A novel approach for high selective micro-sampling of organic painting materials by Er:YAG laser ablation

Maria Perla Colombini a,*, Alessia Andreotti b, Giancarlo Lanterna c, Maria Rizzi c

a Dipartimento di Scienze Ambientali e del Territorio, Università di Milano Bicocca, P.za Scienza 1, 20126 Milan, Italy
b Dipartimento di Chimica e Chimica industriale, via Risorgimento 35, 56126 Pisa, Italy
c Laboratorio Scientifico, Opificio delle Pietre Dure, V.le F. Strozzi 1, 50129 Florence, Italy

Abstract

A new approach for sampling micro-amounts of mainly organic materials from thin layers of a painting is described. A pulsed Er:YAG laser system operating at 2.94 µm was used for collecting ablate materials. The experimental ablation conditions optimised on reference paint layer samples resulted in using laser energy lower than 20 mJ at 15 pulses/s (pps) assisted by water/ethanol mixtures. The ablate materials condensed on glass coverslips were characterised by Fourier transformed infrared spectrometry (FT-IR) and gas chromatography-mass spectrometry (GC-MS) procedures. The results showed that the laser energy did not significantly degrade the ablate organic material collected which can be successfully identified. The procedure, tested and calibrated on reference paint layer specimens, was applied for the sampling and characterisation of two old paintings. The presence of overpaintings consisting of egg and Venice turpentine in one case, and of “beverone” over a varnish (linseed oil and Venice turpentine) in the other one was highlighted. © 2002 Éditions scientifiques et médicales Elsevier SAS. All rights reserved.

Keywords: Laser micro-sampling; Organic binders; FT-IR and GC-MS analysis; Paintings

1. Research aims

A painting is made up of several paint layers of thickness ranging between 5 and 40 µm which are difficult to collect selectively. When a characterisation of the organic matter is needed, sampling is a particularly difficult analytical step since organic matter is present in small percentages with respect to inorganic matter (pigments, ground, etc.). During the tests on the evaluation of a new Er:YAG laser system (λ = 2.94 µm) for cleaning paintings, many materials were ablated from the paint surface. In such cases, being able to collect the ablate material is a precise and selective sampling which would be a definite improvement on traditional methods based on mechanical tools. The main aims of this research were:

• to collect reproducible samples of the ablate material under optimised experimental conditions;
• to characterise the collected organic materials by means of sensitive analytical techniques, such as FT-IR and GC-MS;
• to check any changes in the matter composition induced by pulsed laser energy;
• to identify binders and varnishes in the ablate material from different paint layers.

Reaching these objectives entailed devising a powerful and innovative sampling method with limited invasivity in agreement with conservation guidelines.

2. Introduction

The characterisation of organic materials is one of the most difficult steps in the study of paintings [1]. Organic substances used as binders, adhesives or coatings are widespread in all the paint layers; moreover ageing and pollution processes may have strongly altered their original composition giving rise to products that are not easy to identify [2]. These factors make the work of modern restorers particularly difficult. In fact, before deciding on what conservation techniques to employ, they need accurate information on the organic and also the inorganic compounds employed both in the original painting and in any overpaintings due to subsequent restorations. While inor-
ganic compounds can be determined quite easily with non-invasive spectrometric techniques or just by taking microscopical amounts of samples, organic compounds entail collecting relatively large heterogeneous samples (0.5–1 mg) whose content of organic compounds does not exceed 200 µg. The collection of lower amounts often hampers the identification of the organic media. Since painting materials are present in a layered form with a thickness of a few micrometers, a selective collection by mechanical samplers is very difficult, and often the sample includes materials from adjacent layers, thus affecting the interpretation of the results. There is thus the need to have a sampling methodology that can selectively collect organic material from the different thin paint layers, to reduce the invasivity of the procedure and to obtain sufficient material to be analysed giving reliable results.

