Assessing the comparative efficacy of forensic torch excitation wavelengths for dental restoration differentiation: a pilot study

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KEYWORDS

Forensic Odontology, Dental Fluorescence, Restorative materials, Canonical Discriminant Analysis, Post-mortem identification

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ABSTRACT

Background: The increasing sophistication of restorative materials in replicating natural tooth fluorescence poses a challenge for both antemortem and postmortem dental charting. This study evaluated the efficacy of various discrete excitation wavelengths, including those from common forensic torches, in differentiating modern restorative materials from tooth. Methods: Conducted within the Institute for Photonics and Advanced Sensing (IPAS) laboratory framework, six extracted human molars and five contemporary A3-shade restorative materials were examined via spectrophotometry across five excitation wavelengths: 266, 300, 365, 395 and 426nm. Canonical Discriminant Analysis (CDA) was applied to Peak Intensity and Peak Emission Wavelength data to quantify the multivariate statistical separation between groups. Results: UV-A wavelengths of 365nm and 395nm produced the most robust group separation. While 365nm excitation produced the highest absolute emission intensities for visual detection, 395nm excitation elicited unique spectral red-shifts in composite resins, providing the highest mathematical discrimination. Visible violet excitation at 426nm was the least reliable differentiator. Conclusion: For forensic practitioners, UV-A torches (365–395 nm) remain effective for detecting the tooth-coloured restorations tested here. While 365nm is optimal for rapid visual screening 395nm provides a more distinctive spectral fingerprints for formal characterisation. Despite improved fluorescence matching under visible light, the modern restorative materials tested here retain unique spectral signatures under UV-A illumination, though variable fluorescence intensity influenced detectability and may limit generalisation to other restorations or teeth.

INTRODUCTION

Forensic odontology has become a well-established and respected dental specialisation, providing a highly reliable means of identification.[1] This efficient method relies on the systematic comparison of clinical records with post-mortem examinations, identifying unique markers such as patterns of missing teeth and discernible restorations.[1,2] In modern dentistry tooth-coloured restorations have become the norm and their increasing aesthetic quality makes them, in some instances, remarkably difficult to distinguish from natural tooth. This evolution, while aesthetically pleasing, poses

significant challenges for both clinical practitioners and forensic odontologists. The limited detectability of tooth-coloured restorations complicates accurate dental charting and may result in incomplete or erroneous records. Detection of composite fillings during forensic examination has been shown to be possible using ultraviolet (UV) torches of various types.[3-6] However, how well this works on modern materials, which are designed to mimic natural tooth fluorescence has not been thoroughly investigated.

The physical phenomenon of UV-induced fluorescence from refracted sunlight was first described in 1845 by Frederick Herschel, but its application in dental science emerged significantly later. While early investigations in the 1920s reported the red fluorescence of dental tissues, the documented history of dental fluorescence as a rigorous scientific subject truly began in the mid-twentieth century. Research from the 1950s onwards into enamel and dentine fluorescence established its relevance for clinical diagnostics, and it has since been extensively studied for diagnostic and aesthetic purposes, such as caries detection, restorative material design and polymerisation control.[7-10]

Historically, research has relied on UV excitation (300-400nm), which is favoured for its high-energy photons that efficiently induce fluorescence in both natural teeth and restorative materials.[11, 12] Under UV illumination, human teeth typically exhibit peak fluorescence around 440nm, and dental materials are often engineered to mimic this wavelength for aesthetic compatibility[13-16]. However, a major limitation in existing published research is the variable nature of excitation sources, which complicates comparisons between studies. Most laboratory investigations employ wide-band spectra that do not replicate the discrete wavelengths used in common forensic UV torches.[17-19] While such devices are favoured in forensic settings because they are portable and battery-operated, their usefulness in their current form may decline as modern materials increasingly mimic tooth fluorescence.

High-energy visible violet light (around 405-430nm) is gaining interest as a safer and potentially more discriminative light source.[20, 21] Recent findings indicate that 426nm excitation induces maximum fluorescence emissions in human teeth[11], suggesting that visible violet wavelengths may elicit distinctive responses not yet fully considered by manufacturers of tooth-coloured restorative materials. Natural tooth fluorescence originates predominantly from dentine because of its organic content.[22-24] Restorative materials are comparatively homogeneous and largely derive fluorescence from added phosphors.[25-27] Although manufacturers aim to produce materials to reproduce the blue fluorescence of teeth under UV, their behaviour of these materials under visible violet light remains relatively unexplored.

