Interpreting stains: Assessing the possibility of post-mortem interval estimation through taphonomic skeletal discolouration

by Morgan L. Graumann

Within the field of forensic anthropology, a primary goal for investigators is the estimation of a decedent's post-mortem interval (PMI). Given its importance, numerous unique techniques exist to aid in accurate and precise PMI determinations. However, very few of these methods thoroughly account for the timeline upon which osteologic material becomes stained by its depositional environment. The organic structure of bone allows for a unique pattern of discolouration, but it can be difficult to interpret these properly. In order to discuss the widest array of discolouring factors, the following comprises results from varied individual studies, with the specifics of each process briefly described in order to present a wider overview of each factor. Four key headings allow an organized approach to the material—(i) Soil Contents, (ii) Atmospheric Processes, (iii) Human Traces, and (iv) Nefarious Acts—with each heading discussing three common sub-discolourants. Stains arise either by natural or nefarious means, varying drastically by geographical location and climate, producing a range of observable results. The analysis concludes with an integrated discussion of the factors detailed herein, to ultimately evaluate the question of whether or not discolouration can be utilized as a reliable PMI-estimation methodology.

Estimating a decedent's post-mortem interval (PMI) is one of the primary goals and initiatives of medicolegal and forensic anthropological investigations (Haglund and Sorg 1997; Stewart 1979). Given its importance, numerous unique techniques exist to aid in accurate and precise PMI determinations. However, very few of these methods thoroughly account for the timeline upon which osteologic material becomes stained by its depositional environment, as influenced either by natural or nefarious means (Byers 2017; White et al. 2012).

As a porous, pale, and organic matter, bone is quick to adopt the colour of its surroundings (Lee Lyman 2022; White et al. 2012). During life, the proximity of bone to muscle, fat, and soft tissues typically produces a yellowy-white hue; however, the range of situation-specific post-mortem taphonomic possibilities increases this variability (Lee Lyman 2022; Stewart 1979). Recognizing that each scenario is unique and involves overlapping factors, there are common discolourants which have known methodological timelines for their rates of skeletal alteration. By researching an array of the more common, well-tested methodologies, the results should lend themselves as an additional tool in estimating PMI. Overall, the following examines common patterns of discolouration from both natural and unnatural realms in order differentiate between their origins and rates of deposition to assess whether bone stain interpretation would be an effective tool for estimating PMI.

In order to discuss the widest array of discolouring factors, the following is organized under four key headings: (i) Soil Contents, (ii) Atmospheric Processes, (iii) Human Traces, and (iv) Nefarious Acts. Each heading discusses three specific sub-discolourants, comprising results from various studies to include a brief overview

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of the involved processes and expected results. The analysis concludes with an integrated discussion of the factors detailed herein, in order to ultimately evaluate the question of whether or not discolouration can be utilized as a reliable PMI-estimation methodology.

(i) Soil Contents

Soil is an active, porous, organic collection of naturally occurring chemicals, mineral substances, and biological life (Pokines and Spiegel 2022). Pedology—the study of soil specifics—is extremely well understood, therefore offering the most robust research pathway within this study (Pokines and Spiegel 2022). The following specifically discusses those aspects of soils which are known to commonly discolour osteological material: (a) Tannins, (b) Plants, and (c) Minerals.

Tannins

Tannins are complex, natural chemical compounds which originate within the soil and groundwater around plants and trees (Kraus, Dahlgren, and Zasoski 2003). Tannins can stain everything from tree bark, to bodies of freshwater, to the wine-staining of human enamel (Pokines and Baker 2022; Pollock, Pokines, and Bethard 2017). The compound's structure allows it to seep into the porosities of organic material, accumulating over time to produce a darkened colour through a process termed 'soil staining' (Pokines and Baker 2022). Due to regional variance in soils, Munsell Soil Color Charts® are used to maintain uniformity in the description of colour variants within scientific reports and experiments, as the Chart® accounts for hue, darkness and saturation (K.I.C. 1994).

Tannins discolour along a scale of yellowy-orange to chocolate-brown, depending on their concentration, with lighter soils containing fewer tannins (Kraus, Dahlgren, and Zasoski 2003). They are more potent in warm, moist climates, and are particularly active around fleshed remains because the nitrogen released during decomposition fuels the staining process (Evans 2022; Kraus, Dahlgren, and Zasoski 2003; Pollock, Pokines, and Bethard 2017). Bones exposed to tannins in a warm, humid climate can develop initial staining within 2 days (Kraus, Dahlgren, and Zasoski 2003). In warm, dry climates it may require upwards of 1 week to achieve this same stain, while in cold, dry climates the process can take over one month (Pollock, Pokines, and Bethard 2017; Woolen 2019). Red and yellow colour spectrums increase over time, consistently observable as stains after the 8-week mark, while soil discolourants on the blue colour spectrum degrade over time and are typically only blatantly observable until the eighth week after deposition (Sauerwein 2011; Schultz and Dupras 2022).

The specific stain colour directly reflects the depositional environment—whereas metals or minerals may result in differently discoloured bones, those in soil or sand contexts will take on the specific surrounding soil colour (Nicholson 1998; Schultz and Dupras 2022). Whether buried or surface, dry or wet, hot or cold, a bone will not become darker than its surrounding soil regardless of the duration of exposure (Kraus, Dahlgren, and Zasoski 2003; Schultz and Dupras 2022). To form a stain, initial discolouration appears on the outermost layer of cortical bone, the point of tannin contact (White et al. 2012). Over a period of 50 to 80 years, stains gradually affect the cortical surface and then progress to the trabecular bone, simultaneously increasing the surface porosity (Schultz and Dupras 2022). Whiter, lighter soils feature higher sodium and quartz contents with less tannins and therefore

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less pigmentation with a lower ability to deeply stain (Kraus, Dahlgren, and Zasoski 2003). These paler environments require an average of 4 to 6 weeks before yellowy-orange surface stains may be observed (Sauerwein 2011; Schultz and Dupras 2022).

