The Fluorescence Properties of Contemporary Tooth-Coloured Restorative Materials: Its Forensic and Clinical implications

by

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Abstract

Introduction

Rising demand for aesthetic dental restorative materials and advancements in material science have led to the development of a variety of tooth-coloured restorative materials. The development of novel clinical techniques such as layering has enabled clinicians to create dental restorations that replicate the optical nuances of the natural tooth structure. However, this also means that identification of such restorations can be challenging. Accurate recording of materials used for restorations is essential for clinical records in patient care, as well as for epidemiological purposes and for forensic dental comparisons.

Fluorescence is an optical trait of tooth-coloured dental materials, which is intended to provide life-like aesthetics to restorations. The work described in this thesis explores the fluorescence properties of tooth-coloured dental restorative materials, and how these can be used to identify the presence of restorations. It also includes studies of how fluorescence is affected by moisture and by exposure to high temperatures.

Methods

In study 1 & 2 (Chapter 3 & 4) of this thesis, the fluorescence properties of dry and wet samples of various tooth-coloured restorative materials were evaluated using a fluorescence-based digital single-lens reflex (DSLR) camera. Following on from this, in study 3 (Chapter 5), the diagnostic reliability and validity of fluorescence-aided identification of restorations (FAIR) was compared with another optical method

(digital imaging fibre optic transillumination (DiFOTI) using near infrared light) and with conventional dental examination using a mirror and probe, for identifying tooth-coloured restorations. Given the strong performance of the FAIR method, in study 4 (Chapter 6), FAIR was then compared with conventional white light illumination (CM) for the selective removal of tooth-coloured RBC restorations, measuring the extent of loss of tooth structure, the amount of residual restorative material, and the time taken to remove the restorations. Finally, in study 5 (Chapter 7), the effect of heat on the fluorescence properties of contemporary tooth-coloured restorative materials and of natural tooth structure was assessed, using a range of temperatures (200 ◦C, 500 ◦C, 900 ◦C, and 1200 ◦C).

Results

Examination of the fluorescence properties of dental restorative materials in study 1 revealed that fluorescence properties were influenced by water sorption of the materials. Violet light (405 nm) produced the greatest range of luminosity values (10–204) amongst the tooth-coloured restorative materials and was the best wavelength for discriminating between restorations and tooth structure. The optimal optical filter combinations with violet light were orange or yellow filters. In study 2, significant variations were found in fluorescence luminosity amongst different shades for a given brand and material type. There were also significant variations among the red, green and blue colour channel values for all materials, according to their shade, with the exception of Fuji IX. In study 3, both the sensitivity and specificity of the FAIR method (95% and 97%, respectively) were found to be significantly higher than those for DiFOTI (82% and 82%) and for conventional inspection (71% and 82%). In study 4, fluorescence visualisation was found in study 4 to aid the selective removal

of tooth-coloured restorative materials. In comparison to working under white light illumination, when using FAIR showed a lower increment in inter-cuspal cavity width (FAIR: $p = 0.17$, CM: $p = 0.0025$). In study 5, all tooth-coloured restorative materials were found to undergo changes in their colour and in their fluorescence properties, at each of the temperatures used. Resin-based restorative materials still fluoresced after treatment at 200^{0}C, and after treatment at 500^{0}C underwent major colour changes due to volatilisation of resin. Materials containing inorganic fluorophores still fluoresced after treatment at 900^{0}C, while after treatment at 1200^{0}C, none of the materials tested in this study showed any fluorescence emissions.

Conclusions

The work presented in this thesis shows that information about the unique fluorescence characteristics of tooth-coloured restorative materials has several key applications. Firstly, all materials tested could be detected and discriminated from natural tooth structure when using 405 nm light for excitation. Secondly, a fluorescence-based examination was superior to conventional examination and to DiFOTI for identifying tooth-coloured restorations. This fluorescence method allowed such materials to be removed selectively, with less damage to adjacent health tooth structure. Finally, dental restorative materials containing inorganic fluorophores still fluoresced after treatment at 900 $^\circ$C, and this feature could aid in their identification, for forensic purposes, in victims of fires.

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List of publications arising from the thesis

Publications during candidature

Peer reviewed papers

1. Kiran R, Chapman J, Forrest A, Tennant M, Walsh LJ. Forensic applications: Fluorescence properties of tooth-coloured restorative materials using a fluorescence DSLR camera. *Forensic Sci Int*. 2017;273:20–8. https://doi.org/10.1016/j.forsciint.2017.01.022
2. Kiran R, Chapman J, Tennant M, Forrest A, Walsh LJ. Direct tooth coloured restorative materials: A comparative analysis of the fluorescence properties among different shades. Accepted for publication in *Int J Esthet Dent, on* 26/11/2019.

3. Kiran R, Chapman J, Tennant M, Forrest A, Walsh LJ. Detection of tooth-coloured restorative materials for forensic purposes based on their optical properties: an in vitro comparative study. *J Forensic Sci* 2019;64:254–9. https://doi.org/10.1111/1556-4029.13851

4. Kiran R, Chapman J, Tennant M, Forrest A, Walsh LJ. Fluorescence-aided selective removal of resin-based composite restorative materials: An in vitro comparative study. J Esthet and Rest Dent. 2019 Oct 16. First published: 16 October 2019 https://doi.org/10.1111/jerd.12536

5. Kiran R, Chapman J, Tennant M, Forrest A, Walsh LJ. Effect of Heat on the Fluorescence Properties of Tooth-Colored Restorative Materials and Their Forensic Implications. Journal of forensic sciences. 2019 Nov;64(6):1698-706. https://doi.org/10.1111/1556-4029.14122

Conference abstracts

Detection of tooth-coloured restorative materials for forensic purposes based on their optical properties: an in vitro comparative study.

1. Poster presentation

   IADR Conference, Australia and New Zealand Division 2017
   Adelaide, Australia

2. Paper presentation

   Indian Prosthodontic Society Conference 2017

   Bali, Indonesia

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>>*<<

“Reality is merely an illusion, albeit a persistent one.”

- *Albert Einstein*

Chapter 1 Literature review and Rationale

1.1. Introduction

The objective of this chapter is to explore aspects of the fluorescence and phosphorescence behaviour of human teeth under ambient and artificial ultraviolet (UV) and visible light that are critical for achieving an aesthetic restoration of tooth structure. In this chapter, the major fluorescence traits of hard tissues of teeth and tooth-coloured restorative materials are reviewed, drawing on relevant peer-reviewed papers. This chapter will also discuss the various methods used to analyse and quantify the fluorescence traits of human tooth structure and tooth-coloured restorative materials, with a particular emphasis on the forensic and clinical implications. It also discusses in brief where clinical techniques using fluorescence characteristics have been implemented in dental practice.

1.2. Fluorescence of teeth and restorative materials

1.2.1. Fluorescence phenomena

Photoluminescence is a common term that is used for the process of the emission of visible light from a molecule or substance that is in an electronically excited state, following absorption of radiant energy with a wavelength shorter than that of the visible light spectrum (400–700 nm) (1). The term photoluminescence includes fluorescence (which is a short-term process), phosphorescence (a long-term process), and bioluminescence.

In fluorescence, light is emitted within 10^{-8} seconds after excitation, and the emission process has a short lifetime, effectively ending when the exciting light

source is turned off. On the other hand, phosphorescence has a long lifetime, with the emission of light persisting after the removal of the exciting light source (2, 3).

An essential quality of fluorescence is the interlinked nature of excitation and emission. If the light source is pulsed, the fluorescence emissions will be in bursts, which correspond to those pulses. When fluorescence occurs, the fluorophores absorb the incident light, causing the electrons to move to higher orbits. However, this excited state is very short in duration, as it is quite unstable. The electrons decay quickly to lower levels, and light is given off. There is an associated thermal component to the relaxation of the molecule back to the ground state. The difference in the colour of the emitted fluorescence, compared to that of the excitation light, reflects the difference in energy, as the emitted fluorescence has a lower photon energy and a longer wavelength. For example, when porphyrin molecules are excited with violet light, they emit visible red light. If excited with visible red light, they emit near infrared light. This difference in wavelengths of excitation and emission is known as the Stokes shift, being named after Sir George Stokes (4-6).

In nature, many minerals and some biological molecules fluoresce, including certain amino acids and some molecules found in keratin, bacteria and fungi, and certain amino acids. This feature has been used widely to identify and distinguish such materials from other similar substances.

1.2.2. Ultraviolet light as a source of excitation

The fact that teeth fluoresce when illuminated with ultraviolet light has been known for over 100 years and was first reported by Stubel in 1911 who noted that the teeth of rabbits illuminated with UV light produced blue light (7). The ultraviolet region of the electromagnetic spectrum is divided into three bands: UVA (315–400 nm, also termed black light, long wave or near UV), UVB (280–315 nm, also termed middle UV), and UVC (200–280 nm, also termed short wave UV) (8). Having the longest wavelength and thus lowest photon energy of the three ultraviolet wavebands, UVA has limited effects on human mucosal tissues when only short-term exposures are employed.

UVA and the associated visible violet region is the important range in terms of diagnostic applications in dentistry, and its ability to make biological materials fluoresce is well known in both dentistry and medicine. As well as eliciting fluorescence emissions, light in the UVA range can also activate photo-initiators and thereby initiate chemical reactions that contribute to the photo-polymerisation of some dental materials, such as resin cements and composite restorative materials. The same type of light has been used in industrial settings to photo-polymerise adhesives, coatings, and resins.

In the literature, the fluorescent properties of some older types of composite resin materials have been determined using ultraviolet fluorescent tubes, which are

large, bulky, and fragile (9). Other possible light sources in the UVA region include high intensity discharge lamps, krypton ion lasers, helium cadmium lasers, nitrogen lasers, semiconductor diode lasers and light emitting diodes. Diode lasers and LEDs have high efficiency, are low in cost, and have a longer operational life than traditional UVA light sources. Two further advantages of both diode lasers and light emitting diodes are that they generate little heat and can be operated at low voltages (3.5–4.0 volts). Their small size and high resistance to shock and vibrations makes them ideally suited to use both in the field and in dental equipment, such as on a movable arm on a dental unit, or in a head-mounted light.

1.2.3. Luminescence of teeth

Luminescence of teeth includes both fluorescence and phosphorescence. Initial studies on human teeth focused on determining the fluorophores responsible for luminescence phenomena under UV light excitation (10). Wisotzy in 1963 reported phosphorescence in teeth, appearing as an afterglow in a totally darkened room (3). Assessments of the phosphorescence intensities and lifetime of mineralised tissues and other solid biological materials have reported that human dentine, bone and enamel give phosphorescence emissions with lifetimes as long as 31± 2 seconds. The phosphorescence intensity for enamel is around one third less than for bone or for dentine (11). This difference suggests that the organic matrix of dentine and bone is largely responsible for phosphorescence, particularly components of the polypeptide chains associated with the proteins of

the organic portion of the mineralised tissue (12). However, there is also evidence that the inorganic phase of both bone and dental enamel can generate phosphorescence emissions (10).

When the ambient light source is natural ultraviolet light, normal human teeth will emit light through autofluorescence. Likewise, if irradiated by an artificial source of UV light, they will also fluoresce (13). Regardless of the source of UV or violet light, the same endogenous fluorophores that are present in the dental hard tissues will be excited (14). The typical pattern is that teeth exposed to UVA light (350–400 nm) will emit light spontaneously in the visible blue spectrum (430–450 nm) (15, 16). While this pattern is true for both enamel and dentine, it has been known since the work of Benedict in 1928 that dentine fluoresces more strongly than enamel. Moreover, where mineral has been lost, such as in early carious lesions (white spot lesions), UVA-induced fluorescence is less (17).

Between the 1930s and 1950s, several studies investigated which substances in teeth could be responsible for their fluorescence characteristics (18). As already mentioned, sound human teeth will emit blue light under UVA excitation (11, 19, 20), but there can be occasional situations where longer wavelength green or yellow emissions are also seen (15). With excitation between 340 and 375 nm, the maximum emissions occur at around 450 nm, with the strongest emissions occurring with 370 nm excitation (21). In intact human teeth, a large component of fluorescence emissions is derived from the dentine, which fluoresces much

more intensely than does the enamel (17, 18). The fundamental pattern of fluorescence is not altered dramatically by the shade of the tooth (22, 23).

Which molecules are responsible for the autofluorescence of human teeth has not yet been resolved fully (16, 24). It is apparent from the early investigations into this, which used UV light for excitation, that the endogenous fluorophores were located primarily in the organic matrix (3, 11, 25, 26). Such a view is consistent with the finding of a loss of luminescence in teeth once they had been heated to a temperature of 600^0C, which would alter the organic matrix (27). There is evidence supporting the view that within the organic component of teeth, a major fluorophore is peptides that contain the amino acids tryptophan and tyrosine which are major fluorophores (28). It also appears likely that L-proline is involved. This amino acid is found in relatively large amounts in the collagen component of dentine (3). Fluorometric studies have shown that alkaline hydrolysates of dentine proteins show comparable fluorescence properties to these particular amino acids (11, 16, 29). While molecules such as tryptophan, tyrosine, pyrimidine, hydroxy pyridine and others have all been implicated as fluorophores, it appears that no one molecule in isolation is responsible, and that multiple molecules contribute to fluorescence emissions.

Investigations of the role of the inorganic phase of bone and enamel have shown that these can give rise to phosphorescence for extended periods after X-ray excitation at room temperature (10). This led to the development of the technique

of X-ray induced fluorescence (XRF). Furthermore, it has been shown using hard tissue sections of teeth that specific regions within teeth where more proteins are present fluoresce more strongly, such as enamel tufts, enamel lamellae, and the dentino-enamel junction. Likewise, within the dentine itself, intense fluorescence occurs along the dentinal tubules (19). The fluorescence spectra of dentine are similar to those of cross-linked collagen (30). This explanation is also consistent with observed regional variations within a single tooth. A study of such variations within teeth revealed that fluorescence at the gingival one third, where enamel is thinner and there is more dentine, was greater than fluorescence at the incisal third, which is mostly enamel (21).

Further evidence of the role of organic fluorophores comes from the work of Bosch and Spitzer, who evaluated the excitation and emission spectra of human enamel, bovine enamel, and organic material that had been separated from enamel. They concluded that the organic component was the major contributor to the total luminescence of enamel. This was based on their finding that there was no difference in the emission peak of human enamel and of organic material from enamel (31).

