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# Identification of Indigo and its Degradation Products on a Silk Textile Fragment Using Gas Chromatography-Mass Spectrometry

Jennifer Poulin

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*This article describes a new technique for the analysis of natural dyes on textile fibres. The use of m-(trifluoromethyl)-phenyltrimethylammonium hydroxide (TMTFTH) extraction and derivatization, followed by gas chromatography-mass spectrometry (GC-MS), has proven to be a simple and fast technique for the analysis of indigo dye on textiles. Not only does the procedure allow for the identification of the main dye component, indigotin, but it also provides information on the degradation of indigo on textiles, a topic that has received limited attention in conservation literature. This paper discusses compounds formed through the reactions of TMTFTH with indigotin and the main degradation products of indigo, 2-aminobenzoic acid, isatin and isatoic anhydride. In addition, a new degradation product has been tentatively identified as 2-benzyl-3-indolinone. The occurrence of these degradation products was investigated in samples obtained from an Indian textile from the Sultanate period (13<sup>th</sup> - 15<sup>th</sup> centuries) belonging to the Fine Arts Museums of San Francisco, woven, in large part, from indigo-dyed blue silk fibres. During a period of storage, pale yellow discolorations gradually appeared on tissue paper used to wrap the textile. Analysis of the tissue paper determined an abundance of indigo degradation products, in particular anthranilic acid (2-aminobenzoic acid). These compounds formed on the textiles fibres through oxidative degradation and subsequently volatilised to the surface of the paper, and also to the neighbouring fibres.*

*Cet article décrit une nouvelle technique d'analyse pour identifier des colorants naturels sur les fibres textiles. L'extraction et la dérivatisation à partir de l'hydroxyde de m-(trifluorométhyl)phényltriméthylammonium (HTMTFT), suivi du couplage chromatographie en phase gazeuse-spectrométrie de masse, constituent une méthode simple et rapide pour l'analyse de l'indigo sur des textiles. La procédure permet d'identifier non seulement la composante principale de ce colorant, l'indigotine, mais aussi les produits de dégradation de l'indigo fixé aux textiles, un sujet qui est rarement considéré dans la littérature. Cet article discute des composés créés lorsque le HTMTFT réagit avec l'indigotine, ainsi que des produits de dégradation principaux de l'indigo: l'acide 2-aminobenzoïque, l'isatine et l'anhydride isatoïque. De plus, un nouveau produit de dégradation a été identifié de façon préliminaire comme étant le 2-benzyl-3-indolinone. Des échantillons d'un textile indien de la période du sultanat (du 13<sup>e</sup> au 15<sup>e</sup> siècle), appartenant aux Fine Arts Museums de San Francisco et composés de fibres de soie teintes avec de l'indigo, ont fait l'objet d'examen afin de déterminer si ces produits de dégradation étaient présents. Le papier de soie qui enveloppait ce tissu durant sa mise en réserve pendant un certain laps de temps, fut retrouvé avec des taches jaunes. L'analyse du papier jauni révéla une abondance de produits de dégradation de l'indigo, et en particulier l'acide anthranilique (acide 2-aminobenzoïque). Ces composés se forment sur les fibres textiles selon les mécanismes de dégradation par oxydation, et par la suite peuvent se volatiliser et se déposer sur des surfaces adjacentes tel que le papier de soie, ou aussi sur des fibres voisines.*

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## Introduction

### *Current Methods for the Analysis of Natural Dyes*

For the past two decades, one of the most widely used methods for analysing natural dyes on textiles has been photodiode array high performance liquid chromatography (PDA-HPLC). For mordant dyes on textiles, acid-hydrolysis followed by PDA-HPLC is used. This procedure allows the vat dye indigo to be detected, but with very low yields. For indigoid dyestuffs on textiles, the more common method is hot solvent extraction using pyridine or dimethyl formamide to strip the colourant from the fibre, followed by PDA-HPLC.<sup>1,2</sup> Therefore, when analysing textiles containing both indigoid and mordant dyes, two separate sample preparation procedures are generally utilized.<sup>3</sup>

More recently, liquid chromatography-mass spectrometry (LC-MS) has come into use in the analysis of dyestuffs on textile

fibres. For the analysis of yellow flavonoid dyes and anthraquinones, the same multi-step acid-hydrolysed extraction procedure that was developed for PDA-HPLC is used.<sup>4,7</sup> For the analysis of indigoid dye compounds, as in analysis by PDA-HPLC, a different sample preparation procedure is required to dissolve the extracted indigo.<sup>8</sup>

### *Potential of TMTFTH Derivatization Followed by GC-MS in the Analysis of Natural Dyes*

There is a growing popularity in the use of m-(trifluoromethyl)-phenyltrimethylammonium hydroxide (TMTFTH) for the analysis of organic materials by gas chromatography-mass spectrometry (GC-MS) in the field of conservation. This reagent has the advantage that it is quick and offers a method of derivatization without complex sample preparation. Not only a methylation reagent, it hydrolyses triacylglycerides as well to form fatty acid methyl esters - essential in the analysis of drying oils. However,

one drawback of the technique is that TMTFTH is known to produce multiple derivatives from individual compounds, which may complicate analysis or result in lower sensitivity. TMTFTH is presently used in the analysis of many substances found in fine art and archaeological objects, including drying oils, fatty acids and soaps, fatty alcohols, waxes, natural resins, and alkyd paint media.<sup>9-12</sup>

