Forensic Science International xxx (2011) xxx–xxx

Contents lists available at SciVerse ScienceDirect

Forensic Science International

journal homepage: www.elsevier.com/locate/forsciint

Mapping the lateral extent of human cadaver decomposition with soil chemistry

J.A. Aitkenhead-Peterson [a,*], C.G. Owings [b], M.B. Alexander [a], N. Larison [c], J.A. Bytheway [d]

[a] Department of Soil and Crop Sciences, Texas A&M University, College Station, TX 77843, United States
[b] Department of Entomology, Texas A&M University, College Station, TX 77842, United States
[c] College of Criminal Justice, Sam Houston State University, Huntsville, TX 77341, United States
[d] Southeast Texas Applied Forensic Science Facility, Sam Houston State University, Huntsville, TX 77341, United States

ARTICLE INFO

Article history:
Received 23 November 2010
Received in revised form 4 May 2011
Accepted 17 September 2011
Available online xxx

Keywords:
Cadaver decomposition islands
Soil
DOC
DON
Nutrients
Cations

ABSTRACT

Soil below decomposing cadavers may have a different lateral spatial extent depending upon whether scavengers have access to the human cadaver or not. We examined the lateral spatial extent of decomposition products to a depth of 7 cm of soils beneath two decomposing corpses, one in which the subject was autopsied, unclothed and placed under a wire cage to restrict scavenger access and one in which the subject was not autopsied, unclothed and exposed to scavengers. The two bodies had accumulated degree days (ADD) of 5799 and 5469 and post mortem interval (PMI) of 288 and 248 d, respectively. The spatial extent for dissolved organic carbon (DOC) and organic nitrogen (DON) for both bodies was large but similar suggesting some movement off site for both compounds. Mean DOC was 1087 ± 727 and 1484 ± 1236 $\mu g\ g^{-1}$ dry soil under the two corpses relative to 150 ± 68 $\mu g\ g^{-1}$ in upslope control soils. Sulfate tended to have 'hot spots' of lower values relative to the control soils indicative of anaerobic respiration. pH was lower and electrical conductivity was higher in the soil under both decomposing cadavers relative to control soils. Some of the nutrients examined downslope of the human remains were significantly higher than control soils upslope suggesting movement of decomposition products off-site which could be an important factor when using human remains detector dogs.

1. Introduction

A cadaver decomposition island (CDI) is defined as a highly concentrated island of fertility [1]. Cadaver decomposition islands are those regions in the soil below and around a decomposing corpse. While Carter et al. [1] produced an excellent review on the CDI under various mammals; the spatial extent of the human decomposition islands has not been addressed. Understanding and mapping spatial extent of the CDI is important particularly if water soluble chemistry moves 'off-site' which can complicate location of clandestine graves particularly when human remains detection (HRD) dogs are used. In addition, movement of these products off site or their downward migration may impact local freshwater and groundwater chemistry.

Soil chemistry under decomposing or dry remains of pigs and humans have been used to estimate post mortem interval (PMI) [2–4]. Unfortunately every grave site is different in terms of its soil series which is based upon local environmental factors such as vegetation and its associated soil microbiology, climate, geology, topography or aspect and time for formation [5]. Furthermore, very little research has been conducted on the soil solution chemistry under human remains in different soil orders [2]. The Vass et al. [2] study examined seven un-autopsied, un-embalmed cadavers at a research facility at the University of Tennessee. Soils in the Vass et al. [2] study were a fine, mixed, thermic Typic Paleudalf sampled at a depth of 3–5 cm and samples were taken from beneath the corpse for a period of approximately 2 years. Vass et al. [2] suggested that soil solution volatile fatty acids (VFA), anions and cations could be used to estimate post mortem interval. Much more research has concentrated on volatile organic carbon [6,7] where the major classes of compounds detected were alcohols and aldehydes [6], and cadaver decomposition islands of other mammal species [3,8].

The main objective of this study was to examine the mapped spatial extent of the CDI of two above-ground human grave sites, one accessible to scavengers and un-autopsied and one inaccessible to scavengers and autopsied and compare them to control soils. A secondary objective was to examine soil down slope of grave sites to determine the potential for water soluble movement of chemical constituents off site.

2. Materials and methods

2.1. Site description

Our research was conducted at the Southeast Texas Applied Forensic Science (STAFS) research and training facility in Huntsville, TX, USA. The facility, which opened in 2007, is a human decomposition research facility, located in the Center

* Corresponding author. Tel.: +1 979 845 3682.
E-mail address: jpeterson@ag.tamu.edu (J.A. Aitkenhead-Peterson).

0379-0738/$ – see front matter © 2011 Elsevier Ireland Ltd. All rights reserved.
doi:10.1016/j.forsciint.2011.09.007

for Biological Field Studies at Sam Houston State University. It is adjacent to the Sam Houston State Forest which has a dominant vegetation of loblolly and shortleaf pines. Maximum security fencing surrounds the 1-acre outdoor research facility which is set in a forest clearing with an additional 8 acres (32,000 m^2) of minimum security reserved for other types of forensic training such as search and recovery maneuvers. Soil at the facility is classified as loamy, silicious, semiactive, thermic arenic plinthic paleudalfs and fine, mixed, semiactive, thermic aquic paleudalfs of the Depcor and Huntsburg series. Both soil series are moderately well drained and have slow permeability. The slope within the 1-acre facility is approximately 4%. Soils at the top of the slope are Depcor series with a depth to water table of 60–107 cm. Soils at the bottom of the slope are Huntsburg series with the water table between 15 and 46 cm below the surface. Vegetation within the facility comprises sparsely scattered grass and weeds with some research units having vegetation and some bare soil. Research units of 3.7 m × 3.7 m within the outdoor facility are designated for research studies on human decomposition. At the date of soil collection the facility had been accepting donations for approximately 3 years. Both units used in our study were located on the upper slope of the facility to avoid contamination from water soluble chemical constituents' derived from upslope graves. The units used in our study had not been used for research prior to this study, in other words, the decomposition products presented in this study are from one cadaver in each unit.

Mean temperature and summed precipitation during the study period was 20.1 and 532.4 mm for unit 1 and 22.1 °C and 452.4 mm for unit 2. The accumulated degree days (ADD) [2] and post mortem intervals (PMI) were 5799 ADD and 288 d for unit 1 and 5469 ADD and 248 d for unit 2, representing a difference of 330 ADD and 40 d between the two units.

The subject in unit 1 was autopsied and the organs removed before placement, unclothed and placed under a wire cage to protect from scavengers. The subject in unit 2 was not autopsied and was placed unclothed and un-caged thereby open to scavenger influence. Scavenger activity was generally from small rodents and birds. A bobcat had gained entry into the facility prior to our sampling of the soil. Some vulture activity has been observed but this was minimal. Observed activity from scavengers includes movement of upper and lower limbs, removal of fingers and chewing of bones once the subject has reached the dry remains stage.

