

[Online Resources]

A Kingdom with Blackened Teeth 2,000 Years Ago: Tracing the Practice of Tooth Blackening in Ancient Vietnam

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Contents

Online Resource 1: Methodological protocols (Figs. S1–S5; Table S1).....	2
Online Resource 2: Scanning Electron Microscopy with Energy Dispersive X-ray Spectroscopy (SEM-EDS) results (Figs. S6–S13; Tables S2–S7).....	10
Online Resource 3: Portable X-ray Fluorescence (p-XRF) results (Table S8).....	17
Online Resource 4: Ethnographic review (Table S9)	18
Online Resource 5: Historical review (Table S10)	27
References.....	34

Abbreviations

ASCOGT, Aborigines Survey Committee, Office of the Governor-General of Taiwan
WTBHB, Writing Team of the Brief History of Blang

Online Resource 1: Methodological protocols (Figs. S1–S5; Table S1)

Data collection

This study conducted detailed macroscopic and microscopic examinations across all surfaces of the studied teeth. To minimize contamination prior to chemical analysis, the surfaces of the studied samples were cleaned with ethanol using Kimwipes, as ultrasonic cleaning was avoided due to the potential risk of damage. Multiple points were measured on each surface to allow comparison and to ensure the representativeness of the chemical data. From these observations, a set of key results from critical sampling areas (Fig. S1) was chosen for presentation and discussion. These areas were identified based on their visibly dark coloring and their limited exposure to burial environment influences observed at the micromorphological level.

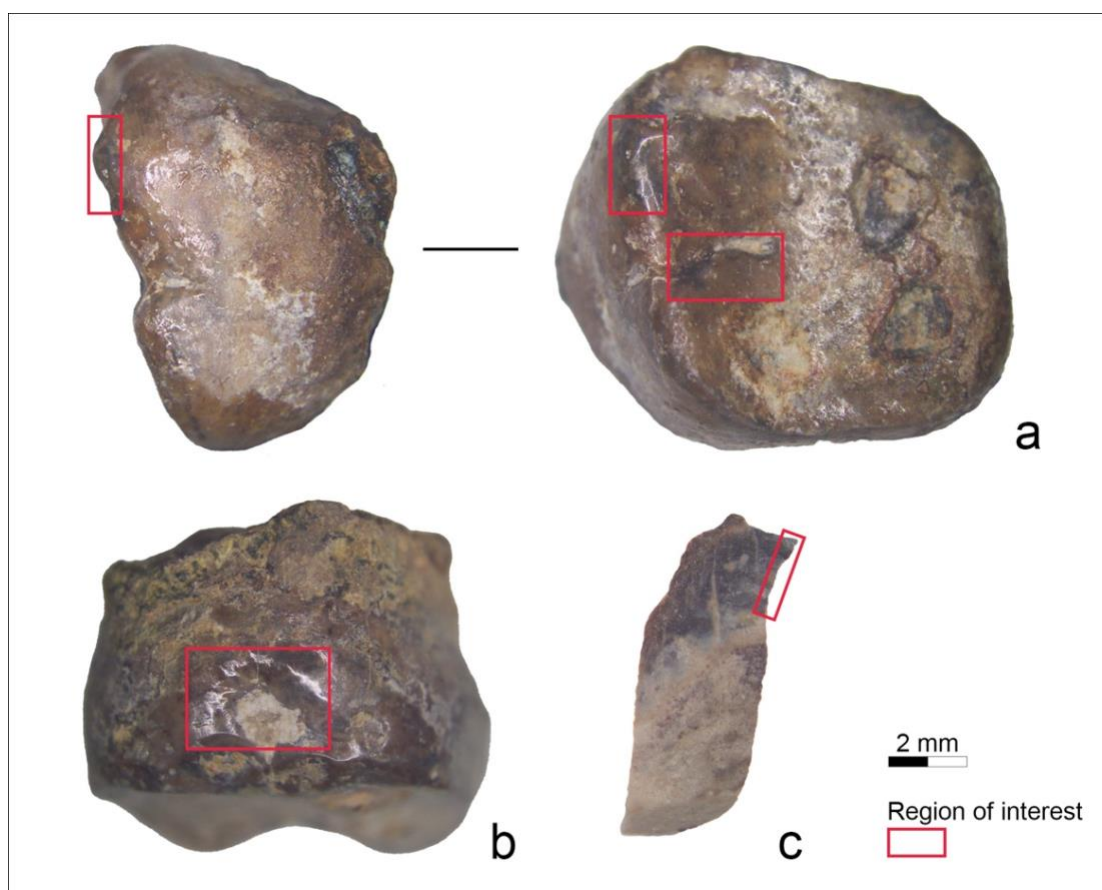


Fig. S1 Ancient tooth samples and the region of interest for technical analysis: **(a)** Sample #1 (04DXD1K2), dental cusp and occlusal surface; **(b)** Sample #2 (04DXUI2), buccal surface; **(c)** Sample #3 (04DXMPK), post-excavation fractured longitudinal section of enamel

Over hundreds to thousands of years of burial, the ancient samples underwent postmortem taphonomic alterations and accumulated varying amounts of adherent sediment, necessitating particular caution during Scanning Electron Microscopy with Energy Dispersive X-ray Spectroscopy (SEM-EDS) analysis to avoid misinterpretation.

The identification of dental enamel was achieved partly by comparison with a confirmed control sample and by recognizing characteristic morphological features. Backscattered electron (BSE) imaging revealed a consistent bright matrix with an orderly, fish-scale-patterned enamel prism or rod structure, observed in both modern and ancient samples (Fig. S2). This pattern may be influenced by the thin-sectioning angle; in longitudinal sections, the prisms align horizontally along their long axis (e.g., part of Fig. S2c & Fig. S11). Additionally, surface cracks are evident regardless of the age of the sample. On the other hand, EDS spectra and the relative elemental proportions provide more concrete evidence for the identification of dental enamel, with major signals corresponding to those of the modern sample (Fig. S3b-1 & S3d).

These criteria can be combined effectively. Analyses of areas where BSE imaging reveals a consistent, bright, and smooth matrix (Fig. S3a & S3c) often yield EDS profiles comparable to those of experimental enamel samples (Fig. S3b-1). In contrast, analyses of surfaces that appear mottled, darkened, or rich in impurities tend to produce spectra with greater variability and deviation (Fig. S3b-2 & S3b-3), likely resulting from environmental and taphonomic factors. Following this determination, the present study emphasizes the display and interpretation of alterations in the enamel proper where both criteria were satisfied, thereby reducing the likelihood of confusion with signals from diagenetic deposits.

Noted that the aluminum (Al) signal observed across samples is likely due to the aluminum foil used to stabilize the samples and reduce charging effects during the experiment, as this signal is absent where no foil was used.

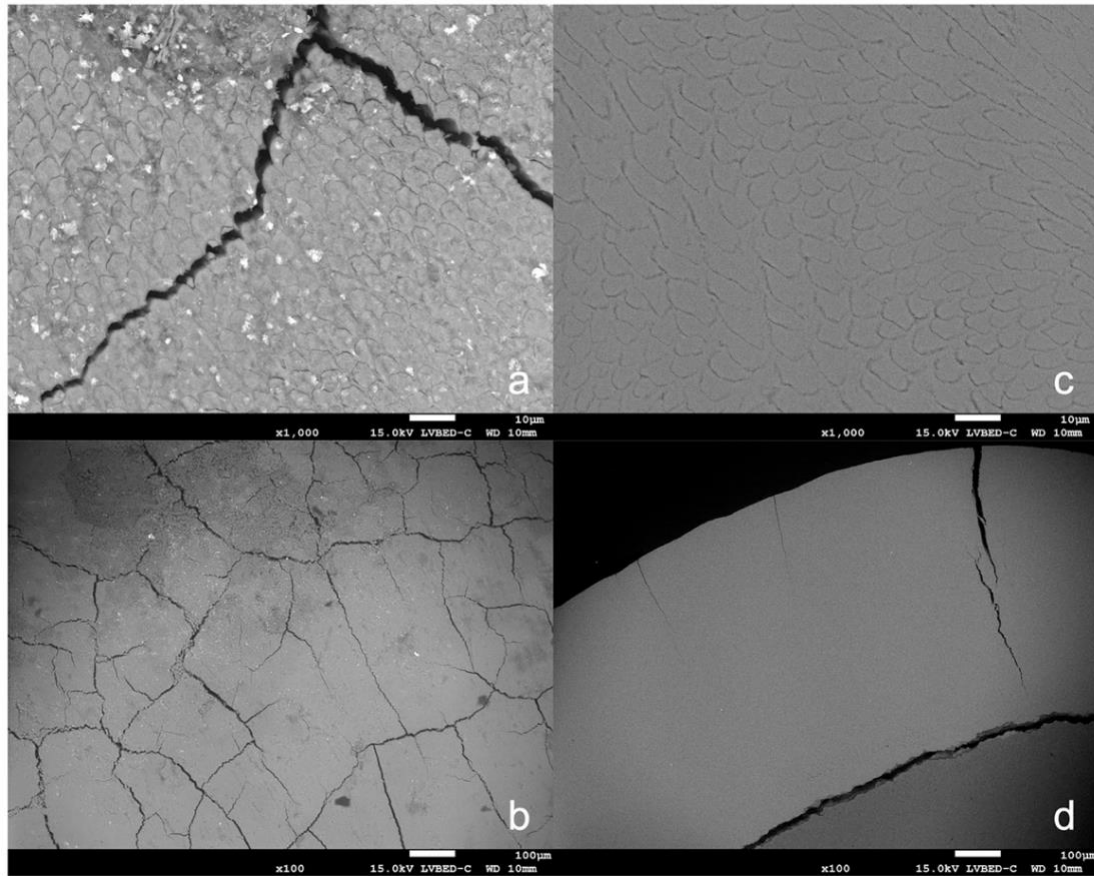


Fig. S2 BSE images of enamel microstructure at different magnifications ($\times 1000$ and $\times 100$, respectively).
(a–b) Sample #2; **(c–d)** experimental control sample