Given the sensitivity of fluorescence spectra to excitation methods, this pilot study compares the emission spectra of natural teeth and a selection of contemporary restorative materials under discrete UV wavelengths corresponding to forensic torch LEDs (266, 300, 365 and 395nm) and a visible violet wavelength (426nm).[11, 18, 19] The primary aim is to assess the efficacy of these different wavelengths in differentiating natural teeth from a selection of contemporary tooth-coloured restorative materials and to contribute to best practices for material identification in forensic odontology.

MATERIALS AND METHODS

This study was conducted in accordance with the *National Statement on Ethical Conduct in Human Research 2007 (Updated 2018)*. The research protocol, encompassing the evaluation of forensic torch wavelengths (Project ID: 35804), was authorised as exempt from formal ethical review by the University of Adelaide Human Research Ethics Committee. This exemption was granted on the basis that the research involved only negligible risk using non-identifiable, archived dental specimens.

Sample Selection and Preparation: To facilitate integration into the Institute for Photonics and Advanced Sensing (IPAS) laboratory workflows and ensure high-throughput spectral consistency, a standardised sample set was used. Six intact, disease-free human adult molars were sourced from the University of Adelaide Dental School archives. Molars were selected primarily for their large, relatively flat surface areas, which permitted stable orientation during spectrophotometric measurement and provided a consistent volume of dentine to induce fluorescence.

Similarly, all restorative materials were prepared exclusively in A3 shade. As a common mid-range

clinical shade, A3 served as a representative baseline to establish the fundamental discriminative capacity of the selected wavelengths across different brands and compositions.
While it is acknowledged that tooth and material type and shade variations may influence fluorescence intensity and peak emission, this narrowed scope was essential for the primary objective of this pilot investigation. This evaluation establishes a protocol that can be expanded in future research to include a broader range of shades, materials and different tooth morphologies.
The materials included:

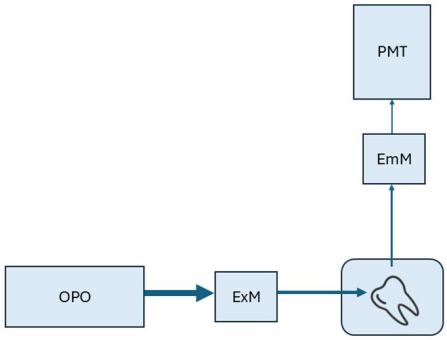
- capsule-delivered Filtek Z250 composite (3M)
- syringe-delivered Universal Flo composite (Gaenial)
- syringe-delivered Universal Injectable composite (Gaenial)
- syringe-delivered Supreme XTE Flowable composite (3M)
- capsule-delivered Fuji2 LC glass ionomer cement (GC Corporation)

Each material was dispensed and shaped into an approximate molar crown and light-cured according to the manufacturer's instructions. Post-curing, samples were not altered with polishing discs. Materials and teeth were stored in deionised water at room temperature for a minimum of 24 hours and air dried prior to spectral analysis.

Optical Excitation Setup
Fluorescence excitation was generated using a tuneable Optical Parametric Oscillator (OPO) (Opotek OPO), pumped by a Q-switched Nd:YAG laser integrated into an Edinburgh Instruments FLS980 Spectrometer.
As illustrated in the schematic Figure 1, the raw laser output was directed through an excitation monochromator (ExM) to isolate discrete wavelengths at 266, 300, 365, 395 and 426 nm. Emissions were processed through an emission monochromator (EmM) and detected via an air cooled R928 photomultiplier tube (PMT) (Hamamatsu Company, USA) to ensure high signal-to-noise ratio for lower-intensity UV-C and UV-B excitations. This dual-monochromator configuration ensured that the data used for subsequent Canonical Discriminant Analysis (CDA) represented true material fluorescence, free from scattered excitation light.

Statistical Analysis
The multivariate spectral differences (Peak Intensity and Peak Emission Wavelength) were analysed using Canonical Discriminant Analysis (CDA) to determine the potential for separation between the Molar and Material groups. CDA was selected for its efficacy in pilot studies with small sample sizes to provide Wilks' Lambda, classification accuracy and canonical function score plots. Analysis was performed using IBM SPSS Statistics v28

Figure 1. Schematic representation of the experimental optical geometry using the Edinburgh Instruments FLS980 Spectrometer. The setup illustrates the discrete excitation path (incident at 45° to the sample surface) and subsequent fluorescence emission collection at a 90° angle to minimise specular reflection while maximising signal-to-noise ratios.



