The influence of rehydration on decomposition in the Highveld region of South Africa—Using a pig model

Claire Lynne du Toit [1,*], Jolandie Myburgh [2], Desiré Brits [1]

[1] Human Variation and Identification Research Unit, School of Anatomical Sciences, Faculty of Health Sciences, University of the Witwatersrand, Johannesburg, South Africa.

[2] Department of Anatomy, School of Medicine, Faculty of Health Sciences, University of Pretoria, Pretoria, South Africa.

* **Correspondence:** Claire Lynne du Toit, Human Variation and Identification Research Unit, School of Anatomical Sciences, Faculty of Health Sciences, University of the Witwatersrand, Johannesburg, South Africa. Email: clairedtt5@gmail.com

ABSTRACT

Researchers have observed that rainfall may re-initiate decomposition in desiccated tissue; however, no conclusive research-based evidence exists on the specific effects of rehydration on decomposition. Therefore, this study aimed to assess the effects of artificial rehydration on the progression of decomposition following the advanced stage of decomposition. Twelve adult pig cadavers (8 experimental; 4 controls) were placed in the central Highveld of South Africa during cooler (April–July 2021) and warmer (August–November 2021) months. Decomposition was scored approximately biweekly to obtain the total body score, and accumulated degree days (ADD) were calculated for each pig. All pig cadavers were covered by chicken wire cages with transparent tarps to control for natural rehydration and scavenging. Once the experimental pig cadavers reached a three-visit stasis in the advanced phase of decomposition, they were artificially rehydrated, and changes in the progression of decomposition between the control and experimental groups were plotted (ADD against TBS) for observation. The rehydrated experimental pig cadavers showed re-initiation of decay and insect re-colonization, while the control cadavers mainly remained in a state of stasis with insect activity ceased altogether. Greater cadaver decomposition islands and a color change post-rehydration were also noted in some experimental cadavers. This supports the need for future research on the impact of rehydration, including associated soil moisture on decomposition rates, progression, and invertebrate colonization, which will enhance our understanding of the effects these environmental factors have on the accuracy of post-mortem interval estimation.

Keywords: decomposition; forensic anthropology; pig model; post-mortem interval; rehydration; taphonomy.

HIGHLIGHTS

- Rehydration in the advanced stage of decomposition reinitiates the decay process.
- Insect prevalence increased, and recolonization was triggered following rehydration.
- Cadaver decomposition islands increased post-rehydration.

1 INTRODUCTION

A major focus in forensic taphonomy is the process of decomposition and the biotic and abiotic elements that play an active role in its progression and cessation. Soft tissue decomposition is described as a systematic breakdown of a whole body until the point of skeletonization [1]. The process of decomposition consists of various stages, and although these stages have been delineated, there is significant overlap [2]. Knowledge of the decomposition process, and the various factors that influence the progression thereof, is vital when assessing the post-mortem interval (PMI).

PMI estimation is a critical tool utilized in death investigations that can provide context regarding recovered remains. It can allude to the post-mortem environmental conditions a body may have been exposed to, and PMI could help to narrow down the list of unidentified individuals logged in missing person databases based on the time these remains were found, estimated times of death calculated and the date individuals that have been reported missing [3]. In South Africa, the number of unidentified remains is high, and of the 18,324 bodies stored at 11 medico-legal laboratories across Gauteng in 2020, approximately 1173 (6.4%) remained unidentified [4]. However, the high number of cases involving unidentified decomposed individuals is also a global problem [5], highlighting the need for better means of identification, including more accurate methods to estimate the PMI.

Current methods used to estimate the PMI are linked to the stages of decomposition and require a thorough knowledge of the factors that can influence the progression and rate of decay of human remains [3, 6, 7]. Temperature, humidity, rainfall, soil, invertebrate colonization, vertebrate scavenging, trauma, clothing, body size and weight are some of the main factors influencing the decomposition process [8]. Rainfall/moisture has been attributed to influencing decomposition [9]. However, there is limited research looking at the direct effects of rainfall/moisture on the progression of decomposition. The effects of rain (rehydration) have mainly been observed as a part of seasonal variation and it has been suggested that rehydration will result in the re-initiation of active decomposition after desiccation [9]. This is forensically significant, as remains are likely to dry out and stall in the advanced stage of decomposition during dry seasons and resume decomposition after rehydration (rainfall) in the wet season. If these remains are, therefore, only discovered in the wet season, a shorter PMI estimation than the actual PMI might be calculated, resulting in inaccurate estimations. This is specifically the case in the Highveld region of northern South Africa, with its colder and drier winter months (June–August) and warmer, wetter summer (December–February). This study, therefore, aimed to assess the influence of rehydration on remains that have entered a state of stasis in the advanced stage of decomposition.

2 MATERIALS AND METHODS

This study was conducted at the Forensic Anthropology Body Farm (FABF) on the Miertjie Le Roux experimental farm, University of Pretoria. The experimental farm consists of 570 hectares (5.7 km^2), with a ±0.5 hectare (5000 m^2) fenced-off section designated to the FABF in the Cullinan District, Gauteng Province, in the central Highveld plateau of South Africa [9]. The Köppen-Geiger climate classification of this region is Cwb; (C) temperate, (w) dry winter, (b) warm summer [10, 11]. Rainfall mainly occurs during the summer months in this area, with limited winter showers [12].

A total of 12 domestic pig (*Sus scrofa domesticus*) cadavers (deceased remains) weighing approximately 40–100 kg were used in this study. Ethical clearance for the use of animal material in this study was obtained from the animal research ethics committee, the University of the Witwatersrand (AESC20-05-003; 2020/05/05/O) and the University of Pretoria research ethics committee (REC104-20). The specific weight of the pig cadavers was chosen to represent the average weight of an adult human, as body size and weight have been shown to affect the rate of decomposition [13]. Pigs were collected within 1 day of death in the fresh stage of decomposition. Only pig cadavers with no external trauma or wounds were accepted for this research, as an increase in invertebrate colonization has been observed in cases with trauma or external wounds [14]. Pigs that died of natural causes, including diseases such as *Lawsonia intracellularis* and *Haemophilus parasuis*, which are common amongst pig farms [15], were used. A pig model was used due to the similarities between human and pig decomposition and the similarities regarding the skin, weight, diet, and intestinal flora [16]. Pigs are still not the perfect proxy for human decomposition studies due to the difference in limb proportions and body composition [17]. However, due to South Africa's lack of human decomposition facilities, the use of pig models is necessary. They provide valuable information on the factors that influence decomposition, which can then be used to support PMI estimations in human cases.

Four placements occurred (Table 1), with three pigs placed each time (April, July–August, and early September 2021). The placement of the pigs during this time frame was mainly due to limitations and travel restrictions caused by the COVID-19 pandemic, but also fell outside of months with high summer rainfall while allowing for the direct comparison to the study by Myburgh et al. [9], which did not exclude rainfall as a variable. In three instances, only two fresh pig cadavers were available for placement, with the third pig cadaver placed within the same week. To accommodate for the different placement dates and potential temperature differences, the ADD for each pig was scored individually. Of the three pig cadavers placed for each trial, one pig cadaver served as the control pig (CP), and the other two pig cadavers served as the experimental pigs (EP), which were rehydrated.

TABLE 1. Comparison of minimum, maximum, and average daily temperatures, as well as total rainfall for each trail, over each experimental period respectively, including the month of placement.

Temperature (°C)	Trial 1 (April)	Trial 2 (July)	Trial 3 (August)	Trial 4 (September)
Minimum	2	2	4	10
Maximum	27	36	36	36
Daily average	15.4	19.9	21.1	21.9
Total rainfall (mm)	22.2	23.4	23.4	22.3

Cages were used in this study to cover the pig cadavers during the progression of decomposition (Figure 1) to prevent scavenger activity of larger vertebrate scavengers known in the area, such as stray dogs (*Canis lupus familiaris*) and black-backed jackals (*Canis mesomelas*) [15]. The top of each cage was covered with a transparent plastic tarp to prevent direct rainfall on the cadavers and surrounding soil while allowing natural sunlight. The base of the cage was left open for the cadaver to have contact with the ground, while the sides of the cages were covered with chicken mesh to allow air circulation and insect access so as not to hinder the normal decomposition process. Soil moisture, however not fully eliminated, was reduced.

FIGURE 1. Custom metal cages, 0.9 m × 0.9 m × 2.0 m, surrounded with chicken mesh to prevent the entry of large scavengers and covered with a transparent plastic tarp, fastened to the cage, to prevent natural rehydration.