The fluorescence properties of human teeth may be influenced by variables such as the tooth type, the age of the person, and the environmental conditions in which extracted teeth are stored (such as moisture and temperature). No studies have found any significant differences between corresponding teeth in the

mandibular and maxillary arches in the same person. However, human dentine shows stronger fluorescence emissions with increasing age, as the dentine undergoes sclerosis (32). As the dentine becomes thicker and the dental pulp volume decreases, there is a greater overall bulk of dentine that can fluoresce. These same changes influence the chroma of dentine and reduce its opacity (33). Since optical properties of teeth are inherently driven by their physical structure of the teeth, any such changes could alter not just the fluorescence but also the tooth shade (34). A final factor to be considered is that as a person ages, the enamel may also become thinner because of attrition and other forms of non-carious tooth structure loss. This would allow the optical features of the underlying dentine to become more prominent.

The literature on the topic of the influence of ageing on the fluorescence intensity of teeth has been controversial. Initially, it was believed that an increase in fluorescence intensity occurred that was directly proportional to chronological age, and it was suggested that dentine fluorescence could even be used for the purposes of age estimation in forensic examinations (24, 34). However, more recent studies have demonstrated that the fluorescence of teeth increases in early adulthood and reaches its peak at around 26.5 years of age, and then declines steadily after this point (34). The changes in dentine could reflect degradation of organic components. Studies using experimental accelerated ageing models have noted that while the fluorescence of human dentine samples decreases over time, the fluorescence of enamel does not change. However, the

results from such models cannot be extrapolated to physiological ageing, which is far more complex in terms of changes that occur to potential fluorophores over time (35).

1.2.4. Fluorescence of tooth-coloured restorative materials

A successful aesthetic restoration in a tooth not only duplicates the appearance of the adjacent natural tooth structure, but also is minimally invasive, in order to conserve tooth structure (36). In order to mimic the appearance of natural tooth structure, an ideal aesthetic dental restorative material needs to replicate the optical traits of natural teeth, such as opalescence and translucency (37).

Hypothetically, an ideal restorative material would also replicate the fluorescence properties of natural tooth structure, both under natural ambient light, as well as under conditions where there is artificial light. When choosing a restorative material, wherever possible shade matching is done under natural light or using daylight-corrected light sources (38–40). Even if a restoration appears well matched under daylight, there could still be issues with how the restoration appears under blue, violet or UVA light (41).

It was reported in 1977 by Panzeri and colleagues that the major constituents of the composite resin materials used at that time did not fluoresce under UV light (42). Without any fluorescence traits, they appeared dull rather than life-like, particularly under conditions such as in night clubs where ambient UVA or violet

light was being used (32, 43, 44). The solution to this was to add in exogenous fluorophores (45–47). However, the total fluorescence emission is not simply the sum of the fluorescence emissions of each fluorophore that is added (47).

1.2.4.1. Fluorescence of resin-based restorative materials

Reis and co-workers in 2007 assessed the fluorescence properties of 10 commercial resin-based composites and compared these to natural tooth structure. They reported a significant difference between the fluorescence of the composite materials and tooth structure (48). While early composite resin materials used in dentistry did not fluoresce under UV light, other materials used in anterior restorations in the 1970s did show some fluorescence. As was later done with resin-based composites, it was suggested that adding in exogenous fluorophores would improve the quality of such fluorescence emissions (42).

The optical properties of resin-based composites are influenced by a range of parameters including the size, shape and composition of filler particles, and the chemical constituents that are present in the resin matrix phase. The colour of a restoration is the result of a complex interaction of the incident light and the matrix and filler particles, and it is influenced by whether the material is translucent or opaque. Manufacturers add in various pigments to obtain the desired shade, and these pigments can influence the fluorescence properties of commercially available composite resins (49). There have even been attempts to improve on the fluorescence appearance of resin-based materials by adding in

quantum dots, so that the materials fluoresce in a way that more closely resembles natural tooth structure (50). There can be considerable variation in fluorescence amongst resin-based materials from the same manufacturer, which can be explained by physical and chemical differences between those materials (51).

A range of environmental factors can alter the fluorescence properties of a resin-based restoration. These include exposure to oxidants (such as in bleaching treatments) or to coloured foods, degradation over time, and exposure to higher temperatures (34, 38, 52–54). External staining from coloured foods or from highly charged mouth-rinses or tobacco stains could interfere with light entering and light leaving a restoration. Higher temperatures could cause breakdown of the resin polymer matrix.

1.2.4.2. Fluorescence of Ceramic/Porcelain restorative materials

Compared to resin-based restorative materials, the fluorescence behaviour of dental ceramics has not been investigated to the same extent. Baran and colleagues experimented with adding rare earth element oxides into dental porcelain to act as fluorophores and improve fluorescence emissions under UV illumination (46). This was an improvement on previous work where uranium or cerium compounds were used for the same purpose (55).

Eventually, rare earth element oxides (ytterbium, europium, cerium and terbium) superseded the older approach which relied on uranium compounds, and this

was desirable as it avoided issues with radioactivity. The use of uranium in dental ceramics was eventually banned for that very reason. However, when using rare earth element oxides as fluorophores, the intensity of fluorescence emissions are typically less than those of natural teeth (46, 56, 57). This explains why when the fluorescence emissions of dental ceramics are compared to those of natural tooth structure, the typical finding is that the ceramics will fluoresce less, by around half (45, 47, 58, 59), despite the fact the exogenous fluorophores have been added (60).

Studies that examined the effect of shade have revealed that variations in the shades of dental porcelain affect their fluorescence. Those ceramics with greater colour saturation have less intense fluorescence emissions (58, 59).

A particular issue with ceramics is the temperature stability of the fluorophore. Ecker and co-workers, who investigated the effect of repeated firing on the fluorescence behaviour of dental porcelains, found that repeated firing reduced the fluorescence of porcelains. The effect was similar to the influence of high temperature on the fluorescence of resin-based composites (45).

1.2.5. Techniques for analysis and quantification of fluorescence of tooth-coloured restorative materials

In the past, a range of devices and methods have been employed to explore and quantify the fluorescence emissions of tooth-coloured restorative materials.

Devices that have been employed include fluorimeters (35, 41, 45–47, 53, 56, 61–64), spectrophotometers (42, 50, 54, 58), and photographic cameras (48, 51, 65–67) Fluorimeters are useful for analysing the wavelength distribution of fluorescence emissions. They employ filters to remove the reflected incident light (2). Fluorimeters were used by Benedict (17) and by Forziati and colleagues (68). Spectrofluorometers can provide detailed data sets on fluorescence patterns, by generating a total luminescence contour map. They have been used widely for analysing the fluorescence properties of materials and of individual fluophores, particularly when the latter have been dissolved into different solvents (42, 50, 54, 58).

Image analysis has also been used to analyse and quantify the fluorescence properties of tooth-coloured restorative materials. Fluorescence images are taken using a camera that is fitted with appropriate filters (48, 51, 65–67). These long pass filters remove the reflected light from the excitation light source, allowing the longer wavelength fluorescence emissions to be recorded. The colour information in the digital images is then evaluated in terms of colour channel values for red, green, blue and luminosity, which together make up each pixel.

1.2.6. Summary

Fluorescence properties of restorative materials should ideally be matched to natural tooth structure to better simulate the optical traits of tooth structure. The

closer the match, the more natural the restoration will appear, but the harder it will be to identify. Since the major constituents of tooth-coloured restorative materials do not fluoresce, in most materials the manufacturer would have added in fluorophores, such as rare earth oxides. Different patterns of fluorescence could identify particular types or brands of material.

It is essential that dental professionals can differentiate between restorations and tooth structure, for accurate charting of previous dental treatment, as well as for forensic dental identification. As restorative materials and methods for placing them have improved over time to give better aesthetics, methods to identify the presence of tooth-coloured restorations in teeth become ever more important. Thus, it would be desirable to have a simple method for the identification of tooth-coloured restorative materials, particularly one that is based on the difference in their fluorescence compared to that of tooth structure, as this would be a simple and easy method. It would be without significant risks, since non-ionising radiation would be used.

Since any method for fluorescence identification needs to be simple to use at the point of use, that is, at the chairside or in the field, complex devices such as spectrophotometers would be unsuitable, but a camera would be suitable. To illuminate the teeth with light in the UVA to visible violet range, LEDs seem ideal, because of their high conversion efficiency of electrical energy into light, the high stability of their light output over time, and their low power requirements. Light

sources in the UVA range (e.g. 365 and 380 nm) have been found useful in previous studies (69, 70).

Given the potential use of fluorescence to aid in detecting restorative materials, it will be essential to determine an appropriate wavelength for excitation and suitable filter combinations to improve the signal to noise ratio and provide the greatest differentiation between the range of contemporary restorative materials that could be encountered compared to adjacent tooth structure. The range of materials to be examined in this thesis will include contemporary tooth-coloured restorative materials.

The final point in this summary is that the fluorescence properties of a particular tooth-coloured restorative material are likely to vary according to the shade of the material, as different tints will have been used to create the final colour of the material. This aspect has not been examined in any previous studies.

1.3. Forensic considerations

1.3.1. Significance of dental features in human identification in forensic sciences

One of the major tasks within forensic odontology is human identification. Positive identification of deceased individuals is important for several reasons, extending beyond closure for their immediate family members, to other considerations which are humanitarian, ethical, psychosocial and legal (71). The

identification of a deceased person might be required when the body is decomposed, deformed, or mutilated beyond recognition.

When the soft tissues of the body are decomposed totally, personal identification methods such as physical marks, photographs, iris characteristics, retinal scans, fingerprints and DNA might not be useful (72). Identification by comparison of that individual's dental features can play an important role. Teeth are stable under situations in which the soft tissues undergo decomposition, because of their composition and their position within the body (73). Posterior teeth are well protected from this standpoint. However, dental pattern identification may not be possible in cases where the victim was involved in an explosion or was incinerated to the point where only a few fragments of teeth and bone remain (74). The INTERPOL Disaster Victim Identification (DVI) Guide states that principal methods of human identification are fingerprints, DNA and dental comparisons (75). Human identification by comparison of dental records is often used in conjunction with other methods of identification.

The details of a person's dentition, such as the presence or absence of particular teeth, restored or unrestored surfaces on teeth, or missing and decayed surfaces of teeth on the dental records can be exclusive to that person, due to the large number of possible combinations that could theoretically exist (76). In conjunction with the dental variables already mentioned, additional information on the brand or type of restorative materials that are present could add additional points of

data and further improve the level of certainty and precision, as this would give additional unique features (77).

Methods that have been employed in specific brand identification of restorative materials include light-based fluorescence methods, X-ray fluorescence (XRF) methods, and scanning electron microscopy (SEM) combined with energy dispersive X-ray spectroscopy (EDS) (71).

1.3.2. Methods employed to identify tooth-coloured restorations in forensic odontology

Various methods have been advocated to detect the presence of composite resin restorations, including radiographs, dyes, light-induced fluorescence (78), XRF, and SEM with EDS. Several studies have assessed the role of radiographs in ante-mortem and post-mortem comparisons (79–87). A key limitation is that not all tooth-coloured restorations are radiopaque. Sainio and co-workers presented three cases to demonstrate the valuable role that radiographs could play in ante-mortem and post-mortem dental comparisons, when used for identification of unknown persons (79). Likewise, a study that analysed simulated post-mortem and ante-mortem bitewing radiographs and used subtraction radiography showed that bitewing radiographs could facilitate the identification of a single individual within a group of individuals by following certain fixed criteria (80, 84).

A positive identification could occur if the post-mortem X-rays were an exact replica of the ante-mortem radiographs, particularly in terms of the outline shapes of the composite resin restorations (85). Arguably having a single restoration with a unique morphology could give an important clue to identity if it gave a perfect match in both ante-mortem and post-mortem radiographs (84).

Radiographs may also provide some information on the types of restorative material that may be present in a tooth. An investigation of the ability to discriminate between different restorative materials based on their radiopacity showed that of the three radiographic systems used (conventional film, phosphor plate and CCD-based sensors), the conventional film-based method performed best for differentiating between restorative materials (81).

Despite their advantages, radiographs may have limited usefulness for determining the extent of a tooth-coloured restoration. A study by Chesne and colleagues found that radiographic examination alone could not confirm the location of the restorations accurately (82). Jackowski and co-workers evaluated the radiopacity of different restorative materials from computed tomography (CT) images and suggested a method that allows for 3D colour encoded visualisation to improve differentiation between types of restorations (83). Likewise, Sakuma and colleagues attempted to differentiate between normal dental enamel and composite resin restorations, based on their differences in Hounsfield units on multi-detector CT images. They found this very difficult to do, since their

Hounsfield values were identical. Nevertheless, they still were successful in visualising the composite resins from CT reconstructed images (86).

A systematic review of the performance of radiological methods advocated for human identification in forensic dentistry, which included intra-oral radiography and CT imaging, argued that appropriate knowledge of the strengths and weaknesses of different methods was essential for choosing the best method in any given situation (88).

Bush and colleagues have initiated two databases for dental resins. One uses SEM with EDS. This laboratory method is a well-established technique for compositional analysis of materials. This second uses XRF, a method that can be taken into the field. XRF is able to discriminate between resin materials, teeth, bone and other substances (89). In fact, it can help locate and identify composite resin restorations that are present and provide information to help determine their brand or type. Such information could aid victim identification. XRF is normally used with teeth, but it can also be applied to cremains of resin restorations (90).

A novel technique for the post-mortem detection of tooth-coloured dental restorations was proposed by Benthaus and colleagues in 1998. In this method, teeth were etched with 37% phosphoric acid, then stained with dyes, making the restoration margins easier to see, and improving recognition of the presence and

extent of tooth-coloured restorations (91). Such simple clinical methods may be useful for the field.

Post-mortem detection of tooth-coloured dental restorations using UV light was advocated by Clark and Ruddick in 1985 (92). The same approach was also used by Tani and co-workers, who investigated the ability to differentiate composite resin restorations from adjacent healthy tooth structure using fluorescence in the context of large-scale dental health inspections (43). The use of quantitative light-induced fluorescence (QLF) to identify composite restorations in forensic inspections was also advocated by Pretty and colleagues in 2002. Using this method, they detected more filled surfaces than with conventional clinical examination methods (78).

Hermanson and colleagues in 2008 assessed the fluorescence properties of 15 commercially available resin-based restorative materials, using UV LED light sources. They reported that materials belonging to different brands fluoresced at varying wavelengths and intensities, using 365 and 380 nm for illumination (70). A similar approach was employed by Guzy and Clayton, who used a UV LED flashlight as a light source for identifying composite resin restorations (9).

1.3.3. Alternate optical methods used to identify tooth-coloured restorations

The traditional method of detecting tooth-coloured restorations in dental practice is visual inspection under white light, supplemented in some cases with

radiographs. Visual inspection has poor sensitivity. Clinicians may struggle to distinguish the margins of the restoration from the surrounding normal tooth structure (43, 69, 82, 87).

