

La mer emporte, les os se souviennent: FTIR-ATR analysis of bioapatite preservation in skeletal remains from the 18th-century Fortress of Louisbourg to inform coastal archaeological rescue operations

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Abstract

Ongoing coastal erosion over the last three centuries has prompted rescue excavations of the Fortress of Louisbourg Rochefort Point Cemetery in Atlantic Canada. Given the imprecise nature of 18th-century record-keeping and mapping, the exact location of the cemetery is unclear; therefore, determining where to excavate is vital to ensure a timely and efficient recovery of these individuals before the site is destroyed and their stories are lost. Fourier-Transform Infrared (FTIR) spectroscopy was applied to femoral cortical bone from 51 individuals to assess diagenetic alteration and to inform future excavation planning. Visual preservation scores averaged 2.4/3, closer to poor preservation, while molecular preservation showed greater variability [IRSF: 2.17–3.95; C/P: 0.14–0.80; Aml/P: 0.10–0.77; API: 0.13–1.12; BPI: 0.31–1.32]. Significant correlations between worsening skeletal preservation and proximity to the northern coast confirm that saltwater drainage impacts bone more substantially than burial soil conditions. This study provides a framework of factors to be considered when excavating human remains in different burial conditions to maximize recovery.

L'érosion côtière des trois derniers siècles a conduit à des fouilles de sauvetage du cimetière de la Pointe Rochefort de la Forteresse de Louisbourg (RPC), au Canada atlantique. La localisation précise du cimetière demeure incertaine; déterminer où fouiller est donc essentiel avant que le site ne soit détruit et que ses histoires ne soient perdues. La spectroscopie infrarouge à transformée de Fourier (FTIR) a été appliquée aux os corticaux fémoraux de 51 individus afin d'analyser l'altération diagenétique. Les scores de préservation visuelle étaient en moyenne de 2,4/3, ce qui se rapproche d'une préservation mauvaise, tandis que la préservation moléculaire présentait une plus grande variabilité [IRSF : 2,17 – 3,95 ; C/P : 0,14 – 0,80 ; Aml/P : 0,10 – 0,77 ; API : 0,13 – 1,12 ; BPI : 0,31 – 1,32]. Des corrélations significatives confirment que le drainage d'eau salée près de la côte nord du site affecte la diagénèse osseuse d'avantage que les conditions du sol. Cette étude offre un cadre de référence des facteurs à considérer lors de fouilles de restes humains dans diverses conditions d'enfouissement afin d'en maximiser la récupération.

1. INTRODUCTION

1.1 Fortress of Louisbourg Skeletal Collection

Situated at the Fortress of Louisbourg National Historic Site of Canada in Unama'ki (Cape Breton Island), Nova Scotia, the Rochefort Point Cemetery (RPC) was in use between 1738 and the early 1760s. Over the last three centuries, the RPC has experienced significant coastal erosion with almost 32 m of shoreline lost (CBCL Limited 2010; Dunham 2014; Johnston 2001; Taylor et al. 2011). This ongoing erosion is directly impacting the stability of the RPC and has prompted the rescue excavation and relocation of these individuals before this land mass becomes inaccessible due to rising sea levels (see Fig. 1).

Since excavation of the RPC began in 2017, skeletal preservation has varied widely, ranging from complete skeletal preservation to no skeletal preservation within the excavated areas ($\sim 520 \text{ m}^2$) (Scott 2019a, 2019b, 2021, 2026a, 2026b, 2026c). This inconsistency in skeletal preservation has been partially attributed to variations in soil pH across the cemetery due to the varied uses of the site before it became a cemetery and the associated structures that were left behind (e.g., lime production and storage) (Scott and Fonzo 2019). While soil composition certainly plays a major role in skeletal preservation, it is not the only factor to consider. For instance, there have been a number of individuals excavated where soil composition and pH would suggest good preservation, but in actuality were visually quite poor and vice versa (Scott 2019a, 2019b, 2021, 2026a, 2026b, 2026c). Therefore, an exploration of additional factors and their impact on preservation, particularly at the molecular level, is warranted.

The measurement of molecular preservation in skeletal material at a site can provide a detailed, objective assessment of the environmental conditions (e.g., seawater drainage) that can affect those buried long before any visual signs appear. Because an incongruity between the visual presentation of the skeletal remains and their molecular preservation is evident (see Hughes and Scott 2023; Scott and Fonzo 2019), there is a need to explore factors beyond soil acidity that may be affecting skeletal preservation. Specifically, seawater needs to be explored as proximity to the ocean and constant exposure to seawater from the worsening coastal erosion is bound to impact the skeletal remains of those at the RPC. To explore the impact of seawater on molecular skeletal preservation, this study uses Fourier-Transform Infrared Spectroscopy (FTIR) to test the hypothesis that burials closer to the coastline at Rochefort Point will exhibit decreased molecular and, potentially, visual skeletal preservation. This will also test our hypothesis that simple visual assessments of archaeological remains are an imperfect measure for predicting molecular preservation and do not lend the same statistical power to evaluate preservation trends at archaeological sites. This molecular data in tandem with previously explored pH levels will not only assist with excavation planning for the RPC but also provide an efficiency framework for other bioarchaeological rescue excavations.

1.2 Diagenetic alteration and the environmental impact on bone

When visually assessing skeletal preservation, we often note the presence, strength, brittleness, and recognizability of skeletal elements (Williams et al. 2025). However, when assessing the molecular preservation of bone, we typically only consider the integrity of the original chemical structures that comprise the skeletal material (Smith et al. 2023). Bone is a complex tissue, comprised of both mineralized inorganic components (i.e., calcium hydroxyapatite) and organic components—predominantly type-1 collagen (Boaks et al. 2014; Keenan 2016). The variety of calcium hydroxyapatite found in bone, referred to as bioapatite, incorporates carbonate into its chemical structure, creating a disordered crystal structure that provides greater structural integrity than standard calcium hydroxyapatite (Dobberstein et al. 2009; Tătar et al. 2014). The process by which bone is structurally and chemically altered by exposure to the burial environment is known as diagenetic alteration or diagenesis, and is caused in part by leaching; when water-soluble ions from the soil are absorbed and exchanged

with the bone's bioapatite (Keenan 2016). This causes a shift from the disorganized crystalline structure of bioapatite to a more ordered one, making the bone increasingly brittle and prone to damage and dissolution (France et al. 2020; Kendall et al. 2018; Nielsen-Marsh et al. 2000).

