

CASE REPORT

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Analysis of Suspected Trace Human Remains from an Indoor Concrete Surface

ABSTRACT: This paper describes the sequence of analyses used to determine the nature of a stain located on the floor of room in the former Athens Mental Health and Retardation Hospital in Athens, OH. The location of the stain was reported to be the position in which a decomposing body was discovered on January 11, 1979. The current stain is found to contain strong evidence for both natural decomposition products and deliberate adulteration. Microscopic analyses, solubility tests, FTIR, ICP-OES, pyrolysis-MS, and derivatization GC-MS were consistent in determining the removable parts of the stain to be composed mostly of calcium and sodium salts of free fatty acids, such as palmitic acid, consistent with previous descriptions of adipocere. The free fatty acids could have been formed via known bacterial degradation pathways or via saponification through the basic environment caused through contact with the concrete. To our knowledge, adipocere formation on an exposed indoor environment has not been described before. The stain and concrete also show signs of being chemically modified with an acidic reagent, such as Blu-Lite—a phosphoric acid-based cleaner that was a commonly used cleaner in the building from the time of discovery to the present day. The chemical etching appears to have been restricted to an area resembling the shape of a human body, which is consistent with deliberate adulteration of the appearance of the stain.

KEYWORDS: forensic science, adipocere, fatty acids, fatty acid methyl esters, human decomposition, human remains, trace analysis, pyrolysis mass spectrometry

On December 1, 1978, 53-year-old Margaret Schilling—a patient of the Athens Mental Health and Retardation Center located in Athens Ohio—went missing (1). Initial searches were unsuccessful, but on January 11, 1979 her body was found on the concrete floor, near a window, in an abandoned wing of the building. An autopsy revealed that she had died of natural causes and based on the significant decomposition, she was estimated to have been dead for 4–5 weeks prior to discovery (1). Eye-witnesses report that her body was naked and that her clothes were neatly folded nearby (2). The average temperature and humidity inside the room during the 6 weeks that she was missing is not known. However, because significant decomposition was noted at the time of discovery, the temperature in the room was certainly warm enough to facilitate bacterial degradation.

Although the floor was supposedly cleaned after removing her body, there now (December 2007) is present a very distinct white mark on the floor in the shape of a human body (Figs. 1 and 2). This “stain” has fueled numerous ghost stories and urban legends in the community, and has been featured in a TV documentary/drama (3). Until recently, however, the stain has never been chemically identified or explained. We were asked in March 2007 to identify the nature of the stain and report on whether or not the stain is authentic, e.g., a product of human decomposition, or has been deliberately modified at some point since February 1979, perhaps for dramatic effect. Numerous chemical and physical analyses (reported herein) revealed that the chemical composition of the stain is indistinguishable from adipocere.

When animals, such as mammals, undergo postmortem bacterial degradation in certain conditions (e.g., moist and anaerobic), triglycerides found mostly in adipose tissue undergo hydrolysis to

produce saturated and unsaturated free fatty acids (4–8). When the fatty acids are present mostly as insoluble calcium salts, the by-product is often termed adipocere, and it is a commonly observed product when humans are exhumed from grave sites (9,10). Initial formation of adipocere can take place in as short as a few weeks (1,11–13), but hydrogenation and hydroxylation of the unsaturated fatty acids may continue to occur for decades, depending on the conditions (5,13,14).

It is interesting to note that Margaret’s body was found in relatively dry conditions (e.g., on the fourth floor of a brick building) and in aerobic conditions (e.g., on the floor, exposed to air), which are not ordinarily conducive to adipocere formation. However, examination of exhumed human remains from dry vaults (15) reveals that excess moisture—beyond that found naturally in the body—is not required for adipocere formation. Many of the physical and chemical tests lead us to consider this process as a potential explanation for the occurrence of the removable residue from the stain. Because we could not find a similar case of adipocere formation on an exposed body in an indoor environment, we are making the results available to the forensic community.

