Sacrifice of Isaac by Antonio di Donnino del Mazziere: Technical Analysis and Conservation Treatment
Introduction

The Sacrifice of Isaac (acc. 1977.177) entered the collection of Harvard Art Museums in 1977. The work (c.1525) is attributed to Antonio di Donnino del Mazziere (1497–1547), an artist about whom relatively little is known, with only a handful of paintings and drawings attributed to him. He is one of a group of painters that art historians have dubbed the “Florentine eccentrics”, who were followers of Fra Bartolomeo and Andrea del Sarto. The provenance for the work is incomplete.

The painting (figure 1) is in the style and format of a spalliera, a genre particularly related to the Italian renaissance. Intended for the domestic interiors of patrician homes, spalliere are painted panels wall-mounted at shoulder or eye level, the name being derived from the Italian spalla, meaning shoulder. Usually spalliere were created in cycles or groups, rather than individual stand-alone panels. Their purpose was both decorative and narrative, often including more than one episode in a single panel (episodic narration). (Barricault pp.1-6).

The work shows two episodes from the biblical narrative of Abraham and Isaac. At the left, an angel appears to a kneeling Abraham, either telling him he will have a son, Isaac (Genesis chapter 18), or instructing him to sacrifice Isaac (Genesis chapter 22). A tent, figures and sheep at the left background represent the nomadic herding lifestyle of the clan. In the top right a later episode is shown, where an angel stops Abraham at the point of carrying out the instruction and sacrificing the kneeling Isaac. To the left of Abraham a grazing sheep pre-empts the end of the drama, that Abraham sacrifices the sheep in place of his son as instructed by the angel. At the lower right corner the two servants and donkey that accompany Abraham and Isaac to the mountain (Genesis chapter 22 verse 3) balance the composition, while at the centre a vista shows a typical skyline for an Italian Renaissance town on a hill. Two small figures and a donkey at the centre of the work assist to create a sense of depth, as well as implying the journey Abraham and Isaac take to the place of sacrifice.

The main compositional elements are closely related to Lorenzo Ghiberti’s depiction of the same narrative in The Gates of Paradise for the Florence Baptistery, c. 1435 (figure 2). As a Florentine, Mazziere would surely have been familiar with this work. In Ghiberti’s version the eye is drawn to the strong diagonal movement from bottom left to top right corner. In this version the seated figures at the bottom right corner balance the overall composition without drawing attention to their prominent position. In the Mazziere version the diagonal
connection is lost by the greater space (including a void) between the two episodes. This results in the seated figures appearing to take a prominent role, although they do not take an active part in the narrative.

The *Sacrifice of Isaac* is strikingly similar in terms of its format, composition, and subject matter to a painting in the Galleria Borghese in Rome, the *Life of Joseph: Jacob Receiving the News of the Presumed Death of Joseph* also by Mazziere (figure 3). The *Life of Joseph* has been thought to relate to the significant *spalliera* group executed for the Borgherini family in 1515, comprising fourteen panels illustrating scenes from the life of Saint Joseph, but it has not yet been convincingly integrated into the cycle. It seems more likely that both the *Sacrifice of Isaac* and the *Life of Joseph* were created as pendants, as suggested by their close dimensions (the Borghese panel is 78 x 183 cm, the Harvard panel is 75 x 143 cm), comparable design elements (see the line of hills at the left edge of *Isaac* and the right side of *Joseph*, and the seated/kneeling figures at the bottom right of both works) and unifying genealogical theme (both are father/son stories from Genesis).

The commission for both paintings and their intended function remain unknown. In order to determine whether it and the *Life of Joseph* were indeed created for a common domestic setting, a combination of archival research on both works and technical examination of the *Life of Joseph* to compare with that of the *Sacrifice of Isaac* is required.