RESULTS

This spectrophotometric analysis revealed distinct fluorescence profiles for natural tooth and restorative materials across five excitation wavelengths. Wide variability in intensities was noted within groups.

UV-C and UV-B Excitation (266nm and 300nm)
At 266nm and 300nm, both natural teeth and restorative materials exhibited relatively low-intensity fluorescence. While the groups remained statistically distinct, overlapping spectral ranges resulted in lower

canonical scores that indicate less robust differentiation compared to longer wavelengths.

UV-A Excitation (365nm and 395nm)

The 365nm wavelength achieved correct classification for all samples in the exploratory transformed spectral space (Figure 2, Panel A). 365nm produced the highest absolute emission intensities for restorative materials, with peaks reaching up to 50,000 counts. However, the emission peaks demonstrated considerable variability, appearing staggered across a range from 439nm to 490nm (Figure 2, Panel B). In contrast, 395nm excitation demonstrated the greatest statistical separation as evidenced by the largest distance between cluster centroids on the first canonical function (Figure 2, Panel C), yet it resulted in significantly lower photon counts (Figure 2, Panel D). This 395nm separation was driven by a consistent, uniform red-shift in the peak emission of the composite resins, which created a more predictable spectral signature than the high-variance peaks observed at 365 nm.

Visible Violet Light Excitation (426nm)

The 426nm visible light excitation proved to be the least reliable differentiator among the wavelengths tested. While two specific composite resins exhibited significantly higher intensities and distinct spectral morphologies, the remaining four restorative materials, including the Fuji 2 LC glass ionomer cement (GIC), demonstrated a high degree of spectral overlap with the natural molar cluster (Figure 3).

The high degree of separability observed between natural tooth structure and restorative materials was driven by distinct spectral signatures, particularly under UV-A excitation. As shown in Table 2, natural molars exhibited a consistent fluorescence peak in the 440–460nm region. In contrast, several composite resins demonstrated a characteristic red-shift in their peak emission wavelength, moving towards the 480–510nm spectrum. This wavelength-dependent shift was most pronounced at 395nm excitation, which provided the greatest mathematical distance between cluster centroids.

Figure 2. Canonical Discriminant Analysis (CDA) scatter plots and corresponding fluorescence emission spectra for natural molars and restorative materials under 365nm and 395nm excitation. A. CDA for 365 nm, illustrating clear group clustering and high classification accuracy. B. CDA for 395nm demonstrating the greatest statistical separation, evidenced by the increased distance between the Molar and Material centroids. C. Emission spectra for 365nm, highlighting high absolute photon counts and the variable, staggered peak positions among the different restorative resins. D Emission spectra for 395nm, illustrating the lower signal magnitude and the consistent, diagnostic red-shift observed in restorative materials. Note the significant difference in y-axis scales between B and D.