2.2. Sample collection and treatment

Upslope control soils (n = 9 samples) were taken at upslope areas of the 1-acre facility. Vegetation here comprised grass and weed growth and there was no potential for contamination of decomposition products. Downslope control soils (n = 6 samples) were taken at downslope areas of the 1-acre facility. These sampling positions had bare soil and no vegetation and a greater potential for receiving runoff contamination from decomposition products. All the control soils (n = 15) collected were >23 m away from units that had housed human remains. Samples were taken to a depth of 15 cm with a 2 cm diameter sureshot auger and each soil core taken was treated as one sample.

Within each of our research units, perimeters 101 cm wide and 254 cm long were set around the two grave sites. The subject assigned to unit 1 was placed on 17th December 2009 and removed on 23rd September 2010. Unit 1 had skeletal remains up to one week prior to our sampling campaign on 30th September 2010. The obvious visual region of decomposition was split into quarters and five random samples were taken from each quarter. Three of these samples were taken from within the decomposition region and two of the samples were taken outside of the decomposition region for each quarter (Figs. 1–3). Sample positions were recorded (Figs. 1–3). The subject assigned to unit 2 was placed on 26th January 2010 and was still in place when the unit was sampled on 30th September 2010. Unit 2 had dry

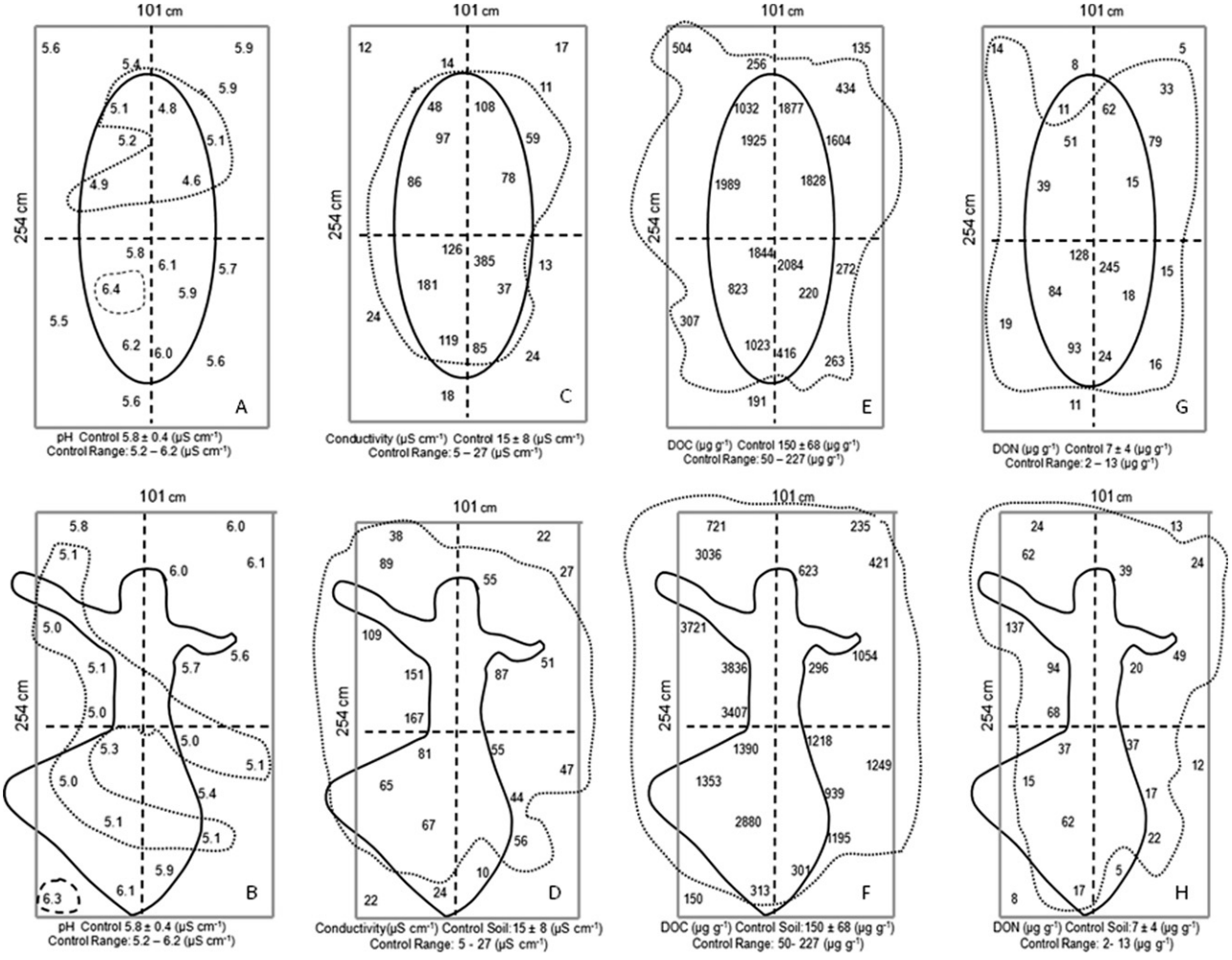

Fig. 1. Mapped human cadaver decomposition islands autopsied and protected from scavengers (upper panel) and open to scavengers (lower panel) for (A) and (B) pH, (C) and (D) electrical conductivity, (E) and (F) DOC and (G) and (H) DON. The head of the remains is positioned at the top for both sets of human remains. The black line outlines the subject and the grey dotted line delineates significantly higher or lower values relative to the range of upslope control samples. Numeric values are the pH, conductivity or mass of constituent µg g^{-1} soil at our sampling positions. Upslope control mean values ± 1 standard deviation and upslope control ranges and units are included at the foot of each individual figure.