Materials and Methods

Sample Collection

The stain is composed of a light-colored area in the shape of a human body which is outlined by a 4–6 inch wide darker residue. Selected areas of the white and dark residue are quite thick (up to 2 mm) and could be collected by scraping the residue off the floor with a razorblade. Samples were placed in labeled plastic vials and stored at room temperature. Samples were collected from several different locations of the light area (hereafter referred to as the light sample), the dark area outlining the body (hereafter referred to as the dark sample), and from the surface of concrete from an area on the other side of the room (a control). For comparisons, we also

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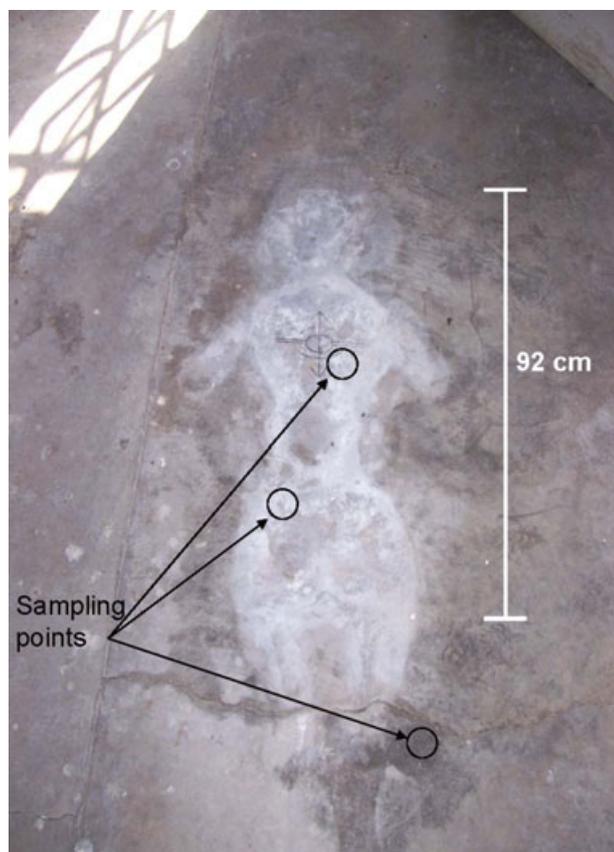


FIG. 1—Photograph of the questioned stain on the concrete floor showing the light area surrounded by a darker border.

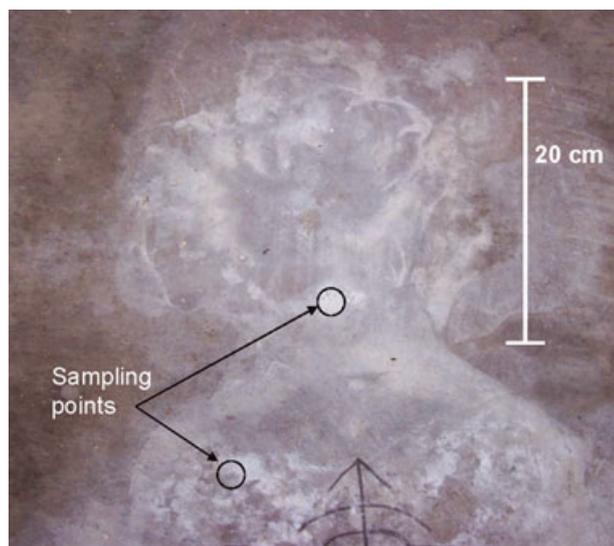


FIG. 2—Photograph of the head and shoulders of the questioned stain.

obtained and tested commercial bar soap (Camay, Proctor and Gamble, Cincinnati, OH) and human skin (from edge of fingernail of a consenting adult). We were informed by custodial staff that the spot was (at least) once cleaned with a phosphoric acid-based cleaner called Franklin Blu-Lite™ (Great Bend, KS). This cleaning agent is advertised as a cleaner for concrete surfaces and contains 20.5% phosphoric acid and 0.25% didecyl dimethyl ammonium

chloride. To obtain a reference field-sample of Blu-Lite, we poured a small amount of Blu-Lite on the concrete floor on the opposite side of the room to the stain and left it to dry. The residue was then collected in a similar manner to the other surface residues.

Microscopic Observations

A small amount of the light sample was placed on a microscope slide and observed under reflectance and transmittance light under a polarizing modular laboratory microscope (LOMO Polam L-213M, Northbrook, IL).