Remains buried directly in soil often become evenly discoloured due to the direct, equal access to the cortical surface, whereas surface finds result in spottier, localized discolouration (Behrensmeyer 1978; Pokines and Baker 2022). Remains buried in bags or non-porous coverings are only available to discolour once the covering degrades or breaks, resulting in spotty discolouration on whichever surface the tannins are able to reach. As such, bagged remains are subject to differing staining schedules depending on the occurrence of leakage (Jaggers and Rogers 2009; Pokines and Baker 2022). Alternatively, coffin remains consistently produce more evenly discoloured bones than plain soil burials, yet are often more damaged due to coffin wear (Janaway 2008; Pokines and Baker 2022; Pollock, Pokines, and Bethard 2017). Remains buried in coffins often result in darker discolourations after two months of skeletal exposure because darker wood contains a higher tannin concentration and over 70% of wooden coffins sold in the United States are produced using darker varieties such as mahogany or walnut (Kraus, Dahlgren, and Zasoski 2003; Pokines and Baker 2022). Equally dark colours may be achieved in environments of increased organic and tannin content, such as composting piles, initially observable within 2 to 3 days (Nicholson 1998). In general, soil-strewn bone will become a uniform chocolate-brown after approximately 25 years of exposure (Pokines 2016; Schultz and Dupras 2022; Ubelaker 1997).

Plants

Plants, as organic matter, are able to grow on anything from which they may collect nutrients, including bones (White et al. 2012). Growing specific plants on osseous tissue results in a distinct array of discolouring traits occurring along a relatively consistent timeline.

Burial produces a higher localized moisture content than surface deposition, resulting in darker stains over time, particularly of the green and black spectra (Ubelaker 1997). In particular, mosses and fungi present in the soil can grow in as little as 10 days (Huculak and Rogers 2009; Sauerwein 2011). Similar to soil staining, fleshed remains result in deeper discolourations due to the wide range of chemicals released during decomposition (Huculak and Rogers 2009; Pokines and Baker 2022). Additionally, mold can grow within the first week of exposure if conditions are appropriately moist, whether buried or on the surface, and can be found equally on both fleshed and skeletonized remains (Pokines 2016). Interestingly, North American mold creates a unique dark pink-mauve coloured stain (Schultz and Dupras 2022).

Surface plant materials become evident around two months after exposure, with different plants flourishing in different environments (Schultz and Dupras 2022). For example, shade promotes a dampened environment, allowing mosses, lichens, and algae to thrive. Mosses of the green, yellow, and red varieties gain their colours from chlorophyll, xanthophyll, and carotenoids, respectively (Davies 2004; Sauerwein 2011). Surface fungi cause dark black discolourations, similar to burnt bone (Ubelaker 1997). However, early fungi staining only permeates the uppermost layers of cortical bone, whereas burning pervades the full thickness (L’Abbé et al. 2022; Renke 2010). Peat moss produces a dark red-yellow stain, often initially visible between 4 to 8 weeks after skeletal exposure (Sauerwein 2011). Algae, while typically found in submersed aquatic environments, can also grow terrestrially in moist locations (Evans 2022; Sauerwein 2001; Ubelaker 1997). Terrestrial algae produce a deeper green stain and can begin populating a fleshed decedent within days, though it may take up to 2 months to be observed on skeletonized remains (Pollock, Pokines, and Bethard 2017; Ubelaker 1997; Renke 2010). Shaded remains decay at slower rates than elements under direct sunlight (Schultz and Dupras 2022). A case in Papua New Guinea records exposed surface remains as unaffected by weathering for over 60 years due to their placement in shade (Tappen 1994; Pokines and Spiegel 2022).

On the sun-drenched surface, grassy and leaf-strewn soils produce yellow-tinged stains around 8 weeks of exposure (Davies 2004; Sauerwein 2011). Leaves, acorns, roots, and shed tree bark discolour at the same rate (Huculak and Rogers 2009; Pollock, Pokines, and Bethard 2017). Leaves tend to produce larger-sized, lighter yellow-red stains than acorns, which are smaller and darker brown (Pollock, Pokines, and Bethard 2017). Pine needles create unorganized linear green-brown stains and may take up to 3 months to appear depending on the moisture level and season of deposition (Pokines and Spiegel 2022; Schultz and Dupras 2022). Discolouration from root etching occurs through the acid decalcification of the bone's external cortical surface over prolonged contact, resulting in darkened brown stains typically requiring over a decade to appear (Pokines 2016; Schultz and Dupras 2022). Grasses stain bone in the same manner as clothing, through high-velocity contact. However, it is unlikely to see grass growing on bone (Jaggers and Rogers 2009).

Minerals

Minerals permeate bone in the same manner as tannins, but result in unique, textured surface concretions with the added ability to form encrustations on the exterior of bone surfaces (Mann et al. 1998; Schultz and Dupras 2022). Minerals may interact with buried bone, yet are unable to leave visible residue until the moisture evaporates (Pokines and Baker 2022).

Sodium chloride (NaCl) is the most common soil mineral to affect bone, as it is water-soluble and can thus travel through both soil and marine contexts (Schultz and Dupras 2022). NaCl forms spicules on the bone's surface in the same way salt crystals form following the evaporation from water (Sutton 1981; Pokines and Higgs 2022; Schultz and Dupras 2022). Depending on the natural humidity of the environment and the flow of NaCl within the area, these spicules may appear within 48 hours (Evans 2022; Pokines and Baker 2022).