This paper describes a new sampling method based on the recovery of ablated materials on a glass coverslip after the paint surface has been radiated with a pulsed Er:YAG laser operating at 2.94 µm [3]. Reference paint layer specimens were employed to test and calibrate the overall procedure. The material collected at different laser energies and in the absence or presence of some wetting agents were characterised by applying analytical procedures based on FT-IR and GC-MS [4–6]. Comparisons between the proposed method and the scalpel sampling method highlight the potential of pulsed Er:YAG laser sampling. The optimised experimental condition found allowed the sampling of old paintings. In particular, the results related to samples from a 17th century canvas copy of Caravaggio’s “Christ crowned with thorns” (five repeated samples at 13 mJ laser energy with wetting agent U1);

The samples of paintings which are under restoration at the Laboratories of OPD in Florence were collected from:

1. A 17th century canvas copy of Caravaggio’s “Christ crowned with thorns” (five repeated samples at 13 mJ laser energy with wetting agent U1);
2. A 13th century panel “Virgin with Child” by Anonymous (one sample from the brown ground at 4 mJ laser energy, three repeated samples from the green curtain at 20 mJ laser energy with wetting agent U1 and two samples from the same area at 10 and 20 mJ laser energy with wetting agent U1).

Sampling were performed with scalpels along with the laser procedure in the same areas. According to the restorers, these paintings presented thick overpaintings, patinas and varnishes.

### 3.2. Old painting samples

### 3.3. Laser equipment and sampling procedure

A “Conservator 2940” model Er:YAG laser by Schwartz Electro-Optics (Orlando, FL, USA) operating at a wave-length of 2.94 µm was used [3]. The laser impact zone has an area of about 1 mm². The laser pulse rate was set to 15 pulses/s (pps); each laser pulse delivered a variable energy with wetting agent U1 and two samples from the same area at 10 and 20 mJ laser energy with wetting agent U1).

**Table 1**

<table>
<thead>
<tr>
<th>Sample name</th>
<th>Support</th>
<th>Ground layer</th>
<th>Paint layer</th>
<th>Surface layer</th>
<th>Composition thickness a (µm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>E1</td>
<td>Inert ceramic</td>
<td>Gypsum/animal glue (1:15)</td>
<td>Yellow ochre/lead white in whole egg tempera</td>
<td>Whole egg tempera</td>
<td>12</td>
</tr>
<tr>
<td>E2</td>
<td>Inert ceramic</td>
<td>Gypsum/animal glue (1:15)</td>
<td>Yellow ochre/lead white in whole egg tempera</td>
<td>Dammar resin</td>
<td>12</td>
</tr>
<tr>
<td>E3</td>
<td>Inert ceramic</td>
<td>Gypsum/animal glue (1:15)</td>
<td>Yellow ochre/lead white in whole egg tempera</td>
<td>Burnt umber/linseed oil</td>
<td>25</td>
</tr>
<tr>
<td>E4</td>
<td>Inert ceramic</td>
<td>Gypsum/animal glue (1:15)</td>
<td>Yellow ochre/lead white in whole egg tempera</td>
<td>Burnt umber/linseed oil on mastic resin</td>
<td>25</td>
</tr>
<tr>
<td>LSV Glass</td>
<td>None</td>
<td>None</td>
<td>None</td>
<td>Linseed oil/sandrac</td>
<td>–</td>
</tr>
<tr>
<td>DV Glass</td>
<td>None</td>
<td>None</td>
<td>None</td>
<td>Dammar</td>
<td>–</td>
</tr>
</tbody>
</table>

a The mean thickness of the surface layers was estimated by examining cross-section from the reference samples using an optical microscope.
energy to the target ranging between 5 and 50 mJ. A silver-coated hollow glass waveguide tube (1 mm i.d., 1.5 m length), whose end part looks like a pen, delivered the light to the target.

A large set of microscope glass coverslips (15 × 15 mm), used as sampling devices, was cleaned by washing with deionised water, methanol and acetone, dried in an oven and stored in cleaned tray boxes avoiding any contact with organic substances. Cotton gloves were used to handle the sampling devices. These were placed on the painting, and after laser impact, the ablated material condensed on the glass, thus giving the opportunity to collect the surface paint material for chromatographic and spectroscopic analyses (Fig. 1). Samplings on the specimens (described in Table 1) and on old paintings were performed at the following laser working conditions:

• a single pass at variable energies of 4, 10, 12, 13, 13.5, 15, 20, 30, 45 mJ;
• multiple passes at a fixed energy of 10, 13, 20 mJ repeating up to four times the treatment in the same area;
• a single pass at a fixed energy of 10, 12, 13, 15, 45 mJ with the wetting agents U, U1 and U2.

3.4. Apparatus

The following instrumentations were used:

1. A 5890 Series II gas-chromatograph (Hewlett Packard, Palo Alto, CA, USA) coupled with a quadrupole mass spectrometric detector mod. 5971A (electron impact 70 eV, ion source temperature 180 °C, interface temperature 280 °C).

