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Ink and support characterization of typologically established papyrus groups from the Palau-Ribes collection

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Abstract

Most of papyrus documents have reached us in a very fragmentary state due to their excavation conditions and, because of the early history of papyrology, without an archaeological contextualisation. Traditionally, papyrologists have only relied on the bibliological characterisation of the documents they work on to provide such a chronological, geographical and socio-cultural context within which to fully understand the texts they edit and study. This study, following previous research on ink characterisation of papyrus documents, uses infrared spectroscopy and SEM/EDX for the analysis of ink and the papyrus surface. To this purpose, 67 samples of coherent groups of papyri from the Palau-Ribes collection in Barcelona, organised according to chronological, geographical and bibliological criteria, have been analysed to identify trends in their composition that may contribute to their characterisation and further enlarge our knowledge of written culture in Antiquity. The samples have been categorised into seven groups: Papyri that are (1) dated to II CE; (2) dated to VI CE; (3) from Oxyrhynchus; (4) from the Monastery of Bawit; (5) written in chancery hands; (6) written in bookhands, and (7) brown ink. All samples have been analysed using infrared spectroscopy, and SEM/EDX has also been used for analysis of a small subset of sample in order to confirm the presence of iron, sulphur and other elements. The results confirm previous findings in iron-gall ink distribution for texts written in bookhands from the Byzantine period, and adds to the characterisation of other groups, such as the Bawit one, with high levels of lignin in the composition of the writing surface, or the degradation of the cellulose shown for the Oxyrhynchus group, suggesting peculiar traits in the manufacturing and conservation of written documents from ancient times.

Keywords: Ink analysis, Ancient papyrus manuscripts, Iron-gall ink, Carbonic ink, Infrared spectroscopy, SEM/EDX, Palau-Ribes collection, Bibliological characterization, Oxyrhynchus, Bawit, Roman period, Byzantine period

Introduction

Application of elemental and molecular analytical techniques, especially those which are non-destructive, to studying materials related to cultural heritage has dramatically increased during the last few years [1–4]. Determining the composition of papers, parchments, papyrus and inks of ancient manuscripts has been one of the goals in museums, archives and libraries housing valuable documents for some decades. The possibilities offered by this knowledge when combined with that provided by the study and the analysis of the texts themselves in terms of their contextualization and interpretation has recently started to reveal itself to the papyrological community [5–8].

Unlike medieval codices and other written materials that in one form or another have been kept in libraries or archives, be it public or private, to the present day of their history, papyri are for their most part archaeological findings: they have come back to light after having been buried in the sand for centuries as the result of mere chance or excavations more or less methodologically

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conducted according to archaeological criteria [9]. This is determining for the history of papyrology and its methods, since particularly at its beginning at the turn of the twentieth century, scholars were almost uniquely interested in recovering the texts, but not so much the objects, that had been long lost from antiquity. Papyrus was for centuries the noblest writing material used in the Mediterranean basin. However, the immense majority of papyrological findings come from Egypt, because of the low soil humidity and because large areas in the desert were left undisturbed for centuries after many towns and cities were abandoned in late antiquity [10]. When these papyri have reappeared in modern times, they are often very fragmented and, in most cases, severely damaged. This is due not just to the conditions endured under the sand, but also because many of them come from rubbish deposits where they had been disposed of in ancient times.

The general lack of documentation, especially during the first decades of papyrological activity, about the circumstances that surrounded the discovery of the papyri and their arrival in modern collections make it difficult to place these objects and the texts they carry into their geographical, chronological and cultural context.

Except for the relatively few cases where the document carries a date or specifies its provenance, papyrologists rely on internal evidence or the palaeographical analysis to contextualize the document in terms of time, space or cultural milieu. This is almost always so in the case of literary papyri, i.e., those containing pieces of pagan or Christian literature which, unlike medieval codices, never provide the circumstances of the copy: scribe, place or date when it was executed, etc. Thus, the study of aspects such as the quality of the *charta* (the manufactured papyrus stems that produce the writing material), the *mise en page* of the text, the use of the space, or the style of writing chosen to produce the document, among others, is essential to understand the text by anchoring it to the circumstances that may ultimately explain its existence. For instance, it has been long known to papyrologists that changes in the manufacture of the papyrus plant as a writing support and the composition of the inks took place in the transition from the Roman (s. I–III) to the Byzantine period (s. IV–VII) of Greco-Roman Egypt [11, 12]. This is how the use of analytical techniques for the characterisation of inks and writing supports of ancient documents may be of valuable help in order to determine the circumstances of their production: by elucidating the composition of different inks for different purposes or particular characteristics in the process of manufacturing the papyrus plant we may come to identify very significant aspects of the material culture that produced our oldest written heritage.

In this paper, following previous work conducted by our team on ink characterization of documents belonging to the Palau-Ribes collection [5, 7, 13], we have tried to identify significative patterns in the elemental and molecular composition of the inks and writing material of a selected number of specimina from the collection. These specimina have been organized according to papyrological criteria into coherent groups from either a chronological, geographical or cultural point of view, in the hope that this type of analysis may reveal itself as a valuable tool to support papyrological findings. This study has used infrared spectroscopy for characterisation of papyrus and inks in 67 samples from the Palau Ribes papyri collection. A subset of samples have been also analysed using SEM/EDX in order to confirm the presence of iron, sulphur and other elements.

The particles collected for infrared spectroscopy are not destroyed allowing other analytical determinations, in this instance characterisation using SEM/EDX. This has contributed to sample economizing, as has the fact that, in many cases, one and the same sample has been included as representative of different papyrus groups. For the rationale behind sample choosing see below.

Materials and methods

Instrumentation

Infrared spectrometer

An infrared microspectrometer Thermo IN10MX instrument (Thermo Fisher Scientific, Waltham, MA, USA) has been used. The instrument has a mercury cadmium telluride (MCT) detector, KBr beamsplitter and Ever-Glo air-cooled long lasting source. The wavenumber range measured was between 4000 and 650 cm^{-1} with a spectral resolution of 4 cm^{-1}. A total of 64 scans were accumulated for each of the spectra shown in this work. Samples have been measured in transmission mode with a Thermo Scientific™ Micro Compression Cell and Diamond Window. Background was collected from a clear spot in the diamond window, using the same aperture as that used for samples.