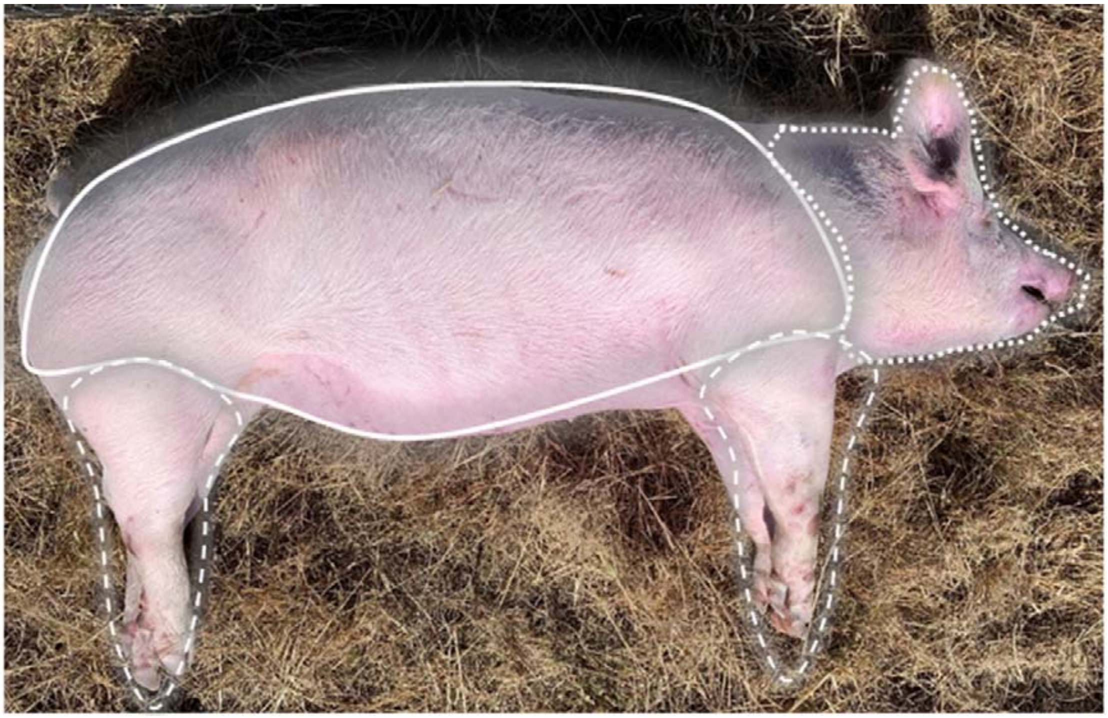

FIGURE 2. Regions of the body, including the head and neck (dotted line), trunk (solid line) and limbs (dashed line) assessed to establish the TBS following guidelines as set out by Keough et al. [18].

Decomposition scores were collected two to three times a week until the cadaver reached the advanced stage of decomposition and thereafter once a week until remains had desiccated and decomposition completely stalled. The progression of decomposition was assessed by defining decomposition as a change in the total body score (TBS)—a method developed by Megyesi et al. [3] and adapted for a pig model by Keough et al. [18]. The TBS was collected during each visit using the adapted description and scores proposed by Keough et al. [18]. Each section of the body was assessed according to the delineation of body regions described in Myburgh et al. [9] and Keough et al. [18] (Figure 2), including the head and neck—from snout to the start of the shoulder girdle, trunk—from the shoulder girdle to the pelvic girdle, including the thorax and abdomen; and limbs—where the limbs meet the trunk, excluding the shoulder and pelvic girdles. The sum of the scores from each region gave the TBS value for that stage of decomposition [3].

FIGURE 3. Method of watering a cadaver once the TBS stalled in the advanced stage of decomposition.

Decomposition was scored until the cadaver was in the advanced stage of decomposition, and there was no observable change in the TBS value over three consecutive site visits (a TBS value between 18 and 20) [18]. Once the experimental cadavers had stalled in advanced decomposition, they were rehydrated with an amount of water that is equivalent to the average rainfall (10 mm = 10 L per square meter) during a typical rainstorm in the Highveld region [19]. An area of 1 m^2 was marked out around the cadaver, and the water was poured with a sweeping motion over the area where the cadaver lay using a watering can (Figure 3). Spring water was used due to its availability and to resemble natural rain as it is typically devoid of excessive chemicals used in the general water supply (e.g., high chlorine levels). Thereafter, the progression of decomposition was followed by scoring the TBS until the control and experimental cadavers reached skeletonization and/or a state of complete stasis in the TBS scores for three consecutive visits.

Due to the small sample size, statistical analyses were not conducted. However, an analysis and description of the trends were conducted to observe the changes in the progression of decay by plotting the ADD value against the corresponding TBS.

During decomposition, basic insect activity was also noted in the three regions of the pig cadavers. A retrospective analysis, using photographs and site notes from each visit, was done by assigning a score for each of the three body regions. A score of 0 was recorded for the absence and 1 for the presence of insect activity. The scores were totalled for every visit, with 1 representing insects' presence in only one region of the body, 2 representing insect activity in any 2 regions, and 3 representing insect activity in all three body regions. The regional scores represented insect activity over the experimental phase and were plotted against ADD as a timeline.

3 RESULTS

The placements of the study pigs were spread over the trial period (Tables 1 and 2) and resulted in two distinctive placements as done by Myburgh et al. [9], one cooler (Trial 1) and three warmer (Trial 2, 3, and 4) placements based on the maximum temperatures recorded and the natural rainfall. The natural rainfall that occurred during each trail was very similar, ranging between 22.2 and 23.4 mm with a difference of 1.2 mm. The temperatures between the two sets of placements differed by a minimum average daily temperature of ±4°C and a maximum average daily temperature of ±9°C (Table 2). Over the study period, the minimum temperature was ±2°C, the maximum temperature was ±36°C, and the mean average daily temperature was ±18°C, with a recorded total of 45.6 mm of rain [20].

TABLE 2. Comparison of the minimum, maximum and average daily temperature for the cooler and warmer trials.

Temperature (°C)	Cooler trial (Trail 1)	Warmer trials (Trails 2–4)
Minimum	2	2
Maximum	27	36
Daily average	15.4	19.9