This article describes a new technique for the analysis of natural dyes on textiles. It was suspected by the author that the alkaline nature of the TMTFTH reagent would allow extraction and methylation of natural dyes, which could then be analysed by GC-MS. The reagent was tested on reference fibres dyed with many natural dyestuffs including: indigo, madder, cochineal, Persian berries, old fustic, weld, lac, barberry root, tannins, alkanna, henna, brazilwood, logwood and red sandalwood.<sup>13</sup> The results were very promising for many of the dyestuffs. The application of TMTFTH with GC-MS to the characterization of indigo and its breakdown products on an Indian textile fragment from the Sultanate period (13<sup>th</sup> - 15<sup>th</sup> centuries) (**Figure 1**) is outlined in this article.

#### *History and Characteristics of Indigo as a Dyestuff*

Before describing the analysis of indigo on the Indian textile fragment, the history and characteristics of indigo as a dye will be briefly summarized. The dyeing of textiles with extracts of the indigo plant is a tradition dating back to ancient times. The word indigo derives from the Greek *indikon*, meaning “substance from India.”<sup>14</sup> The main colouring component in indigo (*Indigofera* sp.) is indigotin and it is present as the colourless precursor glycoside indican. Indigotin may also be obtained from woad (*Isatis*

*tinctoria*), where it is present in the precursor form of isatin B and indican.<sup>15</sup> The molecular structure of indigotin is shown in **Figure 2**. Indigo and woad require different growing conditions, and this was the prime factor in their distinct geographical usage prior to the 16<sup>th</sup> century. Indigo is grown in India and other tropical regions, and woad in the more temperate climate of Europe. For this reason, indigo did not come into extensive use in Europe until the 17<sup>th</sup> century when trading routes were more firmly established; at which point, for a number of reasons, indigo supplanted woad as the principal blue dyestuff in use.<sup>14,16</sup>

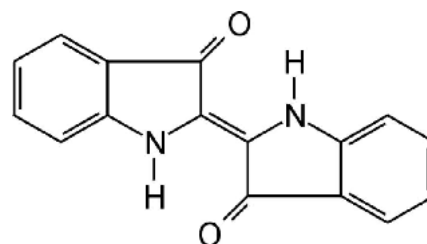
Indigo is classified as a vat dye; it does not need a mordant to bind it to textile fibres. During the dyeing process, both an alkaline and reducing medium are required to convert the insoluble indigotin to its soluble leuco-indigotin form, thus permitting it to enter into the fibre. After dyeing, exposure to oxygen in the air oxidizes the dye back to indigotin.<sup>16</sup> Deep shades of indigo are produced through multiple immersions in the dye vat that effectively deposit layer upon layer of indigo on a yarn or textile. Each layer is strengthened through intermolecular hydrogen-bonding between adjacent indigotin molecules. However, the layers themselves are susceptible to sliding apart and this can result in transfer through rubbing.<sup>14</sup>

Indigo is considered a very lightfast natural dye, earning a British Standard (BS) rating of 5 on wool, in comparison with flavone-containing old fustic and Persian berries, which both rate 2.<sup>17</sup> Although the degradation of indigo is not often discussed in relation to the substance as a dye, as this blue is one of the most predominant natural dye colours remaining in textile collections, its fading as a pigment has recently received greater attention.<sup>18,19</sup> In fact, when present in an upper paint layer, indigo is quite unstable and likely to fade from vibrant blue to a pale greyish colour, as observed in many 17<sup>th</sup> century oil paintings.<sup>20</sup>

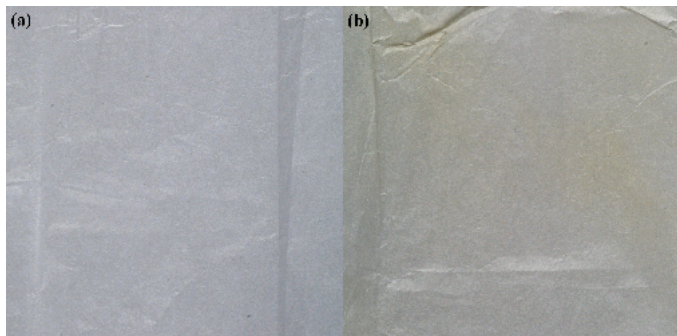
Many of the factors that influence the degradation of indigo as a pigment relate to physical characteristics such as particle size, thickness of pigment layers, and the presence of impurities.<sup>19</sup> The degradation of indigo used as a pigment has been observed in the absence of light,<sup>21</sup> however, the principal oxidation processes are more pronounced in the presence of light. The oxidative degradation of the main colouring matter in indigo, indigotin, results in cleavage into smaller compounds. These have been identified by mass spectrometry, and include isatin, isatoic anhydride, and anthranilic acid (2-aminobenzoic acid).<sup>19</sup>



**Figure 1.** Photograph of textile fragment *Warrior Procession with Standard Bearer*. Sultanate period, India, 13<sup>th</sup>-15<sup>th</sup> century, silk; weft-faced compound twill (samit), dimensions: 559 mm x 495 mm; Fine Arts Museums of San Francisco, Gift of George and Marie Hecksher, Acc. no. 2000.186.2. Image courtesy of the Fine Arts Museums of San Francisco.