The rate and severity of diagenesis in bone depends heavily on extrinsic factors in the burial environment. For example, leaching as a chemical process is highly dependent on equilibrium, meaning that when skeletal material enters the depositional environment, there will be an initial surge of diagenesis as the skeletal material exchanges ions with the environment. However, if external factors (e.g., soil hydrology) remain stable, the decline in skeletal preservation will eventually level off (Hedges 2002; Nielsen-Marsh et al. 2000). However, if these extrinsic factors are in flux (e.g., frequent storm surges, changing tidal flows, sea-level rise), the chemical reaction between bone and the burial environment is accelerated (Peris 2022). Both components of bone (collagen and bioapatite) degrade more quickly in saltwater, especially bioapatite, since it is an active ion exchanger with sodium and chloride ions. Because of the sodium and chloride content in seawater, bone will leach more quickly than in fresh water or in an arid burial environment (Nielsen-Marsh et al. 2000; Stiner et al. 2001; Trujillo-Mederos et al. 2012).

While diagenetic changes in bone can contribute to the physical changes associated with poor visual preservation, (France et al. 2020; Kendall et al. 2018; Nielsen-Marsh et al. 2000); it is important to clarify that although the decline in visual and molecular preservation of skeletal material often occurs concurrently, they are distinct processes that can be independent from each other. For example, external physical changes to the skeleton (e.g., scavenging) would have very little to do with the molecular integrity of the skeletal material, and similarly, if certain chemicals are present in the soil (e.g., excess carbonate), skeletal preservation will be visually good even if the chemical structure of the skeleton is poor (France et al. 2020; Williams et al. 2025). While compromised molecular preservation often predicts poor visual preservation, a growing number of researchers caution against using visual assessment to predict the state of molecular preservation in skeletal material (see Chesson et al. 2021; Hollund et al. 2013; Smith et al. 2023).

1.3 Fourier-transform infrared (FTIR) spectroscopy

FTIR spectroscopy with attenuated total reflection (ATR) is a commonly used analytical instrument for the characterization and quantification of heterogeneous materials (Tiernan et al. 2020). Samples are exposed to a beam of infrared radiation, in which chemical functional groups (e.g., carbonate or phosphate) characteristically absorb or transmit the radiation within a specific range of wavenumbers (cm^{-1}) (Gong et al. 2024; Kazarian and Chan 2006). When measured in absorbance, the peaks of an infrared spectrum can serve as a proxy for the concentration of the chemical functional group that absorbs in that wavenumber range (Gong et al. 2024).

Well-established metrics have been developed to measure the severity of diagenesis occurring in bone, based on the spectra of unaltered bone and the predictable way in which bone spectra change as diagenesis occurs (see France et al. 2020; Shemesh 1990; Smith et al. 2023). Among the various metrics

developed, the infrared-splitting-factor (IRSF), carbonate-phosphate ratio (C/P), amide-I-phosphate ratio (Aml/P), and the types A (API) and B (BPI) carbonate-phosphate indices are among the most frequently used (see Table 1).

Table 1

Calculations utilized to assess diagenetic alteration along with their ranges of Well-Preserved archaeological remains.

Measurement	How Diagenesis is Measured	FTIR Calculation	Approximate Wavenumber Calculation	Ranges for "Well-to-Acceptably Preserved"	Reference
Infrared Splitting Factor (IRSF)	Measures the mean length between the ν_4 phosphate peaks as a proxy for crystal organization. Increased crystal order means the bone has experienced increased recrystallization.	(Phosphate ν_4 +Phosphate ν_4) / (The valley separating them)	$(605\text{cm}^{-1} + 565\text{cm}^{-1}) / (590\text{cm}^{-1})$	2.5-4.0 (France et al., 2020)	(Beasley et al. 2014; France et al. 2020; Shemesh 1990; Smith et al. 2023; Tătar et al. 2014; Termine and Posner 1966; Weiner and Bar-Yosef 1990)
Carbonate-Phosphate ratio (C/P)	Measures the ratio between bone carbonate and phosphate where deviations from the known ratio suggest degree of diagenesis.	(type-B Carbonate) / (Phosphate ν_3)	$(1415\text{cm}^{-1}) / (1015\text{cm}^{-1})$	0.05–0.32 (France et al., 2020)	(Boskey et al. 2016; Grunenwald et al. 2014; Iacumin et al. 2022; Isaksson et al. 2010; Mohd Noor et al. 2020; Smith et al. 2023; Wright and Schwarcz 1996)
Amide I - Phosphate Ratio (Aml/P)	Measures the ratio of collagen's amide group in relation to ν_3 phosphate as a determinant of bone's collagen preservation.	(Amide I group) / (Phosphate ν_3)	$(1650\text{cm}^{-1}) / (1415\text{cm}^{-1})$	< 0.08 (Hollund et al., 2013)	(Hollund et al. 2013; Kontopoulos et al. 2020; Scott et al. 2020; Tamara et al. 2022; Trueman et al. 2004)

Measurement	How Diagenesis is Measured	FTIR Calculation	Approximate Wavenumber Calculation	Ranges for "Well-to-Acceptably Preserved"	Reference
type-A Carbonate-Phosphate Index (API)	Measures the ratio between type-A carbonate and bone's phosphate where deviations from the known ratio suggest degree of diagenesis.	(type-A Carbonate lesser peak) / (Phosphate ν_4)	$(1540\text{cm}^{-1}) / (600\text{cm}^{-1})$	0.04–0.2 (France et al., 2020)	(Snoeck et al. 2014; Sponheimer and Lee-Thorp 1999; Tamara et al. 2022)
type-B Carbonate-Phosphate Index (BPI)	Measures the ratio between type-B carbonate and bone's phosphate where deviations from the known ratio suggest degree of diagenesis.	(type-B Carbonate) / (Phosphate ν_4)	$(1415\text{cm}^{-1}) / (600\text{cm}^{-1})$	0.1–0.65 (France et al., 2020)	(France et al. 2020; Snoeck et al. 2014; Tamara et al. 2022)

2. MATERIALS AND METHODS

2.1 Sample

Fifty-one individuals were selected from the RPC for this diagenesis analysis. The femur was selected as a universal element for its density and thus its resistance to degradation, allowing for the maximum number of individuals to be considered in this study (McEwan et al. 2004). It has been previously described that deeper burials contribute to better molecular preservation as they experience less exposure to fluctuating conditions above ground, and the opposite applies to shallower burials (see Kravanja et al. 2025; Rai et al. 2022). With this in mind, and to capture the most typical burials at the RPC, only individuals buried within two standard deviations of the mean burial depth (22.40 cm to 86.77 cm) were analyzed to better understand the site-wide impact of seawater drainage while minimizing confounding factors (Wescott 2018).