Please cite this article in press as: J.A. Aitkenhead-Peterson, et al., Mapping the lateral extent of human cadaver decomposition with soil chemistry, Forensic Sci. Int. (2011), doi:10.1016/j.forsciint.2011.09.007

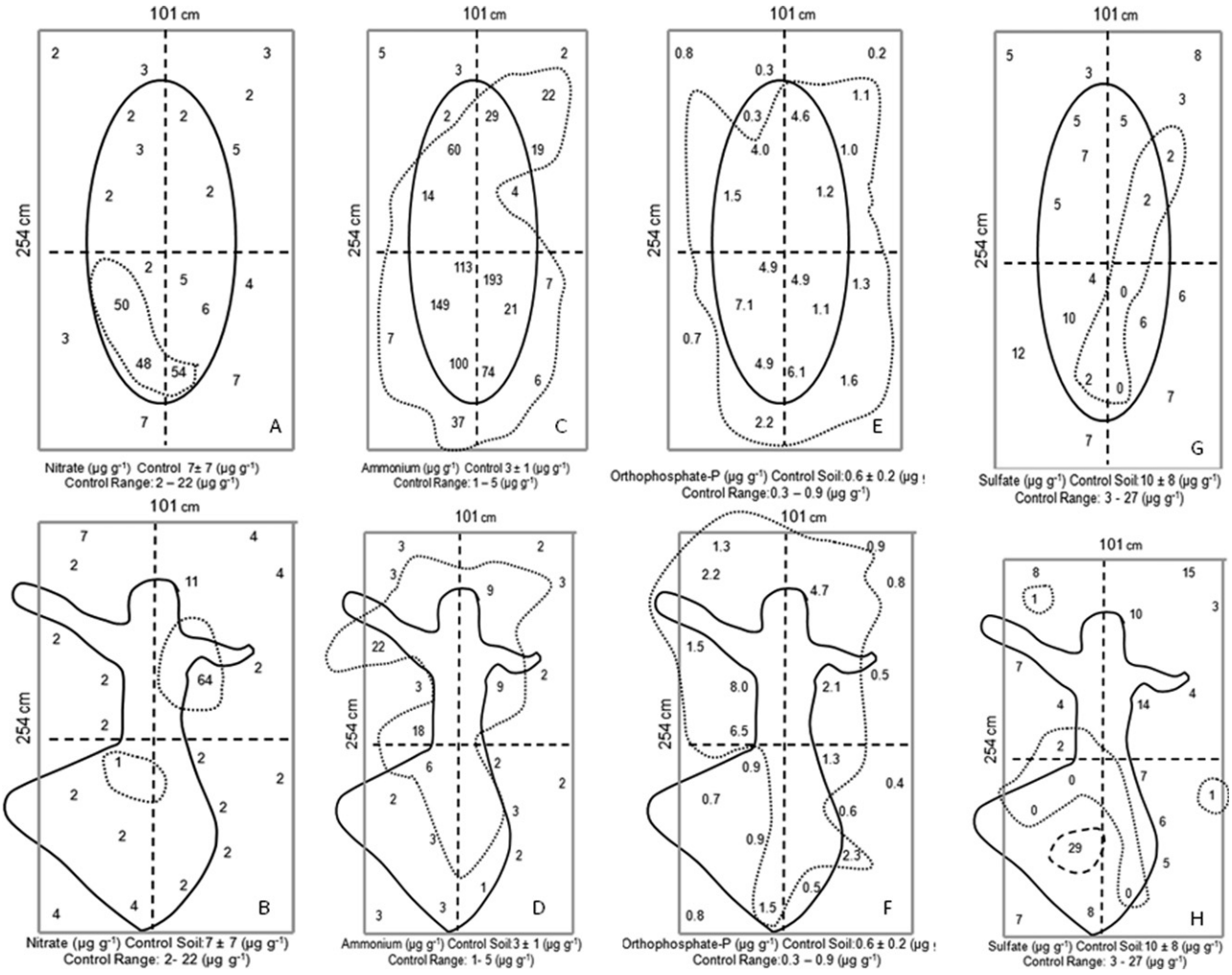

Fig. 2. Mapped human cadaver decomposition islands autopsied and protected from scavengers (upper panel) and open to scavengers (lower panel) for (A) and (B) nitrate-N, (C) and (D) ammonium-N, (E) and (F) orthophosphate-P and (G) and (H) sulfate. The head of the remains is positioned at the top for both sets of human remains. The black line outlines the subject and the grey dotted line delineates significantly higher or lower values relative to the range of upslope control samples. Numeric values are the mass of constituent $\mu g\ g^{-1}$ soil at our sampling positions. Upslope control mean values ± 1 standard deviation and upslope control ranges and units are included at the foot of each individual figure.

skeletal remains. The 101 cm × 254 cm perimeter area was split into quarters with the vertical line at the center of the remains and the horizontal line at the pelvic region (Figs. 1–3). Due to scavenger accessibility the upper and lower limbs were no longer in their original supine placement position. Five random samples were taken from each quarter and the sampled positions relative to the dry remains were recorded (Figs. 1–3). All samples were taken using a 2 cm diameter sureshot auger to a depth of 7.5 cm. Each soil core retrieved was considered a sample and individually bagged. A depth of more than 7.5 cm was not possible at the grave sites due to the formation of an impenetrable, assumed adipocerous layer.

Soils were air dried for one week prior to sieving through a 2 mm sieve to remove stones and root debris. Immediately after sieving an aliquot of 3 g soil from each sample collected was placed into a 50 mL centrifuge tube and 30 g of ultrapure water was added. The soil:solution units were shaken for 22 h at 400 rpm prior to centrifugation at 10,000 × *g*-force for 20 min at room temperature.

The supernatant solution was removed and pH and electrical conductivity were recorded for each solution sample prior to filtering through an ashed Whatman GF/F filter (nominal pore size 0.7 µm) for nutrient analysis and through a 0.25 µm Pall cellulose acetate filter for anion and cation analysis. Samples were analyzed for nutrients immediately after their water extraction and analyzed for anions and cations within 4 weeks of collection.

The procedures used for extraction coupled with air drying and sieving would aerate soil that was likely anaerobic prior to collection. This may have induced bacterial activity but because all control soils and sample soils were treated the same we believe that all chemistries will be relative.

2.3. Sample analysis

Dissolved organic carbon (DOC) and total dissolved nitrogen (TDN) were measured using high temperature Platinum-catalyzed combustion with a Shimadzu TOC-VCSH and Shimadzu total measuring unit TNM-1 (Shimadzu Corp., Houston, TX, USA). Dissolved organic carbon was measured as non-purgeable carbon which entails acidifying the sample (250 µL 2 M HCl) and sparging for 4 min with C-free air. Ammonium-N was analyzed using the phenate hypochlorite method with sodium nitroprusside enhancement [9] and nitrate-N was analyzed using Cd–Cu reduction [10]. Orthophosphate-P was quantified using the ascorbic acid, molybdate blue method. Alkalinity was quantified using the methyl orange method [11]. All colorimetric methods were performed with a Westco Scientific Smartchem Discrete Analyzer (Westco Scientific Instruments Inc., Brookfield, CT, USA). Calcium, magnesium, potassium and sodium were quantified by ion chromatography using an Ionpac CS12A analytical and Ionpac CG12A guard column for separation and 20 mM methanosulfonic acid as eluent at a flow rate of 1 mL min^{-1} and injection volume of 25 µL (DIONEX ICS 1000). Fluoride, chloride, and sulfate were quantified using Ionpak AS20 and Ionpak AG20 analytical and guard columns for separation with 35 mM KOH as eluent at a flow rate of 1 mL min^{-1} and an injection volume of 25 µL (DIONEX ICS 1000; DIONEX Corp., Sunnyvale, CA, USA). Dissolved organic nitrogen was estimated as the product of TDN – (NH_4-N + NO_3-N). NIST traceable and control standards plus replicate samples were run every 10th sample on all analyses to monitor instrument precision and for quality assurance and control.