Infrared Spectroscopy

A small amount of the solid sample was placed on the center of an ATR cell (SensIR Technologies Dura SamplIR ATR, Danbury, CT) and analyzed without modification or dissolution using a Shimadzu Advantage FTIR-8400 (Columbia, MD). A total of 24 scans were averaged in absorbance mode with a resolution of 4 cm^{-1} .

Direct Insertion Probe-Mass Spectrometry (DIP-MS)

A small amount of sample was placed in a direct insertion vial. A direct insertion probe (Thermo Scientific, Waltham, MA) was used in chemical ionization (CI) mode using isobutane (99%, Airgas, Radnor, PA) as the CI gas on a Finnigan Polaris Q quadrupole ion trap (Thermo Scientific). The initial temperature of the probe of 50°C was held for 1 min. The probe temperature was then ramped at $150^\circ\text{C}/\text{min}$ to 450°C where it was held for 5 min. The ion source temperature was 200°C .

Gas Chromatography/Mass Spectrometry (GC/MS)

Saponification and derivatization was performed on each sample to convert the fatty acids to fatty acid methyl esters (FAME). Three reagents were prepared as follows: Reagent 1 contained 45 g NaOH, 150 mL methanol, and 150 mL distilled water. Reagent 2 was a mixture of 325 mL of 6.0 N HCl and 275 mL methyl alcohol. Reagent 3 contained 200 mL of hexane and 200 mL of methyl tert-butyl ether. Approximately 30 mg of each sample and 1 mL of reagent 1 were vortexed briefly and heated in a boiling water bath for 5 min. The samples were removed and vortexed briefly again and returned to the water bath for 30 min. After heating, the samples were cooled and 2 mL of reagent 2 was added. The samples were briefly vortexed and heated in an oven at 80°C for 10 min. After samples were removed and cooled, 1.25 mL of reagent 3 was added and the samples were shaken lightly for 10 min. The top hexane layer was removed and placed in a GC vial for analysis.

Chromatographic analysis was performed using a Trace GC (Thermo Scientific) coupled with a Finnigan Polaris Q quadrupole ion trap (Thermo Scientific) in EI mode. A $1\ \mu\text{L}$ aliquot was injected using a TriPlus autosampler (Thermo Scientific) and analyzed on a $28\text{ m} \times 0.25\text{ mm} \times 0.25\text{ mm}$ DB5-MS fused silica capillary column (Agilent, Santa Clara, CA). The carrier gas was ultrapure (99.999%) helium (Airgas, Radnor, PA) at a constant flow of $1.0\text{ mL}/\text{min}$. The initial column temperature was 70°C for 1 min. The temperature was then increased by $20^\circ\text{C}/\text{min}$ to a final temperature of 280°C and held for 5 min. A split injection of 10:1 was used with the injector temperature at 260°C . The transfer line temperature was 280°C with the ion source temperature at 200°C . A FAME standard (catalog number 47080-U; Sigma Aldrich, St. Louis, MO), containing 26 different FAMEs from 12:0 to 20:0 was

run as a positive control to identify the retention time of each FAME. Chromatograms were collected in full mass scanning mode and were evaluated using extracted ion chromatograms at m/z 55, 87, and 129. The main fatty acids of interest were the saturated fatty acids: lauric (12:0), myristic (14:0), palmitic (16:0), oleic acid (18:1) and stearic acid (18:0), which are the most commonly found homologues in living systems and decomposing matter.

Inductively-Coupled Plasma Optical Emission Spectrometry (ICP-OES)

A calibration curve was obtained from 0.1 to 25 ppm using multi-element ICP-MS calibration standard (catalog number CLMS-2N; Fisher Scientific, Waltham, MA) in 5% nitric acid. For each sample, approximately 5 mg was precisely weighed out and placed in a 15 mL plastic screw top vial. A total 300 μ L each of concentrated HNO_3 , concentrated HCl , and 30% H_2O_2 (all from Fisher Scientific), was then added. The samples were then heated until they dissolved and allowed to cool. After cooling, the samples were each diluted with distilled water to 10 mL. Some samples were further diluted in 5% nitric acid as necessary. The samples were analyzed using a Vista-MPX ICP-OES (Varian, Palo Alto, CA).