Figure 1: Antonio di Donnino del Mazziere. *Sacrifice of Isaac*. Harvard Art Museums, Cambridge MA.
Figure 2: Ghiberti, Lorenzo. *Sacrifice of Isaac* panel from *The Gates of Paradise* c.1435. Florence Baptistry (Barricault p. 63)

Figure 3: Antonio di Donnino del Mazziere. *Life of Joseph: Jacob Receiving the News of the Presumed Death of Joseph*. Galleria Borghese, Rome. (Moreno and Stefani p. 283)
Analysis and Methodology

The technical analysis of the *Sacrifice of Isaac* was driven by three aims. The first was to investigate the possibility of a connection between it and the *Life of Joseph*. A detailed description of Mazziere’s materials and technique on the *Sacrifice of Isaac* would lay the groundwork for a technical comparison to be made with the *Life of Joseph*, to see how closely they correspond on a material level. Technical comparisons with other works by Mazziere would also assist in positioning the *Sacrifice of Isaac* within the artist’s oeuvre.

The second reason was to investigate some highly unusual material features on the *Sacrifice of Isaac*. During initial treatment stages the anomaly of a layer of metal leaf underlying the paint layers was found throughout the work. Further investigation was desired in order to find out the extent of the metal leaf and the reason for its extravagant use, to see if it indicated any changes to the work, and investigate the possibility of other equally surprising features in materials or technique.

The third reason was to inform the conservation treatment of the work. Greater knowledge of materials and techniques comprising an artwork leads to more informed decisions about its treatment. In this case some treatment stages were not attempted until analysis had been conducted, and the results of these analyses impacted the treatment plan.

The work was examined with a variety of analytical tools, including x-radiographing, microscopy techniques, the mounting of paint samples to examine in cross-section, micro-chemical stains, Fourier-transform infrared spectrometry, gas-chromatography-mass spectrometry, x-ray fluorescence spectrometry, and infrared reflectography. Technical specifications for each of these analyses are given in Appendix 1.
Results summary
The technical analysis enabled the layer sequence comprising the artwork’s materials and technique to be determined, shown in figure 4. Each layer will be discussed in detail, beginning at the support.

Figure 4: The layer structure of the Sacrifice of Isaac by Mazziere

<table>
<thead>
<tr>
<th>Restoration Varnish and Overpaint</th>
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<td>Lead-white Preparatory</td>
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<td>Vermeil (Shellac)</td>
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<td>Silver Leaf</td>
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<td>Red Bole</td>
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<td>Gesso Ground</td>
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<td>Wooden Panel (Poplar)</td>
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Support
The support is a wood panel, measuring 75.1 x 142.9 x 3.2 cm (H x W x D). The grain runs horizontally. X-radiographs of the panel show that it is constructed of two planks joined at the horizontal axis (figure 5). The panel appears to be held together solely by adhesive, with no joining devices such as dowels, nails or splines being visible along the join in the x-radiographs.

The wood was identified as a variety of Poplar (*Populus* sp) by microscopic examination of slivers of wood cut in three directions to the grain; cross-section, transverse and radial, as described by Hoadley (p. 24-25). These were compared to known samples. The cross-section revealed an even distribution of numerous uniform pores, including some multiple pores. The
transverse section showed only uniseriate ray cells, along with large, rounded intervessel pits. These features identify the wood as Poplar (Hoadley p.37), and were confirmed by comparison with known samples for Black Poplar and Grey Poplar (figure 6).

There are three vertical grooves averaging 1.0 cm deep, evenly spaced across the panel verso (figure 7). They have a longitudinal taper, with the narrow end at the top for the left and centre groove and at the bottom for the right groove, and are cut with a trapezoidal cross-section (figure 8).

The character of these grooves are entirely consistent Uzielli’s observation that “In the late fifteenth and early sixteenth centuries, dovetailed crossbeams (trapezoidal in cross section), inserted in tapered…parallel grooves mortised into the thickness of the panel, were often used” (p.122). In addition “grooves are mortised across the grain into the planking, as deep as approximately one-third of its thickness…also widely known to have a longitudinal taper…adjacent crossbeams were placed with the larger ends orientated toward opposite edges of the support” (ibid, p.126). The tapering allowed the tightening of the dovetail join simply by moving the crossbeam further along the axis (ibid).