2.2 Visual Assessment of Skeletal Remains

As previously described, there was anecdotal incongruity between the visual and molecular preservation of certain individuals from RPC (Hughes and Scott 2023; Kaufman 2024; Scott 2019a, 2019b, 2021, 2026b, 2026c, 2026a; Scott and Fonzo 2019). To explore this phenomenon and investigate how measuring molecular preservation provides a more detailed understanding of archaeological site patterns, a visual skeletal preservation score was assigned to each individual on a scale from 1 to 3, adapted from the visual assessment score system used by Scott and Fonzo (2019), which was originally adapted from Gordon and Buikstra (1981) (see Table 2).

Table 2

Skeletal preservation classification criteria adapted for this study from Scott and Fonzo (2019; page 206), originally adapted from Gordon and Buikstra (1981)

Visual Score	Criteria
1	Skeletal elements are whole and undamaged; no evidence of postmortem cortical destruction; all elements are identifiable.
2	Skeletal elements are fragmentary but can be reconstructed; mild to moderate postmortem cortical destruction: all elements are identifiable.
3	Skeletal elements are extremely fragmentary; severe postmortem cortical destruction; some elements may not be recognizable.

The visual assessment score system used by Scott and Fonzo (2019) comprised a scale of 1 to 4; with a score of 4 indicating that “skeletal elements are missing, do not hold shape when removed from soil; tooth crowns may be present but are fragile; most elements are absent/unrecognizable” (see Scott and Fonzo, 2019; page 206). Given the selection criteria of this study (i.e., a present and identifiable femur), a score of 4 was not applicable to the samples included and was therefore removed from the scoring classification.

2.3 Sampling procedure

Using a Dremel rotary tool (model 300) with a rounded carbide burr, the outer cortical surface of the posterior-lateral portion of the femur was removed to expose approximately 3 cm² of inner cortical bone that was free from soil or debris. Approximately 5 mg of the inner cortical bone was then collected from each individual. The carbide burr was sanitized in an ethanol flame both before and after each sample was collected. The powdered samples were then passed through a series of sieves to ensure a uniform particle size across individuals (20–63 microns) (Udvardi et al. 2017). Sieves were sanitized with ethanol before and after each sample was collected.

2.4 Spatial Measurement of Burials

To measure the proximity of each burial to the northern shore of the RPC, the latitude degrees of the selected burials were determined from GIS data collected at the time of excavation, using a Topcon ES-Series total station. These degrees of latitude were compared with both visual assessments of skeletal preservation and FTIR diagenetic metrics to determine whether any correlation exists between the visual and/or molecular preservation of the skeleton and the proximity of the burial to the northern shore. Beyond exploring a correlation between skeletal preservation and burial proximity to the northern shore, we also wanted to evaluate whether these patterns meaningfully differentiated. To do this, we took the total south-to-north axis of the included individuals, spanning 2.54×10^{-4} degrees of latitude, and divided it equally into four latitude groups. To facilitate statistical testing by geographic location, all burials were assigned to one of these four groups.

2.5 FTIR-ATR

Infrared spectra were generated using a Bruker Alpha II FTIR spectrometer equipped with an attenuated total reflection (ATR) accessory that compressed the samples at approximately 100 psi. Twenty-four spectra scans were taken per sample with a resolution of 8 cm^{-1} and analyzed using Opus FTIR software, capturing the absorbance readings for wavenumbers between 450 and $4,000\text{ cm}^{-1}$. An automatic baseline correction for peak height was applied within the Opus software to every collected spectrum. Based on the spectra collected for each femoral sample, the extent of diagenesis was determined using five commonly used and established metrics (see Table 1). While each of these metrics, except for Aml/P, has an upper and lower threshold indicating whether skeletal material is within a range of well-to-acceptably preserved (see Table 1) and as values decrease, molecular preservation typically declines. The exception is IRSF, where a higher IRSF score indicates more extreme diagenesis. Each metric was calculated using the absolute peaks or troughs within the range of each relevant functional group, in accordance with the recommendations of Smith and colleagues (2023).

3. RESULTS

A total of 51 individuals qualified under the selection criteria, with all collected data presented in Table 3.

Table 3
A summary of all preservation (visual and molecular) data collected

Burial #	Latitude group	Visual preservation score	C/P	IRSF	Aml/P	API	BPI
14/2017	3	2	0.412	2.169	0.382	0.223	0.308
18/2017	3	2	0.213	3.546	0.152	0.242	0.452
19/2017	3	2	0.338	2.896	0.297	0.342	0.551
22/2017	3	2	0.211	3.506	0.225	0.301	0.549
24/2017	2	2	0.186	3.500	0.115	0.155	0.355
26/2017	1	1	0.222	3.312	0.137	0.170	0.409
30/2018	1	2	0.206	3.493	0.174	0.242	0.426
33/2018	1	2	0.510	2.754	0.481	0.682	0.885
36/2018	1	3	0.496	2.886	0.487	0.705	0.944
38A/2018	1	3	0.185	3.498	0.146	0.349	0.401
65/2019	1	2	0.240	3.476	0.224	0.686	0.527
66/2019	1	3	0.484	2.821	0.451	0.396	0.908
71/2019	1	3	0.299	3.261	0.281	0.403	0.606
72/2019	3	1	0.311	3.291	0.288	0.479	0.614
75/2019	2	2	0.363	3.065	0.317	0.374	0.698
76B/2019	2	3	0.243	3.420	0.216	0.433	0.504
76C/2019	2	3	0.287	3.361	0.279	0.405	0.568
77/2019	2	1	0.332	3.296	0.310	0.409	0.645
79/2019	2	3	0.311	3.393	0.317	0.279	0.632
81/2019	1	3	0.301	3.221	0.275	0.463	0.585
82/2019	1	2	0.234	3.440	0.196	0.440	0.455
84/2019	2	1	0.306	3.205	0.270	0.409	0.582
85/2019	2	1	0.795	2.352	0.767	1.121	1.321
87/2019	1	1	0.342	3.243	0.335	0.555	0.683
90/2019	3	1	0.355	3.276	0.320	0.241	0.680
91/2019	2	3	0.295	3.331	0.292	0.310	0.581