**Figure 2.** Molecular structure of indigotin.



**Figure 3.** Side-by-side photographs of different areas of the tissue paper wrapping, comparing: (a) a site close to the edge of the paper where no visible discoloration was observed; and (b) a central area of the paper with obvious yellowing. The discolorations are not homogeneous, and the observed patches of darker yellowing may correspond to areas of the tissue paper that were in close proximity to areas of the textile (see **Figure 1**) with darker blue fibres.

The chemistry of the fading of indigo on textiles is a topic that has only received limited attention in the conservation literature since its fading is not often noticeable. However, the textile industry has given the topic significant consideration, especially as it relates to the degradation of indigo on cotton as in the case of indigo-dyed blue jeans.<sup>22,23</sup> The demand for blue jeans that have been selectively faded resulted in the development of an oxidative process known as acid-washing. A problem arose in the yellowing of these garments over time after treatment. It was determined that the degradation of indigo produced isatin and anthranilic acid.<sup>22</sup> As anthranilic acid crystals are pale yellow and isatin is orange in colour, both of these compounds were found to play a role in the yellowing of the denim in treated areas.<sup>22</sup>

Dyes are deposited inside textile fibres as a collection of differently sized particles ranging from finely dispersed molecules to large aggregates of compounds. The fastness of a dye is found to be directly related to the formation of large aggregates, as they present a smaller surface area than more dispersed compounds and, therefore, have fewer molecules exposed to oxidation processes. The greater the porosity of the fibre, the larger the dye aggregates that may form, and thus the more stable the dye.<sup>24</sup> The order of decreasing fastness with relation to porosity of natural fibres has been proposed to be wool, jute, linen, silk, and cotton.<sup>24</sup> This supports the observation that indigo is quite stable on wool fibres, and fades more rapidly on cotton fibres.<sup>25</sup> Therefore, theoretically, the permanence of indigo on silk should lie between that of wool and cotton.

#### *Background on the Sultanate Textile Fragment*

The textile fragment from the Fine Arts Museums of San Francisco originates from the Sultanate period, India, which dates from the 13<sup>th</sup> to 15<sup>th</sup> centuries A.D. The textile is constructed of a complex samit weave consisting of complementary wefts with inner warps. The intricate weft contains six different coloured silk

yarns, many of which are blue and dark blue.<sup>26</sup> The warp consists of red silk yarns. A photograph of the textile fragment is shown in **Figure 1**. The fragment measures approximately 560 mm x 500 mm and features a warrior procession led by a standard bearer. Each of the segments is constructed of a monochromatic figure on a monochromatic background of contrasting colour. There are four repeating patterns: a green warrior on a red background, white on dark blue, blue on red, and yellow on dark blue.

The textile was donated to the Fine Arts Museums of San Francisco in 2000 and has been in storage at the museum since that time. It is stitched to a silk backing and, at some point between 2003 and 2005, was wrapped in Light Impressions® wrapping tissue, no. 3700, and stored in a drawer of a powder-coated metal cabinet. In April of 2006, pale yellow discolorations were observed on the tissue in areas that were in contact with the textile. **Figure 3** shows a photograph of both yellowed and non-yellowed areas of the tissue paper. Samples of the fibres and tissue paper wrapping from the warrior motif textile were analysed to identify the dyes and to determine the cause of the yellowing on the paper.

#### **Methods**

##### *Preparation of Textile and Paper Samples*

Fibre samples of approximately 2 mm lengths were cut from a Schwebbe indigo on wool reference sample.<sup>27</sup> Very small samples from the warrior motif silk textile were removed by a conservator and sent to the Canadian Conservation Institute for analysis. For the yellowed tissue paper wrapping from the warrior motif textile, a strip of approximately 3 mm by 50 mm was removed from an area of concentrated discoloration and subsequently cut into smaller pieces. All samples were treated with a 1:1 solution of TMTFTH and toluene and heated at 65°C for 45 minutes in a capped 2 mL glass vial. The samples were then centrifuged at 1500 rpm for 2 minutes and the resulting liquid pipetted into a microvial insert, replaced in the original vial and capped.

