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Inorganic elemental analysis of decomposition fluids of an *in situ* animal burial

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ABSTRACT

In homicide investigations, it is critically important that post-mortem (PMI) and post-burial interval (PBI) of buried victims are determined accurately. However, clandestine graves can be difficult to locate; and the detection rates for a variety of search methods can be very low. This paper presents elemental analysis results of 18 months of decomposition fluids from an *in situ* buried animal cadaver used as a human clandestine burial proxy. Study results showed potassium, sulphate and sodium are key detectable elements which mirror observed conductivity temporal changes from this and other studies. Seasonal rainfall has a strong influence on both fluid generation and subsequent concentration which needs to be accounted for. Study implications suggest inorganic elements could provide both detection and potential dating of discovered clandestine burials.

1. Introduction

Geoscientific methods are being increasingly utilized by forensic search teams for the detection and location of clandestine burials [1–3]. Clandestine graves of murder victims are usually shallow, less than 3 m and typically 0.5 m below ground level (bgl) [4,5], but current detection rates are low and, without locating the victim's body, obtaining a successful conviction is more difficult [6,7]. Search investigators will typically use a variety of methods, which include scenario-based, feature-focused, intelligence-led, and systematic standard operating procedures [6,7]. Standard operating procedures require investigators to follow sequential workflows, from reviewing case information, sourcing background/intelligence information, and remote data analysis. This process occurs before determining search strategies, undergoing site reconnaissance and phased site investigations, and then intrusively investigating anomalous areas [2,6,8]. Geoscientific site investigation methods vary depending upon the specific case, search site, and numerous other factors that are reviewed elsewhere [2,3], but can include scent-trained human remains detection dogs [8,9], forensic geomorphology [10], forensic botany [11,12] and entomology [13,14], near-surface geophysics [15–23], intrusive probing [24,25] and soil geoscience analysis [26–28].

After a body has been found, it is natural for investigators to focus on determining time since death. There has been extensive research on estimating the postmortem interval (PMI) of very recently deceased individuals discovered aboveground that has been reviewed elsewhere [28], commonly using body cadaver temperatures [29,30], entomology [31], entomofauna [32] and thanatochemistry [33]. For longer deceased individuals, other common PMI dating methods include tissue decomposition [34], skeletal remains [35] and tooth odontology [36]. Below ground decomposition rates of discovered individuals have been shown to be highly variable [37], depending on organic content [38], various local environmental factors such as soil type [26,39–41] and organism accessibility [42], among other factors. These factors complicate the estimation of PMI for buried remains. Furthermore, it may be useful to estimate the post-burial interval (PBI) as a guide to the PMI. However, the PMI and PBI may be different: A victim might not be buried immediately after death. In such cases, the PBI can be used as an estimate of the lower limit of the PMI. The presence of a decomposing cadaver has also been shown to be detectable on the surrounding soil. For example, changes in soil chemistry [38,43,44], such as changes in the levels of methane [45], phosphates and nitrates [46], ninhydrin-reactive nitrogen [47,48], volatile organic compounds [27,38,43] and pH [46,49] can all be detected. Changes in these soil properties can be used to estimate time since death. The decay of other items such as materials associated with a grave has also been suggested to allow a PBI to be estimated [34,41]. Although relatively poorly understood, grave soil has been shown to be detectable by

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near-surface geophysical search methods, specifically electrical resistivity [19,21,50,51], and its reciprocal, bulk ground conductivity [16]. Geophysical research using simulated clandestine grave burials can provide critical information, for example, on optimal geophysical detection methods and equipment configurations [15,28,52–54], as well as providing continuous datasets for comparison with real cases [54–60]. Recent research has found that electrical resistivity anomalies over burials are predominantly due to conductive fluids in grave soil that vary temporally that may be due to decomposition [28,54,61]. It has been shown that it is possible to repeatedly extract *in situ* decomposition fluids from both a buried pig cadaver and background soil water, without the need for repeated disturbance or numerous replicate samples as other authors have performed. The resulting fluids can be simply analyzed for conductivity using a handheld meter [28]. However, it is uncertain what elements are causing these observed temporal changes in conductivity, hence the focus of this repeat study.

The aims of this study are to therefore; *firstly* to sample and present results from an 18 month monitoring study of a burial pig carcass, analysing the leachate and background soil water for the major inorganic chemical components and *secondly*, perform a systematic statistical analysis of the resulting element parameters to determine the contributions from individual inorganic elements responsible.

2. Methodology

2.1. Study test site

The selected burial site was located in a restricted area on Keele University campus, approximately 200 m above sea level, close to Newcastle-under-Lyme town in Staffordshire, UK (Fig. 1). Part of the study site was initially used for simulated clandestine graves monitoring and geophysical investigations (see Refs. [54,55,61]). Daily climatic records were obtained from a nearby weather observation station within Keele University, with a temperate local climate that is typical for the UK [62]. The study site was a small plot of land approximately 25 m by 20 m, covered with grasses and surrounded by deciduous trees on three sides. The study site scenario is a typical representative of a semi-rural environment.