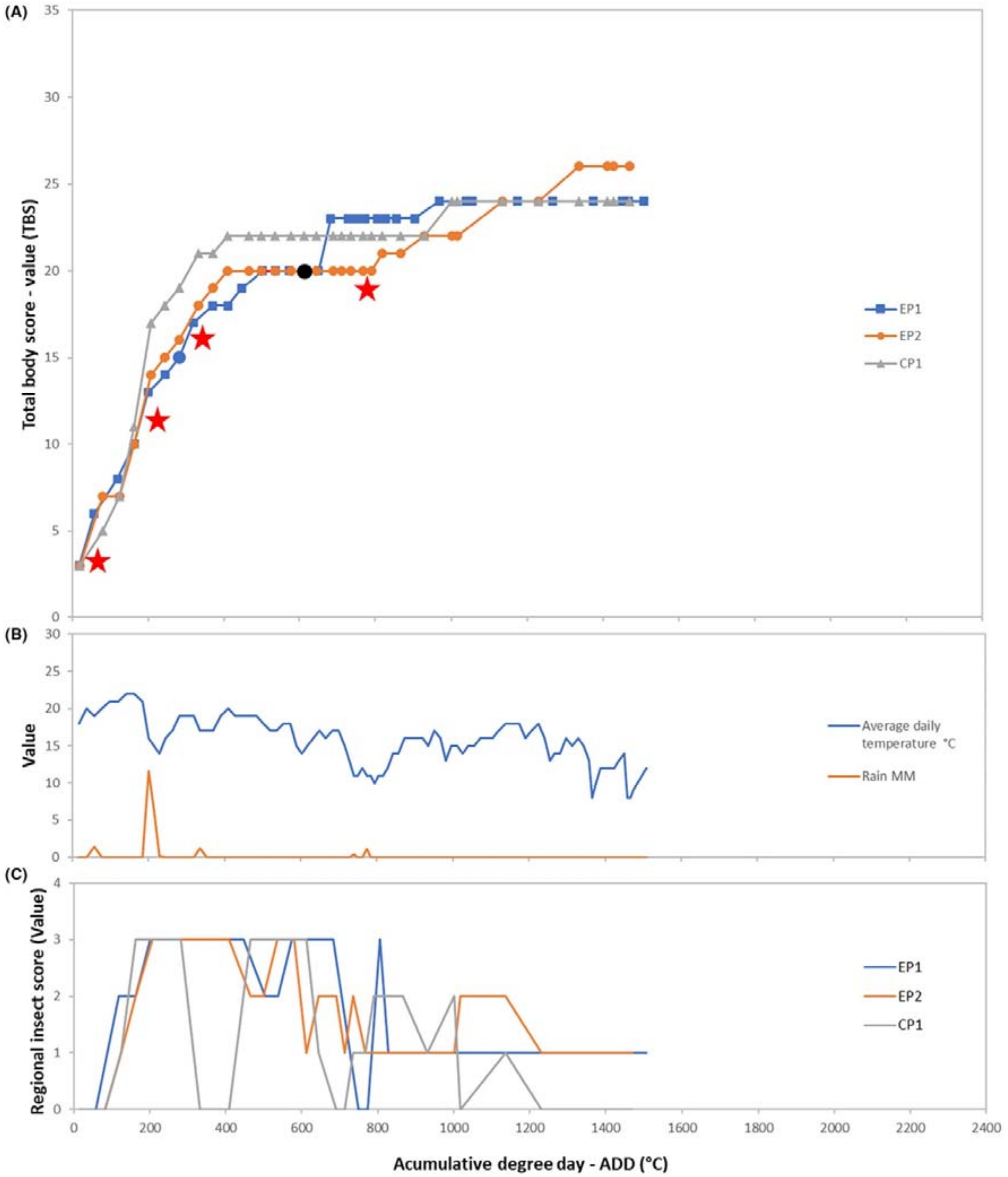

FIGURE 4. (A) Scatter plot representing accumulative degree days (ADD) against the TBS for the decomposition cycle of Trial 1 with the blue and orange lines indicating the experimental pigs (EP1 and EP2 respectively), and the gray line the control pig 1 (CP1). The rehydration event for EP1 and EP2 is represented with a black dot, and indirect rehydration events (rain) identified with stars. (B) Representation of the average daily temperature and total daily rainfall against ADD, and (C) a graphical representation of the insect activity scored by the regional presence of insects (head and neck, trunk, and limbs). A score of 1 represents the presence of insects on one body region, 2 means the presence in two regions and 3 on all regions. The regional scores represent insect activity over the experimental phase plotted against ADD as a timeline.

As seen in Figures 4-7, rehydration affected the decomposition progression in the experimental pig compared to the control pig for both the cooler and warmer trials. Rehydration resulted in reinitiated decomposition, with the TBS values increasing after a state of stasis. Post-rehydration differed between Trial 1 and Trials 2–4, where the TBS/ADD data points are far more clustered in Trial 1 than in the warmer Trials 2–4. The cooler Trial 1 averaged 12.5 days (±135ADD) to change 2 TBS points post-rehydration. The warmer trials (Trials 2–4) averaged ±9 days (±242ADD) to observe a change in decomposition by approximately 2.8 TBS points. The two pigs that took the longest to change in TBS post-rehydration were EP2 (20 days, ±201ADD, by 1 TBS point) and EP3 (25 days, ±575ADD by 2 TBS points). Both the larger pigs (EP3 and EP6) took longer to re-initiate decomposition post-rehydration, increasing their TBS by 2 TBS points over 25 (±182ADD) and 11 days (±575ADD), respectively. The experimental sample for all four trials displayed a trend of continued decay after rehydration. The decomposition of the control sample in all four trials progressed uniformly, and, once stasis was reached, remained unchanged or displayed small increases which corresponded with natural rehydration (rainfall) events until complete desiccation or skeletonization occurred.

An interesting observation noted during Trials 2 and 3 included the experimental pigs (EP4 and EP5) displaying a greater cadaver decomposition island (CDI) when compared to the respective control pigs of the same trials, which had not been directly rehydrated (Figures 8 and 9). A CDI is the visible deposition of broken-down tissue from decomposition, observed as a dark stain surrounding a cadaver [21]. Smaller CDIs were observed for all pigs. However, the length of the grass surrounding some of the pigs made the size of the CDI unclear.

Although efforts were put towards placing pigs in the dry seasons in the Highveld, natural rehydration (rainfall) and/or indirect rehydration occurred during the experimental phases of the research project. Indirect rehydration affected the decomposition progression of all four trials' experimental and control pig cadavers. The tarps covering the top of the cages prevented direct rehydration from rain. However, some changes were still observed due to indirect rehydration likely from the soil, e.g., increased insect activity and re-initiation of decomposition post-rehydration, as indicated by changes in TBS. In addition to the CDI color change on the ground surrounding the cadavers, color changes were also observed post-rehydration in experimental pigs EP7 and EP8, for Trial 4 (Figures 10 and 11). Rehydration for Trial 4 differed from that of Trials 1–3. The pig cadavers of Trial 4 were placed within secondary cages, however, due to excessive wind, the tarps protecting the cadavers from direct natural rehydration shifted and the two experimental pig cadavers (EP7 and EP8) were fully rehydrated with approximately 2.8 mm of rain (Figures 7, 10 and 11). However, due to the displacement of the tarp, it is thought that the tarp protected the other areas of the cadaver and the water was concentrated onto the hind limb of the pig cadavers and, therefore may have exposed them to more rain than recorded. Prior to the rehydration, the pig cadavers had dried out, leaving the tissue brown and dehydrated (Figures 10A and 11A). The skin of both pig cadavers notably changed color post-rehydration. The cadavers then subsequently dried out again and the color change did not persist as the tissue dried out and returned to the brown color. Furthermore, the different trials had rainfall occurring during different phases of decomposition, although most were very small showers (<5 mm). The indirect natural rehydration was associated with an increase in the progression of decomposition at ±600ADD for all the pigs of Trial 2 (Figure 5A,B). Trial 3 demonstrated an increase in the progression of decomposition observed for EP5 (ADD 500–600). In Trial 4, the rainfall which occurred on PMI day 32 (673ADD) contributed to the wetting event of the two experimental pigs (Figure 7A,B), which led to a re-initiation in the progression of decomposition. Although the pig cadavers were not controlled in their rehydration, the rehydration occurred during the stagnant (advanced) phase of decomposition. It is also important to note that during rainfall, the average daily temperature briefly dropped (Figures 4-7).

