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Chemical evidence for the spread of lac colorant in Xinjiang from the 2nd to 9th centuries CE

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Lac dye, indigenous to South and Southeast Asia, is historically accepted to have entered Xinjiang during the Han Dynasty through trade routes established between India and China, and later from the 5th century CE, when Buddhist art started flourishing in the region. The dynamics of trade and colorant applications of lac in the region has been constrained by the limited availability of historical written sources, archeological materials, and scientific investigations. Using ultra-performance liquid chromatography with photo diode array detection and mass spectrometry (UPLC-PDA-MS), this study explores the characterization of lac in a small group of archeological textiles, wall paintings, and painted sculptures, dated between the 2nd and 9th century CE. It aims to undertake a first attempt to historically trace the dissemination of the insect dye between India and Northwest China, and within the broader framework of trade and cultural exchange along the Silk Roads in the first millennium CE.

Insect dyes have been highly regarded since ancient times for providing lasting and bright shades of red color, representing luxury and prestige. These dyes were among the most expensive colorants, and the raw insects were also one of the most profitable trading goods among continents in history. The most common sources of Eurasian insect dyes found in ancient historical literature are scale insects of the superfamily Coccoidea, including lac (*Kerria lacca* Kerr and other *Kerria* and *Paratachardina* species), kermes (*Kermes vermilio* Planchon), Polish cochineal (*Porphyrophora polonica* Linnaeus), and Armenian cochineal (*Porphyrophora hamelii* Brandt). The term "cochineal" specifically refers to Polish and Armenian cochineal in the following text.

Red colorants extracted from these insects are anthraquinone molecules, such as kermesic acid, carminic acid and laccaic acid A, which have been used to dye wool and silk since antiquity, producing colors ranging from purple to orange, depending on dyeing processes and mordants[1]. Kermesic acid and carminic acid colorants only exist within the female scale insect's body, which means that kermes and cochineal insects need to be killed and crushed to extract the red dyes, while lac dye can be extracted from the resin without killing the insects. To isolate the dye from the resin, crude lac was traditionally ground into a fine powder and treated with a dilute base solution, which extracted the coloring matter. This extract was subsequently precipitated by adding an alum solution and then evaporated to produce lac dye[1].

Kermes insects occur in coastal regions around the Mediterranean Sea. Polish and Armenian cochineal (along other *Porphyrophora* species) were widespread throughout the Palearctic region, including North of the Himalayas, and both Central and East Europe, where a characteristic biotope of steppe environment dominates[1]. Polish cochineal appears to have been used as a textile dye by the 1st century BCE by Central European communities, and would only be levied from the middle of the first millennium CE by the Roman-Byzantine authorities[2]. Further east, Armenian historians and geographers wrote in the 5th century BCE that insects grew in the Ararat valley and were sold for dyeing purposes. These insects were very likely Armenian cochineal, which was historically recorded to be found among the booty plundered by the Assyrians around 714 BCE[3]. Lac dye is part of

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the resinous secretion produced by scale insects of the *Kerria*, *Paratachardina* and other species, of which the Indian *Kerria Lacca* is the most common one in human culture. This dye has been used in India for more than 4000 years, being appreciated for its excellent dyeing ability since the end of the Vedic period[4].

At the turning to the 1st millennium CE, kermes and cochineal were the main sources of insect dyes in Europe, West Asia and North Africa, as furthermore attested by the identified presence of these dyes in textile fragments found in Roman archeological sites located in Egypt and Syria[5,6]. In contrast, lac-dyed textiles, imported from South and Southeast Asia, appear to have been sparsely available in these regions[7]. In Xinjiang and its surrounding regions, lac, as well as kermes and cochineal, have been reported in a few excavated textiles[8–11], suggesting the travel of these insect materials from West and South Asia into Northwest China from the 3rd century BCE to the 10th century CE.

Xinjiang, located in the hinterland of the vast central Eurasian continent, is a historically significant crossroad of various cultures and trade routes since ancient times. This territory not only bridges the West Asian and the Eastern civilizations, but is also open to the Steppe of the North, and has long-lasting ties with India. Previous archeological studies indicated that as early as the 2nd millennium BCE, the Steppe peoples of the Late Bronze Age and Early Iron Age mingled with the indigenous peoples in Xinjiang[12]. Agricultural plants, animals, technologies, religious beliefs, artefacts and trade subsequently traversed into East Asia through Xinjiang along with the migration and mixing of people at that time[13,14]. With the rise of the Silk Road, Xinjiang played an increasingly important role in the cultural exchanges between East and West. Especially around the 1st century BCE, Buddhism began to spread from India northwards, gradually entering Xinjiang. As an important stop for the spread of Buddhism, Xinjiang witnessed the prosperity and development of Buddhist culture. The Kizil and Simsim Grottoes in the Kucha area are outstanding representatives of Buddhist art in Xinjiang. These caves are not only places of Buddhist practice and worship, they are also surviving treasure repositories of ancient artistry, with their intricate wall paintings, richly-made textiles, and painted sculptures and potteries. The decorative patterns, styles, and dyes found in many of these objects reflect the influence of foreign cultures, such as those of West Asia or the Eurasian Steppe[15–17].