The consistency of the grooves with Uzielli’s description indicates that their purpose was to contain sliding crossbeams inserted to strengthen the join and combat warping. The wood is lighter at the cut faces of all the grooves than the rest of the panel’s verso, suggesting that the crossbeams remained in place for some time, protecting the inner wood here from age-darkening. An alternate reason could be that all the grooves were cut at a later stage than the panel’s original construction, however given that such grooves are so consistent with the manufacturing techniques of the time, and there being no other means of supporting the adhesive join, this seems unlikely.
Figure 5: X-radiograph of the bottom right corner, showing horizontal join in the panel.

Figure 6: Wood identification
**Figure 7: Verso of panel showing vertical grooves**

**Figure 8: Trapezoidal cross-section (dovetail) of grooves in panel verso.**

**Gesso Ground**

A thick white gesso layer is the first visible preparatory layer applied to the panel (figure 9). It is visible at the bottom of many losses across the panel. There is a single drip at the center of the right edge that appears to be gesso, which would indicate it was applied while the panel was laid flat.

Initial media analysis was conducted with micro-chemical stains applied to paint samples mounted in cross-sections. Positive results were given for both protein and lipids. This
suggests a medium of the typical animal glue (containing protein) with the less typical addition of an oil binder (containing lipids).

XRF was conducted on an area of exposed gesso, giving strong signals for calcium and sulfur. FTIR spectra indicated the presence of gypsum, confirming the XRF result for calcium sulfate, and confirmed the likelihood of an oil binder. From this it can be deduced that the ground is a typical early Italian gesso comprising animal glue and calcium sulfate ground, with the less typical addition of oil.

Figure 9: Paint sample shown in cross-section (sampled from area of green background)

**Bole layer**

There is an orange-red layer applied directly over the white gesso (figure 9). It is a continuous layer throughout the work, as indicated by cross-sections and where it is visible in paint losses. Micro-chemical stains indicated that the layer contains protein but no lipids. XRF gave signals for mostly iron, with traces of aluminium and silicon.

The presence of protein suggest at an animal glue medium, while the XRF results are typical for a bole. This layer is therefore consistent with the bole layers used as a preparatory layer for gilding.
Silver leaf

Initially metal leaf was observed as a nimbus surrounding the central angel. Next it was observed where there were paint losses throughout the work, lying between the bole and paint layers. It was found in all paint samples taken from the work and mounted in cross-section (figure 9). Finally its presence as being across the entire surface in a continuous layer on top of the bole was confirmed during XRF analysis of all major areas.

In cross-sections the colour of the leaf could be seen to be silver when observed in bright field (figure 10). The leaf was confirmed as being silver metal with XRF analysis. Grid-like dark marks visible in infrared might reveal gaps between the leaves, or where they overlap (figure 11). If this is the case, then the size of the leaves would be approximately 6 – 6.5 cm square.

Silver leaf was used in medieval times for its own colour, such as depicting armour in battle scenes (Thompson p. 190), or to represent silver brocade (Cennini pp. 88-89). Another use was to apply transparent paints over a layer of silver leaf to give an effect of heightened reflectance to certain areas (Nadolny p.153, Van de Graaf pp.43-45).

Silver leaf was also a popular substitute for gold leaf, as it was a much cheaper metal. Cennini (p.88) makes the recommendation “And, indeed, I advise you, if you want to teach boys or children how to do gilding, have them lay silver, so as to get some experience with it; for it is less expensive.” To imitate gold-leaf the silver would be covered with a transparent yellow coating.

It seems likely that the silver in the exposed nimbus surrounding the central angel was originally intended to look gold, due to the presence of yellow shellac residues in this area. However, the question remains of why was silver applied to the entire panel when only a relatively small area was to remain exposed? If the artist desired to use the reflective qualities of the silver it would be expected that transparent paints would be used throughout the work, but this is not the case. Instead an opaque lead-white layer has been applied to the silver and shellac throughout the rest of the work, seemingly to block out the undesired silver. This would imply that the panel was gilded with silver for some other purpose than this painting (maybe as a gilding exercise), and the artist decided to simply cover it up rather than remove it for this work.
Figure 10: Paint sample shown in cross-section (sampled from area of exposed gilding surrounding central angel)