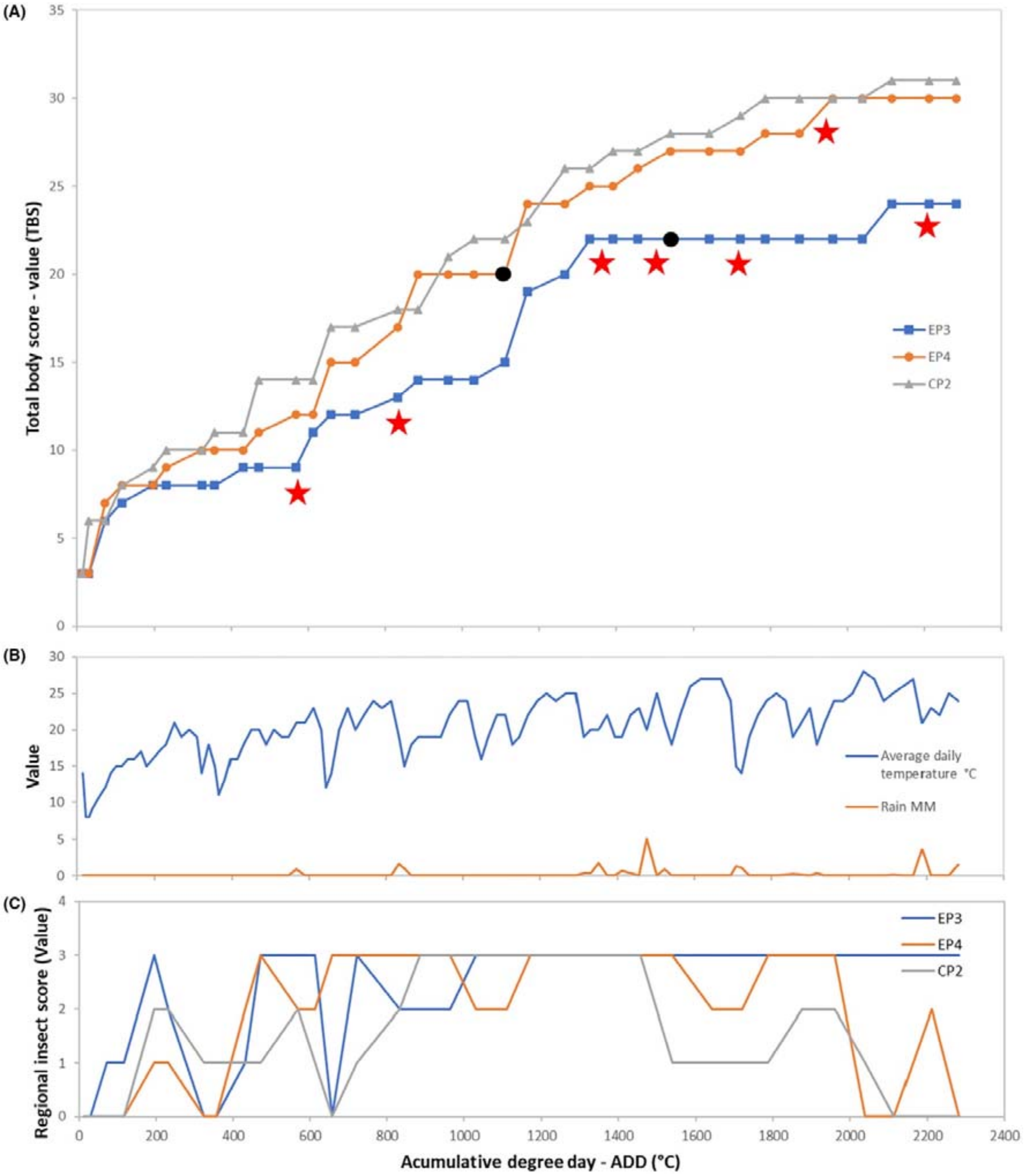

FIGURE 5. (A) Scatter plot representing accumulative degree days (ADD) against the TBS for the decomposition cycle of Trial 2 with the blue and orange lines indicating the experimental pigs (EP3 and EP4 respectively), and the gray lines the control pig 2 (CP2). The rehydration event is represented for EP3 and EP4 with a black dot, and indirect rehydration events (rain) identified with stars. (B) Represents the average daily temperature and total daily rainfall against ADD, and (C) a graphical representation of the insect activity scored by the regional presence of insects (head and neck, trunk, and limbs). A score of 1 represents the presence of insects in one region, 2 means the presence in two regions, and 3 on all regions. The regional scores represent insect activity over the experimental phase plotted against ADD as a timeline.

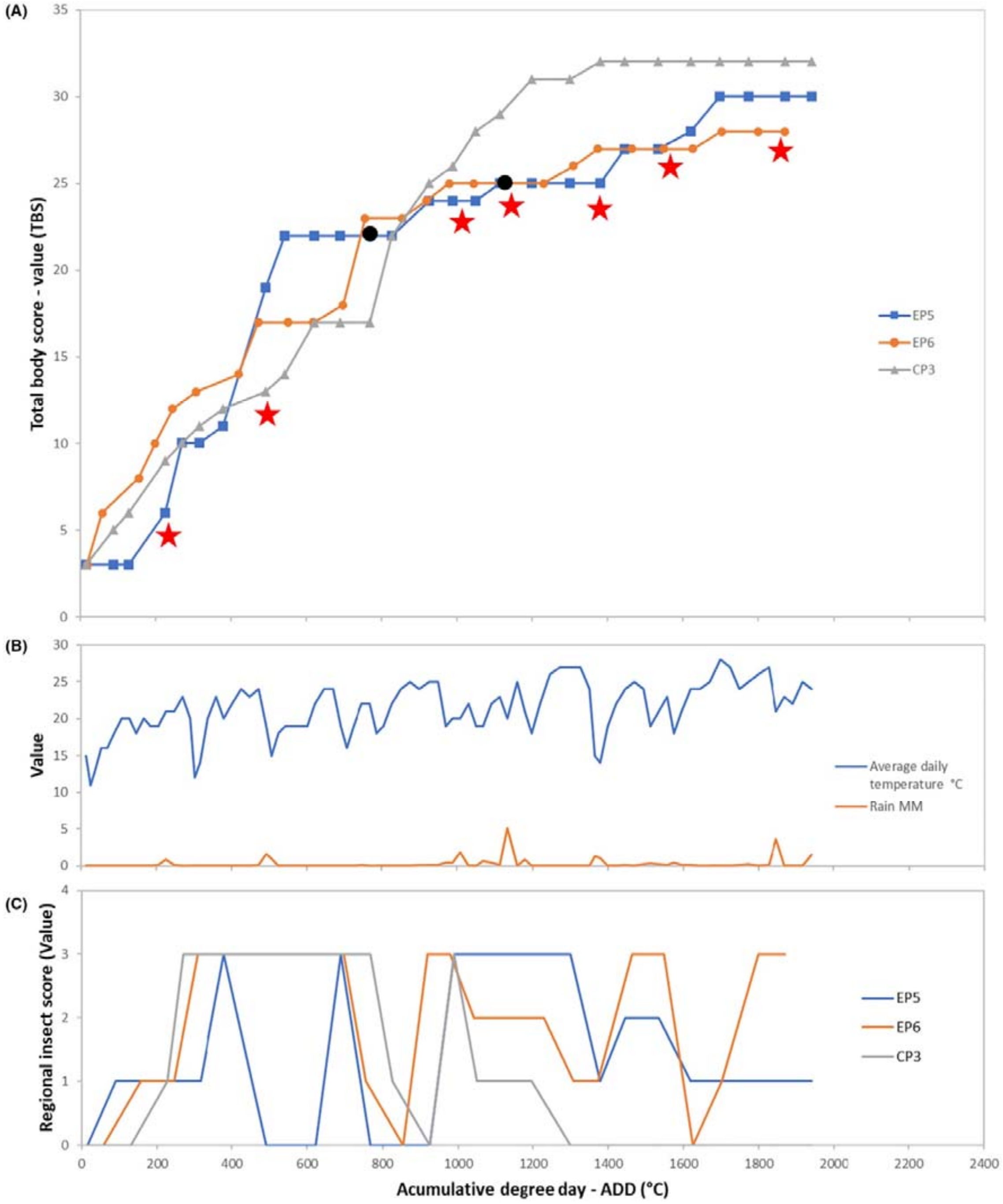

FIGURE 6. (A) Scatter plot representing accumulative degree days (ADD) against the TBS for the decomposition cycle of Trial 3 with the blue and orange lines indicate the experimental pigs (EP5 and EP6 respectively), and the gray line the control pig 3 (CP3). The rehydration event is represented for EP5 and EP5 with a black dot, and indirect rehydration events (rain) identified with stars. (B) Represents the average daily temperature and total daily rainfall against ADD, and (C) a graphical representation of the insect activity scored by regional the presence of insects (head and neck, trunk, and limbs). A score of 1 represents the presence of insects in one region, 2 means the presence in two regions, and 3 on all regions. The regional scores represent insect activity over the experimental phase plotted against ADD as a timeline.