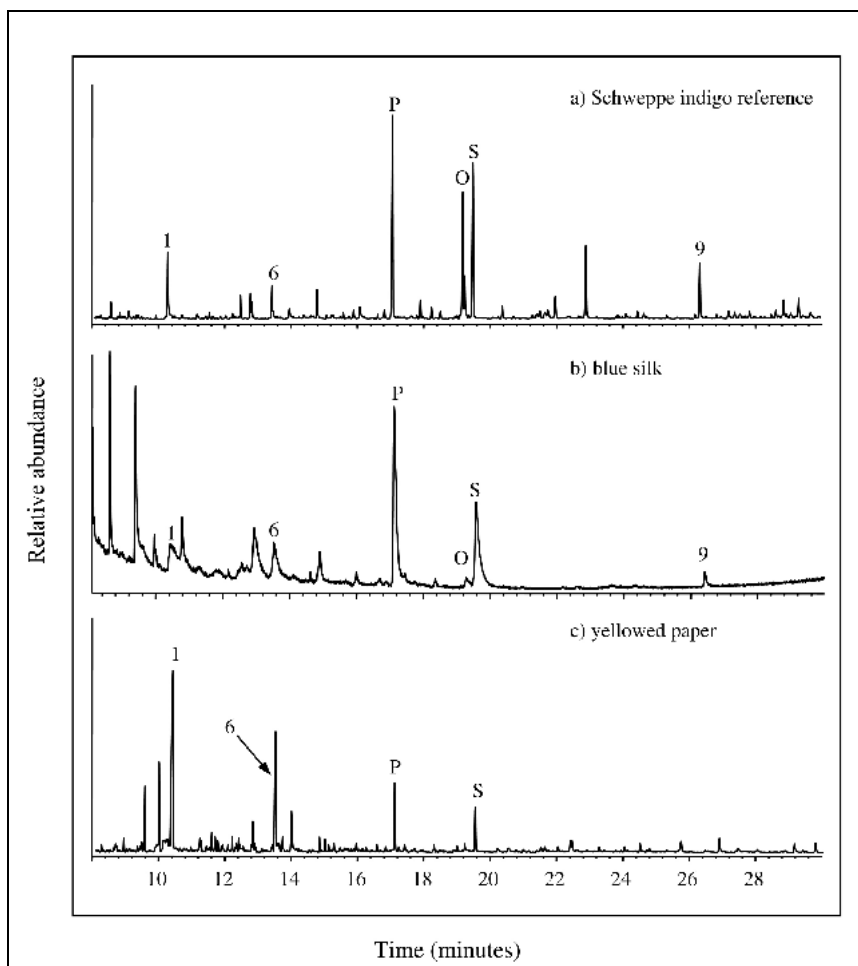
##### *Instrumentation*

An Agilent 6890 gas chromatograph (GC) with 7683 auto injector was used with an HP-5MS capillary column ((5% phenyl)-methylpolysiloxane; 30.0 m, 0.25 mm internal diameter, 0.25 µm film thickness), interfaced to a 5973 turbo pump mass spectrometer (MS) (Agilent Technologies, Inc., Santa Clara, CA 95051, USA). The GC inlet temperature was set at 250°C and the MS interface at 280°C. The GC oven was programmed from 50°C to 200°C at a rate of 10°C/min and then from 200°C to 300°C at a rate of 6°C/min and a final hold time of 15 min; total run time was 46.67 min. Ultra-high purity helium carrier gas was used with a constant flow of 1.0 mL/min. The MS was run in scan mode from 50-500 amu, with the source and quadrupole temperatures set at 230°C and 150°C respectively. The MS was operated in the electron impact (EI) positive ion mode (70 eV). Data were processed using Agilent Chemstation software (v.D.02.00.275).

## Results and Discussion

**Figure 4a** shows the partial total ion chromatogram (TIC) of the Schweppe indigo reference sample, extracted and derivatized with TMTFTH. As indigotin is the main colourant in indigo, the peak eluting at 26.36 minutes (labelled 9 on the chromatogram) is of prime interest. The peak, which represents the methylated derivative of indigotin, is identified as 2-bis-(N-methylindole-3-methoxy) from its mass spectrum. The structure of this compound is given in **Figure 5** (structure 9). It is interesting to note that the same compound was also identified by Prati et al. during the analysis of indigo pigment by pyrolysis GC-MS (py-GC-MS) with derivatization using tetramethylammonium hydroxide (TMAH).<sup>18</sup> **Figure 6** is an extracted ion chromatogram ( $m/z = 320$ ) of the Schweppe indigo reference, cropped to detail the eluting methylated indigotin peak. Along with the major indigotin peak (labelled b), three additional minor peaks (labelled a, c and d) are present. The corresponding mass spectra for these compounds are recorded in **Figure 7**. Based on the similarities in the fragmentation patterns, these minor components may be isomers of the methylated indigotin originating from the TMTFTH derivatization.

Other major peaks of interest in the Schweppe indigo reference sample are the methyl ester of anthranilic acid (labelled 1 in **Figure 4a**), and also a compound tentatively identified as 2-benzyl-3-indolinone (labelled 6 in **Figure 4a**), eluting at 10.32 and 13.46 minutes respectively. **Table I** gives the characteristic fragment masses corresponding to



**Figure 4.** Partial total ion chromatograms after derivatization with TMTFTH of: (a) Schweppe indigo wool reference sample; (b) blue silk fibres from the warrior motif textile (shown in **Figure 1**); and (c) yellowed tissue paper wrapping (shown in **Figure 3**). The numbers on the chromatograms are explained in **Table I**. The labels P, S and O indicate the peaks for the methyl esters of palmitic, stearic and oleic acids, respectively.