Scanning electron microscope

A JEOL Scanning Electron Microscope Model JSM-6480LV (JEOL Ltd., Akishima, Tokyo, Japan), equipped with an energy-dispersive X-ray detection system (Oxford D6679 EDX detector), operated under recommended conditions (15 kV acceleration voltage and 5 nA probe current). The collection time was 400 s, whilst the working distance was 10 mm. In the first place, several high-quality photographs were taken, for which the spot size employed was 20 nm. Later, in order to analyze with the Energy-Dispersive Spectrometry, it was convenient to

open the spot size (40 nm) to maximize the amount of energy that reached the detector.

Papyri description

We have taken papyri specimina belonging to the Palau-Ribes collection and grouped them according to four criteria: chronology, geography, palaeography and aspect of ink colour, resulting in seven groups:

1. Dated to II CE; the peak of written production in the Roman period.
2. Dated to VI CE, the Byzantine period.
3. Oxyrhynchus, capital of nome in Middle Egypt which produced the largest number of papyrus documents to the present date.
4. Monastery of Bawit, in Upper Egypt, representative of the monastic life and its socio-cultural impact on the surrounding areas in the Byzantine period.
5. Chancery hands, i.e., writing styles typically used in the Egyptian administration.
6. Bookhands, i.e., writing styles typically used in book production of high standards.
7. Brown ink, a feature perceived by papyrologists to have changed in the characterization of inks in the Byzantine period.

The composition of the ink and writing support of each of the selected papyri have been studied with a view to finding patterns matching some of the above criteria. Additional file 2: Table S1 shows the groups of samples analysed in this study. In some cases, the same sample may belong to different groups (e.g., groups 2 and 6; a papyrus dated to VI CE and written in a bookhand). We should add in this respect that the basic papyrological opposition between literary texts and documentary texts runs across the sampling criteria (except, naturally, the chancery and bookhands groups, that contain only documentary and literary papyri, respectively. See also the commentary about the Bawit group in the conclusions). Also originating from the history of the discipline, due to the especial attention that the first papyrologists paid to texts containing literature, either pagan or Christian, which were thus specifically labelled as "literary" as opposed to the rest, that remained simply "documentary", this opposition, although challenged because of oversimplistic, still remains operative in papyrology, inasmuch as other aspects, apart from the contents, tend to be specific of each group. We will see that the distinction is relevant also to the purpose of this paper.

Sample handling

Fibres of clean papyrus and ink particles were selected from each of the specimina. At least three particles from the same sample were chosen to confirm homogeneity. A careful selection of 11 samples were also measured by SEM/EDX.

In many cases, two or three different particles taken randomly from the same sample were analysed to determine reproducibility.

Single particles or fibres, as small as 20 μm, were removed from the ink or clean papyrus using a scalpel or tungsten needles. They were placed on diamond cells (2 mm diameter) and pressed against another diamond window to reduce the thickness and allow the sample to be measured in transmission. Figure 1 shows details of the sampling method and the final image of the particle on the diamond window after pressure. The original particle was ~ 50 μm and the final sample size corresponds to ~ 200 μm.

The ink particles for SEM/EDX measurements were collected in the same manner as those for infrared spectroscopy. After collection they were fixed to a graphite adhesive to avoid being removed from the chamber by the vacuum.

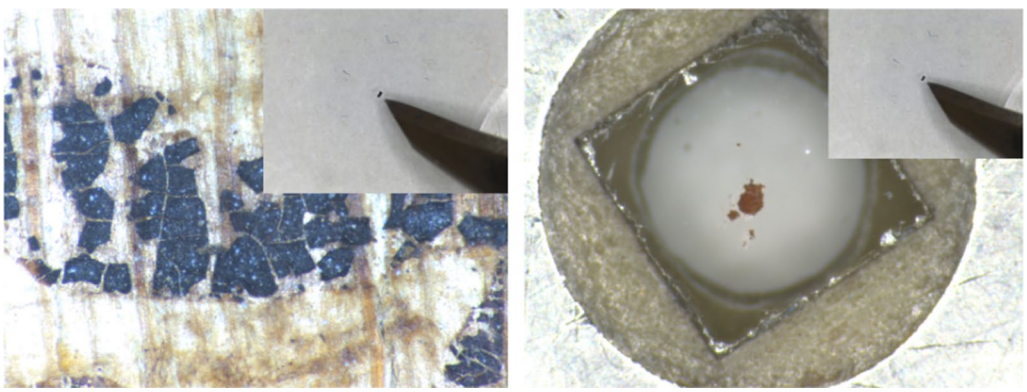

Fig. 1 Different examples of sampling performed to extract very small fragments from inks

Statistical analysis

We used the principal components analysis (PCA) and linear discriminant analysis (LDA) to find the best way to classify the papyri (*Bawit* and *Oxyrhynchus*). These useful tools for data analysis allowed us to better understand the differentiation of the twenty-two papyri between two groups according to their origin. PCA uses the information provided with the data and obtains the majority of variance. By the LDA, the number of groups must be known initially. A careful revision of the classifications obtained by LDA should report, a priori, an expected logical classification. Otherwise, the number of groups can be modified until a satisfactory differentiation is obtained. In LDA tool, the conditional probability density functions are presupposed to present normal distributions with mean and covariance. LDA presents a good precision when homoscedasticity is established. However, the groups obtained in absence of this condition can also reproduce the data classification properly. Another difference between PCA and LDA refers to the fact that LDA tries to generate differences between the data by the group's classification. However, PCA does not consider the differences between the data for the analysis. The software used for the multivariate analyses were OriginLab 9.0 and SPSSv.26.

Results and discussion

What can we find in ink and papyri samples?

Analysing inks on cellulose support has the problem of the strong interference of the matrix. Molecular analysis of cellulose using infrared or Raman spectroscopy has the inconvenience of interfering intensive bands in the fingerprint region, where the spectroscopic information is more useful. This problem could be solved by isolating ink from the support but, in many cases, and depending on the kind of ink and the papyrus manufacturing process, the mixture of ink with fibres is so strong that it is difficult to avoid cellulose information. In some samples, the ink seems to be covering the surface of the papyrus and can be removed easily with no interference of papyrus fibres. In many other cases, the ink and fibres are deeply inter-mixed and it is difficult to remove only the ink.

In addition, differences in writing strokes are sometimes present in the same papyrus. Homogeneity of ink content could be inconsistent depending on many factors, such as how recently the ink was prepared, how much ink remained in the inkwell, or the precipitation of salts at the bottom of the inkwell. Therefore, our FT–IR spectra will be used as an example of the possible interferences that can be found in an antique ink sample. Identified materials include: cellulose, lignin, protein, starch, oxalate, sulphate, carbonate, kaolin and quartz. The last three materials could originate from the surrounding environment, and the remainder are typically found in manuscripts from different origins, either in the writing supports or as components in inks.

