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Unveiling materials and origin of reused medieval music parchments by portable XRF and ER-FTIR

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ABSTRACT

In this research, we analyzed two handwritten music parchments, dated back between the late 14th century and the beginning of the 15th century, using non-invasive multiband (MBI) and reflectance transformation imaging (RTI), portable Energy Dispersive X-Ray Fluorescence (p-XRF), and External Reflection FTIR spectrometry (ER-FTIR). The two parchments were discovered in different libraries in Northern Italy, in Milan and Pavia, respectively, as covers of paper books (a manuscript and a printed book). Since the findings represent a valuable evidence of rare Lombard polyphonic music of the late Middle Ages, they have aroused great musicological and philological interest leading to the fusion of humanistic disciplines with multiple analytical set-ups to get insights into their story. The paleographic, stylistic, and musical similarity of the two manuscripts raised questions about their origin: whether the two fragments were originally part of the same musical manuscript disassembled at a later period and reused as book covers, and whether different portions of the manuscripts were written at different stages of time. The study addressed the origin of the parchments through the non-invasive spectrochemical and superficial characterization of the inks and parchment substrate. The results of the proposed multi-analytical approach proved effective and reliable in the morphological and chemical distinction of medieval inks from inks applied in later periods and identifying parchment manufacturing processes and degradation products. In addition, relative ratios Cu/Fe and Zn/Cu were used as fingerprints for black inks confirming the common origin of the two medieval musical manuscripts and the writing of different portions of the manuscripts using the same ink, likely within a short period. Overall, this study highlights not only the synergic contribution of a multidisciplinary approach, but also limitations and analytical strategies to overcome critical drawbacks in the analysis of historical manuscripts.

1. Introduction

Being a durable and stable material, parchment was the most used writing support for documents, books, and manuscripts during the Middle Ages in Europe [1,2] and successively, a key material for recycling. Starting from the 12th century though, parchment was gradually displaced by the cheaper paper [1], a transition that culminated with the advent of printing in the 15th century. Consequently, from the mid-15th century until the late 17th century the practice of dismantling obsolete books and recycling their pages became widespread in Western Europe. Thus, durable well-crafted parchments emerged as a key material for covering printed volumes, preserving numerous manuscript fragments from destruction [3].

Investigating manuscript parchment fragments that have been disassembled from the original volumes and reused for other scopes involves queries about their origin, provenance, dating, and contents. For reused manuscripts that still retain textual significance, recovering their original historical and cultural context is indeed of utmost importance and such queries can only be adequately addressed through a multidisciplinary approach. A useful method to trace this information

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combines a historical and paleographical approach with scientific investigations aimed at identifying the writing materials and the production technologies to gain valuable insights, in the framework of the recently-established research field called "Fragmentology" [4].

From a chemical point of view, parchment is derived from the thickest and innermost layer of skin of calf, goat, and sheep, and the main component is collagen [5–7]. In Europe, the manufacturing process for parchment involved a chemical treatment with lime (alkali) to dehair the skin and remove moisture and grease before scraping the surface and stretching the parchment [5,7]. The so-prepared parchment sheet, or *folio,* turned out to have optimal thickness, solidity, and opacity for writing on both *recto* and *verso* sides several times, scraping, and reusing the pages.

Beyond the parchment substrate, the other essential writing material was the ink. Between the 12th and 19th centuries, the palette of black inks in Western countries was dominated by iron-gallic products [8]. Although different recipes were used in the Middle Ages, iron gall ink was typically produced by mixing four main components: galls, vitriol, Arabic gum, and an aqueous medium. Oak galls were the source of the highest quantity of hydrolysable tannins, gallo-tannins, from which gallic acid oligomers are derived [9]. Consisting of iron sulphate ($FeSO_4 \cdot 7H_2O$), or a mixture of metal sulphates such as iron and copper sulphates ($CuSO_4 \cdot 5H_2O$), the vitriol was obtained from different mines and by various processes [10,11]. Therefore, vitriol was contaminated to varying degrees with many other metals such as copper, aluminium, zinc, and manganese, which could be indicators of the origin of the raw materials [10,12,13]. Despite the comprehension of the chemistry beyond iron-gall inks remains an ongoing topic of research, recently reviewed by Caterino et al. and Melo et al. [14,15] it is widely known that the color of the mixture changes when Fe^{2+} from vitriol reacts with gallic acid to form iron gallate upon oxidation. This product is the stable, water-insoluble, and black complex of Fe^{3+} [16,17]. Finally, Arabic gum was the suspension medium for the colored complex within the aqueous solution.

The use of non-standardized recipes, as well as the source and purification procedures of the raw materials, all contributed to creating a great variability of ink compositions and formulations [8,14] that can be used to distinguish distinct inks within the handwritten text. In this view, the valuable contribution of the analytical techniques in studying reuse handwritten fragments become essential. In recent years, the analytical approach applied to the study of ancient manuscripts has been mostly represented by non-invasive techniques, such as multispectral imaging, X-ray Fluorescence (XRF), Raman, and Fourier-Transform Infrared (FTIR) spectroscopies, to ensure the preservation of the manufact's integrity [6,18–21]. Using different light sources, multiband and hyperspectral imaging allowed the localization of inhomogeneity in ink compositions and the visual recovery of altered texts [22,23]. In certain instances, XRF enabled the qualitative or quantitative investigation of minor and trace components of the ink gaining the identification of fingerprint composition that can be used to propose chronological connections or determine the origin of raw materials [10,11,13,24–26]. Complementary molecular information are achieved by Raman and FTIR spectroscopic analyses which are able to characterize different classes of inks, including organic and iron gall inks [27,28,6], degradation products, and alteration of the paper or parchment substrate [16,29,30]. In addition to those techniques, Proton Induced X-ray Emission (PIXE) [31–33] was occasionally applied to detect impurities and light elements in the inks of historical manuscripts aimed at reconstructing the chronological layering of their application.