Burial #	Latitude group	Visual preservation score	C/P	IRSF	Aml/P	API	BPI
94/2019	2	3	0.416	2.978	0.377	0.200	0.774
97/2019	2	3	0.211	3.495	0.177	0.262	0.421
100/2019	2	3	0.247	3.381	0.208	0.526	0.506
101/2021	2	2	0.176	3.640	0.162	0.403	0.368
104/2021	3	2	0.220	3.379	0.185	0.405	0.447
105/2021	3	2	0.382	3.008	0.364	0.373	0.722
106/2021	4	2	0.289	3.203	0.272	0.471	0.602
111/2021	3	2	0.326	3.146	0.284	0.150	0.616
112/2021	3	3	0.296	3.335	0.280	0.187	0.570
113/2021	4	3	0.356	3.176	0.361	0.305	0.690
115/2021	3	3	0.162	3.658	0.137	0.208	0.329
116/2021	4	3	0.174	3.657	0.152	0.160	0.373
121/2021	3	3	0.239	3.438	0.255	0.126	0.492
125/2022	4	3	0.141	3.953	0.126	0.177	0.435
132/2022	4	2	0.162	3.672	0.148	0.134	0.317
135/2022	4	3	0.146	3.767	0.105	0.191	0.320
136/2022	3	3	0.157	3.885	0.129	0.219	0.385
140/2022	3	3	0.150	3.854	0.103	0.165	0.322
141/2022	4	3	0.180	3.540	0.152	0.153	0.365
144/2022	4	3	0.195	3.720	0.197	0.187	0.436
145/2022	3	3	0.161	3.834	0.115	0.216	0.353
151/2022	4	3	0.165	3.667	0.130	0.168	0.340
152/2022	4	3	0.197	3.556	0.155	0.216	0.411
159/2022	4	3	0.206	3.497	0.181	0.315	0.427
162/2022	4	3	0.150	3.827	0.119	0.343	0.345

3.1 Visual Assessment of Remains

For the 51 individuals included in this study, the average visual skeletal preservation score was 2.4/3 with a median score of 3/3 (see Table 3). There was no statistical significance when comparing the degrees of latitude of the samples per visual score (H: 3.31, $p = 0.191$) or the prevalence of a particular visual score within each of the four latitude groups (Fisher Exact Test: 6.42, $p = 0.38$) (see Fig. 2). However, there was greater variability in the degrees of latitude as visual preservation worsened (i.e., visual score of 1 versus 3) and no well-preserved individuals (i.e., visual score 1) in the latitude group closest to the northern coastline (i.e., latitude group 4) (see Fig. 2).

3.2 Molecular Preservation of Remains

When we look at the molecular integrity of the samples included in this study, we see a variable range in the FTIR-based metrics [IRSF: 2.17–3.95; C/P: 0.14–0.80; Aml/P: 0.10–0.77; API: 0.13–1.12; BPI: 0.31–1.32]. The site-wide averages, with the exception of API, are considered acceptable or well preserved archaeological remains [IRSF: 3.36; C/P: 0.27; Aml/P: 0.25; API: 0.33; BPI: 0.53] (see Tables 1 and 3) (Colmenares-Prado et al. 2025; France et al. 2020; Hollund et al. 2013). A Spearman rank correlation test was conducted to assess the correlation between the collected FTIR metrics for each burial and the northward progression in degrees of latitude toward the RPC coastline. This test showed a significant correlation between worsening preservation (note: IRSF, generally as it increases, denotes worse preservation in this context [see France et al., 2020]) with proximity to the northern shoreline (see Fig. 3).

Through a Kruskal-Wallis test, there was a significant relationship between the latitude groups and each FTIR metric of diagenesis ([IRSF; H: 11.28 P: 0.010], [C/P; H: 12.05 P: 0.007], [Aml/P; H: 8.02 P: 0.046], [API; H: 15.65 P: 0.001], [BPI; H: 9.62 P: 0.022]) (see Fig. 4).

This result shows that burial position, especially as it shifts from south to north with increasing progression toward the coastline, is linked to consistent differences in molecular preservation across all five metrics. Additionally, as the space between latitude groups increased (e.g., latitude group 1 vs. latitude group 4, compared to latitude group 1 vs. latitude group 2), a significant difference was found between these groups when tested using a Dunn's Post-Hoc test and Bonferroni correction. The only exception was that of Aml/P (see Figure and Table 4). These comparisons further corroborate the results of the Spearman's Rank correlation tests, which demonstrates that our observations are not only a detectable continuous trend but also a discrete, group-level phenomena that becomes more pronounced with increasing spatial separation.

Table 4

P values of Dunn's post hoc test per comparison of the south-to-north group to each employed FTIR-based metric of diagenetic alteration. Significant p-values are bolded.

Latitude Group Comparison	IRSF	C/P	Aml/P	API	BPI
1 vs. 2	P: 1.00	P: 1.00	P: 1.00	P: 1.00	P: 1.00
1 vs. 3	P: 0.99	P: 1.00	P: 1.00	P: 0.02	P: 0.76
1 vs. 4	P: 0.012	P: 0.015	P: 0.99	P: 0.005	P: 0.047
2 vs. 3	P: 1.00	P: 1.00	P: 1.00	P: 0.20	P: 0.97
2 vs. 4	P: 0.04	P: 0.017	P: 0.08	P: 0.06	P: 0.06
3 vs. 4	P: 0.34	P: 0.22	P: 0.57	P: 1.00	P: 1.00

Using the visual assessment score as the grouping criterion, rather than the latitude group, there was only a statistically significant association between visual preservation and the C/P and IRSF FTIR metrics when evaluated with a Kruskal-Wallis test. There was also very little variation in the collected FTIR metrics across the visual scores of skeletal preservation, meaning that we could only establish a limited statistical connection between visual and molecular preservation observed at the RPC (see Fig. 5).