**Table I: Compounds and Mass Spectral Data Corresponding to Indigotin and its Degradation Products Identified by GC-MS after Derivatization with TMTFTH.**

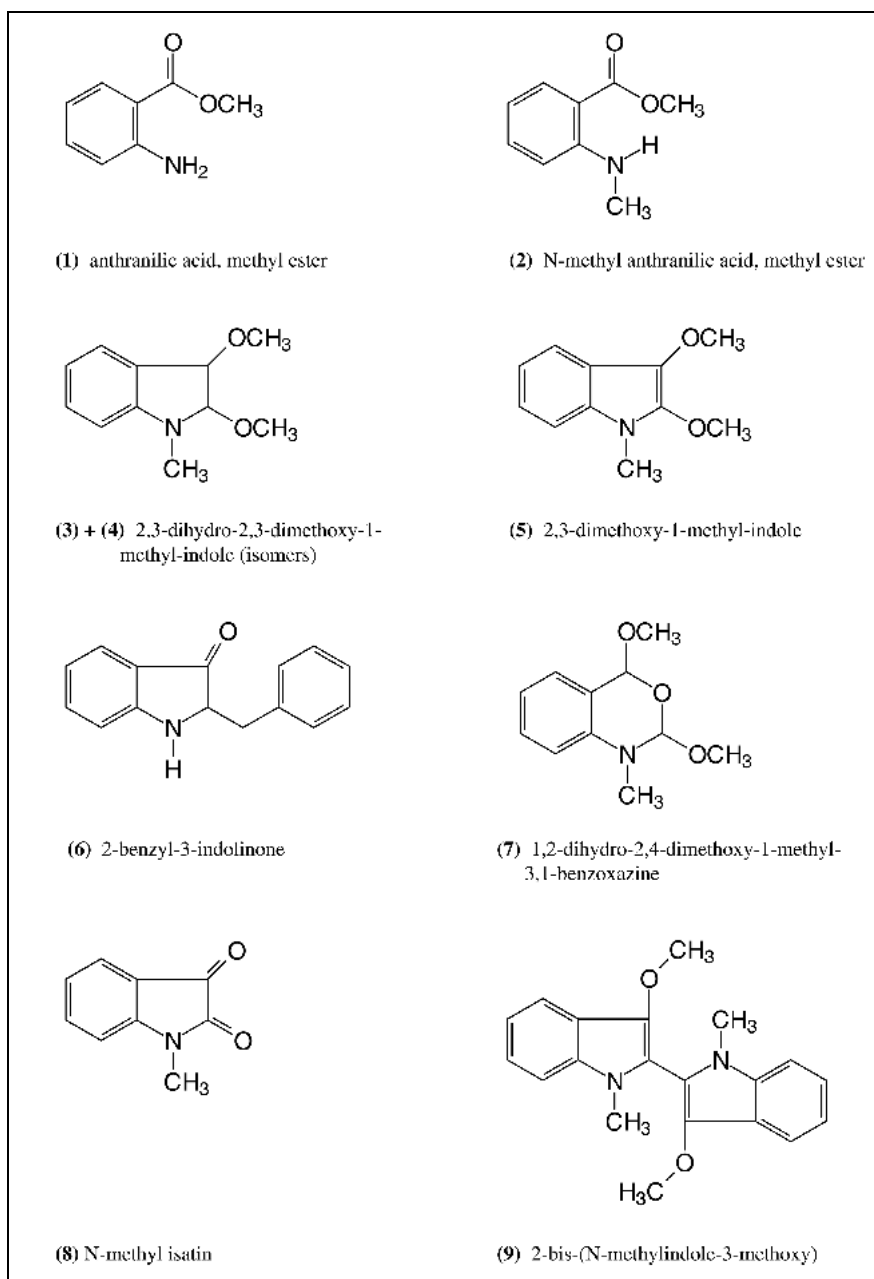
Peak Label	Compound*	Molecular Weight	$m/z$ (mass-to-charge ratio) Values of Characteristic Fragment Ions; % Abundance given in parenthesis
1	anthranilic acid, methyl ester	151	119 (100), 151 (59), 92 (46), 120 (31), 65 (18)
2	N-methyl anthranilic acid, methyl ester	165	165 (100), 105 (93), 104 (88), 132 (60), 77 (44)
3	2,3-dihydro-2,3-dimethoxy-1-methyl-indole (isomer)	193	146 (100), 161 (32), 130 (31), 118 (28), 178 (16), 193 (13)
4	2,3-dihydro-2,3-dimethoxy-1-methyl-indole (isomer)	193	161 (100), 118 (91), 146 (68), 132 (28), 178 (1), 193 (1)
5	2,3-dimethoxy-1-methyl-indole	191	161 (100), 160 (92), 132 (68), 148 (65), 191 (57), 176 (46)
6	2-benzyl-3-indolinone	223	132 (100), 164 (33), 77 (29), 105 (26), 223 (24), 104 (23)
7	1,2-dihydro-2,4-dimethoxy-1-methyl-3,1-benzoxazine	209	146 (100), 177 (36), 209 (25), 90 (12), 120 (8)
8	N-methyl isatin	161	104 (100), 161 (68), 105 (62), 78 (33), 133 (32), 77 (19)
9	2-bis-(N-methylindole-3-methoxy)	320	320 (100), 305 (86), 275 (43), 290 (25), 146 (25), 145 (23)

\*These compounds correspond to the marked peaks in **Figure 4** and **Figure 8**, and the molecular structures in **Figure 5**. Assignments were made based on mass spectral library matches and elucidation by the author from the mass spectral data.

these compounds and other peaks of interest. The proposed structures for these compounds are depicted in **Figure 5**. Anthranilic acid is a known degradation product of indigotin. Upon exposure to air and/or light, indigotin oxidizes to form isatin, which then further degrades to anthranilic acid in a secondary reaction.<sup>19,21</sup> It is possible that these breakdown products result either from the degradation of indigotin on the textile fibre, or from reaction conditions within the instrument. Other peaks of interest in the Schwebpe dyed wool sample, which do not pertain to the degradation of indigotin, are the methyl esters of palmitic, stearic and oleic acids (labelled P, S, and O on the chromatogram, **Figure 4a**), as well as lower levels of

branched-chain fatty acids, consistent with ruminant fats.<sup>28</sup> The Schwebpe reference fibres were also found to contain a significant level of sterols with main peaks identified as cholesterol and its methyl ether. These compounds originate from lanolin, which is naturally present on wool fibres.