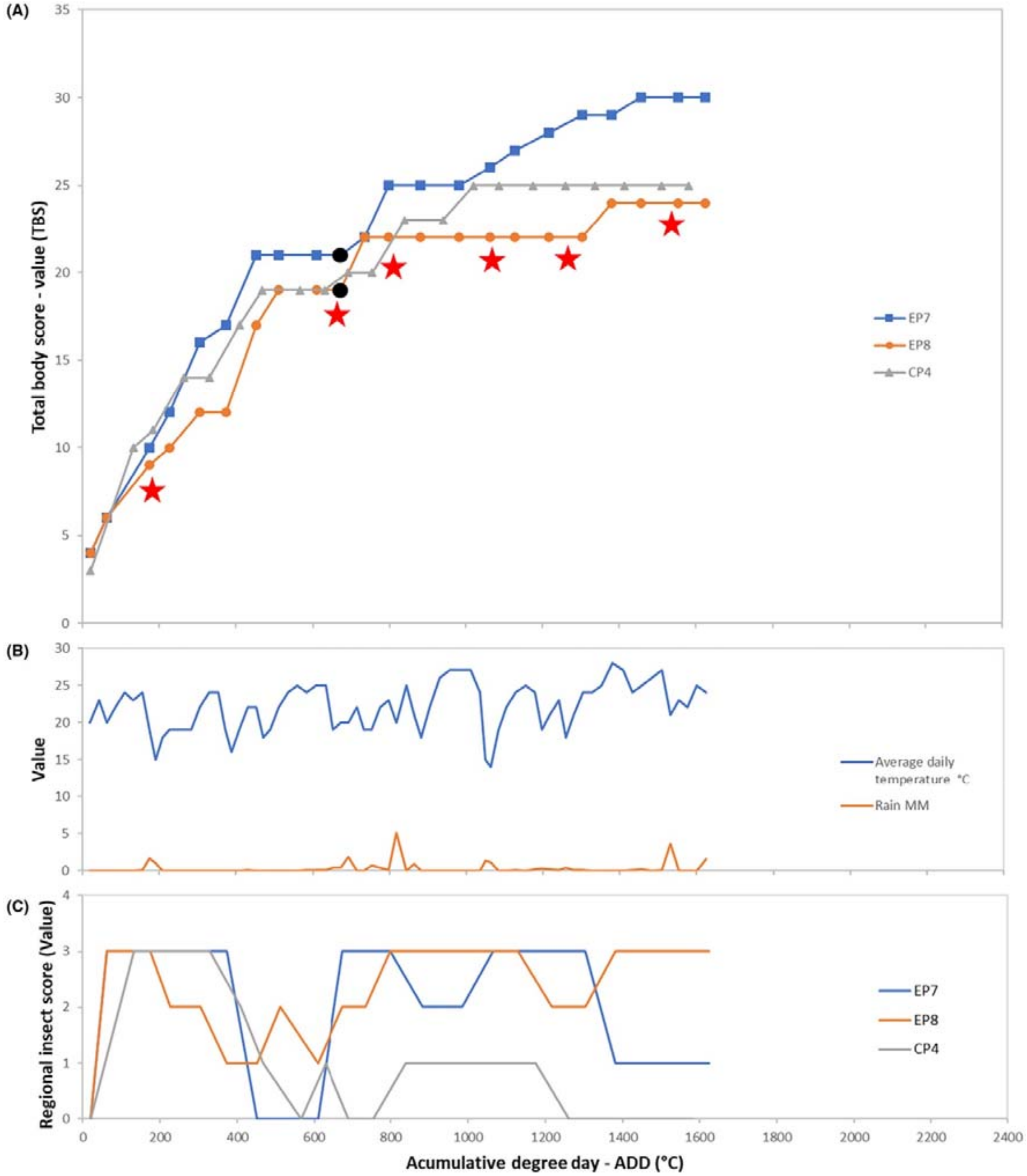

FIGURE 7. (A) Scatter plot representing accumulative degree days (ADD) against the TBS for the decomposition cycle of Trial 4 with the blue and orange lines indicate the experimental pigs (EP7 and EP8 respectively), and the gray line the control pig 4 (CP4). The rehydration event is represented for EP7 and EP8 with a black dot, and indirect rehydration events (rain) identified with stars. (B) Represents the average daily temperature and total daily rainfall against ADD, and (C) a graphical representation of the insect activity scored by the regional presence of insects (head and neck, trunk, and limbs). A score of 1 represents the presence of insects in one region, 2 means the presence in two regions, and 3 on all regions. The regional scores represent insect activity over the experimental phase plotted against ADD as a timeline.

Insect activity was seen to decrease after rainfall as well as directly after the direct rehydration. However, the insect activity then re-instated for a short period of time. The insects observed post-rehydration mainly included Hide beetles (*Dermestidae maculatus*). Flies (*Diptera*) were present post-rehydration and after indirect rehydration events, but no maggot re-colonization and migration were observed in or around the pig cadavers. The increase in activity was observed after smaller bouts of indirect natural rehydration and with increased average daily temperatures. It was also observed that, since the pig cadavers were only covered at the top, hydration from natural rehydration (rain) resulted in the soil surrounding the pig cadaver to become moist with a subsequent increase in beetle activity on the pig cadavers (Figure 12).

4 DISCUSSION

This study explored the effects of rehydration on decomposition progression and its subsequent observed effects on insect activity. To accomplish this aim, an experimental group of pig cadavers were artificially rehydrated after a period of decomposition. Artificial rehydration served as a means of controlling the timing and the quantity of water. As this study set out to replicate the Myburgh et al. [9] study, all rainfall was prevented to control for this variable as far as possible, for the whole duration of decomposition. By removing the earlier rainfall, we could see if there were any differences compared to Myburgh et al. [9], which may be attributed to rainfall from start to finish. The progression of decomposition was compared to that of a control group of pig cadavers, which were protected from hydration. A better understanding of the effects of rehydration on the progression of decomposition may provide a better understanding of the effects thereof on the PMI, regardless of the ADD. This could potentially justify the need for rain/rehydration/moisture to be incorporated into the PMI estimation methods and not only temperature, as is current practice [3].

Rehydration took place during the advanced phase of decomposition. This stage of decomposition is characterized by the remains entering a state of desiccation. Desiccation is the drying out of carrion tissue and inhibition of bacterial and insect activity resulting in the cessation of active decomposition [1, 22]. After rehydration, the experimental pig's decomposition progression for all trials increased (±2.62 TBS over ±10 days) compared to the control samples. The effects of rehydration changed the typical progression of decomposition, i.e., the experimental pigs had a re-initiation of decay after direct rehydration and then entered a second stagnant phase. The stagnant period mostly persisted until the end of the study in the control pigs. The amount of time that the cadavers are desiccated may result in a change in the progression of decomposition and needs to be further explored. Slight increases in TBS scores were also observed after cases of indirect rehydration (rainfall) for both the experimental and control pig cadavers. These results are in line with the suggestions by Myburgh et al. [9], who stated that once a body has entered a “stagnant period”, rainfall would mask this period, and the estimation of PMI would be incorrectly interpreted to be shorter. Archer [23] concluded that higher temperatures and more rainfall result in a faster progression of decomposition; this was proposed as a possible difference between the initial progression of decay in all the pigs placed in the warmer and cooler trials.

The increased environmental temperatures recorded during this research (max ±36°C) resulted in increased progression of decay in both the cooler and warmer trials, promoting insect and bacteria activity as critical thermal energy is required for increased activity to take place [3, 8, 24-26]. During rainfall events, the average temperature dropped for all the trials, with such changes indicating an effect of rainfall on the environmental temperature of the cadavers during decomposition (Figures 4-7). Megyesi's TBS/ADD formula, which is the most commonly used method of PMI estimation, attributes temperature as accounting for 80% of the variation seen in physical changes occurring during the decomposition process [3, 26-29]. When assessing the increased temperatures combined with increased humidity in this research study, the progression of decomposition was possibly altered. Temperature, as a factor affecting the progression of decomposition, cannot always be isolated from other factors, such as soil moisture [30], humidity, and rainfall, which are dependent or co-dependent on temperature [1, 8]. The factors played a combined role in the progression of decomposition along with insect and bacterial activity [26]. However, the accuracy of the method is highly variable between seasons, and the possible effects of seasonal variation are not fully understood in the case of rainfall and need to be further explored with longitudinal studies continued from this baseline research [31, 32]. Seasonality was however introduced in this study to assist in the comparison with Myburgh et al. [9], where all variables were replicated with the exclusion of rainfall. The standardized methods of PMI look solely at temperature as a factor to model formulae to establish the time since death [3, 33-35]. Rehydration and, therefore seasonality need to be considered in PMI estimations.

Additional changes noted post-rehydration included color changes in the tissue, which were specifically seen in experimental pig cadavers EP7 and EP8 for Trial 4 (Figures 10 and 11). This has also been noted in other studies where rehydration (exposure to humidity) of desiccated tissue visually changed the tissue appearance with tissue often appearing devoid of pigmentation, white or ashen, like tissue observed in arid environments that was previously browned/blackened before rehydration [36]. Victims whose remains are submerged in water also display a change in skin color [37]. Dalal et al. [37] noted that the pig cadavers that had been submerged appeared "grayish" in color along with sloughing of the skin. Therefore, it is possible that the time of exposure to rainfall might affect the rehydration of the dried decomposed skin, causing a color change. As such, whitened saturated tissue color could be indicative of desiccated tissue exposed to rain, which could affect the interpretation of post-mortem events and the timing of PMI. However, the interpretation depends on the timing of discovery as the color change did not persist once the cadavers were dehydrated once again.

Another qualitative observation made during the study is the greater CDI related to direct rehydration (Figures 8 and 9). The presence of CDIs is not uncommon in taphonomy research [21, 38, 39]. Cogswell and Cross [39] noted that CDIs differed based on the surfaces on which the cadavers were placed, e.g., concrete, grass, and gravel [39]. The authors also noted that CDIs present around a cadaver on a grass surface persisted after being exposed to rainfall compared to other surfaces, such as gravel and concrete. Soil chemistry of CDI is currently being explored as a method of PMI estimation [21, 38]. The current study supports further research into the effect of rehydration on CDI and the subsequent effects on the PMI estimation methods, as the CDI may serve as an indicator of rehydration, including how the size and depth of the pig cadaveric nutrients in the soil could affect the interpretation of PMI estimation after rehydration.