Some papyrus samples show the typical absorption signals of lignin, among which stand a shoulder at 1715 cm^{-1}, probably due to carbonyl groups [14], and two bands centred at 1515 and 1270 cm^{-1} which could be attributed to a stretching and deformations vibrations, respectively, of different aromatic components of lignin [15, 16], as seen in Fig. 2A. Even though the first assignment could be due to oxidation of the papyrus and the others should be taken with care as the samples are not as clean as desired, we have verified experimentally, after carrying out many spectra, that these three bands always stand out when lignin is present. Other bands are similar to those of cellulose. Lignin is a complex natural organic polymer responsible for the structural support tissue of plants forming cell walls. Its detection in some samples could be due to a certain papyrus quality; the FT–IR spectra of all papyri analysed from Bawit were found to have lignin bands, see below for further discussion.

Other spectra collected from a few samples have bands due to protein and starch (Fig. 2B). Both are considered binders and have been found in manuscripts, mixed either with papyrus or ink [17]. The presence of protein could also be a consequence of contamination during the manufacturing of the papyrus support or the handling of the specimina, even sampling by technicians. Protein has two characteristic amide absorption bands [18] at 1650 cm^{-1}, assigned to C=O stretching vibration [19, 20], and at 1550 cm^{-1}, associated to C–N stretching [19, 21] and N–H bending vibrations [19, 20]. Starch shows a pattern similar to carbohydrates like cellulose, but with different relative intensities at 1156, 1080 and 1019 cm^{-1}, related to C–O stretching vibration [22].

Sulphates and oxalates are commonly found in many ink particles, as is observed in Fig. 2C which shows a spectrum of sample 53 *Bookhands*. Sulphates have been found mostly in inks belonging to the *Bookhands* and *Brown* groups. Particles found to contain sulphate were perfectly separated from the papyrus, forming like a superficial crust. Sulphates have a characteristic band at ~ 1100 cm^{-1}, with high intensity, assigned to S–O symmetric stretching [13, 23], and a small, sharp absorption peak at ~ 1000 cm^{-1} [13, 24], which changes position according to the cation involved in the molecule [25]. Sulphates in inks could be due to components used in its manufacturing process. Iron-gall inks are prepared using iron sulphate. Different sulphates have been found in some samples. Ink spectra of samples 6, 53 (*Bookhands* group), 134 (*Bookhands* and *Brown* groups), 31 (*Brown* group), and 172 show clear bands of sulphate. For this

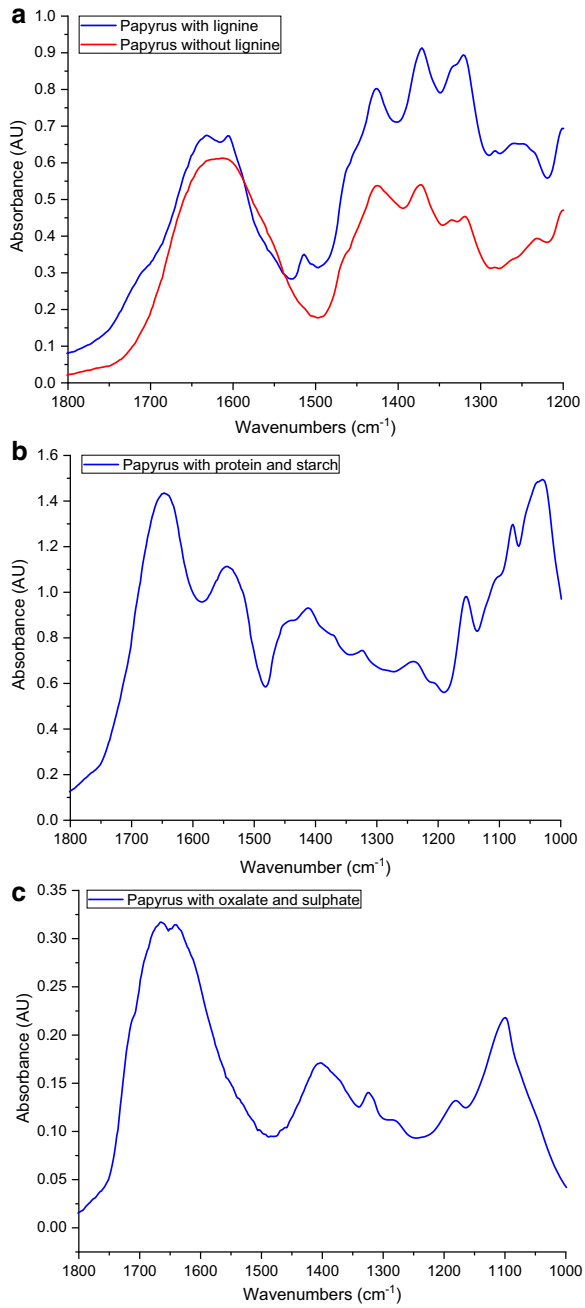

Fig. 2 FT–IR spectra of papyrus (**A**) with lignin (red) and without lignin (blue), **B** with protein and starch and **C** ink containing oxalate and sulphate

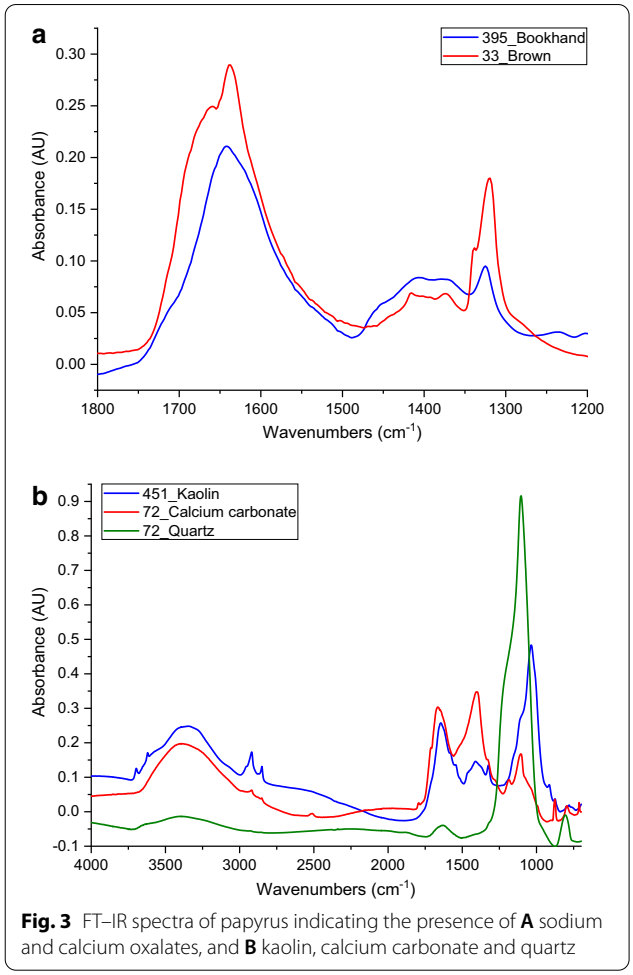

Fig. 3 FT–IR spectra of papyrus indicating the presence of **A** sodium and calcium oxalates, and **B** kaolin, calcium carbonate and quartz