Results and Discussion

Although it has been claimed that the stain could have been caused by some kind of sunlight-induced photographic negative of Margaret's body (2), such claims have not been supported with any chemical or physical evidence. If authentic, the stain is much more likely to be a degradation product of the skin or fatty tissue covering Margaret's body, one example of which is adipocere. Although we frequently refer to the image as a stain, closer examination with a magnifying lens revealed the image to be a combination of a waxy residue on/in the concrete, and concrete with a significantly lighter tint than the surrounding area; e.g., the concrete itself appears to be chemically altered in the light areas of the stain.

Figures 1 and 2 show photographs of the stain at the supposed location where the body of Margaret Schilling was found, and the sampling locations. Initial observations of the physical appearance of the spot were: (i) there is a very obvious man-made marking in the center of the torso that is considered to be of no interpretive value, (ii) there are no forearms or legs (below the knees) in the image, (iii) there are very smooth edges defining the boundary of the light colored stain, and (iv) the light-colored stain is outlined by a larger darker stain. The off-white stain measured 94 cm (37") from the top of the head to the bottom of the buttocks, which is consistent with common anatomical dimensions of an adult female. Before questioning the shape and physical appearance of the stain, we performed significant chemical analyses to determine whether the composition of the stain was consistent with adipocere formation, or whether it was a foreign substance deliberately or unintentionally placed on the floor sometime between 1979 and 2007. We did not perform DNA analysis of the stain for several reasons: (i) we did not have a reference DNA sample for Margaret Schilling, (ii) the presence of DNA would not have proven that the entire stain was authentic or natural, only that part of it was originally of human origin, and (iii) the absence of DNA would not have proven the stain was completely synthetic or artificial—the DNA might have been present originally, but since degraded.

Figure 3 shows a photomicrograph of the light sample that was scraped from a thick (c. 2 mm deep) residue near the buttocks. The image is at 100 \times magnification in reflected light mode. It appears



FIG. 3—Photomicrograph of the white sample under a reflected light at 100 \times magnification.

to be an amorphous waxy substance with no distinct crystalline features. Observation under crossed polars in transmitted-light mode did not reveal any significant contributions from anisotropic matter. The substance contains very small dark particles and man-made fibers on occasion, which is to be expected for a residue that has been on the floor and exposed to air for more than 27 years. The majority of the wax-like substance was insoluble in water, 5% nitric acid, chloroform, benzene, hexane, acetone, and methanol. It was somewhat soluble in 1N NaOH and dissolved completely during saponification (boiling in concentrated NaOH).

Figure 4 shows the IR spectra of the light sample and of bar soap to show the similarities between the absorption bands of the two samples. Both samples are quite consistent with former FTIR studies of substances like soap, human skin or adipocere (16,17). Both the residue and the bar soap shows two peaks at c. 2916 and 2849 cm^{-1} , which are characteristic of C-H stretching of long carbon chains. The bar soap has a large peak at 1556 cm^{-1} and a triplet between 1470 and 1420 cm^{-1} which indicate the presence of carboxyl groups, probably sodium salt, with a long hydrocarbon chain (17). The spectrum for the light sample shows peaks at 1576 and 1539 cm^{-1} and a triplet between 1470 and 1420 cm^{-1} , which are also indicative of a carboxylate group, but apparently not the sodium salt. The light sample spectrum shows a very small broad

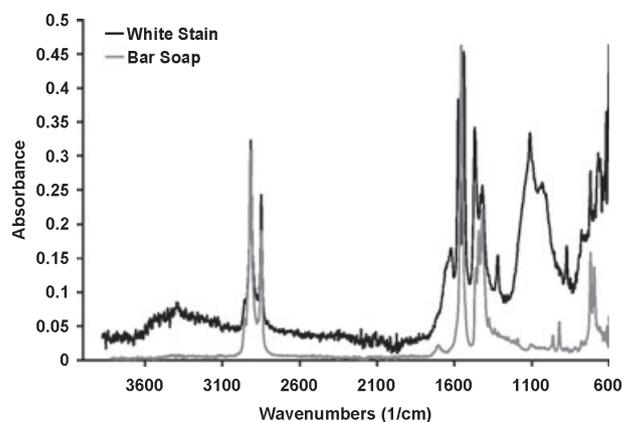


FIG. 4—FTIR spectra of (■) the light sample and (■) commercial bar soap.

