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Title

Decomposition variability between the scene and autopsy examination and implications for post-mortem interval estimations

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Abstract

Forensic experts rely on scene and/or autopsy photographs to estimate the post-mortem interval (PMI) when an *in-situ* assessment of decomposition is unfeasible. The degree of decomposition may vary between the scene and autopsy, which importantly could affect estimations of the unknown PMI in forensic casework. This study aimed to investigate decomposition variability between the scene and autopsy and assess the subsequent effect on the accuracy of PMI estimations. Scene and autopsy photographs from 94 cases with known PMI were used from the Allegheny County Office of the Medical Examiner in Pittsburgh, United States. The total decomposition scoring (TDS) method measured the overall decomposition level, and 28 markers of decomposition were recorded as a percentage of the total body surface area (TBSA). In 60% of cases the TDS had increased at autopsy causing significant overestimations of the autopsy PMI and 86% of decomposition markers varied between the scene and autopsy. Decomposition progressed during mortuary time lags (MTL) of 3 to 44 hours, where bodies were stored in a pre-autopsy refrigerator at 4°C, suggesting that refrigeration may not always delay decomposition. This research also assisted in validating photographs as a proxy for real-time decomposition assessments. While the autopsy photographs conferred higher quality than the scene photographs, the scene photographs produced more accurate PMI estimations. Forensic experts should exhibit caution when estimating the PMI from autopsy photographs alone, as they may not accurately reflect scene decomposition. To prevent misinterpretation of the PMI estimation, both scene and autopsy photographs should always be requested.

Key Words

Post-mortem interval

Decomposition

Forensic pathology

Photographs

Autopsy

Accumulated degree days

Introduction

The estimation of the post-mortem interval (PMI) aids medico-legal death investigations by identifying suspect(s) in homicide cases, establishing the cause of death, and identifying the deceased [1, 2]. The forensic pathologist, as well as the forensic physician, forensic anthropologist, and forensic entomologist, are considered specialists in estimating the PMI using a variety of techniques [3-7]. Within all these disciplines, it can be considered best practice to attend both the scene and autopsy examination and report a PMI estimation [8-10]. However, when scene and/or autopsy attendance is not possible, photographs routinely captured by crime scene investigators (CSI) may be the only source available for the forensic experts to estimate the PMI [1, 11, 12]. To date, no study has investigated the validity of scene and autopsy photographs as a proxy for real-time taphonomic observations to estimate the PMI in forensic casework.

Depending on the case circumstances, there may be several reasons that prevent forensic experts from attending the scene. In the UK this could include: i) police delays in determination of suspicious deaths and requesting the specialist ii) expert unavailability or difficulties in accessing a geographically remote scene location, and iii) in outdoor deaths where poor weather is imminent or hazardous factors are present, the rapid removal of the body from the scene to the mortuary in favour of evidence preservation [8]. In cold case reviews, the prolonged time between the date of recovery and reinvestigation, creates further reliance on scene and autopsy photographs to provide visual evidence of the decomposition state *in-situ* [13]. This arguably renders the estimation of the PMI even more problematic if experts have not attended the scene and the assessment of decomposition is based only on photographs.

Similarly, there may be instances where an autopsy examination is unattended by the forensic expert. For example, hazardous deaths imposing a CBRN risk may require autopsies with a high degree of biosecurity, thereby limiting the number of attending personnel, with the exception of the forensic pathologist. Deaths involving UK and US military casualties may have an initial autopsy conducted in a foreign country (or not conducted at all) and the body is only later repatriated, by which time decomposition has progressed with the number of days or weeks passed. In UK cases requiring a second defence autopsy, several days or even weeks could have elapsed after the primary autopsy, and if the suspect has been charged after the disposal of the body, this will be restricted to a review of the initial autopsy photographs [14]. Collectively, this reiterates the need to evaluate the use of decomposition case photographs as an appropriate substitution for real-time taphonomic observations and PMI estimations when either scene or autopsy attendance is not possible.

Following the recovery of the body from the scene and before the commencement of an autopsy examination, the body will be transported to the mortuary for cold storage preservation. The purpose of refrigerated mortuary storage to delay decomposition is widely canonised in forensic taphonomic literature and is routinely practiced [1, 15]. The degree and rate of decomposition are likely to change over this period due to: i) the environmental transition of the body into cold storage, and ii) the duration of cold storage which correlates with the PMI. Entomological case studies report extensive blowfly larvae development at standard mortuary refrigeration temperatures of 4°C, and air subsequent caution on PMI interpretation from autopsy entomology samples [16, 17]. Research has also explored changes to algor mortis [18] and abdominal gas formation [19] that occur during mortuary storage. However, there are a multitude of decomposition markers and the rate at which decomposition may progress during mortuary storage is yet to be quantified. Investigating this could prevent misinterpretation of the PMI estimation when only the scene or autopsy photographs are available in cases where the PMI remains completely unknown [11].

The use of scene and autopsy photographs has long proven viable to assess soft-tissue decomposition in experimental taphonomy [7, 20-23]. Galloway et al. [20] used autopsy photographs from 189 decomposition cases to create their landmark sequence of five decay stages. These stages were later modified by Megyesi et al. [21] in their development of the 'Total Body Score' (TBS) method from photographs of 68 scene and/or autopsy cases, which modelled decomposition as a function of the PMI and accumulated degree-days (ADD). While the TBS model has repeatedly failed to be validated for use in forensic casework, this is commonly attributed to the rigid ordering of stage-specific decomposition markers and discrepancies between the estimated ADD and the actual ADD; rather than any issues with the photographic origin of the model [22, 24-28]. More recently, Gelderman et al. [12] 'Total Decomposition Score' (TDS) was developed from post-mortem photographs and is arguably an improvement of the TBS method due to the flexibility that is introduced in accounting for the non-binary sequencing of decomposition markers.

In recent years, a small body of research has explored the validity of decomposition photographs as a substitute for *in-situ* assessments in experimental taphonomy research. High concordance rates between observers have been found when using images of porcine analogues and donated human cadavers [29, 30]. Comparison of photographs to *in-situ* observations of porcine samples [11] and donated human cadavers [31] have also found high inter-observer reliability, agreeing that digital images can be used as a proxy for experimental taphonomic observations. However, the sample observers in the aforementioned studies were derived from an academic pool of individuals at a human taphonomy facility (HTF), rather than forensic experts who may rely on photographs to assess decomposition for PMI estimations in forensic practice. Gelderman et al. [7] found poor agreement between forensic physicians when estimating the PMI based on their experience from scene and/or post-mortem photographs. While they recognised that the photographs limited the identification of some decomposition markers (due to clothing, scent, and palpation), the effect this conferred on the accuracy of PMI estimations was beyond the scope of their study [7].

The aim of this study was to investigate decomposition variability between the scene and autopsy and assess the subsequent effect on the accuracy of PMI estimations. Two key research questions were asked: i) is decomposition variability present between the scene and autopsy photographs and if so, ii) does this effect the accuracy of subsequent PMI estimations when only scene or autopsy photographs are available? This research assists in validating scene and autopsy decomposition photographs for PMI estimations, when in-situ scene or autopsy attendance is not possible in forensic casework.

Method

This study was conducted at the Allegheny County Office of the Medical Examiner (ACOME) in the metropolitan area of Pittsburgh, Pennsylvania, US. The ACOME provides medico-legal death investigation services for all sudden, unexplained, and unnatural (including suicidal, homicidal, and accidental) deaths within a population of 1.2 million people [32, 33]. It has a unique infrastructure in housing a morgue operation service (comprised of forensic investigation, autopsy, and histology departments) and a suite of forensic laboratory services for evidence evaluation to assist law enforcement and judicial court systems [32]. For the purpose of this study, resources from the forensic investigation and autopsy departments were utilised.

<u>Case Selection</u>

A total of 94 cases over a 10-year period (2007 to 2016) were selected for analysis from the ACOME Medical Examiner's Information Management System (MEIMS). The MEIMS contains death investigation and autopsy reports along with scene and autopsy case photographs for each case. The ACOME is responsible for allocating 'morgue dispositions' for cases, whereby complete (full) or external autopsy examinations are conducted by a forensic pathologist to determine the manner and cause of death. In cases where internal examination of organs is not believed to affect the determination of cause and manner of death, an external examination is conducted alongside toxicological analysis [32].

There was a total of 12,278 cases received at the ACOME from 2007 to 2016, of which 12,090 were morgue disposition cases, meaning a pathological autopsy examination had been completed (Figure 1). Cases were first excluded if the death was imminent following a natural disease process ('no jurisdiction') or the ACOME could issue the death certificate if the cause of death could be determined without an autopsy ('we will issue') (n=188). Cases were then selected based on their 'decomposition code': a one-word descriptor of the decay level that is assigned by autopsy technicians in consultation with forensic pathologists at the start of the autopsy exam. To retrieve decomposition cases, the following decomposition code filters were applied: *early decomposition, moderate decomposition, advanced decomposition, mummification, and skeletonization,* which resulted in 2,078 cases (Figure 1).

Only adult cases were included that had not been subject to burning or burial, as these conditions can affect the interpretation of taphonomic changes [16]. All bodies were complete with no missing body parts. Of the remaining 2,032 cases; a PMI was available in 1,216 cases. Cases were excluded if the PMI was reported as a range (e.g., 3 to 4 hours or 3 to 4 days) including vague terminology, such as: 'several weeks'. Cases were also excluded if the PMI was based on a source that conferred lower confidence (e.g., circumstantial scene evidence such as newspaper dates or shelf-life dates of food items). This resulted in 320 cases where the PMI was reported to a specific date and originated from a high confidence source (e.g., concluded police reports and technology-derived methods such as last known mobile phone activity, and access to social media accounts). The final inclusion criterion required temperature data recorded from the scene, which resulted in a final sample size of 94 cases (Figure 1).