Calcium carbonate ($CaCO_3$) is less naturally abundant in soil than NaCl (Mann et al. 1998; Schultz and Dupras 2022). Instead of spicules, $CaCO_3$ produces a distinct off-white flaked coating on the bone's surface (Pokines and Higgs 2022). Given sufficiently dry surface conditions, single-layer $CaCO_3$ concretions can form within 2 days (Ubelaker 1997). However, harder, more visible concretions require numerous layers of built-up evaporates, typically requiring upwards of a month (Schultz and Dupras 2022).

Lastly, the rarest of the discolouring soil minerals is vivianite ($Fe^{2+}3[PO_4]_2$). From the iron phosphate family, vivianite causes dark blue-green surface encrustations within anerobic mud or marine environments (Mann et al. 1998). It turns a brilliant bright blue once exposed to oxygen (Schultz and Dupras 2022). Vivianite has recently been uncovered in connection with long-term cold-climate burials in Europe and is currently understood to require between 2 to 5 decades to form (Rodriguez et al. 2021).

(ii) Atmospheric Processes

Bones can become exposed to nature through both natural and nefarious means. The most common discolouring atmospheric factors are (a) Sun, (b) Water, and (c) Cold Temperatures.

Sun

Bleaching is the result of the sun interacting with exposed bone, resulting in the degradation of the organic matrix to produce an increasingly paler, whitened colour (L'Abbé et al. 2022; Schultz and Dupras 2022). Sun bleaching is notably more yellowy-white in tone than burned bone, with the latter becoming terribly fragile and often featuring grey ash in addition to its snow-white hue (Krap et al. 2019; L'Abbé et al. 2022; Prawira and Triyono 2019). Most commonly, shade-free boreal depositions result in sun bleaching only to the exposed bone surface with soil staining present on the inferior aspect (Pokines and Spiegel 2022; Ubelaker 1997). While soil staining may permeate the entirety of a porous bone, sun bleaching is restricted to areas of direct exposure (Stokes, Márquez-Grant, and Greenwood 2020; Schultz and Dupras 2022). Inner trabeculae can only be bleached if the cortical shell is damaged or absent (Pokines and Baker 2022).

Sun bleaching may occur on any exposed bone. However, the effects of this process are most pronounced on freshly skeletonized bones (Behrensmeyer 1978; Pokines and Baker 2022; Sauerwein 2011). The natural oils and fats present in recently de-fleshed remains causes sunrays to be magnified in the same way sunlight produces fire if magnified through a mirror (Stokes, Márquez-Grant, and Greenwood 2020; White et al. 2012). This initial moisture causes the degree of bleaching to be more pronounced in less time (Behrensmeyer 1978). A freshly skeletonized bone can develop notable surface bleaching after 3 weeks of exposure in the Northern Hemisphere, in countries such as Canada and the United Kingdom, requiring as little as 1 week in desert, equatorial environments (Sauerwein 2011; Schultz and Dupras 2022; Stokes, Márquez-Grant, and Greenwood 2020). In contrast, dry bone exposed to sunlight requires an average of two months to become visibly bleached in humid environments and four months in dry or cold climates (Huculak and Rogers 2009; Stokes, Márquez-Grant, and Greenwood 2020). This creates difficulty in determining PMI, as the effects of primary contexts can alter the final interpretation in cases of shorter intervals (Huculak and Rogers 2009; Sauerwein 2011).

In addition to the duration of exposure, the intensity of the sunlight in a given location also impacts the speed and extent of the bleached stain as the intensity of ultraviolet exposure differs globally (Stokes, Márquez-Grant, and Greenwood 2020). Sun bleaching can range anywhere from brilliant white nearer to the equator with minimal organic content remaining, to off-white in warmer, moist, tropical environments, to grey in the far North and South indicating the highest level of retained organic content (Behrensmeyer 1978; Schultz and Dupras 2022). For example, an exposed femur in Egypt can show signs of bleaching within a single week, becoming brilliant white through sun exposure after only 1 year (Sauerwein 2011; Stokes, Márquez-Grant, and Greenwood 2020). Alternatively, an exposed femur in the United Kingdom requires over 6 weeks of summer exposure and over 12 weeks of winter exposure to achieve the most minimal visual bleaching, yet it will never attain the brilliant white observed in the desert (Pokines and Baker 2022; Schultz and Dupras 2022;

Stokes, Márquez-Grant, and Greenwood 2020).

Water

Water is one of the most uniquely penetrative substances on earth, able to infiltrate the smallest porous crevices. In addition to its role in mineral and chemical transport, the complete waterlogging of osseous remains results in a specific set of observable traits due to the constant moisture and unique underwater marine environment (Evans 2022; Pokines and Higgs 2022).

One of the greatest differences between marine and terrestrial environments is the array of organisms. Marine life decomposes fleshed elements in more rapid and predictable succession than scavengers in terrestrial settings, thus resulting in quicker access to the skeleton (Brock and Madigan 1988; Pokines and Higgs 2022). However, encasing a body in non-porous material, such as a plastic bag, restricts organismal access to the remains (Evans 2022; Renke 2010). As well, newly decomposing bodies float, restricting the access of underwater scavengers (Evans 2022; Pokines and Higgs 2022). The most common marine invertebrates known to form on osteological remains are barnacles, branching corals, and bryozoan zooids, visible in approximately 2 weeks, 1 to 2 months, and 1 decade, respectively (Brock and Madigan 1988; Checa et al. 2019; Hageman et al. 1998; Pokines and Higgs 2022; Renke 2010; Wang et al. 2021).