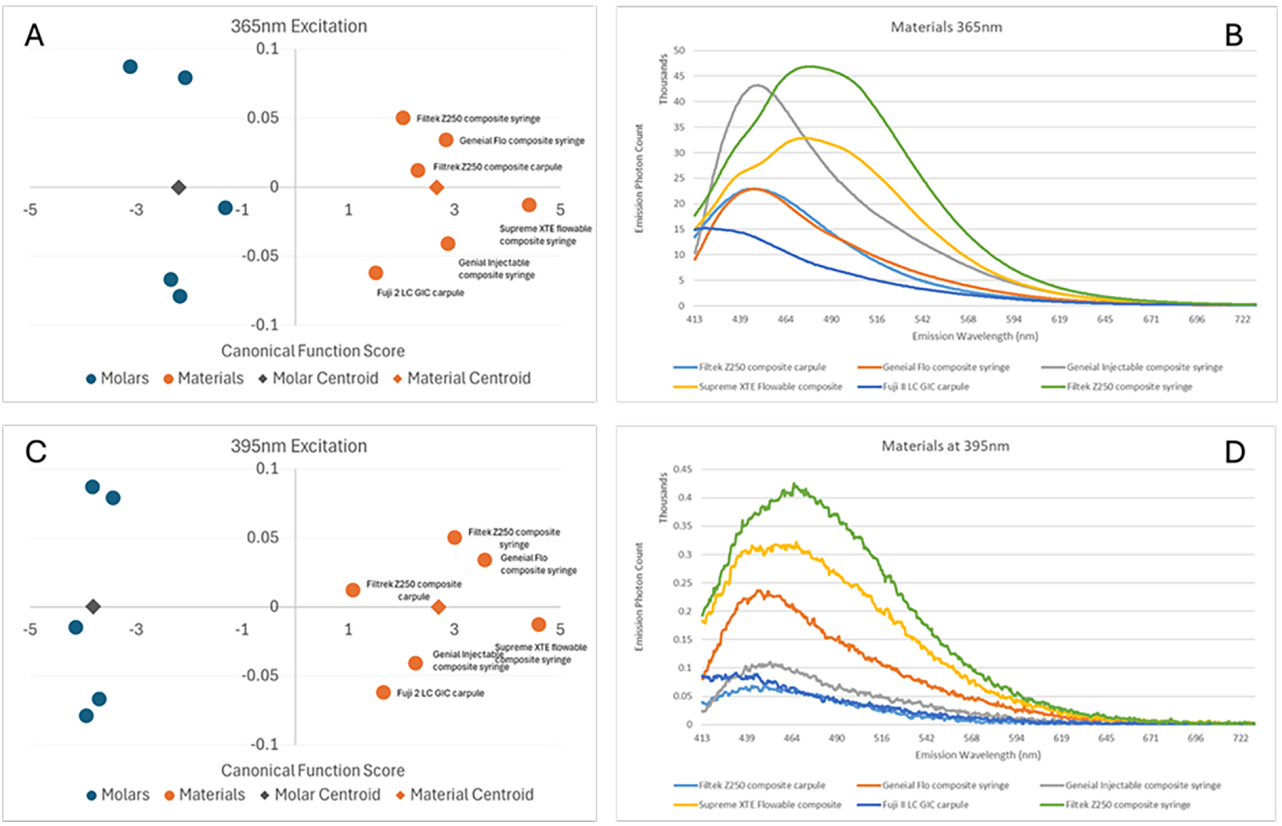


Figure 3. Canonical Discriminant Analysis (CDA) scatter plot and corresponding fluorescence emission spectra for 426nm excitation. A illustrates that 426nm is a less effective wavelength for fluorescence differentiation, evidenced by the misclassification of the 'Fuji II LC GIC carpule' within the natural molar cluster. B displays the emission spectra, where although Genial Flo and Genial Injectable showed higher relative photon counts, the overall signal magnitude and overlapping spectral characteristics limit the accuracy of material differentiation.

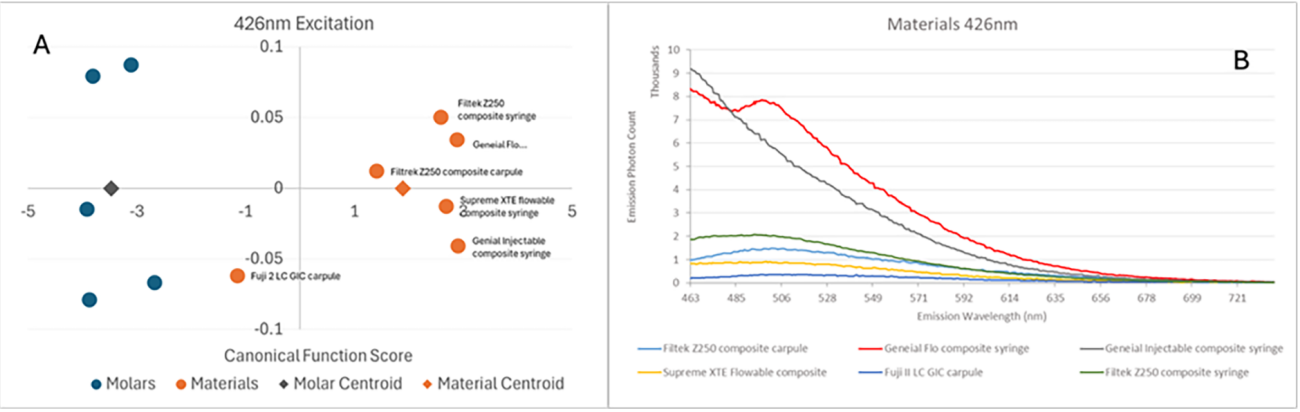


Table 1. Canonical Discriminant Analysis results showing statistical significance and classification accuracy for Molar vs. Restorative Material differentiation across five excitation wavelengths