last one, from the *Oxyrhynchus* group, it has been possible to determine that the spectrum has bands attributable to calcium sulphate dihydrate or gypsum ($CaSO_4{\cdot}2H_2O$). See below under conclusions.

Figure 3A shows spectra with oxalate absorptions collected from *Bookhand*s and *Brown* samples. Oxalates are present in most ink particles, but their intensity is higher in the *Bookhands* and *Brown* groups. In most spectra a typical band at ~ 1320 cm^{-1} can be detected and corresponds to the carbonyl symmetric stretching absorption of calcium oxalate [13]. Other oxalates, such as sodium or iron, show small shifts in the position or twin bands, as seen in the *Bookhands* group which shows a twin peak around 1340 cm^{-1}, which is closer to sodium oxalate bands instead of calcium [26]. Differences can be observed and compared by magnifying spectral areas. Other absorptions bands correspond to carbonyl asymmetric stretching (1638 cm^{-1}) and scissoring vibrations (800 cm^{-1}) [13, 27].

Oxalates are often the final degradation products of carbohydrates, due to hydrolysis and oxidation processes, so they could appear as a degradation product from tannins or gum Arabic. In addition, they can be formed by fungal degradation of carbonates [24, 28]. This molecule could react with cations forming different oxalate salts. It is very common to find them in

ancient samples such as paintings, sculptures, tools, etc. [29, 30]. Despite this, oxalates were not found in any clean papyrus fibre of the analysed collection.

Other chemical compounds such as quartz, carbonates and kaolin are found randomly in the collection of samples analysed. Sand coming from the surrounding environment could contribute to particle deposition of this nature. Figure 3B shows a green spectrum with a large kaolin content: this spectrum presents two weak but sharp peaks at 3696 and 3622 cm^{-1}, assigned to O–H stretching vibration [31].

The blue spectrum is collected from a sample enriched in quartz content which shows a high intensity band at around 1080 cm^{-1} and a small one at 780 cm^{-1} attributed to asymmetric Si–O-Si stretching vibration [32]. The red spectrum presents two characteristic bands of calcium carbonate centred at 1454 (stretching vibration of carbonate) and 876 cm^{-1} (O-C-O symmetric bending vibration) [33].

Apart from the above-mentioned molecules, there is another infrared region to be considered. Carbonyl groups (C = O) absorb in a region around 1700–1750 cm^{-1} [24] and can be attributed to either papyrus or ink oxidation. Previous studies in our lab concluded that iron-gall inks showed stronger absorptions around 1700 cm^{-1} compared to carbon inks [34]. Corrosive inks (iron cations and sulphuric acid) can degrade carbohydrates, which are part of cellulose or gums. As a consequence, hydroxyl groups are oxidized to carbonyl.

Figure 4 shows a comparison of two representative spectra of iron-gall and carbon inks. The principal difference between them is the stronger carbonyl band in iron-gall inks FT–IR spectra compared to carbon inks. There is also a difference in band position and shape.

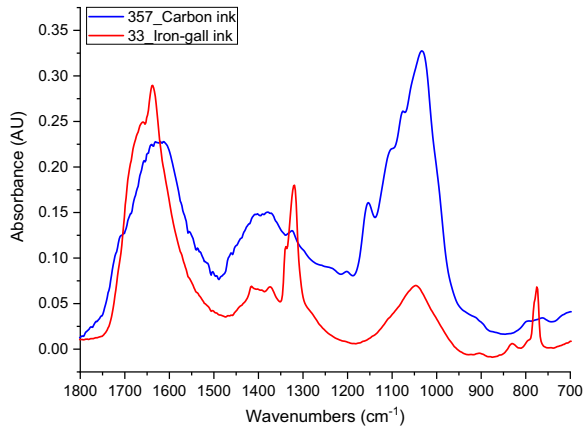

Fig. 4 Representative FT–IR spectra of iron-gall (red) and carbon inks (blue)

Degradation of papyrus support

Visually it is possible to observe the state of cellulose: sane or degraded fibres. Differences in the shape and intensity of carbonyl groups bands (1750–1700 cm^{-1}) correlate with the visual degradation [34]. It is difficult to extract pure ink particles from degraded samples, and cellulose bands are always observed in the ink spectra.

Figure 5A shows the 3D PCA graph obtained from the region 1500–1900 cm^{-1} corresponding to the papyri classified as *Bawit* and *Oxyrhynchus* groupings. The three Principal Components (PC) explain the 95.5% of the total variance, being PC1 the one which explains most of it (88.3%). The combination of these three components