In this research, we discuss the scientific investigation of two medieval music parchments, discovered in two distinct renowned libraries in Lombardy (University Library in Pavia and Trivulziana Library in Milan), that were reused as covers for paper books. The extraordinary finding holds significant value as the two parchments contain rare and important evidence of previously unknown polyphonic compositions with sacred and profane French texts of probable Lombard origin dated back to ca. 1400. The codicological, paleographic and musicological similarity of the two music parchments, although they were discovered in different libraries of Northern Italy, suggested to musicologists and philologists that the two fragments could be part of the same codex, and raised the question whether different handwritten portions were coeval, i.e. music and texts on *recto* and *verso* sides [34]. Achieving this information is relevant for identifying the regional features of Lombard medieval music as precisely as possible, in order to study and understand a hitherto unknown repertoire of great musicological interest. For this reason, the aim of the analytical study presented here was to determine the origin of the two music parchments and to establish a potential chronological reconstruction of the writing of the texts and music. To achieve this aim, the potential of an integrated non-invasive and portable multi-analytical set-up was explored to preserve the integrity of the valuable document. Diverse photographic techniques, portable X-Ray Fluorescence and External Reflection Infrared spectroscopy were combined for the first time to reliably characterize the superficial micro-scale features and the chemical composition of the writing materials with a particular focus on the inks. The peculiarity of this research on reused manuscript fragments offered a novel opportunity to highlight limitations and analytical strategies to overcome critical drawbacks in the analysis of historical manuscripts, such for instance the loss of the original historical-cultural context, the adverse state of conservation, and the immovability of the manufact.

2. Material and methods

2.1. The music parchments: TRIV and PV

Two historical music parchments were analyzed in this study. The first parchment is currently conserved at the Archivio Storico Civico e Biblioteca Trivulziana (Castello Sforzesco, Milan, Italy), referred to as TRIV in the paper. It is a bifolio used as a cover of a hand-written manuscript and it contains musical notations and texts written with black ink on red staves Fig. 1-a. The parchment fragment, which is still attached to the book, was discovered by Anne Stone (City University of New York) and tentatively dated between the end of the 14th century and the first decades of the 15th century [35].

The second music parchment bifolio served as a cover of the book *Tractatus de irregularitatibus* by Bartolomeo Ugolini, printed in Venice in 1601, and it is conserved in the Biblioteca Universitaria di Pavia (Pavia, Italy). At the time of discovery, it was only possible to partially read the French-texted polyphonic pieces transcribed in the visible outer part of the cover. For conservative purposes, the bifolio was detached from the hosted volume enabling the display of a polyphonic Credo in Latin on the inner side of the cover. In the article, this fragment is referred to as PV, which included the music bifolio tentatively dated between the end of the 14th century and the first decades of the 15th century [34], and an extra parchment strip successively attached to reinforce the book's spine (PV1 and PV2 in Fig. 1-b, respectively). Apart from the medieval music, PV also includes later handwritten additions: (i) the title of the book inscribed when the book became part of a library's collection occurring after 1601 (referred to as PV1_b), and (ii) the book's title displays in PV2 (referred to as PV2_b).

Inventory numbers of the two medieval manuscripts are reported in Table 1, while high-resolution images are in Supporting Information (Fig.S1 and Fig.S2).

2.2. Analytical approach and techniques

2.2.1. The analysis of historical music parchments

High-resolution photographs at different wavelengths were acquired to enhance the legibility of the texts and help the discrimination of different inks; while non-invasive and portable spectroscopic techniques were selected to gain chemical identification of the parchment substrate, the ink of notes, texts, and red staves, as represented in Table 1 by

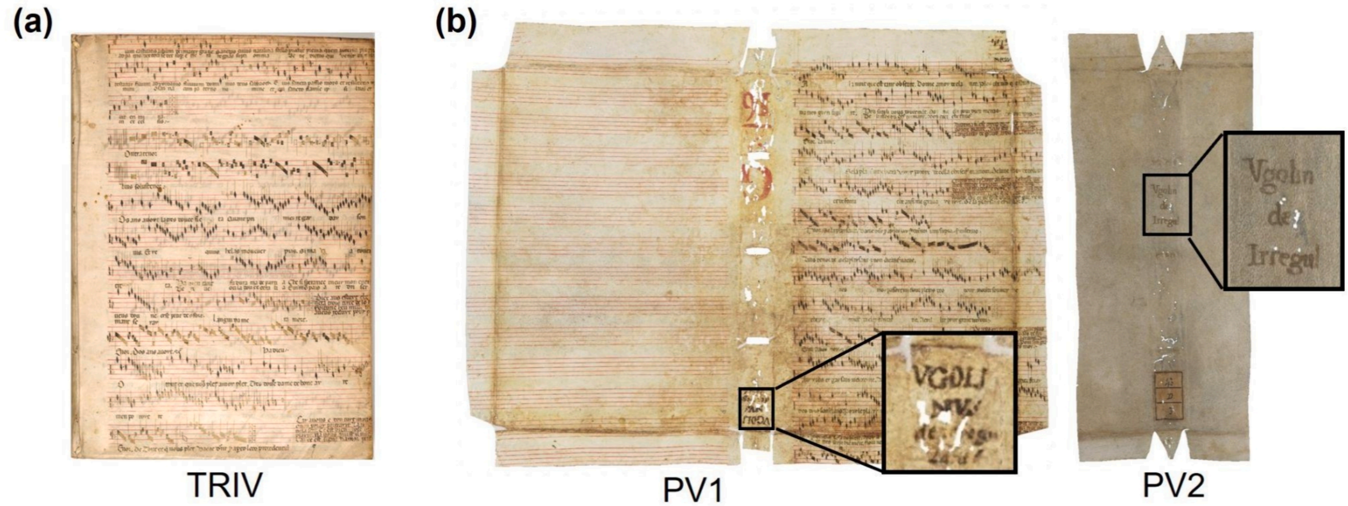
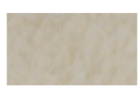