Excitation Wavelength (nm)	Wilks' Lambda (Λ)	p-value	% Correctly Classified	Classification Result
266 (UV-C)	0.052	0.003	100%	Perfect differentiation
300 (UV-B)	0.057	0.004	90.9%	One molar misclassified
365 (UV-A)	0.057	0.004	100%	Perfect differentiation
395 (UV-A)	0.016	<0.001	100%	Most statistically significant
426 (Visible violet)	0.089	0.012	90.9%	One material misclassified

Table 2. Intensity (Photon Count) and corresponding Emission Wavelength (nm) at the peak of the spectral curve for all five excitation wavelengths

Sample ID	266nm		300nm		365nm		395nm		426nm	
	Intensity	λ_{Em}	Intensity	λ_{Em}	Intensity	λ_{Em}	Intensity	λ_{Em}	Intensity	λ_{Em}
Molar 1	143	439.9	300	444.2	5,414	451.5	194	442.0	1,589	487.1
Molar 2	343	463.6	122	473.9	3,038	466.3	168	452.3	1,088	487.5
Molar 3	101	455.9	99	515.2	1,680	477.4	50	456.9	2,865	502.7
Molar 4	138	448.9	201	447.7	4,172	461.9	94	446.4	959	501.8
Molar 5	265	461.9	905	439.9	12,524	451.9	8,023	451.1	2,401	500.3
Material 1	1,183	436.0	1,440	439.9	23,002	446.8	69	441.6	1,477	500.9
Material 2	3,493	477.7	15,185	434.7	22,094	446.4	237	444.7	9,338	463.0
Material 3	984	443.3	4,393	446.4	4,230	443.8	111	451.9	9,173	445.5
Material 4	6,040	487.3	17,037	477.1	32,074	476.3	322	457.9	933	496.0
Material 5	290	441.6	3,724	412.6	15,230	436.3	91	432.4	364	515.6
Material 6	3,519	479.1	20,294	477.4	46,095	477.8	426	485.8	3,085	499.3

DISCUSSION

The present study advances our understanding of material identification in forensic odontology by elucidating the discriminative power of different excitation wavelengths in detecting restorative materials. The findings underscore the critical role of wavelength selection in fluorescence analysis. While the findings of this pilot study indicate distinct fluorescence profiles between the teeth and materials tested, the variance observed in peak intensities and spectral morphology suggests that a larger sample size, encompassing different tooth morphologies and a broader range of restorative shades, would likely reveal points of spectral overlap. This variability is important, since the discriminative boundaries apparent in a controlled laboratory setting are likely to become less distinct in clinical or forensic scenarios.

Although modern restorative materials are engineered to mimic the fluorescence of natural teeth, the spectral signatures of the materials tested here remained distinct under specific UV-A excitation wavelengths. A key finding is the superior performance of 365nm and 395nm excitation wavelengths, which correspond to standard emissions from commonly used forensic torches. The high photon counts at 365nm facilitate rapid, preliminary screening, enabling practitioners to swiftly identify restorative materials in a clinical or forensic setting. This wavelength's high-intensity output ensures that restorations remain clearly visible even when viewed through filtered lenses required for UV torch use. Conversely, the subtle red-shift, a phenomenon likely linked to the specific phosphors and photo-initiators, such as Lucirin TPO or Ivocerin, used in modern universal shades[15, 25], induced by 395nm excitation offers a unique molecular fingerprint for synthetic materials, providing a robust signature for confirmatory, computer-aided analysis. Ultimately, the high inter-sample variability at 365nm ensures that most materials will exhibit some form of intense fluorescence, whereas the subtle but reliable red-shift at 395nm ensures that these materials are fundamentally distinguishable from natural tooth structure on a molecular level. Because natural dentine fluorescence is primarily driven by collagen and amino acid cross-links that do not exhibit this specific shift, 395nm provides a unique optical fingerprint for synthetic materials.[8, 23] The Canonical Discriminant Analysis demonstrated that separation between the Molar and Material clusters is not only based on intensity variations, but on fundamental differences in peak emission wavelength.