Optical methods that are based on the interaction of light with hard tissues may aid in distinguishing restorations from natural tooth structure. Optical methods in current clinical use include light-induced fluorescence, fibre-optic trans-illumination (FOTI) and digital imaging fibre-optic trans-illumination (DiFOTI) using near infrared light, as in the KaVo DIAGNOcam™.

With both FOTI and DiFOTI, patterns of the reflection, scattering, transmission, and absorption of light are used to discriminate between the internal structural features of teeth (93). FOTI typically uses broad spectrum high intensity white light, which gives strong scattering effects. In contrast, DiFOTI systems such as the DIAGNOcam™ use coherent near-infrared light from a diode laser. Compared with visible light, near infrared light penetrates better through sound tooth structure, and gives less scatter (94–96). While DiFOTI has been used primarily to detect lesions of dental caries on approximal enamel surfaces, the method may also be applied for detection of tooth-coloured restorations, because these transmit light differently to natural tooth structure.

1.3.4. Summary

The identification of a deceased person by comparison of ante-mortem and post-mortem dental records plays a pivotal role when dental structures are the only source of information that remains (77, 97, 98). Since teeth are highly mineralised tissues, they may be relatively stable under conditions where the soft tissues might decompose (76). Information about restorations placed and previously present in teeth are a key part of dental records. Along with dental features such as missing and decayed surfaces, and the presence or absence of teeth, the presence of a restoration (its location and extent) is vital information. Recording the types and brand type of restorative materials used may improve the level of certainty and precision in human identification (77).

Employing radiographs for ante-mortem and post-mortem is a well-established method, however not all resin-based materials are radiopaque. XRF and SEM with EDS can identify the brand of material based on variations in elemental composition. SEM with EDS is a laboratory-based method and is not suited to clinical or field settings. In situations such as mass fatalities due to natural calamities or man-made disasters, methods which are quick, easy and reliable in identifying restorations would be very useful. The XRF method of Bush and colleagues is one such method, as it is portable and can be used in the field (90).

An alternative method which is promising is using UVA or visible violet light (405 nm) from an LED light source. This has potential for differentiating between

materials and natural tooth structure. However, the reliability of fluorescence-aided identification of restorations (FAIR) needs to be determined in a formal manner. This aspect will be addressed in this thesis.

Both fibre-optic trans-illumination (FOTI) and digital imaging fibre-optic trans-illumination (DiFOTI), as in the DIAGNOcam™, are already employed in clinical situations for caries diagnosis. The difference in the light transmission of healthy tooth structure and carious tooth structure is the basis for caries diagnosis. The same principle could theoretically be applied for detecting restorations. This thesis will compare FAIR and DiFOTI to conventional methods, to assess their accuracy for detecting restorations.

1.4. Resin-based restorative materials and their selective removal

1.4.1. Resin-based composites and their role in restorative dentistry

Since their introduction in 1968, resin-based composites have progressively become more widely used, and nowadays would be regarded as the material of choice for conservative direct anterior aesthetic restorations (99). Sequential improvements in the physical properties of resin-based restorative materials over time make RBC the preferred material for large posterior restorations as well (100, 101). Several studies have found that there is an increasing trend for dentists and patients to choose RBC rather than silver amalgam for posterior restorations (102).

Due to occlusal wear and other processes during normal function, all RBC restorations will experience signs of degradation after extended periods of service in the mouth. Modern RBC have low annual failure rates of between 1% and 3% (101, 103). Regardless of their type, failing restorations are a common situation that is seen in dental practice, and clinicians spend a large amount of time replacing them. This accounts for 57% to 60% of all operative work (104–106). There are many reasons why a restoration may need to be replaced, including secondary caries, marginal discolouration, marginal fracture, restoration fracture, tooth fracture, and defective anatomical form (106–110).

During the process of removing and replacing a restoration, it is likely that a significant amount of natural tooth structure may be lost. This is particularly the case when the dentist cannot determine, as they are working at the cavity margin, what is tooth and what is restorative material. Unnecessary removal of healthy tooth structure weakens the remaining tooth and contributes to a ‘death spiral’ of breakdown of the tooth. Because RBC restorations can be very difficult to differentiate from tooth surface when the shade match is accurate (111–113), methods that facilitate selective removal of composite resin materials from teeth would be very useful.

1.4.2. Methods employed for guided removal of composite resin restorations

Lasers are used in many dental procedures, including soft tissue surgery, decontamination, cavity preparation, caries removal and caries prevention (114–

120). The use of infrared lasers for removing dental hard tissues is based on a combined photomechanical and photothermal ablation mechanism (121). Laser systems commonly used for hard tissue cavity preparation are the Er,Cr:YSGG laser (λ = 2.780 μm), the Er:YAG laser (λ = 2.940 μm), and the carbon dioxide laser (λ= 9.3 or 10.6 μm) (122). All these lasers have been used for selective removal of composite materials from tooth structure, based on the much more rapid ablation of the resin matrix of RBC materials than human enamel. Recent studies have demonstrated that the 9.3 μm carbon dioxide laser is capable of achieving rapid and selective removal of resin-based composites, with less than 20 μm loss of enamel (126, 127). Nevertheless, water spray is essential during lasing to prevent thermal damage to adjacent tooth structure or to the dental pulp (123–125).

Hard tissue laser systems are ‘line of sight’, and so the beam must be angled into undercuts to remove composite material from undercut regions of a cavity. Current hard tissue laser systems do not ablate ceramic materials, but they can weaken and fragment the adhesives and cements which bind them to teeth. All hard tissue lasers are expensive and require specific operator training for their safe use (128, 129). These issues have limited their uptake in clinical practice.

1.4.3. Summary

Because a well-placed tooth-coloured restoration is difficult to detect, there is an inherent risk that some sound tooth structure may be removed inadvertently when the restoration needs to be replaced. Thus, methods to help identify the location and extent of tooth-coloured restorations would make operative dentistry more efficient and safer and reduce damage to teeth. Such methods need to be simple to use, quick, inexpensive, cause no damage to the teeth, and facilitate the removal of a range of types of material.

Since many restorative materials do not have identical fluorescence properties when compared to healthy tooth structure when illuminated with UVA or violet light (405 nm), a technique based on fluorescence-aided identification of restorations (FAIR) could be a valuable aid in guiding the removal of tooth-coloured restorative materials (49, 130). The FAIR technique is based on the phenomenon of illuminant metameric failure, whereby two materials match when viewed under one type or wavelength of light, but not under other type or wavelength of light (37, 131). Amongst most tooth-coloured restorative materials, restorations may match the colour of the adjacent natural tooth surface under daylight or when viewed by a daylight-corrected light source (such as the operating light of a dental unit), but may not match under a violet light source which induces fluorescence (132, 133). This thesis includes studies which explore whether the FAIR technique is of value for the guided selective removal of resin-based restorations, and whether it is able to minimise the loss of healthy tooth structure, when compared to conventional methods.

1.5. Effect of heat on tooth-coloured restorative materials

1.5.1. Incinerated remains

As previously mentioned, in cases of charred and incinerated remains, the use of fingerprints and DNA for the identification of individuals may be problematic or impossible (134, 135). In such cases, it is essential to be able to differentiate charred restorative materials from charred tooth structure, for victim identification using dental records. The highly mineralised composition of dental enamel makes it able to withstand relatively high temperatures before degrading.

Certain dental materials such as the ceramics used in tooth-coloured restorations, and the titanium alloys used in dental implants, are manufactured or fabricated using high temperature production methods, and thus these materials are inherently quite heat resistant. In contrast, restorations based on composite resin materials degrade at high temperatures, in the same manner as other similar organic polymers and plastics. As discussed earlier, because of their location in the body, the posterior teeth are protected somewhat by the adjacent hard and soft tissues, such as the cheeks, tongue, and alveolar bone, and these provide some measure of heat protection for the crowns and roots of teeth during fires (135, 136).

The specific effects of heat on teeth and on restorative materials within teeth are influenced by a range of factors. These include whether or not there is direct exposure to flames, the maximum temperatures achieved, the duration of the

heating event, the presence of any materials (in addition to the soft tissues) interposed between the teeth and the fire, and temperature alterations caused by any substances used to quench the fire (137, 138).

The bodies of deceased persons may be subjected to different temperatures depending on the source of heat and the nature of the fire event. House fires may reach temperatures of around 700 to 900 ◦C (136, 139, 140). Cremation occurs at temperatures between 871 and 982 ◦C (139, 140). The temperatures in burning motor vehicles may range between 800 and 1100 ◦C, especially when petrol is present as an accelerant, while in aviation accidents the associated fires can reach temperatures up to 1538 ◦C (141, 142). In bushfires and fire storms, temperatures may reach as much as 2000 ◦C (139, 143, 144).

The duration and peak intensity of a fire is determined by the presence of combustible material and the availability of oxygen or another oxidant (145). In some cases, fires can smoulder for days or even weeks, as reported in Victorian bushfires, and in the World Trade Centre after the 9/11 attacks (141, 143).

1.5.2. Effects of heat on teeth

In cases of long duration fires with temperatures above 700 ◦C, dental analysis may be the only reliable method of identification of victims, as other materials that would normally be used as primary means of identification (such as

fingerprints and DNA) or as a secondary means of identification (for example, clothing and personal effects) have been destroyed (143, 145).

When subjected to extensive heat, teeth lose hydration, and may fall from their sockets as the periodontal ligament becomes degraded (146, 147). In fire incidents, the posterior teeth are affected the least because they are more protected due to their location in the oral cavity (146). Posterior teeth with multiple roots tend to remain in place within their alveolar bone sockets, while single-rooted teeth are most often found to be displaced and isolated (147).

Several studies have assessed the response of teeth and restorative materials to elevated temperatures and tracked the corresponding macroscopic and microscopic changes. Fire-induced injuries to the teeth and jaws were classified in 1995 by Anderson and colleagues into six grades, as follows: (0) no injury, (1) injury to anterior teeth, (2) injury to anterior and posterior teeth (unilaterally), (3) injury to anterior and posterior teeth (bilaterally), (4) fragments of jaw bone including teeth and/or roots, and (5) no dental remains.

Macroscopic changes to teeth from elevated temperatures include fragmentation and colour changes. Enamel will fragment when teeth are heated slowly at 800 ◦C (142). The separation of the coronal portion of the tooth from the root, a phenomenon referred to as ‘popping off’, may occur at around 450 ◦C when teeth are heated for 30 to 60 minutes (148, 149). This effect occurs because of

differential thermal coefficients of expansion of enamel and dentine, and the pressure effects generated as free water in the dentine is converted into steam (150).

As a tooth is heated, colour changes occur as water is lost, and protein and other organic structures in enamel and dentine degrade, particularly those chemical bonds which hold strands within helical collagen molecules together (145, 151). When unrestored teeth are exposed to elevated temperatures upward of 150 ◦C, their colour becomes a lighter yellow, then changes to bluish white as water is lost. The teeth then become progressively brown at 300 ◦C, dark grey-black at 500 ◦C, and finally almost white at 1000 ◦C and above (145, 151, 152).

1.5.3. Effect of heat on dental materials

At extreme temperatures of up to 1600 ◦C, dental remains may be the only remnants of a person which survive and that are suitable for analysis (153, 154). Dental implants that survive could provide additional information to support positive victim identification (155–157). Metallic dental implants, metallic dental prostheses and metallic and ceramic dental restorations can survive temperatures above 1000 ◦C (134, 141, 142, 158, 159). Orthodontic brackets and wires, depending on their composition, can survive temperatures from 700 to 980 ◦C (160). Silver points used as root canal fillings can also survive prolonged heat exposure, and these may also aid in the identification process (143, 160). Different tooth-coloured restorative materials exhibit distinctive features upon

exposure to heat (140). Macroscopic changes observed when certain materials are subjected to heat are summarised in Table 1.1.

The colour changes which occur in dental tissues, bone and tooth-coloured restorative materials after exposure to heat could make distinguishing restorations by visual inspection difficult, especially if only fragments of teeth are found (74, 161). As discussed previously, in forensic odontology, several methods have been employed to aid the detection of tooth-coloured restorative materials, including SEM with EDS, and XRF. These can help identify the brand of dental restorative material used (89, 90).

Use of UVA or violet light-induced fluorescence may be of limited use for identifying tooth-coloured composite resin filling materials in incineration situations, as the organic components of the matrix (and the accompanying fluorescence properties) would be expected to be lost at temperatures above 300 ◦C, while the inorganic filler components may withstand temperatures well over 1090 ◦C (162). This aspect will be explored in this thesis.

Table 1. 1. Macroscopic changes observed in restorative materials when subjected to different temperatures

Sl. no	Restorative material	Macroscopic changes observed	Temperature in ◦C	Reference
1.	GIC Fuji II	Distinctive red hue when decomposed	200	*Robinson et al. 1998*
		Appeared brown	1000	
2.	Composite resin materials	Became vitrified masses		
	Herculite®XRV™	Appeared as ash white discs	1000	*Robinson et al. 1998*
	Occlusion™	A wide variety of shades: ash white,		
	Silux Plus™	brown, gray or black	400–600	*Moreno S et al. 2009*
	Geristore™			*Robinson et al. 1998*
3.	Silver amalgam	Loss of glaze, expansion, globule formation and cracking	400	*Meralti G et al. 2004*
			1100	*Gunthe and Schdmit*
				Patidar K A et al. 2010
4.	Ni Cr and metal ceramic crowns	Loss of glaze and slight loss of morphology	1100	*Patidar K A et al. 2010*
5.	Porcelain alloys	No changes	1100	*Norlander AL 1995*

1.5.4. Summary

In a fire, the effects on tooth-coloured restorative materials are expected to differ between the various classes of material. Glass ionomer cements (GIC) contain water in their matrix and so should be sensitive to temperatures above the boiling point of water. The resin matrix of composite resin materials will become volatile at higher temperatures, while pure ceramics, and hybrids of ceramics and resins should be more heat resistant.

When considered from the standpoint of fluorescence emissions, in natural healthy tooth structure, the amino acids and other organic components that generate most of the blue-green fluorescence emissions under UVA or visible violet (405 nm wavelength) light will degrade at high temperatures. As a result, fluorescence should decline as the temperature rises into the higher ranges.

The inorganic filler components of tooth-coloured restorative materials are glasses or glass-like in nature and provide much of the high compressive strength and low wear attributes of modern dental materials. It is hypothesised that the glass fillers in GIC and in RBC and the pure ceramics would begin to melt and fuse when heated to temperatures above 1000 ◦C. Any inorganic fluorophores, such as those based on rare earth element oxides, should retain some fluorescence properties after exposure to a temperature of 900 ◦C.