Fig. 1. Historical parchment fragments (a) TRIV discovered in Milan, (b) PV discovered in Pavia containing the bifolio PV1 and the reinforcement parchment PV2. The squares display later additions of writings (PV1_b and PV2_b, respectively).

Table 1
Inventory numbers of the historical parchments and acronyms are reported, along with explicative images representing the different type of materials: parchment substrate, black ink for text or notes, red ink of staves, black ink of book title.

Inventory #	*Name*		*Type & acronyms*				
			Substrate	*Ink text*	*Ink note*	*Ink stave*	*Ink book title*
Cod. Triv. 1759 (binding)			TRIV_s	TRIV_t	TRIV_n	TRIV_p	none
	TRIV						
			PV1_s	PV1_t	PV1_n	PV1_p	PV1_b
		PV1					
Pergamene sparse, scatola 4, n.8	**PV**		PV2_s	none	none	none	PV2_b
		PV2					

typology. In addition, reflectance transformation imaging technique was applied in PV1 to enhance the visualization of the inks thickness, or even layering of inks [36]. In TRIV parchment 170 measurements were acquired with p-XRF, and 13 with ER-FTIR, in correspondence with the representative areas on the *recto* side. In PV1 a similar number of measurements were conducted on both *recto* and *verso* of the bifolio. During analyses, the parchments have been placed on customized supports to prevent the X-ray beam from reaching other surfaces besides the parchment and analysis spots were carefully selected in areas without interference from overwriting or labels. Acquired data were compared to investigate a possible temporal sequence in the writing of different parts (i.e. text and notes, recto and verso), and the origin of the two medieval parchments.

2.2.2. Multiband imaging

Multiband images were acquired with a Phase One IQ3 photographic system (100 MP detector, 16-bit RAW files of 11608 × 8708 pixels), extended to detect radiation from 350 to 1000 nm. For all the acquisitions, a Schneider Kreuznach 120 mm LS f/4.0 Macro lens was used in combination with different filters and light sources to select the appropriate wavelength range for the investigation. Lighting was chosen according to the different purposes, and different filters were mounted on the lens to correctly acquire the images. Two Godox Witstro AD360 flashlights (360 W with light diffuser) were used for the visible and near-infrared images, two UV LED lamps (emission 365 nm ± 10 nm, 3 W power) were used for UV reflected images and visible fluorescence induced by UV radiation.

2.2.3. Reflectance transformation imaging (RTI)

RTI is an imaging technique used for estimating the intensity and direction of the light reflected from an object, with the aim of representing that object under different directions of the incident light, through the interactive re-lighting of the subject [37]. RTI images are created with multiple photographs of the subject shot from a fixed camera position and different but known light directions. The object is then represented by a series of images with varying highlights and shadows that are used to generate a geometrical model of the surface using the normal map rendering. For each parchment, 100 images were acquired corresponding to 100 different light positions using a flashlight source (Godox Witstro AD360). Each image set was processed using RTI builder software (Version 2.0.2) and the output file was visualized interactively with RTI viewer [38].

2.2.4. Portable X-ray Fluorescence (p-XRF)

Analyses were performed using a portable energy-dispersive spectrometer ELIO (XG Lab, Milan, Italy; Bruker Optics, Billerica, MA, USA) equipped with a rhodium anode. The diameter of the beam spot on the surface is approximately 1.3 mm and measurements were performed at 40 kV tube voltage, 60 μA tube current for 90 s. Data were processed by ELIO 1.6.0.29 software. Due to the working conditions in air, the detection limit is Z<13 [39]. The discrimination of black inks was based on similar values of copper/iron, zinc/iron, and zinc/copper ratios. For data processing, the net area counts for each element were normalized to the values of Rh-Kα (20.21 keV).

2.2.5. External reflection FT-Infrared spectroscopy (ER-FTIR)

A portable Alpha spectrometer (Bruker Optics, Etringen, Germany; Billerica, MA, USA) equipped with a SiC globar source, a permanently aligned RockSolid interferometer with gold mirrors, and a DLaTGS detector was used. A gold flat mirror was used as the background. Measurements were performed in external reflection mode with a 23°/23° optical layout, at a working distance of 15 mm and a diameter of 3 mm. Measurements were acquired within the range 7500–365 cm^{-1} with a spectral resolution of 4 cm^{-1}. For each point, 145 spectra (3 min scan time) were acquired, averaged, and transformed in log(1/R) (R=reflectance) or absorbance by Kramers-Kronig transform using OPUS 7.2 software package.