2. “Trace gas-chromatograph 2000” (Thermo Quest, USA) coupled with a quadrupole ion trap mass analyser Polaris Q.

3. A Perkin-Elmer 1725× FT-IR spectrophotometer, a DMCS detector, an X-Y-Z micrometric sample holder for a 1.5 mm diameter pellets, KBr (Schilling spectroscopic grade). Spectra collection and calculations were made with PE Spectrum software (release 2.1).

3.5. Analytical procedure

3.5.1. GC-MS analysis

The glass sampling device with the ablate material was inserted into a vial, covered with ammonia solution 2.5 M and sonicated for 3 h. The ammonia extracts were analysed for the amino acid content and the residue for the neutral and acidic organic compound content according to a well-established analytical procedure [4–6], that has been slightly modified.

Chromatographic separation was performed on a chemically bonded fused silica capillary column HP-5 MS (Hewlett Packard) and Rtx-5MS (Thermo Quest, USA).

3.5.2. FT-IR analysis

The glass sampling device with the ablate material was gently scraped with a scalpel, the recovered material was admixed with about 5 mg of KBr in a mortar, milled and pressed to form 1.5 mm diameter micro-pellets.

4. Results and discussion

The FT-IR spectra of the ablated materials from the reference paint layer specimens in all the working conditions used were very similar to those of the material directly sampled with a scalpel. Except a small shift of carbonyl and double bonds stretching bands in reference specimens containing oil and dammar varnish, no important modification seems to have occurred during laser radiation. This is highlighted in Fig. 2, where spectra for the E2 specimen collected with the two sampling methods are compared. Generally speaking, a bulk sample is obtained by using the scalpel sampling method which means that traces of the background binders may interfere with the absorption of the compounds of the outer paint film as shown in Fig. 2. On the other hand, the Er:YAG laser, under controlled conditions, removes a very thin layer of the paint film thus reducing the
above interference and simplifying the identification of the ablate paint layer.

A deeper inspection of the ablate material was obtained by GC-MS analysis. The chromatograms for all the analysed samples showed that the binders and varnishes present in the specimens were selectively recovered and identified, in particular:

- **Proteinaceous media.** The ablate samples from E1 specimen exhibited an amino acid pattern quite similar to the one expected both in the presence and absence of wetting agents. The increase in the laser energy up to 45 mJ caused a loss of nearly 50% of serine (the simplest hydroxy amino acid) and an increase in proline and glutamic acid contents. The identification of the protein obtained by applying principal component analysis to the relative amino acid percentage contents of each sample (Fig. 3) showed that the egg protein is recognised for all the experimental conditions, though the 45 mJ samples are slightly shifted from the egg cluster. The increase in laser power also led to a general increase in the total amount of proteinaceous matter recovered, as shown in Fig. 4. The use of U1 wetting agent seems to favour the removal of the surface paint layer.

- **Lipids.** The fatty acid profile of linseed oil and of egg was, respectively, found in all the lipidic ablate material from LSV, E1 and E3 specimens for all the experimental conditions tested. Cholesterol and cholestanes were also detected in specimens containing egg. The increase in laser power increased the total amount of fatty acids recovered. Specifically, removing dicarboxylic acids seems more efficient than removing triglyceride units containing monocarboxylic acids, as shown in Fig. 5.

Polar wetting agents such as U and U1 seem to enhance this effect probably because they dissolve polar free dicarboxylic acids entrapped in the three-dimensional cross-linked polymer structure, thus facilitating their removal from the paint. Provided laser energies lower than 15 mJ are employed for sampling, these results highlight that it is still possible to identify the lipidic matter. In fact, as reported in Table 2, ratio values of palmitic to stearic acid ($P/S$), of azelaic to palmitic acid ($A/P$) and the sum of dicarboxylic acids ($\Sigma D$) agree with those reported in the literature for linseed oil and egg [1,4].

- **Natural resins.** The ablate matter from LSV specimens containing sandarac resin showed the distribution of pimamic, sandaracopimamic, isopimamic acid and to-
tarolone with no significant changes in any of the experimental conditions tested: chromatograms obtained for materials from scalpel and laser sampling are basically identical as shown in Fig. 6. As far as the ablate dammar resin is concerned, the neutral and acidic terpenoid compounds determined allowed its identification on the basis of the presence of shoreic and ursonic acids, \( \alpha \) and \( \beta \) amyrone. A mean ratio value of 9 between shoreic and ursonic acids was found; \( \alpha \) and \( \beta \) amyrone contents seem to increase with the laser energy.