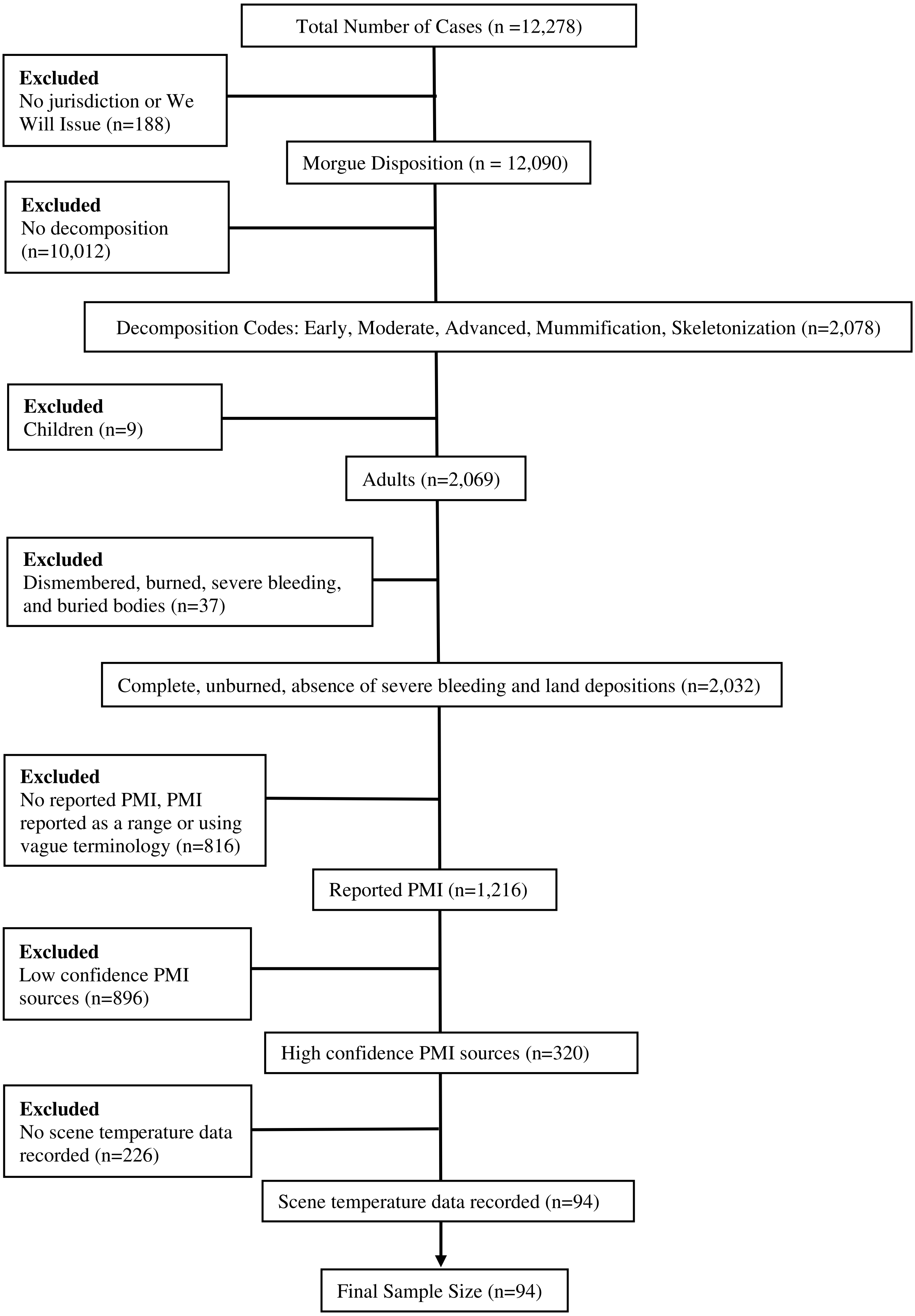

Fig. 1 Criteria for Decomposition Case Selection

Recorded Variables

Death investigation case reports were used to record intrinsic, scene, and environmental variables (Table 1). Intrinsic variables were characterised by the demographics of the sample and internal factors that vary between individuals. Scene variables related the body to the context of the scene, and extrinsic variables comprised environmental factors that could influence the state of decomposition.

	Variable	Description
Intrinsic	Age	Recorded age at death.
	Weight (pounds)	Recorded weight upon intake at the ACOME.
	Height (inches)	Recorded height upon intake at the ACOME.
	Body mass index (BMI)	Calculated as BMI = body weight/height $^{\text{squared}}$ Classified as follows: Underweight (<18.5), normal (18.5 to <25), overweight (25 to <30), class I obesity (30 to <35), class II obesity (35 to <40), class III obesity (>40).
	Cause of death	There are an infinite number of causes of death determined by the forensic pathologist.
	Manner of death	ACOME pathologists can assign one of five manners of death: natural, accidental, suicide, homicide, undetermined [32].
Scene	Death location	Recorded as indoors/outdoors, then further sub-categorised to include residence, vehicle, bath water (indoors); and woodland, garden, river water, road (outdoors).
	Body Position	Recorded as supine, prone, lateral, kneeling, seated, or suspended.
	Clothing	Recorded as fully clothed, partially clothed, unclothed, and covered (e.g., bedding material).
Environmental	Temperature	Recorded at the scene by medico-legal death investigators. For indoor cases, a single thermostat reading was taken within the residence or by using a portable ambient air thermometer (for vehicle cases) and water thermometer (for bathwater cases). For outdoor cases, the mean daily temperature was recorded using data from the nearest location weather station for land deaths and a water thermometer for river deaths. In all cases the temperature readings were recorded by photographs at the start of the investigation.
	Season	Meteorological seasons were recorded as winter (December, January, February), spring (March, April, May), summer (June, July, August), and autumn (September, October, November).
	Mortuary time lag (MTL)	Recorded as the time (in hours) between the recovery of the body from the scene and the commencement of the autopsy examination.

Table 1 Recorded intrinsic, scene, and environmental variables

The time period between the recovery of the body from the scene and the commencement of the autopsy examination was referred to as the ‘mortuary time lag’ (MTL). The MTL was recorded to evaluate its effect on decomposition variability between the scene and autopsy photographs. During the MTL, bodies were stored in a mortuary refrigerator at 4°C until the autopsy examination.

Recording Decomposition

In addition to the assigned autopsy decomposition codes, two methods were employed to record observable changes in decomposition between the scene and autopsy photographs. It is protocol for medico-legal death investigators to document post-mortem changes including livor mortis, rigor mortis, the degree of decomposition, and insect activity at the scene [34]. This information was contained in the death investigation reports and was used in conjunction with the forensic pathology reports to confirm the presence of decomposition markers.

Gelderman et al. [12] total decomposition scoring (TDS) method was used to score the decomposition state across three anatomical regions of the body: facial decomposition score (FDS), body decomposition score (BDS) and limbs decomposition score (LDS). Each anatomical region comprises a six-point scale ranging from a score of 1 (no visible changes) to 6 (complete skeletonization) and the scores are then summed to obtain a total decomposition score (TDS), that reflects the overall level of decomposition. To score cases, the TDS system was used in descending order. This meant that all cases started with a score of 6 in each anatomical region (complete skeletonization) and points were deducted if the descending decomposition criteria were absent. When the first criterion was present, the corresponding class score was assigned. For example, if a body presented 'skin having a leathery appearance', the BDS started at 6 (complete skeletonization) until this characteristic was visible in the BDS criteria (4.2) and the case would then be assigned a BDS of 4. This approach was taken to avoid bias in ascending scoring, where it was possible that more advanced characteristics could be missed if the scoring stopped at the first criterion visible in the photographs.

The scene and autopsy photographs were scored separately, resulting in a 'scene TDS' and 'autopsy TDS'. This facilitated a comparison of the accumulated decomposition between the scene and autopsy photographs for each case. All cases were scored, except for drowning cases (n=8), as the TDS method is designed for use on land depositions only [12]. This resulted in a total of 86 cases that were scored using TDS. In cases where the body was found 'fully clothed' (n=24), photographs of the anterior and posterior body surfaces were taken by lifting the clothing up, or removing the clothing, if it restricted the conduction of the body evaluation. This facilitated the ability to assign the BDS and LDS. The ACOME follow the US Department of Justice death investigation guidelines which state that additional photographs should be taken after the removal of any items that obstruct the external body examination [34]. The body examination requires the investigator to document scars, tattoos, injuries, and post-mortem decomposition changes, which in fully clothed cases, could only be observed by lifting or removing the clothing. However, if an area of the body surface was unobservable in fully clothed cases, the medico-legal death investigator scene notes were used in addition to the photographs, which detailed the decomposition characteristics present. The inter-observer reliability of the TDS scoring method has previously been researched finding high concordance rates between assessors [7, 12].

The second recording method employed a modified version of the Giles et al. [28] independent system which was originally developed to assess the onset of 28 decomposition markers by recording their presence or absence on the body. This recording method was modified to measure the extent to which certain decomposition markers were displayed as a percentage of the total body surface area (TBSA) between the scene and autopsy (Table 2). The TBSA method was used to score all 94 cases as it included the 8 water cases which were excluded from the TDS scoring method. For example, 'rigor mortis fixed' was marked as present or absent, whereas 'skin slippage' could also be scored using TBSA to account for the gradation of this marker.

Category	**Decomposition Markers**
Skin Appearance	Skin Slippage* (SS), Marbling* (MAR), Blistering (BLI)*, Desiccation (DES)*, Livor Mortis (LM), Skin Degloving (DG)
Skin Discoloration	Green Discoloration* (GD), Brown/Black Discoloration* (BRBL)
Insect Activity	Adult Blowflies (ABF), Oviposition (OVP), Larvae Activity (LV), Pupae Casings (PU), Cadaver Decomposition Island (CDI)
Events	Rigor Mortis Fixed (RMF), Rigor Mortis Lysed (RML), Facial Bloating (FB), Abdominal Bloating (AB), Post-bloating (PB), Hair loss (HL), Fluid purge from facial orifices (FP), Tongue Protrusion (TP), Putrefaction (PUT), Scavenging* (SCA)
Advanced Decay	Tissue Sloughing* (TS), Tissue Desiccation* (TD), Bone Exposure* (BE), Adipocere (ADP), Mould formation (MOU)

Table 2 Individual Decomposition Markers (modified from Giles et al. [28]) *indicates the 10 gradable decomposition markers where a TBSA score was also calculated to estimate the proportion (%) of the total body surface area (TBSA) that presented the decay marker.