The discolour of waterlogged bone may be mistaken for discolouration caused by soil burial, as both instances allow equal access to the circumference of the bone (Schultz and Dupras 2022). In the same manner as in soil, bone assumes the colour of the surrounding water (Kraus, Dahlgren, and Zasoski 2003). Warmer water conditions result in quicker decay and discolouration, such that waterlogged elements within at-or-below-freezing temperatures can appear fresh for over twelve months (Evans 2022; Jagger and Rogers 2009). As well, water of a lower pH (pH 1–4) results in quicker alteration (Evans 2022). Depending on the saturation of the water, a bone in a low pH environment may demonstrate signs of initial discolouration within an hour of exposure (Evans 2022; Schultz and Dupras 2022).

Some of the most common discolourants in fluvial environments are tannins, which wash from soil into groundwater and discolour lakes, rivers, bogs, and freshwater environments (Goffer 2007; Pollock, Pokines, and Bethard 2017). Waterlogged tannins produce the same dark-brown discolouration observed in terrestrial soil burials (Evans 2022; Kraus, Dahlgren, and Zasoski 2003; Schultz and Dupras 2022). Water allows tannins to travel more readily and continuously within bone; however, their lowered concentration in water rather than in soil ultimately slows the process (Evans 2022). Considering both factors, waterlogged fluvial tannin discolouration follows a slightly slower timeline than terrestrial staining, typically observed within 2–3 weeks after skeletal submersion, though colder temperatures result in longer delays (Evans 2022; Renke 2010). In contrast, the salt of marine environments bleaches human remains rather than darkening them, due to the chemical reaction of the saltwater environment (Pokines and Higgs 2022). Marine bleaching can be distinguished from sun bleaching in that it affects the entirety of the bone evenly and almost always correlates to an overall rounded, smoothed appearance of the cortical surface (Evans 2022; Pokines and Higgs 2022).

In deeper marine environments, acidic erosion becomes an additional discolouring factor (Pokines and Higgs 2022). The bone surface is altered in two steps over periods of 2–3 years. First, acids darken and harden the cortical layers into a rough, cracked covering (Amadasi et al. 2015). This is followed by the gradual disintegration of portions of the covering, revealing clean, pale underlayers for the process to repeat (Brock and Madigan 1988; Evans 2022). In the anaerobic portions of the world's oceans, altogether different colour patterns are observed. Dark sulphides are formed, resulting in deep purples, blues, and browns, and anaerobic iron oxides cause a dark black-red-brown staining (Cheney 2021; Evans 2022; Pokines and Higgs 2022; Rodriguez et al. 2021). It is common for these two to mix, presenting as a calico stain (Pokines and Higgs 2022). Bones may withstand marine contexts for over 300 years before being degraded by the frigid saltwater environment, whereas bone in fresh water can only survive for 1 to 5 decades (Brock and Madigan 1988; Pokines and Higgs 2022; Steptoe and Wood 2002).

In fully submerged freshwater ecosystems, brown and red algae flourish in addition to green algae which also grows terrestrially (Pokines and Higgs 2022; Zimmerman and Wallace 2008). Brown algae develops at a similar rate to green algae and may be observable on submerged bone around 1 month from initial exposure and up to 3 months in colder climates (Evans 2022; Pokines and Higgs 2022; Zimmerman and Wallace 2008). Conversely, red algae, although it develops at a similar rate, does not stain the bone. Instead, it forms hardened, calcified crusts which attach to the bone's outermost cortical layers (Brock and Madigan 2008; Pokines and Higgs 2022; Zimmerman and Wallace 2008).

Cold Temperatures

Particularly in the polar regions, there is a drastic change in temperature and humidity between the summer and winter months. Discolouring and decompositional factors often thrive in warm, moist climates, therefore most studies take place in these idealized conditions. However, it is worth studying the effects of frigid temperatures on bone appearance, given the variation in global climates and depositional environments. Colder climates impede the decomposition of soft tissues, making it difficult to assess the staining of underlying osteological material (Byers 2017; Pokines and Baker 2022). The majority of post-mortem information recorded in cold-weather studies assess that which happens to the soft tissue, though there are some which detail osteological alterations (Komar 1998; Mellen, Lowry, and Micozzi 1993; Woolen 2019).

Cold temperatures, particularly those below freezing, inhibit normal decompositional processes such as access of scavengers to remains, with fewer plants or microorganisms in drier cold-weather locales (Brock and Madigan 1988). Maggots may still be found on winter remains, though in fewer numbers (Byers 2017; Goffer 2007). Altogether, this results in soft tissue remaining on the bones for longer periods, sometimes fully adhering (Pinkowski 2021; Woolen 2019). Such environments produce desiccated black-purple soft tissue or mummified grey soft tissue, while exposed bone can develop greasy sections of adipocere and become distinctly bleached through repetitive freezing cycles (Byers 2017; Mellen, Lowry, and Micozzi 1993; Woolen 2019).

Buried remains in winter conditions require 2–3 months before the soft tissue separates from the bone, at which point the bone surface assumes black, grey, and red patterns of discolouration which darken gradually with prolonged exposure (Alfsdotter and Petaros 2021; Pokines and Baker 2022; Woolen 2019). However, buried elements in non-porous coverings, such as plastic bags, often remain intact with soft tissue discolouration resulting in white skin and deep brown muscles after approximately 3 months (Alfsdotter and Petaros 2021; Woolen 2019). Bagged bones often require over 6 months to become exposed, after which time they may discolour as per the decompositional fluids within the bag itself (Pokines and Spiegel 2022; Woolen 2019).