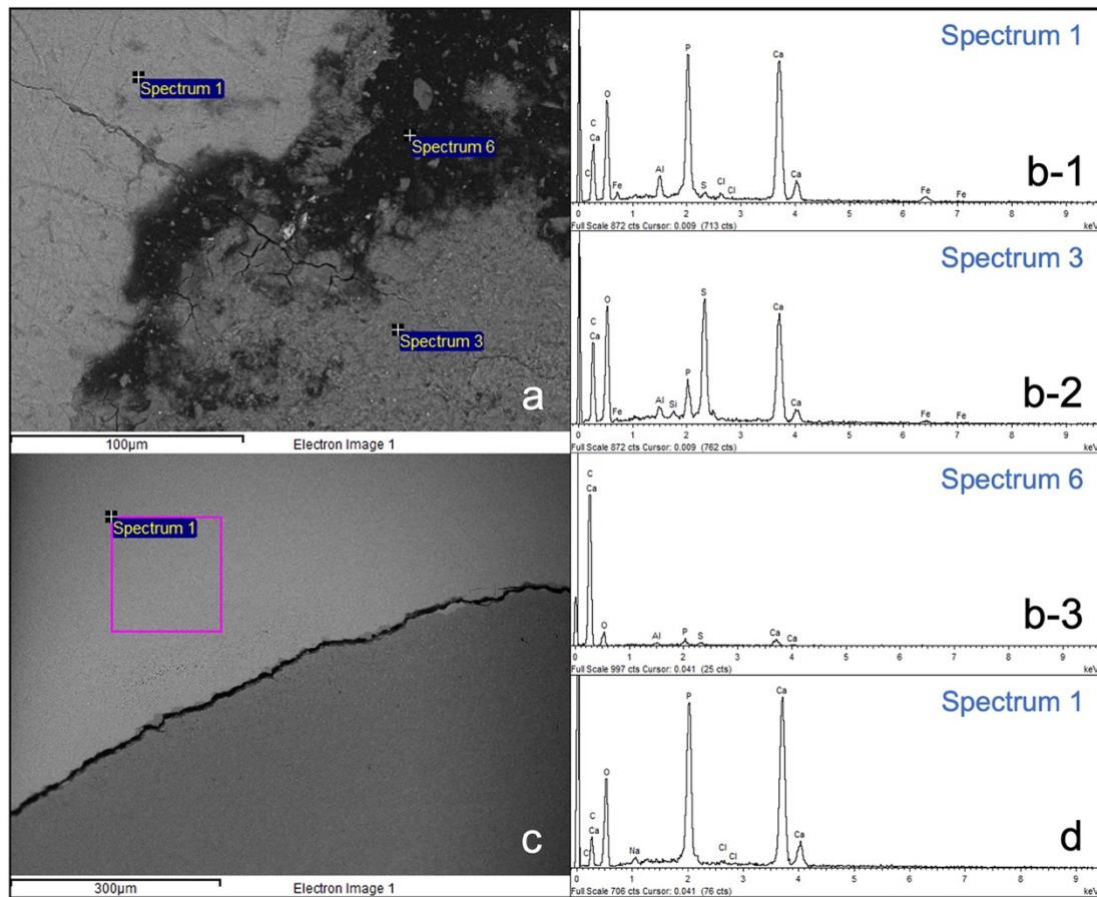


Fig. S3 Determination of enamel using BSE imaging and EDS spectra: **(a–b)** Sample #1; **(c–d)** experimental control sample

Experimental sample preparation

To provide realistic and reliable analytical controls, an experiment was performed on a modern animal tooth, aiming to characterize more clearly about the micromorphological changes and chemical alterations induced by the anthropogenic tooth blackening process. The experimental sample was prepared as a longitudinal thin section and examined using SEM-EDS both before and after the manual modification.

The dyeing procedure was informed by ethnographic analogies; however, due to difficulties in sourcing and importing exact ethnological materials, a commercial traditional iron-gall ink (Rohrer & Klingner Eisen Gallus Tinte Scabiosa) was used as a substitute. Although the ink is designed to be blackish purple, its chemical composition, iron salts and tannic acids, is comparable to that of traditional dyeing pastes. The sample was soaked in the ink for one, ten, and 60 minutes, followed by two cycles of ultrasonic cleaning in water. This process enabled observations of color

changes and a comparative spectral analysis before and after the formation of a darkened layer on the dental surface (Fig. S4).

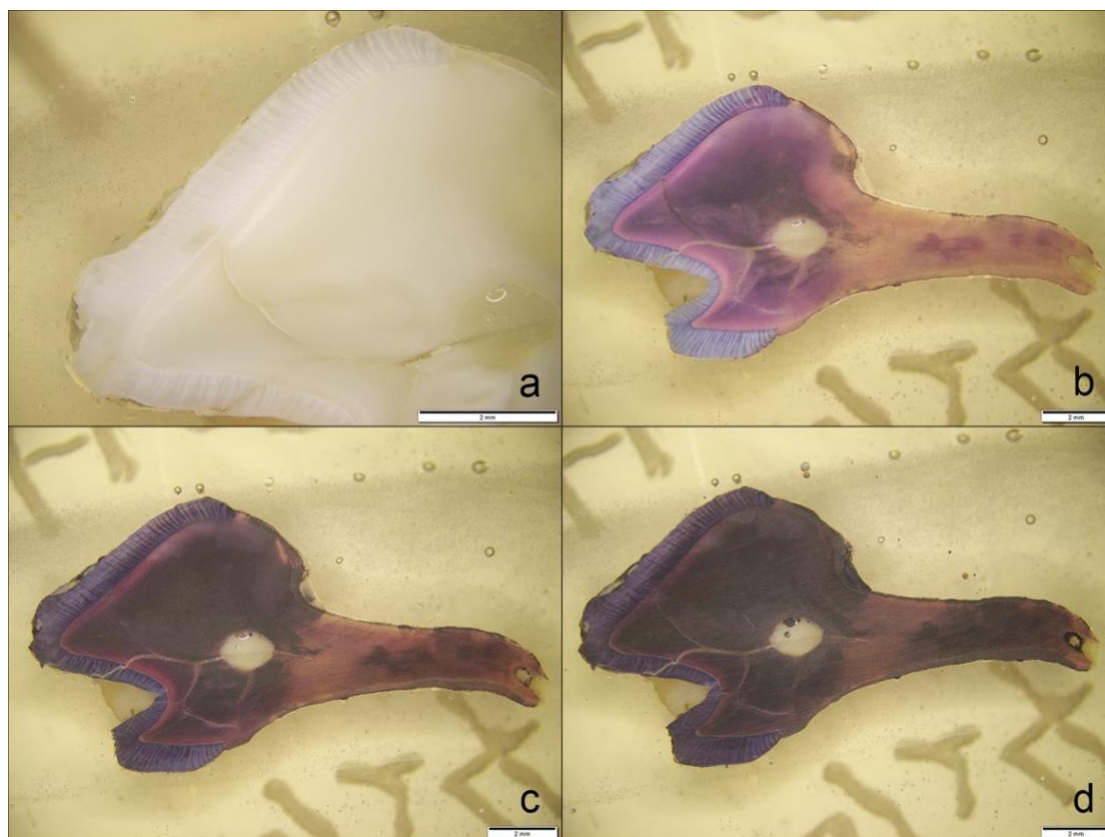


Fig. S4 Experimental sample at different stages of treatment: **(a)** unaltered; **(b)** soaked in iron-gall ink for one minute; **(c)** soaked in iron-gall ink for ten minutes; **(d)** soaked in iron-gall ink for 60 minutes

Scanning Electron Microscopy with Energy Dispersive X-ray Spectroscopy (SEM-EDS)
SEM-EDS analyses were conducted on three archaeological samples and one experimental modern sample, with multiple analysis points per sample. Surface observation and chemical identification were performed using a Field Emission Scanning Electron Microscope (FE-SEM, JEOL JSM-7100F-LV) equipped with Energy Dispersive spectrometer (EDS, Oxford Instruments X-max 80) at the EPMA Laboratory, Institute of Earth Sciences, Academia Sinica. The analyses were performed under low vacuum (50Pa), with an accelerating voltage of 15 kV, a probe current of 0.1 nA, and a working distance of 10 mm for 30 seconds.

Portable X-ray Fluorescence (p-XRF)

The p-XRF analysis was performed on three archaeological samples using a Bruker Tracer 5g machine (serial number 900G10034) at the Australian National University. The machine was set to 40 keV and 20 μ A, with the ‘yellow’ Ti 25 μ m: Al 300 μ m filter in place to optimize the detection of metals ranging from Ti to Ag and W to Bi in non-metallic samples (Drake et al. 2022). Each sample was analyzed three times, and the results were averaged to improve representativeness. The resulting spectra were normalized to Rh K α 1, and deconvolution was performed using Bruker’s ARTAX Spectra 8.0 software. The calibration utilized the Certified Reference Materials in Table S1, with certified values obtained from the GeoReM database (Jochum et al. 2005). The in-house standard ANU2000 was run for precision control, and a regression curve was generated to calibrate Fe concentration (Fig. S5).