4. DISCUSSION

Given the significant correlations and association testing across multiple FTIR-based metrics of molecular preservation in the studied individuals and their physical location within the RPC, it can be confidently stated that seawater drainage is a major contributing factor to the wide range of preservation observed at the RPC. The northern part of the RPC located at the base of a small hill is closest to the ocean and is consistently exposed to seawater. Additionally, this northern edge would be directly affected by any storm surge runoff water on Rochefort Point. This increased exposure to seawater is a likely contributor to the observed worsening molecular preservation, and a similar trend was observed visually in the more northern burials. The impact of leaching and water exposure, especially saltwater, is well documented for accelerating diagenesis, as the inorganic component of bone rapidly undergoes ion exchange and degrades under these conditions (Hedges and Millard 1995; Kendall et al. 2018; Martin 2021; Trujillo-Mederos et al. 2012).

The soil at Louisbourg is characterized as sandy-loam (Cann et al. 1963; CBCL Limited 2010), which means it is highly permeable, making it especially susceptible to mineral leaching (Vinten et al. 1994). Hydrology, which refers to how water is retained or flows through soil, is a key indicator of the extent of leaching in this environment. This is seen in two patterns: diffusive water patterns where minimal water movement has a gentle effect on bone, compared to recharge water patterns that fluctuate between wet and dry cycles and is more disruptive and damaging to bone (Hedges 2002; Hedges and Millard 1995; Nielsen-Marsh et al. 2000). Sandy-loam, like that found in the RPC, has been shown to be significantly

affected by recharge patterns in soil hydrology (Zomlot et al. 2015), which, in this case, leads to increased disruption of the depositional environment creating increased rates of diagenesis. Given this knowledge, it is unsurprising to see that the burials closest to the ocean and with increased water drainage exhibit the most extreme diagenesis amongst the RPC burials studied.

Because the visual preservation scores suggested a higher frequency of poorly preserved individuals as they approached the northern shore of the RPC (see Fig. 2), it was surprising that these findings were not statistically significant, unlike our FTIR-based metrics of diagenesis (see Figs. 3 and 4). This may be due, in part, to the limited statistical power that results from dividing data into smaller subsamples for analysis (Serdar et al. 2021). The most significant limitation of this study and a likely contributor to our weaker correlations is sampling bias and only including individuals with a femur present. While this decision was made to build a robust and representative sample, the thick cortical bone could have biased this sample towards those that were already better preserved. For example, in the case of those with no bone (only teeth) remaining at the time of excavation were not included, and many of which were seen in the latitude group closest to the ocean (latitude group 4) (Scott 2019a, 2019b, 2021, 2026c, 2026a, 2026b). This is also where we see the worst overall visual preservation as well as the fewest candidates available for inclusion in this study (i.e., had a present and identifiable femur), which is unsurprising given the results of this study due to their proximity to the coastline (see Fig. 2) (Scott 2019a, 2019b, 2021, 2026c, 2026a, 2026b). As previously discussed, visual preservation is not necessarily analogous to molecular skeletal preservation. However, because they often correlate (see Smith et al., 2023), selecting a skeletal element more likely to physically preserve would still likely skew molecular measurements of diagenesis towards indicating a better overall state of preservation. This sampling bias toward physically surviving elements likely compressed the range of visual preservation scores available for this analysis, which may explain why visual assessment failed to establish a statistically significant trend as burials approached the northern shore. The FTIR-based measurements of diagenesis employed in this study, however, were still able to detect significant diagenetic trends within this same, skewed sampling, further demonstrating that molecular analysis captures the granularity of preservation trends that visual assessment alone cannot.

Additionally, the fact that our visual preservation scores did not statistically associate with most of our FTIR-based metrics for measuring diagenesis (see Fig. 4) also demonstrates that the visual assessment of skeletal remains is an imprecise and frequently inaccurate predictor of the molecular integrity of bone (Beasley et al. 2014; France et al. 2020; Smith et al. 2023). Researchers, such as Bell and Jones (1991) and Hollund and colleagues (2017), have long raised a similar concern that the visual presentation of bone is an imperfect predictor of the molecular state or the degree of diagenesis. This was something we observed firsthand: the visual preservation of a burial was concealing the environmental impacts on the individual at the molecular level in the case of Individual 85/2019. Individual 85/2019 is a 12–14-year-old non-adult female (see Scott 2021) located in our latitude group 2, with excellent visual preservation, which was scored at the highest visual preservation score of 1. However, this individual also showed the highest, or, in the case of IRSF, the second-lowest values out of our entire sample,

indicating a high degree of diagenetic alteration, which puts them well outside the established ranges of what are considered well-preserved archaeological remains (see Tables 1 and 3, Fig. 6).

The most typical diagenetic process observed in skeletal material is the complete dissolution of bioapatite and its replacement with environmental mineral components, as reflected by high IRSF scores and low values for our other four metrics (France et al. 2020; Smith et al. 2023). However, in the case of Individual 85/2019, who was buried in close proximity to a hearth, the extraordinarily high C/P metrics indicate that the excess carbonate from their burial environment has infiltrated the chemical structure of their skeletal material (France et al. 2020; Scott 2021; Scott and Fonzo 2019), which can be worsened by the established fact that this type of chemical exchange is statistically higher among non-adults than their adult counterparts (see Leskovar et al. 2024). Because their individual FTIR-based diagenesis metrics were notable outliers to the rest of the cohort, it can be confidently stated that the skeletal material from Individual 85/2019 was chemically compromised despite appearing in excellent visual condition. Our inability to establish a statistical relationship between visual preservation and most metrics of molecular preservation (see Fig. 5), along with examples such as Individual 85/2019, further demonstrates that visual preservation may not be a reliable proxy for chemical integrity. This reinforces the value of integrating techniques such as FTIR into standard bioarchaeological assessments and archaeological rescue efforts.