The TBSA method is commonly used to calculate the extent of burn injuries and has previously been applied to estimate the expression of mummification in forensic cases [35]. It anatomically divides the body into 11 areas to include the anterior and posterior of the head, upper limbs, torso, back, and lower limbs; each of which has an assigned value of 9% (except for the groin at 1%). The 10 gradable decomposition markers were assigned an individual TBSA score. For example, if 'skin slippage' was present on both arms (18%) and the anterior left leg (9%) this would give a TBSA of 27% for that decomposition marker. In cases where the body was 'fully clothed' and when body surfaces were restricted by clothing, the medicolegal death investigator scene notes provided information on the decomposition characteristics observed on the body at the time of the scene examination. The TBSA method does not produce a PMI estimation and was primarily used to measure the extent of change in individual decomposition markers between the scene and autopsy.

<u>PMI</u>

Two calculations of the known PMI (PMI_{actual}) were made for each case. The first PMI, referred to as the 'scene PMI', was calculated as the time difference (in days) between the date of death and the date of discovery. The second PMI, referred to as the 'autopsy PMI', was calculated as the time difference (in days) between the date of death and the date of the autopsy, and therefore included the MTL period. This distinction was needed to give an accurate reflection of the true PMI for the scene and autopsy photographs.

The PMI was then estimated, for the scene and the autopsy, using the Gelderman et al. [12] ADD formula for both indoor and outdoor cases (Table 3). A distinction was also made between the estimated 'scene ADD' (based on the scene TDS) and 'autopsy ADD' (based on the autopsy TDS) to account for any differences in TDS between the scene and autopsy.

Indoors	Outdoors
ADD = 10^ (- 0.05 + 0.23*TDS) ± 29.6 ADD days	**ADD =** 10^ (0.03 + 0.19*TDS) ± 52.1 ADD days

Table 3 ADD Linear Regression Formula for Indoor and Outdoor Cases (Gelderman et al. [12])

Two PMI estimations ($PMI_{estimated}$) were made for each case to measure the effect of PMI estimation error between the scene and autopsy photographs against the known PMI (PMI_{actual}). The 'scene PMI' was estimated by summing the daily temperatures (above 0ºC) from the date of body discovery until the accumulated sum reached the estimated ADD value ($ADD_{estimated}$) [12]. The estimated scene PMI was the day that the estimated ADD value was reached. The second PMI estimation was referred to as the 'autopsy PMI' and was calculated in the aforementioned way, but the summing of daily temperatures also included the MTL period to account for any effect of the 4°C refrigeration on decomposition. The estimated autopsy PMI was the day that the estimated ADD value was reached.

For indoor deaths, the ambient indoor temperature was recorded at the time of discovery. Given that indoor temperatures are less subject to extreme fluctuations than outdoor temperatures, it was assumed that bodies were exposed to this temperature for the duration of the PMI. For outdoor cases, the mean daily temperatures were taken from the nearest weather station for land deaths and by using a water thermometer for river deaths.

Statistical Analysis

The statistical analysis was conducted using XLSTAT software (version 2020.5.1) in EXCEL. To test the correlation between: i) PMI and TDS and ii) ADD and TDS and iii) MTL and TDS, the Spearman rank-order correlation coefficient (rs) was used. This non-parametric statistical test can be used on continuous or ordinal data. The rs values range from -1 to +1 and indicate the strength of the correlation between two variables, ranging from 'very weak' (0.00 – 0.19) to 'very strong' (0.80 – 1.00) [36]. Linear regression was then used to determine if each independent variable: ADD, PMI and MTL was an accurate measure of the TDS (dependent variable). All independent variable values were log-transformed to create a linear relationship with TDS and plotted separately against the TDS values for the scene and autopsy. A paired two sample t-test determined any significant differences between ADD_{actual} and $ADD_{estimated}$, and the PMI_{actual} and $PMI_{estimated}$, from the scene and autopsy for each case, with statistical significance considered at $p<0.05$. Paired two sample t-tests were also used when assessing the differences in accumulated decomposition between the scene and autopsy (as measured by TDS and TBSA) and to determine significant decomposition variation between the recorded intrinsic, scene and environmental variables.

Confidence intervals of 95% were not reported for PMI estimation in this study. They are arguably not applicable to PMI estimations in real forensic cases because they are derived from specific experimental samples and not the whole population [37]. An accurate PMI estimation within the 95% confidence interval does not mean that there is 95% probability of finding the true PMI in an unknown case. Our calculations of PMI accuracy were based on assessing the number of cases that could predict PMI with 100% accuracy, or within one PMI day. Any greater than this would arguably be too wide to be considered useful for PMI estimations in homicide cases, where narrower and more accurate PMI estimations are required [28].

Results

Overall Descriptions

The sample consisted of adult males (n=64) and adult females (n=30), with an age range of 19 to 88 years. The mean age at death was 52 years (standard deviation (SD) ±17). The BMI of cases extensively ranged from 4.7 (severe thinness) to 52.3 (class III obesity), with a mean BMI of 25.2 (overweight) (SD ± 7.8). The sample was composed of natural manners of death (n=38), accidental (n=26), suicide (n=15), homicide (n=9), and undetermined (n=6). There was a total of 15 different causes of death, which were sub-categorised to 'trauma' (firearms, blunt force trauma, sharp force trauma) (n=14), drugs and/or poisoning (n=20), drowning (n=8), undetermined (n=6), asphyxiation (n=5), and natural causes (e.g., atherosclerotic cardiovascular disease, liver cirrhosis) (n=41).

Bodies were most commonly found in an indoor setting (n=69). Of the indoor deaths, these were further sub-categorised according to their location either inside a residence (n=60), bathwater (n= 3), or a vehicle (n=6). Outdoor deaths made up the remainder of the sample (n=25) and were found in a variety of settings: woodland (n = 11), river water (n=7), garden (n=4), or road (n=3). Bodies were found in a range of different positions: supine (n=39), seated (n=18), lateral (n=16), prone (n=15), suspended (n=3) and kneeling (n=3) position. Most bodies were found fully clothed (n= 57), whereas the rest were partially clothed (n=16), unclothed (n=11), or covered (n=10). The scene PMI for the sample ranged from 1 to 121 days, with a mean PMI of 8.7 days (SD ± 16). The autopsy PMI, which included the mortuary time lag (MTL), ranged from 1 to 122 days, with a mean PMI of 9.7 days (SD ± 17).

The same overall mean temperature was reported for both indoor and outdoor cases at 20°C (SD ± 8.4°C, n=69 indoors cases, and SD ± 7.3°C, n=25 outdoors). A larger temperature range was observed for indoor (-6°C to 37°C) than outdoors cases (-3°C to 28°C) across the dataset, although there was a low frequency of cases where the temperature was ≤ 0°C (indoors n=4, outdoors n=2). Outdoor river water cases had a mean temperature of 17°C (n=7), compared to 27°C for indoor bath cases (n=3).

The Allegheny County has a humid, continental climate with four distinct seasons and wide temperature variations between seasons. All four meteorological seasons and calendar months were evenly distributed, with bodies found in summer (n=26), spring (n=25), winter (n=24), and autumn (n=19). The most common month of body discovery was June (n=11), and the least common months were September (n=6) and October (n=6). Autopsy examinations were conducted for 85% of cases within 24 hrs (n=80). Cases were further sub-grouped into the following MTL ranges: 1-6 hrs (n=9), 7-12 hrs (n=10), 13-18 hrs (n=34) and 19-24 hrs (n=27). In the remaining 14 cases, the autopsy was performed within 25-45 hours (n=12) and 46-72 hours (n=2) of scene recovery.

<u>Decomposition Codes</u>

Of the pre-assigned ACOME decomposition codes, ‘early’ decomposition was most common (n=45), followed by ‘moderate’ (n=30), ‘advanced’ (n=14), ‘mummification’ (n=4) and ‘skeletonization’ (n=1). Cases at 1 PMI day were exclusively assigned ‘early decomposition’ (n=3) (Figure 2). Beyond this, all decomposition codes were represented across extensive PMI ranges.

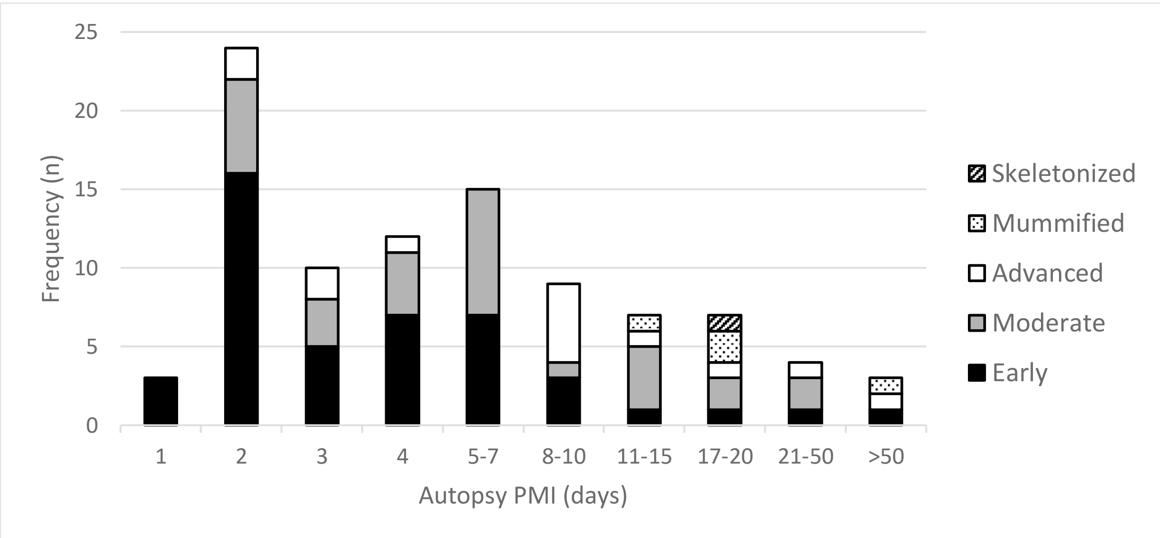

Fig. 2 Frequency of cases in each decomposition stage distributed by the Autopsy PMI (n=94). PMIs greater than 4 days, were randomly sub-grouped into ranges up to >50.