While these results are likely affected by the pilot nature of the study and its limited sample size and type, the significant canonical function scores for both 365nm and 395nm confirm that these wavelengths elicit distinctive optical fingerprints that are statistically superior to those elicited by 266nm or 426nm excitation.

The trade-off between visual accessibility and mathematical distinctiveness is crucial for practitioners to understand, as it guides the appropriate use of these wavelengths in different stages of material identification. While 365nm offers high visual contrast for initial identification, 395nm provides the most mathematically distinct signature for formal, high-certainty material characterisation where mathematics outweighs any need for apparent visual contrast.

The results of this investigation suggests that using a single wavelength alone is not sufficient for tooth coloured differentiation. Therefore, a forensic torch with at least 2 selectable wavelengths of 365nm and 395nm is suggested for differentiation.

Our findings also underscore the limitations of visible violet or blue light torches for restoration detection. The high inter-sample variance and spectral blending with natural molars observed at 426nm render this wavelength unsuitable for consistent forensic differentiation. Methodologically, while our study's exclusive use of molars provided a consistent experimental baseline, it also introduced constraints. Future research should expand sample sizes to include anterior teeth with varying dentine-to-enamel ratios, as these may exhibit greater fluorescence intensity and affect contrast ratios observed in this study.[12] Furthermore, while the decision to use only A3 shade materials was necessary for high-throughput analysis, this limits the generalisability regarding full VITA shade options. Darker shades or highly translucent incisal resins may contain different concentrations of fluorescing agents that alter classification accuracy. Additionally, the consideration of stratified layering techniques, where multiple resins are used in a single restoration, can help determine the extent to

which laminated materials improve or reduce detection efficacy under UV forensic torch use.[28, 29]

CONCLUSION

This pilot study confirms that despite the increasing aesthetic sophistication of modern dental materials, the ones tested here remain statistically and visually distinct from natural tooth structure under specific ultraviolet excitation. However, larger studies are required to determine whether overlaps between tooth and materials fluorescence become apparent when a larger population is sampled. Practitioners must recognise that the use of a UV forensic torch does not guarantee all composites are apparent, given the range of shades and brands available on the marked. Nevertheless, the 365nm and 395nm wavelengths, common to many forensic torches, demonstrated the highest efficacy for differentiation, with 395nm providing the most robust mathematical separation through unique spectral shifts in restorative resins. Conversely, visible violet light at 426nm proved less reliable, particularly for identifying glass ionomer cements, suggesting it should not be the primary tool for forensic dental identification. We recommend that forensic practitioners prioritise the use of 365–395nm UV-A torches during post-mortem examinations to ensure the accurate detection of clinically valuable restorative identifiers. Future research should expand sample sizes and consider stratified layering techniques to enhance understanding of material differentiation under UV torch use.

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SUPPLEMENTARY MATERIAL

The supplementary figures provide additional visualisation of the fluorescence responses observed across the excitation wavelengths investigated in this study. This data supports the primary findings presented in the main manuscript by illustrating the Canonical Discriminant Analysis outcomes at 266 nm and 300 nm excitation (Figure S4, S5), alongside comparative fluorescence emission profiles for both natural molars and restorative materials across the investigated wavelength range (Figure S6, S7, S8,S9).

Figure 4s. Canonical function at 266nm excitation.