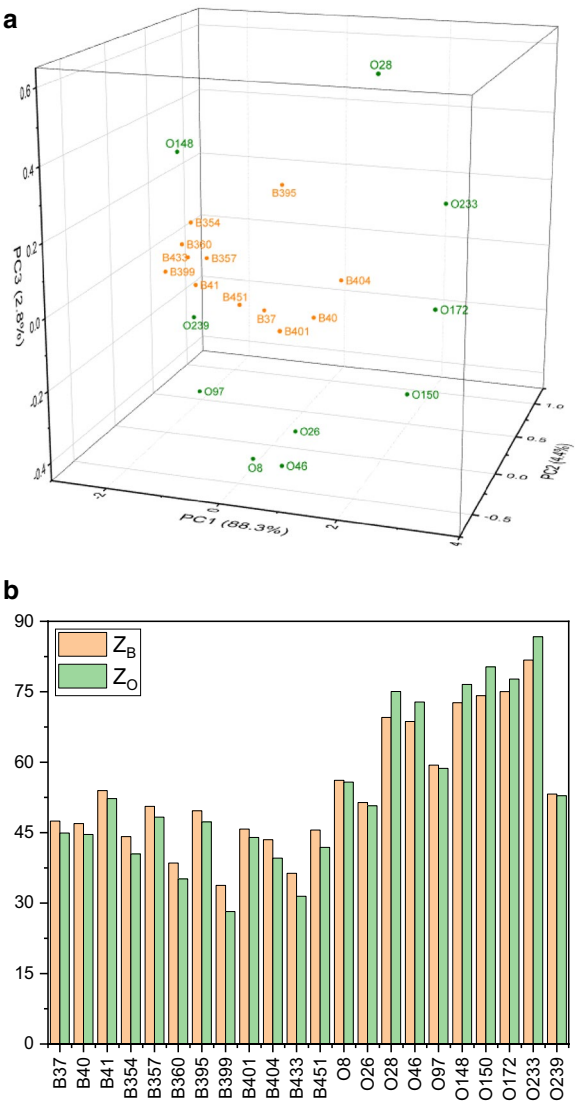

Fig. 5 **A** 3D PCA plot and **B** Z_B and Z_O parameters of LDA for *Bawit* and *Oxyrhynchus* groups

makes possible to conveniently differentiate the samples from *Bawit* and those from *Oxyrhynchus.* As it can be seen in Fig. 5A, the *Bawit* papyri largely form an internal nucleus within the figure, unlike those of *Oxyrhynchus,* which are much more dispersed, not responding in a homogeneous way to the variation of any of the three PC generated. This leads us to suggest that spectra from the *Bawit* papyri are homogeneous and with similar degradation states. *Oxyrhynchus* group shows very different cellulose degradation. This is probably explained by the much wider chronological, and hence cultural, range of the documents here studied coming from *Oxyrhynchus*, right from the Ptolemaic period (sample 172, dated to II BCE) down to the Byzantine period (sample 233, dated s. VI). Instead, *Bawit* texts come all from a very homogeneous environment, both culturally and chronologically; that is, a monastery (and not a whole city in all its diversity, as is the case of *Oxyrhynchus*) funded towards the end of IV CE and progressively declining after the Arab conquest of Egypt, the sixth and seventh centuries representing the peak of its prosperity (and therefore its written production) [5].

The LDA tool reports two classification equations based on the parameters used for the analysis. In this case, we used six parameters obtained from the results of the FT–IR spectra of *Bawit* and *Oxyrhynchus* papyri (see Additional file 1: Fig. S1). These parameters were the intensity and the full width at half maximum (W) of the two main peaks observed in the spectra and their respective ratio (I_{peak1}, I_{peak2}, I_{peak2}/I_{peak1}, W_{peak1}, W_{peak2} and W_{peak2}/W_{peak1}). The first peak was located between ~ 950 and 1150 cm^{-1} whilst the second peak was observed at ~ 1500–1750 cm^{-1}. The SPSS software used the Wilks' lambda as reference, and it reported that the more adequate components to differentiate the papyri were the I_{peak2}/I_{peak1} and the W_{peak2}. Only these two parameters were found statistically significant to parametrize our mathematical model. As LDA results, we obtained two classification functions based on the previous commented parameters, one for the *Bawit* papyri:

$$Z_B = -47.443 + 5.474 \cdot \left(\frac{I_{peak2}}{I_{peak1}}\right) + 0.988 \cdot W_{peak2}$$

and the other for the *Oxyrhynchus* papyri:

$$Z_O = -70.338 + 12.864 \cdot \left(\frac{I_{peak2}}{I_{peak1}}\right) + 1.176 \cdot W_{peak2}$$

When I_{peak2}/I_{peak1} and W_{peak2} were substituted by their numerical value for each papyrus, we obtained a Z_B and a Z_O value to be compared. When $Z_B > Z_O$, the papyri would be classified into *Bawit* group, while if $Z_O > Z_B$, the sample should be considered in *Oxyrhynchus* group. All Z_B and Z_O values were presented in Fig. 5B. At view of our results, all the papyri from the Monastery of *Bawit* were correctly classified, as they presented a $Z_B > Z_O$ pattern. However, the papyri from *Oxyrhynchus* presented a different behaviour, as only the 60% of the *Oxyrhynchus* papyri were correctly classified in its corresponding group. These findings were in agreement to those reported in PCA analysis and let us confirm that the *Bawit* papyri present statistical significative similarities that indicate a close origin. However, as it was expected based on our PCA analysis, the *Oxyrhynchus* papyri presented a heterogeneous trend, allowing us to hypothesize again that the different and more diverse origin of their inks was behind this erratic trend.

Spectroscopic description of some papyri

Next, a description of 11 carefully selected inks is shown as an example of the different groupings carried out in this work.

Sample 31 (Brown group): patristic literature (trinitarian text?)

Additional file 1: Fig. S2 shows the infrared spectrum from sample 31. This infrared spectrum indicates neat bands without cellulose. Sulphates (probably potassium sulphate, as seen in EDX spectra) and silicates, since silicon and aluminium are related, can be observed in infrared spectra. Some particles also show bands that could correspond to organic acids. Both techniques suggest the use of an iron-gall ink.

Sample 33 (Brown and Bookhands groups): Psalms 1,3–6; 2,6–9

The infrared spectrum shows neat bands without cellulose (Additional file 1: Fig. S3). Oxalate bands are probably related to sodium oxalate. A calcium phosphate spectrum from an infrared spectroscopy database is similar to those appearing in the spectra, as prove EDX mappings. In antiquity, some black inks were made using burned bones, and one of their components is calcium phosphate [35]. However, since this is the only papyrus in the study presenting calcium phosphate nothing can be concluded as to the use of animal bones in the composition of the inks in the samples examined.

A large amount of iron found with EDX spectra (around 3.7%) indicates an iron-gall ink, as seen in Fig. 6. In addition, other metals, especially copper and magnesium, have also been found.