‘Early’ decomposition which was most prevalent at 2 PMI days (n=16) and was represented by every subsequent PMI range up to >50 days, with one indoor winter death of 67 PMI days subject to -0.5°C scene temperature. ‘Moderate’ decomposition peaked at a PMI range of 5 to 7 days (n=8) and was relatively widespread between 2 PMI days (n=6) and 21 to 50 PMI days (n=2). No ‘moderate’ decomposition codes were assigned after 48 PMI days. 'Advanced' decomposition was most common at 8 to 10 days (n=5) and was represented as early as 2 PMI days (n=2) where both cases were subject to high indoor temperatures of 27°C and 30°C. The earliest onset of 'mummification' was at 15 PMI days (n=1, represented in Figure 2 in the 11-15 days PMI range). 'Mummification' was also assigned at a PMI range of 17 to 20 days (n=2) and accounted for the highest PMI of 121 days. There was one case of 'skeletonization' at 17 PMI days.

Total Decomposition Scoring (TDS)

In 60% of cases, there was an increase in the autopsy TDS (n=52) and for the remaining 40% of cases, the same TDS was assigned at both the scene and autopsy (n=34) ($p<0.05$, n=86). The autopsy TDS did not decrease below the scene TDS for any case (n=86). The scene TDS varied from scores of 3 to 13 with an mean TDS of 7 (SD ± 2). Similarly, the autopsy TDS ranged from scores of 3 to 13, however, the mean TDS increased to 8 at autopsy (SD ± 2). A TDS of 6 was the most common scene TDS (n=34), which frequently encompassed criterion 2.1 (livor mortis, rigor mortis, and vibices) across the face, body and limbs. Conversely, the most frequent autopsy TDS was a score of 9 (n=29), which was reflective of characteristics such as facial and abdominal bloating, along with skin blisters, skin slippage, and marbling on the limbs. Similarly, there were statistical increases in the FDS, BDS and LDS assigned at autopsy when compared to their respective scene scores ($p<0.05$). The amount by which the FDS, BDS, LDS changed were not statistically different *between* these anatomical categories ($p>0.05$).

In the 52 cases where the TDS increased at autopsy, this occurred across a wide range of scene TDS values (3 - 9 and 12 TDS) (Figure 3). A scene TDS of 6 was the most frequent to increase at autopsy (n=26), as bodies progressed to higher scores characterised by discoloration. The greatest mean increase in TDS shown in Figure 3 represented an 'early' decomposition case that was assigned a scene TDS of 4 and an autopsy TDS of 7, which presented a TDS increase of 3. However, the greatest acceleration in TDS out of the 52 cases was noted on a 'moderate' decomposition case that was assigned a scene TDS of 5 and an autopsy TDS of 9, which presented a TDS increase of 4 (only mean TDS increases shown).

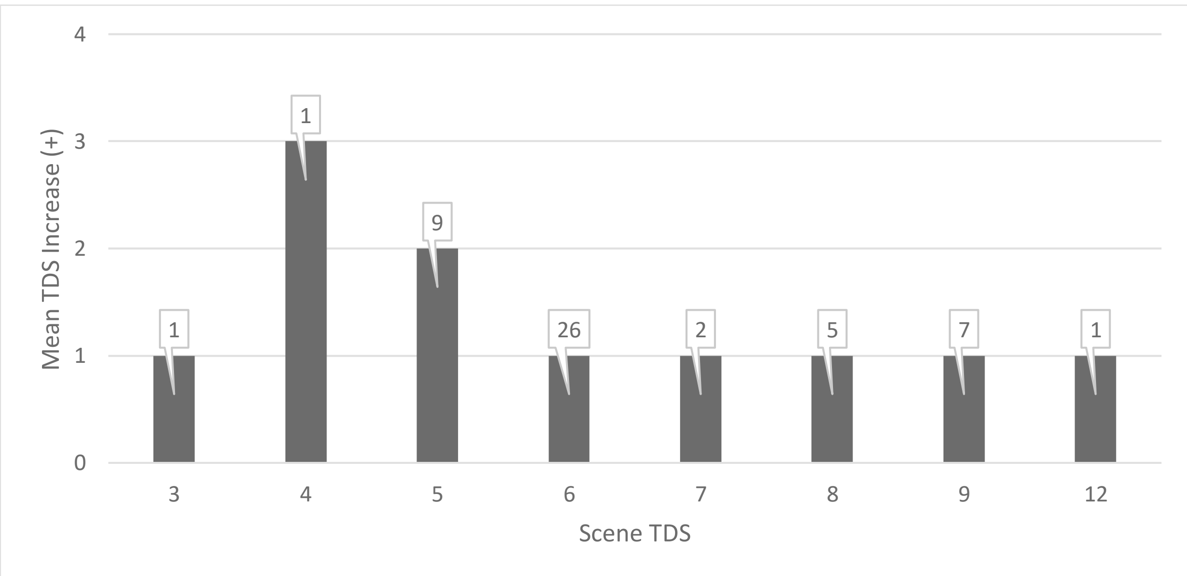

Fig. 3 Mean TDS increase from scene to autopsy TDS (n=52). Number in box above each bar represents the frequency of cases.

The mean change in TDS between the scene and autopsy was examined against the assigned ACOME autopsy decomposition codes. Bodies assigned 'early', 'moderate' and 'advanced' decomposition had a mean TDS increase between scene and autopsy scores of 1 TDS ($p<0.05$, n=52, SD ± 1) (results not shown). Of the 86 cases scored using TDS, 71% of 'moderate' decomposition cases (n=41) had an increased TDS at autopsy (n=29). Similarly, 'early' decomposition cases (n=28) had a comparably high percentage of TDS increase in 70% of cases (n=20). Conversely, 'advanced' decomposition cases (n=12) were least likely to present an increase in TDS at autopsy (25%, n=3). 'Mummification' and 'skeletonization' bodies presented no change in TDS between the scene and autopsy (n=5).

The increase in autopsy TDS was extrapolated to the *autopsy* PMI since the TDS change was measured at the autopsy examination (Figure 4). The increase in TDS between the scene and autopsy was consistently prevalent between 1 to 8 autopsy PMI days (n=44) and most prominent at 2 autopsy PMI days (n=16). There were no changes in scene and autopsy TDS between an autopsy PMI of 9 and 10 days (n=3, data not shown). The increase in TDS became apparent again at 11 to 15 autopsy PMI days (n=4), and 21 PMI days (n=1), which predominantly corresponded to 'moderate decomposition', whereas changes in TDS at 31 and 68 PMI days related to 'early' decomposition cases (n=2) (Figure 4).

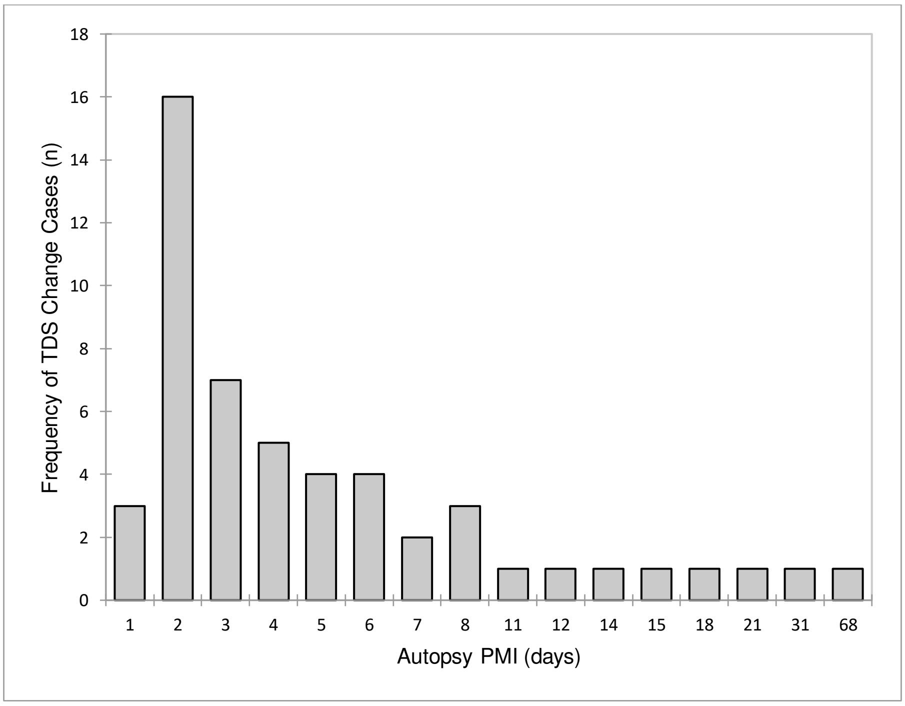

Fig. 4 Distribution of cases where the TDS increased at autopsy by the known autopsy PMI (n=52).

<u>Intrinsic, Scene and Environmental Variables</u>

All taphonomic variables were represented in the 52 cases where TDS changed and, in the 34 cases where it remained stable. However, class II and III obesity (n=6), and the temperature ranges of: 1°C to 5°C (n=3) and >30°C (n=1), presented only TDS increases (Figure 5). Conversely, the TDS remained exclusively unchanged only in cases with a 'suspended' body position (n=3) (Figure 5).