1.6. Research aims and hypothesis for the thesis

1.6.1. Chapter 3 (Study1)

As has been explained, tooth-coloured restorative materials are difficult to differentiate from the tooth structure if the shades are well matched. However, restorative materials cannot replicate the fluorescence properties of natural tooth structure at all wavelengths of light. The aims of study 1 in Chapter 3 of this thesis were: (1) to compare the fluorescence properties of tooth-coloured restorative materials when illuminated with different wavelengths of visible light from LED arrays; (2) to assess the effect of moisture on the fluorescence properties; and (3) to determine if hybrid restorative materials have unique fluorescence emissions.

Study 1 tested two hypotheses: (1) that the greatest differentiation between different materials, and between the materials and the adjacent tooth structure, will occur with short wavelength (UV-A/violet) light; and (2) that hybrid restorative materials would exhibit a recognisably unique emission spectrum, that would be different from that of all other classes of tooth-coloured materials.

1.6.2. Chapter 4 (Study 2)

The fluorescence properties of a particular tooth-coloured restorative material may well vary according to the shade of the material, since different colouring agents and fluorophores will have been used to create the final colour of the material. The aim of the study 2 in Chapter 4 in this thesis was to investigate the

fluorescence properties of different shades of contemporary tooth-coloured restorative materials, under excitation from 405 nm light. The hypothesis was that fluorescence properties would vary with shade.

1.6.3. Chapter 5 (Study 3)

From the results of study 1, it was apparent that the fluorescence properties of tooth-coloured restorative materials vary significantly when exposed to 405 nm light, and that the materials differed from tooth structure. Therefore, it was believed that a technique for the fluorescence-aided identification of restorations (FAIR) should be practicable. Moreover, based on its method of operation, the DiFOTI method (using the KaVo DIAGNOcam™) should be useful for detecting tooth-coloured restorations. Thus, the aim of study 3, in Chapter 5 of this thesis, was to compare the diagnostic sensitivity and specificity of FAIR and DiFOTI methods with conventional visual and tactile examination, for the detection of tooth-coloured restorations.

Study 3 tested two hypotheses: (1) that differences in light transmission through natural tooth structure versus various tooth-coloured restorations would aid in their identification using DiFOTI; and (2) that FAIR using 405 nm violet light would enhance the identification of tooth-coloured restorative materials, to give more reliable identification than either conventional examination, or DiFOTI.

1.6.4. Chapter 6 (Study 4)

From the results obtained in studies 1 and 3, it was predicted that FAIR should be able to facilitate the selective removal of tooth-coloured restorations, since under 405 nm illumination the restorative materials can be differentiated from the adjacent tooth structure. Based on this logic, the aim of study 4 (Chapter 6) of this thesis was to compare FAIR with conventional white light illumination for the removal of tooth-coloured RBC restorations. It was hypothesised (1) that FAIR would reduce the extent of loss of tooth structure, (2) that FAIR would reduce the amount of residual material at the end of cavity preparation, and (3) that FAIR would allow the procedure to be completed in a shorter time.

1.6.5. Chapter 7 (Study 5)

The highly mineralised composition of enamel makes it able to withstand relatively high temperatures before undergoing degradation. Likewise, the inorganic filler components of tooth-coloured restorative materials have high melting points as they are generally glass or ceramic in nature. Based on these concepts, the aim of study 5 (Chapter 7) in this thesis was to assess the effect of various high temperatures on the fluorescence properties of contemporary tooth-coloured restorative materials and on natural tooth structure.

Study 5 tested two hypotheses: (1) that the ceramic and glass components would show minimal degradation until heated to temperatures above 1000 ◦C; and (2) that tooth-coloured restorative materials which contain inorganic fluorophores

would retain some of their fluorescence properties after exposure to a temperature of 900 °C.

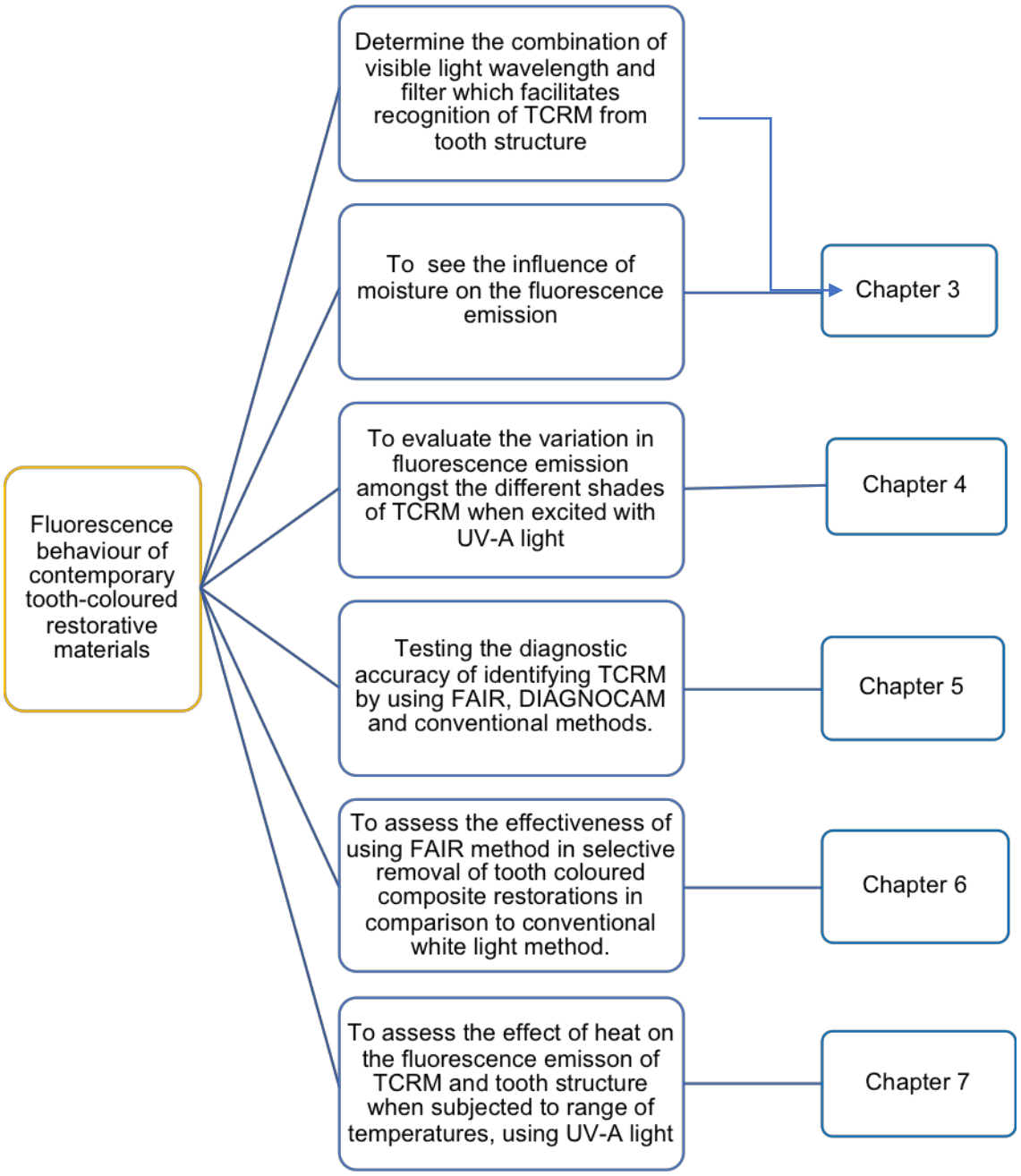

Figure 1. 1. Flow of experiments and corresponding chapters

Chapter 2 General Methods

2.1. Ethics approval

The collection of human extracted teeth and all the experiments included in this thesis were conducted with ethics approval from the Human Research Ethics Committee of Central Queensland University following guidelines established by the National Health and Medical Research Council of Australia (approval number H15/03-035). Clinicians who participated as examiners in the study were informed about the procedures involved in their respective studies, and they all participated voluntarily.

2.2. Collection of extracted teeth

Human permanent teeth were collected from dental extraction procedures (where the reason for extraction included periodontal conditions, orthodontic reasons, apical periodontitis and impacted teeth) in a general dentistry clinic. Immediately following extraction, teeth were rinsed thoroughly under tap water to remove any blood. All surface debris and all visible remnants of soft tissue were removed by mechanical debridement using hand scalers. Teeth were stored in an aqueous solution of 1% thymol (Sigma Aldrich, St Louis MO, USA) and then sterilised by 20 kGy gamma radiation.

2.3. Procedures and protocols

2.3.1. Cavity preparation in extracted teeth

The teeth used included incisors, canines, premolars and molars. While some variations apply for each study conducted in this thesis which involved cavity preparations, in all cases water-cooled diamond burs in a high-speed dental handpiece (model S-Max M600L, NSK, Kanuma, Japan) were used to prepare cavities. In teeth devoid of any pathology, GV Black's class I or class II cavities were prepared in line with the anatomy of the individual teeth. The dimension of cavities varied with the type of teeth. In teeth with previously existing carious lesions (in study 3) all affected and infected carious dentine was excavated, and the cavity margins redefined as per the shape of the original carious lesion.

2.3.2. Direct and indirect restorations

An array of direct and indirect tooth-coloured restorative materials were used to restore the cavities prepared for the studies conducted in this thesis. Based on the material type and brand types, the protocol used differed, in line with the manufacturer's instructions.

For resin-based composite restorations the following standard clinical protocol was used. The cavities were etched with 34% phosphoric acid gel for 20 seconds, to remove the smear layer from the prepared cavities and to promote mechanical adhesion. The etchant was removed by rinsing the cavities with water from a triple syringe for 10 seconds. The cavities were then air dried to remove excess moisture using compressed air. A dentine bonding agent (Scotchbond™ Universal Etchant, 3M-Espe, St Paul, MN) was then applied with

a microbrush, and left for 20 seconds, then thinned by compressed air for 0.5 seconds, and finally photopolymerised using an LED curing light (Mini-LED, Acteon Satelec, Merignac, France) in pulsed mode at a power density of 1250 mW/cm2. The RBC restorative material was placed using an oblique multiple layering technique. Each increment was photopolymerised for 20 seconds. Once completed, the restorations were polished using urethane-coated paper impregnated with aluminium oxide particles and rubber polishers. The restored teeth were then stored in distilled water until used further.

For glass ionomer cement restorations, the prepared cavities were treated with 20% polyacrylic acid solution for 10 seconds. This was rinsed away with water from a triple syringe for 10 seconds, and the cavity dried with compressed air. The GIC was mixed in a mechanical mixer and syringed into the cavity. Resin-modified GIC materials were photopolymerised for 20 seconds. Auto-curing GIC materials were allowed to set over 4 minutes, in line with the manufacturer's instructions. Once fully set, a layer of unfilled resin glaze was applied and photopolymerised for 20 seconds.

For teeth that received indirect restorations, the prepared enamel was etched with 30% phosphoric acid gel for 30 seconds. The etchant was removed by rinsing the cavities with water from a triple syringe for 10 seconds. The cavities were then air dried to remove excess moisture using compressed air. The bonding agent was applied with a micro brush, and left for 20 seconds, then

thinned by compressed air for 0.5 seconds, and finally photopolymerised using an LED curing light for 10 seconds. A second coat of bonding agent was then applied and photopolymerised in the same manner. The inner surface of all ceramic inlays, full crown restorations and veneers was etched with 37% phosphoric acid for 15 seconds, rinsed with water for 60 seconds, and then air dried with compressed air to remove any moisture. A silane coupling agent was then applied. Following this, the bonding agent was applied, left for 5 seconds and then photopolymerised. The respective luting cement was then applied onto the fitting surface of the fabricated restorations, and the restorations seated onto the appropriate tooth. After removing excess cement, the preparation margins were light-cured for 30 seconds.

All the ceramic/Vita Enamic inlays, full crowns and veneers used in the studies in this thesis were fabricated in a CAD-CAM milling machine (inLab MC XL, Sirona Dental Systems GmbH, Bensheim, Germany). There were some variations in the types of indirect restoration used, and variations in fabrication methods which are presented in detail in the respective chapters.

2.3.3. Disc samples

Samples for each restorative material used in the studies conducted for this thesis were fabricated. For studies 1 and 2, the disc samples were made by compressing unset material between glass microscope slides. For study 5, the

unset material was compressed between a glass microscope slide and an acetate matrix separating strip.

The thickness of the discs varied from 2 mm to 5 mm, however the width was always 10 mm in diameter. Each direct restorative material was packed into a rigid plastic mould and then covered with a microscope glass slide to obtain a flat surface. Samples were then light cured for 60 seconds using an LED curing lamp After the samples had been retrieved from the mould, they were photopolymerised for a further 60 seconds, to ensure uniform curing of the entire thickness of the sample. A total of 3 samples were prepared for each shade type of each material. The ceramic samples were prepared by slicing the ceramic blocks provided by the manufacturer with a precision diamond saw (IsoMet™ 1000, Buehler, Lake Bluff, IL, USA). Once prepared, all samples of materials were coded and stored in airtight containers at room temperature.

2.3.4. Histogram analysis of images

For studies where image analysis using histogram data was part of the methodology, the samples were photographed in a standardised manner with a digital single-lens reflex camera (Canon model EOS Rebel T2i/EOS 550D, Canon, Tokyo, Japan) fitted with a 60 mm macro lens, and customised arrays of 405 nm LEDs and white LEDs with selectable orange and yellow optical filters (Figure 2.1). The camera was used in a dark environment at a fixed distance of 20 cm from the sample. The light used for illumination of the sample, and the chosen filter combination varied between studies. The same exposure

parameters (white balance, shutter setting and aperture setting) were used throughout.