2.4. Statistical analysis

Mass of nutrients per gram of dry soil was calculated and the spatial distribution was plotted to enable visual observation of the cadaver position and the mapped decomposition islands for each unit (Figs. 1–3). The values for the mapped decomposition islands were determined as any sample nutrient mass that was greater than the highest value in the upslope control soil dataset. Two sample *t*-tests with $\alpha < 0.05$ were run for (a) each nutrient within the mapped decomposition island and (b) each nutrient in the individual units to compare against the upslope control soils. Some gravesoil nutrients did not display masses that were higher than the highest control soil value. Other nutrients displayed

Please cite this article in press as: J.A. Aitkenhead-Peterson, et al., Mapping the lateral extent of human cadaver decomposition with soil chemistry, Forensic Sci. Int. (2011), doi:10.1016/j.forsciint.2011.09.007

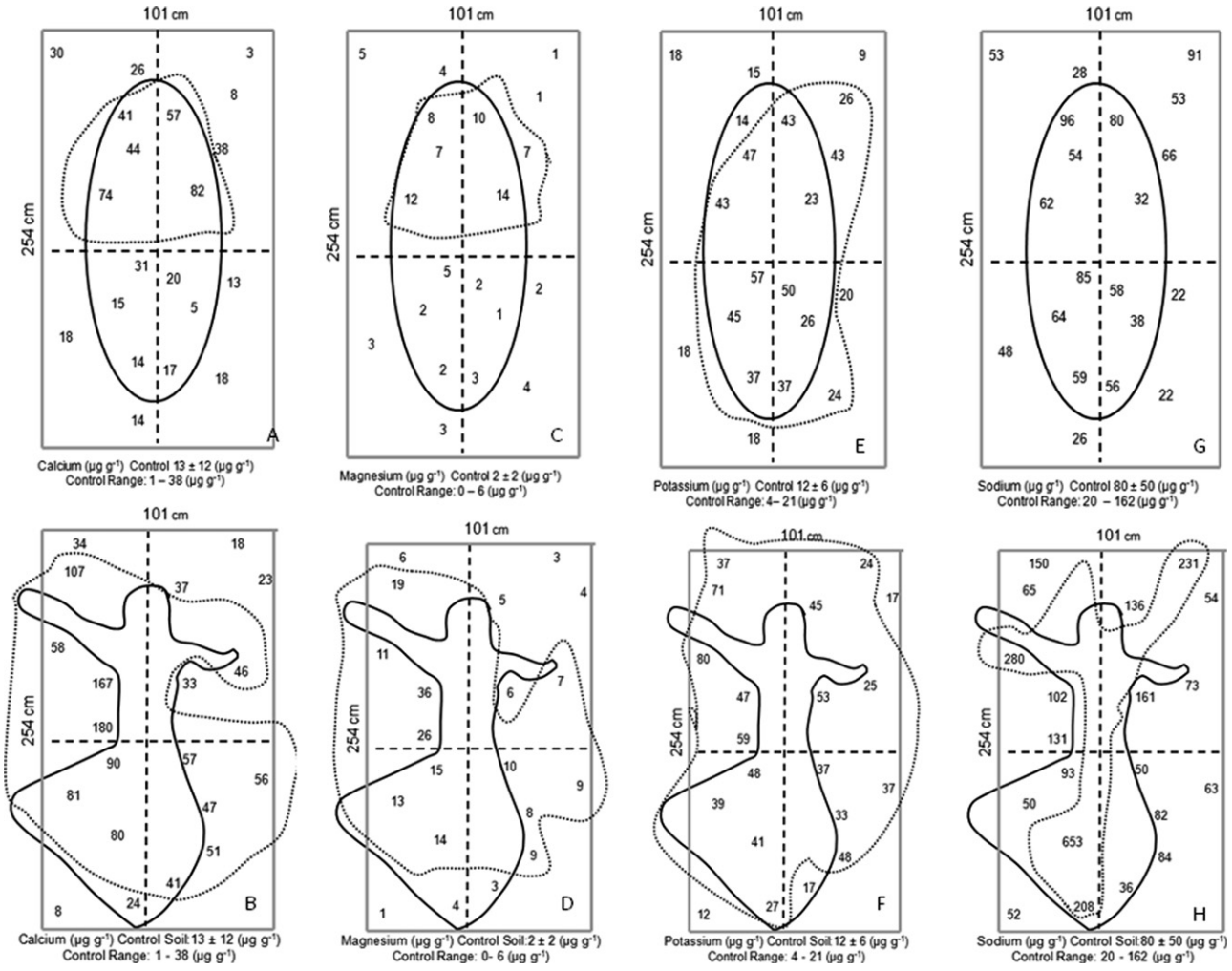

Fig. 3. Mapped human cadaver decomposition islands autopsied and protected from scavengers (upper panel) and open to scavengers (lower panel) for (A) and (B) calcium, (C) and (D), magnesium, (E) and (F) potassium and (G) and (H) sodium. The head of the remains is positioned at the top for both sets of human remains. The black line outlines the subject and the dotted grey line delineates significantly higher values relative to the range of upslope control samples. Numeric values are the mass of constituent $\mu g\ g^{-1}$ soil at our sampling positions. Upslope control mean values ± 1 standard deviation and upslope control ranges and units are included at the foot of each individual figure.

values lower than the lowest control soil value. To test potential runoff from grave sites we applied two sample, one-tailed *t*-tests to the upslope control and downslope control data.

Regression analysis was used with electrical conductivity as the independent variable with all chemistries analyzed in turn for all gravesoils in an effort to produce a simple empirical predictive model for gravesoil.

3. Results

We observed evidence of water soluble decomposition products moving off site for pH ($p = 0.008$), conductivity ($p = 0.02$), DOC ($p = 0.02$), orthophosphate-P ($p = 0.001$), ammonium-N ($p = 0.03$), DON ($p = 0.009$) and potassium ($p = 0.01$). The higher significance for DON, orthophosphate-P and potassium moving off site suggests more research should be devoted to this phenomenon.

3.1. Cadaver decomposition islands

The lateral spread of the CDI was mapped for each of the grave sites within each unit for each nutrient analyzed to determine the extent that the soil around the grave site was affected (Figs. 1–3). One of the most interesting findings was that the mapped CDI differed considerably depending upon which nutrient was plotted and whether the subject was open to scavenger activity or had been autopsied. The effect of scavenger activity was evident by relocation of limbs relative to their original placement (Figs. 1–3) likely by the bobcat and some vulture activity which likely extended the CDI.