If bones remain encased in soft tissue in an exposed winter environment, the tissue can become a desiccated, mummified purple-blue-grey within 1 month, progressing to leathery, blackened and entirely hardened soft tissue after 3–4 months of exposure (Bostock, Parkes, and Williams 2019; Pokines and Spiegel 2022; Woolen 2019). In such situations, bones may not become exposed barring external taphonomic factors, such as scavenging. Tissues may also undergo a partial process of saponification, which is a modification of putrefaction similar to adipocere formation (Alfsdotter and Petaros 2021; Komar 1998; Schultz and Dupras 2022). Depending on the severity of the temperature, freezer burn may result in the bleaching of osseous elements after 1–2 months of exposure (Alfsdotter and Petaros 2021; Woolen 2019). Adipocere results from the anaerobic bacterial hydrolysis of adipose tissue and is common of warmer, moister climates; however, it can be observed as a white-yellow addition to bones after roughly 2 months of exposure in winter conditions, often described as resembling a ‘cheese type’ consistency (Alfsdotter and Petaros 2021; Komar 1998; Mellen, Lowry, and Micozzi 1993; Woolen 2019). In most cases of surface exposure, remains are typically skeletonized after 2–3 years in freezing conditions (Alfsdotter and Petaros 2021; Woolen 2019).

(iii) Human Traces

While substances in the immediate depositional environment are the most common discolourants, traces of a decedent’s material lifestyle may also exert a role in post-mortem bone staining. This section examines three factors which are commonly on or within humans, and thus are likely to remain with them after death: (a) Blood, (b) Metal, and (c) Clothing.

Blood

Blood is a particularly interesting discolourant because, contrary to most other factors, blood stains decrease in saturation over time, being darkest just after death (Capella et al. 2015; Haglund and Sorg 2002). During initial soft tissue decomposition, red blood cells go through hemolysis wherein they degrade and release hemoglobin into the body exterior to the typically confined circulatory system (Schultz and Dupras 2022). The concentration of hemoglobin is highest immediately after death and diminishes over time with the cessation of metabolic processes, resulting in this inverse staining pattern (Capella et al. 2015; Schultz and Dupras 2022).

Producing a dark reddish-brown colour on bone, hemoglobin lightens over time and is most easily evaluated by the decade (Huculak and Rogers 2009; Sauerwein 2011). The first decade after death features the darkest, most obvious stains, decreasing to a paler yet discernable stain over the second decade (Huculak and Rogers 2009; Schultz and Dupras 2022). Under both luminol analysis and gross observation, all traces of hemoglobin are entirely absent by 25+ years after death (Capella et al. 2015; Sauerwein 2011). Over three independent studies, these results remained consistent in over 80% of cases, providing quite reliable estimation data (Capella et al. 2015; Huculak and Rogers 2009; Sauerwein 2011; Schultz and Dupras 2022).

Metal

Corrosion occurs as certain metals face prolonged contact with air or water, and stains occur if the corrosion process occurs near organic material, including bone (Cheney 2021; Schultz and Dupras 2022). Apart from noble metals such as gold or platinum, the majority of metals are oxidizable and, therefore, susceptible to staining (Schultz and Dupras 2022). In forensic contexts, copper and iron are the most common discolourants (Cheney 2021; Janaway 2008; Schultz 2012). As is commonly the case, moist, humid environments are associated with higher concentrations of staining over quicker timeframes, as moisture accelerates the corrosion process (Janaway 2008; Schultz and Dupras 2022). Similarly, metal staining occurring in fertile soil contexts, as well as those in close proximity to fleshed remains, tend to produce deeper hued discolourations, as the natural elements in these contexts increase the potency of corrosion (Cheney 2021; Cronyn 1990; Pokines and Baker 2022; Schultz and Dupras 2022; Ubelaker 1997).

Copper has natural preservational properties as a result of its salty composition, often delaying putrefaction (Cheney 2021; Schultz 2012). Along with its bronze and brass alloys, copper staining typically results from buttons, zippers or bullets and affects those bones in closest proximity (Bostock, Parkes, and Williams 2019; Janaway 2008; Schultz and Dupras 2022). Beginning as a red-brown penny colour, copper eventually corrodes to produce a green-blue stain (Cheney 2021; Schultz 2012; Schultz and Dupras 2022). Before turning green-blue, copper produces a light pink-red stain within the first week of oxidization. However, as osseous material requires a minimum of 3 to 4 weeks of metallic contact to become stained, this pink-red colour is very rarely observed osteologically (Cheney 2021; Goffer 2007; Janaway 2008; Schultz and Dupras 2022). A pale but discernible green stain is often present after 4 weeks of copper contact, with a dark green stain appearing after eight weeks (Cheney 2021; Goffer 2007; Schultz 2012).

Corroded iron can produce a light rust-coloured orangey-brown stain on bone within 1 week of contact in warm and moist environments (Cheney 2021; Cronyn 1990; Goffer 2007; Schultz and Dupras 2022). In moist, well-oxygenated environments, there often appears a larger concentration of red within the orangey-brown stain (Cheney 2021; Goffer 2007; Schultz 2012; Schultz and Dupras 2022). Iron is commonly present in temperate and boreal soils, resulting in this same orangey-brown discolouration (Pokines 2016; Ubelaker 1997). Although rare in forensic settings, iron corrosion can cause pitch black or marbled red staining if subjected to wet, anaerobic conditions such as at the ocean's depths (Evans 2022; Goffer 2007; Pokines and Higgs 2022). Importantly, iron discolouration tends to affect a surface area larger than the dimensions of its original contact area (Cheney 2010; Schultz and Dupras 2022). As well, iron phosphate produces a unique, rare blue-green discolourant—vivianite, discussed above.