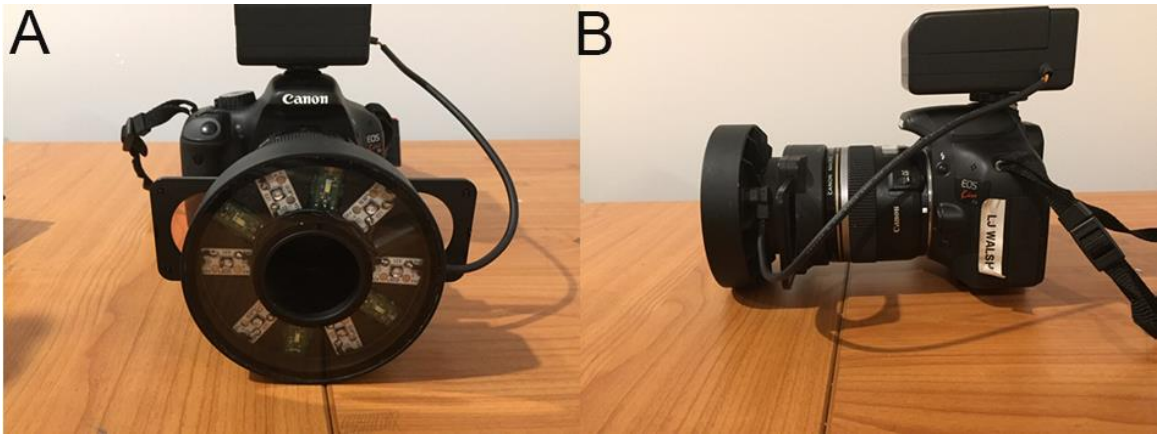

Figure 2. 1. Images of the customised DSLR camera for the study (Courtesy of Professor LJ Walsh). A. Front view showing the array of 405 nm LEDs and white LEDs, and the sliding lifter assembly which allows different filters or no filter to be selected. B. Side view of the setup. Details of the construction of the LED array and filters are published in Walsh LJ Takada M, Shinjo T. Intraoral inspection apparatus and method for operating intraoral inspection apparatus. US patent application 20130034826. 2013.

All studies used the same digital image analysis method. Digital image analysis was undertaken using Adobe Photoshop™ Creative Cloud software. For each image, colour channel data (that is, red, green, blue and luminosity) was collected from the sample, using the histogram tool, with a fixed number of pixels (Figure 2.2). The values for colour channel data ranged from 0 to 255 (8 bit colour). Mean values for red, green, blue and luminosity were used for statistical analysis. Because luminosity, the intrinsic brightness, is the outcome of reflectance and fluorescence phenomena, all fluorescence images were taken with a long pass filter to eliminate reflected light, so that only fluorescence

emissions were recorded. Therefore, the luminosity values in the study correspond to those for fluorescence emissions.

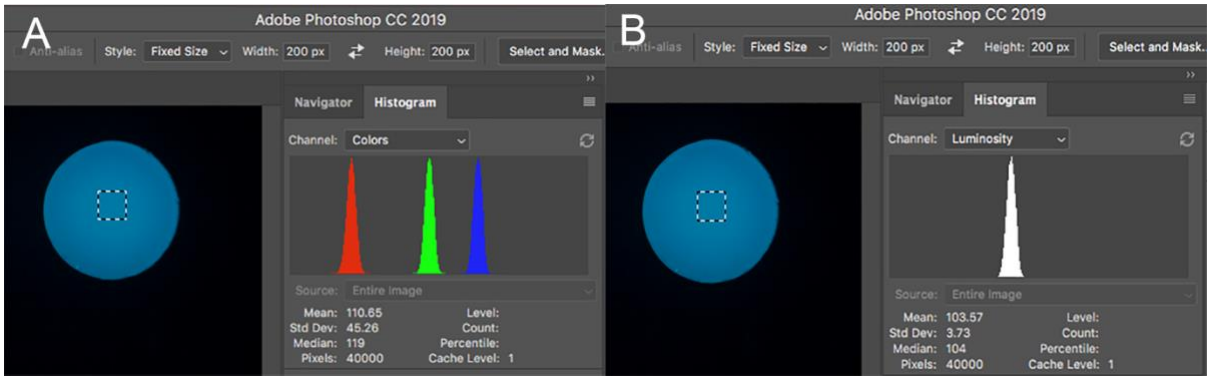

Figure 2. 2. Example of image analysis. A violet light fluorescence image of a disc sample is shown, with the sample selection area (a square of 40,000 pixels in area). The histogram analysis tool shows data for red, green and blue (A) and luminosity (B).

2.3.5. Disposal of extracted teeth used in the study

Once evaluated, the extracted human teeth used in the studies conducted for this thesis were disposed of according to the 2015 Australian Dental Association Infection Control Guidelines and in line with Queensland environmental protection laws on waste management. The teeth were placed into disposable cups and covered with setting plaster of Paris. Once set, the cups were disposed of in the general waste stream for landfill.

2.4. Methodology

2.4.1. Study 1 and 2 —Fluorescence emissions of tooth-coloured restorations

Study 1: The fluorescence properties of selected tooth-coloured restorative materials were analysed in studies 1 and 2. For study 1, a total of 27 tooth-coloured restorative materials were selected (Table 1), and three discs for each material type were prepared, as described earlier. The samples were stored in airtight containers, coded to de-identify them, and then stored at room temperature until further analysed.

Each sample was photographed in a dry state in a dark environment. The illumination source was a programmable multi-colour LED array with a remote controller (model 1R-1627S, Tristar, Colour Stars Inc, Irvine, CA, USA). The wavelengths tested included red (670 nm), orange (635 nm), yellow (585 nm), green (535 nm), cyan (470 nm), royal blue (450 nm), blue-violet (430 nm) and violet/UVA (405 nm). At each wavelength, the samples were photographed with either no filter, an orange filter or a yellow filter. Then the samples were stored in distilled water for eight weeks to hydrate them fully, after which they were photographed again.

Data for both dry and wet samples were collected using Adobe Photoshop™ Creative Cloud 2014 software, using histogram analysis.

Study 2: Based on the results from study 1, the 405 nm wavelength and the orange filter were chosen as suitable for study 2. Samples of 10 mm diameter and 5 mm thickness were fabricated for each shade and type of material used in

the study in a standardised manner. The prepared samples were then coded and stored in airtight containers at room temperature for 20 days, before being photographed in a dark environment using 405 nm light with an orange filter.

2.4.2. Study 3 — Comparative study of diagnostic methods

The study was based on the results of studies 1 and 2, used 405 nm light with an orange filter for identification of tooth-coloured restorations.

Three sets of maxillary and mandibular complete dental arches were fabricated by mounting extracted teeth in typodont models (model D95SDP-TRM.670, Nissin Dental Products Inc, Kyoto, Japan). The prepared models were stored in distilled water to maintain hydration of the teeth. To simulate clinical conditions, the models were then mounted in phantom head dental mannequins (Columbia Dentoform, Long Island City, NY, USA). Cavity preparations and restorations were completed in 55 mounted teeth with the selected tooth-coloured restorative materials. Details of the type of cavity and the restorative materials used are listed in Table 2.1.

Table 2. 1. List of restorations placed, and type of tooth-coloured restorative material used

Tooth number	Surfaces restored	Classification /Type of restoration	Brand Name	Shade	Batch Number
M1 15	D/O	Inlay	Vitabloc	1M2C	33450
M1 13	D/M/B/L	Class III & V	3M Filtek Supreme XTE	A2E	N710711
M1 11	B/I	Labial veneer	Vitabloc	1M2C	33450
M1 21	D/M/B/L /I	Full crown	Vitabloc	1M2C	33450
M1 23	M/B/L/D	Class III & V	Gradia	A3.5	1509172
M1 24	B	Class 1	Fuji IX	A2	1511101
M1 25	D	Class I	Voco Amaris	O1	1452078
M1 26	OBP	Class I	Admira Fusion	A2	1525583
M1 37	DO	Class II	Gradia	A3.5	1509172
M1 36	O	Class I	Voco Amaris	O1	1452078
M1 35	MOB	Class II & V	Voco Admira	OA2	1527294
M1 33	DLB	Class II & V	3M Filtek Supreme XTE	A2 E/B	N710711/ N767183
M1 31	D	Class II	Admira Fusion	A2	1525583
M1 43	BM root	Class V	Admira Fusion	A2	1525583
M1 44	B root	Class V	Admira Fusion	A2	1525583
M1 45	BM	Class V	Admira Fusion	A2	1525583
M1 46	O	Class I	3M Filtek Supreme XTE	A2 E/B	N710711/ N767183
M1 47	O	Class I	Herculite Ultra	A2D	5582394
M2 18	B	Class V	Voco Amaris	O1	1452078
M2 17	DO	Class II	Voco Admira	A2	1519084
M2 16	O	Class I	Gradia	XWT	1510201
M2 15	BO	Class I	Herculite Ultra	A2D	5582394

M2 13	B	Class V	Fuji VIII	A2	1504021
M2 12	DMBLI	Full crown	Vita Enamic	2m2T	52210
	DMBLI+	Full crown			5580988
M2 11	L	Class V	Vita Enamic/ Herculite XRV	E	
M2 21	B	Class V	Fuji VIII	A2	1504021
M2 22	LDMB	Class III + V	3M Filtek Supreme XTE	A2E/B	N710711/ N767183
M2 23	ML	Class III	Spectrum TPH Dentsply	A3	151020
M2 26	BO	Class I	Grandioso	A2	1512560
M2 27	MOD	Class II	Herculite XRV	D/E	5476946/ 5580988
M2 37	O	Class I	Gradia	A1	1512091
M2 36	O	Class I inlay	Vita Enamic	2M2T	52210
M2 34	D	Class II	Spectrum TPH Dentsply	A3	151020
M2 33	MBDLI	Class IV	Grandio	A2	1445171
M2 32	MLD	Class III	Gradia	A1	1512091
M2 43	MBDLI	Class IV	Grandio	A2	1445171
M2 45	O	Class I	Voco Amaris	O1	1452078
M2 47	O	Class I	Voco Admira	OA2	1527294
M2 48	MO	Class II	Voco Amaris	O1	1452078
M3 18	O	Class I	Herculite XRV	D	5476946
M3 17	MO	Class II	Voco Admira	OA2	1527294
M3 15	O	Class I	Voco Amaris	O1	1452078
M3 13	MBDLI	Full crown	Vita Enamic	2M2HT	
M3 12	BDLI	Class IV	Herculite XRV	D/E	5476946/ 5580988
M3 21	MBDLI	Class IV	Admira Fusion M: Herculite Ultra	A2 A2D	1525583 5582394
M3 22	MBDLI	Class IV	3M Filtek Supreme XTE	A2E /B	N710711/ N767183

M3 23	MBDLI	Full crown	Vita Enamic	2M2HT	40550
M3 24	DBO	Class II	Spectrum TPH	A3	151020
M3 26	O	Class I	Herculite Ultra	A2D	5582394
M3 28	O	Class I	Gradia	XWT	1510201
M3 38	O	Class I	Vocoamaris	T1	1447393
M3 37	O	Class I	Admira Fusion	A2	1525583
M3 36	OB	Class I	Gradia	A1	1512091
M3 44	OB	Class I	Voco Admira	A2	1519084
M3 45	DO	Class II	Grandio	A2	1445171
M3 46	OB	Claas I	Herculite Ultra	A2E	5444508

1 M1= Model 1, M2 = Model 2, M3 Model 3. B = buccal; M = mesial; D = distal; P = palatal; O = occlusal; L = lingual; I = incisal

Manufacturer details:
Vita Zahnfabrik, Bad Sackingen, Germany
Kerr, Uxbridge, UK
Voco, Cuxhaven, Germany
GC Corp, Tokyo, Japan
Dentsply, York, PA, USA

Four clinicians participated in the study. The models containing restored and unrestored teeth were mounted in a phantom head dental mannequin for examination purpose. Each examiner charted the restored and unrestored surfaces using three different diagnostic methods: conventional, FAIR and DIAGNOcam™ methods. The examiners repeated the whole process after four weeks, following the same protocol as before. The examiners were not involved in cavity preparation, restoration placement and fabrication of the models.

2.4.2.1. Conventional method

Conventional visual and tactile examination was performed using a double-ended sharp explorer, mouth mirror, three-way triple air/water syringe, and under regular white light illumination from a simulator-mounted dental unit lamp with 30,000 Lux light intensity (A-dec white HB-LED dental light, Newberg, OR, USA).

2.4.2.2. DIAGNOcam™

The DIAGNOcam™ system consists of a handpiece (Figure 2.3) with a built-in laser light source and a camera, and a customised stand to hold the handpiece. The handpiece is linked by a USB connection to a computer to receive the images. (Figure 2.4). The camera is a charge coupled device (CCD) sensor. The handpiece head has two plastic sheaths through which the optic fibers pass that deliver the laser light. When the handpiece head is placed on a tooth, the plastic sheathes extend over the tooth and the adjacent alveolar processes. Near infra-red laser light passes through the teeth and reaches the CCD sensor. A grey scale image is captured and saved (Figure 2.4). The handpiece is then moved along the arch length to examine all the teeth.

Figure 2. 3. The DIAGNOcam™ handpiece. Image adapted from the KaVo dental official website.

Figure 2. 4. DIAGNOcam™ in use.

2.4.2.3. DIAGNOcam™ examination method

Examination using the DIAGNOcam™ was performed following the manufacturer's recommended standard dental caries diagnosis technique, as described above, however instead of scoring dental caries, the examiners were looking for tooth-coloured restorations. Each examiner followed a specific protocol. The examination always started from the most distal tooth in the upper right quadrant and moved along the arch to the upper left quadrant. Then, in the lower arch, the teeth were examined starting from the most distal tooth in the lower left quadrant and moved along the arch towards the right side. Three images were taken for each tooth, with the point of focus being on the distal third in the first image, then the middle third, and finally the mesial third. The images were coded and saved. The examiners then examined all images on the computer screen and scored filled and unfilled surfaces. While doing this, they used features of the software including grey-scale evaluation, and image enlargement.

2.4.2.4. Examination by the FAIR method

Examination using the FAIR technique was conducted with the aid of a mouth mirror. The illuminating light was from an array of LEDs (405±10 nm, model SEFL33UV-405, SE Electronics, Shanghai, China) that was attached to the handle of the dental unit light, so that the examiner could adjust the incident light according to their convenience, in the same way as they would position a conventional dental unit lamp. Examiners wore orange eyewear that acted as a

long cut-off filter (λ ≥ 520nm). Other light sources in the room were turned off. This method was adopted as it could be applied clinically without changes to a regular dental surgery setup. The sensitivity and specificity of identifying filled and unfilled surfaces was determined.

2.4.3 Study 4 — Guided removal of restorations

A total of 60 extracted human permanent posterior teeth devoid of any pathology were used in this study. The teeth were embedded in poly-siloxane-based silicone laboratory modelling material (Lab-putty, Coltene Whaledent AG, Altstätten, Switzerland). These were located in 15 sets of models (4 teeth in each).

2.4.3.1 Model preparation

Class I GV Black cavities 2 mm wide and 2 mm deep were prepared in the molar teeth, and similar cavities 1.5 mm wide x 2 mm deep prepared in the premolar teeth. In each tooth, the cavity outline varied according to the anatomical complexity of the tooth. All the 15 models were then coded. A pre-operative digital 3-dimensional scan of each model was performed using a 3Shape TRIOS® 3 intraoral scanner. This scanner was calibrated in line with the manufacturer's recommendations. The scanned three-dimensional images were identified and saved in STL (stereolithography) format.