3.2. Soil pH and conductivity

pH in the mapped CDI averaged 4.9 ± 0.2 ($n = 5$) in unit 1 and 5.1 ± 0.1 ($n = 9$) in unit 2 which were both significantly lower than the control soil pH which averaged 5.8 ± 0.4 ($p < 0.001$; Table 1). There was no significant difference in pH between units 1 and 2 ($p = 0.27$). There was a significant difference in pH between the two mapped CDI's ($p = 0.01$). Our mapped CDI for pH tended to be limited to the upper torso in unit 1 but was spread across the area of unit 2 (Fig. 1A and B). Conductivity in the control soils averaged 15 ± 8 $\mu S\ cm^{-1}$ and ranged from 5 to 27 $\mu S\ cm^{-1}$ (Table 1). The mapped CDI conductivity in units 1and 2 averaged 118 ± 93 $\mu S\ cm^{-1}$ ($n = 12$) and 77 ± 38 $\mu S\ cm^{-1}$ ($n = 15$) respectively. The mapped CDI conductivity in units 1 and 2 were both significantly higher than the control soils ($p < 0.001$) but there was no significant difference in conductivity between the two units ($p = 0.14$). We observed a much larger spread of mapped conductivity in unit 2 relative to unit 1 based on our expected decomposition island (Fig. 1C and D).

Please cite this article in press as: J.A. Aitkenhead-Peterson, et al., Mapping the lateral extent of human cadaver decomposition with soil chemistry, Forensic Sci. Int. (2011), doi:10.1016/j.forsciint.2011.09.007

Table 1
Mean, standard deviation and ranges of chemical constituents of the upslope and downslope control soils, mapped decomposition islands and the two units for this study. unit 1 comprised an autopsied subject who was placed unclothed and caged to protect from scavenger activity and unit 2 comprised an un-autopsied subject placed unclothed and subjected to scavenger activity. Italicized *n* values are the number of samples collected within each unit that fell above or below the range observed for the upslope controls and represent the number of samples within the unit that were allocated as our proposed CDI mean values. Values for *n* under the samples column indicate the number of upslope, downslope samples taken within the facility and the number of samples taken within each unit. Differences in superscript letters are significant differences in mean chemical constituent between the mean upslope control and mean for each unit or downslope control samples or significant differences between the two CDI samples at $\alpha < 0.05$.

Samples		pH	EC (μS cm^{-1})	NH_4-N	NO_3-N	DOC	DON	PO_4-P	SO_4^{2-}	Ca^{2+}	Mg^{2+}	K^+	Na^+	Cl^-
				μg g soil^{-1}										
Control	Mean	5.8^a	15^a	3^a	7^a	150^a	7^a	0.6^a	10^a	13^a	2^a	12^a	80^{ab}	50^a
Upslope	Std Dev	0.4	8	1	7	68	4	0.2	8	12	2	6	50	34
n = 9	Range	5.2–6.2	5–27	1–5	2–22	50–227	2–13	0.3–0.9	3–27	1–38	0–6	4–21	20–162	12–127
Control	Mean	6.3^b	28^b	4^b	14^a	215^b	12^b	2^b	11^a	20^a	4^a	28^b	115^a	47^a
Downslope	Std Dev	0.2	11	1	13	17	2	1	4	12	2	18	50	23
n = 6	Range	6.0–6.5	15–40	3–6	2–32	190–235	9–14	1–3	7–18	6–36	1–7	19–64	53–189	17–83
Unit 1	Unit mean	5.6^a	77^c	42^c	10^a	952^c	48^c	2.5^d	5^b	28^a	5^a	31^c	55^a	33^a
n = 20	Std Dev	0.5	87	56	17	745	58	2.2	3	22	4	14	22	11
5799 ADD	CDI Mean	4.9^d	118^c	52^c	49^b	1087^c	58^b	3.2^d	0^c	59^{bc}	10^c	38^c	–	–
Autopsied	Std Dev	0.2	93	58	5	727	61	2.1	0	18	3	11	–	–
No Scavenger Access	*n*	*5*	*12*	*16*	*3*	*17*	*16*	*15*	*4*	*5*	*6*	*13*	*0*	*0*
Unit 2	Unit mean	5.5^a	63^c	5^a	6^a	1417^c	38^c	1.9^{abd}	7^{ab}	61^c	10^b	40^c	140^b	69^a
n = 20	Std Dev	0.4	41	6	14	1240	33	2.1	7	46	9	17	137	55
5469 ADD	CDI Mean	5.1^c	77^c	13^c	64^b	1484^c	45^b	3.1^d	1^c	85^c	15^{bc}	44^c	343^c	272^b
Not Autopsied	Std Dev	0.0	38	7	0	1236	33	2.4	1	46	9	15	208	0
Scavenger Access	*n*	*9*	*15*	*5*	*1*	*19*	*16*	*10*	*6*	*12*	*12*	*17*	*4*	*1*

3.3. Dissolved organic carbon

Water extractable DOC from the control soils had a mean value of 150 ± 68 μg g^{-1} which was significantly lower than mapped CDI soils on unit 1 ($p = 0.003$) which had a mean DOC mass of 1087 ± 727 μg g^{-1}. The mapped CDI in unit 2 had a mean DOC mass of 1484 ± 1236 μg g^{-1} and was also significantly higher than the upslope and downslope control soils ($p < 0.001$). There was no significant difference in DOC between the two units (Table 1). The lateral spread of mapped CDI for DOC was quite extensive for both units (Fig. 1E and F).

3.4. Soil nitrogen species

Dissolved organic nitrogen was widespread around the gravesite (Fig. 1G and H). While the control soils had 7 ± 4 μg g^{-1} DON, it was an order of magnitude higher in the mapped CDI and unit soils (Table 1). DON was significantly increased in the mapped CDI of the gravesoils relative to the control soils ($p < 0.01$). There was no significant difference in DON between the two mapped CDI or the two unit soils (Table 1).

Nitrate tended to display 'hot spots' of high nitrate-N at both units (Fig. 2A and B). The mapped CDI at unit 1 for nitrate-N was 49 ± 5 μg g^{-1} ($n = 3$) and the CDI on unit 2 was 64 ± 0 μg g^{-1} ($n = 1$) both significantly higher than the upslope control nitrate-N which averaged 7 ± 7 μg g^{-1}. There was no significant difference among the units and control soils for nitrate-N (Table 1).

Ammonium-N in the upslope control soils had a mean of 3 ± 1 μg g^{-1} dry soil with a range of 1–5 μg g^{-1} dry soil. Ammonium-N in the mapped CDI of unit 1 and had a mean of 52 ± 58 μg g^{-1} dry soil ($n = 16$) and a mean of 13 ± 7 ($n = 5$) in the CDI of unit 2 (Fig. 2C and D). Unit 1 had significantly higher ammonium-N relative to both control soils and unit 2 (Table 1).

