavenging was infrequent and limited to small areas of soft tissue loss.

The decomposition rates of the pigs and humans were compared using two methods. For the first comparison, the mean, median, and coefficient of variation of ADD were calculated for each TBS point and visually assessed (Table 1, Figs 1–3). Additionally, a linear mixed model (LMM) using maximum likelihood estimates was used to determine if decomposition rate varied between the human and pig groups. The use of LMM was advantageous to account for unbalanced design and repeated measurements on single remains, which has the potential to highlight both inter- and intra-sample variation. In the LMM, the dependent variable was TBS (TBS^2 transformation). The independent fixed variables were ADD (Log_{10} (ADD+1) transformation), an indicator variable for human (indicator = 1) or pig (indicator = 0) remains, and an interaction term (Log_{10}(ADD+1) x Indicator). The random effects for both intercepts and slopes were ADD and the individual remains. The interaction term was used to test for a statistical difference between slopes. The *p*-value for the interaction was determined by a likelihood test comparing the models with and without the interaction term.

TABLE 1—*The mean and coefficient of variation of ADD at each TBS point for humans and pigs. "N/A" indicates that no specimens were scored at that TBS interval.*

TBS	Pig ADD	Pig Coefficient of Variation	Human ADD	Human Coefficient of Variation
3	9.42	109.74	11.43	100.28
4	N/A	N/A	8.74	122.98
5	15.32	92.30	25.57	75.83
6	27.42	70.57	23.26	86.77
7	31.19	50.08	15.18	84.57
8	47.38	53.52	39.77	80.75
9	46.41	67.36	35.47	77.56
10	63.71	56.01	55.30	77.40
11	74.97	51.40	94.13	68.61
12	108.36	68.76	91.35	70.91
13	118.22	55.18	127.69	58.32
14	145.80	57.24	175.93	53.59
15	201.89	59.79	221.76	56.94
16	206.93	46.56	236.01	58.76
17	513.89	177.73	241.09	59.26
18	466.68	172.0	379.91	131.51
19	1194.82	126.38	519.92	115.21
20	993.65	131.86	594.17	83.86
21	2356.82	96.35	1296.11	92.20
22	1482.82	156.92	2081.71	68.79
23	1979.29	131.01	3175.87	63.06
24	3881.73	76.94	5218.36	59.21
25	5751.12	61.77	5614.57	58.01
26	4225.05	81.23	6042.73	60.24
27	4963.49	86.71	6711.08	59.99
28	5159.25	69.79	4711.35	46.19
29	6991.28	58.76	4501.79	27.94
30	8002.54	59.00	4543.18	18.63
31	8738.27	59.15	5270.26	5.15
32	10515.97	43.99	N/A	N/A
33	9263.33	47.18	N/A	N/A
34	9711.82	36.12	N/A	N/A
35	13246.84	27.61	N/A	N/A
36	12168.09	0.16	N/A	N/A

ADD, accumulated degree days.

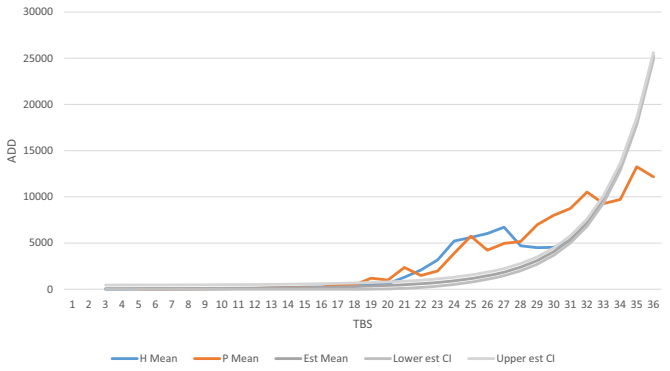

FIG. 1—*Pig and human ADD mean for each TBS score compared to the estimated ADD from the TBS model (1). ADD, accumulated degree days; TBS, total body score. [Color figure can be viewed at wileyonlinelibrary.com]*

The analysis was conducted in Program R (version 3.4.0) using the lme4 package (17,18).