Table S1 Certified Reference Materials utilized for p-XRF calibration

Standard	Description	Preparation
SiO2 100%	Pure silica (international)	Powder
45d	Soil (international)	Powder
BHVO-1	Hawaiian basalt (international)	Epoxy mounted
BHVO-2	Hawaiian basalt (international)	Pressed powder
BIR-1	Icelandic basalt (international)	Pressed powder
DNC-1 A	North Carolina diabase (international)	Pressed powder
DNC-1 B	North Carolina diabase (international)	Pressed powder
SBC-1	Brush Creek shale (international)	Pressed powder
SCO-1	Cody shale (international)	Pressed powder
SDC-1	Mica schist (international)	Pressed powder
SGR-1	Green River shale (international)	Pressed powder
STM-1	Nepheline syenite (international)	Pressed powder
SARM-1 NIM-G	Granite (international)	Pressed powder
JA-2	Japanese andesite (international)	Powder
JB-1b (1)	Japanese basalt (international)	Powder
JB-1b (2) / JB-1b (p)	Japanese basalt (international)	Pressed powder
JB-2	Japanese basalt (international)	Pressed powder
JB-3	Japanese basalt (international)	Pressed powder
JG-3	Japanese granodiorite (international)	Pressed powder
JR-1	Japanese rhyolite (international)	Powder
JR-2 (1)	Japanese rhyolite (international)	Powder
JR-2 (2) / JR-2 (p)	Japanese rhyolite (international)	Pressed powder

JR-3	Japanese rhyolite (international)	Powder
JR-3 #3 / JR-3 (p)	Japanese rhyolite (international)	Pressed powder
9.5 Obsidian	Obsidian	Pressed powder
NBS688	Nevada basalt (international)	Pressed powder
NIST688	Basalt (international)	Pressed powder
NIST278	Obsidian (international)	Pressed powder
NIST2710a	Soil (international)	Powder
NIST2711a	Soil (international)	Powder
ANU2000	Obsidian (in-house ANU)	Raw obsidian
GC-188	Tongan basalt (in-house ANU)	Powder
T-10	Tongan basalt (in-house ANU)	Powder
T-11	East Polynesian basalt (in-house ANU)	Powder
T-13	Tongan basalt (in-house ANU)	Powder
T-14	Samoa basalt (in-house ANU)	Powder
T-16	Fijian basalt (in-house ANU)	Powder
T-17	Samoa basalt (in-house ANU)	Powder
T-22	Samoa basalt (in-house ANU)	Powder
T-23	Tongan basalt (in-house ANU)	Powder
T-28	Samoa basalt (in-house ANU)	Powder
T-7	Samoa basalt (in-house ANU)	Powder
MQ001	Marquesan basalt (in-house Auckland)	Pressed powder
MQ007	Marquesan phonolite (in-house Auckland)	Pressed powder
MQ1529	Marquesan basalt (in-house Auckland)	Pressed powder
MQ5296	Marquesan basalt (in-house Auckland)	Pressed powder
MQ5337	Marquesan basalt (in-house Auckland)	Pressed powder
MQ5567	Marquesan basalt (in-house Auckland)	Pressed powder
MQ5875	Marquesan basalt (in-house Auckland)	Pressed powder

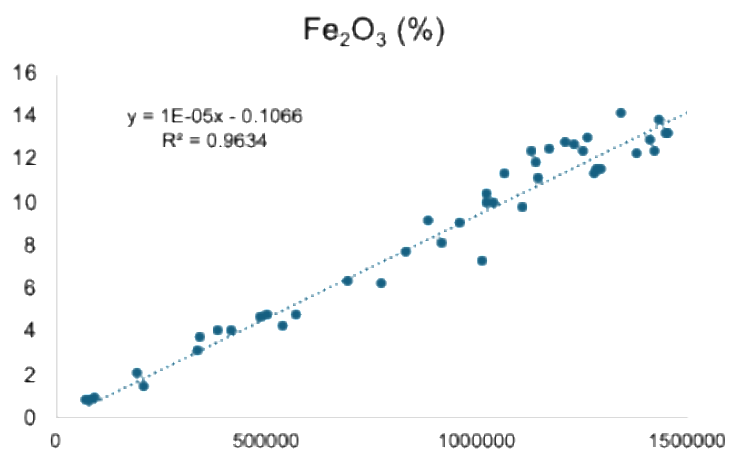


Fig. S5 Regression curve for Fe (expressed as Fe_2O_3), including the equation and R^2 value

Online Resource 2: Scanning Electron Microscopy with Energy Dispersive X-ray Spectroscopy (SEM-EDS) results (Figs. S6–S13; Tables S2–S7)

Sample #1

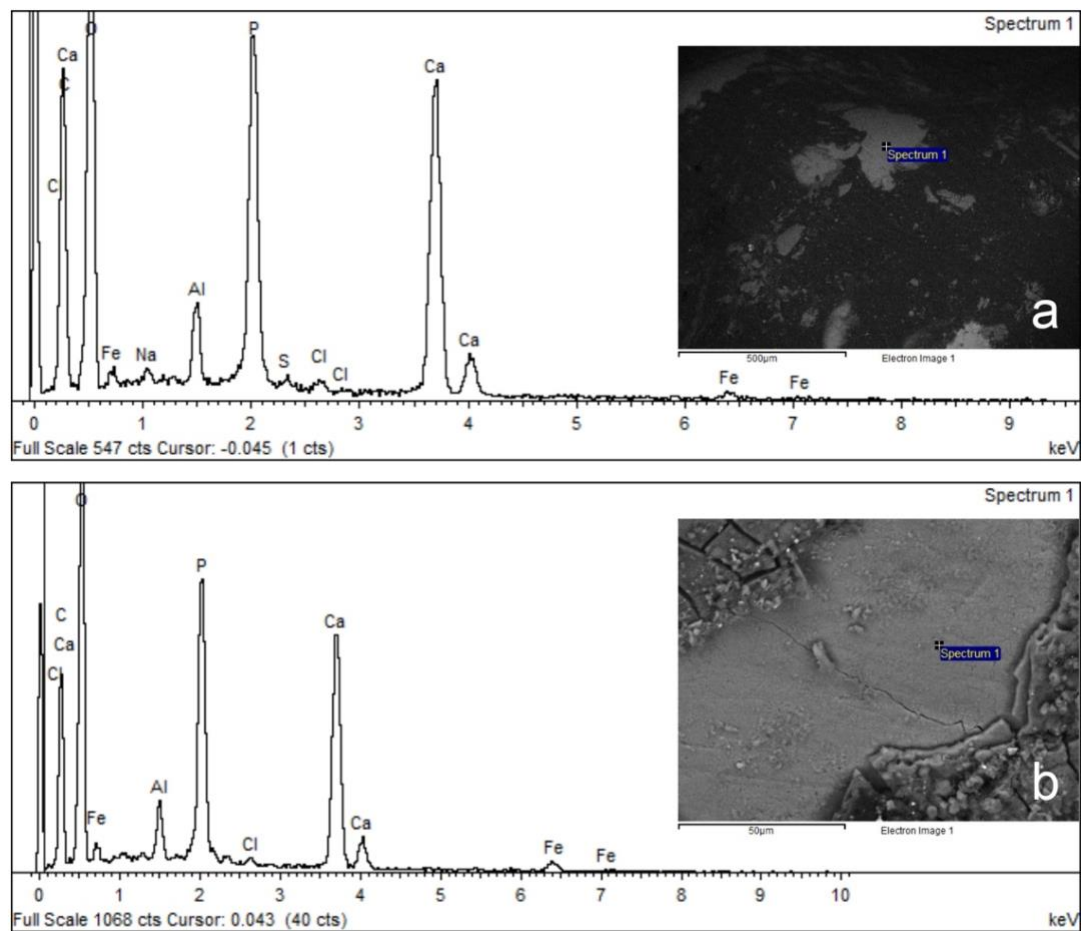


Fig. S6 Representative EDS spectra and BSE images of Sample #1

Table S2 EDS measurement results of major elements (in weight %) for Fig. S6a

Spectrum	Na ₂ O	Al ₂ O ₃	P ₂ O ₅	SO ₃	Cl	CaO	Total Fe-oxide	SUM
Spectrum 1	0.84	4.93	34.21	0.61	0.76	34.6	2.03	77.98