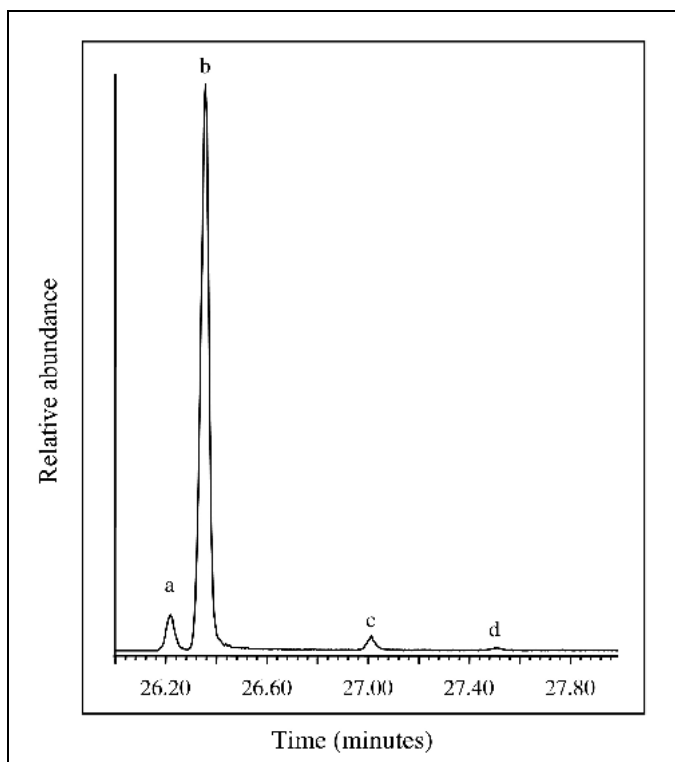
A chromatogram of a sample of blue fibres from the weft of the warrior motif textile is shown in **Figure 4b**. Although the sample consisted of only a few 2 mm long filaments, the indigotin peak is clearly visible (labelled 9 on the chromatogram). Also present are the methyl ester of anthranilic acid (labelled 1) and the compound tentatively identified as 2-benzyl-3-indolinone (labelled 6).



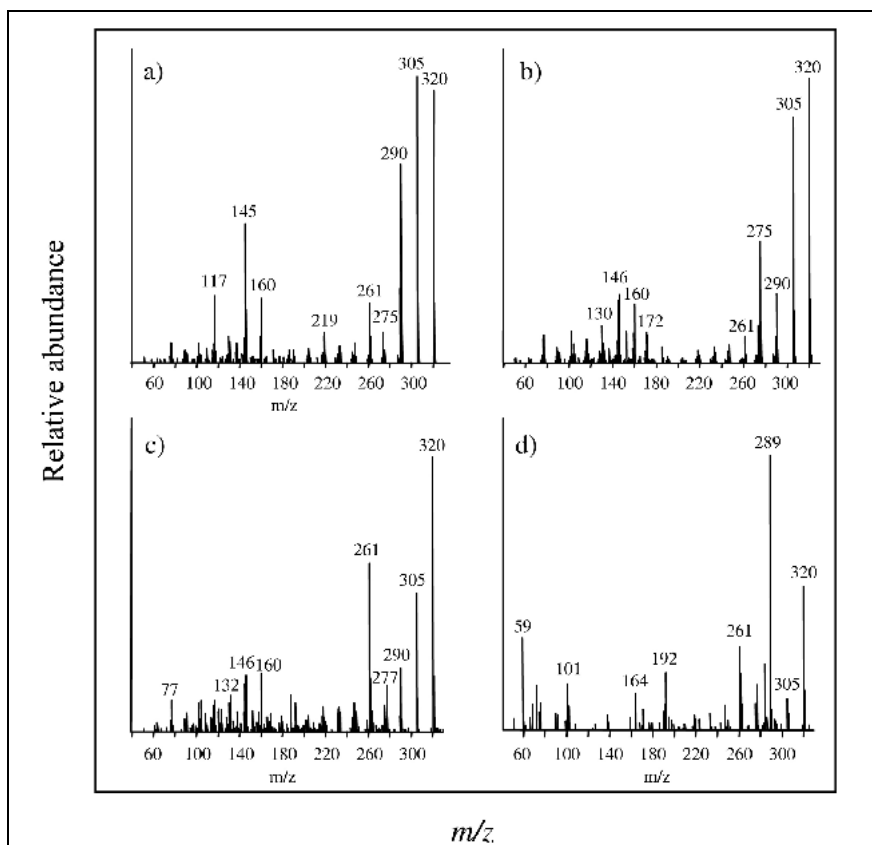
**Figure 5.** Molecular structures of indigotin and its degradation products identified by GC-MS after derivatization with TMTFTH. These molecular structures correspond to the compounds listed in **Table 1**. The structures were either identified from a mass spectral library or elucidated by the author from the mass spectral data.