5. CONCLUSION

While previous research at the RPC identified an unsurprising correlation between burial soil pH and skeletal preservation, this relationship could not fully explain all the patterns observed (e.g., poor visual preservation in alkaline soils and excellent preservation in acidic soils) (Scott 2019a, 2019b, 2026b, 2026a, 2026c, 2026a; Scott and Fonzo 2019). Additionally, many of the same individuals had visual and molecular scores that were incongruent with what we would expect (Hughes and Scott 2023; Scott and Fonzo 2019). Through a larger-scale evaluation of molecular preservation at the RPC, we observed that individuals closer to the northern shore of the cemetery exhibited poorer preservation, likely due to increased seawater drainage and subsequent dissolution of their skeletal bioapatite. By examining the limited correlation seen between our visual preservation assessment, and the trend of worsening preservation near the ocean, and FTIR-based diagenesis metrics, we demonstrate that chemical analysis offers a more detailed understanding of preservation trends than visual observation alone. Our findings also support the growing view among bioarchaeologists that visual assessments are an imperfect predictor of molecular preservation, as these two types of observations reflect related, but still distinct processes (see Hollund et al. 2017; Smith et al. 2023; Tătar et al. 2014; Williams et al. 2025).

When conducting rescue archaeology efforts in the wake of climate change-related coastal erosion, time is a precious and limited resource. Deciding where to excavate each season is vital for maximizing the recovery of individuals before the site is fully destroyed. Relying solely on visual assessments or soil pH does not carry enough statistical power to make confident predictions about the state of preservation for individuals buried throughout a site with varying environmental factors, namely, differing levels of

seawater exposure. Fourier-transform infrared spectroscopy is a relatively affordable and accessible analytical chemistry instrument. Portable FTIR models are also available, and the collection of FTIR spectra to calculate FTIR-based metrics of diagenetic alteration can be done quickly and with minimal destruction (i.e., ~ 2 mg of powdered bone). Regarding applicability to RPC, based on the trends observed in this study and derived from FTIR-based preservation metrics, we can conclude that continuing to excavate near the RPC shoreline, past latitude group 3 and latitude group 4, is likely to produce diminishing returns over time, which visual evaluation alone would not reveal with the same clarity. Although we plan to excavate all burials on Rochefort Point during the tenure of this project, studies such as this one enable a more robust strategy for selecting the best course of action each field season as the shoreline continues to recede and dictates how we successfully excavate this ever-changing historic landscape.

Declarations

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Figures



Figure 1

(Top) 1745 map of Rochefort Point showing the approximate location of the RPC. (Bottom) 2026 aerial photo of Rochefort Point with dotted lines indicating the approximate location of the RPC. This comparison highlights how the coastal erosion on Rochefort Point has closed the distance between the northern edge of the cemetery and the shoreline.

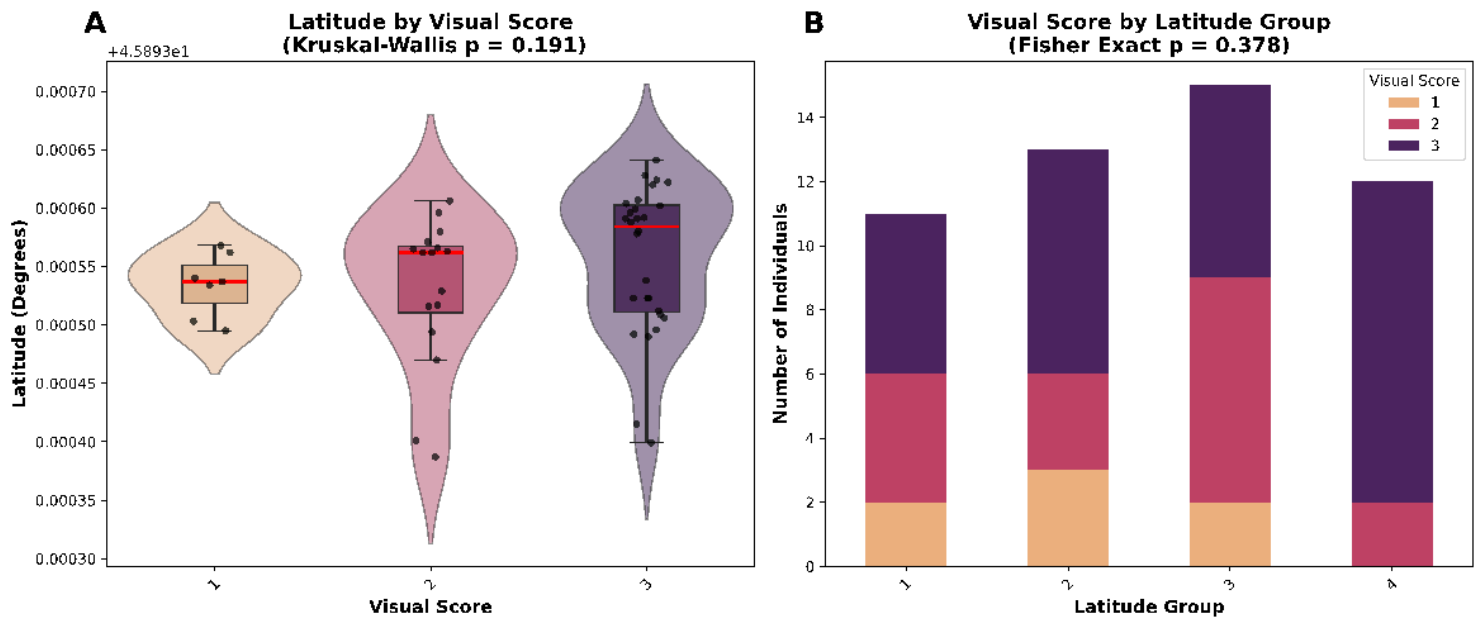


Figure 2

(A) Violin plot showing the distribution of degrees of latitude per score of visual preservation for individuals buried at the RPC, and (B) bar plot showing the distribution of visual preservation scores per latitude group.