Despite great efforts in describing the spread and exchange of metals, cash crops and pottery in Eurasia, little scientific research has been paid to insect-derived materials with regional characteristics, such as lac. Historical context about trade and colorant applications of lac dye in Xinjiang is usually limited by the scant availability of historical written sources, archeological materials, and scientific investigation. This study attempts to historically trace the dissemination of lac dye in Xinjiang, by combining the geographical location and cultural background of lac, with chemical analysis of surviving archeological objects. Using ultra-performance liquid chromatography with photo diode array detection and mass spectrometry (UPLC-PDA-MS), scanning electron microscope (SEM) and Raman spectroscopy, this study characterizes the presence of lac in fifteen samples taken from archeological textiles, wall paintings, and painted sculptures dated between the 2nd and 9th century CE.

Methods

Objects and samples

Samples were obtained from three archeological textiles from three burial sites in Xinjiang, Miran and Yingpan, located on the southern and eastern rims of the Taklamakan Desert, and Astana, located in the Turpan Basin (Fig. 1). These textiles are extraordinarily well preserved, due largely to the very dry climate of Xinjiang (Fig. 2a, d). An additional silk samite robe lining fragment with horses' roundels of Sogdian origin (MC1) was collected by the China National Silk Museum through intermediaries in Gansu or Qinghai province. In addition, eleven samples of nine wall paintings and painted sculptures (Fig. 2b, c, e–k) were obtained mainly from the Kizil and Simsim Grottoes and Astana Tombs (Fig. 1). The list of samples and associated descriptions are presented in Table 1, and the background of the listed archeological sites is described below:

- Yingpan, Yuli (CE 140 ± 30): The Yingpan Cemetery is located in the Lop Nor area of Xinjiang. In 1995, The Cultural Relics and Archaeology Institute of Xinjiang Uygur Autonomous Region led a team to excavate the cemetery and many textiles (wool, silk, cotton), wooden objects, pottery and ironware were found. The cemetery can be dated back to Eastern Han and Jin Dynasties (CE 25–420). The finding of weaving tools and loom components found in tombs indicate that the recovered woolen textiles could have been produced locally [18].

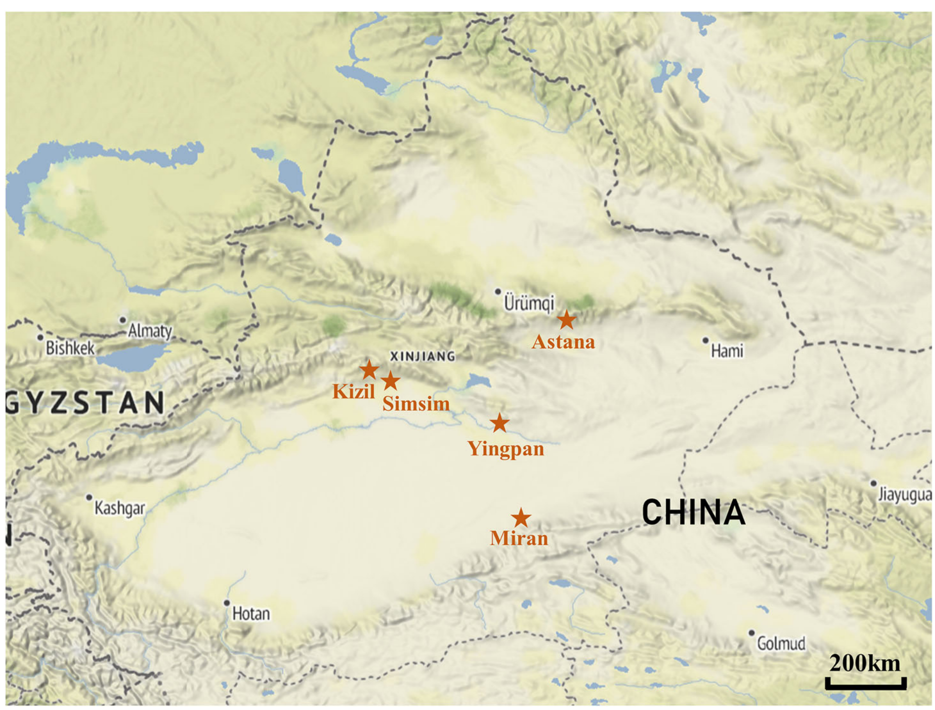

Fig. 1 | Geographic location of the archeological sites studied in this work.