![Sample 15: visible light, dark-field, 200x]

![Sample 15: visible light, bright-field, 200x]

![Sample 15: ultraviolet light, 200x]

Figure 11: Underdrawing and possible silver-leaf borders revealed in infra-red (composite image of bottom left corner comprising infrared digital reflectograms).
Shellac

In all cross-sections taken from painted areas there is a yellow/orange layer immediately on top of the silver leaf. This layer has no visible pigment particles, and fluoresces yellow/orange in ultraviolet light (figures 9 and 10). Sampling showed that although the material appears to form a continuous layer elsewhere, only residues of this layer remain on top of the gilding comprising the main angel’s nimbus.

Considering the FTIR analysis of this layer (figure 12) in association with its distinctive orange fluorescence in ultraviolet light an identification of shellac was determined. Shellac is a natural resin of insect origin. The resin is secreted by the lac insect Laccifer lacca (native to India) onto twigs, harvested as ‘sticklac’, which is the raw material from which shellac or lac dye can be extracted (CAMEO).

This instance of a shellac layer would appear to be an early example of its use as a coating. According to Masschelein-Kleiner (p.90), lac dye was known in Europe as early as the 10th century, while Williams (p.89) asserts there is no clear indication for the first use of shellac, or if the resin was used as a coating at the same time as being used as a dye. Yet Williams states that shellac was not imported to Europe until the 17th century (ibid), while Masschelein-Kleiner (p.90) states that gum lac was not used in Europe until the end of the 16th century. (Here it is assumed that Masschelein-Kleiner’s reference to ‘gum lac’ is the purified shellac resin that Williams refers to.)

From this it seems that shellac was not widely available in Europe at the time of this work’s execution (c.1525), however it is clear that the unrefined sticklac was. According to Kirby and White (p.68) a Florentine merchant, Pegelotti, writing in the 14th century fully describes the “tubular form, colour and grainy appearance” of sticklac. In Kirby, Spring and Higgitt’s study of medieval recipes for lac lake pigments, they found that for most recipes it was the entire raw material (the sticklac) that was ground and extracted with alkali (p.75). Kirby and White draw the following conclusion about lac lake; “it appears that lac lake was the ‘standard’ lake used for easel painting in the fifteenth century in Florence, and probably other parts of Italy as well” (p.68). This indicates that sticklac was well-known and used in Europe prior to the execution of the Mazziere painting.
Imitating gold leaf with silver and a colouring layer was a common medieval practice, as gold-leaf could be 3 to 7 times more expensive than silver-leaf (Nadolny p.165). The colouring layer, referred to here as ‘vermeil’ after Nadolny (p.146) was a transparent yellow to orange-red varnish, its sole function being to make silver or tin surfaces appear gold. Recipes for vermeil vary widely in their constituents, and include references to linseed oil, gums, resins, and a range of yellow pigments and dyes, including red lake (Nadolny pp.149, 189-190). The terms for gum, resin, and varnish appear often, but are either no more specific than the English equivalents used here, or it is not clear which specific material they might refer to. Sticklac may have been known as either a gum or a resin in medieval times, and may have been trialled as a major constituent in a vermeil, as could the lac dye. The FTIR spectral similarities to shellac and to Indian lake (lac) could be expected for a coating largely comprised of sticklac.

Another reason for the shellac application could have been to protect the silver from tarnishing, as it was well-known to do so. Cennini states “…above all you are to work with as little silver as you can, because it does not last; and it turns black, both on wall and on wood…” (p. 60).
Figure 12: FTIR spectra for yellow resin layer (shellac) and close matches

White layer
A white layer of paint is present on top of the shellac layer throughout the work, except where the silver leaf and shellac were left exposed around the central angel. The white layer was visible in some losses to the paint layers, and was observed in cross-sectioned paint samples taken throughout the work (figure 9). It is also apparent as a white ridge at the edge of the exposed silver leaf surrounding the angel, clearly visible in raking light (figure 13).