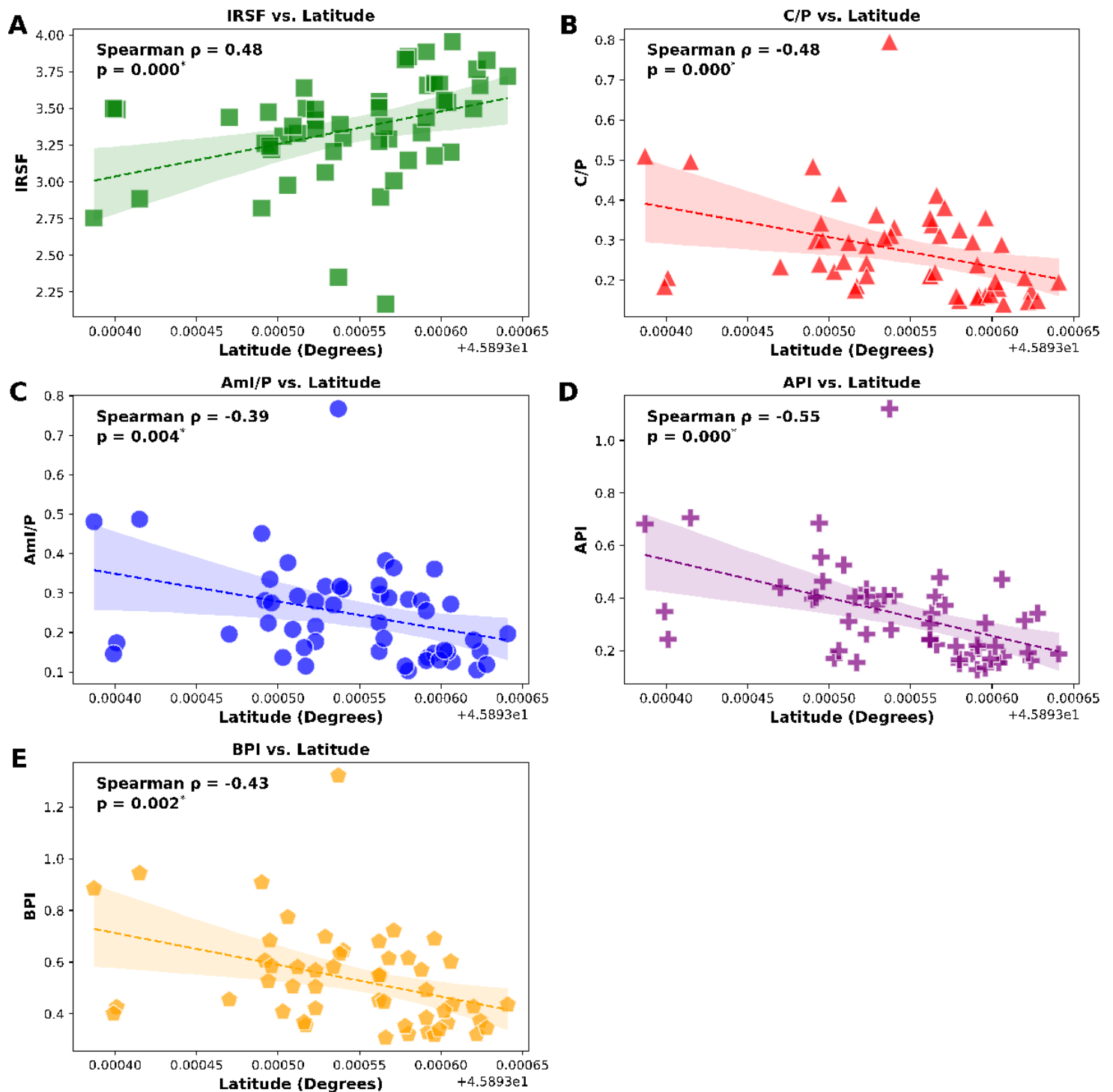


Figure 3

Spearman Rank Correlation Tests for IRSF (A), C/P (B), Aml/P (C), API (D), and BPI (E) against degrees of latitude. *Indicates significant P-Value within 95% confidence.