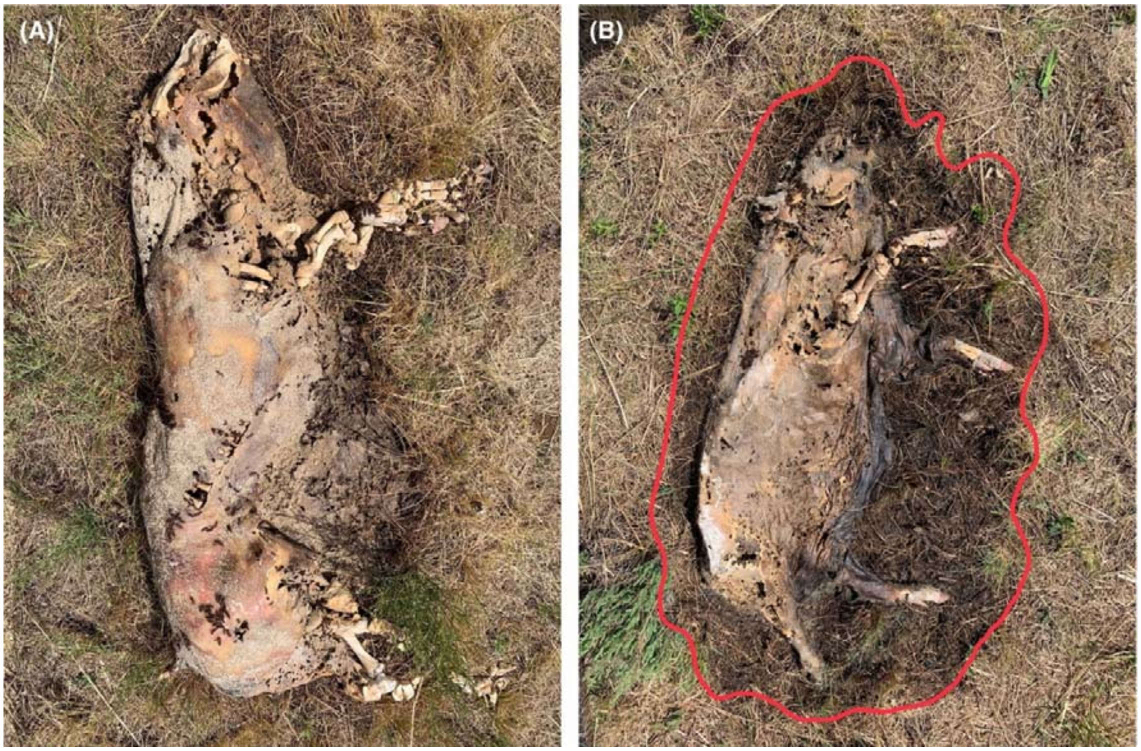

FIGURE 8. (A) Control pig 2 (CP2) on PMI 108 days (2115ADD), and (B) experimental pig 4 (EP4) on PMI 108 days (2115ADD), post-rehydration of Trial 2 displaying a greater CDI (dark stain on ground surrounding the pig cadaver outlined in red).

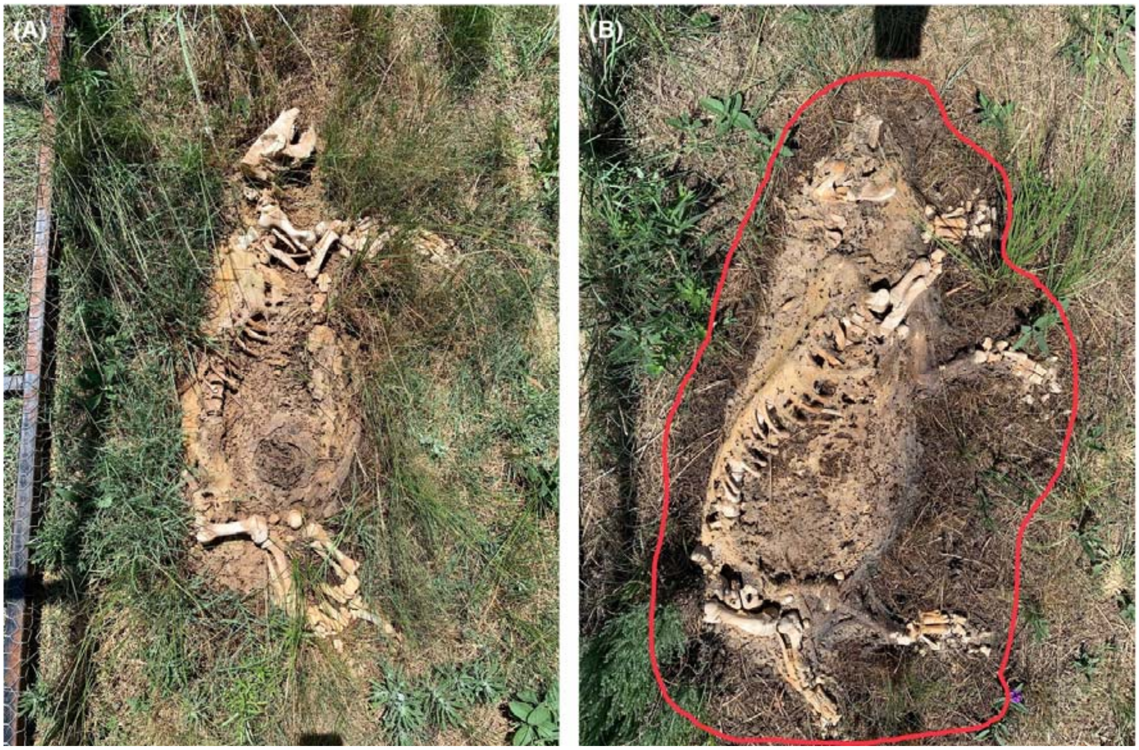

FIGURE 9. (A) Control pig 3 (CP3) on PMI 82 days (1697ADD), and (B) experimental pig 5 (EP5) on PMI 82 days (1697ADD), post-rehydration of Trial 3 displaying a greater CDI (dark stain on ground surrounding the pig cadaver outlined in red).

FIGURE 10. Hind limbs and associated trunk of experimental pig 7 (EP7) from Trial 4 illustrating skin discoloration post-rehydration. (A) EP7 on PMI 29 (609ADD) with brown dried out tissue, and (B) EP7 on PMI 32 (673ADD) post natural direct rehydration (rain) with white water saturated tissue and increased insect activity visible.

FIGURE 11. Hind limbs and associated trunk of experimental pig 8 (EP8) from Trial 4 illustrating skin discoloration post-rehydration (A) EP8 on PMI 29 (609ADD) with brown dried out tissue, and (B) EP8 on PMI 32 (673ADD) post natural direct rehydration with white water saturated tissue and increased insect activity visible.