3. Results and discussion

3.1. TRIV music parchment

The elemental composition of the substrate TRIV_s is dominated by calcium (Ca-Kα 3.69, Kβ 4.01), together with low counts of iron (Fe-Kα 6.41, Kβ 7.05), potassium (K-Kα 3.31), and sulfur (S-Kα 2.33) Fig. 2. As commonly reported in literature for Western parchment, the use of lime, i.e. $Ca(OH)_2$, for skin dehairing justifies such high counts of Ca, while Fe, K, and S may be related both to the substrate composition and to contamination from the inked areas [40]. To establish which element significantly increased in comparison with the substrate, a threshold limit was calculated and reported as the limit of detection (LOD) in Table 2. LOD corresponds to the averaged net area counts + 3σ (where σ is the standard deviation), normalized to Rh-Ka counts, of each element measured in the parchment substrate.

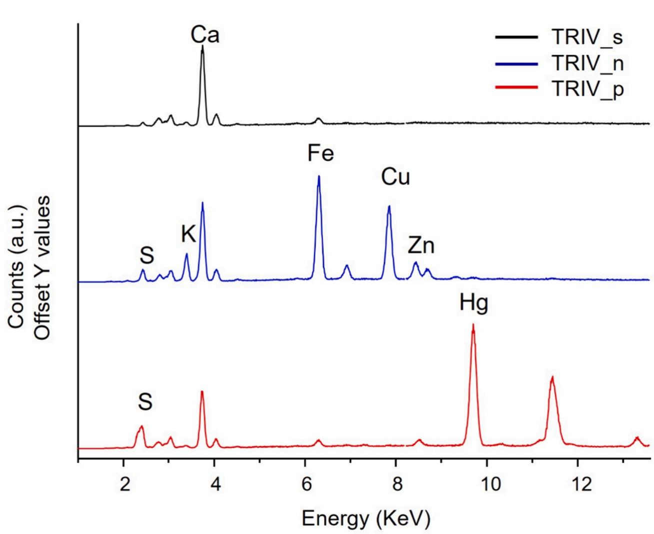


Fig. 2. XRF spectra of the parchment substrate (black line), black ink of notes (blu line), red ink of the staves (red line) of TRIV. Characteristic elements are reported for each spectrum. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

Table 2
Average of net area counts normalized to Rh-Ka [39]. The limit of detection (LOD) was calculated for the parchment substrate as [average + 3σ], where σ is the standard deviation. Values below LOD are marked with asterisks; n.d. = not detected.

element	TRIV_s	LOD	TRIV_n	TRIV_t	TRIV_p
S	0.92 ± 0.06	1.10	3.9 ± 1.2	4.0 ± 0.8	10.9 ± 1.2
K	1.08 ± 0.13	1.47	8 ± 3	3.3 ± 2.1	0.79 ± 0.23*
Ca	27.5 ± 2.6	35.3	29 ± 7*	42 ± 9	21 ± 3*
Mn	0.62 ± 0.08	0.86	0.80 ± 0.25*	0.88 ± 0.17	0.59 ± 0.05*
Fe	2.2 ± 1.1	5.5	38 ± 15	22 ± 9	2.64 ± 0.26*
Cu	0.28 ± 0.11	0.61	27 ± 11	14 ± 7	0.10 ± 0.05*
Zn	0.28 ± 0.16	0.76	5.7 ± 2.3	4.2 ± 2.6	0.38 ± 0.03*
Hg	n.d.	n.d.	n.d.	n.d.	55 ± 19

According to the normalized counts of the elements in Table 2, p-XRF measurements on notes and texts (TRIV_n and TRIV_t, respectively), both written with black ink, revealed statistically significant increases in Fe, copper (Cu-Kα 8.05, Kβ 8.91), and zinc (Zn-Kα 8.64, Kβ 9.57) counts, compared to the parchment substrate. Besides these elements, which showed values above the LOD, counts of S and K also appreciably increased. The detected elements are consistent with the hypothesis of iron gall ink: Fe, Cu, Zn, and S are attributable to the vitriol, containing likely Fe and Cu sulphates with Zn impurities, while K is attributable to Arabic gum [41]. The hypothesis of iron-gall inks is supported by multiband imaging as well, showing a dark color in UVIF image while starting to lose opacity in IRR image at 850–1000 nm (Fig. S1-c), although it does not become transparent as tannin ink behaves in the near-IR range [12].

A completely different composition was found for the red staves: mercury (Hg-Lα 9.98, Lβ 11.83, Lγ 13.82) and S were principally detected, along with traces of lead (Pb), while Hg and Pb remained undetected in the parchment substrate. This elemental composition results in the possible use of natural cinnabar, as often reported in the literature [42,43], or vermillion mixed with lead red [44]. The evidence of cinnabar/vermilion is consistent with the transparency of the staves in the IRR image due to the reflection of the near-IR radiation by these pigments.

According to p-XRF results, ER-FTIR spectra acquired on the substrate TRIV_s clearly reveal the signal of proteins of the parchment. Diagnostic bands of collagen are recognized in the pseudo-absorbance spectrum by combination bands and overtones at 5140, 4893, 4597, 4385, 3985 cm^{-1} [45], and by the derivative-like signals of amides marked with asterisks in Fig. 3-a. In the KKT absorbance spectrum in Fig. 3-b, amide peaks are visible with maxima at 1661 for amide I (ν C=O and δ N-H of polypeptides backbone), 1551 cm^{-1} for amide II (ν C-N and in plane δ N-H), at 1450 cm^{-1} (δ CH_2), and 1230 cm^{-1} for amide III (complex mix of in plane δ N-H and CH_2 wagging) [30,46]. In addition to protein, the pseudo-absorbance spectrum displays a Reststrahlen band at 1420 cm^{-1} together with a derivative-like signal with maximum at 878 cm^{-1} that unequivocally characterizes the presence of calcium carbonate (ν_3 and ν_2 of CO_3^{2-} vibrations [47], marked with triangles in Fig. 3-a) likely formed from the reaction between the lime of the dehairing treatment and atmospheric CO_2 transforming into $CaCO_3$ [1].