Sample #2

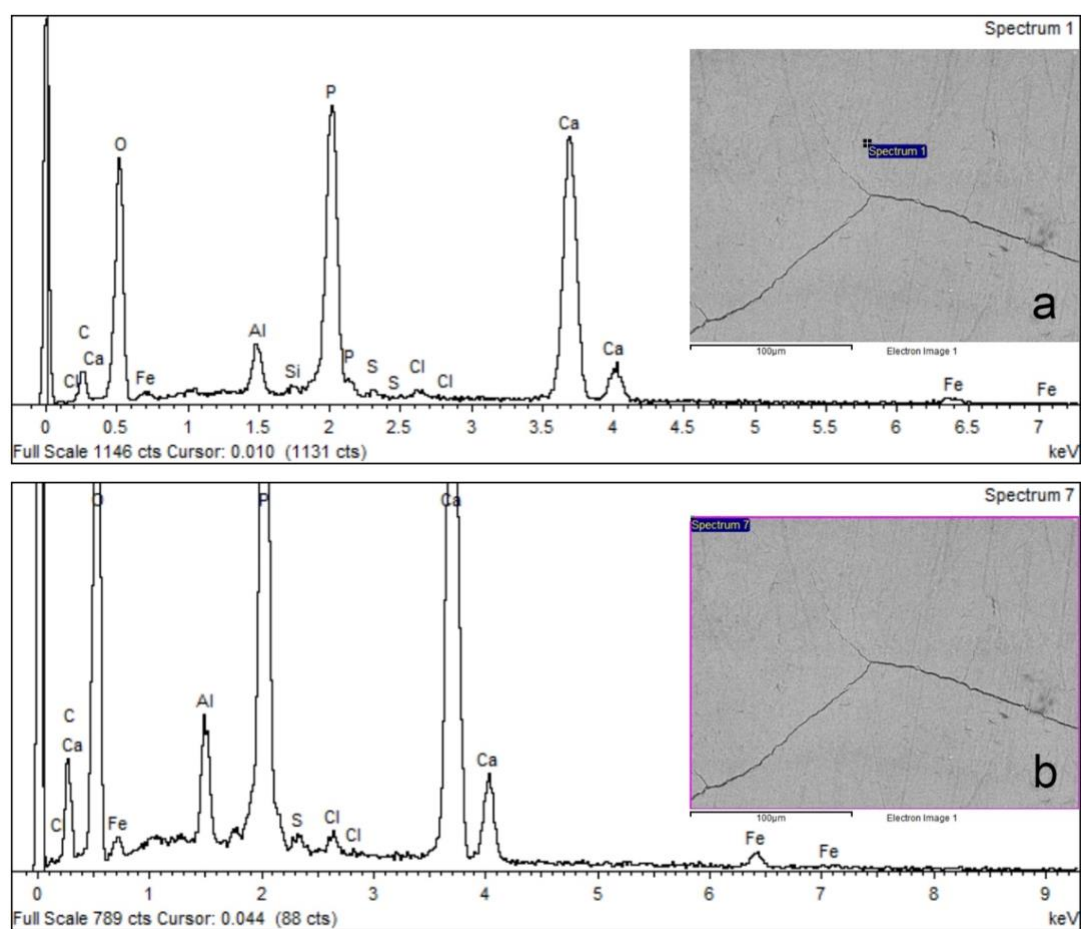


Fig. S7 Representative EDS spectra and BSE images of Sample #2

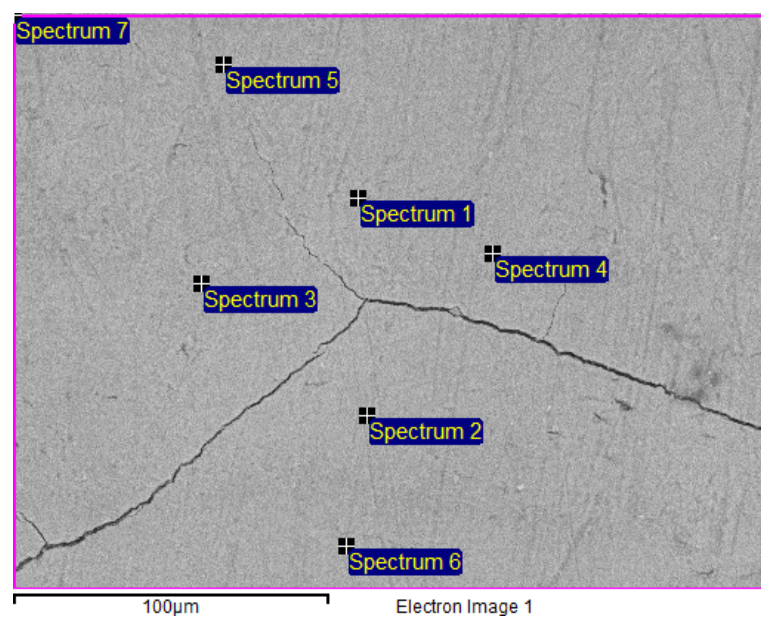
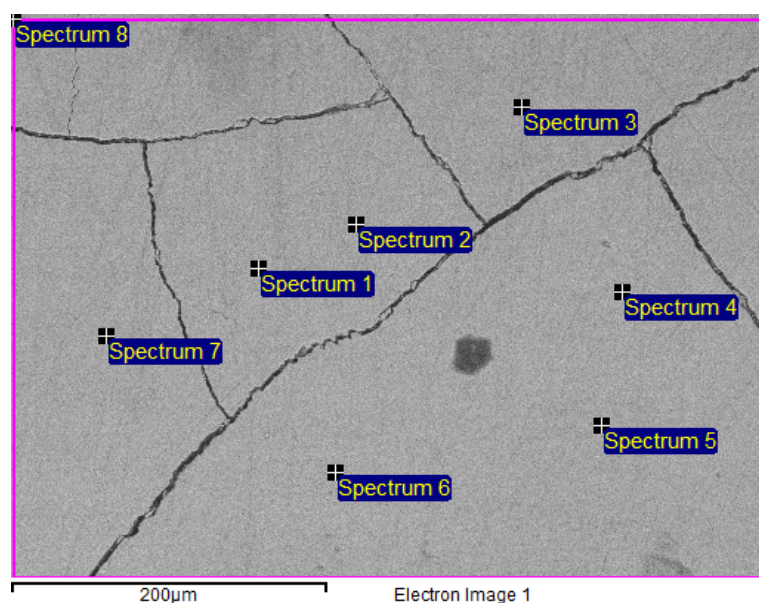


Fig. S8 Spectra corresponding to Table S3

Table S3 EDS measurement results of major elements (in weight %)

Spectrum	F	Na ₂ O	Al ₂ O ₃	SiO ₂	P ₂ O ₅	SO ₃	Cl	CaO	Total Fe-oxide	SUM
Spectrum 1	0.31	0.47	4.18	0.54	39.63	1	0.63	40.92	2.49	90.17
Spectrum 2	0.84	0.32	4.27	0.34	40.34	0.98	0.61	42.19	2.41	92.3
Spectrum 3	1.29	0.62	3.03	0.23	39.52	1.03	0.61	43.18	1.85	91.36
Spectrum 4	0.53	0.4	4.14	0	39.47	1.24	0.74	41.64	1.93	90.06
Spectrum 5	1.2	0.53	3.9	0.29	39.1	1.33	0.55	41.46	2.02	90.38
Spectrum 6	1.31	0.5	4.41	0.26	40.5	1.09	0.75	42.61	3.02	94.45
Spectrum 7	0.3	0.25	4.28	0.35	39.56	1.11	0.62	41.63	2.87	90.97
Mean	0.83	0.44	4.03	0.29	39.73	1.11	0.64	41.95	2.37	91.39
Std. deviation	0.45	0.13	0.47	0.16	0.5	0.13	0.07	0.76	0.46	1.56

**Fig. S9** Spectra corresponding to Table S4**Table S4** EDS measurement results of major elements (in weight %)

Spectrum	F	Na ₂ O	Al ₂ O ₃	P ₂ O ₅	SO ₃	Cl	CaO	Total Fe-oxide	SUM
Spectrum 1	1.03	0.38	6.78	39.68	1.57	0.49	36.35	3.81	90.09
Spectrum 2	1.97	0.55	7.17	40.01	1.74	0.56	35.44	3.38	90.82
Spectrum 3	1.12	0.55	6.63	39.95	1.85	0.83	37.69	3.41	92.03
Spectrum 4	1.45	0.28	6.62	40.31	1.92	0.67	38.17	3.73	93.15
Spectrum 5	0.94	0.56	7.07	38.39	1.87	0.47	35.76	6.49	91.55

Spectrum 6	1.03	0.52	6.70	40.87	1.47	0.58	37.55	3.64	92.36
Spectrum 7	0.87	0.49	6.86	38.95	2.00	0.62	36.39	3.43	89.61
Spectrum 8	1.08	0.50	6.78	39.47	1.58	0.64	37.54	3.68	91.27
Mean	1.19	0.48	6.83	39.70	1.75	0.61	36.86	3.95	91.36
Std. deviation	0.36	0.10	0.20	0.78	0.19	0.11	1.00	1.04	1.17