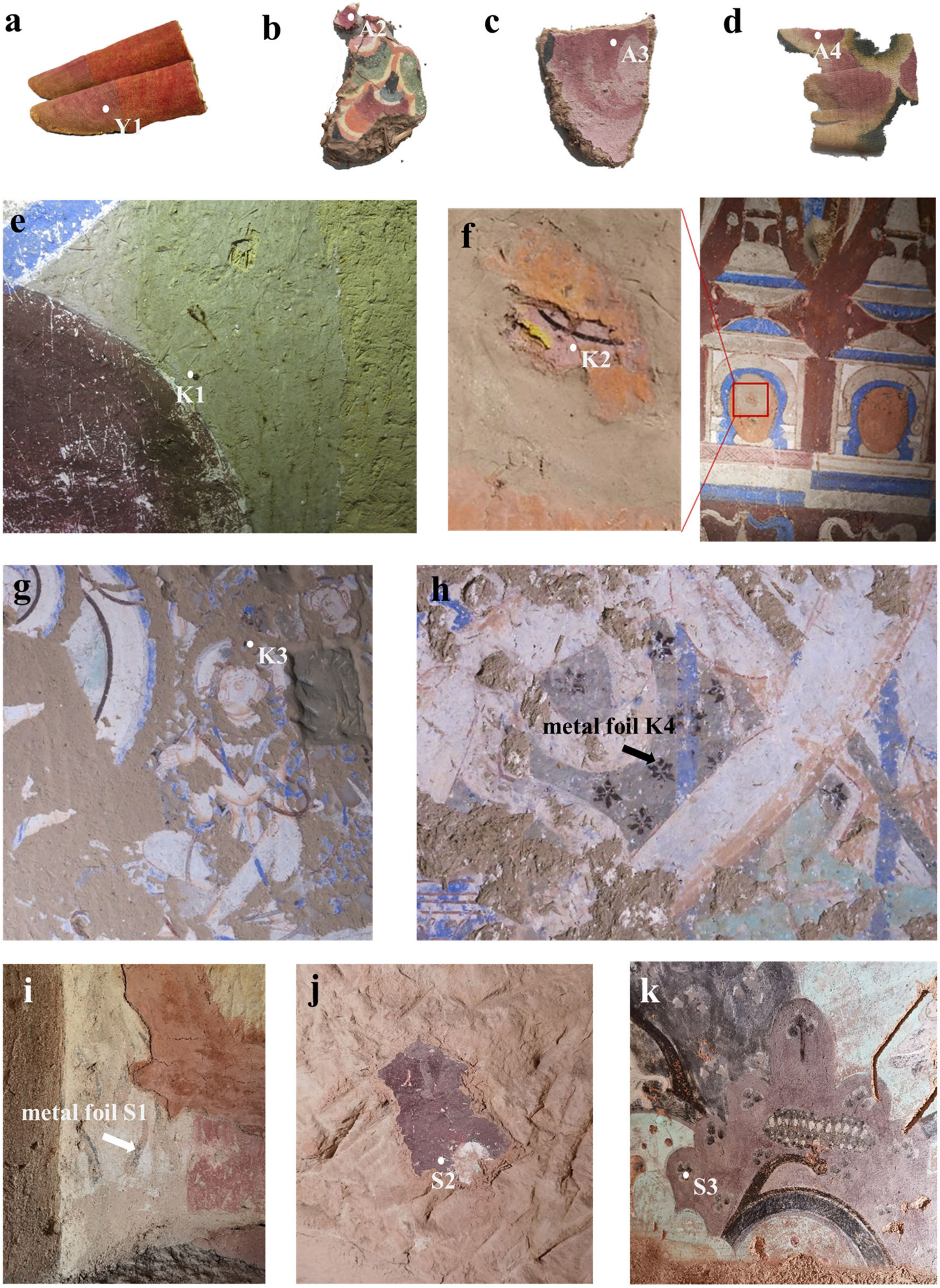

Fig. 2 | Sampling locations of the objects. **a** Y1 from the Yingpan Cemetery; **b** A2 from 60TAM336, **c** A3 from 72TAM187, and **d** A4 from 73TAM227, from the Astana Tombs; **e** K1, **f** K2, **g** K3, and **h** K4 from Cave 171 in the Kizil Grottoes; and **i** S1 from Cave 1, **j** S2 from Cave 9, and **k** S3 from Cave 14, in the Simsim Grottoes.

– Kizil (CE 5th–7th) and Simsim (CE 5th–9th), Kucha: The Kizil and Simsim Grottoes are two cave complexes located in the north of the Taklamakan desert, in the ancient Kucha area. The Kizil is the earliest major Buddhist cave complex in China, with more than 5000 m^2 wall paintings left. The Simsim is equally covered with colorful wall paintings and architectural cave structures following different styles from Central China, India and even Greece[19,20].

– Astana, Turpan (CE 7th–9th): The Astana Tombs are located near the Turpan basin, and were the public graveyards of the ancient Gaochang people. Thousands of tombs dating from the 3rd to the 9th centuries CE were excavated by the Xinjiang Uyghur Autonomous Region Museum and other institutes during 1959 and 1975. These campaigns recovered from the tombs many painted clay sculptures, pottery, and metallic objects. Due to the extremely dry

Table 1 | List of the archeological textiles and painted objects investigated in this study

Archeological site	Object No.	Description
Yingpan, Yuli (CE 140 ± 30[a])	Y1	Red woolen socks
Kizil, Kucha (CE 5th–7th)	K1 in Cave 171	Red colorant of wall painting
	K2 in Cave 171	Red colorant of wall painting
	K3 in Cave 171	Red colorant of wall painting
	K4 in Cave 171	Gilding adhesive of wall painting
Simsim, Kucha (CE 5th–9th)	S1 in Cave 1	Gilding adhesive of wall painting
	S2 in Cave 9	Red colorant of wall painting
	S3 in Cave 14	Red colorant of wall painting
	S4 in Cave 18	Red colorant of wall painting
Museum collection (CE 6th–9th)	MC1	Robe lining with horses' roundels
Astana, Turpan (CE 7th–9th)	A1 from 72TAM187	Ocher colorant of Painted sculpture
	A2 from 60TAM336	Purplish colorant of Painted sculpture
	A3 from 72TAM187	Dark red colorant of Painted sculpture
	A4 from 73TAM227	Pink yarn
Miran, Charklik (CE 8th–9th)	MR1	Red woolen thread

[a]The textile sample was radiocarbon dated by accelerator mass spectrometry (AMS) at the Beta Analytic Radiocarbon Dating Laboratory.