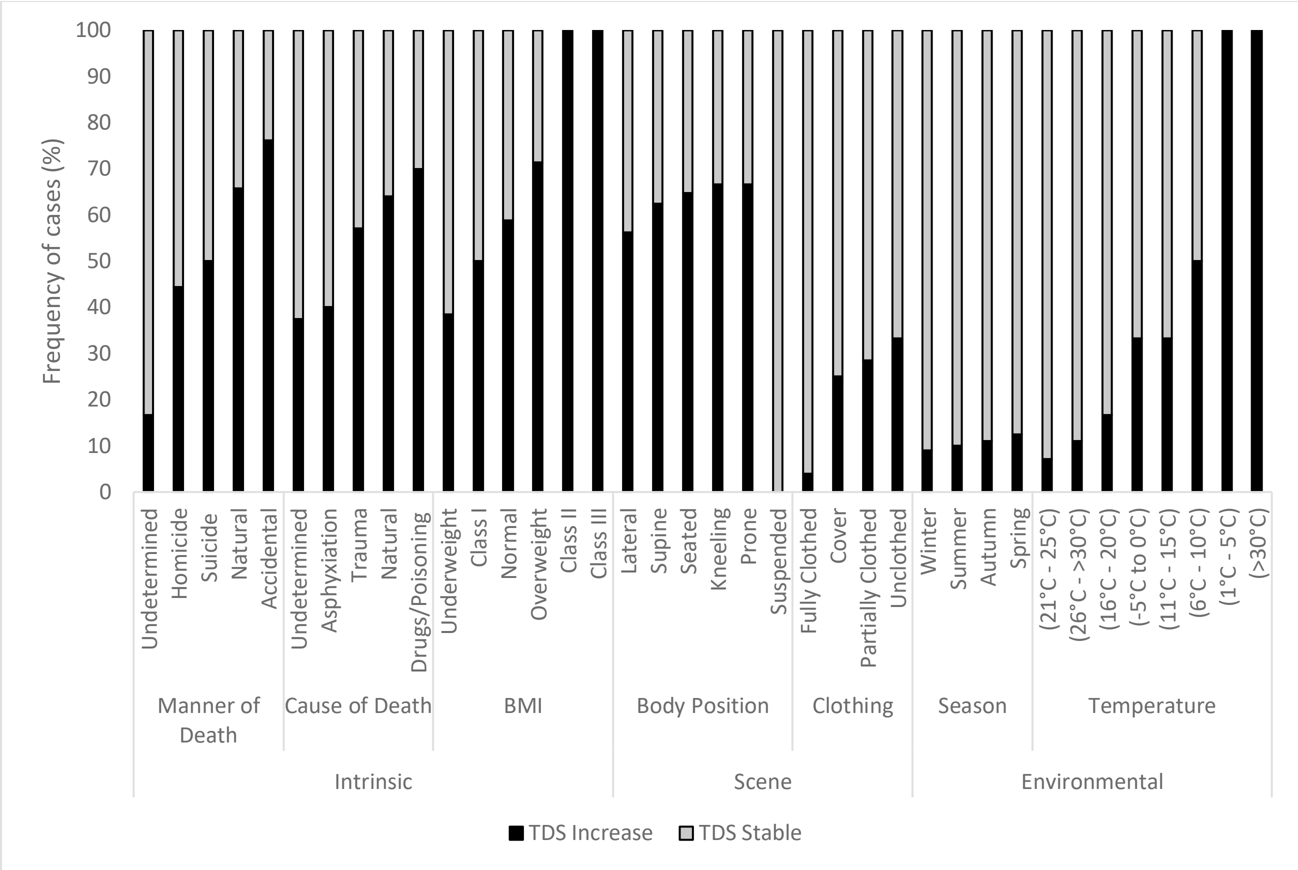

Fig. 5 Frequency of cases where the TDS increased or remained stable between the scene and autopsy by intrinsic (manner of death, cause of death, BMI), scene (body position, clothing), and environmental (season, temperature) variables (n=86).

The intrinsic variables of manner, cause of death, BMI, and the scene variable of body position, conferred a higher frequency of cases where the TDS increased, whereas the environmental variables of season and temperature, and the scene variable of clothing represented more cases of TDS stability. (Figure 5).

Cases of TDS increase were more frequently observed in 'natural' (66%, n=25) and 'accidental' (76%, n=16) manners of death, and trauma (57%, n=8), natural (64%, n=25), and drugs and/or poisoning (70%, n=14) causes of death (Figure 5). Along with class II (n=3) and class III (n=3) obesity cases, which exclusively presented TDS increases, 71% of cases 'overweight' BMI presented the next highest frequency of TDS increase (n=15).

Interestingly, TDS increase cases were more frequently observed in unclothed (33%, n=7) than fully clothed (4%, n=28) bodies (Figure 5). This finding could potentially support the viability of scene photographs to observe decomposition markers on fully clothed bodies. However, the TDS remained predominantly stable over all clothing categories. Cases of TDS change were uniformly distributed across all body positions, with the exception of 'suspended' bodies (n=3) which exclusively presented TDS stability.

Season appeared to have minimal effect on the TDS increase, where the TDS remained uniformly unchanged between the scene and autopsy in the majority of cases in winter (91%, n=10), summer (90%, n=9), autumn (89%, n=8), and spring (88%, n=7) (Figure 5). The TDS increased across every scene temperature range from -5°C to 0°C (n=4) through to >30°C (n=1), with the exception of 1°C to 5°C (n=3), >30°C (n=1). However, TDS increase cases were in the minority for all other temperature ranges, with only 7% of cases falling in the 21°C to 25°C temperature range (n=19).

In the 52 cases where the TDS increased at autopsy, the mean TDS increase was compared between each intrinsic, scene and environmental variable. Suicide (n=6), undetermined (n=1) and trauma cases (n=8) had the highest mean increase of 2 TDS, when compared to other manner and causes of death (results not shown) 'Class 1 obesity' (n=6) was the only BMI classification that presented a TDS increase of 2. However, there were no significant differences in TDS increases between the aforementioned variables and their corresponding inter-class intrinsic variables ($p>0.05$, two sample t-tests).

The scene variables of 'body position' and 'clothing' presented no significant effect on the TDS increase ($p > 0.05$). However, partially clothed bodies (n=10) and prone bodies (n=10) had a greater mean TDS increase of 2, compared to the remaining body positions and clothing types.

Seasonal variables also appeared to have minimal influence on mean TDS changes, with a mean TDS increase of 1, reported across all seasons of death. Scene temperature also had no significant effect on the TDS increase ($p > 0.05$). The highest mean TDS change of 2 was found between bodies exposed to a scene temperature ranges of 1-5°C (n=3) and 6-10°C (n=2). Moreover, in cases where the TDS increased, the reported scene temperature extensively ranged from -5°C to 35°C (n=52); which was comparable to cases with no TDS change, where the scene temperature varied from -1°C to 30°C (n=34).

<u>Mortuary Time Lag (MTL)</u>

The MTL showed a 'weak' negative correlation with the increase in TDS between the scene recovery and autopsy examination ($r_s = 0.25$, $p<0.01$, n=86) (Figure 6).

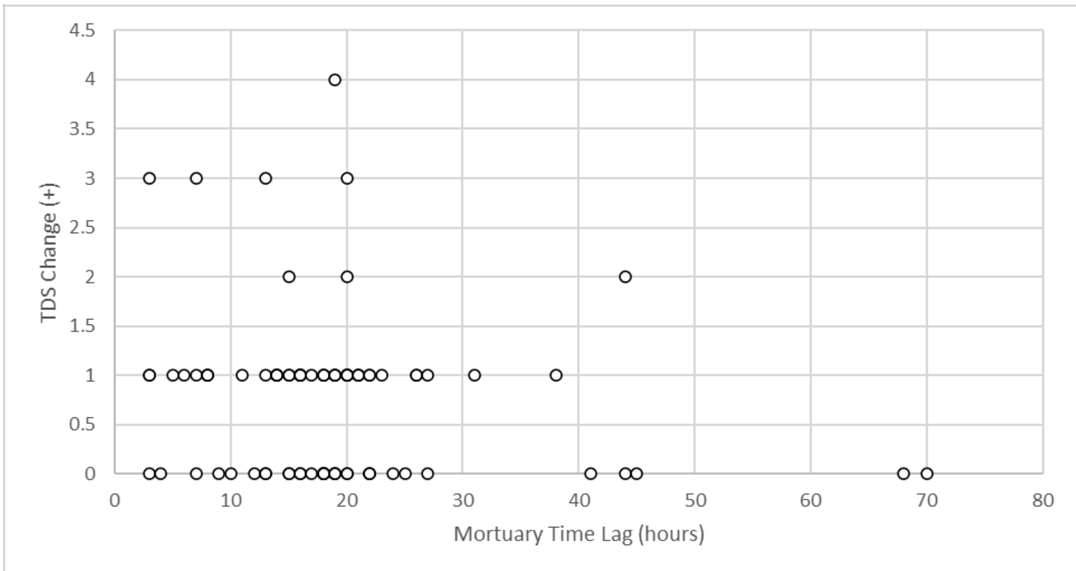

Fig. 6 The effect of the mortuary time lag (MTL) on the total decomposition score (TDS) change between the scene and autopsy examination (n=86)

In 60% of cases, marked increases in TDS were observable between a MTL of 3 to 44 hours (n=52). In the remaining 40% of cases, the TDS remained stable between 2 to 70 hours MTL (n=34). After a minimal MTL of just 3 hours, a TDS increase of 3 between the scene and autopsy, was reported for a drugs and/or poisoning death displaying early decomposition. The greatest TDS changes occurred between 3 to 20 hours and the highest TDS change of 4 was represented in a case of moderate decomposition with a natural cause of death and an MTL of 19 hours (Figure 6).

There were significant differences between the scene and autopsy TDS within each MTL group as follows: 1-6 hrs (n=7), 7-12 hrs (n=10), 13-18 hrs (n=30), 19-24 hrs (n=26) and 25-45 hours (n=11) ($p<0.05$). However, there were no significant differences between the scene and autopsy TDS when the autopsy was conducted between 46-72 hours of scene recovery ($p>0.05$, n=2). This was not surprising given that both cases had no change in TDS over the MTL (Figure 6).