Sample 53 (Bookhands group): Gospel of John

Infrared spectra are clean and without cellulose, which leads to think that it is an iron-gall ink (Additional file 1: Fig. S4). Firstly, calcium oxalate and potassium

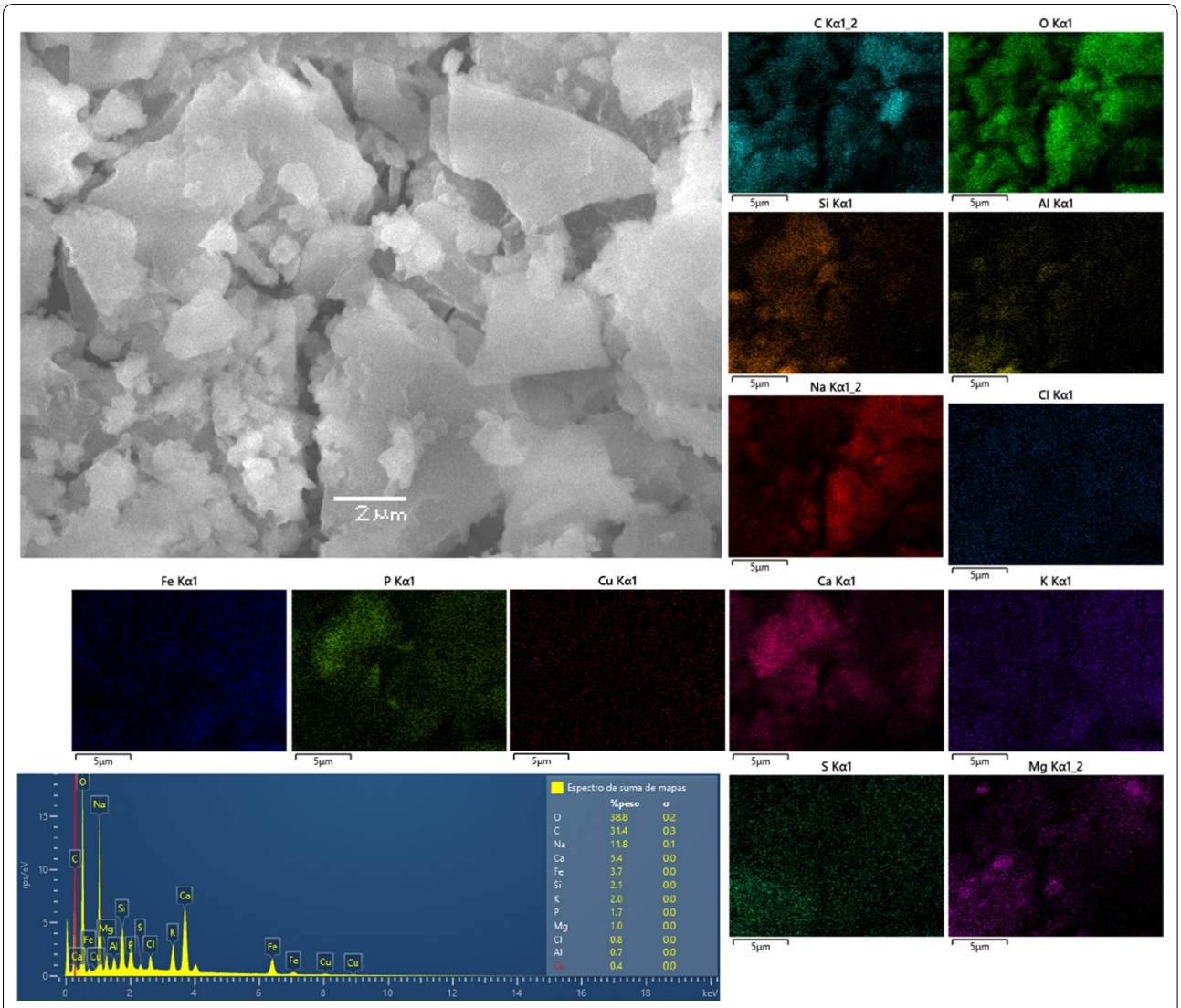

Fig. 6 SEM micrograph, EDX elements mappings and EDX spectra of sample 33

sulphate are identified by IR spectroscopy and EDX mappings confirm the correlation between potassium and sulphur and calcium-enriched particles. Secondly, EDX mappings illustrate homogeneous occurrence of iron and a similar iron amount to that present in sample 33 has been detected. However, negligible copper was detected. This sample seems to belong to an iron-gall ink.

Sample 90 (Brown and Bookhands groups): Gospel of Matthew 26, 56–57, 62–63

Figure S5 shows infrared spectrum from sample 53. Infrared spectra confirm no cellulose bands but, unlike the previous sample, neat strong bands of sodium oxalate appear. There are also absorption signals in the wavenumber region where sulphates, phosphates or silicates appear.

SEM/EDX and infrared spectroscopy results were found to agree. Important concentration of iron is common in iron-gall inks.

Sample 133 (Brown group): Christian literature

It was difficult to avoid cellulose fibres when we tried to remove ink particles, nonetheless spectra were clean and with very high calcium oxalate bands, as brought out in Additional file 1: Fig. S6.

The EDX mappings show that silicon and aluminium appear together (aluminosilicates), which could be due

also to external contamination. In addition to the calcium oxalate, in some particles, calcium and phosphorus are clearly related. Sodium chloride is also observed, which is common to find in Egyptian materials [36]. Copper and calcium phosphate particles were also detected.

The large absorption of calcium oxalate and the presence of iron and copper suggest an iron-gall ink.

Sample 233 (Oxyrhynchus, S.VI and Chancery groups) contract: loan of land

Clean particles of ink could not be obtained as the ink was deeply mixed with cellulose.

The only molecule identified in the infrared spectrum was calcium carbonate (Additional file 1: Fig. S7). Sodium chloride was detected by EDX, although the rest of the elements are homogeneously mixed in the analyzed ink particles.

The absence of iron and the background absorption of the infrared spectrum is normally due to carbon content. The EDX spectra confirm the non-presence of both iron and copper, which tend to also appear in metallic inks. These results make us conclude that it is carbon ink.

Sample 241 (S.VI group) contract: beginning of an agreement

As it happened with sample 233, cellulose and ink are closely mixed.

The infrared spectra of this sample show a high background absorption and the only molecule identified was kaolin (Additional file 1: Fig. S8), a common silicate found in environmental samples. EDX mappings show that aluminium appears together with silicon (Fig. 7).