To ensure precise shade matching for the restorations that were to be placed in these cavities, a custom-made shade guide was fabricated for all the three brands and shades of material used. The cavities were then restored using one of the three different brands of tooth-coloured restorative material: Admira® Fusion (Voco Gmbh, Cuxhaven, Germany) (10 shades), or GRADIA® DIRECT X (GC Corp., Tokyo, Japan) (7 shades), and TPH Spectra® LV (Dentsply DeTrey, Konstanz, Germany) (13 shades). Shade selection corresponded to that of the individual teeth, and the restoration technique used was oblique layering, with the goal being a perfect match to the tooth shade. The models with the restored teeth were then stored in distilled water at room temperature until used.

2.4.3.2. Removal of restorations

Five experienced dentists were then tasked with removing the restorations from the teeth. In total, 12 teeth were assigned to each operator (six teeth for the conventional method, and six teeth for the FAIR method). In the conventional method, the operators used a dental unit white LED lamp with an intensity of 30,000 lux (white HB-LED dental light, A-dec, Newberg, OR, USA) as their working light. With the FAIR method, they used the light from an array of 9 LEDs (405±10 nm, model SEFL33UV-405, SE Electronics, Shanghai, China). The LED array was attached to the handle of the dental unit light. Additional aides such as double-ended sharp explorer, conventional mouth mirror, and a three-way triple air/water syringe were also used during restoration removal for identification of remnants of composite resin material.

The restorations were removed using water-cooled straight fissure diamond burs in a high-speed contra angled dental handpiece (model S-Max M600L, NSK). The total time needed to remove each restoration was recorded in seconds. After completion of restoration removal, a post-operative optical three-dimensional scan was performed for each model.

2.4.3.3. Inter-cuspal cavity width measurements

3D Tool version 13.2 CAD software was used to compare the pre- and post-operative 3D digital scans. Inter-cuspal cavity width measurements were taken using the measure and mark-up distance tool. To quantify the extent of lost tooth structure during restoration removal, the linear distance between the cavity walls at their external marginal line angles was measured, to the nearest 0.01 mm. In total, four repeat measurements were taken for each tooth at specific sites, in both pre- and post-operative 3D scans, and the differences between the mean values used to assess changes in cavity shape.

2.4.4. Study 5 — Effect of heat on restorations

2.4.4.1. Sample preparation

A total of 132 extracted human permanent teeth (premolars and molars) devoid of any pathology were selected for this study. The teeth were randomly divided into 11 groups of 12 teeth each, allowing one group for each of the tested restorative material types used in the study. One group of 12 teeth was retained

as controls (unrestored teeth). A total of 24 teeth were prepared for full crown restorations. The teeth were then restored using the relevant tooth-coloured restorative materials. Placement of restorations was performed according to the standard protocol for each material type. A detailed description of the protocol followed for each material type is described in section 3.3.2. The restored teeth were then mounted into individual custom trays containing heat-stable dental investment for ease of manipulation.

As a further control for effects of heat on the material alone (that is, outside of a tooth), three discs 10 mm in diameter and 2 mm in thickness, were prepared for each restorative material, using a glass microscope slide to compress unset material against an acetate strip.

2.4.4.2. Methodology

To determine visual colour changes, changes in positions of restorative margins, and to measure the baseline colour, the prepared samples were photographed in a dark environment, first under white light (WL) without any filters, and then under 405 nm light with an orange and yellow filter combination. The light source was perpendicular to the sample. All the images were taken under standardised conditions with a fixed camera to a sample distance of 200 mm, against a dark background.

The discs and the restored teeth were randomly divided into four sub-groups of three teeth each, and allocated to different heat treatments (200 ◦C, 500 ◦C, 900

◦C, or 1200 ◦C). Each tooth was placed in a 30 mL glazed porcelain crucible with a lid that was designed for high temperature use (Cole-Parmer™) (Figure 3.5). Discs of materials were placed in a custom-made tray fabricated by using heat-stable dental investment material (Bellavest®, BEGO Bremer Goldschlägerei Wilh. Herbst GmbH & Co., Bremen, Germany).

The porcelain containers and custom trays containing discs were then placed in a silicon carbide muffle chamber furnace (ModuTemp™) (Figure 3.5), and were exposed to the predetermined temperature, at an increasing rate of 10 ◦C per minute, with a holding time of 30 minutes at the pre-set temperature. The furnace was then turned off and the samples allowed to cool slowly. Once the oven chamber reached room temperature, the samples were removed and photographed in the same manner as before. Features such as macroscopic changes, changes in colour, and alterations in restoration margins were recorded.

Figure 2.5. Crucibles and custom trays. These were used for tooth samples and disc samples, respectively. The furnace is shown on the right image.

Chapter 3 Variation in the fluorescence of tooth-coloured restorative materials and the natural tooth structure

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Hybrid restorative materials might present unique emission spectra

This chapter is published as a peer reviewed article (Appendix D):

Kiran R, Chapman J, Forrest A, Tennant M, Walsh LJ. Forensic applications: Fluorescence properties of tooth-coloured restorative materials using a fluorescence DSLR camera. Forensic Sci Int. 2017;273:20-8.
https://doi.org/10.1016/j.forsciint.2017.01.022

This chapter assessed the fluorescence properties of tooth-coloured restorative materials using a fluorescence camera, when illuminated with a range of wavelengths of LED lights and viewed under specific optical filters. The difference between the wet and dry samples was also assessed to assess the influence of moisture on the fluorescence properties of the tested tooth-coloured restorative materials.

3.1. Introduction

Demand for aesthetic materials in restorative dentistry has led to the evolution of new types of tooth-coloured restorative materials, which attempt to accurately mimic the optical nuances of natural tooth structure. Various combinations of tooth-coloured materials now exist including resin modified and reinforced glass-ionomer cements (GIC), ormocers (organically modified ceramics) and hybrid ceramic materials, which are combinations of polymers and ceramic materials.

In forensic odontology, victim identification using dental records is an efficient and well-established method and may be used in combination with other means of identification. Information recorded on dental charts of restored, non-restored, missing and decayed surfaces of teeth can be used for comparison with post-mortem dental features. Additional individuation may be possible if the details of the brand and type of aesthetic restorative materials are accurately recorded in the treatment notes (164–166).

Metallic restorations are easily distinguished from sound tooth structure both by direct vision and by radiographic appearance. When restorative materials mimic the appearance of tooth structure very closely, however, this raises the challenge as to how they can be detected reliably both clinically and during radiographic examination (78, 87). Not all tooth-coloured restorations have sufficient radiographic contrast with tooth structure to allow detection on dental radiographs (82).

Quick, accurate methods to detect their presence and to correctly classify their type and brand would be an asset for both routine clinical examination and forensic identification purposes (77, 90).

Comparing the different fluorescence properties of sound and decayed teeth is a well-established method for finding early dental carious lesions, and the same method has been applied to distinguish tooth-coloured restorations from adjacent normal tooth structure (77, 78, 163).

Aesthetic dental restorative materials vary considerably in their fluorescence properties, as it is difficult for them to replicate all the fluorescence properties of natural tooth structure at all wavelengths of light. Light in the ultraviolet and visible violet range has been found useful for detection of resin restorations (70,78). However historically, light sources used for eliciting such fluorescence were high-intensity fluorescent light sources and lasers. The latest generation of

UV-emitting LEDS (Light-Emitting Diodes) have several advantages over these, including high electrical efficiency, small size and low cost, as well as long operating life. Given the potential for fluorescence to aid in recognising tooth coloured restorations, the objective of this study was to compare the fluorescence properties of dry and wet samples of contemporary tooth-coloured restorative materials when exposed to different wavelengths of visible light from LED sources. To remove the light used to excite the fluorescence, samples were viewed through coloured filters.

The study tested two hypotheses: (1) that short wavelength (UV-A/violet) light will give the greatest differentiation between different materials and between the materials and the adjacent tooth structure; and (2) that hybrid restorative materials would exhibit a recognisably unique emission spectrum different from that of all other classes of tooth-coloured materials.

3.2. Materials and methods

In this study, a series of 27 selected tooth-coloured restorative materials and three human permanent and three human deciduous extracted teeth were included (Table 1). The restorative materials were all prepared according to the manufacturer's instructions, and each was formed into the shape of a disc 2 mm thick and 10 mm in diameter, using a rigid plastic matrix. The samples were coded to de-identify them, and then stored in sealed containers at room

temperature. An LED curing light (Mini LED, Acteon Satelec, Merignac, France), which emitted visible blue light over the wavelength range of 420 to 480 nm, was used in pulsed mode at a power density of 1250mW/cm2 for curing the composite restorative materials. The prepared samples were photographed in a standardised manner with a digital single-lens reflex camera (Canon model EOS Rebel T2i/EOS 550D, Tokyo, Japan) fitted with a 60 mm macro lens in a dark environment illuminated by the various LEDs, in combination with clear, orange and yellow filters. Both the light source and the camera lens were kept at a fixed distance of 200 mm and an angle of 85 degrees from the surface of the samples.

Table 3. 1. Brand names, shade and generic type of tooth coloured restorative materials used in this study

Sl No	Sample material	Shade	Generic type of material
1	Permanent Teeth-		
2	1		
3	Permanent Teeth -		
4	2 Permanent Teeth-3 Vita Enamic	3M2HT	Hybrid: ceramic reinforced with polymer network
5	Vita Enamic	2M2T	Hybrid: ceramic reinforced with polymer network
6	Vita Enamic	2M2HT	Hybrid: ceramic reinforced with polymer network
7	Vitablocs	OM1C	Feldspar ceramic blocks
8	Vitablocs	1M2C	Feldspar ceramic blocks
9	Voco Admira	A2	Hybrid: Ormocers-organic modified ceramic
10	Voco Admira	OA2	Hybrid: Ormocers-organic modified ceramic
11	Herculite XRV	D	Composite: Microhybrid filled
12	Herculite XRV	E	Composite: Microhybrid filled
13	Herculite Ultra	A2E	Composite: Nanohybrid filled
14	Herculite Ultra	A2D	Composite: Nanohybrid filled
15	Gradia	A3.5	Composite: Microhybrid
16	Gradia	A1	Composite: Microhybrid

17	Gradia	B1	Composite: Microhybrid
18	Gradia	XWT	Composite: Microhybrid
19	VocoAmaris	O1	Compoiste: Nanoreinforced hybrid
20	VocoAmaris	T1	Compoiste: Nanoreinforced hybrid
21	Grandio	A2	Composite: Nanohybrid filled
22	Admira Fusion	A2	Hybrid: Nanohybrid Ormocer
23	GrandioSO	A2	Composite: Nanohybrid
24	Fuji II	A1	Hybrid: Resin modified Glass -Ionomer cement (light cured)
25	Fuji VIII	A2	Hybrid: Resin modified Glass -Ionomer cement (self cured)
26	Fuji IX	A2	Conventional Glass-Ionomer cement
27	Spectrum TPH Dentsply	A2	Composite: sub-micron filled
28	Spectrum TPH Dentsply	A3	Composite: sub-micron filled
29	3M Filtek Supreme XTE	A2E	Composite: Nanofilled
30	3M Filtek Supreme XTE	A2B	Composite: Nanofilled
31	Deciduous teeth-1		
32	Deciduous teeth-2		
33	Deciduous teeth- 3		

The camera was used with the following settings (speed ISO 400, aperture F2.8, and shutter speed 1/30 second). The camera was set to use the Adobe RGB1998 colour space. Images were recorded using 8 bits per colour channel (16,777,216 colours) with an image size of 18 megapixels, using a constant white balance setting of white fluorescent light with white balance correction set to zero. The samples were imaged with the incident light rays being perpendicular to the surface of samples (Figure 3.1).

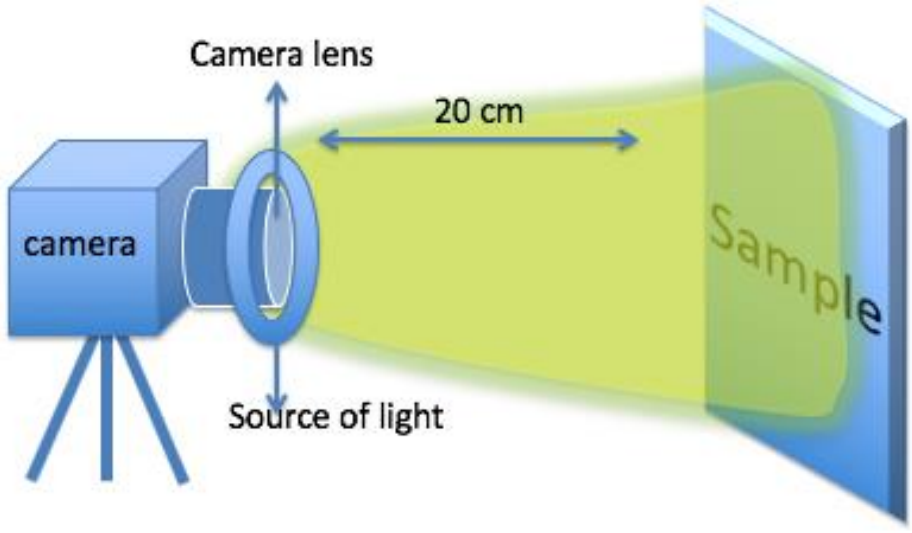


Figure 3. 1. Schematic representation showing the camera, light source and sample position in the laboratory

A programmable multi-colour LED array was used with a remote controller (model 1R-1627S, Tristar, Colour Stars Inc, Irvine, CA) to deliver the chosen wavelengths to illuminate the samples. From longest to shortest, the wavelengths were red (670 nm), orange (635 nm), yellow (585 nm), green (535 nm), cyan (470 nm), royal blue (450 nm), blue-violet (430 nm) and violet/UV-A (405 nm). Each light source had a spectral bandwidth of 30 nm. Samples were photographed with and without the use of filters (clear, yellow, or orange).

In order to assess the effect of water sorption on the fluorescence emissions of the various tooth-coloured restorative materials, each sample was first photographed in the dry state, then stored in distilled water for eight weeks at room temperature and photographed again. Additional readings were then made

after the samples had been returned to dry storage for a further 60 days. Extracted human permanent teeth were included as positive controls so that fluorescence patterns could be compared with the natural enamel of human teeth. The use of extracted teeth for this study was approved by the institutional human research ethics committee (approval number H15/03-035).

3.2.1. Statistical analysis

Digital image analysis was undertaken using Adobe Photoshop™ Creative Cloud 2014 software, applying the histogram tool to collect colour channel data for each sample. The software was set to the RGB 1998 colour space to match the camera setting. Values varied from 0 to 255 for each colour channel data as the images were recorded in 8-bit colour.