2-Benzyl-3-indolinone (**Figure 5**, structure 6) has not, to this point, been documented in the literature as a potential degradation product of indigotin. The reaction pathway may proceed through initial loss of the carbonyl group followed by ring opening and loss of the amino group. Both anthranilic acid and 2-benzyl-3-indolinone are routinely found in indigo-dyed textiles analysed by GC-MS with TMTFTH derivatization in our laboratory, independent of the age of the textile. For this reason, it was initially assumed that the products were likely the result of reaction conditions within the instrument.

However, the analysis of the yellowed tissue paper wrapping from the warrior motif textile provided more information about the formation of these degradation products. A total ion chromatogram of a sample from a white area of the tissue paper did not indicate the presence of anthranilic acid or any other compounds linked to the degradation of indigotin. An extract from the yellowed paper, on the other hand, was found to contain these same two indigotin degradation compounds. The absence of available indigotin for degradation in the paper, coupled with the abundance of both anthranilic acid and 2-benzyl-3-indolinone, points to a gradual degradation of indigo on the textile fibres that then transferred to the paper, rather than degradation being due to analysis conditions. In the chromatogram of the yellowed paper, shown in **Figure 4c**, the main peak in the mixture is anthranilic acid (labelled 1 on the chromatogram), with a lesser amount of 2-benzyl-3-indolinone (labelled 6). According to the mass spectral data, the 2-benzyl-3-indolinone peak is co-eluting with an unidentified compound likely originating from the tissue paper. For the anthranilic acid and 2-benzyl-3-indolinone to be present in the tissue paper, they would have had to form first on the textile due to oxidative degradation of indigotin. As the paper was wrapped completely around the textile, the degradation products must have volatilised and deposited on its surface. The



**Figure 6.** Extracted ion chromatogram ( $m/z = 320$ ) of Schweppe indigo reference. The chromatogram has been cropped to highlight the four eluting peaks corresponding to derivatized indigotin and possible isomers. Mass spectra of peaks a, b, c and d are shown on **Figure 7**.



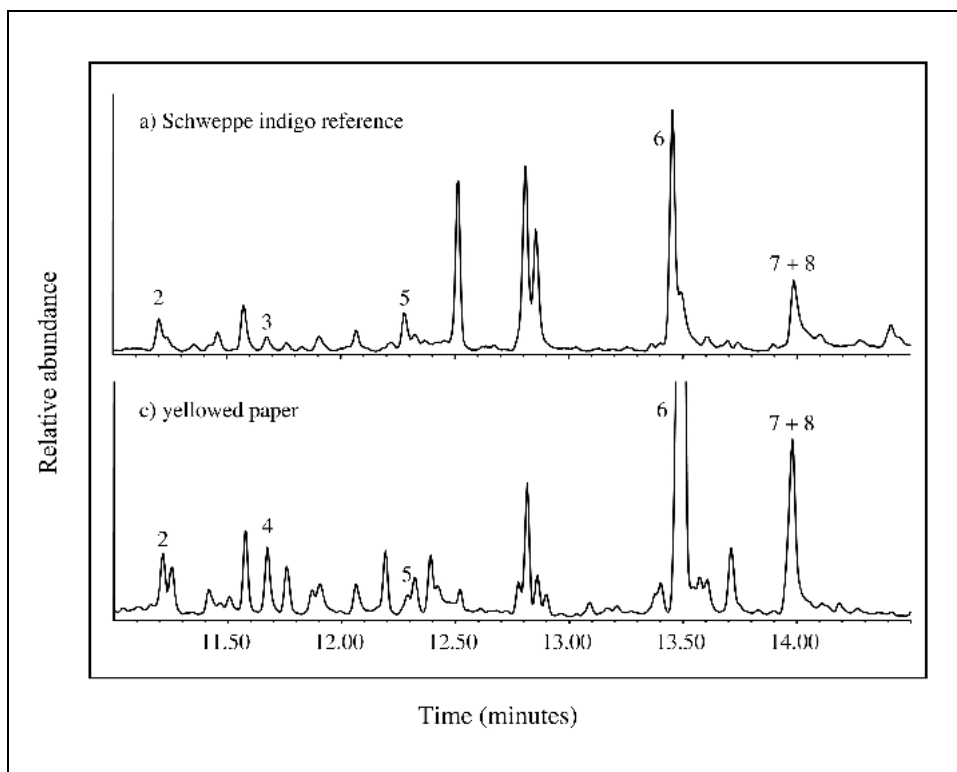
**Figure 7.** Mass spectra of indigotin and possible isomers after derivatization with TMTFTH.  $m/z$  = mass-to-charge ratio.

process happened gradually over a period of one to three years, at which time their increased concentration made them visible to the naked eye as a yellow discoloration. The most abundant compound present in the discoloured tissue paper, anthranilic acid, is pale yellow in colour and it is the presence of a high concentration of this compound that likely accounts for the yellow discoloration on the tissue paper. The warm extraction of the tissue in TMTFTH and toluene removed all visible colour, leaving white paper pieces in a yellow solution. It is interesting to note that low levels of these two main volatile oxidation products were also detected on neighbouring yellow-dyed weft fibres in the warrior textile.