Results

Visual assessment of the TBS means and coefficients of variation in pigs and humans against the model (Fig. 1) made it apparent that the data from FIRS were no longer represented by the model after a TBS of 18. The human and pig mean ADD and median ADD for each TBS point also differed with humans requiring a greater ADD to reach a specific TBS until about a TBS of about 27, and then, the pigs required a greater ADD to reach a specific TBS (Fig. 2, Table 1). A visual assessment of the coefficient of variation suggests that initially the human sample varies more than the pig in the number of ADD required to reach a specific TBS, after a TBS of approximately 15, when the pig sample generally has a greater variation (Fig. 3, Table 1).

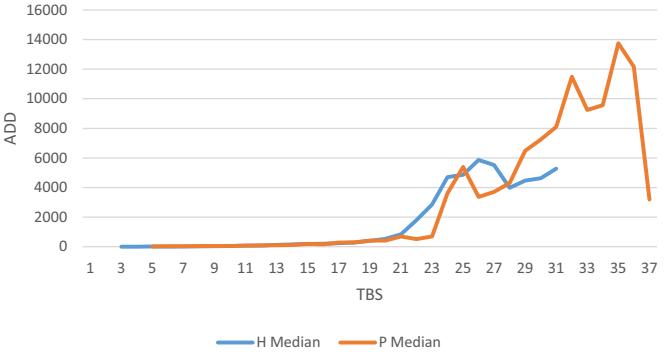

FIG. 2—*Pig and human median ADD for each TBS score. ADD, accumulated degree days; TBS, total body score. [Color figure can be viewed at wileyonlinelibrary.com]*

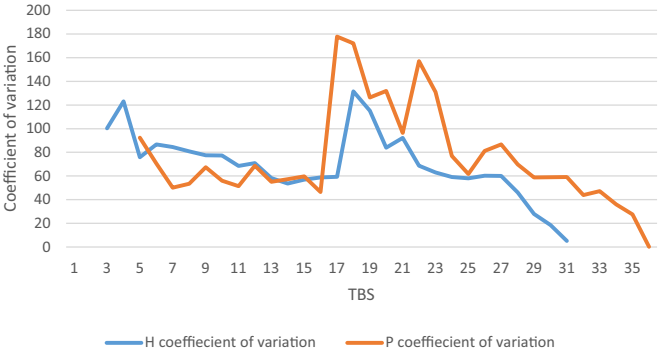

FIG. 3—*Pig and human ADD coefficient of variation for each TBS score. ADD, accumulated degree days; TBS, total body score. [Color figure can be viewed at wileyonlinelibrary.com]*

The LMM indicated that decomposition rates significantly differed between human donors and pig remains based on a comparison of slopes of the interaction coefficient $\chi^2_{(1)} = 5.662$, $p = 0.017$ (Fig. 4). The LMM model used 2626 observations for the 22 humans and 1101 observations for the 17 pigs. The LMM for the fixed effects with standard errors in parentheses was

$$\begin{aligned}\mathrm{TBS}^2 = {} & 351.59(49.55) + 304.66(21.57) \cdot \mathrm{Log}_{10}(\mathrm{ADD}{+}1) \\ & + 171.25(66.05) \cdot \mathrm{Indicator} - 85.88(28.80) \\ & \cdot \mathrm{Log}_{10}(\mathrm{ADD}{+}1) \cdot \mathrm{Indicator}\end{aligned}$$

marginal $R^2 = 0.67$ (fixed effects) and conditional $R^2 = 0.90$ (both fixed and random effects).

Discussion

In approximately 60% of the pig specimens, the intestines ruptured through the abdomen during the bloat phase (Fig. 5);