Sample #3

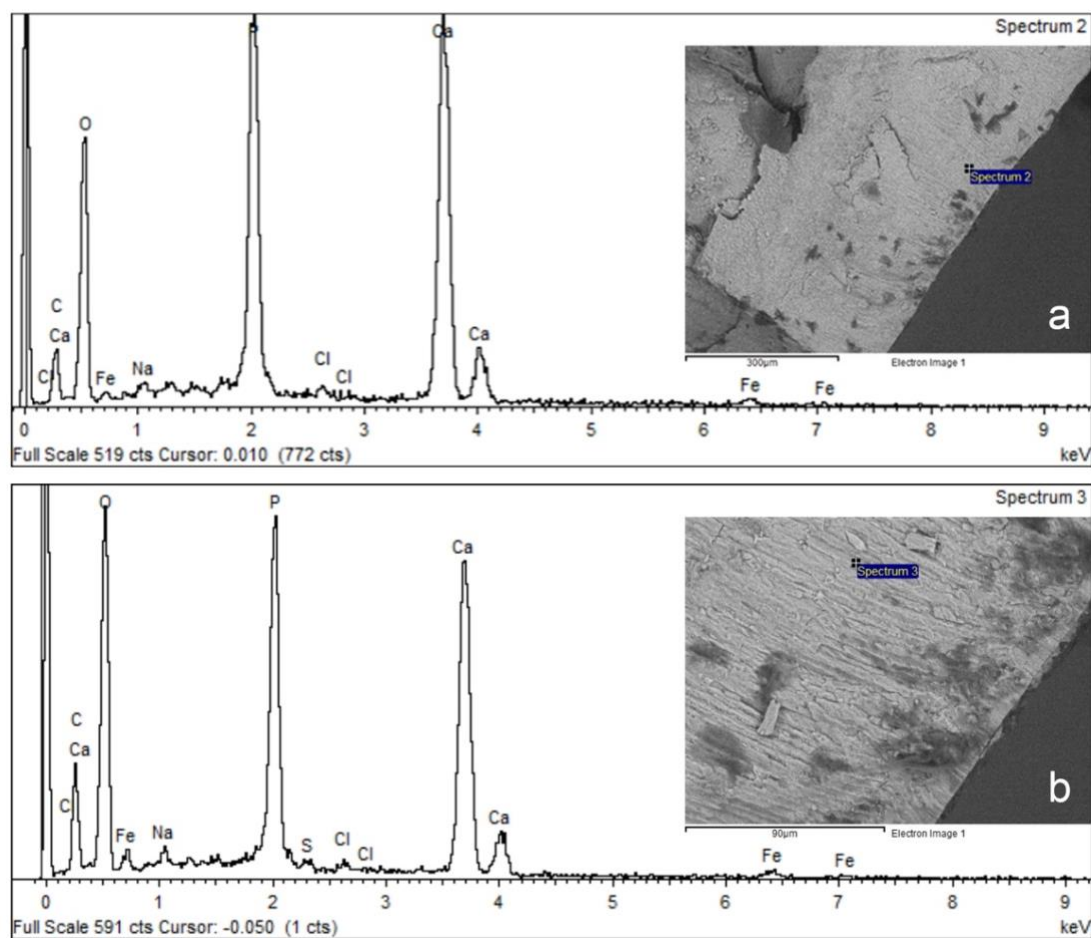


Fig. S10 Representative EDS spectra and BSE images of Sample #3

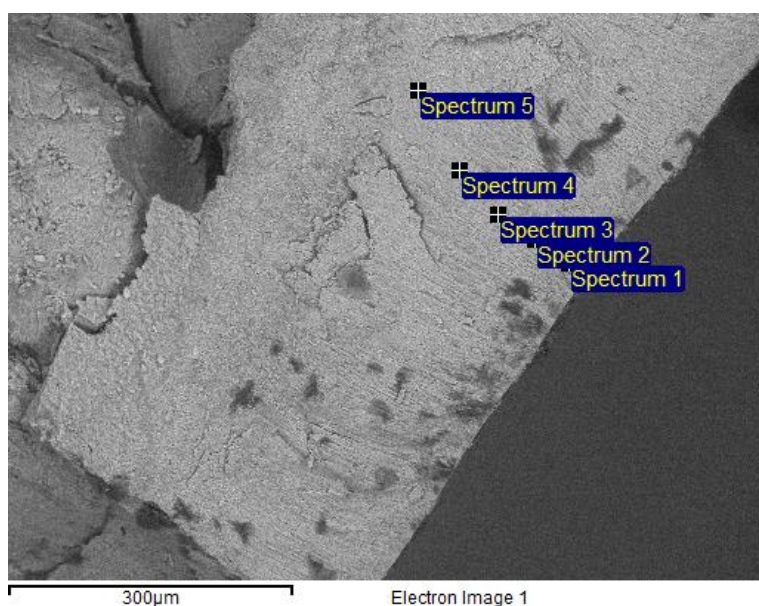


Fig. S11 Spectra corresponding to Table S5

Table S5 EDS measurement results of major elements (in weight %)

Spectrum	Na ₂ O	P ₂ O ₅	SO ₃	Cl	CaO	Total Fe-oxide	SUM
Spectrum 1	0.53	27.78	0.31	0.4	34.74	0.5	64.26
Spectrum 2	0.67	34.87	0.1	0.41	39.4	1.72	77.17
Spectrum 3	0.97	36.27	0.91	0.41	43.29	0.57	82.42
Spectrum 4	0.49	28.81	1.26	0.16	39.18	1.1	71
Spectrum 5	0.78	33.67	0.7	0.24	43.4	1.09	79.88
<i>Mean</i>	<i>0.69</i>	<i>32.28</i>	<i>0.66</i>	<i>0.32</i>	<i>40</i>	<i>1</i>	<i>74.95</i>
<i>Std. deviation</i>	<i>0.20</i>	<i>3.77</i>	<i>0.46</i>	<i>0.12</i>	<i>3.57</i>	<i>0.49</i>	<i>7.33</i>

Experimental sample

After dyeing, the modern enamel's microstructural matrix remained intact, but the signals of Fe and S appeared in the elemental profile (Fig. S12a & S12b) (mean composition of 4.81%±0.74% Fe, 0.91%±0.21% S), closely matching those of the ancient samples. Although the iron-gall ink used in the experiment contains various elements (Fig. S12c), compounds composed of Fe and S appear to be the primary agent involved in chemical reactions with the dental surface, facilitating ion substitution and leaving pigmentation marks. The significant increase in Fe-oxide and SO₃ (Table S6 & S7) suggests enhanced iron and sulfate absorption or bonding changes as a result of the dyeing process.

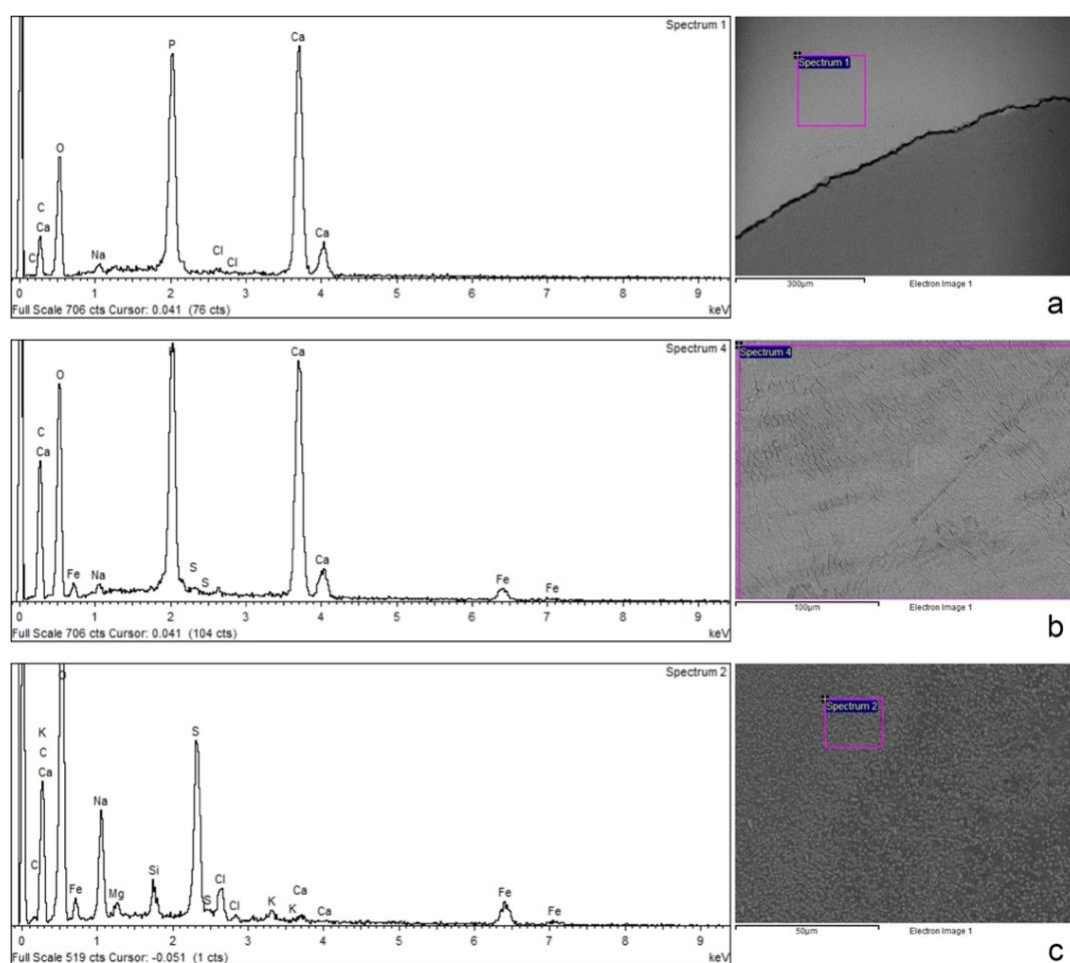


Fig. S12 Representative EDS spectrum and BSE image of the experimental sample and iron-gall ink: **(a)** unaltered enamel; **(b)** altered enamel (soaked in iron-gall ink for 60 minutes); **(c)** iron-gall ink

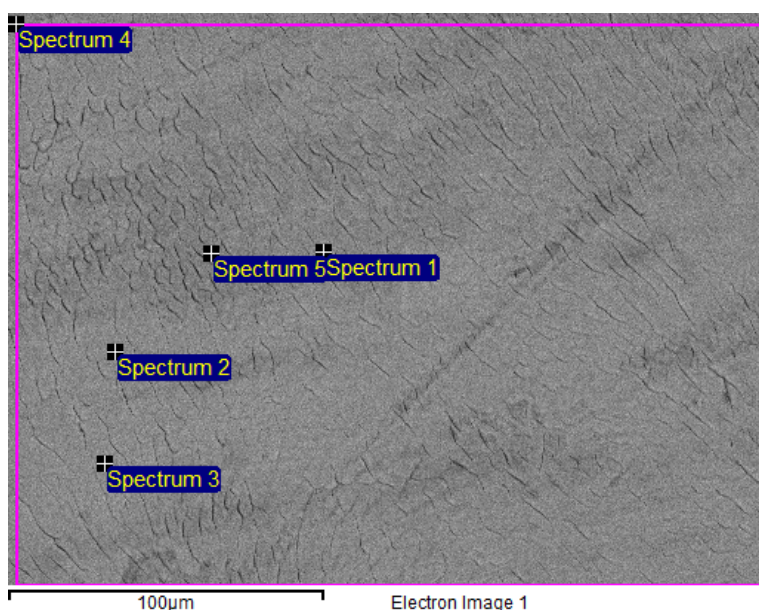


Fig. S13 Spectra corresponding to Table S6

Table S6 EDS measurement results of major elements (in weight %) in the altered experimental sample's enamel