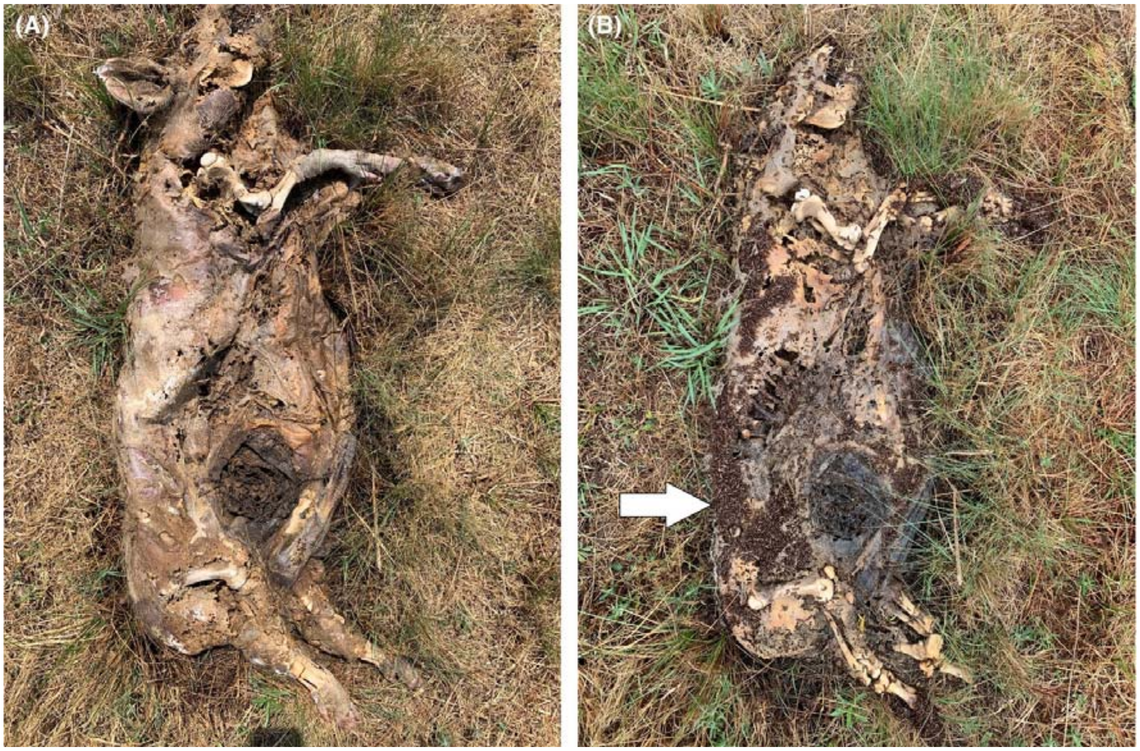

FIGURE 12. (A) Control pig 3 (CP3) on PMI day 47 (925ADD) and (B) CP3 on PMI day 50 (989ADD) colonized by beetles (*Coleoptera dermestidae*) indicated by the white arrow, post indirect natural rehydration.

Indirect rehydration occurred during the experimental phase, where the soil around the cages was rehydrated by rain even though the tarps on the cages attempted to limit the area around the cadaver. Following the indirect hydration events, the pig cadavers displayed different progressions of decomposition. In some cases, the extent of the indirect natural rehydration had no direct effect when the pig cadaver (EP3) stalled in decay, as no change was observed until after direct rehydration. Other pig cadavers experienced an increase in decay and insect activity, such as EP1, EP2 and the control pig (CP1), during the fresh to the early stages of decomposition. This may also explain the initial increased progression of decay for Trial 4 pig cadavers who experienced indirect natural rehydration during the early phase of decomposition along with warmer temperatures. This may indicate that both indirect and direct rehydration influence the progression of decomposition due to the saturation of the ground surface underneath the cadaver with increased humidity and soil moisture [1, 8, 30, 32, 40].

Insect activity was altered post-rehydration in the experimental pig cadavers. After the tissue dried, flies no longer deposited eggs on the pig cadavers. In keeping with the literature, this is due to the flesh no longer containing enough moisture to promote maggot growth and, as such, is no longer palatable for maggot consumption [26, 41]. Hide beetles, however, are biologically equipped to consume harder material such as dry skin/hide [42], although the Hide beetle larvae are very susceptible to environmental factors, and cessation of *Dermestidae* can occur once the tissue is entirely devoid of moisture or subject to waterlogging and predation [43]. The current research suggests that the increased visibility of beetles post-rehydration may be due to either an excess of water forcing the beetles inside the pig cadaver to rise to the surface and become visible to the researcher or due to the re-colonization of insects on the pig cadaver post-rehydration of the tissue. This contrasts with the work of Ayers [41], who did not observe any return of fly activity in rehydrated tissue. The current study noted flies' post-rehydration, but no visible eggs or maggot masses. Flies are associated with the fresh to the early advanced stages of decomposition, where the tissue is still moist [6]. A possible reason for the re-attraction of flies without eggs may be associated with the rehydrated tissues releasing volatile organic compounds (VOCs).

The study was limited to the analysis of rehydration during the advanced stage of decomposition. However, the analysis of rehydration during different stages of decomposition is essential to further understand the effects of rehydration on the pattern of decomposition and the possible effects on the estimation of PMI. Direct rainfall and soil moisture were limited as this research project aimed to assess the baseline effects of rehydration on decomposition, allowing a more focused analysis of the subsequent effects of rainfall when introduced during advanced decomposition. Although the effects of soil moisture were noted, this was not specifically assessed and needs to be considered in future research. This study was also restricted to using a pig model; therefore, it is essential to explore the effects of rainfall and rehydration on human cadavers. The sample size and number of pigs placed at one time were limited by the availability of the pig cadavers from the pig farm due to the need for fresh cadavers used in the study. The number of pigs per trial were further limited by the number of cages and the need to have a batch placement of the pig cadavers. The sample size of this study limited the statistical robusticity and it is recommended studies with increased samples quantify the differences observed. This pilot study supports the need for further research with placement of cadavers over multiple years as a longitudinal study. The pig cadavers in the current study were placed over two seasons, however, longitudinal studies would allow for a deeper understanding of the effects of seasonality on a larger scale. Larger sample sizes in conjunction with longitudinal studies would allow for more robust statistical analysis and the possible incorporation of rehydration into PMI estimation formulae.

In conclusion, this study found that rehydration influences the progression of decomposition as well as insect activity. The findings of this research have provided baseline data relating to the effects of rehydration on decomposition. However, the limitations associated with this study, which necessarily affected the study design, should be considered when interpreting these results and should be addressed in future studies. Notwithstanding, the study highlights the need for research involving the effect of rainfall due to the possible substantial alterations that could occur in the decomposition process. Also, rehydration (rainfall) should be taken into consideration when a body is found as it can mask the PMI of the remains when calculated based on the degree of decomposition, possibly resulting in a shorter PMI estimation.

ACKNOWLEDGMENTS

We thank Mr Luan Fouche (Farm manager; G.H. Braak farm in Rooipoort, Bronkhorstspruit) for donating the experimental sample (pig carcasses) along with assistant staff sourcing and transporting pig carcasses used in this research. We thank Mr Roelf Coertze (Senior farm manager; Department of Natural Science and Agriculture, University of Pretoria) and his assistant staff for granting access to the Forensic Anthropology Body farm on Miertjie Le Roux Experimental farm. We would also like to thank Mr Jacob Mekwa (Senior technician; School of Anatomical Sciences, University of the Witwatersrand), Mr Alfred Sibara (Workshop/Technical assistant; School of Anatomical Sciences, University of the Witwatersrand), and their team for constructing the cages used in the research.

FUNDING INFORMATION

This work was supported by the National Institute of Justice and the Forensic Technology Centre of Excellence, the American Academy of Forensic Sciences, and the Humanitarian and Human Rights Resource Centre (HHRRC).

CONFLICT OF INTEREST STATEMENT

The authors have no conflicts of interest to report.

REFERENCES

1. Galloway A, Birkby WH, Jones AM, Henry TE, Parks BO. Decay rates of human remains in an arid environment. *J Forensic Sci*. 1989; 34(3): 607–616. https://doi.org/10.1520/JFS12680J
2. Tabor KL, Brewster CC, Fell RD. Analysis of the successional patterns of insects on carrion in southwest Virginia. *J Med Entomol*. 2004; 41(4): 785–795. https://doi.org/10.1603/0022-2585-41.4.785
3. Megyesi MS, Nawrocki SP, Haskell NH. Using accumulated degree-days to estimate the post-mortem interval from decomposed human remains. *J Forensic Sci*. 2005; 50(3): 618–626. https://doi.org/10.1520/jfs2004017
4. Bloom J. 1,173 unidentified bodies in Gauteng in 2020. PoliticsWeb 2021. https://www.politicsweb.co.za/politics/1173-unidentified-bodies-in-gauteng--jack-bloom. Accessed 05 Mar 2024
5. da Silva LAF, Vilaça W, Azevedo D, Majella G, Silva IF, Silva BF. Missing and unidentified persons database. *Forensic Sci Int Genet Suppl Ser*. 2009; 2(1): 255–257. https://doi.org/10.1016/j.fsigss.2009.08.090
6. Clark M, Worrell M, Pless J. Post-mortem changes in soft tissue. In: W Haglund, M Sorg, editors. Forensic taphonomy: the post-mortem fate of human remains. Broca Raton, FL: CRC Press; 1997. p. 151–164.
7. Galloway A. The process of decomposition: a model from the Arizona-Sonoran Desert. In: W Haglund, M Sorg, editors. Forensic taphonomy: the post-mortem fate of human remains. Broca Raton, FL: CRC Press; 1997. p. 139–150.
8. Mann RW, Bass WM, Meadows L. Time since death and decomposition of the human body: variables and observations in case and experimental field studies. *J Forensic Sci*. 1990; 35(1): 103–111. https://doi.org/10.1520/jfs12806j
9. Myburgh J, L'Abbé EN, Steyn M, Becker PJ. Estimating the post-mortem interval (PMI) using accumulated degree-days (ADD) in a temperate region of South Africa. *Forensic Sci Int*. 2013; 229(1–3): 165.e1–165.e6. https://doi.org/10.1016/j.forsciint.2013.03.037
10. Conradie DC. South Africa's climatic zones: today, tomorrow. In: proceedings of the international green building conference and exhibition: future trends and issues impacting on the built environment; 2012; Sandton, South Africa. http://hdl.handle.net/10204/6064
11. Engelbrecht CJ, Engelbrecht FA. Shifts in Köppen-Geiger climate zones over southern Africa in relation to key global temperature goals. *Theor Appl Climatol*. 2016; 123(1–2): 247–261. https://doi.org/10.1007/s00704-014-1354-1
12. South African Weather Service. Weather SA. https://www.weathersa.co.za/home/aboutclimateatsaws. Accessed 19 Feb 2022.