In addition to these more common metals, there are a range of other, lesser-known metals which can stain human bones if kept in close enough proximity. In non-forensic burial settings, eye caps made of aluminium were commonly used by the 1930s, producing a dull white-grey coating around the orbits (Mayer 2012). Soils containing high quantities of magnetite may result in brownish-black stains within 2 weeks, whereas soils low in this metal can result in blueish-black discolouration within the same timeframe (Goffer 2007; Schultz and Dupras 2022). Manganese is also naturally present in soils and may produce stains different in colour, depending upon the manganese compound (Janaway 2008). For example, manganese dioxide (MnO_2) stains black, manganese carbonate ($MnCO_3$) creates a lighter pink-brown colour, and permanganate ions ($KMnO_4$) result in deep purple-blue discolourations, all within 2–3 months (Goffer 2007; Schultz and Dupras 2022). Manganese dioxide and iron dioxide do not stain bones in the typical manner; rather, they penetrate and coat the cortical surface in a manner similar to red algae (Pokines and Higgs 2022; Schultz and Dupras 2022). These dioxides are often located in dolomite-rich areas, such as Italy and the southeastern United States, and may hinder medicolegal identification and natural taphonomy (Pokines 2016; Pokines and Spiegel 2022).

Clothing

As above, clothing with metallic elements may produce corrosion stains on exposed bone after as little as 2 weeks, with the specific stain location informing jewelry patterns and, thus, PMI (Bostock, Parkes, and Williams 2019; Janaway 2008; Schultz and Dupras 2022). For instance, if a decedent has a zipper stain, it is likely a modern case. Earrings and bracelets tend to be the most common elements of jewelry to produce discolouration and affect the mandible and carpals, respectively, occasionally observed on the distal ulnae and radii (Schultz 2012; Schultz and Dupras 2022).

In addition to metals, the types of fabric and the specific dyes used in clothing can have different effects on fleshed and skeletonized remains. Specifically, non-commercial dyes tend to be more potent—for example, tie-dying a shirt retains a more dye than a shirt coloured commercially, resulting in a greater concentration of dye for a stain (Miller 2002; Janaway 2002, 2008). Similarly, darker hues such as reds, blues and black are more easily captured on bone's pale surface in the same way washing a single dark item in a load of whites discolours the entire load of laundry (Miller 2002; Janaway 2002:387, 2008; Pokines and Baker 2022).

Fabrics decay at different rates, releasing the dyes from their fibers as they decompose (Miller 2002; Janaway 2002). Fabrics which take longer to decay tend to produce deeper coloured stains, as there is an increased period of exposure to both the fleshed elements as well as to the exposed bone surface (Miller 2002; Pokines and Baker 2022). In some instances, clothing remnants may adhere to the bone surface, although particularly degradable fabrics are susceptible to the chemicals released during decomposition (Janaway 2002). This occasionally stains the flesh but has no impact on the bones, as the clothes decay faster rate than skeletonization occurs (Byers 2017; Janaway 2008; Rowe 1997). These degradation processes occur more rapidly in moist, warm climates (Pokines and Spiegel 2022; Woolen 2019). Plant-based fabrics decay much more quickly than synthetic fabrics. For example, linen takes an average of only 2 to 3 weeks to degrade fully—less, if it is undyed—and is often removed in the decomposition of the fleshed body (Arnett 2019; Bostock, Parkes, and Williams 2019; Miller 2002; Nicholson 1998). Cotton breaks down between 4–6 weeks in warm, humid environments, yet can require over 6 months in colder climates. Rayon requires 8–10 weeks in the same warm environment, while wool remains largely intact for 8–10 months (Arnett 2019; Byers 2017; Miller 2002; Janaway 2002; Rowe 1997). It is important to note that the layering of garments introduces variability. For example, a thick jacket or plastic rain boots will alter the rate of decomposition. Even layers of basic cotton will take longer to degrade than single-layered cotton (Janaway 2002; Rowe 1997).

Regarding non-plant-based materials, nylon can survive roughly 40 years of exposure, leather can withstand over a century, and polyester is estimated to exceed 500 years, making these some of the more useful materials to assess in forensic cases (Arnett 2019; Pokines and Baker 2022; Janaway 2008; Rowe 1997). Further, the presence of these fabrics indicates a modern decedent, as ancient societies did not have access to modern synthetic fabrics (Pokines and Baker 2022; Ubelaker 1997). The colour of stains produced by clothes of any fabric type is directly dependent on (and reflective of) the associated dye colours—that is, red dyes make red stains, and so on (Miller 2002; Pokines and Spiegel 2022).

(iv) Nefarious Acts

Of particular importance in forensic investigations are the uniquely unnatural events associated with an untimely passing. While some nefarious acts leave physical traces, such as blunt or sharp force traumas, other actions leave coloured stains on the remains. The most common of these processes include: (a) Acids, (b) Bases, and (c) Burning / Heat Exposure.

Acids

A range of acids may be employed by perpetrators in forensic scenarios cases as attempts to destroy evidence (Stewart 1979). Contrary to previous discolouring factors, acids tend to discolour flesh and bone on contact and go on to destroy the remains entirely (Cope and Dupras 2009; Schultz and Dupras 2022). Further, acid staining does not vary by climate, humidity, or temperature, nor does the presence of flesh—by natural or nefarious means—make a difference on the colour of the resultant stain (Savage 2018; Schultz and Dupras 2022).

The most common acid in forensic settings is hydrochloric acid (HCl), which produces dark brown stains after brief contact with bones or flesh (Hartnett, Fulgihiti, and Di Modica 2011; Schultz and Dupras 2022). The colour comes from the acid's corrosive nature, similar to rusting iron (Ubelaker 1997; Ubelaker and Sperber 1988). Contrary to its portrayal in media, it takes an extended period of time to dissolve a body in HCl—roughly 12 hours for human teeth and 24 hours to finish the entire body (Cope and Dupras 2009; Hartnett, Fulgihiti, and Di Modica 2011; Schultz and Dupras 2022). Nitric acid (HNO_3) follows the same dissolution pattern as HCl, but is more effective at destroying DNA and physical fragments of a decedent (Ubelaker and Sperber 1988; Schultz and Dupras 2022). However, HNO_3 is prone to explosions and emits extremely toxic fumes, making it a more dangerous substance with which to work (Cope and Dupras 2009; Hartnett, Fulgihiti, and Di Modica 2011).