There were also statistical differences between the scene and autopsy scores between each MTL group ($p<0.05$). However, this could also be due to variety in the combination of variables (e.g., decomposition stage, temperature, cause of death) that were recorded within, and between, each MTL group. When the TDS change was examined between each MTL group, statistical differences were only present between MTL 1-6 (n=7) and all other MTL groups ($p<0.05$).

<u>TDS and PMI</u>

A ‘moderate’ correlation found between the scene TDS and the scene PMI (rs = 0.434, p = <0.0001) and autopsy TDS and the autopsy PMI (rs = 0.401, p = <0.0001) (results not shown). The FDS, BDS and LDS were also moderately correlated with the scene PMI with rs values ranging between 0.442 (LDS) – 0.528 (BDS) (p = <0.0001). The FDS and BDS were also moderately correlated with the autopsy PMI, with an rs of 0.509 and 0.435, respectively (p= <0.0001), whereas the LDS was weakly correlated with autopsy PMI (rs = 0.34, p = <0.0001).

There appeared to be a very weak linear relationship between TDS and PMI ($r^2 = 0.2$) (Figure 7). When both the scene and the autopsy PMI were log-transformed this did not achieve a linear relationship with the scene TDS (r^2 =0.26, n=86) or the autopsy TDS (r^2=0.23, n=86). The low r^2 values indicated that 26% and 23% of the variation in the PMI was estimated by the TDS for the scene and autopsy, respectively.

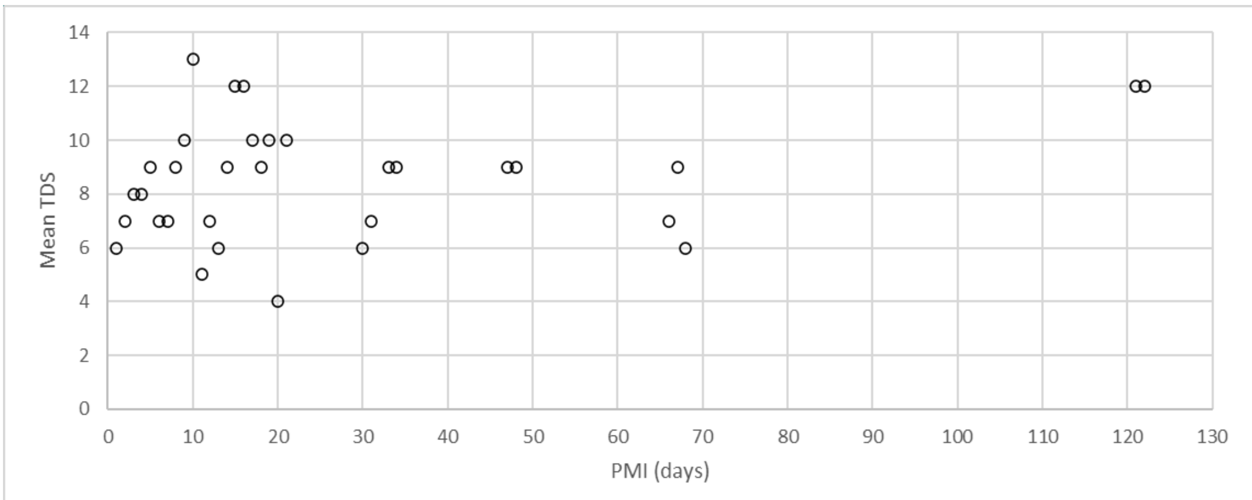

Fig. 7 Mean Scene and Autopsy TDS by Scene and Autopsy PMI (r^2 = 0.2, n=172)

TDS and ADD

TDS had a ‘very strong’ correlation with $ADD_{estimated}$ for both the scene (rs = 0.986, p = 0.0001, n=86) and the autopsy (rs = 0.977, p=0.0001, n=86). A curvilinear relationship was found between TDS and $ADD_{estimated}$ with r^2 values of 0.85 (scene TDS) and 0.84 (autopsy TDS). Log-transforming $ADD_{estimated}$ produced an effective linear regression with both scene TDS (r^2 = 0.98) and autopsy TDS (r^2 =0.97).

The TDS of outdoor cases had the strongest relationship with $ADD_{estimated}$ (r^2 = 0.91, n=19) for both scene and autopsy which increased to an r^2 value of 0.99 when $ADD_{estimated}$ was log-transformed. Indoor cases indicated that 82% of the variability in $ADD_{estimated}$ was attributed to the TDS for both scene and autopsy, which also increased to r^2 of 0.99 (n=67) when $ADD_{estimated}$ was log-transformed (results not shown).

Conversely, there was a ‘weak’ correlation observed between TDS and the ADD_{actual} for both the scene (rs = 0.263, p = 0.01) and the autopsy (rs = 0.296, p = 0.04). No linear relationship was observed between ADD_{actual} and the TDS of both the scene and the autopsy, with r^2 values of <0.05. Log-transforming ADD also did not achieve a linear relationship and only marginally improved the r^2 values to <0.08 which indicates that variation in the TDS could not be attributed to ADD_{actual}. Furthermore, there was a significant overestimation of $ADD_{estimated}$ compared to ADD_{actual} values, for both the scene and the autopsy (p<0.05, n=86).

PMI Estimation

The TDS-ADD model overestimated the PMI at both the scene and autopsy (Figure 8).

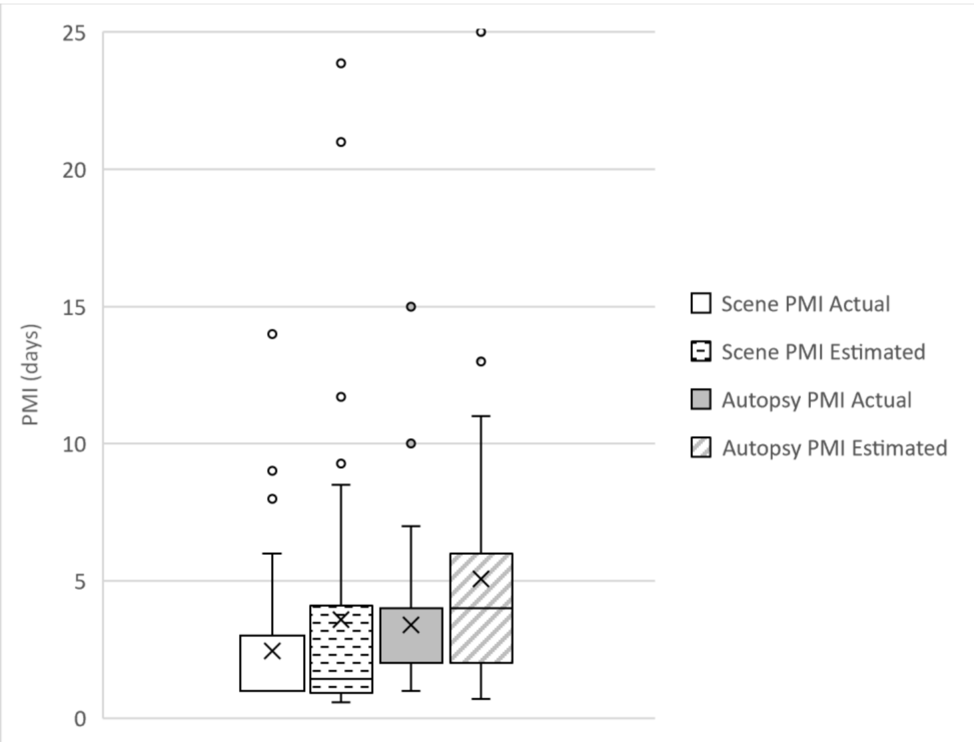

Fig. 8 Box and whisker plot comparing the PMI_{actual} to the $PMI_{estimated}$ for the scene and autopsy (n=78). Values above the upper quartile range are presented by circles. The x and horizontal line within the box plots represent the mean and median PMI, respectively. Figure 8 excludes outliers >25 PMI days (n=8) and therefore the PMI means are readjusted in the box plot and differ from the reported PMI means in-text.

This overestimation was insignificant between the scene PMI_{actual} and $PMI_{estimated}$ ($p>0.05$, n=86) but significant between the autopsy PMI_{actual} and the $PMI_{estimated}$ ($p<0.05$, n=86). This marked difference is also reflected in the comparison of the autopsy means for PMI_{actual} (3 days ± 2) and the $PMI_{estimated}$ (7 days ± 11) (n=86).

Indoor cases also had a significant overestimation of the autopsy PMI only ($p<0.05$, n=67) whereas outdoor cases presented no significant differences between PMI_{actual} the $PMI_{estimated}$ for both the scene and autopsy ($p>0.05$, n=19) (results not shown).

The scene PMI was accurately predicted in 30% of the 86 cases (n=26), which increased to 44% of cases when including an error of ±1 PMI day (n=38). Conversely, the autopsy PMI was accurately predicted in 19% cases (n=16) and conferred a 38% accuracy rate when also predicting the autopsy PMI within ± 1 day (n=33). The accuracy of predictions was confined between 1 to 6 days for scene PMI estimations and 1 to 4 days for autopsy PMI estimations. At PMIs beyond this, PMI predictions had reduced accuracy with an error greater than ± 2 days in 56% of scene estimations (n=48) and in 62% of autopsy predictions (n=53).

Decomposition Markers and TBSA

Of the 28 decomposition markers that were independently assessed, 86% (n=24) were subject to variation between the scene and autopsy in terms of the frequency of bodies that expressed them. Facial bloating (n=19), oviposition (n=12), and the cadaver decomposition island (an exclusive scene characteristic) (n=2) were the only 3 decomposition markers that remained unchanged between the scene and autopsy (n=33). Figure 9 displays only those decomposition markers that were *significantly* different in their presence (or absence) on bodies between the scene and autopsy.