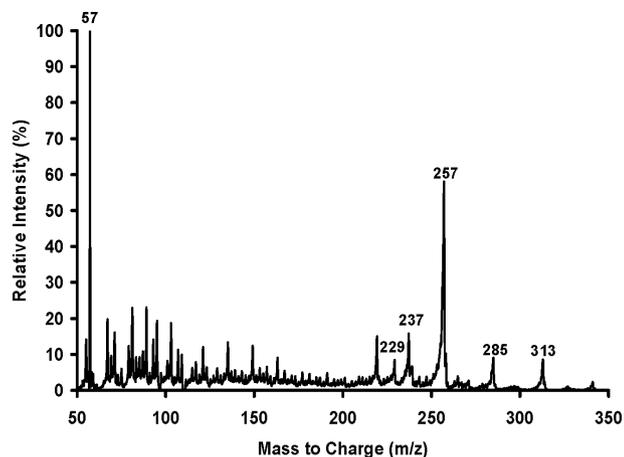


FIG. 5—Time-averaged (1.5–2.6 min) chemical ionization mass spectrum obtained during direct insertion probe analysis of the light sample.

peak above 3000 cm^{-1} , which indicates that only a small fraction of the carboxylic acid groups are in the protonated form; the O-H stretch of carboxylic acids is usually found in this region. The FTIR spectra indicate that the light sample and the bar soap both contain deprotonated long-chain carboxylic acids, although they may contain different metal cations. Indeed, the insolubility of the light sample is consistent with the hypothesis that the salt is not the sodium salt, but perhaps a dicationic insoluble salt, such as calcium or magnesium.

Figure 5 shows a time-averaged mass spectrum that was obtained during a direct insertion probe analysis of the light sample in CI mode. The sample was not modified in any way prior to loading two small grains into the $3\text{ }\mu\text{L}$ glass vial of the insertion probe. The spectrum is relatively uncomplicated, and shows a base peak at m/z 57, which corresponds to one of the reagent ions in the CI source. The peaks of interest appear above m/z 200 and are consistent with the following molecular ion designations: m/z 229 = protonated myristic acid (14:0); m/z 257 = protonated palmitic acid (16:0); m/z 285 = protonated stearic acid (18:0), and m/z 313 = eicosanoic acid (20:0). Closer observation of the pyrogram showed that the lower molecular weight fatty acids of myristic acid and palmitic acid were dominant during the initial increase in the temperature ramp, whereas the higher molecular weight species were somewhat enhanced at higher temperatures. Based on the DIP analysis alone, palmitic acid appears to be the most abundant fatty acid with the other fatty acids peaks making up less than 40% of the total fatty acid composition. A search for cholesterol (m/z 387 = $[\text{M} + \text{H}]^+$) and epicoprostanol (m/z 389 = $[\text{M} + \text{H}]^+$) (18) were also performed on the pyrogram but were negative, but this does not rule out the possibility of original human origin. Cholesterol was observed during the pyrolysis of human skin. The pyrolysis-CI-MS results also indicate that the light sample is composed largely of fatty acids, and is dominated by palmitic acid. To confirm these results and obtain a more sensitive and definitive analysis, we then performed derivatization and GC-MS analysis.

Saponification and derivatization were performed to convert bound and free fatty acids to FAME. The SIM chromatograms of m/z 87 for bar soap and the light sample are shown in Fig. 6. The soap and the light sample each contain almost exclusively fatty acids, but there is a considerable difference in the relative distribution of the fatty acids present. Different brands of bar soap are known to have different distributions of fatty acids, depending on their source, but we are not aware of any searchable database that