When compared to the substrate, the ER-FTIR spectrum acquired on large black inked areas (TRIV_n) showed the broadening of the C=O band from ca. 1660 to 1670 cm^{-1} in the KKT spectrum. This behavior could be explained by the overlapping of ester groups of iron gallate (ν C=O around 1680 cm^{-1} [8]) to the amide I of the parchment substrate, supporting the hypothesis of iron-gall ink. To highlight the shift of the carbonyl peak, pseudo-absorbance spectra and the first derivative of reflectance of TRIV_s compared to TRIV_n are reported in Supporting Information Fig. S3. Unfortunately, other characteristic peaks of iron-gall ink, reported in the literature, were not clearly distinguishable in the ER-FTIR spectra because of distortion and overlap with other signals. In fact, distortions occurred due to oxalates: Reststrahlen bands at 1630

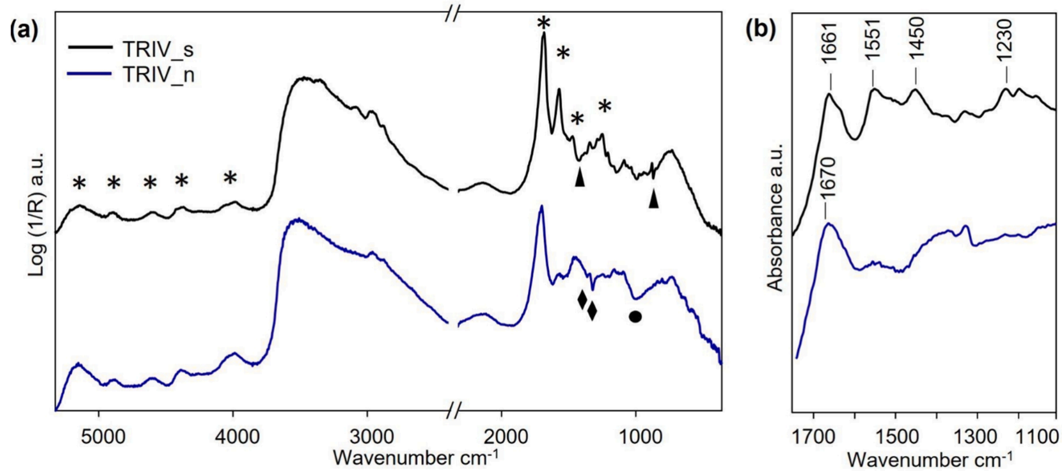


Fig. 3. ER-FTIR spectra in (a) pseudo-absorbance and (b) KKT absorbance of the parchment substrate TRIV_s (top black line) and ink of notes TRIV_n (bottom blue line). Diagnostic signals are marked for proteins (asterisks), calcium carbonate (triangles), oxalates (rhombus), and Arabic gum (circle). (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

and 1320 cm^{-1} (respectively for ν_{as} C=O and ν_s C=O of calcium oxalates CaC_2O_4 [48]), and at 1360 cm^{-1} (marked with rhombus in Fig. 3-a) likely attributed to copper oxalate [49,50]. Numerous authors reported the generation of oxalates as a byproduct of the degradation of Arabic gum and gallnut tannin within the ink; these components undergo glycine degradation, ultimately yielding oxalic acid that reacts with available cations [28,29,50]. Therefore, the detection of oxalates provided further confirmation for the gall ink, together with the detection of the broad Reststrahlen band around 1010–1020 cm^{-1} possibly related to Arabic gum (ν C-O [51]).

Regarding a possible differentiation between the ink used for notes and texts, we referred to p-XRF data. Despite the same primary elements Fe, Cu, and Zn, little difference was found in their counts: lower counts of Fe, Cu, and Zn resulted in TRIV_t than TRIV_n, as displayed in Fig. 4. A reasonable explanation of this behavior was attributed to the thickness or density of the ink mixture, and thus to the amount of material detected by p-XRF. A visible difference in the density of the two groups of ink was observed: the notes resulted with a more inhomogeneous texture and thicker appearance compared to the texts and light ink strokes (e.g. the stem of the notes and other musical notations like sharp # sign), shown in Fig. S1-d. In support of this observation, we analyzed the areas where the ink was visibly altered, likely detached from the substrate, or eroded, but still recognizable in visible and UV light (referred to as “altered ink” in Fig. 4-a and Fig. S1-e). Measurements on visibly altered ink spots revealed an expected distribution of Fe, Cu, Zn counts ranging from high to low values, overlapping with those of unaltered notes and letters. In accordance with this observation, the slope of regression lines in Fig. 4-a described comparable trends of Fe-Cu for texts, notes, and altered ink areas, which means comparable elemental ratios, i.e. Cu/Fe or Fe/Cu, thus suggesting the use of inks with the same recipe. Another evidence of a diverse dilution degree of the inks was described by the Ca counts which resulted to be higher in TRIV_t, if compared to TRIV_n; such a trend could be explained by the presence of an extra medium added to dilute the mixture. Overall, these results suggested that music and text were possibly written using the same ink, likely with different dilutions, along a period where ink recipe and raw materials were maintained constant.