The selectivity of sampling was checked on E4 specimens, which consist of three layers: the ground, a mastic resin varnish layer with a mean thickness of 12 \( \mu \)m and an overpainting consisting of Burnt umber in linseed oil with a mean thickness of 25 \( \mu \)m. The overpainting was removed with a single pass at 45 mJ laser energy using U1 as a wetting agent. In 12 repeated experiments of ablation, only in one case was a trace of moronic acid (marker of mastic) of the underlying resin detected. The reproducibility of the overall proposed method was tested by sampling six times the E3 specimen with a single pass at a laser energy of 45 mJ with a U1 wetting agent. The amount of each fatty acid of the linseed oil film, determined in the ablates, ranged between 1 and 70 \( \mu \)g with a variance (CV) of 25–35%. Considering that the analytical procedure alone has a relative standard deviation of 15–20%, the uncertainty introduced by the sampling procedure would seem to be acceptably low. All these evidences highlighted that the best sampling conditions to employ are a laser energy lower than 20 mJ, and a water/ethanol mixture as a wetting agent: in these conditions, the ablate areas of about 100 mm\(^2\) contained maximally 80 \( \mu \)g of organic material.

Since Er:YAG laser operates at 2.94 \( \mu \)m, it mainly interacts with –OH groups [3]. Polar compounds such as dicarboxylic acids strongly absorb the radiation and are easily ejected from the surface. Polar wetting solvents help the ablation mechanism as shown by the experimental results obtained on reference paint layer specimens. In fact, the sampling exploits a complex mechanism of material transfer from the surface [9]. The thermal effect due to the radiation absorption produces an increase in temperature and pressure which facilitates the vaporisation and gas expansion in micro-bubbles on the surface. The removal of material may follow several mechanisms: steam distillation, sublimation, explosion and mechanical transport. Since

Fig. 5. Dependence of amounts (\( \mu \)g) of recovered dicarboxylic acids (sum of azelaic, suberic and sebacic acid contents), monocarboxylic acids (sum of lauric, myristic, palmitic, stearic and oleic acid contents) and total acidic amount from the ablation of LSV specimens on various laser energies (12, 13.5, 15 and 18 mJ) in the absence of wetting agents.

<table>
<thead>
<tr>
<th>Sample name</th>
<th>Laser energy (mJ)</th>
<th>( P/S )</th>
<th>( A/P )</th>
<th>( \Sigma D )</th>
</tr>
</thead>
<tbody>
<tr>
<td>LSV</td>
<td>10, 12, 13.5, 15, 18</td>
<td>1.0</td>
<td>1.0</td>
<td>30</td>
</tr>
<tr>
<td>E1</td>
<td>13, 15, 45</td>
<td>2.4</td>
<td>&lt;0.05</td>
<td>0.9</td>
</tr>
<tr>
<td>E3</td>
<td>13</td>
<td>1.8</td>
<td>1.0</td>
<td>32</td>
</tr>
<tr>
<td></td>
<td>40</td>
<td>2.1</td>
<td>1.4</td>
<td>45</td>
</tr>
<tr>
<td></td>
<td>45</td>
<td>1.7</td>
<td>1.8</td>
<td>50</td>
</tr>
<tr>
<td>E4</td>
<td>13</td>
<td>1.5</td>
<td>1.1</td>
<td>31</td>
</tr>
<tr>
<td></td>
<td>40</td>
<td>1.6</td>
<td>2.5</td>
<td>62</td>
</tr>
<tr>
<td></td>
<td>45</td>
<td>2.1</td>
<td>2.4</td>
<td>65</td>
</tr>
<tr>
<td>Linseed oil</td>
<td>*</td>
<td>1.5</td>
<td>1.1</td>
<td>29</td>
</tr>
<tr>
<td>Egg *</td>
<td></td>
<td>2.2</td>
<td>&lt;0.1</td>
<td>3</td>
</tr>
</tbody>
</table>

\( \ast \) Literature mean values.

Table 2

Mean values of palmitic to stearic acid (\( P/S \)) and azelaic to palmitic acid (\( A/P \)) ratio values and the sum of dicarboxylic acids (\( \Sigma D \)) for the lipidic material ablated from reference paint layer specimens in the absence and presence of wetting agents at different laser energies.
compounds due to pyrolysis or to chemical transformation have not been observed in the ablative materials, significant photo-decomposition phenomena do not seem to occur during irradiation, thus avoiding doubts in the identification of the main components of a paint.