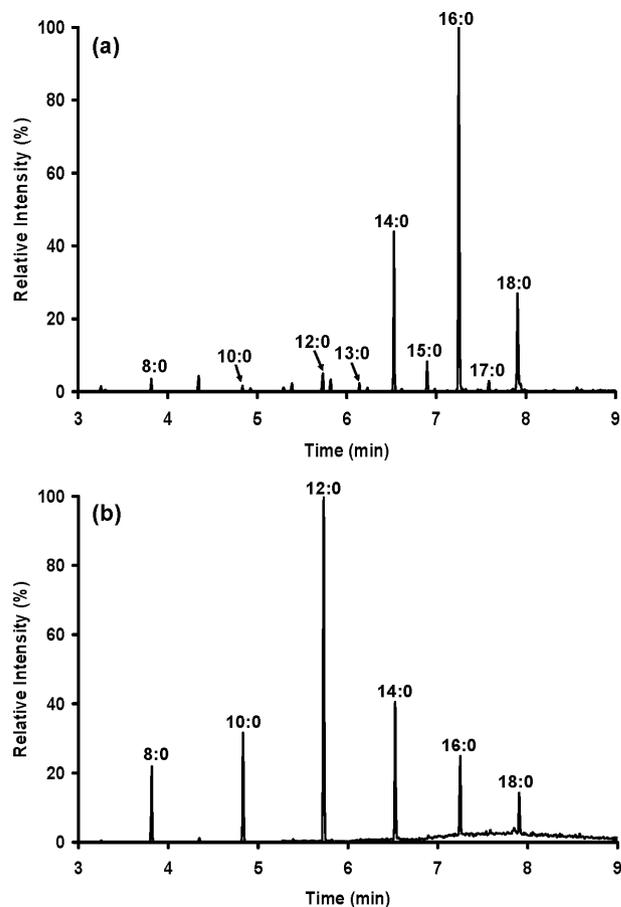


FIG. 6—Selected ion monitoring (SIM) chromatograms of m/z 87 for (a) the light sample and (b) commercial bar soap.

could be used to identify a possible bar-soap with a fatty acid composition similar to our stain sample. The closest example of a survey of chemical analyses of bar soaps involved elemental, headspace GC-MS, UV-Vis, and FTIR analyses (17). The relative distributions of fatty acids were not determined.

Table 1 shows the relative percent compositions of the four most abundant fatty acids present in each sample. For comparative purposes the light sample, dark sample, bar soap, skin, and a concrete surface from an adjacent corner of the room were also tested. Because Blu-Lite was suspected as being used as a cleaning agent for the stain at some point since the removal of Margaret's remains, we also tested Blu-Lite liquid and Blu-Lite that had been deposited on a remote part of the concrete floor, dried, and removed.

Untreated concrete from the other side of the room did not show any significant levels of fatty acids. When the concrete was treated with Blu-Lite, effervescence occurred and the concrete appeared

TABLE 1—Relative (%) fatty acid composition of samples determined with GC-MS (totals may not add up to exactly 100% due to rounding).

	Lauric (12:0)	Myristic (14:0)	Palmitic (16:0)	Oleic (18:1)	Stearic (18:0)
Light sample	2	19	60	0.1	19
Dark sample	6	9	68	<0.1	17
Bar soap	50	21	17	0.3	12
Skin	5	10	43	1.4	41
Blu-Lite™	6	11	35	<0.1	48
Blu-Lite™ on concrete	2	6	44	<0.1	48

considerably paler, when dry. The effervescence is caused by the acidic decomposition of concrete and the release of CO₂. Treatment of concrete with Blu-Lite therefore chemically etched the surface of the concrete in such a way that it left a white mark that could not be removed. Blu-Lite, and Blu-Lite recovered from the concrete showed low levels of fatty acids and did not show any significant differences in the distribution of fatty acid before and after exposure to the concrete surface. Because fatty acids are not listed as an active ingredient in Blu-Lite, we hypothesize that the fatty acids present in Blu-Lite could be an impurity, such as from lubricants or cleaning agents used in the production facilities. The fatty acid composition of Blu-Lite was significantly different from that of samples recovered from the stain, indicating that the light sample could not have originated purely from Blu-Lite. However, all the fatty acid components of Blu-Lite were identifiable in the light sample, including all the odd-numbered fatty acids present in trace amounts.