Spectrum	F	Na ₂ O	MgO	P ₂ O ₅	SO ₃	Cl	CaO	Total Fe-oxide	SUM
Spectrum 1	0.1	0.83	0.2	28.84	0.87	0.3	30.49	5	66.63
Spectrum 2	0	0.92	0.26	29.82	0.97	0.43	30.98	3.84	67.22
Spectrum 3	0	1.06	0.1	30.22	0.7	0.36	31.47	4.3	68.21
Spectrum 4	0	0.86	0.05	30.75	0.77	0.27	33.33	5.26	71.29
Spectrum 5	0	0.64	0.14	27.45	1.25	0.3	27.11	5.67	62.56
Mean	0.02	0.86	0.15	29.42	0.91	0.33	30.68	4.81	67.18
Std. deviation	0.04	0.15	0.08	1.30	0.21	0.06	2.26	0.74	3.15

Table S7 Comparison on normalized mean results (in weight %) in the experimental sample's enamel before and after alteration

Spectrum	F	Na ₂ O	MgO	P ₂ O ₅	SO ₃	Cl	CaO	Total Fe-oxide	SUM
Unaltered	0.00	1.17	0.37	43.97	0.31	0.46	53.44	0.28	100.00
Altered	0.03	1.28	0.22	43.79	1.36	0.49	45.66	7.17	100.00

Online Resource 3: Portable X-ray Fluorescence (p-XRF) results (Table S8)

Table S8 Mean composition (in weight %) in the ancient teeth samples analyzed using p-XRF

Sample	Na ₂ O	MgO	Al ₂ O ₃	SiO ₂	P ₂ O ₅	CaO	TiO ₂	MnO	Fe ₂ O ₃	SUM
Sample #1 (DX04D1K2)	2.51	6.05	15.86	7.05	8.92	15.67	0.12	0.06	18.63	76.27
Sample #2 (DX04001)	2.83	7.19	17.08	6.72	11.67	14.27	0.1	0.2	25.48	87.65
Sample #3 (DX04MPK)	3.22	4.57	12.65	3.62	17.31	39.12	0.06	0.07	0.83	80.73

Online Resource 4: Ethnographic review (Table S9)

Table S9 Classification of various materials and methods used for alterations of dental color across representative geographical regions and ethnic groups

Scientific names of the materials (converted from common names where applicable) and unrecognized materials' names in local languages are presented in italics. Ethnic names are specified when detailed in the reference, based on the original description and/or official categorization, noting that these ethnic classifications may overlap due to temporal and local variations

Ethnic group; Category	Sanitization		Red dyeing		Black dyeing (Primary dyeing)		Polish		Reference	
	Material	Method	Material	Method	Material	Method	Material	Method		
Vietnam										
Kinh; 4	dried betel nut peel, charcoal powder, lime, rice wine	scrub, rinse	stick-lac, lime juice/ rice vinegar, rice wine, banana leaf	powder, mix and heat for paste; spread on leaf and cover overnight	<i>Phyllanthus reticulatus</i> , <i>Galla chinensis</i> , dried pomegranate rind, rice wine, glutinous rice, cinnamon, star anise, clove	powder, heat and mix into paste; cover overnight	coconut shell, metal knife	heat for tar and collect on knife; smear	Vu 1937	
	Kinh; 4	coral powder, rice vinegar	scrub		honey, bone char, agarwood powder	mix into paste; smear				D’Enjoÿ 1898

Kinh; 4	textile, knife	scrub	stick-lac, water, lime juice, banana/ coconut/ palm leaf	powder and mix for paste; spread on leaf and cover overnight	<i>Galla chinensis</i> , pomegranate rind, vitriol, glutinous rice, rice wine, cinnamon, star anise, clove, water	powder, mix, and heat into paste; spread on leaf and cover overnight	a. coconut shell ash b. tobacco, betel nut	a. scrub and polish b. smoke / chew	Bonifacy 1907
Kinh; 4	textile, glass powder	scrub	stick-lac, lime juice, coconut leaf	powder and mix for paste; spread on leaf cover overnight	a. stick-lac, lime juice, metal knife, coconut leaf b. <i>Phyllanthus</i> <i>reticulatus</i> , <i>Galla</i> <i>chinensis</i> , dried pomegranate rind, star anise, coconut leaf	a. powder and mix into paste; collect on knife and rest; spread on leaf and cover overnight b. powder and mix into paste; spread on leaf and cover overnight	coconut shell ash, lard	mix into paste; scrub	Crevost 1907
Yao; 3					bamboo node, piece of iron/ knife	prick bamboo nodes and cover one side by wet iron piece; burn the other side and collect soot on iron piece; smear			Wang 2012
Tai Lue; 3					wood, bamboo node, piece of iron	put wood of local plant in bamboo nodes and cover one side by iron			Huyen 2021

			piece; heat and collect soot on iron piece; smear daily			
Tho, Man, Lolo, Lachi, Tai; 1	fruit (<i>Paederia foetida</i>)	scrub		tobacco, betel nut	smoke/ chew	Bonifacy 1907
<i>Vietnam and Laos</i>						
Khmu; 3	nut (<i>Dracontomelon dao</i>), wood (<i>Cratoxylum formosum</i> , <i>Croton cascarilloides</i>), cast iron	burn and collect soot on cast iron; smear				Tayanin and Bratthall 2006
<i>Laos</i>						
Phu Noi; 3	wood, bamboo node, piece of iron	put a burning piece of wood in bamboo nodes and cover by iron piece; collect soot; smear				Roux 2011
Thai; 3	hard wood, bamboo node, piece of iron	cover one side of bamboo node by iron piece; burn wood on the other side and collect soot on iron piece; smear		vitriol	scrub	Vu 1937
<i>China</i>						

Dai; 3				<i>Paederia foetida</i> , vitriol, young pomegranate, banana leaf	mash and mix into paste; spread on leaf and cover overnight	wood	heat for tar; scrub	Zheng 2001
Dai; 2				wood	burn for tar; smear	betel nut, tobacco, limestone	chew	Zheng 2001
Dai; 3		leaf (<i>Hedyotis</i>), fruit (<i>Paederia foetida</i>)	scrub	<i>Citan</i> (subsistence from charcoal), betel nut, pomegranate rind, water, leaf	mash and mix into paste; boil; spread on leaf and cover overnights			Xu 2006
Bulang, Dai, Jino; 3				wood (chestnut, pear), piece of iron/ wok	burn and collect soot/ tar on iron piece; smear			Song et al. 1983; WTBHB 1984
Bulang, Dai, Hlai; 2 Palaung; 2				wood (<i>Gi</i> , <i>E'aiy Kwy Jikl</i>) charcoal ash, plant, sand, limestone	burn for soot/ tar; smear chew			Song 1975; Zheng 2001 Xu 2006
Hani; 2	sour food	chew		stick-lac	smear			Xu 2006

Myanmar

?	3	sour food, chew vitriol	oil, lampblack, iron	mix the ingredients and applied with hot iron; smear?	Malcolm 1839
<i>Borneo</i>					
?	3		a. coconut shell charcoal powder b. young betel nut, copper sulphate	a. burn for tar; smear b. powder and mix; smear	Gleichauf 1979
Dusun;	2		coconut shell/wood	burn for tar; smear	Roth 1896
<i>Philippines</i>					
Gaddang, Ifugao, Isneg;	3		wood (guava, coffee, <i>Teqom</i> , <i>Eurya</i> <i>acuminata</i> , jackfruit), iron implement (knife, axe)	burn and collect tar on iron; smear	Zumbroich and Salvador- Amores 2010
Bontoc;	2		wood charcoal (guava, benguet pine), cane sugar juice, <i>Basi</i> wine (fermenting sugar cane juice or rice)	mix; smear	Zumbroich and Salvador- Amores 2010

Tingguian; 3	tanbark, iron salt	mix; cover overnight	Zumbroich and Salvador- Amores 2010
Dumagat; 3	wood (<i>Strombosia philippinensis</i>), iron implement (knife)	burn and collect tar on iron implement; smear	Headland 1977
<i>Taiwan</i>			
Amis, Rukai, Paiwan, Puyuma; 3	wood (<i>Buxus sinica</i> , <i>Urraya paniculata</i> , <i>Kaladakalr</i> , <i>Adu'do</i> , <i>Jigura</i>), iron implement	burn and collect tar on iron implement; smear	ASCOGT 2009; Shen 2011; ASCOGT 2015b
Pingpu, Amis, Tsou, Hla'alua, Paiwan, Rukai; 1	fruit (<i>Paederia scandens</i> , <i>Dageru</i> , <i>Tamoroho</i> , <i>Daqir</i>), stem (<i>Potentilla leuconota</i> , <i>Paederia scandens</i> , <i>Tunguca</i> , <i>Tamudrung</i> , <i>Ketsun</i>), flower (banana, <i>Takeru</i>), shoot (<i>Qlceng</i> , <i>Ketsun</i>)	scrub	Yuasa 2000; Shen 2011; ASCOGT 2015a, b
<i>Japan</i>			

Japanese; 4			iron nail/ shaving, rice wine/ tea, <i>Galla chinensis</i> powder	mix and ferment; boil; mix with <i>Galla chinensis</i> powder; smear			Hayashi 1940; 1943
<i>Micronesia</i>							
Palauan; 2			plant, banana leaf	extract tar and spread on leaf; cover overnight			Zumbroich 2015a
Yapese; 3			soil (peat swamp), leaf (<i>Sonneratia acida</i> , <i>Terminalia catappa</i>)	mix; smear			Zumbroich 2015a
<i>Melanesia</i>							
Kaulong, Sengseng; 3			rock (<i>Egit</i>), lime, bark	chew (mix and heat); spread in bark and cover			Zumbroich 2015a
Sulka; 3			rock (<i>Kat</i>), sap (<i>Girpil</i>)	mix; smear; heat			Zumbroich 2015a
Kawaio; 3	betel nut peel	scrub	a. leaf (<i>Akwasi</i>), rock powder (<i>Oogo</i>) b. masticated betel nut/betel nut powder	a. chop, heat, and mash the leaf; mix with rock powder; cover overnight b. chew/ suck	betel nut powder	scrub	Bailit 1968
Lau; 3			rock (<i>Fou Oko</i>), clam shell, leaf (<i>Garuga floribunda</i> , <i>Rhus</i>	powder rock in shell; heat leaves; mix and wrap in red dracaena			Zumbroich 2015a