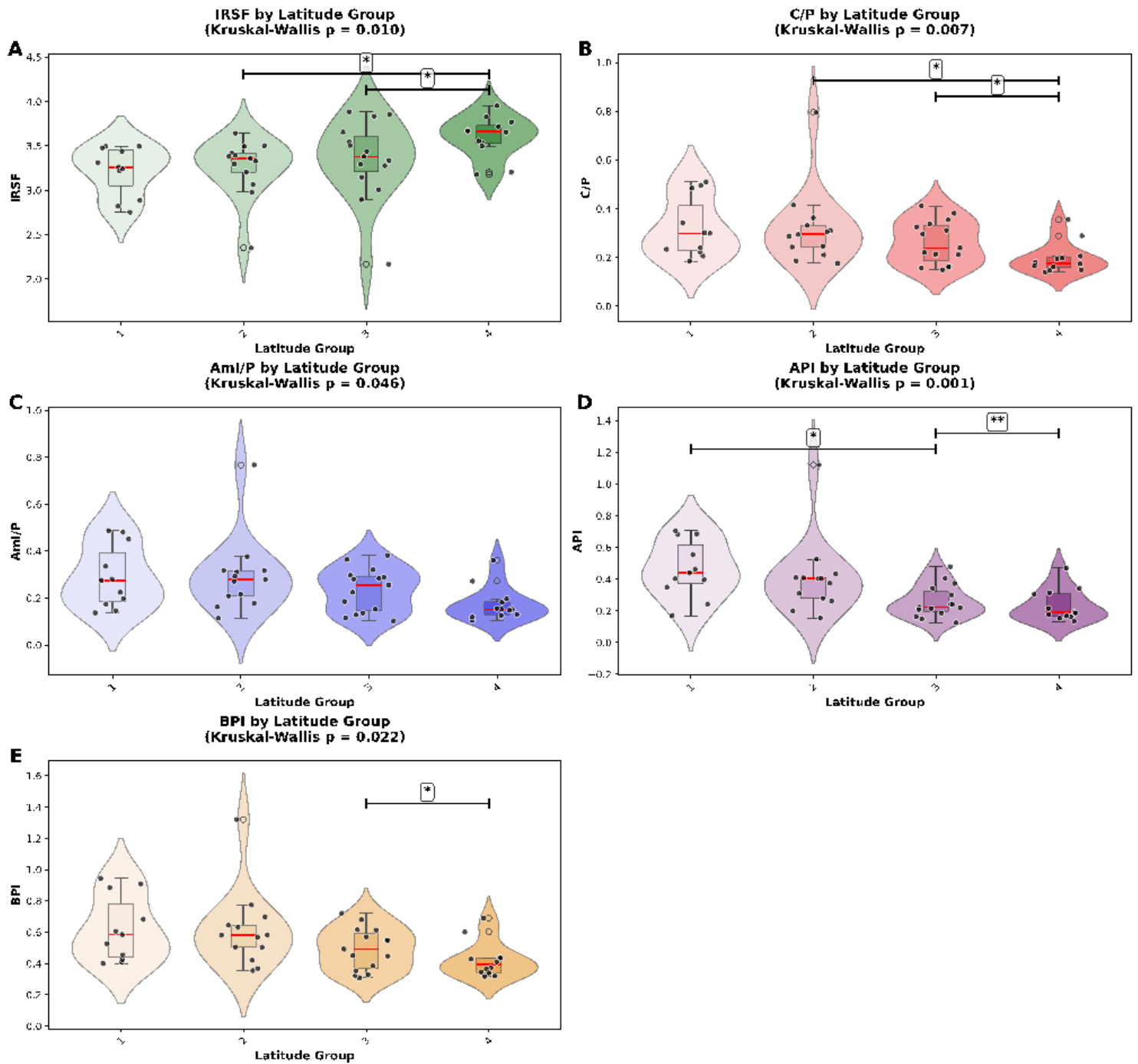


Figure 4

(A-E) Violin plots of IRSF (A), C/P (B), Aml/P (C), API (D), and BPI (E) in relation to their latitude groups. Each Violin plot shows the P-values of Kruskal-Wallis tests. *Indicates significance <0.05 , **indicates significance <0.001 of Dunn's Post-Hoc Test with Bonferroni correction on a 95% confidence interval.

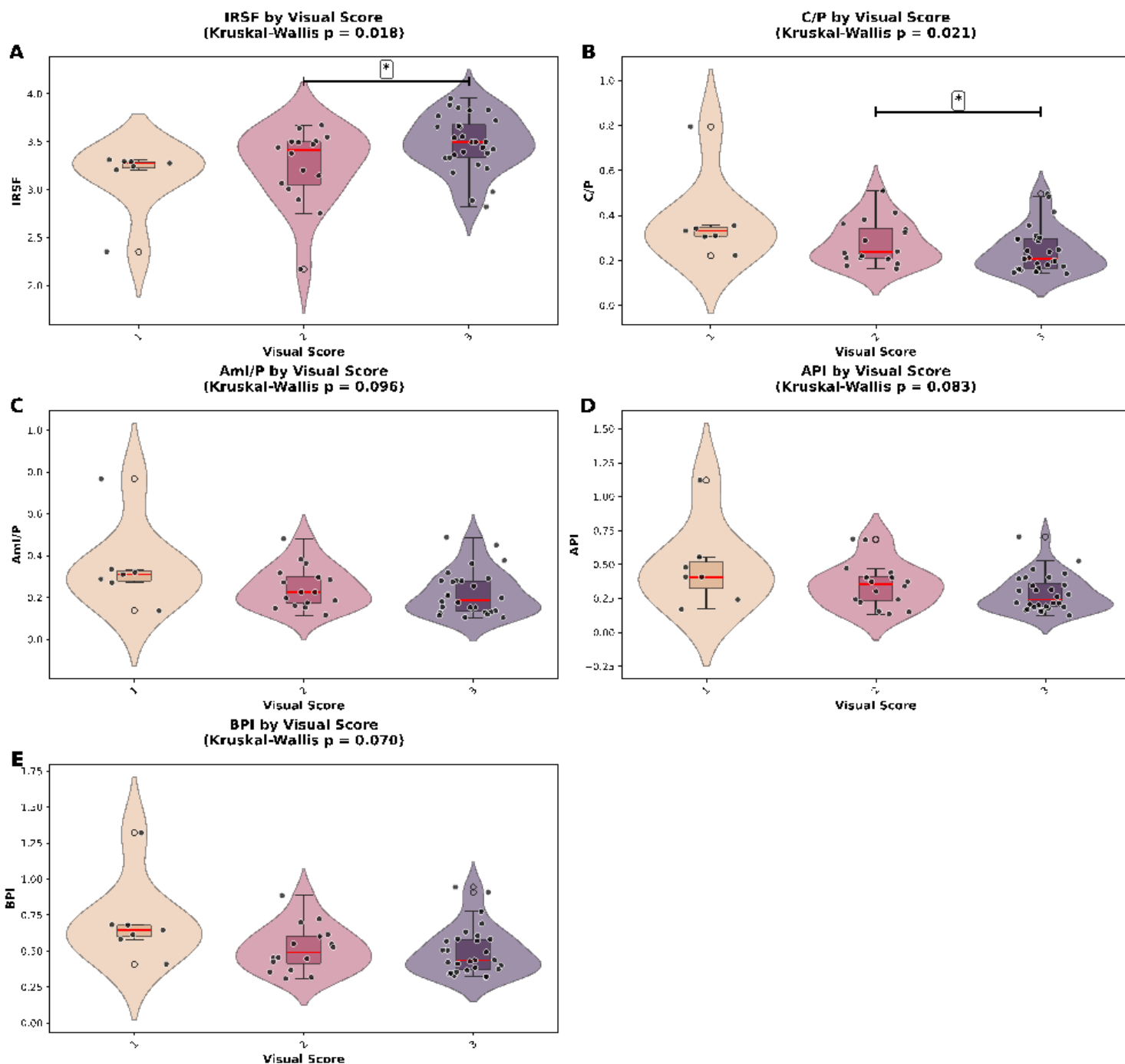


Figure 5

(A-E) Violin plots of IRSF (A), C/P (B), Aml/P (C), API (D), and BPI (E) in relation to their visual score of preservation groups. Each Violin plot shows the P-values of Kruskal-Wallis tests. *Indicates significance <0.05 , **indicates significance <0.001 of Dunn's Post-Hoc Test with Bonferroni correction on a 95% confidence interval.



Figure 6

Burial photo of Individual 85/2019. A sub-adult female aged 12-14 years old