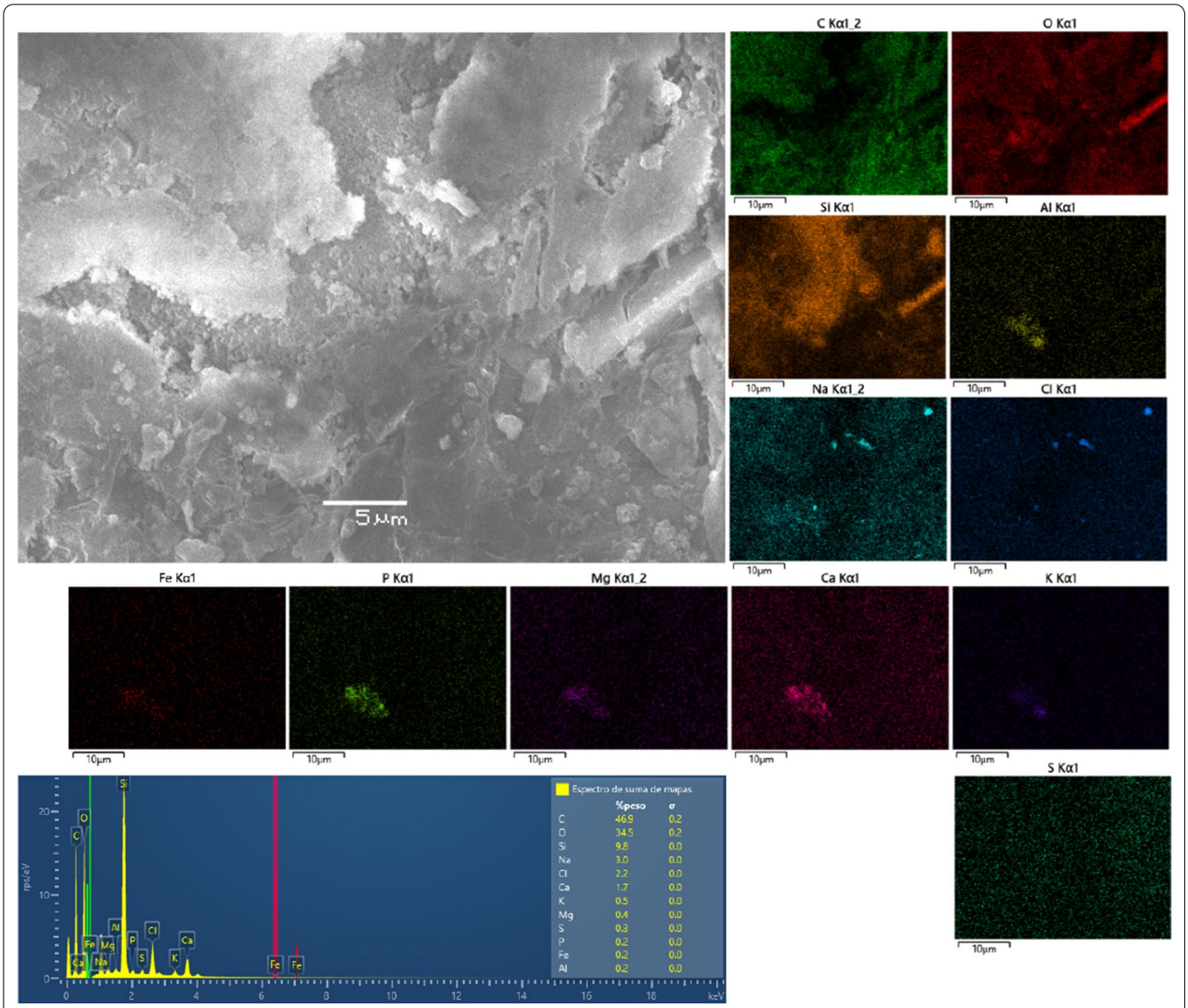

Fig. 7 SEM micrograph, EDX elements mappings and EDX spectra of sample 33

In this figure calcium, potassium and phosphorous are related, and sodium chloride also appears, as seen in other samples.

Few amounts of iron, but not clearly assigned, are also detected by SEM/EDX. EDX mappings indicate its presence could be due to aluminosilicates contamination. Results indicate that this ink is carbon based.

Sample 243 (S.VI group) contract: loan of money

The composition of this sample is the same as samples 233 and 241. Additional file 1: Fig. S9 brought out that only calcium oxalate was detected in the infrared spectrum which exhibited a high background. High absorption is observed in some regions of the spectrum, and it is not possible to identify the broad bands. Silicon, aluminium, potassium, and iron are related in EDX mappings. In addition, it seems some calcium sulphate particles have been formed and magnesium and phosphorous are homogenously distributed. Sodium chloride also appears.

The infrared results coupled with high carbon content and the close relationship among iron, silicon and aluminium suggest a carbon ink.

Sample 395 (Bawit and Brown groups) letter

Ink and cellulose are mixed also in this sample. The infrared spectra showed broad bands and we could only find calcium oxalate (Additional file 1: Fig. S10). In this case, background baseline is not high compared to other carbon inks. EDX mappings show a possible relationship among calcium, magnesium, and sulphur. Sodium, potassium, and chloride seem to be related, although copper could be attached to chloride or sulphur.

The EDX results indicate little iron, related to silicon and aluminium, and, especially, copper wealth, which leads us to doubt about the ink's nature.

Sample 427 (Bookhands group)

Like other *Bookhand* samples, ink was easily removed from the papyrus surface. Spectra are clean and strong infrared absorptions of sodium oxalate are present (Additional file 1: Fig. S11). Calcium sulphate and sodium chloride are identified by EDX mappings.

EDX results display particles with a high iron content. Although oxygen is related, it is difficult to be sure which salt is formed. The results obtained suggest this is an iron-gall ink.

Sample 712 (S.VI group) petition to defensor

Iron is present in EDX graphics, but associated to silicon and aluminium, so it could be due to aluminosilicates. This finding is confirmed by the infrared spectra (Additional file 1: Fig. S12), which have broad bands, high background, and we could not identify any molecule apart from cellulose, which is impossible to remove from particle inks. In this case, results lead us to doubt about the ink's nature.

Conclusions

According to our results, we can conclude that there are some correlations considering some groups and their ink's nature.

Bawit group

Analysis results of clean papyrus fibres indicate that they are very homogeneous and most have a high lignin content. This suggests lower manufacturing standards, where techniques for the preparation of papyrus strips might not have been so careful as to prevent the outer more lignin rich layers of the stem added into the papyrus *charta* [37]. A decrease in the quality of the *charta* can be explained in relation to the scarcity of papyrus as a writing material in Byzantine monastic environments [5] when compared with other periods and social milieux such as, for instance, the urban written culture from the Roman period.