Sulfuric acid (H_2SO_4), particularly infamous among the mafia in both North America and Europe, produces a reddish-purple stain on contact with flesh or bone, making it discernable from other common acids (Hartnett, Fulgihiti, and Di Modica 2011; Schultz and Dupras 2022). An undiluted batch of H_2SO_4 can dissolve a body in just over 2 days, while a less potent (and safer) batch requires over 1 week (Cope and Dupras 2009; Hartnett, Fulgihiti, and Di Modica 2011). Another recently popularized option is hydrofluoric acid (HF), commonly touted on television yet actually extremely ineffective at destroying harder tissues. HF results in a pale white-bluish staining on bones and teeth, and will retain portions of cartilage if the solution is diluted (Palmer 2009; Hartnett, Fulgihiti, and Di Modica 2011).

Bases

Sodium hydroxide (NaOH) is the most popular dissolving base among criminals, particularly among the Mexican mafia, as it is a cheaper and safer alternative to acid (Savage 2018). However, bases have little effect on the skeleton, only dissolving soft tissues such as fingerprints or identifying features (Cope and Dupras 2009; Palmer 2009; Schultz and Dupras 2022). Typically, NaOH (lye) is mixed with water and heated to above 300ºC, after which the solution can turn soft tissues into the consistency of mineral oil within 3 hours (Savage 2018). This produces yellow-orange, coarse-grained bleaching stains on the cortical surface (Hartnett, Fulgihiti, and Di Modica 2011).

Potassium hydroxide (KOH) and chlorine bleach (NaClO) are two additional bases commonly used in criminal settings (Cope and Dupras 2009). Characteristic of bases, neither KOH or NaClO will dissolve bone and they require over 6 and 24 hours, respectively, to dissolve human flesh and create discernable bleaching on the bones (Hartnett, Fulgihiti, and Di Modica 2011). Bases are less effective than acids at dissolving hard tissues, but are favoured for their increased safety (Schultz and Dupras 2022). Typically, skeletal remains are removed from bases and dissolved in smaller batches of acid, making the process safer yet extended (Palmer 2009; Savage 2018).

Heat Exposure

The burning of human remains can be intentional, accidental, or the result of natural causes. Each context has its own set of specific, attributable factors. Unlike most discolourants, burning follows a reasonably predictable progression, although the charred and calcined effects of burnt bone may be confused for fungal growth, manganese contact, or sun bleaching (Byers 2017; L'Abbé et al. 2022; Schultz and Dupras 2022).

The change in colour associated with burning bone reflects the progressive loss of organic matter. The first stage is carbonization, when the bone becomes visibly blackened. During this stage, much of the organic material is expelled from the bone with merely the carbon remaining. As carbon is naturally black, the bone appears charred (Goffer 2007; L'Abbé et al. 2022). Bodily fluids and tissues may be retained throughout carbonization, appearing dark brown-black as they sear to the bone surface (L'Abbé et al. 2022). Carbonized charring notably infiltrates the thickness of the bone, whereas black stains from minerals or soils tend to remain on the outermost layers. Additionally, carbonization is uniquely non-luminescent, further differentiating burnt bone from other stains (L'Abbé et al. 2022; Schultz and Dupras 2022; Shahack-Gross, Bar-Yosef, and Weiner1997). This stage occurs at temperatures above 500°C after periods of 4 to 6 hours. Natural brushfires typically burn between 400 and 600°C, progressing too quickly to carbonize bone (L'Abbé et al. 2022; Alaoui et al. 2014). The cooking of bones, either human or animal, only reaches approximately 100°C, which does not facilitate any carbonization (L'Abbé et al. 2022). At these lower temperatures, bone becomes progressively paler as lipids are rendered and become increasingly smooth and translucent after 5 to 7 hours of boiling (Bosch et al. 2011; L'Abbé et al. 2022).

Continued exposure to fire at temperatures between 500–900°C eventually releases the blackened carbon as either CO or CO_2, leaving only the bone's original hydroxyapatite behind (L'Abbé et al. 2022). Alone, the hydroxyapatite mineral is a fragile white-grey, resulting in the formation of ceramic-like calcined bone (Alaoui et al. 2014; L'Abbé et al. 2022; Prawira, and Triyono 2019). Professional crematoriums operate between 800 and 980°C, requiring roughly 3 hours to achieve a state of powdered, calcined ash for a single decedent (Alaoui et al. 2014). However, bone fragments still need to be separately ground down following cremation (L'Abbé et al. 2022). Notably, cleaned bone burns roughly 30% quicker than fleshed bone in crematorium settings, requiring less than 2 hours to reach calcination (L'Abbé et al. 2022; Triyono, Prawira, and Triyono 2019). If heating is extremely localized, a single bone may exhibit two or more of these stages at once (L'Abbé et al. 2022). Altogether, there are a myriad of ways to discolour post-mortem remains under both natural and unnatural circumstances.

Discussion

After analyzing a subset of common discolourants, it becomes necessary to understand the aspects of currently standardized PMI techniques in order to evaluate the value of discolouration as its own PMI methodology. In order to be effective, a PMI technique must be specific, repeatable, and based in a varied and robust body of evidence. The following examines how post-mortem bone discolouration reflects these criteria by discussing the findings above, and comparing against existing field methods.