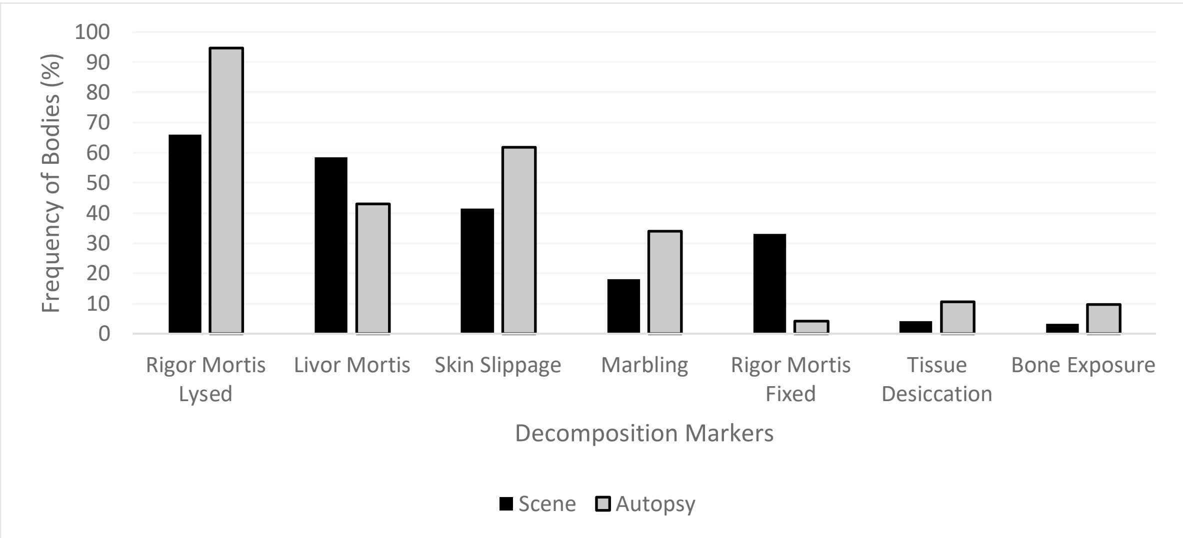

Fig. 9 Key Decomposition Markers Subject to Change between the Scene and Autopsy (n=94)

Rigor mortis was fixed at the scene in 31 bodies and reduced to only 4 bodies at autopsy (Figure 9). The remaining 27 bodies progressed to 'rigor mortis lysed', coupled with a further 62 bodies where 'rigor mortis lysed' remained unchanged from the scene. Similarly, there was a significant decrease in the presence of 'adult blow flies', reducing from 10 to 2 cases at autopsy ($p<0.05$). Conversely, there was a significant increase in the number of bodies presenting 'skin slippage' (n=19) and 'marbling' (n=15) at autopsy ($p<0.05$). Interestingly, the additional bodies exhibiting advanced decay markers of 'tissue desiccation' (n=7) and 'bone exposure' (n=7) all had an MTL >18 hours. Conversely, significantly fewer bodies displayed 'livor mortis' at autopsy (n=43) compared to the scene (n=55) ($p < 0.05$) (Figure 9).

In total, 66% of bodies at the scene exhibited TBSA-scoring decomposition markers (n=62), which increased to 87% of bodies at autopsy (n=82). A TBSA was calculated for each of the 10 gradable markers of decomposition to further quantify their extent of coverage on the human body (Figure 10).

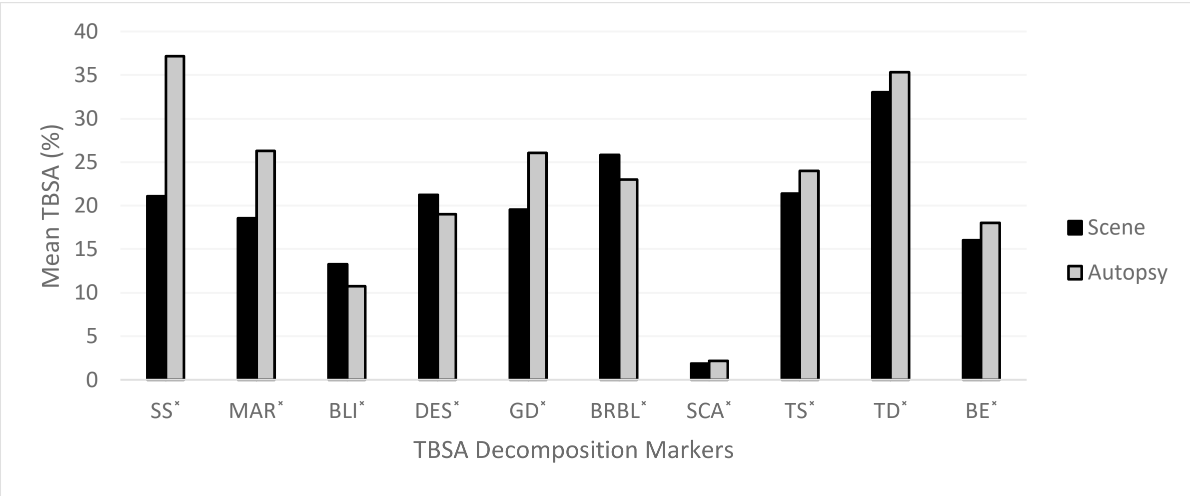

Fig. 10 Mean TBSA Scores for the scene (n=62) and autopsy (n=82) by select decomposition markers. Key of decomposition markers from left to right: SS = Skin Slippage, MAR = Marbling, BLI = Blistering, DES = Desiccation, GD = Green Discoloration, BRBL = Brown/Black Discoloration, SCA = Scavenging, TS = Tissue Sloughing, TD = Tissue Desiccation, BE = Bone Exposure

'Skin slippage' showed significantly increased expression on the body from the scene to autopsy, with a mean TBSA of 21% (n=42) increasing to 37% (n=57), respectively ($p < 0.05$) (Figure 10). Similarly, the mean scene TBSA for both 'green discoloration' (20%, n=18) and 'marbling' (19%, n=17), increased at autopsy by 6% (n=33) and 7% (n=35), respectively. Advanced decomposition markers such as: 'tissue sloughing' (n=9), 'tissue desiccation' (n=10), and 'bone exposure' (n=9), had an increased TBSA between 2 to 3% at autopsy, albeit this increase was insignificant ($p > 0.05$). The mean TBSA between the scene and autopsy of 'blistering' (13%, n=17 vs 11%, n=20), 'desiccation' (21%, n=33 vs 19%, n=34), and 'brown/black discoloration' (26% n=34 vs 23% n=39) found these markers had a *reduced* expression at autopsy, albeit this decrease was insignificant ($p > 0.05$).

Discussion

The first research question investigated if decomposition variability was present between the scene and autopsy photographs. The results showed that both decomposition accumulation (measured by TDS) and the presentation of individual decomposition markers (measured by TBSA) were subject to significant acceleration between body recovery at the scene and the subsequent autopsy examination, in what was termed the 'mortuary time lag' (MTL). Importantly, there was no relationship between the duration of the MTL and TDS, as decomposition acceleration effectively occurred during a wide range of MTLs from 3 hours to 44 hours, where bodies were stored in a mortuary refrigerator at 4°C. Given that decomposition processes are reported to slow down in colder temperatures, and even cease, between 0-4°C [38, 39] it would follow that all bodies should present a similar level of decay, or only minimal changes, between the scene and autopsy, regardless of the MTL duration. However, our results suggest that mortuary storage may not always delay decomposition.

The second research question assessed if decomposition variability between the scene and autopsy would affect the accuracy of PMI estimations. The scene TDS was more accurate at estimating the scene PMI whereas the autopsy PMI was significantly overestimated as result of the increased TDS at autopsy. This research confers important application when forensic experts are using the decomposition apparent in autopsy photographs to inform their PMI estimations and assists in validating photographs as a proxy for real-time decomposition assessments.

Recording Decomposition

Gelderman et al. [12] TDS method proved valuable in quantifying the overall level of accumulated decomposition between the scene and autopsy photographs. It is arguably an improvement on Megyesi et al. TBS method [21] by removing the rigid sequential ordering of co-associated decomposition markers that restrict the progressive scoring of decomposition when all stage-specific criteria have not been met [12]. For example, in the TDS method, the LDS criteria 3.1 is described as 'skin blisters and/or skin slippage and/or marbling'. These options facilitate a true reflection of the decomposition state and provide greater flexibility in accounting for differential decomposition.

The recording of the presence or absence of 28 individual decomposition markers has previously been recommended to assist in removing some of the subjectivity terminology embedded in decomposition scoring models, such as 'some bloating to the neck' [21, 28]. The additional application of the TBSA method to 10 of these decomposition markers meant that it was possible to quantify the extent to which certain decomposition markers displayed as a percentage of the body's total surface area. In doing so, it enabled further discrimination of decomposition marker expression and recognised that two bodies displaying 'skin slippage' and 'marbling' may do so to varying degrees that are not currently accounted for in current scoring models [12, 21, 23].

Scene and Autopsy Decomposition

Decomposition significantly progressed over the mortuary time lag, as measured by TDS and the individual decomposition markers. The increased autopsy TDS correlated strongest with the early decomposition stage (TDS 6). This is reflected by markers such as livor mortis and rigor mortis, which are widely reported to have a rapid onset and limited duration [20, 28, 40]. The lysation of rigor mortis typically occurs between 24 to 36 hours [41] and livor mortis dissipates after 40 hours from onset [42]. While it was not possible to observe the onset of these decay markers due to the retrospective nature of this study, the before (scene) and after (autopsy) 'snapshots' meant that the resolution of rigor mortis and livor mortis had occurred at some time during the mortuary time lag. Given that bodies were stored at 4°C, these findings initially contest the consensus that low storage temperatures prolong the duration of these early post-mortem indicators [40, 42]. However, entomological research has previously found differences in mean temperatures between the inside of the body bag (13.71°C) and the mortuary cooler (5.51°C) [15]. In this study, we also cannot exclude that flucations in the 4°C mortuary refrigeration temperature (caused by personell access and egress to the mortuary refrigeratior) could have contributed to decomposition variability between the scene and autopsy. It is therefore recommended that any further study should take temperature measurements inside and outside of the body bag at regular intervals, to confirm the minimum temperature sufficient for decomposition progression.