Table 2 | List of compounds identified in the archeological textiles by UPLC-PDA-MS (negative ion mode)

Samples	Peak no.	Possible compounds	Chemical formula	Calculated mass $[M-H]^-$	Experimental mass $[M-H]^-$
Y1	1	Laccaic acid A	$C_{26}H_{19}NO_{12}$	536.08	536.00
	2	Laccaic acid B	$C_{24}H_{16}O_{12}$	495.06	494.99
	3	Laccaic acid C	$C_{25}H_{17}NO_{13}$	538.06	538.02
	4	Laccaic acid E	$C_{24}H_{17}NO_{11}$	494.07	494.03
A4	1	Laccaic acid A	$C_{26}H_{19}NO_{12}$	536.08	536.05
	2	Laccaic acid B	$C_{24}H_{16}O_{12}$	495.06	494.99
	3	Laccaic acid C	$C_{25}H_{17}NO_{13}$	538.06	538.06
	4	Laccaic acid E	$C_{24}H_{17}NO_{11}$	494.07	494.07
MR1	1	Laccaic acid A	$C_{26}H_{19}NO_{12}$	536.08	536.05
	2	Laccaic acid B	$C_{24}H_{16}O_{12}$	495.06	494.98
	3	Laccaic acid C	$C_{25}H_{17}NO_{13}$	538.06	538.06
	4	Laccaic acid E	$C_{24}H_{17}NO_{11}$	494.07	494.05
MC1	1	Laccaic acid A	$C_{26}H_{19}NO_{12}$	536.08	536.02
	2	Laccaic acid B	$C_{24}H_{16}O_{12}$	495.06	495.06
	3	Laccaic acid C	$C_{25}H_{17}NO_{13}$	538.06	538.06
	4	Laccaic acid E	$C_{24}H_{17}NO_{11}$	494.07	494.05

Calculated masses and experimental masses of the deprotonated molecules $[M-H]^-$ and chemical formulas are reported.

climate in the Turpan basin, many organic remains have been preserved[21].

- Miran, Charklik (CE 8th–9th): The Miran site, located on the Southern edge of the Taklamakan desert, is well known for its "winged angel" wall paintings and diverse cultural artifacts dating from the Han Dynasty to the Tubo Kingdom (~2nd BCE–9th CE). Although the site was excavated in the late 19th century by European archeologists, most remains have been recovered by teams from the Chinese Academy of Social Sciences and the Cultural Relics and Archaeology Institute of Xinjiang Uygur Autonomous Region since 1950[22].

Chemicals and reagents

All solvents were HPLC grade or higher. Methanol and acetonitrile were purchased from Merck (Germany), formic acid and ultra-pure water from Fisher Scientific (USA), and pyridine from ANPEL CNW (China). Oxalic acid (≥98%) was purchased from Acros Organics (USA).

Extraction procedure

Colorants were extracted from about 0.1–0.2 mg of yarn from archeological textiles and about 0.5 mg of sample from painted surfaces by heating in 200 µL of pyridine/water/0.1 M oxalic acid in water (95/95/10) at 80 °C for 30 min. The supernatant solution was transferred to a 1.5 mL centrifuge tube (Eppendorf, Germany), and the extract was evaporated to dryness under a stream of nitrogen, after which the residue was dissolved in 50 µL of MeOH/H_2O (1/1). Subsequently, the solution was centrifuged for 5 min, and 30 µL of supernatant was transferred to a 200 µL microvial (Shimadzu, Japan) for analysis. Then, 20 µL was injected into the UPLC column by an autosampler.