The differences in fluorescence for each sample under different combinations of incident light wavelengths and filters was analysed using the mean and standard deviation values for a constant sample area of 40,000 pixels, which corresponded to a sample area of 53 mm^2. The mean values for luminosity were used for statistical analysis. Luminosity refers to the brightness of the object, which is the result of both fluorescence and reflectance phenomena. Appropriate filters can remove the reflected light and hence all the luminosity measured arises from fluorescence emissions.

Analyses were undertaken to show the influence of material type, variations due to differences in shades for the same material, and differences from natural tooth enamel. The statistical analysis for a given material compared the influence of moisture (dry versus wet samples), the choice of wavelengths of light used for excitation, and the effects of filters. Analyses were undertaken using ANOVA or repeated measures ANOVA as appropriate, with post-hoc Bonferroni tests. The threshold for significance was set at $P < 0.05$.

3.3. Results

3.3.1. Effect of excitation wavelength and the effect of applying filters

After imaging all the samples and extracted teeth selected for this study, using the complete array of LEDs ranging from 405 nm to 670 nm with and without filters, we found that the samples exhibited within a narrow range of emission spectra for a given light and filter combination with few exceptions. Yellow light excitation with orange filter showed the maximum emission ranging from 210 to 255 for all samples, while characteristically under blue light excitation (450± nm) and with orange filter, all the materials exhibited smaller luminance values in the range of 20 to 75. In general, when imaged under the orange filter most of the materials exhibited less emission spectra; that is, all the luminosity values were less in comparison to without filter and the yellow filter for a given excitation light. It was also observed that the UV-A/Violet light (405± nm) produced the greatest range of emission spectra (10 to 204) among the tooth-coloured restorative

materials and, in comparison, with tooth structure (Figure 3.2). The analysis of

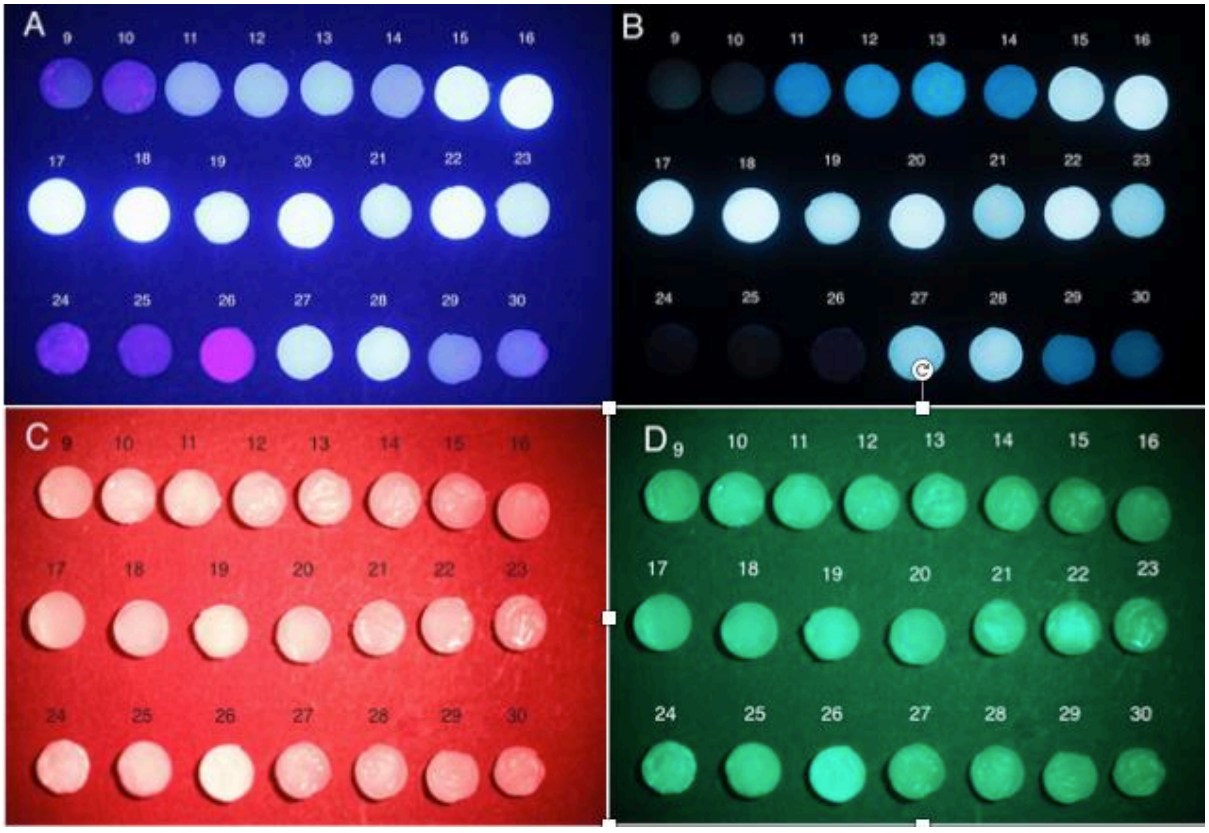


Figure 3. 2. A: Images obtained using Canon EOS camera, following irradiation with UV-A/violet light (405nm) and using yellow filter (Shorter wavelength fluorescence emissions), B: UV-A/violet (405nm) light with orange filter (longer wavelength fluorescence emissions), C: Orange light-635nm with clear filter, D: Green light-535nm with orange filter. The numbers in the images represent the materials in the Table 3.1.

the variance test also showed statistically significant variation in fluorescence emission pattern by the restorative materials, a variance value of 6605 for UV-A/Violet light + the orange filter and 5560 for UV-A/Violet+ the yellow filter when compared to other combinations of light and filter which ranged from 149 to 1130 (Table 2). In comparison with UV-A/Violet + the yellow filter, UV-A/Violet + the orange filter showed the maximum differentiation visibly (Figures 2a, 2b, 2c and

2d) and statistically, which confirms the first of the study hypotheses: that short wavelength (UV-A/Violet) light will give the greatest differentiation between different materials and between the materials and the adjacent tooth structure. The fluorescence emission spectra plotted for each light wavelength and filter clearly demonstrated greater variation for UV-A/Violet light as compared to other wavelengths (Figure 3.3a and 3.3b). The repetitive data analysis for mean luminosity values of dry samples, which were re-recorded after 2 months, statistically did not show much variation ($p>0.05$) (see Table 3).

Table 3. 2. Overview of the ANOVA analysis for variance in peak fluorescence emission spectra of dry samples when illuminated with different combinations of light and filter

Light and filter combination	*Sum*	*Average*	*Variance*
Red with clear filter	4659.1	141.2	1129.5
Red with orange filter	4314.6	130.7	784.9
Red with yellow filter	4561.3	138.2	907.2
Orange with clear filter	7680.9	232.8	540.1
Orange with orange filter	6169.9	187.0	687.3
Orange with yellow filter	7118.3	215.7	473.8
Yellow with clear filter	8217.5	249.0	149.2
Yellow with orange filter	7116.8	215.7	484.5
Yellow with yellow filter	7975.4	241.7	191.7
Green with clear filter	7502.3	227.3	613.4
Green with orange filter	5369.2	162.7	676.9
Green with yellow filter	6849.0	207.5	536.7
Cyan with clear filter	7678.0	232.7	387.4
Cyan with orange filter	5544.0	168.0	712.9
Cyan with yellow filter	7063.5	214.0	442.5
Royal blue with clear filter	5140.0	155.8	831.5
Royal blue with orange filter	1497.0	45.4	187.9
Royal blue with yellow filter	4110.4	124.6	487.2
Bluish violet with clear filter	7387.6	223.9	1251.7
Bluish violet with orange filter	4578.1	138.7	574.6
Bluish violet with yellow filter	6672.6	202.2	1050.6
Ultra violet A with clear filter	7178.6	217.5	2268.4
Ultra violet A with orange filter	3998.8	121.2	6605.4
Ultra violet A with yellow filter	5532.1	167.6	5560.8

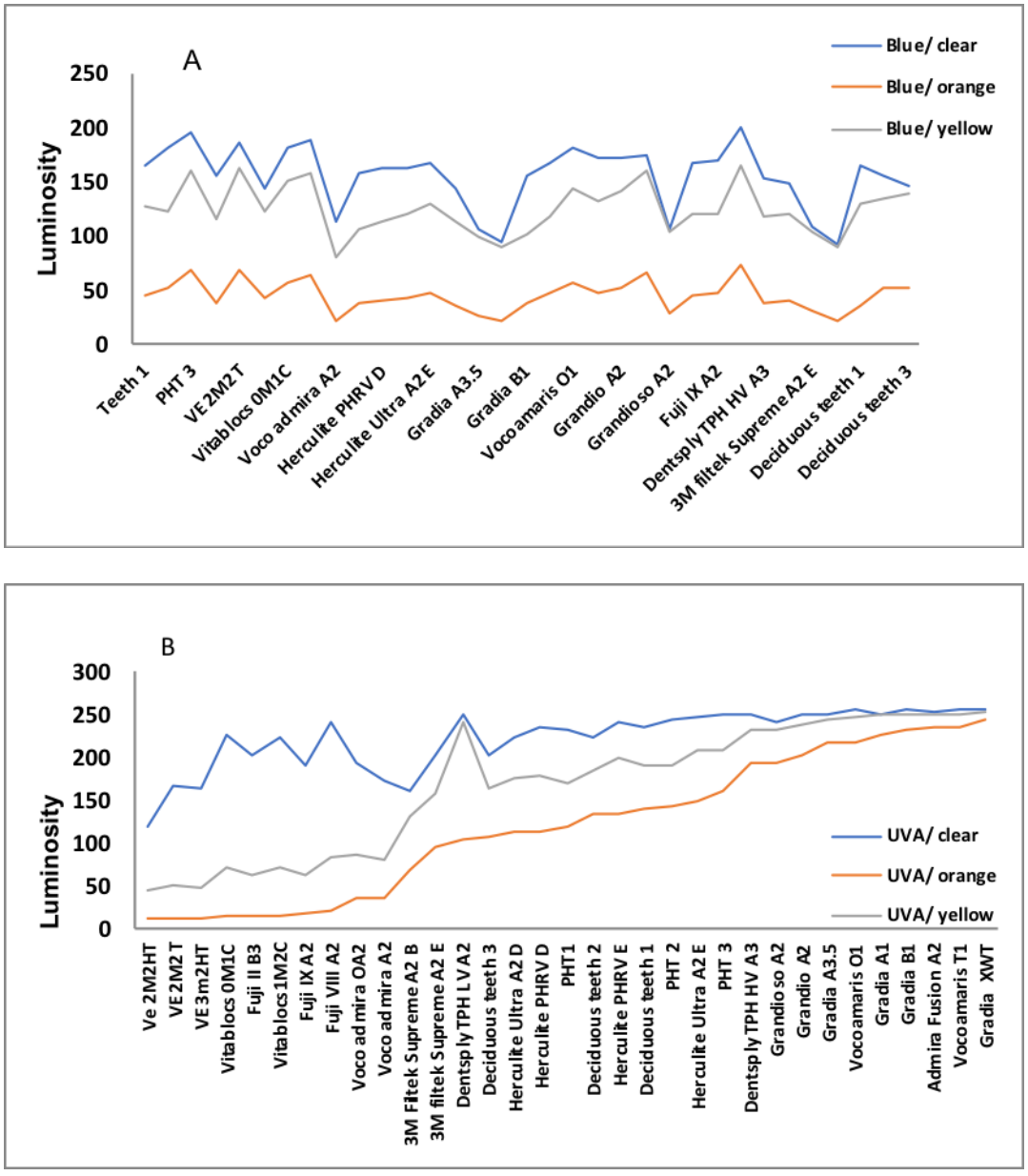


Figure 3. 3a. Peak fluorescence emission of restorative materials and teeth samples when excited with blue (450 nm) light and imaged with clear (straight line), orange (striped dotted line) and yellow (dotted line) filters. Figure 3.3b. Peak fluorescence emission of restorative materials and teeth samples plotted in ascending order when irradiated with Ultraviolet-A light (405) nm and imaged with clear (straight line), orange (striped dotted line) and yellow (dotted line) filters.

Table 3. 3. ANOVA repeated measures test results of two repetitive data of dry samples

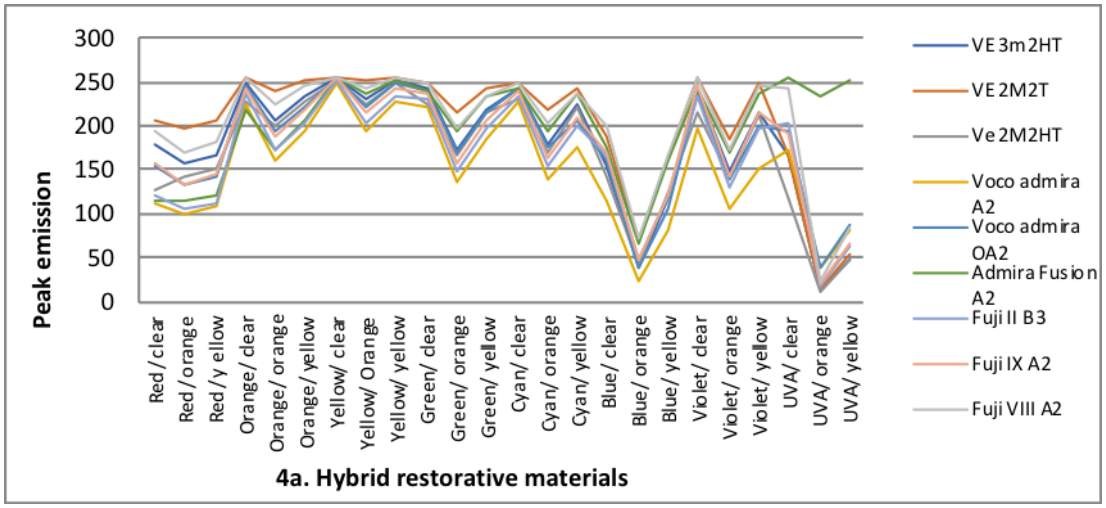
ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Sample	211.2321394	1	211.2321394	0.181999588	0.669722811	3.847783877
Columns	3685300.221	22	167513.6464	144.3313252	0	1.549313996
Interaction	1692.048576	22	76.91129894	0.066267495	1	1.549313996
Within	1708430.843	1472	1160.618779			
Total	5395634.345	1517				

3.3.2. Material type by classification

The restorative materials included in this study can be grouped into three categories of material type: resin composites, hybrids, and ceramics. Ceramic restorative materials very characteristically exhibited the lowest luminosity intensities under UV-A/Violet + orange filter (15) and blue + orange filter combination (44 to 63) (Figure 3.4). Interestingly, when illuminated with UV-A/Violet light and imaged with the orange filter, the hybrid materials such as VitaEnamic™, ormocers and resin modified glass-ionomer cements exhibited very low luminance values (Figure 3.4). This addresses one of the hypotheses of this study, which was to determine whether hybrid restorative materials would exhibit a recognisably unique emission spectrum different from that of all other classes of tooth-coloured materials. VocoAdmira™, an ormocer, had low emission whereas Admira Fusion™, which is a newer ormocer from the same brand, showed high fluorescence emission, that is, around 234.63±. Among the

GICs, Fuji-VIII A2 had the highest emission peak and Fuji-II the lowest. When illuminated with UV-A/Violet light and imaged with a yellow filter, Fuji VIII A2 exhibited unique bright pink visible fluorescence, and other hybrid materials such as VitaEnamic™, ormocer and glass-ionomer cements exhibit bluish-pink emission (Figure 3.3a). Among resin composites, Herculite brand materials showed fluorescence emission (112 to 150), which was closer to tooth structure (108 to 162) when imaged under the UV-A light and orange filter combination. The rest of the composite materials showed emission peak in a higher range (that is, above 200) without much variation.