Information from a nearby borehole records identified the Carboniferous (Westphalian) Butterton Sandstone bedrock geology approximately 2.6 m below ground level (bgl). The local soil is predominantly a made-ground, due to the presence of demolished greenhouses. Initial soil sampling showed a vertical site succession of a shallow (0.01 m) organic-rich, top soil (Munsell colour chart colour (mccc): 5 YR/2/2.5), with underlying ‘A’ Horizon (Mccc: 5 YR/3/3) comprising largely of a natural sandy loam that contained approximately 5% of isolated brick and coal fragments [55]. The natural ground ‘B’ Horizon was located at approximately 0.45 m bgl, consisted mainly sandstone fragments from the underlying bedrock, which suggested a shallower bedrock depth. Also further investigation on soil particle analysis shown that the typical sandy loam soil texture, i.e. approximately 72% sand, 26% silt and 2% clay, in combination with slow lateral water flow led to moderately high moistness of the soil approximately 34%.

2.2. Experimental procedures

The simulated grave was constructed at the eastern part of the site (Fig. 1), which involved removal of the turf and then ground excavation, of a hole up to ~0.6 m deep, ~1.5 m length and ~0.5 m wide. The use of human cadavers was prevented due to the ethical issues involved in the use of human cadavers for research in the UK (Human Tissue Act, 2004); therefore a pig cadaver of the species *Sus scrofa* was used as a proxy for a human cadaver, whilst not ideal, they are considered to be similar to humans in weight, fat to muscle ratio, hair coverage, biochemistry and physiology [15,26,63]. A 90 kg pig of length ~1.5 m, sourced from a local abattoir, was therefore killed by bolt gun; this prevented excess blood being lost as they usually despatched by electrocution and draining. The pig carcass was interred on 18th March 2014 and a lysimeter model 1900 inserted inside the made-slurry at the base of the hole between the two hind limbs and the grave wall (see Fig. 2). After internment of the pig carcass, the grave was backfilled with the same excavated soil, tamped firmly and leaving a slight mound to account for later settlement, before the turf was replaced to simulate a clandestine grave. A control lysimeter was installed on the same day, approximately 16 m away from the pig grave and uphill to prevent any potential contamination (Fig. 2d). For this control lysimeter, a hole of ~0.3 m by ~0.3 m wide and 0.6 m deep (the same depth as the pig grave) was excavated and refilled. The lysimeters were then left in place throughout the monitoring period. Generating a suction pressure within the lysimeter causes soil water to be drawn through the ceramic cup and into the PVC tube. Leachate and soil samples can then be extracted using a plastic syringe with a narrow tube attachment inserted through the stopper assembly (Fig. 2e).

2.3. Sample collection and on-site measurements

Initial sample extraction was conducted two days before the sampling day, to enable a fresh accumulation of leachate fluid in the grave which should be representative during the sampling period. The lysimeter clamp ring used to fold the neoprene tubing was removed, giving access to extract any fluid present in the grave, before a vacuum hand pump was employed to generate a vacuum pressure of approximately 65 centibars (kPa) (Fig. 2e). This pressure is capable of causing moisture to move from the soil through the porous ceramic cup, and into the vacuum sampler [64]. The same extraction procedure was repeated on each sampling day. Samples were extracted from both the pig grave and the control once a month for a period of 18 months, except for the first month that was sampled fortnightly, to enhance and validate the initial soil and leachate conditions. The samples were transferred to 100 ml labelled plastic sample bottles (Fig. 2e) after a portable WTW^{TM} Instrument Multi-line P4 conductivity meter was used on-site to measure conductivity and temperature values (Fig. 2e). These were automatically corrected by the conductivity meter to a reference temperature (25 °C) and are 0.1 °C accurate, thus avoiding any potential temperature variation effects when collecting samples. This procedure was repeated to check reading repeatability and reliability. Samples pH was also measured onsite in the laboratory with standards at pH 4, 7 and 10 before each use. Approximately 10 ml of the collected samples were used for the ICP-OES analysis, which was conducted within 1 h after sampling and the remaining portion kept frozen until further Dionex laboratory analysis was conducted.

2.4. Climatological data collection

Climatological information was obtained from the closest weather station, ~0.2 km from the test site managed by the U.K. Meteorological Office. The recorded data include average daily rainfall and air temperature reading over the corresponding monitoring periods (see Table 1). It measured monthly minimum, maximum, and average total rainfall of 15 mm, 113 mm, and 66 mm, respectively, over the 549 day monitoring period. Calculated monthly total rainfall data of the site were used to correct measured soil water measurements for local rainfall variation, in which conductivity values were multiplied by a rainfall correction