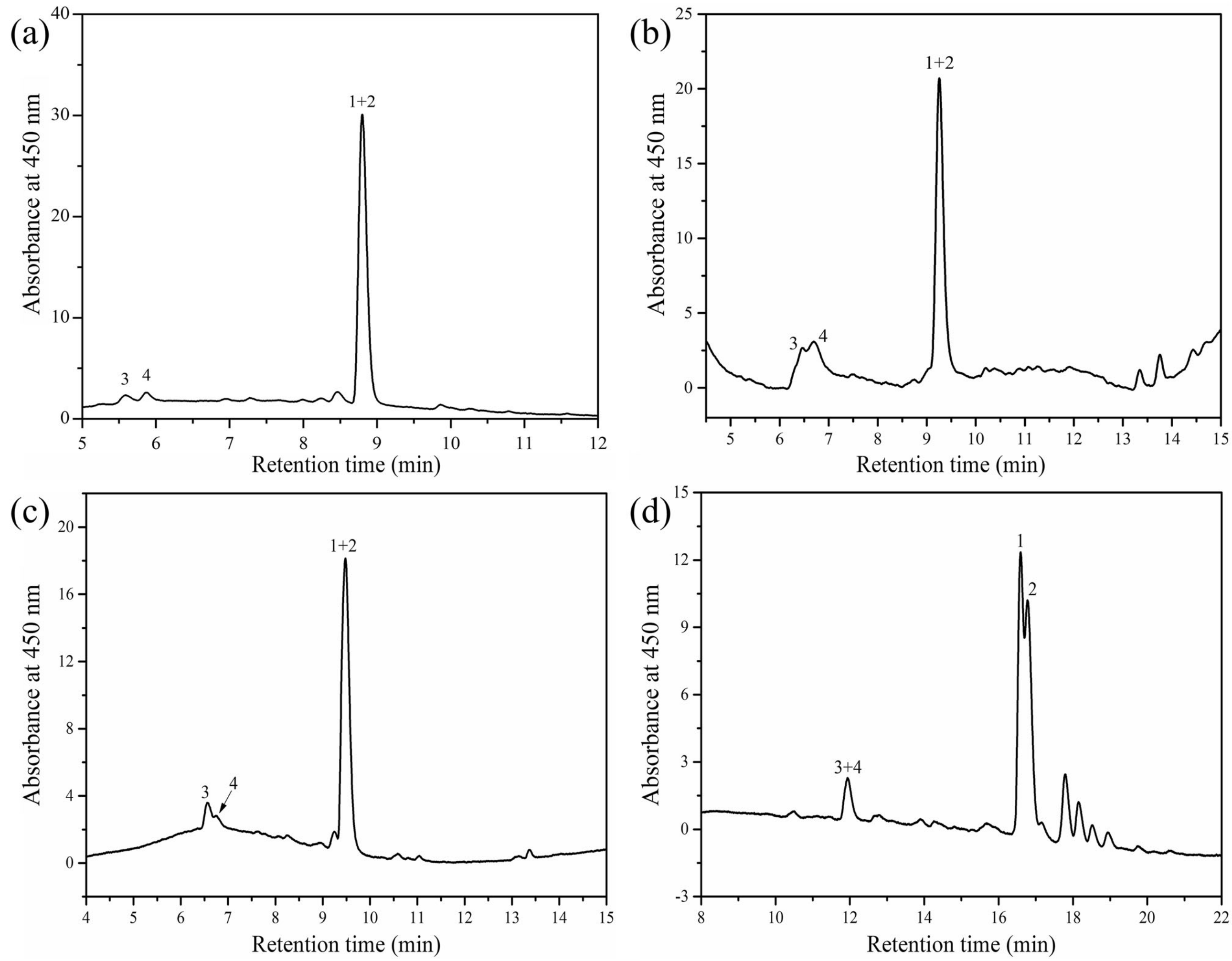

Fig. 3 | HPLC profile of the red dye extracted from the following samples. a Y1, **b** A4, **c** MR1, and **d** MC1. Compounds 1–4 are laccaic acids A, B, C and E, respectively.

Analysis of colorant by UPLC-PDA-MS

Colorant analysis was performed using a UPLC-PDA-MS system consisting of two LC-20AD delivery units, a SPD-M20A photodiode array detector (all Shimadzu, Japan) coupled with an LTQ XL linear ion trap mass spectrometer with an ESI ion source (Thermo, USA). The extract was separated on a Shim-pack XR-ODS column (3.0 mm × 75 mm, 2.2 μm particle size) or a Phenomenex Luna C18 column (2.0 mm × 150 mm, 3.0 μm particle size). The column was eluted with water-acetonitrile gradients containing 0.1% formic acid at a flow rate of 0.3 mL/min[8]. During this research, though several gradient programs were used, the peaks always eluted in the same order, since the types of stationary and mobile phases were not changed.

Multiple-stage mass spectra were obtained and processed by Xcalibur 2.1 software (Thermo, USA) in the mass range of m/z 50-1000. The mass spectrometer parameters were set up as follows: ion spray voltage −2.5 kV for the negative mode; capillary temperature 350 °C; nitrogen gas as sheath and auxiliary gas at pressures of 35 and 15 psi, respectively; capillary voltage −50 V for the negative mode. MS/MS spectra were obtained by data-dependent acquisition (DDA) using collision-induced dissociation (CID).

Microscopic observation and Raman analysis

Digital micrographs were acquired using a three-dimensional digital microscope VHX-5000 (Keyence, Japan) with magnifications between 250 and 2500×. For the cross-sectional observation, sample K4 was embedded in epoxy resin, and then sanded and polished.

Scanning electron microscopy and energy-dispersive X-ray spectroscopy (SEM-EDS) was carried out with a S-3700N tungsten filament scanning electron microscope (Hitachi, Japan) and an energy dispersive X ray spectrometer (Oxford, England). Before the inspection, the cross-sections of samples were sprayed with gold using a high-speed sputter-coating instrument connected with a conductive sheet. The accelerating voltage was set at 20 kV and the working distance was 15 mm.

Raman analysis was performed using a Nova2S Raman spectrometer (Ideaoptics, Shanghai, China) and a BX-43 confocal microscope (Olympus, Tokyo, Japan) with excitation at 532 nm. The laser power applied to the samples ranged between 1 and 3 mW. The Raman spectra were acquired using a 50× objective lens, a spectral range of 86–2986 cm^{-1}, and a resolution of 8 cm^{-1}. The signal was integrated twice, with each collection lasting 8–16 s.

Results

Textile samples

UPLC-PDA-MS analysis showed that all the textiles investigated contained laccaic acids A ($[M-H]^-$ = m/z 536.08), B ($[M-H]^-$ = m/z 495.06), C ($[M-H]^-$ = m/z 538.06) and E ($[M-H]^-$ = m/z 494.07), indicating the use of lac dye as colorant[23] (Table 2). The identification of lac dye in the red yarn sampled from the Yingpan woolen socks (Y1, dated to CE 140 ± 30) represents so far the earliest evidence of the use of lac dye in Xinjiang (Fig. 3a). In a previous study[8], three red dyes including cochineal, lac dye, and dyer's madder (*Rubia tinctorum L.*) were simultaneously identified in a single woolen yarn from a pair of embroidered woolen boots excavated from the Niya site (95MNM3:53), which is dated to the 2^{nd}–3^{rd} centuries CE.