3.2. PV1 music parchment

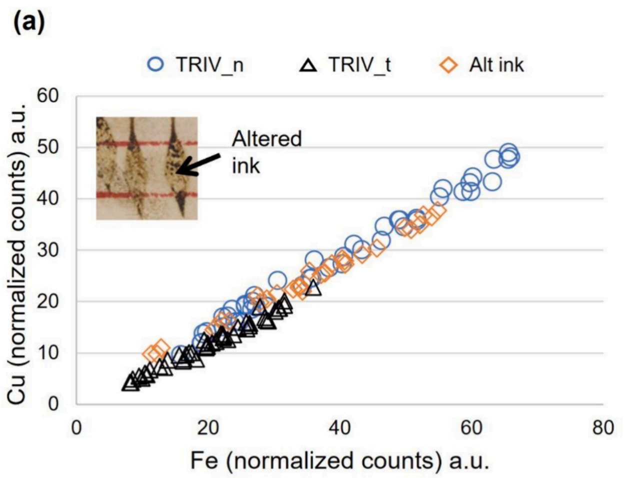
Analyses performed with p-XRF on the substrate (PV1_s) revealed an elemental composition dominated by Ca with traces of Fe, K, and S. Consistently with the observations made for the substrate of TRIV, this result implies a manufacturing process of the parchment where a lime treatment was used to dehair the skin. Concerning the black ink of notes and texts, a chemical differentiation was researched also between the *recto* and *verso* sides of PV1, to investigate whether a possible temporal sequence of writing was observed between the two pages written in French and Latin, respectively. Based on multi-band images displayed in Fig. S2, all the black inks showed the characteristic behavior of gallic inks: dark/black in visible and UV light, while starting to lose opacity in the near-IR region, although it is reported that gallic inks became transparent over 1200 nm [12]. In accordance with the imaging examination, p-XRF analysis proved the same elemental composition for PV1_n and PV1_t, with Fe > Cu > Zn peaks as characteristic features. Fig. 5-a displays Fe and Cu counts measured on notes and texts in both *recto* and *verso* sides of PV1. Lower counts of the elements were measured in the texts of PV1 compared to the notes, independently from the sides. As for TRIV music parchment, this behavior was interpreted as a difference in the thickness or dilution of the inks rather than in the chemical composition. The processed RTI image in Fig. 5-b reinforces this hypothesis: a difference in thickness between the notes and texts is evidenced, as well as the ink detachment in some areas of the notes. PV1_n and PV1_t, either written on the *recto* or *verso*, resulted chemically comparable to each other with similar slopes in the regression line for Fe-Cu, thus describing comparable ink formulation (slope for *recto* 0.7655, and slope for *verso* 0.7193).

When compared to the substrate PV1_s, ER-FTIR signals of the black ink PV1_n and PV1_t display the decreasing of the protein bands and the broadening of the carbonyl signal towards longer wavenumbers. In addition, the formation of calcium oxalate around 1615 and 1320 cm^{-1} showing higher intensity in PV1_n than PV1_t was also observed. Pseudo-absorbance and the first derivative of reflectance are reported in Fig. S3 with marked bands for protein and oxalate. These results suggested that iron-gall ink was used to handwrite both the music notes and texts.

Consistently with the observations for TRIV parchment, the elemental composition of the red staves of PV1 was characterized by high counts of Hg and S, implying the use of cinnabar or vermillion, as confirmed by IRR image in Supporting Information. Due to the thin line of the stave ER-FTIR measurements were not acquired.

3.2.1. Later addition of inks in PV1 and PV2

For comparison, two other types of black ink added at a later time than medieval music were considered: i) PV1_b applied after 1601, and ii) PV2_b applied on the reinforcement parchment later to PV1_b.



	R^2	Slope
TRIV_n	0.9878	0.7351
TRIV_t	0.9781	0.6366
Altered ink	0.9913	0.6431

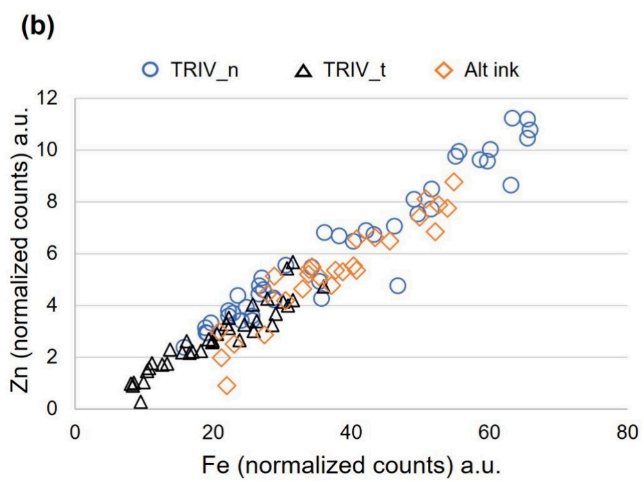


Fig. 4. XRF plots of Fe-Cu (a) and Fe-Zn (b) counts normalized to Rh-Kα are reported for: black notes (blue circle), letters (black triangle), and altered ink areas (orange rhombus). Linear correlation coefficient (R^2) and regression line slope are reported for Fe-Cu. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