The medium of this layer was indicated as oil with microchemical tests, while XRF analysis indicated a strong lead component. These results are consistent with this being a layer of lead-white oil paint.
The presence of this layer seems to negate the possibility of the artist desiring to use the silver layer for some optical effect. If there was any possibility of the silver leaf reflecting light through the paint layers, this opaque white layer would block it out.

The final design of the work closely follows detailed under-drawing (visible in infrared examination). The under-drawing therefore must lie on top of this layer, or else it would not be visible to the artist as they were painting the final design. It seems likely therefore that this white layer was put down in order to provide an appropriate surface for drawing on, and to block out the silver-leaf and shellac layers where they were not required.

Figure 13: detail of central angel in raking light, showing ridge formed by white layer at edge of exposed gilding.

Underdrawing
Extensive under-drawing was revealed in infrared examination, indicating that it is executed in carbon-based media such as charcoal (figure 14). It has been executed carefully with a fine-tipped drawing tool, including cross-hatching to indicate shading in draperies, and has been closely followed in the subsequent painted elements (figure 11).

Minor changes from the under-drawing to the final work include a pair of figures in the background that are not included in the final work (figure 15), more prominence to the wall behind the kneeling Abraham in the under-drawing than the painted version, and alteration in the line of hills in the far background.
The under-drawing in this work provides a good example of the limitations of different infrared imaging techniques. The work was initially imaged in infrared digital photography, however the greater sensitivity achieved in infrared digital reflectograms revealed more detail, and so several main sections of the work were imaged in this way. Details such as the figures shown in figure 15 do not appear in the infrared digital photographic image.

Although infrared digital photography can capture such a painting in a few images, the sensitivity is not great enough to fully capture the underdrawing media in this instance. Infrared digital reflectograms, although relying on a time-consuming process of capturing and mosaicing small images together in this case, have been able to capture much more information from the under-drawing layer.

Figure 14: underdrawing revealed in infrared (infrared digital photograph)

Figure 15: detail at centre, right, figures not appearing in final composition (infrared digital reflectogram)
Paint Layers

Samples of paint taken across the work showed in cross-section that there are usually one to two layers of paint in the areas of design (figure 9). The media was initially indicated as oil with micro-chemical stains, then confirmed with GC-MS. The palmitate/stearate ratio of 1.81 indicates the medium is most likely linseed oil. The use of linseed oil is consistent with early 16th century practice, during which time artists were experimenting with linseed oil as well as continuing the use of egg tempera.

The pigments comprising the artist’s palette were identified using XRF to conduct elemental analysis on different-coloured sites across the work. Table 1 presents some of the XRF results and the pigments they indicate:

<table>
<thead>
<tr>
<th>COLOUR:</th>
<th>BLUES</th>
<th>GREENS</th>
<th>REDS</th>
<th>YELLOW</th>
<th>WHITE</th>
</tr>
</thead>
<tbody>
<tr>
<td>XRF results:</td>
<td>copper</td>
<td>copper</td>
<td>mercury, lead, no result</td>
<td>lead, tin</td>
<td>lead, titanium</td>
</tr>
<tr>
<td>Likely pigments:</td>
<td>azurite</td>
<td>copper, resinate,</td>
<td>vermilion, red lead, organic red</td>
<td>lead-tin yellow</td>
<td>lead white, titanium, overpaint</td>
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Table 1 shows that the palette is consistent with an early 16th century palette. Where no strong signal was received in areas of red, the absence is likely to indicate the use of an organic pigment, such as red lake (lac). The lead signals noted in areas of red and yellow could in part come from the lead-white paint layer underlying the upper layers, however the strength of the signals in these areas indicate the likely presence of other contributing pigments, such as red lead and lead tin yellow. One white stroke with the appearance of overpaint gave a signal for titanium, confirming that it was indeed restoration paint.

Restoration Varnish

The uppermost layer on the painting was a clear continuous coating, coloured an intense dark-yellow. In cross-section it appeared as an unpigmented layer that fluoresces yellow-white in
ultraviolet light (figure 10). It was not an original layer, as discoloured overpaint could be clearly seen to lie beneath it (figure 16).