There are additional compounds present in the reference indigo sample and the yellowed paper extract that represent different methylated derivatives of the indigo oxidation products: isatin, anthranilic acid and isatoic acid anhydride. Chromatograms of these samples have been expanded to show these peaks in **Figure 8**. With the exceptions of the methyl ester of N-methyl anthranilic acid (labelled 2 on the chromatogram), and N-methyl isatin (labelled 8), both of which were identified from a mass spectral library, the proposed structures were tentatively elucidated by the author from their mass spectra, which are listed in **Table I**. Although the sample from the warrior motif textile produced only a weak chromatogram (**Figure 4b**), some of the characteristic mass fragments of these compounds were found by ion extraction, indicating that they are present at trace levels.

Compounds 1 and 2 are derivatives of anthranilic acid, compounds 3, 4, 5 and 8 are derivatives of isatin, and compound 7 is a derivative of isatoic acid anhydride. The presence of multiple derivatives from individual compounds is not unusual when working with TMTFTH, and the result is often a more complicated chromatogram. However, often one of the derivatives forms preferentially, as can be observed in a comparison of the abundances of two derivatives of anthranilic acid: anthranilic acid, methyl ester (1) and N-methyl anthranilic acid, methyl ester (2). In **Figure 4a** the peak representing (1) is clearly visible, yet the peak representing (2) is mostly hidden in the baseline of the chromatogram and is only distinguishable in the cropped version of the TIC depicted in **Figure 8**.

The mass spectra of compounds 3 and 4 share the same molecular ion, as shown in **Table I**, but the observed difference in fragmentation patterns and in relative abundances of fragment ions indicate that they are likely isomers. Interestingly, only compound 3 was present in the Schweppe indigo reference and warrior motif silk textile, and only compound 4 was present in the yellowed paper sample (**Figure 8**).



**Figure 8.** Partial total ion chromatograms of (a) Schweppe indigo reference, and (b) yellowed tissue paper wrapping. The chromatograms have been expanded to show minor components originating from the derivatization of anthranilic acid, isatin and isatoic acid anhydride. The labels on the chromatograms are explained in **Table I**.

## Conclusions

Analysis by TMTFTH extraction and derivatization followed by GC-MS allowed indigotin and its degradation products to be identified on a silk Sultanate warrior motif textile fragment and its discoloured tissue paper wrapping. Methylated derivatives of the indigo oxidation products isatin, anthranilic acid and isatoic acid anhydride were identified. In addition, a new degradation product was tentatively identified as 2-benzyl-3-indolinone. The identified degradation products of indigotin are smaller and evidently more volatile than the parent compound. Although analysis shows that they form and remain, to an extent, on the originating textile fibre, they also volatilise over time and deposit on surfaces in close proximity, such as tissue paper wrappings and neighbouring fibres.

During the period of time in which the yellow discolorations were first observed, the warrior motif textile had been stored in the dark, in a humidity-controlled environment. This, of course, did not preclude the natural oxidative degradation that presumably occurred in the outer most layers of dye on the fibre. From previous research into this process, it is likely that these dark conditions slowed the rate of degradation.

One can clearly see from the photograph of the textile (**Figure 1**) that it has retained its vibrant blue and dark blue hues for over

500 years, and it is likely to do so for many years into the future. The fact that blue-dyed textiles are still so prevalent in museum collections is perhaps partially a product of the dyeing method of layering colour upon colour, a process unique to indigo, and partially due to the manner in which indigotin can enter into the pores of some textile. Thus the loss of indigo due to degradation is generally not very noticeable.

Analysis of natural dyes on textiles using TMTFTH derivatization and traditional GC-MS instrumentation has led to an increased understanding of the degradation of indigotin and the volatile compounds formed through the oxidative process. It has also given valuable insight into the cause and identity of yellow discolorations on materials in close proximity to indigo-dyed fibres. It is useful for conservators to understand that the discolorations originate from indigo dye on a textile, and not from foreign chemical interaction. Because both isatin and anthranilic acid are water soluble, yellow discolorations could potentially be removed by aqueous treatments.<sup>22,23</sup>

The analysis method described in this paper has proven to be an efficient and fast method for identifying indigo dye on textiles. As preliminary studies in our

laboratory have shown, a key advantage to this procedure centres on its effectiveness in the identification of many other natural dyes on textiles as well, including flavonoids and anthraquinones. Therefore, when analysing textiles for dye identification, only one fibre sample and one sample preparation step is required. The sample preparation procedure is very simple, utilizing readily attainable reagents, a short extraction time, and classic GC-MS instrumentation. The TMTFTH reagent has the added advantages of determining further information on dye compound degradation, and identifying additional organic compounds.

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## Materials

Meth Prep II (m-(trifluoromethyl)phenyltrimethylammonium hydroxide, TMTFTH): Alltech Associates, Deerfield, IL, USA, 60015.



Schweppe indigo reference material: see reference # 27.

Toluene, certified A.C.S: Fisher Scientific, Fairlawn, NJ, USA, 07410.

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