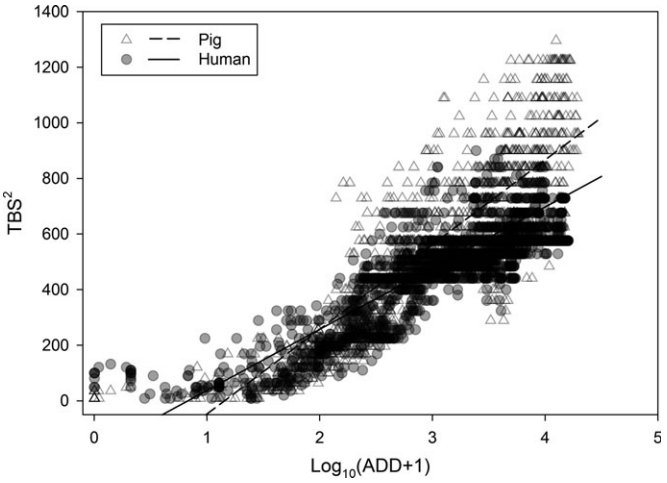

FIG. 4—*Comparison of decomposition rates between humans and pigs. Regression lines were fit using the fixed effects from the LMMs.*

FIG. 5—*Intestinal rupture in pig. [Color figure can be viewed at wileyonlinelibrary.com]*

abdominal rupture did not occur in any of the human specimens. While humans and pigs present anatomical, physiological, and genetic overlap, fundamental differences between the two species exist. While both humans and pigs are monogastric, key differences in the structure and function of the gastrointestinal tract (GIT) may affect the rate, timing, and trajectory of decomposition. On average, the human intestine (large and small) is 7.5 m in length, as opposed to 23 m in the pig. The pig small intestine further presents differences in segment length and branching of mesenteric vessels and attendant anastomoses (which facilitate cellular communication with surrounding tissues). In the human GIT, ileal Peyer's patches (PP) are an aggregate of lymphoid modules that facilitate an immune response within the mucosa by monitoring intestinal bacteria and inhibiting growth and spread of pathogenic strains. Conversely, the ileal PP in pigs is continuous, which not only constitutes a structural difference, but also implies major differences in typical bacterial load, potential exposure to pathogenic bacteria, and enteric colonization. A pyloric diverticulum—a site for microbial metabolism of ingesta—is present in pigs but absent in typical human anatomy (19). Finally, the cecum, ascending and transverse colon, and the proximal portion of the descending colon are oriented in a series of centrifugal and centripetal coils to accommodate length and orientation associated with quadrupedalism, which differ from the folded and "stacked" structure of the human intestines.

The sum of these many subtle differences may contribute dramatically to gross differences in decomposition, such as the abdominal rupture observed in this sample. In more basic terms, while this phenomenon is likely multifactorial, a significantly longer GIT summarily constitutes larger cubic volume of lumina within which decompositional gasses have the potential to aggregate, resulting in greater potential for eruptive response.

Overall similarity in TBS scores in pigs and humans in early decomposition may be an artifact of the overfitting of data to the scoring system; data collection necessitates that a score be applied so the "best choice" category regardless of overall technical accuracy. Only six categories in the head/neck and five in the trunk and limbs describe early decomposition. Limited scoring options may reduce or eliminate the resolution necessary to highlight divergence in gross change. Keogh et al. (20) used the Megyesi et al. (1) TBS to define the typical trajectory of human decomposition and compared this to observations made within a sample of 20 pig carrion. The authors concluded that the gross changes observed between the two groups were sufficiently different to warrant a separate scoring system for pig carrion. However, TBS comparison was limited to pigs and did not include humans in the sample environment, so the differences observed may be a combination of species and environment.

Total body score values for both species plateaued between 21 and 24 for an extended period of ADD. The humans plateaued in moist decomposition for a longer period of ADD. The pigs were a clinically healthy weight, while over half the human sample was overweight or obese. Body fat does impact decomposition, hindering dissipation of heat and providing liquid for bacterial growth and insect oviposition. With regard to the human sample, the presence of diabetes mellitus accelerates decomposition (21); as a strong correlation with obesity and with an estimated seven million undiagnosed cases per annum (http://www.diabetes.org/diabetes-basics/statistics/ (accessed September 30, 2017)), the unreported presence of the disease should be considered in research and in medicolegal investigation. The cause of death in the human sample included chronopathology (cancer and lupus) and traumatic injury. Antemortem infection, wounds, and chronopathology may accelerate putrefaction by inducing hyperthermia secondary to pathogenesis (including tertiary infection such as sepsis), or as the result of medication (21). Wounds also provide additional places for insect oviposition. The plateau observed among both human and pig samples is likely due to environmental factors at the FIRS. In FIRS' arid climate, remains tend to desiccate, rather than skeletonize. Dermal tissue and underlying viscera are hygroscopic in nature and therefore may be expected to react similarly to environmental conditions. Visceral retention following desiccation was noted in several human cadavers at FIRS. These phenomena have not been observed in pigs and may be the result of visceral eruption or another generalized pattern of decomposition.