It was found to be at least partially soluble in polar solvents, including ethanol, isopropanol and acetone, although a swab needed to be rolled many times before removal was effected. It was found that repeated applications of acetone sufficiently degraded the layer to be removable by the mechanical action of a soft crepe eraser, rather than fully dissolving into the solvent (figure 17).

The varnish was examined with FTIR, giving a match for alkyd paint. This result was confirmed with Py-GC-MS, giving the single phthalate peak typical of an alkyd coating.

Coatings based on alkyd resins were not widely available until the 1950’s. Like oils, alkyls eventually harden by polymerization and cross-linking, forming an insoluble coating that can be very difficult to remove, as instanced by Rajer and Kartheiser, Appelbaum, and the author in previous treatments. It is likely that such coatings will become more difficult to remove as they age due to continued cross-linking.

The varnish was recommended for removal in order to retrieve the work’s original colour scheme. It was suggested that the varnish be left untouched on the silver leaf surrounding the angel, as the presence of shellac residues here indicated that this area was originally coated to appear gold, and the restoration varnish was now fulfilling that role. However, when the varnish was identified as alkyd resin it was decided to remove the coating in its entirety while this could be done with minimal risk to the underlying layers, knowing that it is likely to become more difficult to remove over time.

In this instance the response of the coating to solvent action can be understood as the acetone swelling the layer rather than dissolving it, disrupting both its internal cohesion and adhesion to the substrate, eventually enabling its removal by mechanical means.
Figure 16: detail of sky before varnish removal, with discoloured overpaint visible beneath the varnish

![Figure 16](image1)

Figure 17: varnish removal process (swabbing with acetone until the coating deteriorated, enabling its removal with a soft crepe eraser)

![Figure 17](image2)
Conclusion

A full overview of the materials and techniques Mazziere has used in executing the Sacrifice of Isaac has been successfully completed. Apart from their unusual usage, the materials employed are consistent with Italian panel painting of the early 16th century. However it is still unclear as to why the unusual technique of covering the entire panel with bole, silver leaf and shellac prior to applying the paint layers was employed. A discovery of the same technique on another work by Mazziere may clarify this mystery.

The level of detail achieved through analysis provides a good basis for a technical comparison between this and other works by Mazziere, particularly the Life of Joseph in the Galleria Borghese. It is hoped that such a comparative study will be possible in the future. At this point in time it appears that no similar technical study has been conducted on any of Mazziere’s other known works.

Technical analysis has assisted the treatment of the work; identifying the varnish as non-original and potentially problematic to remove in future prompted its entire removal during this treatment. Also, knowing that the silver leaf of the nimbus surrounding the central angel was originally likely to have appeared gold with a coating of shellac encourages the consideration of restoring this appearance by applying a conservation-grade modern equivalent.

In these ways the technical analysis has provided the necessary information to anticipate how the work’s materials will respond in the future, and what the work has been comprised of in the past, influencing conservation decisions in the present.
Acknowledgements
The author thanks the following staff members from Harvard Art Museums for their invaluable assistance throughout this research project; Kate Olivier, Teri Hensick, Kate Smith, Narayan Khandekar, Jens Stenger, Kathleen Kennelly and Tony Sigel from the Straus Center for Conservation, and Stephan Wolohojian and Danielle Carrabino from the Painting, Sculpture and Decorative Arts Department.

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References


Evans, ANAGPIC 2008, 22


Appendix 1: Technical Specifications

X-radiography was undertaken by the author with Teri Hensick, Paintings Conservator, Straus Center for Conservation. FT-IR, GC-MS and XRF analysis was undertaken by Dr Narayan Khandekar, Senior Conservation Scientist, Straus Center for Conservation.

Cross-section Preparation

Cross-sections were mounted in Bio-Plastic liquid polyester casting resin (Ward’s Natural Science, PO Box 5010, San Luis Obispo, CA 93403-5010). Samples were ground and polished to reveal the paint stratigraphy and examined by visible and UV reflected light microscopy (Zeiss Axioskop 2 MAT) using I3 (450-490nm) and D (355-425nm) ultraviolet band pass filters. Images were recorded digitally with a Phase One H-25 digital back (Phase One A/S, Roskildevej 39, DK-2000 Frederiksborg, Denmark).