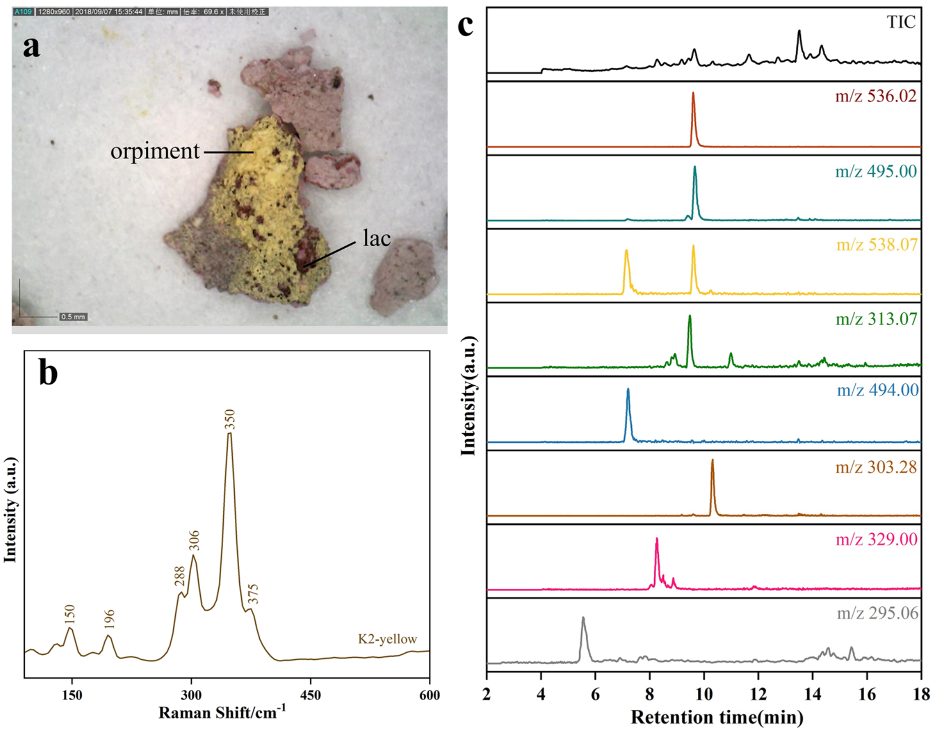

Fig. 4 | The experimental data of sample K2. **a** Microscopic picture. **b** Raman spectrum of yellow pigment (orpiment). **c** Total ion chromatogram (TIC) and extract ion chromatograms (EICs) of the main components by UPLC-PDA-MS analysis (negative mode).

Table 3 | List of compounds identified in the samples collected from Kizil and Simsim Grottoes, and Astana Tombs by UPLC-PDA-MS (negative ion mode)

Possible compounds	Chemical formula	Calculated mass [M-H]-	MS/MS ESI (-) fragment ions	R.T. (min)	K1	K2	K3	K4	S1	S2	S3	S4	A1	A2	A3
Laccaic acid A	$C_{26}H_{19}NO_{12}$	536.08	518.03, 492.13, 448.11	9.5	+	+			+		+	+	+	+	+
Laccaic acid B	$C_{24}H_{16}O_{12}$	495.06	476.98, 451.09, 407.19	9.6	+	+			+		+	+	+	+	+
Laccaic acid C	$C_{25}H_{17}NO_{13}$	538.06	520.02, 494.13, 439.28, 402.20, 365.96	6.4, 9.6		+					+	+	+		+
Laccaic acid D	$C_{16}H_{10}O_{7}$	313.03	293.00, 269.05, 244.75	9.4, 11.2		+	+	+		+	+		+	+	+
Laccaic acid E	$C_{24}H_{17}NO_{11}$	494.07	476.26, 450.15, 405.22	6.5, 9.7		+								+	+
Aleuritic acid	$C_{16}H_{32}O_{5}$	303.21	285.14, 267,16, 201.11, 171.10, 155.08, 127.01	10.4	+	+	+	+	+	+	+	+	+	+	+
Kermesic acid	$C_{16}H_{10}O_{8}$	329.03	283.07, 270.66, 211.26, 187.10, 155.03, 139.93	7.9	+	+	+	+	+	+	+	+	+	+	+
Shellolic acid	$C_{15}H_{20}O_{6}$	295.12	277.12, 251.01, 249.00, 233.19, 207.05	5.6		+	+	+			+	+	+	+	+

Retention times (R.T.), calculated masses of the deprotonated molecules [M-H], chemical formulas and accurate masses of the fragment ions present in the MS/MS spectra are reported.

Laccaic acids were also found in the other analyzed textiles, which are dated to a later period, between the 6^{th} and 9^{th} century CE: the pink yarn (A4) from the Astana Tomb, the woolen thread (MR1) from the Miran tomb, and the Sogdian robe lining silk samite with horses' roundels (MC1) discovered around Gansu or Qinghai province (Fig. 3b–d).

Wall painting samples

Previous studies generally reported that the grotto wall paintings in the Kucha region of Xinjiang were mainly painted with mineral pigments, such as cinnabar or red lead[24]. However, in this study two red areas (K2 and K3) sampled from cave 171 of the Kizil Grottoes indicated the use of an organic colorant which was taken from a pink-hued Buddha face (Figs. 2f and 4a), overlaid on yellow orpiment pigment, as identified by Raman analysis (Fig. 4b). Laccaic acids A, B, C, D and E, were detected, along with a small amount of kermesic acid ($[M-H]^{-}$ = m/z 329.03)[25], demonstrating the presence of lac colorant (Fig. 4c). In addition to these representative compounds, non-colored shellac resin components, such as aleuritic acid ($[M-H]^{-}$ = m/z 303.21) and shelloic acid ($[M-H]^{-}$ = m/z 295.12)[20,26], were observed in relatively high amounts with UPLC-PDA-MS (Fig. 4c). The presence of lac was also confirmed in four samples taken from the Simsim Grottoes dated between the 5^{th} and 9^{th} centuries CE. All these samples retained shellac resin components, including aleuritic acid and shellolic acid. Laccaic acids A-E were partially detectable due to the varying aging degrees of these colorants (Table 3).