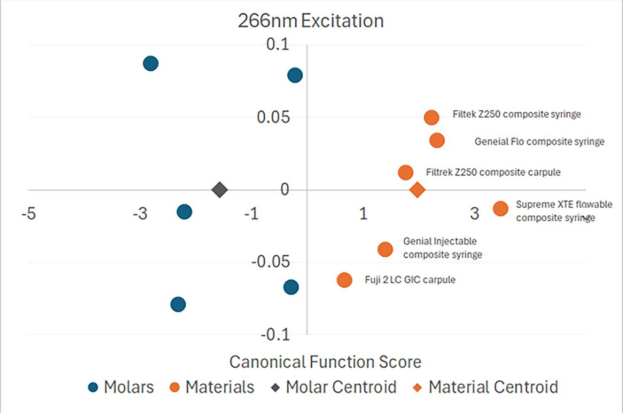

Figure 5s. Canonical function at 300nm excitation

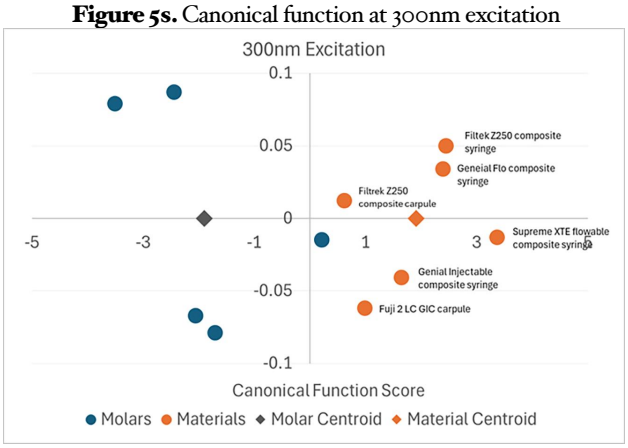

Figure 6s. Fluorescence emission for molar samples at each of the UV excitation wavelengths

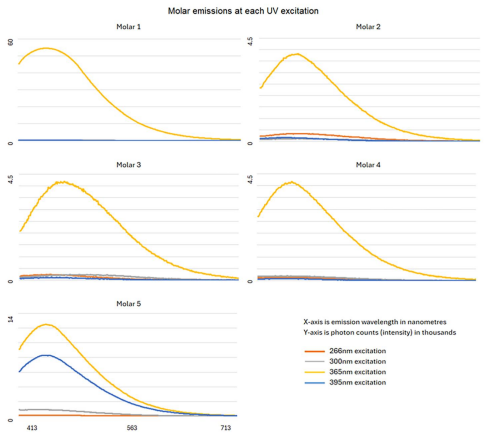

Figure 7s. Fluorescence emission for material samples at each of the UV excitation wavelengths

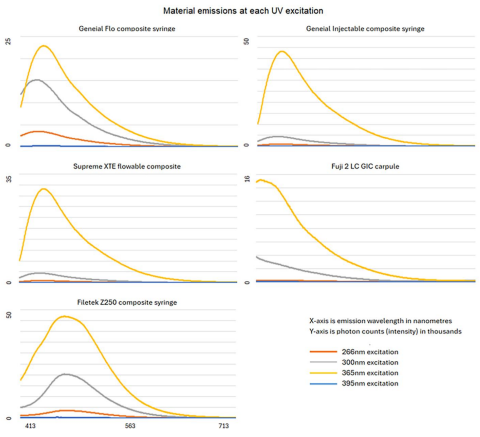

Figure 8s. Comparative emission spectra for materials under each excitation wavelength

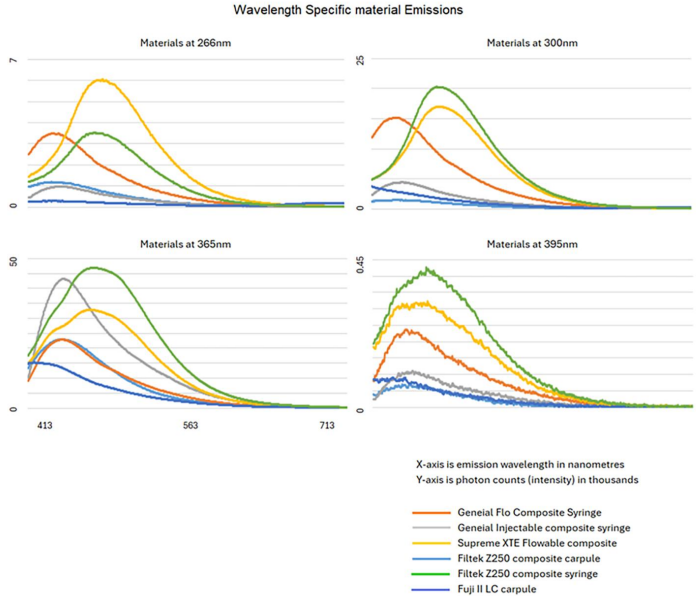

Figure 9s. Comparative fluorescence emission profiles under 426nm excitation. Individual spectral curves for natural molar specimens (grey) and restorative materials (blue) illustrate the significant overlap in emission morphology and intensity at this wavelength. To highlight the lack of diagnostic separability, two high-intensity material outliers were excluded. The dotted trendlines represent the polynomial-smoothed averages for the molar and material groups, respectively, confirming that the mean spectral signatures are nearly indistinguishable. This overlap demonstrates why 426nm was excluded from formal Canonical Discriminant Analysis and is considered unsuitable for consistent forensic differentiation.