		<i>taitensis</i>), water, red dracaena leaf	leaf, then smoke; smear and suck	
<i>South Asia</i>				
Konyak		bamboo, metal blade	burn and collect tar/ soot on wet blade; smear	Zumbroich 2015b
Naga, Wancho				
Naga; 3				
Yogi Nath, Karbi; 2		wood (<i>Melastoma malabathricum</i> , <i>Croton caudatus</i> , <i>Murraya koenigii</i>)	heat for tar; smear	Zumbroich 2015b
Garos; 1		fruit (<i>Paederia foetida</i>)	chew	Zumbroich 2015b
Lepcha, Paharia; 1		fruit (<i>Paederia foetida</i>)	chew	Zumbroich 2015b
Meitei, Maring		fruit (<i>Melastoma malabathricum</i>)	chew	Zumbroich 2015b
Naga; 1				
Indian; 4		lemon juice, iron shaving, fruit (<i>Chebulic</i> , <i>Emblic</i> , <i>Belliric myrobalans</i>), clove	powder and mix	Zumbroich 2015b

<i>East Africa</i>				
?	1	stem/ fruit (<i>Spermacoce</i> , <i>Paederia</i> , etc.)	chew	Zumbroich 2012
Antakarana,		wood (<i>Desmodium</i>), iron	burn and collect tar on	Zumbroich
Tsimihety; 3		tool	iron tool; smear	2012
Tsimihety; 3		bark (<i>Spondias dulcis</i> / <i>Sclerocarya birrea</i>), mud	mix and chew	Zumbroich 2012
<i>South America</i>				
Panobo,		fruit (<i>Manettia</i>	chew and scrub	Zumbroich
Jivaroan,		<i>divaricata</i>), leaf (<i>Piper</i>		2013
Cashinahua,		<i>family</i>)		
Amahuaca; 1				

Online Resource 5: Historical review (Table S10)

Table S10 Key historical documents on the custom of tooth blackening written in Chinese, presented chronologically according to the periods described

¹Source documents are listed with their original titles, followed by common English translations in italics where applicable. ²Authors' names are in bold if the record is based on their firsthand experience. ³Judou (punctuations) of the text are provided by the reference's author/editor or this paper's author, with original annotations and explanatory notes in parentheses. ⁴English translation is provided by this paper's author, along with necessary English interpretations of terminology beyond the original annotations and explanatory notes, shown in italics in parentheses. ⁵Proposed locations are indicated from original script, with modern geographical equivalents in parentheses, and related references are provided in the notes (a–e) below

Source ¹	Author ²	Described period	Original Script in Chinese ³	English Translation ⁴	Proposed location ⁵	Ref.
淮南子 Huainanzi	劉安 Liu, An (179–122 BC)	Emperor Yao, 2356– 2255 BC	堯立孝慈仁愛，使民如子弟。西教沃民，東至黑齒，北撫幽都，南道交趾。	Emperor Yao established a rule of filial piety, compassion, and benevolence, treating the people as if they were his own family. He taught and pacified the Wo (<i>fertile</i>) people in the west, extending eastward to the land of the Heichi (<i>black teeth</i>), the northern region of Youdu, and southward to Jiaozhi.	East of Yao's territory (east of the Middle Yellow River)	Liu 1922

山海經·海外東經 Shanhaijing: Haiwaidongjing <i>Classic of Mountains and Seas: Classic of Regions Beyond the Seas, The East</i>	Anonymou s, pre-Qin (pre-221 BC)	pre-Qin, pre- 221 BC	黑齒國在其（青丘國） 北。為人黑（齒），食稻 啖蛇，一赤一青，在其 旁。一曰在豎亥北，為人 黑手（首、齒），食稻使 蛇，其一蛇赤。下有湯 谷，湯谷上有扶桑，十日 所浴，在黑齒北。	The Kingdom of Heichi (<i>black teeth</i>) is located to the north of it (Kingdom of Qingqiu). The people there are black (teeth) and consume rice and snakes, with one snake being red and the other blueish green. Another account places the Kingdom of Heichi to the north of Shuhai. People there have blackened hands (or heads, or teeth). They consume rice and utilize snakes, one of which is red. To the south is Tanggu (<i>hot spring valley</i>). The Fusang Tree, bathed by ten suns, grows on it. This location is to the north of Heichi.	East overseas of pre-Qin (east overseas of the Middle- Lower Yellow and Yangtze River) ^a	Guo 1986
楚辭·招魂 Chuci: Zhaohun <i>Summons of the Soul</i>	屈原 Qu, Yuan (342–278 BC)	Warring States (Chu), 475/403–221 BC	魂兮來歸！南方不可以 止。雕題黑齒，得人以 祀，其骨為醢。	Soul, return! The south cannot be your resting place. The people there have tattoos and black teeth. They perform human sacrifices and make hai (<i>meat paste</i>) from their bones.	South to Chu State (south to Middle-Lower Yangtze River)	Xiao 1986 a
史記 Shiji <i>Records of the Grand Historian</i>	司馬遷 Sima, Qian (145–86? BC)	Warring States (Zhao), 475/403–221 BC	夫翦髮文身，錯臂左衽， 甌越之民也。黑齒雕題， 卻冠紕紕，大吳之國也。	It is the custom of the Ou-Yue people to cut their hair and get tattoos, and to fold their clothes on the left. It is the custom of the Wu state to dye their teeth black, tattoo their foreheads, without guan (<i>hat</i>), and use rough needlework.	Wu State (Lower Yangtze River)	Sima 1868
異物志（太平御覽）	楊孚	Eastern Han, AD 25–220	屠移在海外，以草染齒， 因號黑齒。 西屠國在海水，以草漆	The Tuyi overseas dye their teeth with plants, which is called black teeth. The kingdom of Xitu is located overseas. The people there dye their	Tuyi/Xitu (central Vietnam) ^b	Li 1936 b;

Yiwuzhi (cited from Taipingyulan)	Yang, Fu (Eastern Han)		齒，用白作黑，一染則歷年不復變，一號黑齒。	teeth with plants, turning teeth into black color with white material. Once dyed, the color remains unchanged for years, a practice named teeth blackening.		1936
吳都賦	左思	Three	烏滸狼臙，夫南西屠。儋	Wuhu and Langhuang, Funan and Xitu. The chief of Daner	Near Daner,	Xiao
Wudufu	Zuo, Si (AD 250–305)	Kingdoms (Wu), AD 220–280	耳黑齒之酋，金鄰象郡之渠。	(<i>drooping ear</i>) and Heichi (<i>black teeth</i>), the leader of Jinlin and Xiangjun.	Jinlin, Xiangjun (Hainan to central Vietnam) ^c	1986 b
蠻書	樊綽	Tang (AD 618–907)	黑齒蠻、金齒蠻、銀齒蠻、繡脚蠻、繡面蠻並在永昌、開（關）南，雜類種也。黑齒蠻以漆漆其齒，金齒蠻以金鏤片裹其齒。有事出見人則以此為飾，食則去之。……茫蠻部落，並是開南雜種也。……从永昌城南，先過唐封，以至鳳藍茸，以次茫天連，以次茫吐薺。又有大賧、茫昌、茫盛、恐茫、薺茫（鮮芒）、	The people of Heichi (<i>black teeth</i>), Jinchi (<i>gold teeth</i>), Yinchi (<i>silver teeth</i>), Xiujiao (<i>feet tattooed</i>), and Xiumian (<i>faces tattooed</i>) are reside between Yongchang and Kainan (Guannan). The Heichi tribe blackens their teeth with lacquer, while the Jinchi tribe cover their teeth with gold pieces. When Jinchi people appear in public, they use this as decoration, but they remove it when eating...The Mang peoples are tribes from Kainan...Traveling south from Yongchang city, one first passes Tangfeng, then reaches Fenglanrong, followed by Mang-Tianlian, and then Mang-Tuhao. There are also Dadan, Mang-Chang, Mang-Sheng, Kong-Mang, Xian-Mang (Zha-Mang), and Shi-Mang, all of whom belong to Mang peoples. They live in stilted houses without city walls. Some lacquer their teeth to black.	Between Yongchang and Kainan (southwestern Yunnan, including parts of Myanmar and Laos) ^d	Fan 1986