In Buddhist art, small layers of metal leaf (such as gold, silver or tin) were usually overlaid on painted areas to give Buddha a 'shining' appearance[20,27,28]. This gilding technique was quite common in the Kizil and Simsim Grottoes. Samples K4 and S1 were taken from metal foil layers, as indicated in Fig. 2. Analytical examination of sample K4 confirmed that this comprises a top thin layer of gold foil (Fig. 5), combined with a purplish red

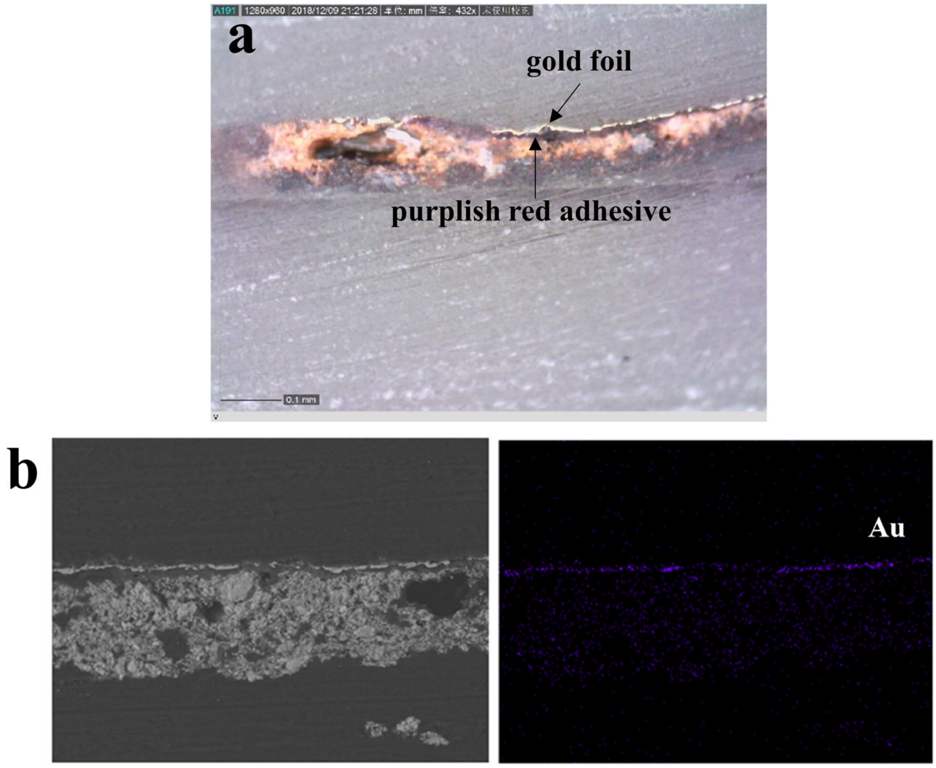

Fig. 5 | The experimental data of sample K4. Top: **a** Cross-sectional micro-photograph highlighting the layers of gold and lac dye-based adhesive; bottom: **b** SEM image (left) and EDS elemental map for gold (right).

resinous substance of lac colorant and shellac (Table 3). These results illustrate a versatile variety of techniques for using lac in wall paintings, not only as red colorant but also as means for gilding.

Sculpture samples

In the purplish red color sculptures (Fig. 2b, c) excavated from the Astana Tombs, lac dye components were furthermore detected (Table 3). The preservation of this organic pigment owes primarily to the incredibly dry and stable environment of the burial chambers, which contributed for the preservation of many sculptures' paint materials.

Discussion

Lac is composed of wax (6–7%), red chromophores known as laccaic acid A-E (4–8%) and resin (70–80%), which consists mainly of terpenic acids, aleuritic acid and several minor fatty acids[29]. Due to the water-solubility of laccaic acid A-E, a lac-dye is prepared by extracting the red chromophores with water-based solvents to produce pure deep red dye[30]. However, in the analyses of the wall paintings from the Kizil and Simsim Grottoes and the painted figurines from the Astana Tombs, both the red pigment and shellac resin components were present, not only as red colorant but also as means for gilding.

Therefore, a rational conjecture is that the shellac components are left on purpose in the formulation of lac pigment in this context. A similar scenario is found in European medieval illuminated manuscripts, where the inclusion of resinous components in lac lake pigment was a desirable feature, as they offered improved binding properties and added sheen to the paint layers[31,32]. It has also been observed in joint investigations of Cave 85 at the Mogao Grottoes (9th-10th century CE) by the Getty Conservation Institute and the Dunhuang Academy Institute, as well as in the wall paintings of Cave 254 at the Mogao Grottoes, that lac might be applied as a red colorant overlaying red minerals to enhance color[33,34].