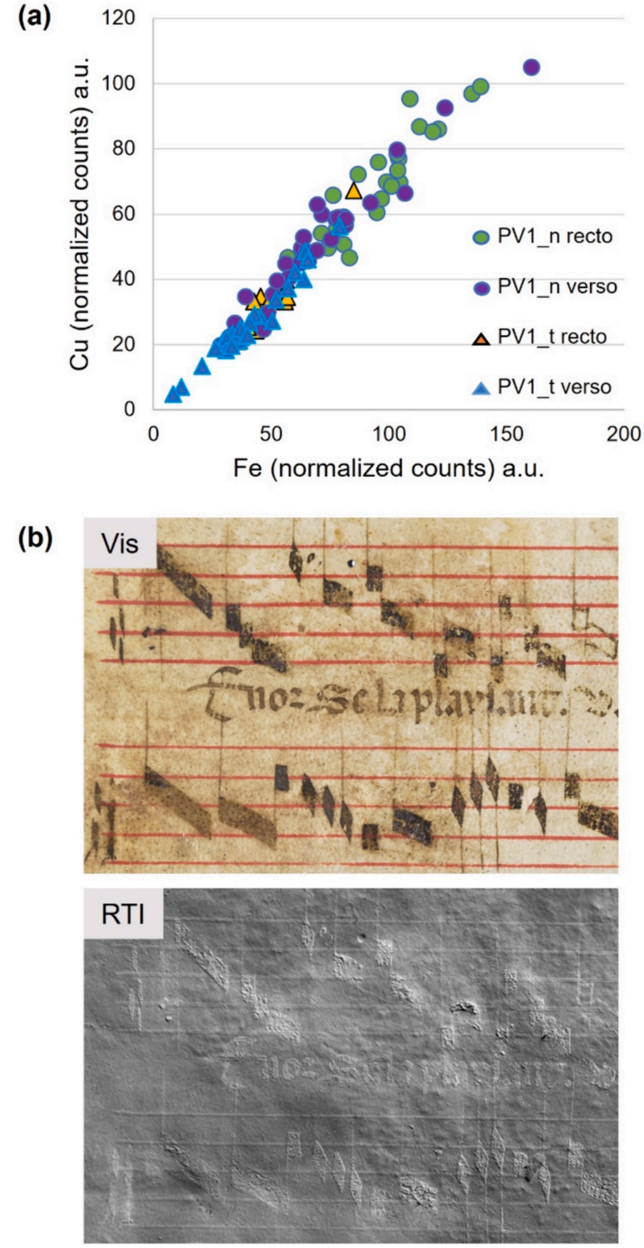


Fig. 5. (a) XRF plots of Fe-Cu counts normalized to Rh-Kα are reported for black notes (circles) and texts (triangles) of PV1, both recto and verso sides; (b) a detail of the notes and text is reported in visible light and processed RTI image.

Significant differences emerged in the p-XRF spectra in Fig. 6: when compared to PV1 black inks, higher counts of K, Fe, and Zn were measured in PV1_b, while a significant increase of Ca and decrease of Fe were measured in PV2_b. No evident differences emerged from the behavior of the two inks by multiband imaging (Fig. 6-b).

Being these inks added later than black inks of PV1_n and PV1_t, a difference in the chemical composition in terms of relative ratios Cu/Fe or Zn/Fe can reasonably suggest a temporal/chronological evolution of the inks likely due to the supplying of different ingredients or changing of the recipes.

3.3. Inks comparison and origin attribution of TRIV and PV music parchments

Analytical results of p-XRF in Sections 3.1 and 3.2 revealed that Fe > Cu > Zn counts characterize the medieval black ink of both TRIV and PV fragments. However, since the measurement of elements can vary considerably due to the intrinsic inhomogeneity of the ink (i.e., thickness, degradation, surface properties), it becomes challenging to distinguish different inks that show the same characteristic elements. To help this discrimination we experimentally defined a confidence range within which similarities in ink composition can be reliably identified. A biplot reporting Cu/Fe versus Zn/Cu was used facilitating the grouping of similar ink compositions and accounting for the contribution of both major and minor elements.

Three measurements were acquired on both single notes and letters of TRIV to evaluate the dispersion of the mean value of Cu/Fe and Zn/Cu ratios primarily attributable to the ink measurement. This yielded a relative standard deviation (RSD) mostly below 5 %. The dispersion of the values was then measured across different notes and letters of TRIV,

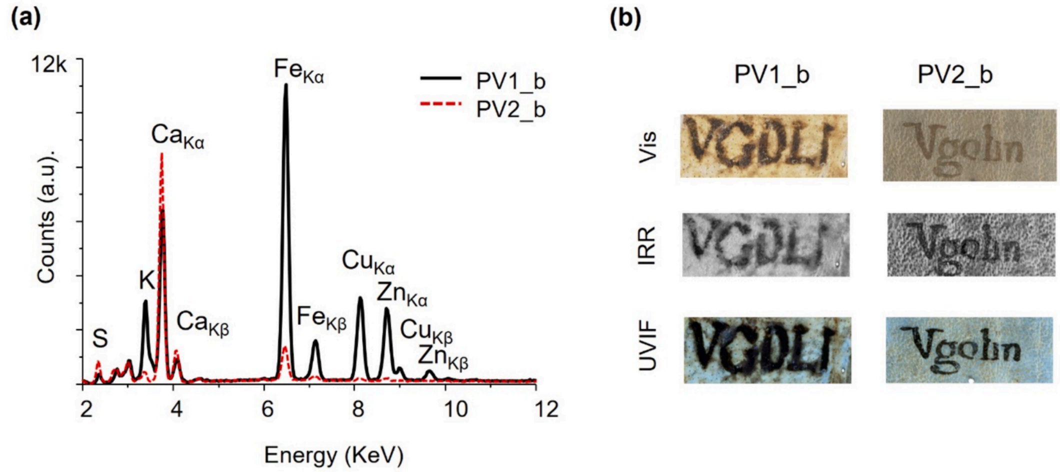


Fig. 6. (a) XRF spectra of later addition of black inks PV1_b and PV2_b, characteristic elements are reported for each spectrum; (b) multiband imaging in visible light (Vis), Infrared Reflectography (IRR), and UV-induced fluorescence (UVIF).