The encouraging results achieved with the reference paint layer specimens, seemed to suggest that the above sampling method could be applied to panel and canvas easel paintings. A copy of one of Caravaggio’s canvases and a 13th century panel were tested by applying the sampling to characterise in the former case a thick overpainting and in the latter some thin patinas and varnishes. In the first case, the results showed that the ablate matter did not contain drying oil and highlighted the presence of a protein and a terpenic resin. The medium of the overpainting was consisted above all of egg tempera identified by PCA analysis (Fig. 7) and Venice turpentine, showed by the presence of large amounts of 7-oxo-dehydroabietic (54.9%), dehydroabi-
etic (23.0%) and di-dehydroabietic (22.1%). To assess the depth of the overpainting, five samplings at 13 mJ laser energy were performed on the same area. In Fig. 7, the statistical treatment of amino acid data highlights that all the ablates belong to the egg cluster; moreover the presence of Venice turpentine was always observed showing the same terpenoid percentage profile. The thinning of the overpainting corresponds to a lower and lower amount of protein recovered after each sampling; in particular in the fifth sample, the amino acids were revealed at the detection limit of the analytical method. This sampling method not only allows the identification of the medium, but by removing layers selectively, it also provides data on how to safely use the laser in restoration cleaning. In the above case, a sixth laser pass for cleaning would be dangerous since it might remove the paint binder.

Concerning the panel “Virgin with Child”, the analyses showed a widespread presence of animal glue, along with traces of Venice turpentine and egg protein. Fig. 8 shows the amino acid patterns for samples collected by mechanical and laser methods. Using a single pass at 4 mJ laser energy removes just a very thin layer which is clearly mainly made
up of animal glue with only a trace of egg (confirmed by the presence of cholesterol) weighing 2.6 µg (as a sum of amino acid contents). The matter collected by the scalpel sampling contains several substances which also come from the substrate where the binder was egg. In fact, the amino acid profile included 9.9 µg (sample size was about 400 µg) and the amino acid profile resulted as a mixture of egg and collagen and was difficult to identify. Thus, the mechanical sampling method hampers the correct identification of substances in single layers. Moreover, the ablate samples from the green curtain of the painting exhibited single layers. Moreover, the ablate samples from the green layer in addition to a typical varnish consisting of Venice turpentine and linseed oil. In fact, in further samplings operated at one 20 mJ pass laser energy, beverone was no longer found, confirming that laser ablation is able to draw out selectively very thin films. 

5. Conclusions

The organic compound compositions which constitute paint layers were not affected by the laser energy in the ablation. In both reference paint layer specimens and in the old paintings, the organic compounds before the laser ablation and in the ablate material were found to nearly coincide. This means that Er:YAG laser equipment with threshold conditions of energies lower than 20 mJ at 15 ps assisted by wetting polar agents such as water/ethanol mixture (U1), can be safely used for selectively collecting thin layers from paintings. This allows the binders and varnishes in the painting to be correctly identified. The procedure is quite simple and fast. Moreover, since the Er:YAG laser acts mainly on hydroxyl groups, polar organic materials, which generally increase as a painting ages [2,10], absorb the laser pulse energy, with a small thermal effect limited to the surroundings of polar molecules. The use of water polar wetting agents which are adsorbed on the paint layer favours the thermal effect, which in turns induces the evaporation of water molecules from the surface. This heating effect leads to an enrichment of organic substances in the ablate mainly through steam distillation; in any case, other mechanisms such as sublimation, explosion and mechanical transport occur at the same time [11]. The result is to obtain a pre-concentration of the analytical sample for each paint layer present: the ablation for areas of about 100 mm² is able to collect a maximum of 80 µg of organic substance in the optimised experimental conditions. This result could not be achieved using traditional mechanical samplers, unless large amounts of material were collected. Thus, from the point of view of conservation, this sampling method has limited invasive effects, and therefore its use is advisable for works of art whenever an analysis of organic materials needs to be carried out.

Finally, since this sampling method offers a unique chance to study the effects between IR radiation and material transformation, more studies are in progress in order to correlate the reactivity of other –OH containing molecules used in paintings.

Acknowledgements

The authors gratefully acknowledge Mauro Matteini, Adele de Cruz and Myron Lee Wolbarsht for their helpful assistance and scientific discussion, and Paola Bracco, Oriana Sartiani and Kyoko Nakahara for the collection and management of the samples. One of the authors (MPC) is grateful to MURST for financial support.

References