Lac has a long history as a natural dye. As described in a 1st century CE manuscript, lac was traded as a commodity, '*lakkos chromatinos*', together with cotton cloth from India into the eastern Mediterranean region[35], and it has been identified as a pigment on 3rd century BCE Greek polychromy[36]. In China, the first record of lac can be found in *WuLu* (吴录), written by Zhang Bo during the Jin Dynasty (4th century CE)[37]. However, as shown in this study, lac dye present in the red yarns found in the Yingpan (Y1) and Niya (95MNIM3:53) sites around the Taklamakan Desert demonstrates that lac was brought into Xinjiang at least from the 2nd century CE. Whether lac was brought into the region in the form of dyestuff (which was then applied in local textiles), or already present as a colorant in imported textiles, is impossible to ascertain without a proper study of the origins of the extant textile fragments.

Trade routes connecting India and China, and passing through Xinjiang, were established during the Han Dynasty, promoting the circulation of a wealth of products along the Silk Roads, namely Indian cotton and Chinese silk[38]. Indeed, cotton cloth imprinted with an Indian Buddhist design has been found near the Niya site in Xinjiang, suggesting a close communication between India and Xinjiang during the Han Dynasty[39]. In the following centuries, lac dye and dyed textiles continued to be traded into Northwest China. For example, many relics containing lac dye have been found at archeological sites such as Astana, Miran, and Dunhuang, showing the wide application of this dye in the region[10,34].

Lac dye was used not only for coloring textiles, but also as a pigment in Buddhist architecture. The earliest identification of lac in Buddhist wall paintings was found in samples from Cave 2 (built around CE 450–500) of the Ajanta Grottoes[40], one of the most representative ancient Indian Buddhist cultural sites. Lac glaze was also confirmed in the Nako monastery (11th–12th century CE) in northern India, which was one of the primary Buddhist centers in the western Himalayas[27]. Furthermore, it is speculated by Dhar that a shellac-based coating was probably applied in the Maitreya temple of Basgo in Ladakh[41]. Lac was also found in the Buddhist grotto wall paintings in the Kucha area (Xinjiang) and at Dunhuang (Gansu). By contrast, other insect-derived red colorants, such as kermes and cochineal, have not been observed so far in wall paintings or other painted surfaces of archeological objects from Northwest China, but only on textiles, with identification of these colorants becoming very sporadic in textiles dating after the 3rd century CE[11,42].

It may be possible to interpret the broad use of lac in Buddhist wall paintings, in opposition to the scant finding of kermes and cochineal. As introduced earlier, the extraction process of lac adheres to the basic Buddhist concept of 'non-killing', whereas the production of kermes and cochineal contradicts this ethos.

Furthermore, in Indian culture, there are records about lac in the *Atharva Veda* written around 1500 BCE, one of the earliest Indian religious sacred texts, where lac was highly praised as 'sister of gods' and 'the shining one' for its medicinal value[43]. This implies that lac already had religious connotations since ancient times. Lac was also appreciated for its excellent dye properties since the end of the Vedic period, and was regarded as one of the *eight major colorants* described in Buddhism sutras [1].

Therefore, an interesting interpretation can be given about lac having a strong relation with artistic Buddhist practices. In opposition, kermes and cochineal are absent in Buddhist wall paintings, and are infrequently found in textiles from the 5th to the 10th century CE. For example, no Dunhuang textile (7th to 10th century CE) has so far revealed the presence of kermes or cochineal, while lac dye was identified in various textiles[8–11].

This possibility strengthens the link between the spread of Buddhism and the adoption of lac dye in Northwest China. Indian Buddhism was introduced into Xinjiang in the 1st century BCE. The creation of Buddhist grottoes first emerged in the Kucha area, where the Kizil and Simsim Grottoes are located, and was an important center of Buddhist culture in Xinjiang. This study confirms that lac was used both as red colorant and/or as a gilding adhesive in the Kizil and the Simsim Grottoes. The caves decorated with lac were built in the flourishing period of Buddhism in the Kucha area (after the 5th century CE). Furthermore, the discovery of lac dye in the painted sculptures unearthed from Astana Tombs suggests that the prevalence of Buddhism in ancient Gaochang may have promoted the use and spread of imported Indian products, such as lac dye. In this context, Shevchenko et al. reported the findings of sesame oil and cattle ghee imported from South Asia in connection with Buddhist cultural items unearthed from the Astana Tombs, and have proposed a close correlation between the development of sesame agriculture and the expansion of Buddhism[44]. Therefore, it is conceivable that besides accelerating the integration of religious ideas, the prosperity of Buddhism also led to cultural and artistic exchanges between ancient India and the Western regions of China, and may as well have contributed to the large-scale application of lac in wall paintings.

Data availability

The author confirms that all data generated or analyzed during this study are included in this published article. Furthermore, primary and secondary sources and data supporting the findings of this study are also available online (https://data.mendeley.com/datasets/7wxgykb78c/1).

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Author contributions
Sihan Zhao: Formal analysis, Methodology. Juan Zhang: Formal analysis, Methodology, Writing - original draft. Ana Albano Serrano: Validation, Writing - review & editing. Diego Tamburini: Validation, Writing - review & editing. Xiaojing Kang: Investigation, Resources. Zhibo Zhou: Investigation, Resource, Writing - review & editing. Yumin Gao: Investigation, Resource. Ling Shen: Investigation. Jiakun Wang: Formal analysis. Jie Yang: Investigation, Resource. Shaowen Shi: Methodology, Validation. Jian Liu and Hui Zhang: Conceptualization, Funding acquisition, Resources, Supervision, Writing – review & editing.

Competing interests
The authors declare no competing interests.

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