As shown in Table 1, palmitic acid is the most abundant fatty acid in both the light and the dark sample. The bar soap contained mostly lauric acid and, as can be seen by SIM chromatogram, a higher abundance of the shorter chain fatty acids. Skin contained significantly more of the longer chain fatty acids such as stearic acid, in agreement with previous results (19). The light sample also contains odd-chain-length fatty acids, which is representative of bacterial activity. In contrast, human adipose tissue (in the absence of bacterial degradation) and bar soap contain almost exclusively even-chain-length fatty acids. Previous research on adipocere formation shows that palmitic acid is often the most abundant fatty acid (5,6,20–23), even though oleic acid is by far the most abundant in adipose tissue during life (5,19,20). The exact distributions of fatty acids in adipocere varies considerably and is often related to the variety of environmental factors during adipocere formation, such as soil type and pH (20). 10-hydroxystearic acid and oleic acid were not present in the light sample, but these fatty acids are not always present in adipocere.

ICP was performed to identify the metal salts of the fatty acids present in each sample. Although the concentrations were determined using an external calibration curve on an absolute scale, the abundance of each metal is reported relative to the total composition of metal ions studied. This practice follows previous reports on the metal ion composition of adipocere (20). The sum of the absolute metal concentrations for the four cations in the solid samples varied from *c.* 6% by mass for skin to *c.* 11% by mass for the light sample, dark sample, and bar soap. Table 2 shows the relative weight percent of the metals calculated for each samples. Blu-Lite contained an excess of sodium before exposure to concrete, but contains a larger relative (and absolute) abundance of calcium after exposure to concrete. Relative to the four metals studied, the light and dark samples collected from the stain contain mostly calcium (55% and 66%, respectively) and sodium (43% and 24%,

respectively). In contrast, the metal ion composition of bar soap and skin is comprised mostly of sodium (99% and 91%, respectively). During adipocere formation, fatty acid salts typically take up K⁺ or Na⁺ from the internal environment of the body (4,20). Over time, when the body is in soil or moist environments, K⁺ and Na⁺ salts are usually replaced by Ca²⁺ or Mg²⁺ (10,24), thereby becoming insoluble.

Whereas Forbes and coworkers found significant levels of magnesium in adipocere samples found in grave sites (20), the magnesium contents in the light and dark samples were more modest and almost certainly reflect the relative contents of calcium and magnesium in the concrete from which the salts originated. We assume that the exposure of the decomposing tissue to the concrete surface—which contains an overwhelming abundance of calcium ions—has caused the sodium and potassium ions to be partially displaced by calcium ions during time periods when diffusion/permeation would have been possible. Such displacement would have to have occurred during the 4–5 weeks that Margaret's body was in contact with the concrete when moisture was present, or during periods when the floor was wetted during any attempted washings. Because we do not have accurate knowledge of the number or duration of washings, we speculate that the majority of the sodium displacement must have taken place during the decomposition of the body. If the stain is in fact entirely fabricated from bar soap (which cannot be corroborated without a comparison sample), the stain would have to have been hydrated for some period to allow the sodium in the bar-soap to become partially displaced by calcium from the concrete. In assessing the authenticity of the stain, there are two factors to consider: the chemical composition and the physical appearance. We have determined that all of the light and dark residues collected from the buttock, lower back, shoulder, and around the stain provide similar results; the residue in each location is composed mostly of calcium and sodium salts of fatty acids. Palmitic acid is by far the most abundant fatty acid present in all the residues tested making up between 60% and 70% of the fatty acid composition. The composition is consistent with what one would expect from adipocere formation (5,6,20–23), especially when in contact with a calcium rich- and basic surface, such as concrete.

Because the dark- and light-colored stains are chemically so similar, it seems reasonable to question why the inner part of the stain is so pale compared to the large dark area surrounding the stain. To see whether the dark areas of the stain could be made lighter, we exposed various areas of the dark stain to different reagents. Water and 3 M NaOH had little effect, but Blu-Lite and 5% nitric acid both had the effect of lightening the appearance of the stain (when dry). The lightening was due, in part, to chemical etching of the concrete below the residue—as witnessed by effervescence—and partly due to bleaching of the residue itself. Acid treatment of the dark residue, such as with Blu-Lite, could therefore provide a mechanism for lightening the color of the residue and chemically etching the concrete itself—both of which are consistent with the appearance of the stain.