			施茫，皆其類也。樓居，無城郭。或漆齒。			
嶺外代答·交趾 Lingwaidaida: Jiaozhi Representative Answers from the Region beyond the Mountains: Jiaozhi	周去非 Zhou, Qufei (AD 1135– 1189)	Southern Song (AD 1127–1279)	《交州記》曰：“交趾之人，出南定縣。足骨無節，身有毛，卧者更扶始得起。”余至欽，見夫黑齒跣足、皂其衣裳者人耳，烏覩所謂足無節、身有毛者哉？	Jiaozhouji (<i>Record of Jiaozhou, a book from Western Jin period, AD 266–316</i>) states, “The people of Jiaozhi, located beyond Nanding, have no joints in their feet and have hair on their bodies. They must be supported to stand from lying.” When I arrived there, I saw people with black teeth, bare feet, and blackened cloths. However, I did not see anyone with jointless feet or hairy bodies?	Jiaozhi (northern Vietnam) ^e	Zhou 1986
西南夷風土記 Xi'nanyifengtuji	朱孟震 Zhu, Mengzhen (AD 1534–?)	Ming (AD 1368–1644)	男女無貴賤，皆穿耳徒跣，以草染齒成黑色。	Regardless of status, both men and women pierced their ears, went barefoot, and dyed their teeth black with plants.	Southwest (southwestern China, between Yunnan and Myanmar)	Zhu 1985
滇略 Dianlue	謝肇淛 Xie, Zhaozhi (AD 1567– 1624)	Ming (AD 1368–1644)	《新唐書》曰：“茫，君號也。永昌南有茫天連、茫鉾、茫施，皆樓居，無城郭，有漆齒、金齒、銀齒三種夷，見人以漆及鏤金銀飾齒，寢食則去	Xintangshu (<i>New Book of Tang, a book from Northern Song period, AD 960–1127</i>) states, “Mang is the title of the ruler. South of Yongchang, there are the Mang-Tianlian, Mang-Zha, and Mang-Shi, all of whom live in stilted houses without city walls. Among them, there are three types of tribes: those with lacquered teeth, gold teeth, and silver teeth. They adorn their teeth with lacquer,	Mangshi (western Yunnan)	Xie 1986

			之。”今芒市夷多以酸石榴皮及藥染齒使黑，初無金銀鏤飾者，黑齒則信然，謂之漆，謂之為飾。見人、寢食脫去皆妄傳也。	engraved gold, and silver when appearing in public, but remove the decorations when sleeping or eating.” In present-day Mangshi, the people often dye their teeth black using sour pomegranate rinds and medicinal substances. There is no trace of gold or silver decorations. The practice of blackening teeth is indeed true, and they refer to it as ‘lacquering’ for decoration. However, the claims that they adorn their teeth to meet people and remove the decorations when sleeping or eating are unfounded rumors.		
大越史記全書 Dai Viet su ky toan thu <i>Complete Annals of Dai Viet</i>	吳士連 Ngô, Sĩ Liên (AD fifteenth century)	Fourth Era of Northern Domination (AD 1407– 1427)	賊從而分州設縣，築城鑿池，居鎮戎之兵二十餘年，變風俗於長髮，白齒胥為北人矣。嗚呼，為亂之極，一至是哉。	The invaders then divided the land into counties, built city walls and moats, and stationed troops for over 20 years. Our people changed their customs to have long hair and white teeth, turning everyone into northerners. Alas, the chaos has reached its peak, culminating in this!	Dai Viet (northern to central Vietnam)	Ngo 2015
倭漢三才圖會 Wakansansaizue <i>Illustrated Sino-Japanese Encyclopedia</i>	寺島良安 Terajima, Ryōan (AD 1654–?)	Edo (AD 1603–1868)	本朝婦人用鐵漿染齒也……其事肇於鳥羽院時。鐵漿，俗云加禰。本細鐵漿以生鐵於器中，清水服餌者，旋入新水，日久鐵上生黃，此也味鹹寒鎮心明目。……今婦人有黑齒，具按鐵漿造法，取古鐵於器中，和米屑少	In our country, women used Haguro (<i>iron liquid</i>) to dye their teeth... This practice began during the reign of Emperor Toba (AD 1107–1123). Haguro (<i>iron liquid</i>), commonly referred to as ‘Kane’, is made by placing raw iron in a vessel filled with clear water and frequently refreshing the water. Over time, a yellow substance forms on the iron, which is salty and cool, calming the heart and improving vision... Today, women who have blackened teeth use the iron liquid method. They place old iron in a vessel with a small amount	Edo (Japan)	Tera shim a 1929

許，漬水，夏三日，冬七日，在煖處則出鏽汁。黃赤色、味微甘為佳。先以五倍子末塗齒，次傳鐵漿，如此數回則齒染真黑。俗傳曰：染齒之日不可食菠薐艸，大忌之。本朝堂上諸臣咸染齒，與地下人以別之。如婦人，無貴賤黑齒，與室女以別之。如孀尼則還磨去成白齒。

of rice bran and soak it in water. In summer, the mixture is left for three days, and in winter, for seven days. If kept in a warm place, rust juice will form. The ideal juice is yellowish-red and slightly sweet. First, powdered wubeizi (*Galla chinensis*) is applied to the teeth, followed by the Haguro (*iron liquid*), and after repeating this process several times, the teeth become truly black. It is commonly said that on the day of teeth dyeing, one must not eat spinach, as it is considered a major taboo. In our dynasty, all court officials dye their teeth to distinguish themselves from commoners. Women, regardless of status, dye their teeth black to distinguish themselves from unmarried girls. However, widows and Buddhist nuns will file the teeth back to white.

Notes:

- a. This juan (*scroll, volume*) of the pre-Qin classical document, *Shanhaijing*, describes the eastern regions beyond the seas. Chinese classics usually regard foreign territories as the lands overseas, and thus, the territory of the contemporary period, including the Middle-Lower Yellow and Yangtze River areas (Tan 1996c), serves as the reference point. A total of 13 kingdoms or regions are mentioned in this juan, listed in order from the southeast to the northeast (Guo 1986), with the Kingdom of Heichi (*black teeth*) being the eighth, located just past the halfway point of the eastern regions. Furthermore, the preceding paragraph describes the god Shuhai walking from the easternmost to the westernmost points, with his left hand pointing northward toward the Kingdom of Qingqiu, located to the south of the Kingdom of Heichi (*black teeth*). This suggests that the Kingdom of Heichi (*black teeth*) is located to the east or slightly northeast of the reference point. Additionally, the region south of the Kingdom of Heichi (*black teeth*) is described as Tanggu Valley and the Fusang Tree, where the suns bathe, indicating an eastern direction. This further supports the placement of the Kingdom of Heichi (*black teeth*) to the east or slightly northeast of the reference point.
- b. The Kingdom of Xitu (西屠國), also known as Xitu-Yi (西屠夷), and written in various forms such as Kingdom of Xitu (西圖國), Xiguo-Yi (西國夷), Tu-Yi (屠夷), or similar variations. The *Wulu (Record of Wu)* from the Western Jin Dynasty describes its location as 30 li (*a Chinese ancient distance unit, approximately 415.8 meters per*

li) south of Xianglin (Zhao 2014), near what is today's Hue in central Vietnam (Tan 1996b). Additionally, the Liangshu (*Book of Liang*) (Yao 1986a), Nanshi (*History of the Southern Dynasties*) (Li 1986a) from the Tang Dynasty, and other historical documents mention the Kingdom of Linyi, which was the Xianglin Xian (*County*) of the Rinan Jun (*Commandery*) during the Han Dynasty and part of the ancient territory of Yuechang. Over 200 li south of its southern border is the Xitu-Yi tribe which also claimed kingship. These records further suggest that the Kingdom of Xitu was located approximately 12–84 km south of Xianglin, in present day central Vietnam.

- c. Fu (賦) is a form of Chinese rhymed prose from the Han Dynasty, characterized by strong parallelism and extensive cataloging of topics. The regions mentioned in the text likely form a single catalog, and the last four regions in pairs from the same sentence show a closer relationship. Although the precise location of Heichi (*black teeth*) remains uncertain, Daner (*drooping ear*) is widely recognized as corresponding to today's Hainan Island. Jinlin (金鄰), also written as Jinlin (金隣), Jinlin (金隣), Jinchen (金陳), and similar variations, is described in the Taipingyulan as being more than 2000 li from Funan, where the people hunt and ride elephants, obtaining ivory after the elephants die (Li 1936a). The Liangshu and Nanshi record a general named Fan Man, whom the Funan people supported as their king. Fan Man's strength, courage, and boldness enabled him to expand the kingdom through military conquests, even building large ships to cross the seas and conquer additional kingdoms. Consequently, Fan Man declared himself the Great King of Funan. However, during a campaign against Jinlin, Fan Man fell ill (Li 1986b; Yao 1986b). Yan Shigu (AD 581–645, Tang Dynasty) annotated the Hanshu (*Book of Han, written in the Eastern Han Dynasty*), mentioning a southern kingdom called Jinlin (金鄰), which is very distant thus named Nanlin (南鄰), also referred to as Linyi (鄰邑) (Ban 1986). Thus, Jinlin is likely close to Funan and Linyi, situated around what is now central Vietnam. Xiangjun from the Han Dynasty is believed to have been located in the vicinity of today's Chongzuo, in western Guangxi, China (Tan 1996e). Therefore, based on the structure of the Fu and research on the locations of other regions, Heichi (*black teeth*) is likely situated somewhere within the area from the border of today's China and Vietnam to central Vietnam.
- d. Yongchang is located in today's Baoshan, Yunnan, and Mang-Tianlian refers to the Menglian Dai, Lahu, and Va Autonomous County, Yunnan (Tan 1996d). Therefore, these indigenous tribes, including those with blackened teeth, likely lived in the area of today's southwest Yunnan, possibly extending into parts of Myanmar, Laos, and even Vietnam.
- e. The author Zhou Qufei held the position of Xianwei (*County Captain*) in Jingjiang Fu (*today's Guilin, Guangxi*) as well as Jiaoshou (*lecturer*) in Qinzhou (*a border city near the Ly Dynasty, Vietnam*) (Tan 1996a). Additionally, Jiaozhi in the Jin Dynasty roughly spanned from the border of Qinzhou to the Red River Delta, extending to the Ma River region (Tan 1996b). Nanding Xian in the original text also referred to the area that is now Ninh Binh province in northern Vietnam (with possible, today's Nam Dinh city). Therefore, the region mentioned in this paragraph likely corresponds to present-day northern Vietnam.

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