3.5. Orthophosphate-P

Upslope control soil orthophosphate-P averaged 0.6 ± 0.2 μg g^{-1}, and 2.5 ± 2.2 and 1.9 ± 2.1 μg g^{-1} for units 1 and 2, respectively. Orthophosphate-P in unit 2 was not significantly different from the control soils or unit 1 (Table 1). Orthophosphate-P in the mapped CDI for both units were significantly higher than orthophosphate-P in the control soils ($p < 0.001$) and there was no significant difference in orthophosphate-P between the two CDI's (Table 1). The lateral spread of the CDI for both units was relatively large (Fig. 2E and F).

3.6. Soil anions: sulfate and chloride

In general sulfate in the mapped CDI's were lower than observed in the control soils. Control soils averaged 10 ± 8 μg g^{-1} sulfate whereas, apart from one 'hot spot of very high sulfate in unit 2 (Fig. 2H) sulfate was generally either non-detectable (0) or around 1 μg g^{-1} in both CDI's (Table 1). Low sulfate tended to be centered within the visual decomposition area in unit 1 (Fig. 2G) and toward the lower torso in unit 2. There were also pockets of low sulfate in the upper and lower quartiles (Fig. 2H). We found no significant difference in chloride among the unit and control soils (Table 1). Chloride was significantly higher in the mapped CDI of unit 2 relative to the control soils and unit 1. All chloride in unit 1 was within the range of our control soils and so no CDI existed for chloride.

3.7. Soil cations

Calcium was significantly elevated in the mapped CDI in both gravesoils relative to the control soils (Table 1) yet the lateral spread of the CDI in the two units was very different (Fig. 3A and B). In unit 1, protection from scavengers and removal of organs during autopsy tended to limit the CDI calcium to the upper torso within the expected CDI (Fig. 3A). In unit 2 the mapped CDI was up to 50 cm away from the remains and surrounded the whole of the remains (Fig. 3B). Unit 2 had significantly higher calcium than unit 1 and the control soils ($p < 0.01$). Magnesium displayed a similar mapped CDI in both units as calcium (Fig. 3C and D), and similarly unit 2 had significantly higher magnesium relative to unit 1 and control soils.

Potassium was significantly higher in both units compared to control soils ($p < 0.001$) and both units displayed a large spread around the remains, the lateral spread being much greater in unit 2 which was open to scavengers (Fig. 3E and F). Unit 1 did not display

Please cite this article in press as: J.A. Aitkenhead-Peterson, et al., Mapping the lateral extent of human cadaver decomposition with soil chemistry, Forensic Sci. Int. (2011), doi:10.1016/j.forsciint.2011.09.007

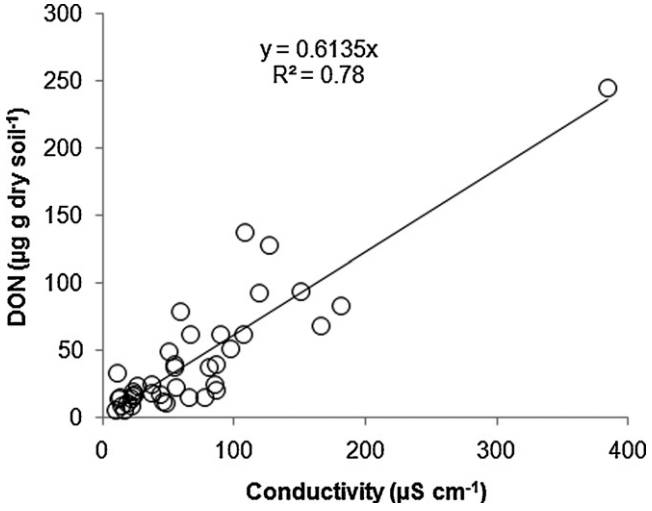

Fig. 4. Relationship between DON and electrical conductivity in gravesoils.

significantly higher sodium relative to control soils and so a CDI could not be mapped (Fig. 3G). Unit 2 and had an unusual CDI spread for sodium which tended to follow the core of the torso and display hot spots either side in the upper quarter (Fig. 3H). Unit 2 and its CDI had significantly higher sodium than the control soils, and the mapped sodium CDI's in unit 2 was significantly higher than in unit 2 (Table 1).

3.8. Relationship between DON and conductivity

Dissolved organic nitrogen was an order of magnitude higher within the mapped CDI's and units than is normally observed in natural environments. Regression analysis with electrical conductivity as the independent variable explained 78% of the variance in soil DON ($p < 0.001$) suggesting that this may be an inexpensive way to determine gravesoil in the field particularly between 5469 and 5799 ADD (Fig. 4). There was no significant relationship between DON and conductivity in control soils.

4. Discussion

pH and conductivity have been shown to increase and decrease in soil below human and other mammal remains [2–4,12,13]. Vass et al. [2] reported an increase in pH under human remains within a few ADD, peaking at 750 ADD and thereafter declining to control soil values at approximately 3750 ADD and declining to below control soil values up to 4500 ADD. Fiedler et al. [13] reported lower pH in graves that were approximately 27 years old relative to their control soils. Wilson et al. [4] reported increased pH from 4.6 to 7.2 during their monitoring of buried pigs up to 378 d whereas Pringle et al. [3] reported so much variability in pH among decomposition islands under pigs that they were not confident in its use as a determinant for PMI. In our study pH was significantly reduced relative to our upslope control soils which ranged from a pH of 5.2 to 6.2 supporting the time frame of lower pH after 4500 ADD observed by Vass et al. [2] and Fiedler et al. [13]. Fielder et al. [13] suggested that high pH in soils affected by adipocere relative to control soils reflect incomplete decomposition. High pH values will solubilize the native soil DOC humic acid fraction [16] but this tends to occur at pH > 8, much higher than the pH values observed in our study. Low pH values may result in iron bound P being solubilized from the soil matrix. Because low pH will solubilize iron, and orthophosphate tends to sorp to iron in the soil matrix, CDI soils with pH values of <5 may contain PO_4^{3-} derived from the soil matrix and not the corpse. Conductivity increased in the soil solution substantially under pigs [4] and was proposed as a way to determine PMI. While our conductivity was also increased relative to upslope control soils (approximately 4× at >5000 ADD) we found that the only chemical constituent that explained a significantly high amount of the variance in conductivity at 5469 and 5799 ADD was DON. It is highly likely that other chemical constituents' might be responsible, and have more importance for enhanced conductivity as the decay process proceeds.