Compared to the human sample, the pigs were a more superficially homogenous sample at the start of the project; control over variables such as weight, cause of death, reported pathology, and immediacy of placement within the facility reduced intragroup variation. Such a homogenous sample may allow researchers to control variables and identify trends in decomposition, but identified trends need to be validated within human samples before their efficacy can be accurately reported and extreme caution is warranted when attempting to validate a study conducted on humans within a pig sample. The use of pigs in research has the potential to complicate judicial proceedings where researchers are asked to testify on the condition of human remains based on

their knowledge of pig decomposition, or where established methods are called into question under the Daubert Standard following publication of research conducted within pig samples.

The human remains were a more variable group, but a more realistic sample of forensic cases. Because forensic cases often involve unknown circumstance, the variation among the human sample was in fact reflective of the variation that any useful decomposition measure must encompass to provide an estimate of the PMI. In this sense, discerning patterns among homogenous proxy populations may provide a false sense that the same conclusions are applicable to a much less homogenous population. Forensic disciplines are still attempting to understand how the interaction of complex variables affects human decomposition—a conservative approach is critical when drawing direct correlations between humans and proxies.

The initial samples of pigs appear more homogenous and humans more varied, but the pigs tend to have a greater coefficient of variation (Table 1, Figs 3, 4), particularly after a TBS of 25. This may partly be due to the fact that the TBS model was created for humans and does not work as well with pigs (20). Furthermore, while a pig sample may present as "healthy" and homogeneous in gross examination, animals reared for consumption are not subject to regular health assessment and should not be assumed to be homogeneously "healthy" at the time of death. Pathological stimulants identified in research include immunocompromise following psychosocial stress, including decreased antibody response to antigenic challenge relative to stock density (22) and increases in viral–bacterial load, such as *Salmonella* following exacerbated norepinephrine excretion which Bearson and Bearson (23) demonstrated may directly impact growth and virulence of *Salmonella*, including the upregulation of genes resulting in enhanced motility. Disease associated with commercial rearing, captivity, and confinement is sufficient to result in research dedicated to locating ever emerging biomarkers for pig disease in an effort to curb economic loss (24). The exercise here was not to provide a comprehensive cross section of pig pathology, but to demonstrate that gross examination and meeting variables dictated by sample requirements are not a sufficient means for determining "health" in a biologically diverse species which has demonstrated strong immunosensitivity to the antemortem environment. The sum of the anatomical, physiological, and pathological differences presented may be manifest in unexpected ways at any time throughout the decomposition event. As researchers struggle to understand the diverse complexity of human decomposition, it is specious to suggest that the intersection of captivity, dynamic zoonotic disease processes, death, and decomposition may be overlooked and a distinct and diverse species defined as homogenous to meet analytical needs.

Finally, the TBS model used by many decomposition studies does not work well with pigs and, for the data presented, fit neither pig nor human data in late-stage decomposition. This was likely due to the desiccation seen in FIRS' semi-arid environment and highlights the need for region-specific validation of models used to estimate postmortem interval.

Conclusion

When compared using the TBS model, the trajectory of decomposition observed between human and pig samples diverged in rate and gross presentation. Additionally, the TBS scoring model did not perform well in the arid environment at the FIRS. Regional decomposition patterns may require regional scoring models. Nonhuman proxies do provide a superficially homogenous sample allowing isolation of individual variables and have the potential to indicate trends in taphonomy. Human samples tend to be more variable, particularly in body composition and cause of death, both of which affect the pattern of decomposition. Pigs may be useful in studying general trends. However, they are not a substitute for human subjects, and caution is warranted when attempting to apply data derived from pigs to human subjects, especially in medicolegal investigation. Above all, reliance on a relatively homogenous proxy sample may make researchers overconfident in their ability to predict the timing and patterns of decomposition.

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