showing an increment of RSD (within 10 %) due to uneven characteristics of the writing. Taking into account visibly altered inks, a range of values was defined for Cu/Fe and Zn/Cu of TRIV to group the inks with unique chemical composition. The range was established as [average value ± 3σ], where σ is the standard deviation. The application of this criterion facilitated the identification of a specific group, which is illustrated with ellipses in Fig. 7, wherein the measured values are deemed not statistically distinct. Both TRIV_n and TRIV_t measurements lay inside the range, partially overlapping each other. Comparing the results of TRIV with those of PV parchments, the measurements of PV1_n and PV1_t inks falling into the same group of TRIV implying that the inks used in the two medieval music parchments were chemically comparable to each other. In contrast, data from the later added inks, i.e. PV1_b and PV2_b, showed a significant distinction from the medieval ones localizing in a different area of the plot.

4. Conclusions

The uniqueness of this research offered the opportunity to propose a methodological non-invasive approach to study reuse manuscripts, specifically two music parchments dated back to the Middle Ages that were reused as covers of different paper books found in two distinct historical libraries in Italy. In support of codicological, paleographic and musicological observations that found similarities between the two musical manuscripts, the proposed analytical approach allowed us to investigate whether the handwritten parchment fragments were originally part of the same music manuscript, and whether different portions of written music and texts were coeval, contributing to increase our knowledge of the rare medieval Lombard musical tradition. The combination of multiband imaging, RTI, p-XRF and ER-FTIR was used for the first time on ancient reuse manuscripts. It enabled the reliable identification of the chemical composition and manufacturing processes of the parchment and medieval inks and helped the visualization of the inks thickness by non-invasive and portable techniques overcoming critical aspects often encountered in the analysis of ancient manuscripts. In agreement with multiband imaging, complementary results of p-XRF and ER-FTIR analysis agreed with the identification of the dehairing treatment of the two parchments with lime, characteristic of the European manufacturing process during the Middle Ages. Also, the analyses carried out on black and red inks revealed the presence of iron-gall used for the music notes and text, and the use of cinnabar/vermillion for the staves of both TRIV and PV1 parchments. A similar chemical composition was identified for the ink of both notes and letters, with Fe > Cu > Zn as a characteristic feature, independently from the historical parchment; while differences in the dilution degree of the ink used for the notes or texts were suggested by the elemental counts and processed-RTI image. Statistical calculations on the relative ratios Cu/Fe and Zn/Cu permitted grouping inks with similar chemical characteristics, assessing a comparable ink composition for different portions of the manuscripts, i.e. notes and texts, *verso* and *recto* of PV1. From these results, it was hypothesized that the writing of music and texts occurred during a short period using the same ink recipe and raw materials. Moreover, this type of data processing supports the conclusion that originally the two bifolios, TRIV and PV1, were part of the same music manuscript that was disassembled for recycling and reusing as the cover of distinct books. By the synergic contribution of humanistic disciplines with the proposed non-invasive multi-analytical set-up relevant information was accessed to unveil musicological, historical, and cultural aspects of the regional features of medieval Lombard music between the late 14th and early 15th centuries. In this view, the contribution of the proposed analytical techniques, here applied for the first time in studying reuse manuscript fragments, stands as a valuable tool for accessing indications about the origin and chronological sequences in the writing, increasing our knowledge about a – so far- unknown musicological repertoire of great relevance.

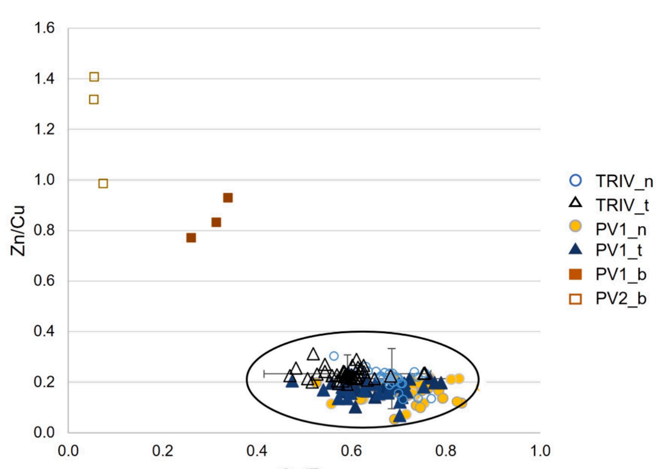


Fig. 7. Plots of Cu/Fe and Zn/Cu ratios measured by p-XRF on black inks of TRIV, PV1 and PV2.

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CRediT authorship contribution statement

F. Volpi: Writing – review & editing, Writing – original draft, Visualization, Supervision, Project administration, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. **G. Fiocco:** Writing – review & editing, Visualization, Methodology, Formal analysis, Conceptualization. **M. Gargano:** Writing – review & editing, Investigation, Formal analysis. **M. Albano:** Writing – review & editing, Methodology, Formal analysis. **A. Calvia:** Writing – review & editing, Investigation. **F. Saviotti:** Writing – review & editing, Investigation. **C. Delledonne:** Writing – review & editing, Formal analysis. **C. Lee:** Writing – review & editing, Formal analysis. **T. Rovetta:** Writing – review & editing, Formal analysis. **M. Malagodi:** Writing – review & editing, Resources.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.microc.2024.111224.

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