Decomposing cadavers represent a resource of extremely high quality due to their low C:N ratio [1]. Our study found that the DOC:TDN ratio of the control soils (10 ± 3) was much reduced relative to our gravesoils (32 ± 21 and 19 ± 22) due to the extremely high inputs of dissolved organic carbon into our mapped CDI. While we did not quantify individual organic carbon compounds electing to measure bulk DOC, we were aware of the large pulses of propanoic, *iso*- and *n*-butanoic and *iso*- and *n*-valeric acids that could be expected based on the report of Vass et al. [2] in Tennessee alfisols soils. For example, Vass et al. [2] reported an approximate mean 1167 $\mu g\, g^{-1}$ propanoic acid and 1000 $\mu g\, g^{-1}$ *n*-butanoic acid at 450 ADD assuming the pre-death weight factor was 1. Work by Fiedler et al. [13] who sampled soils from two approximately 27-year old exhumed graves reported high DOC and evidence of lateral leaching. Our DOC values at 5469 and 5799 ADD suggested that DOC may not degrade significantly over time and that this was evidently the case in the Fiedler et al. study [13]. However, wooden coffin structures, embalming fluids and an anaerobic reducing environment in deep gravesoil may have all contributed to the high water extractable soil DOC reported in the Fiedler [13] study. What surprised us the most in our study was the movement or spread of DOC and DON relative to the decomposed remains. The spatial area of the mapped CDI was increased significantly when we considered DOC and DON compared to some of the other chemical constituents. Although the highest concentrations of DOC were beneath the pelvic region in unit 1 it had moved laterally away from the body at unit 2. Our individual units were relatively level with no obvious slope but the impenetrable adipose layer below 7.5 cm at our sample units may have induced further solubilization and spread of DOC; alternatively, some movement of the body by scavengers at unit 2 may also have been responsible for the extended lateral movement. Dissolved organic nitrogen also had an extended CDI similar to that of DOC. DON was likely comprised of soil microbes and enzymes plus components of the decomposing body, nucleic acids, amino acids, sugars and neutrals and proteins.

Little is understood about the fate of cadaver derived nitrogen and phosphorous in the soil ecosystem [2] but relatively recently ninhydrin-N extraction has been used to detect gravesoil [14,15] which likely is a measure of the DON and ammonium-N components of soil microbial biomass. We observed higher soil DON at 5469 ADD in unit 2 relative to DON at 5799 ADD in unit 1 and suggest that the amino groups had likely been enzymatically cleaved from DON molecules by 5799 ADD resulting in the higher ammonium-N observed in unit 1. Furthermore, the fact that ammonium-N was so high at 5799 ADD led us to speculate that nitrification was not occurring and the soil at this stage was still displaying anaerobic or reduced tendencies. This hypothesis was confirmed by a lack of or lower values for soil sulfate and bicarbonate. At the advanced decay stage the soil system will become anaerobic and anions such as indigenous soil and cadaver-derived nitrate, sulfate, bicarbonate and carbon dioxide will be used by soil micro-organisms for respiration. While sulfate use for anaerobic respiration was likely at both of our units (see Fig. 2) it was not apparent for nitrate. Vass et al. [2] reported an increase of approximately 525 $\mu g\, g^{-1}$ ammonium at 20 d post mortem compared to our ammonium increase of 190 $\mu g\, g^{-1}$ soil at 288 d post mortem in unit 1 and 21 $\mu g\, g^{-1}$ soil at 248 d post mortem in unit 2. Vass et al. [2] also reported that the peak of

Please cite this article in press as: J.A. Aitkenhead-Peterson, et al., Mapping the lateral extent of human cadaver decomposition with soil chemistry, Forensic Sci. Int. (2011), doi:10.1016/j.forsciint.2011.09.007

ammonium occurred at 750 ADD. Thus we should have expected ammonium-N to have returned to control values by the time we sampled, yet both CDI's had significantly greater ammonium-N relative to control soils. What is important here is that the ammonium-N will only be nitrified in aerobic soils and where our soils were undisturbed up until our sampling campaign, the soils in the Vass et al. study were being sampled every 3 d during the summer and weekly in the fall and winter. This continual disturbance of soil will in effect cause aeration of the gravesoil resulting in nitrification and removal of ammonium.

Benninger et al. [12] reported significantly higher mass of extractable phosphorous under pig (*Sus scrofa*) up to 100 d post-burial. Fiedler et al. [13] reported significantly higher total P in exhumed gravesoils relative to control soils. The typical ingredients of embalming fluid are formaldehyde and glutaraldehyde, neither of which contains P. Our PO_4-P was still significantly higher in our CDI's relative to control soils and the lateral spread was beyond the extent of the remains at >5000 ADD which supported the findings of Feidler et al. [13]. We believe that total P and orthophosphate-P may prove to be a stable and viable indicator of PMI with further research.

Base cations increase in CDI's [2]. Potassium, calcium and magnesium values in soil at the advanced decay stage were 300, 50 and 10 $\mu g\ g^{-1}$ soil respectively in alfisols in Tennessee [2]. These were different to our soil potassium values but similar to our soil calcium and magnesium which showed an average increase over mean control soil values of 32, 72 and 13 $\mu g\ g^{-1}$ soil, respectively in our unit open to scavengers and 26, 46 and 8 $\mu g\ g^{-1}$ soil, respectively in our unit protected from scavengers. Vass et al. [2] further suggested that only seven ions, Na^+, Cl^-, NH_4^+, K^+, Ca^{2+}, Mg^{2+}, and SO_4^{2-} might be useful due to their stability and reproducibility among subjects.

Any differences between our study and that of Vass et al. [2] and Fiedler et al. [13] are likely due to the different time-scales, climate, soil series and textures and burial depths among the sites, plus the fact that in the Vass et al. [2] study none of the cadavers were autopsied. Although it would seem logical that autopsied cadavers would decompose faster, this is not always the case. Parkinson [17] reported that in some instances autopsied cadavers decomposed faster and in other instances autopsied cadavers decomposed at a similar rate as un-autopsied cadavers. During autopsy many vital organs may be removed and not replaced, this may have an impact on cadaver decomposition rates and purge contents and may alter the resulting soil chemistry relative to an un-autopsied body. We did not find significantly enhanced sodium or chloride in the subject that had been autopsied and had their internal organs removed prior to placement which may be indicative of the chemical constituents that internal organs contribute to soil chemistry.

Due to the limitation in number of units we used, which was based on our protocol of units being at the top of the slope to (a) avoid contamination by water soluble chemical constituents and (b) wishing to use units that had not been affected with remains before, we were unable to significantly test the affect of scavengers and autopsy on the spread of CDI. Thus it was not possible, through replication to determine if these factors had a statistically significant effect on soil chemistry. We do believe however, based on our mapped CDI's that access to human remains by scavengers will increase the spatial extent of the CDI for some chemical constituents and furthermore, movement of body parts by scavengers will create hot spots of concentrated chemical constituents around the gravesite. Our data supports the studies of Melis et al. [8] who reported that the spatial extent of soil nutrients was quite large around decomposing bison due to scavenging and Fiedler et al. [13] who reported lateral leaching of DOC in gravesoil.