'Marbling' and 'skin slippage', which usually occur between 1 to 5 PMI days [20] were also identified as early decomposition markers that were subject to significant progression during the mortuary time lag. 'Skin slippage' is preceded by fluid-filled vesicles, or blisters, that form on the underlying dermis and further accelerate epidermal slippage upon their eruption [40]. The frequency of bodies displaying 'blistering' was greater at the scene than the autopsy as recorded by the TBSA method. It is possible that the movement of the body into, and out, of the body bag during the mortuary time lag, burst the blisters (thus, accounting for their reduction at autopsy) and subsequently promoted further skin slippage. Conversely, 'marbling', induced by proliferation of bacteria in superficial veins, is indicative of putrefactive changes that occurred during mortuary refrigeration. While this disputes research that putrefactive decomposition processes are inhibited at 4°C [38] our finding supports research showing evidence of progressive abdominal gas formation, the hallmark of putrefaction, during mortuary refrigeration [19].

The onset and development of both 'skin slippage' and 'marbling' are considered to be nonspecific to temperature conditions and have previously been found to occur in both extreme cold and heat-related deaths [23, 43]. It is therefore unlikely that mortuary refrigeration inhibited the development of these markers. Larval development was also not hindered by mortuary refrigeration at 4°C in this study. This is consistent with previous findings that larve can continue developing during morgue storage at low temperatures [16, 17]. Our finding of increased prescence of adult blow flies at the scene also supports research that encourages forensic entomologists to attend the scene for a greater yield of entomological evidence than subequent autopsy collection offers [10].

The progression of individual decomposition markers at autopsy was not limited to the early decomposition stage. While bodies in the advanced, mummification and skeletonization stages presented no significant acceleration in TDS between the scene and autopsy, the latter two decomposition stages were under-represented in our dataset. It is recommended that inclusion of more advanced and skeletonization stages, with longer PMI durations, are needed to corroborate these results. The TBSA method identified increased frequencies of 'tissue desiccation' and 'bone exposure' at mortuary time lags of >18 hours. Studies show that these advanced decomposition markers are associated with longer PMIs and can stablise the decomposition process by persisting for 2 to 9 months under dry, cold conditions [20, 25]. It is therefore possible that the environmental transition of the body to a cold and moisture-deficient mortuary storage, coupled with the longer mortuary time lag duration where the internal body temperature would take time to stabilise with the surrounding mortuary storage temperature, could have triggered the onset of 'tissue desiccation' and 'bone exposure'.

In this study, bodies were exposed to an extensive range of mortuary time lags which did not appear to correlate with the TDS increase. The greatest acceleration in TDS was observed between 3 to 20 hours of mortuary time lag and TDS increases occurred up to 44 hours in cold storage. However, it is notable that in 36% of cases the accumulated decomposition remained stable between the scene and autopsy, even when exposed to the same intrinsic, scene and environmental variables in cases where the TDS changed. Consequently, it was not possible to identify which variables had the greatest impact on TDS change. While the mean TDS change was compared between each variable, this resulted in unevenly distributed and small sub-groups. Furthermore, it is recognised that comparison of single variables does not account for the complexity of the decomposition process [1, 4] and it is more likely that a combination of variables will affect the TDS change. A large sample sizes is therefore needed to facilitate further sub-grouping of taphonomic variables to assess their inter-relationships on decomposition progression over the mortuary time lag.

Scene and Autopsy Photographs

The increase in autopsy decomposition has thusfar been predominantly explained by changes related to the decomposition processes. We cannot conclusively exclude the possibility that clothing restricted the identification of some decomposition markers (e.g., small areas of brown/black discoloration) from the scene photographs that were not detailed in the supporting medico-legal death investigator notes, and that subsquently became observable in the autopsy photographs when the clothing was fully removed. However, our findings showed little variation in the TDS change between unclothed and full/partial/covered bodies when clothing was removed at autopsy.

Decomposition markers in the TDS model relating to subjective colour (e.g., grey to green discoloration) and touch (e.g., rigor mortis, 'leathery texture' to mark desiccation) proved challenging to identify from both scene and autopsy photographs [11, 21, 31]. Cross-referencing our decomposition observations to the death investigator and forensic pathology reports, which accurately documented these post-mortem changes, facilitated confirmation of these decomposition markers that could not be readily determined from the photographs. It is therefore recommended that when in-situ scene and/or autopsy attendance is not possible, the forensic expert should also seek corroboration of these decomposition markers from such reports.

While autopsy photographs arguably confer higher quality due to better lighting conditions, anatomically sequenced shots and a clinically clean environment; the scene photographs in this study, were correctly exposed and of comparable quality. It is protocol for medico-legal death investigators to produce multiple high-quality images and to take additional photographs after removal of objects or items that interfere with photographic documentation of the body [34]. Scene photographs also offer important contextual information regarding the body position *in-situ* and it's relationship to the surrounding environment. It is recommended that when scene and/or autopsy attendance is not possible, experts should use both scene and autopsy photographs to observe and compare decomposition changes, provided the photographs confer high quality (e.g., correct exposure, focus, and good composition of overview, mid-range and closeups [34]).

PMI Estimation

The increase in TDS was predominantly consistent with 1 to 8 PMI days and can partly be explained by the existence of 'early decomposition' markers subject to rapid changes in expression [28]. Beyond this, there was no real pattern in TDS changes by PMI duration. The autopsy TDS increased across a number of PMI days and up to 68 days. This was supported by the absence of a linear relationship between TDS and PMI. One possible explanation for this is the dominance of 'early' decomposition cases in the overall sample which were recorded across extensive PMI ranges (2 to 67 days). Similarly, 'advanced' decomposition cases were observed as early as 2 PMI days and were associated with high scene temperatures. The original researchers of the TDS model aired caution on inaccurate PMI estimations when cases presented a combination of low TDS at long PMIs, and high TDS at shorter PMIs [12]. In our study, such combinations also did not support the linearity of the TDS model of

PMI prediction. However, cases beyond 8 PMI days were underrepresented and a larger sample size at longer PMI days is needed to confirm these findings.

TDS could predict estimated ADD with high confidence when the ADD was log-transformed, a finding that is consistent with previous research [12, 28]. However, PMI predictions were significantly over-estimated at autopsy, which most likely resulted from the increased autopsy TDS that was inputted into the ADD equation. Significant deviations between the actual and estimated ADD may be a further contributory factor of inaccurate PMI estimations, which corroborates previous research [1, 11, 16, 28]. Scene PMI was accurately predicted within 1 PMI day in 44% of cases, whereas autopsy PMI was accurately predicted within 1 PMI day in 38% of cases. Furthermore, no cases conferred accurate PMI prediction beyond 6 PMI days. The increased accuracy of scene PMI estimations could be attributed to the lower scene TDS values assigned between 1 to 8 PMI days that supported the linearity of the TDS model.

Interestingly, outdoor decomposition cases had the strongest relationship with estimated ADD and produced no significant differences in PMI estimations, when compared to indoor cases. It is possible that the daily averages in outdoor temperatures recorded from local weather stations were a more accurate reflection of the scene environment, compared to the single thermostat recordings of indoor cases. Due to the retrospective nature of this study, it was not possible to obtain daily scene temperatures from indoor cases. A single temperature reading was recorded at the scene upon body discovery and summed for each retrospective PMI day, assuming that the scene temperature remained unchanged. Consequently, these temperature estimations may not have been accurate, given that additional variables such as open windows or controlled heating timings could have caused daily temperature fluctuations [21, 44].

Temperature appeared non-influential to the overall decomposition change, which is consistent with previous research [1, 28]. Effectively a wide range of scene temperatures were reported for cases where the TDS changed, and where it remained stable, over the mortuary time lag. Interestingly, the TDS increased in four bodies that were exposed to the -5°C to 0°C scene temperature range. These cases were associated with various mortuary time lags of $>6 - \leq 12$ (n=1), $>18 - \leq 24$ (n=2) and $>24 - \leq 48$ (n=1) which suggests that, even at low scene temperatures, the duration of cold storage does not influence the rate of internal decomposition progression. However, heat-induced micro-environments may be generated from insect metabolism on the decomposing body, which could have produced deviations from the recorded ambient temperature, regardless of the indoor or outdoor setting [16]. In addition, depending on the scene location, bodies were likely subject to variable durations of transit times to the mortuary, so it also cannot be excluded that bodies had different initial temperatures upon entering mortuary storage.

Conclusion

In this study, the accumulation of decomposition and associated markers, were subject to significant acceleration between the scene recovery and subsequent autopsy, suggesting that mortuary refrigeration does not always delay decomposition. The progression of decomposition predominantly occurred during shorter PMI durations and was associated with the early stage of decomposition. Consequently, the autopsy PMI was significantly overestimated using the TDS model. While the TBSA method was not used to estimate the PMI, it sought to decipher the co-association of stage-specific decomposition criteria by identifying key decomposition markers that were subject to statistical change between the scene and autopsy examination. Further research is needed to confirm the causes of these decomposition variable changes. This could include identifying which decomposition markers are temperature-dependent for expression and ascertaining any temperature discrepancies between the interior body bag and the external mortuary refrigeration environment. This study confers important application to forensic experts who may rely on the scene and/or autopsy photographs of the decomposing body to inform their estimation of the PMI. It follows that for best practice, both scene and autopsy photographs should be used in cases where an *in-situ* examination of the body is not possible. If only autopsy photographs are available and the PMI is unknown, the forensic expert should consider that the decomposition observed at autopsy may not correspond to the scene decomposition to avoid any misinterpretation of the PMI.

Declarations

Funding

No funding was received for conducting this study.

Conflicts of interest/Competing interests

The authors have no conflicts of interest to declare that are relevant to the content of this article.

Availability of data and material

The datasets generated and analysed during the current study are available from the corresponding author on reasonable request.

Code availability

Not applicable.

Consent to participate

For this type of study, formal consent was not required.

Consent for publication

For this type of study, consent for publication was not required.

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Decomposition variability between the scene and autopsy examination and implications for post-mortem interval estimations

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