A deep mixture of ink and papyrus in the strokes of letters was noted, making it impossible to remove a single clean particle of ink. This fact, associated as has been seen to documentary texts, may have to do with the fact that all papyri present in the *Bawit* group are, in fact, documents: although it is suspected that the monastery would have also produced a considerable quantity of literary texts used for the liturgy and it might even have had a library, it has not been possible to ascribe with absolute certainty any such text to the monastery [38], which has in itself precluded the entrance of literary texts in the sampling for the group. It is not surprising therefore that *Bawit* samples could be correlated with carbon-based inks, for as will be seen below, the use of iron-gall inks seems to be associated with the production of literary texts.

All samples exhibited the presence of calcium oxalate in the IR spectra and had no strong absorption in the carbonyl region.

Bookhand group

Spectra of clean papyrus with no strokes ink are relatively homogeneous.

Contrarily to the *Bawit* group, ink particles were easily removed from the cellulose support and look like a surface layer covering the papyrus. As a consequence, cellulose bands are not present in the spectra, which helped to identify the presence of other molecules. Strong absorptions in the carbonyl region were detected. Thus, also consistently in opposition to the *Bawit* group, *Bookhand*s samples could be correlated with iron-gall inks. These

results confirm previous findings that iron-gall inks appear to be associated with the production of literary books during the Byzantine period and do not seem to have been extended to the writing of documents [39]. However, two documentary papyri, namely 395, a letter, and 712, a petition directed to an urban magistrate, present some doubts as to their ink composition; it is worth noting in this respect that both letters and petitions are closest to be regarded as a literary genres among all documentary types of text [40–42].

The spectra of all ink samples in this group contain oxalate bands, but in comparison with calcium oxalates, found in other groups of this research and in other studies made in the past in our laboratory [26], oxalates in the *Bookhands* group have a doublet indicative of sodium oxalate instead of calcium oxalate.

Brown group

The only common characteristic of samples belonging to the *Brown* group is the fact that ink particles can be removed easily from the papyrus support, as it happened with the *Bookhand* group. Calcium oxalate is present in many of them and strong absorptions in the carbonyl region were detected.

Therefore, the *Brown* samples also seem to be in agreement with iron-gall inks. Interestingly enough, the only two samples from this group where it is not possible to ascertain the iron-gall nature of the ink are the only two of non-literary character, both from Bawit: samples 395, a letter (see above), and 399, a receipt for which our results do not point to iron-gall ink.

II CE group

It is worth noting that, unlike the literary texts in the *Bookhands* group, all dated to the Byzantine period, literary texts from the Roman period (samples 127, 147 and 164), also written in relatively formal hands, do not present iron-gall inks, but carbonic ones.

Two questions related to the presence of sulphates and oxalates that are of interest for the study of writing practices in Greco-Roman Egypt:

It was pointed out that sample 172 contained calcium, not iron sulphate, which was typical in iron-gall inks. In fact, sample 172, an official document dated to 186 BCE containing a decree of amnesty, would be too early to present such an ink. Instead, the identification of the calcium sulphate dihydrate (gypsum) points to the practice, relatively common in Ptolemaic Egypt, of processing discarded papyrus material to make a sort of *papier-mâché* that would then be used to make mummy cases [43]. Once the discarded material had been given the form of the piece desired, it was covered by a coating of gesso plaster on which painting was applied.

The hypothesis of the presence of tannins raises the question of the origin of the acidic component of iron-gall inks in Egypt. This region is not the natural habitat of the oak tree, typically used as the source of tannic acid to produce iron-gall inks in Europe. We know, however, that wine industry flourished in Greco-Roman Egypt, and, although we do not have any written evidence, we may assume that the extension of cultivated land dedicated to vineyards, an estimate of c. 1250 square kilometres for the Roman period [44], would have allowed for the extraction of tannins eventually used for the fabrication of iron-gall inks. In this respect, it is very interesting to note that ancient sources bear witness to the use of tamarisk galls, rich in tannins, for pharmacological purposes [45]. Since it is also known that this plant was cultivated and exploited in Greco-Roman Egypt [45], the possibility exists that its galls, or its leaves, may have also been used for the production of iron-gall inks.

Finally, it has been observed that iron-gall inks are removed from the cellulosic support in a much easier way than those based on carbon, which is consistent with current knowledge. The chemical processes that favour the greatest degradation of papyri written with iron-gall ink are acid hydrolysis and metal cations oxidation [46]. Hydrolysis is due to the acidic environment generated by bisulphate, released when iron complexes with gallates, leading to a breakdown of the cellulose polymer chains [47]. On the other hand, oxidation occurs mediated by free metal cations, especially iron, but due to the various origins of the iron sulphates with which these inks were made, they could also be due to other cations whose salts contaminated iron sulphate, like copper, manganese, etc. [48].

Supplementary Information

The online version contains supplementary material available at https://doi.org/10.1186/s40494-022-00742-1.

Additional file 1: Figure S1. (A) FT–IR spectra of *Bawit* and (B) *Oxyrhynchus* groups. **Figure S2.** FT–IR spectra of sample 31 (*Brown* group). **Figure S3.** FT–IR spectra of sample 33 (and **Bookhands** groups). **Figure S4.** FT–IR spectra of sample 53 (*Bookhands* group). **Figure S5.** FT–IR spectra of sample 90 (*Brown* and *Bookhands* groups). **Figure S6.** FT–IR spectra of sample 133 (*Brown* group). **Figure S7.** FT–IR spectra of sample 233 (*Oxyrhynchus, S. VI* and *Chancery* groups). **Figure S8.** FT–IR spectrum of sample 241 (*S. VI* group). **Figure S9.** FT–IR spectrum of sample 243 (*S. VI* group). **Figure S10.** FT–IR spectrum of sample 395 (**Bawit** and *Brown* groups). **Figure S11.** FT–IR spectrum of sample 427 (*Bookhands* group). **Figure S12.** FT–IR spectrum of sample 712 (*S. VI* group).

Additional file 2. Table S1: list of sampled papyri together with metadata and analysis results.

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Author contributions
AN defined the criteria to be used in the selection of the papyri to be sampled and formed the corresponding groups; NFF and FJP performed the infrared spectroscopy and SEM/EDX analysis of the samples and interpreted the corresponding results, the statistical treatment was performed by RL, and the three discussed them with AN, who is mainly responsible for the section "Introduction" and wrote the "Papyri description" section, whereas NFF, FJP and RL wrote the sections "Materials and methods", "Instrumentation" and "Sample handling" sections. The sections "Results and discussion" and "Conclusions" sections are the product of the cooperation among the four authors. All the authors read and approved the final manuscript.

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Availability of data and materials
The datasets used and/or analysed during the current study are available from the corresponding author on reasonable request.

Declarations

Competing interests
The authors declare that they have no competing interests.

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