This acidic modification also explains a questionable observation made around the head of the stain: there was very little surface residue found in the head-area of the stain, but significant discoloration of the concrete. A person lying on the ground on his or her back would only have a small surface area of their head in contact with the floor. It is known that liquefaction of the brain tissue can occur, and that brain decomposition fluids can seep out of cranial cavities during decomposition (25); this could have caused pooling of decomposing fluid in the 20 cm × 20 cm (8" × 8") area observed in the head region. However, this does not explain how anaerobic conditions could have been obtained in this area in order

TABLE 2—Relative (%) composition of major cations of samples using ICP-OES. Percentages are reported relative to the sum of the four major cations studied (totals may not add up to exactly 100% due to rounding).

	Ca	K	Mg	Na
Light sample	55	1	1	43
Dark sample	66	3	7	24
Bar soap	0.2	0.3	0.1	99
Skin	4	5	0.7	91
Concrete	64	15	13	8
Blu-Lite™	21	15	<0.1	74
Blu-Lite™ on concrete	97	0.5	1	0.7

to permit anaerobic bacterial hydrolysis of the glycerides into fatty acids. Under the buttocks or torso regions of the body, the weight of the body could have provided regions of oxygen deprivation, but such conditions could not have been established in the region of the stain not concealed by flesh. We therefore cannot explain how fatty acids could have been formed in the region around the head, unless the glycerides were saponified by the basic medium created by contact with the concrete. Because no significant surface residue could be collected from the head region, and because the region is so pale, it seems very likely that the stain in this area is due mostly to chemical etching (acid treatment) of the concrete, with possible physical re-distribution of some of the fatty acids from other parts of the stain.

Conclusion

Based on the chemical analyses of the removable waxy surface residues and close observation of the stain, we have formulated the following explanation for the appearance of the stain; regardless of the manner of death, Margaret's body was probably in contact with the area of the stain for a period of 4–5 weeks. During this time, significant decomposition is known to have occurred, indicating that the room was apparently warm enough to facilitate bacterial degradation. During this time, anaerobic bacterial decomposition could have taken place in the contact areas between the concrete and the heavier, fatty areas of Margaret's body, such as the buttocks, back and shoulders. Bacterial action is supported by the odd-numbered fatty acids found in the residues. Such decomposition, facilitated by the moisture naturally present in Margaret's body, formed free fatty acids from the lipids in her subcutaneous tissue. This process may have been accompanied, in part or in whole, by the basic conditions provided through contact with the concrete. During the 4- to 5-week period in which the free fatty acids were being formed, and in any subsequent washing over the years, at least half of the sodium ions were displaced by calcium ions from the concrete. The result is a waxy residue of mostly calcium palmitate which is up to 2 mm thick in certain areas of the stain. In most areas of the stain, the waxy residue also resides in surface pores in the concrete, consistent with the suggestion that removal of the stain was attempted on at least one occasion.

At some point since the removal of Margaret's remains in January of 1979, the floor has likely been treated with an acidic chemical—probably Blu-Lite (20.5% phosphoric acid)—to lighten the color of the waxy residue and of the concrete. The chemical etching was not uniform across the entire floor surface, however, but was selectively restricted to a shape that resembled the apparent outline of a human body. The mechanism of lightening of the dark residue and the concrete through acidic treatment was verified by exposing different small areas of the dark stain to Blu-Lite or 5% nitric acid. Had the acidic reagent propagated outside the shape of body when applied, accidentally or deliberately, the appearance of the stain would be very different from that observed today. If the acidic reagent was added in an attempt to remove the stain, the insolubility of calcium palmitate in neutral or acidic solutions would explain why the residue was not readily removed in the past. The waxy residue should be readily removable with a hot, soapy, or basic solution, but the etched concrete would indefinitely remain a lighter color than the surrounding areas, unless otherwise modified. While we are not at liberty to comment on any supernatural phenomena related to the stain, the chemical and physical evidence suggests that the stain has both natural and man-made properties. The procedures outlined in this project could be useful for identifying suspected human remains in cases where DNA or other physical evidence is unavailable.

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