Micro-chemical staining protocol

The protein stain, Amido black 2, was prepared and applied according to Martin (1977). The lipid stain, Sudan black B, was prepared and applied according to Johnson and Packard (1972). All other stains were prepared and applied according to Wolbers (2000).

Infrared examination and imaging

The work was imaged in infrared digital photography (IRDP), using a Phase One digital back designed to fit a Rollei medium format (X-Act2) in place of the photographic film cassette. This digital back has a silicon charge couple detector (CCD) sensitive up to ca. 1.1 microns. Digital resolution: Phase One P 45+ (5412 x 7216).

More detail was provided in infrared digital reflectograms (IRDR). Several main sections of the painting were imaged using an Inframetrics Infracam with a 15” lens, capturing images in 4 cm increments, which were then mosaiced together to form composite images. The Inframetrics Infracam (now made by SWIR) has a platinum silicide (PtSi) CCD sensitive between 1.1 and 1.8 microns. (1100-1800 nanometers_Digital array of 256 x 256 pixels).

X-radiography

X-radiography was conducted in February 2008, using Lorad LPX-160 X-radiograph apparatus. Kodak M-1 film was used, exposed at 55 kV, 5 nA, for 3 minutes 45 seconds, at 100” tube distance.
Fourier-transform Infrared Spectrometry

FTIR spectrometric analyses were carried out using a Bruker Vertex 70 infrared bench coupled to a Bruker Hyperion 3000 infrared microscope with a 15× objective. The sample was compressed on to a diamond cell (2 × 2 mm) with a stainless steel roller and the sample area defined by double shutters contained in the microscope. An absorbance spectrum was measured and subtracted against a blank background. The spectrum was compared with the Infrared and Raman Users Group (IRUG) database of artist’s materials.

Gas Chromatography – Mass Spectrometry

Lipid analysis

Samples were weighed and a 2:1 mixture of Methprep II (Alltech Associates, 2051 Waukegan Road, Deerfield, IL 60015) and benzene added to an equivalent of 1:1 weight per volume. The sample was heated to 50°C for half an hour to complete the transesterification of the fatty acids. Samples were injected via autosampler onto a DB-5 MS column (30m x 0.25mm, 1μm phase coating) using a splitless injector heated to 300°C. The Agilent 6890N GC oven heated the column from an initial temperature of 50°C (2 minutes) to 300°C at a ramp rate of 10°C/minute and maintained the final temperature for 10.5 minutes. The mass spectrum of the separated components was collected using an Agilent 5973 mass selective detector.

Polymer analysis

The sample was inserted into a quartz pyrolysis tube (CDS Analytical Inc, 465 Limestone Road, Oxford, PA 19363). The sample and tube was placed inside a platinum heating coil which was then placed into the pyrolysis injector (CDS Pyroprobe 2000) and pyrolyzed at 750°C for 10s. The sample then passed to a DB5-MS column (30m x 0.25mm, 1μm phase coating) through a split-splitless inlet (ratio 23.4:1, split flow 21 ml/min) heated to 300°C. The Agilent 6890N GC oven heated the column from an initial temperature of 40°C (1 minute) to 300°C at a ramp rate of 10°C/minute and maintained the final temperature for 20 minutes. The mass spectrum of the separated components was collected using an Agilent 5973 mass selective detector.

X-ray Fluorescence Spectrometry

Areas were examined in situ using a Rontec ArtTAX μXRF Spectrometer equipped with an electronically cooled X-Flash detector, which contains a silicon drift detector and high-speed,
low-noise electronics with a resolution of 160eV at a count rate of 10kcps. X-rays were produced by a low power tube with a molybdenum target. The beam was focused by polycapillary optics to a spot size of 70μm x 50μm. The analysis area was purged by a stream of helium. Analysis was carried out at 50kV for 200s. Bronk et al. (2001) have published a detailed description of this instrument.