13. Sutherland A, Myburgh J, Steyn M, Becker PJ. The effect of body size on the rate of decomposition in a temperate region of South Africa. *Forensic Sci Int*. 2013; 231(1–3): 257–262. https://doi.org/10.1016/j.forsciint.2013.05.035
14. Kelly JA, Van Der Linde TC, Anderson GS. The influence of clothing and wrapping on carcass decomposition and arthropod succession during the warmer seasons in Central South Africa. *J Forensic Sci*. 2009; 54(5): 1105–1112. https://doi.org/10.1111/j.1556-4029.2009.01113.x
15. Keyes CA, Myburgh J, Brits D. Taphonomic bone trauma caused by southern African scavengers. *Int J Leg Med*. 2020; 134: 1227–1238. https://doi.org/10.1007/s00414-019-02154-6
16. Stokes KL, Forbes SL, Tibbett M. Human versus animal: contrasting decomposition dynamics of mammalian analogues in experimental taphonomy. *J Forensic Sci*. 2013; 58(3): 583–591. https://doi.org/10.1111/1556-4029.12115
17. Matuszewski S, Hall MJR, Moreau G, Schoenly KG, Tarone AM, Villet MH. Pigs vs people: the use of pigs as analogues for humans in forensic entomology and taphonomy research. *Int J Leg Med*. 2020; 134: 793–810. https://doi.org/10.1007/s00414-019-02074-5
18. Keough N, Myburgh J, Steyn M. Scoring of decomposition: a proposed amendment to the method when using a pig model for human studies. *J Forensic Sci*. 2017; 62(4): 986–993. https://doi.org/10.1111/1556-4029.13390
19. Dyson LL. Heavy daily-rainfall characteristics over the Gauteng province. *Water SA*. 2009; 35(5): 627–638. https://doi.org/10.4314/wsa.v35i5.49188
20. World Weather Online. https://www.worldweatheronline.com Accessed 7 Jan 2022.
21. Fancher JP, Aitkenhead-Peterson JA, Farris T, Mix K, Schwab AP, Wescott DJ, et al. An evaluation of soil chemistry in human cadaver decomposition islands: potential for estimating post-mortem interval (PMI). *Forensic Sci Int*. 2017; 279: 130–139. https://doi.org/10.1016/j.forsciint.2017.08.002
22. Pietro CC, Di Vella G, Introna F. Factors affecting decomposition and Diptera colonization. *Forensic Sci Int*. 2001; 120(1–2): 18–27. https://doi.org/10.1016/S0379-0738(01)00411-X
23. Archer MS. Rainfall and temperature effects on the decomposition rate of exposed neonatal remains. *Sci Justice*. 2004; 44(1): 35–41. https://doi.org/10.1016/S1355-0306(04)71683-4
24. Vass A, Bass W, Wolt J, Foss J, Ammons J. Time since death determinations of human cadavers using soil solution. *J Forensic Sci*. 1992; 37(5): 1236–1253. https://doi.org/10.1520/JFS13311J
25. Shean BS, Messinger L, Papworth M. Observations of differential decomposition on sun exposed v. shaded pig carrion in coastal Washington state. *J Forensic Sci*. 1993; 38(4): 938–949. https://doi.org/10.1520/jfs13492j
26. Finaughty DA, Morris AG. Precocious natural mummification in a temperate climate (Western Cape, South Africa). *Forensic Sci Int*. 2019; 303:109948. https://doi.org/10.1016/j.forsciint.2019.109948
27. Marella GL, Perfetti E, Manciocchi S, Arcudi G. A case of “precocious” mummification. *J Forensic Leg Med*. 2013; 20(2): 122–124. https://doi.org/10.1016/j.jflm.2012.06.013
28. Kadej M, Szleszkowski Ł, Thannhäuser A, Jurek T. A mummified human corpse and associated insects of forensic importance in indoor conditions. *Int J Leg Med*. 2020; 134: 1963–1971. https://doi.org/10.1007/s00414-020-02373-2/Published

29. Lennartz A, Hamilton M, Weaver R. Moisture content in decomposing, desiccated, and mummified human tissue. *Forensic Anthropol*. 2020; 3(1): 1–16. https://doi.org/10.5744/fa.2020.1001
30. Ross AH, Hale AR. Decomposition of juvenile-sized remains: a macro- and microscopic perspective. *Forensic Sci Res*. 2018; 3(4): 294–303. https://doi.org/10.1080/20961790.2018.1489362
31. Forbes MNS, Finaughty DA, Miles KL, Gibbon VE. Inaccuracy of accumulated degree day models for estimating terrestrial post-mortem intervals in Cape Town. *South Africa Forensic Sci Int*. 2019; 296: 67–73. https://doi.org/10.1016/j.forsciint.2019.01.008
32. Giles SB, Harrison K, Errickson D, Márquez-Grant N. The effect of seasonality on the application of accumulated degree-days to estimate the early post-mortem interval. *Forensic Sci Int*. 2020; 315:110419. https://doi.org/10.1016/j.forsciint.2020.110419
33. Schiel M. Using accumulate degree days for estimating the post-mortem interval: a reevaluation of Megyesi's regression formulae [dissertation]. Indianapolis, IN: University of Indianapolis; 2008.
34. Moffatt C, Simmons T, Lynch-Aird J. An improved equation for TBS and ADD: establishing a reliable postmortem interval framework for casework and experimental studies. *J Forensic Sci*. 2016; 61(23): 201–207. https://doi.org/10.1111/1556-4029.12931
35. Jeong SJ, Park SH, Park JE, Park SH, Moon T y, Shin SE, et al. Extended model for estimation of ambient temperature for post-mortem interval (PMI) in Korea. *Forensic Sci Int*. 2020; 309:110196. https://doi.org/10.1016/j.forsciint.2020.110196
36. Byard R, Simpson E, Forbes SL. Arid climate adipocere—the importance of microenvironment. *J Forensic Sci*. 2020; 65(1): 327–329. https://doi.org/10.1111/1556-4029.14152
37. Dalal J, Sharma S, Bhardwaj T, Dhattarwal SK, Verma K. Seasonal study of the decomposition pattern and insects on a submerged pig cadaver. *J Forensic Leg Med*. 2020; 74:102023. https://doi.org/10.1016/j.jflm.2020.102023
38. Barton PS, Reboldi A, Dawson BM, Ueland M, Strong C, Wallman JF. Soil chemical markers distinguishing human and pig decomposition islands: a preliminary study. *Forensic Sci Med Pathol*. 2020; 16(4): 605–612. https://doi.org/10.1007/s12024-020-00297-2
39. Cogswell GC, Cross PA. The effects of surface variation on the decomposition of pig carcasses. *J Forensic Leg Med*. 2021; 79:102108. https://doi.org/10.1016/j.jflm.2020.102108
40. Vass AA. The elusive universal post-mortem interval formula. *Forensic Sci Int*. 2011; 204(1–3): 34–40. https://doi.org/10.1016/j.forsciint.2010.04.052
41. Ayers LE. Differential decomposition in terrestrial, freshwater, and salt- water environments: a pilot study [dissertation]. San Marcos, TX: Texas State University; 2010.
42. Schroeder H, Klotzbach H, Oesterhelweg L, Püschel K. Larder beetles (*Coleoptera dermestidae*) as an accelerating factor for decomposition of a human corpse. *Forensic Sci Int*. 2002; 127: 231–236. https://doi.org/10.1016/S0379-0738(02)00131-7
43. Archer MS, Elgar MA. Cannibalism and delayed pupation in hide beetles, Dermestes maculatus DeGeer (*Coleoptera: Dermestidae*). *Aust J Entomol*. 1998; 37: 158–161. https://doi.org/10.1111/j.1440-6055.1998.tb01564.x