Specificity

The level of specificity for taphonomic discolouration is not particularly refined, due, in part, to the current lack of robust research on the topic, alongside the finite limitations of observable colour. With few defined colours and a vast array of potential discolourants, the possibility of specificity is limited. In contrast, forensic entomology offers specific knowledge of insects, their lives and their reproductive cycles. The highly utilized discipline of forensic entomology applies specific knowledge on the timing of insect and arthropod lifecycles to estimate a decedent's post-mortem interval (Byers 2007; Joseph et al. 2011). Depending on the specific assemblage of findings, this method can provide an estimation within the hour and, in some cases, even to the minute, for which its specificity enhances its medicolegal reliability (Monika et al. 2020). For example, the presence of first-generation flat-winged blow flies can only suggest a PMI estimate of 10 minutes, whereas a black stain may originate from fungi, burning, iron oxides, or particularly tannin-heavy soils, each presenting their own varied, individual post-mortem interval estimates (Joseph et al. 2011; Monika et al. 2020).

Repeatability

There are a multitude of extraneous variables influencing any given taphonomic observation such that the distinguishing of causes and repeatability become challenging. This same issue is mirrored in the age estimation technique of pubic symphysis degradation (Meindl et al. 1985). The basic principle is clear that the symphysis degrades over time and therefore can be utilized to estimate age at death; however, the activity levels, health issues, and genetic predispositions of an individual can have distinct effects on the presentation of the symphysis, making the final estimation less precise (Suchey and Katz 1998). Similarly, there are numerous antemortem health conditions that have discolouring effects on osseous tissue which may influence a post-mortem analysis if not properly documented. For example, a pale-yellow to dark green-black may be created by certain tetracycline antibiotics (Eliasson and Wingren 2018:32; Farahnik 2015; Steadman, Brown, and Wall 2021). As studies have not yet accounted for the interaction between post-mortem discolourants upon antemortem stains, this becomes more difficult to interpret. These variabilities make it difficult to replicate results because the addition or subtraction of a single factor may have far-reaching consequences, resulting in an incomplete methodology. Few academic studies account for the role of scavengers on post-mortem remains because it is so drastically varied and subjective. There is a bias towards unnaturally protected circumstances in controlled experiments, but it shifts the discolouration schedule by failing to account for the increased pace at which scavengers expose bone in addition to the regular processes of decomposition. As such, it would appear more appropriate to read Timelines 1 and 2—located in the appendix—as less ‘time since death,’ and more ‘time since skeletal exposure.’

Many studies utilize porcine models due to the extreme likeness between Sus scrofa and Homo sapiens tissues; however, this has prevented study into the discolouring discrepancies between adult and juvenile bones. The pliability of forming juvenile bone tissue alongside their smaller size and markedly lower ratio of primary to secondary osteons within younger bones all likely corresponds to a difference in staining patterns either over time or in the saturation of the hue; however, while these differences might reasonably be assumed, further studies are needed to provide a practical, functional scientific basis. Overall, it is difficult to replicate the endless array of possible discolouring situations a human bone may undergo after death, making a more precise estimation of occurrences from visual discolouration less reliable.

Diversity

Perhaps the greatest discrepancy in the current pool of discolourant research is the limited consideration of the range of environmental and situational variability. For example, one summer study in Egypt and one winter study in Chicago do not offer a full accounting of the situational possibilities, just as uniform chocolate-brown staining rate achieved after 25+ years of exposure likely alters in different environments. There are a number of factors which could be assumed to remain the same between warm and cold climates, such as the rate of polyester degradation, the timeframe of leaf litter staining, or the rate of copper (and copper alloy) oxidization but, to the author’s knowledge, specific studies into such materials have not yet been completed. It is worth noting that the past decade of climate change has had an appreciable effect on the natural degradation of human remains and may impact the validity of older studies. In Edmonton, the average peak summer temperature in the late 1980s was between 24ºC and 26ºC, but the same city is now recording annual stretches above 40ºC (Environment Canada 2023). Similarly, traditionally hot states such as Texas and Florida have begun experiencing atypical snowfall in recent years (NOAA 2023). This alteration in climatic activity alters the rate of processes acting upon deposited remains. As such, the current body of literature discussing natural discolouring agents, such as tannin staining, may no longer prove to be as accurate as they once were.

Lastly, as noted above, a majority of the current information comes from Sus scrofa studies, biasing those results while also lacking an account of juvenile factors. In the Woolen (2019) study, one sample included a limb with markedly more flesh than would exist on a human, introducing variability in the amount of time needed to decompose the flesh and expose the bone.

Concluding Remarks

Overall, it appears many of the current limitations of this methodology could be upgraded to a standardized state with more vigorous experimental and modern research on topics such as wider ranging global climates, alternative seasonal environments, and varied decedent age ranges. Regarding the original research question—can the rates of visual alteration of various discolourants be used as an effective tool by which to estimate a decedent's post-mortem interval —the answer, as the research currently stands, is no. The rates of alteration are specific in certain cases such as clothing degradation, acidic contact and burning, but given the larger limitations of applicable knowledge in this field, this method is not yet an effective tool for PMI estimation.

The study of taphonomic bone staining remains extremely valuable because it provides investigators with information about the environment and post-mortem conditions. However, there are limitations in this research that do not lend this technique to being used as an estimator of PMI. The exacting specificity for the subject, as it stands, has not reached a state whereby this technique may be reliably utilized, though the past decade has seen an increased number of studies directed towards individual discolourants in variable climates. Recent literature on the topic shares this perspective that with additional research, the field will become increasing useful.

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Appendix: Warm and Cold Climate Discolouration Schedules

TIMELINE 1: Warm Climate Discoloration Schedule

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TIMELINE 2: Cold Climate Discoloration Schedule