One of the most interesting findings of this study was the significant movement downslope of some chemical constituents, particularly orthophosphate-P, DOC, DON and potassium. The movement of these molecules downslope of human remains may have implications in terms of interpreting what might appear to be a false positive final response by human remains detection dogs, thus more research should be devoted to this phenomenon. Furthermore, based on the lateral mobility observed of DOC, DON and PO_4-P, downward migration of these decomposition chemical constituents to the water table is also a possibility although not tested in this study. While we have shown movement offsite for some anions more research is needed to assess the potential of adsorption to soil minerals, particularly of decomposition product DOC and DON to assess their potential for migration to the water table.

5. Conclusions

This study represented a snapshot of soil chemistry at 5799 and 5469 ADD of an autopsied and un-autopsied cadaver. Although we observed some similarities in our soil chemistry to those of other published studies we point out that we were examining a specific point in time. Furthermore, any differences observed relative to other studies were likely effects of time, scavenging, autopsy, soil type, and climate differences.

- Cadaver decomposition can have a significant and persistent affect on gravesoil chemistry.
- Electrical conductivity had a strong relationship with dissolved organic nitrogen, which might allow it to be used as a field test for gravesoil in the extended CDI.
- Significant downslope movement of gravesoil chemistry was observed, particularly orthophosphate-P, potassium, dissolved organic carbon and dissolved organic nitrogen.

Contributions

JAAP contributed to chemical and statistical analysis and production of the manuscript; CGO and NL contributed to soil collection and processing; MBA contributed to chemical analysis and JAB, director of the facility contributed to manuscript production and editing. All authors commented on the 1st draft of the manuscript. No funding was provided for this research.

Acknowledgements

We thank the STAFS facility within the Center for Biological Field Studies at Sam Houston State University for the opportunity to examine soils within their facility. This study is a combination of research for undergraduates C. G. Owings and N. Larison and preliminary investigations into CDI by PhD student M. B. Alexander. We thank Ms. N. Stanley for help with chemical analysis. Analysis of soils and manuscript preparation was funded by Texas AgriLife Research.

References

[1] D.O. Carter, D. Yellowlees, M. Tibbett, Cadaver decomposition in terrestrial ecosystems, Naturewissenschaften 4 (2007) 12–24. , doi:10.1007/s00114-006-0159-1.
[2] A.A. Vass, W.M. Bass, J.D. Wolt, J.E. Foss, J.T. Ammons, Time since death determinations of human cadavers using soil solution, J. Forensic Sci. 37 (1992) 1236–1253.
[3] J.K. Pringle, J.P. Cassella, J.R. Jervis, Preliminary soilwater conductivity analysis to date clandestine burials of homicide victims, Forensic Sci. Int. 198 (2010) 126–133.
[4] A.S. Wilson, R.C. Janaway, A.D. Holland, H.I. Dodson, E. Baran, A.M. Pollard, D.J. Tobin, Modelling the buried human body environment in upland climes using three contrasting field sites, Forensic Sci. Int. 169 (2007) 6–18.

Please cite this article in press as: J.A. Aitkenhead-Peterson, et al., Mapping the lateral extent of human cadaver decomposition with soil chemistry, Forensic Sci. Int. (2011), doi:10.1016/j.forsciint.2011.09.007

[5] H. Jenny, Arrangement of soil series and types according to functions of soil-forming factors, Soil Sci. 61 (1945) 375–391.
[6] E.M. Hoffman, A.M. Curran, N. Dulgerian, R.A. Stockam, B.A. Eckenrode, Characterization of the volatile organic compounds present in the headspace of decomposing human remains, Forensic Sci. Int. 186 (2009) 6–13.
[7] A.A.A. Vass, Odor analysis of decomposing buried human remains, J. Forensic Sci. 53 (2008) 384–391.
[8] C. Melis, N. Selva, I. Teurlings, C. Skarpe, J.D.C. Linnell, R. Andersen, Soil and vegetation nutrient response to bison carcasses in Bialowieza primeval forest, Poland, Ecol. Res. 22 (2007) 807–813. , doi:10.1007/s11284-006-0321-4.
[9] USEPA Method 350.1, Determination of ammonia nitrogen by semi-automated colorimetry, in: J.W. O'Dell (Ed.), Revision 2.0 August 1993. Environmental monitoring systems laboratory, Office of research and development, U.S. Environmental Protection Agency, Cincinnati, OH 45268. Available from: <http://www.epa.gov/waterscience/methods/method/files/350_1.pdf> (accessed: 05.03.2011).
[10] USEPA Method 353.2, Determination of nitrate-nitrite nitrogen by automated colorimetry, in: J.W. O'Dell (Ed.), Inorganic Chemistry Branch, Chemistry Research Division Revision 2.0 August 1993 Environmental monitoring systems laboratory, Office of research and development, U.S. Environmental Protection Agency, Cincinnati, OH 45268. Available from: <http://www.epa.gov/waterscience/methods/method/files/353_2.pdf> (accessed 05.03.2011).
[11] USEPA Method 310.2, Alkalinity (Colorimetric, Automated, Methyl Orange). Available at: http://water.epa.gov/scitech/methods/cwa/bioindicators/upload/2007_07_10_methods_method_310_2.pdf> (accessed: 05.03.2011).
[12] L.A. Benninger, D.O. Carter, S.L. Forbes, The biochemical alteration of soil beneath a decomposing carcass, Forensic Sci. Int. 180 (2008) 70–75. , doi:10.1016/j.forsciint.2008.07.001.
[13] S. Fiedler, K. Schneckenberger, M. Graw, Characterization of soils containing adipocere, Arch. Environ. Contam. Toxicol. 47 (2004) 561–568.
[14] D.O. Carter, D. Yellowlees, M. Tibbett, Moisture can be the dominant environmental parameter governing cadaver decomposition in soil, Forensic Sci. Int. 200 (2010) 60–66.
[15] D.O. Carter, D. Yellowlees, M. Tibbett, Using ninhydrin to detect gravesoil, J. Forensic Sci. 53 (2008) 397–400.
[16] F.J. Stevenson, Humus Chemistry: Genesis, Composition, Reactions, 2nd ed., Wiley, NY, 1994.
[17] R. Parkinson, Bacterial communities associated with human decomposition. Ph.D. Thesis, Victoria University of Wellington, 2009.

Please cite this article in press as: J.A. Aitkenhead-Peterson, et al., Mapping the lateral extent of human cadaver decomposition with soil chemistry, Forensic Sci. Int. (2011), doi:10.1016/j.forsciint.2011.09.007