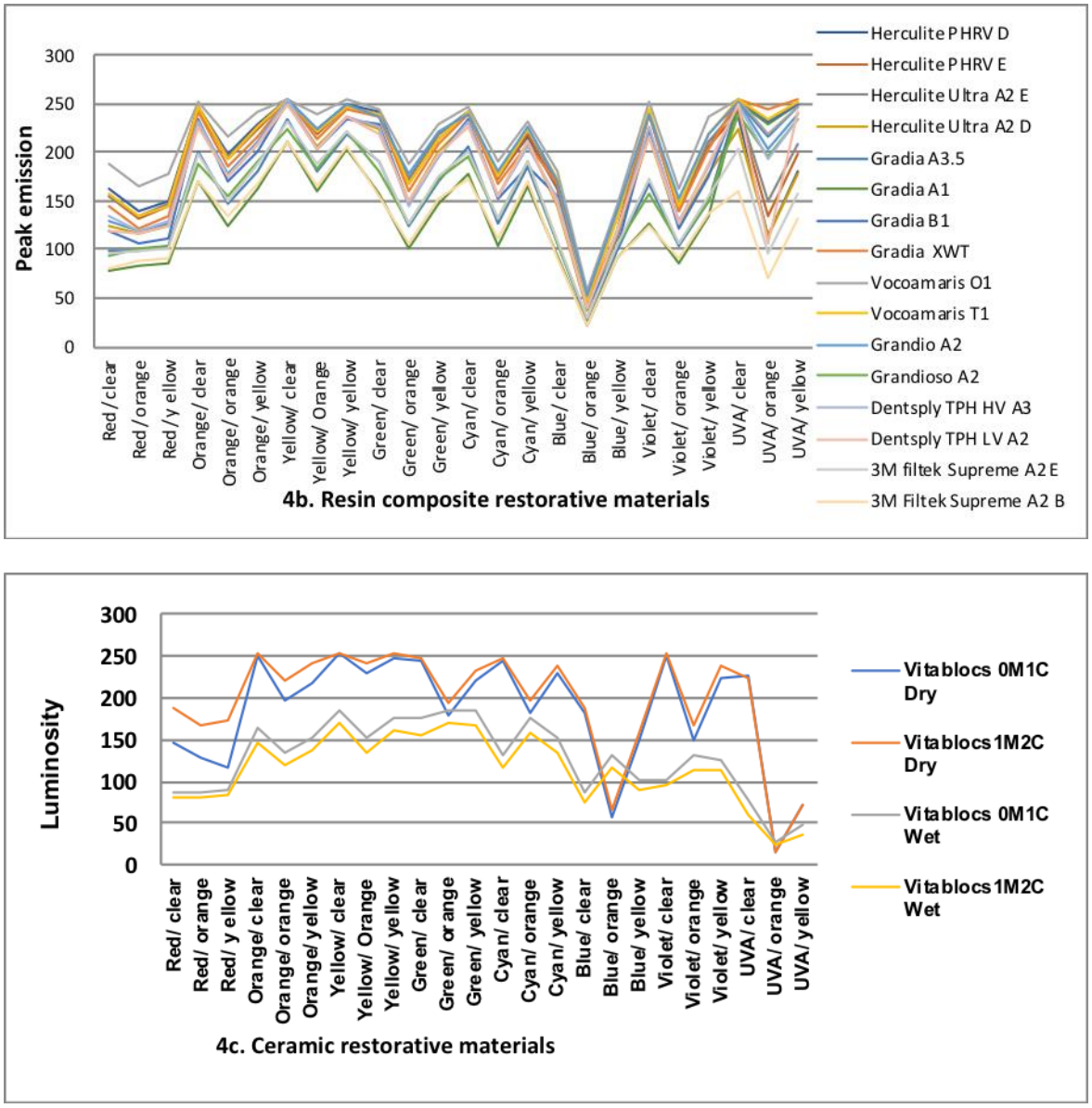


Figure 3. 4. Distribution of peak emission according to material type. 4a: Hybrid restorative materials, 4b: Resin composite materials, 4c: Ceramics

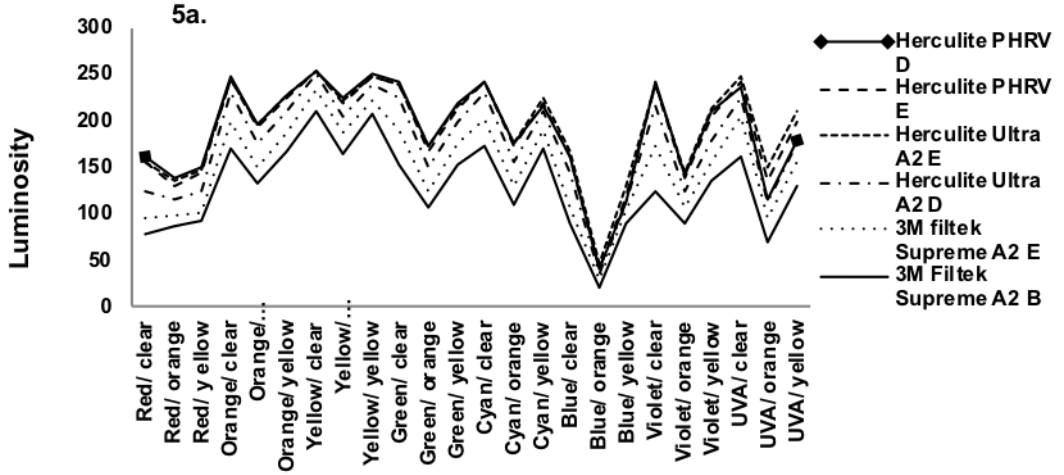
3.3.3. Shade distribution

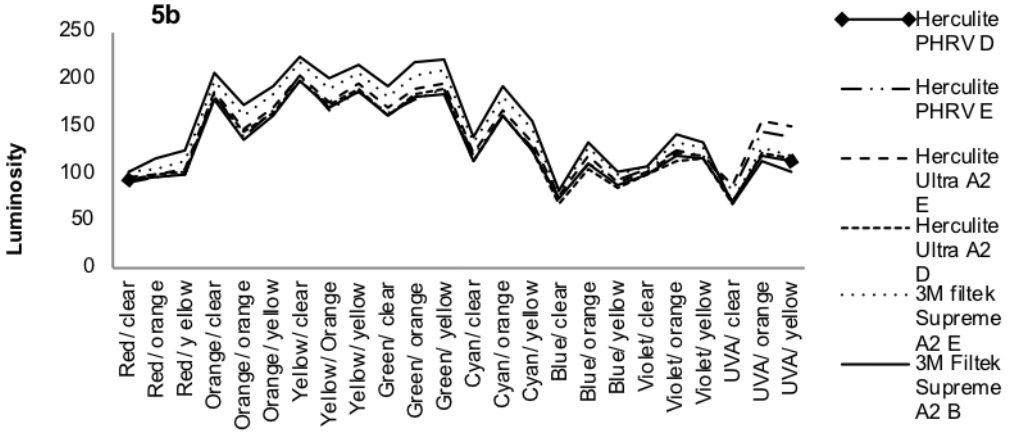
Considering that there are numerous shades for each material type, we focused on selecting enamel/dentine shades and opaque/translucent shades based on the options available for that particular material. The number of available shades

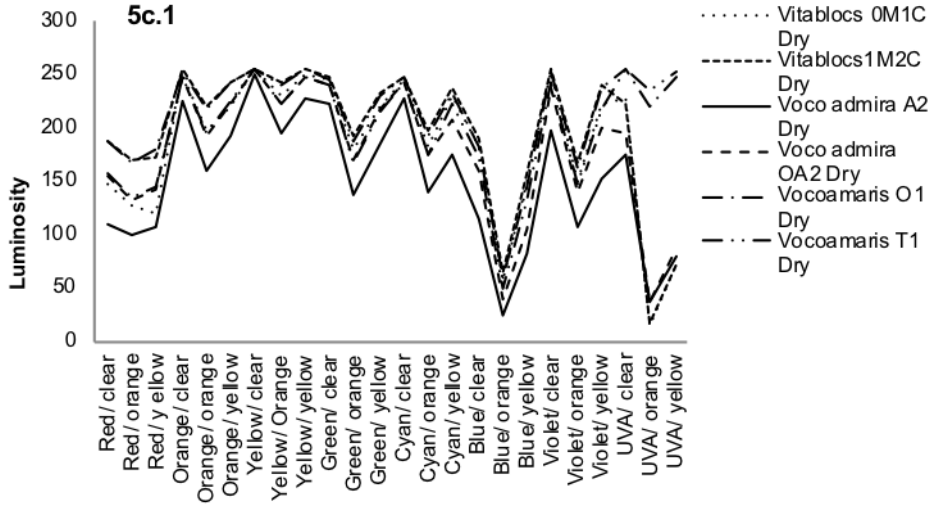
per brand ranged from one to four. Among the brands, which had both enamel and dentine shades, dentine shades exhibited reduced luminance peaks to enamel shades both in dry and wet states except for 3M Filtek Supreme, in which the dentine shade exhibited the highest peak of emission under all combinations of light wavelength and filter with the exception of UV-A/violet light illumination where it had the lowest emission peak (Figure 3.5a). Peak emission for all six materials under the enamel and dentine group was with a yellow light + without filter combination. For brands, which had opaque shades, these demonstrated greater luminance values in both dry and wet samples, except for Vitabloc, which exhibited reduced emission spectra for dry samples of opaque shades (Figure 3.5c). Gradia XWT, which is an extra-white product, very clearly had greater emission spectra under all combinations of light and filter in comparison to other shades of this brand.

3.3.4. Dry versus wet samples

In general, all wet samples of restorative materials demonstrated reduced levels of fluorescence emission in comparison to dry samples when illuminated with


5a.
Luminosity
300
250
200
150
100
50
0
Red/ clear
Red/ orange
Red/ y ellow
Orange/ clear
Orange/...
Orange/ yellow
Yellow/ clear
Yellow/...
Yellow/ yellow
Green/ clear
Green/ orange
Green/ yellow
Cyan/ clear
Cyan/ orange
Cyan/ yellow
Blue/ clear
Blue/ orange
Blue/ yellow
Violet/ clear
Violet/ orange
Violet/ yellow
UVA/ clear
UVA/ orange
UVA/ yellow
Herculite PHRV D
Herculite PHRV E
Herculite Ultra A2 E
Herculite Ultra A2 D
3M filtek Supreme A2 E
3M Filtek Supreme A2 B


5b
Luminosity
250
200
150
100
50
0
Red/ clear
Red/ orange
Red/ y ellow
Orange/ clear
Orange/ orange
Orange/ yellow
Yellow/ clear
Yellow/ Orange
Yellow/ yellow
Green/ clear
Green/ orange
Green/ yellow
Cyan/ clear
Cyan/ orange
Cyan/ yellow
Blue/ clear
Blue/ orange
Blue/ yellow
Violet/ clear
Violet/ orange
Violet/ yellow
UVA/ clear
UVA/ orange
UVA/ yellow
Herculite PHRV D
Herculite PHRV E
Herculite Ultra A2 E
Herculite Ultra A2 D
3M filtek Supreme A2 E
3M Filtek Supreme A2 B


5c.1
Luminosity
300
250
200
150
100
50
0
Red/ clear
Red/ orange
Red/ y ellow
Orange/ clear
Orange/ orange
Orange/ yellow
Yellow/ clear
Yellow/ Orange
Yellow/ yellow
Green/ clear
Green/ orange
Green/ yellow
Cyan/ clear
Cyan/ orange
Cyan/ yellow
Blue/ clear
Blue/ orange
Blue/ yellow
Violet/ clear
Violet/ orange
Violet/ yellow
UVA/ clear
UVA/ orange
UVA/ yellow
Vitablocs 0M1C Dry
Vitablocs1M2C Dry
Voco admira A2 Dry
Voco admira OA2 Dry
Vocoamaris O1 Dry
Vocoamaris T1 Dry

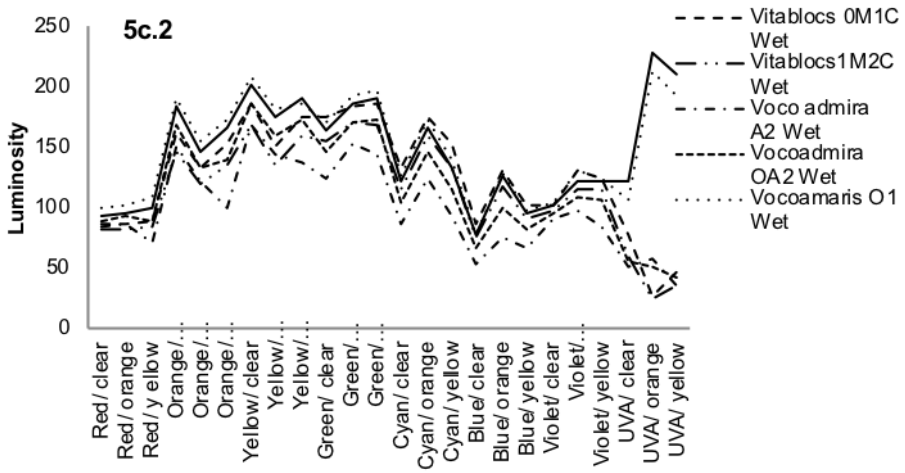
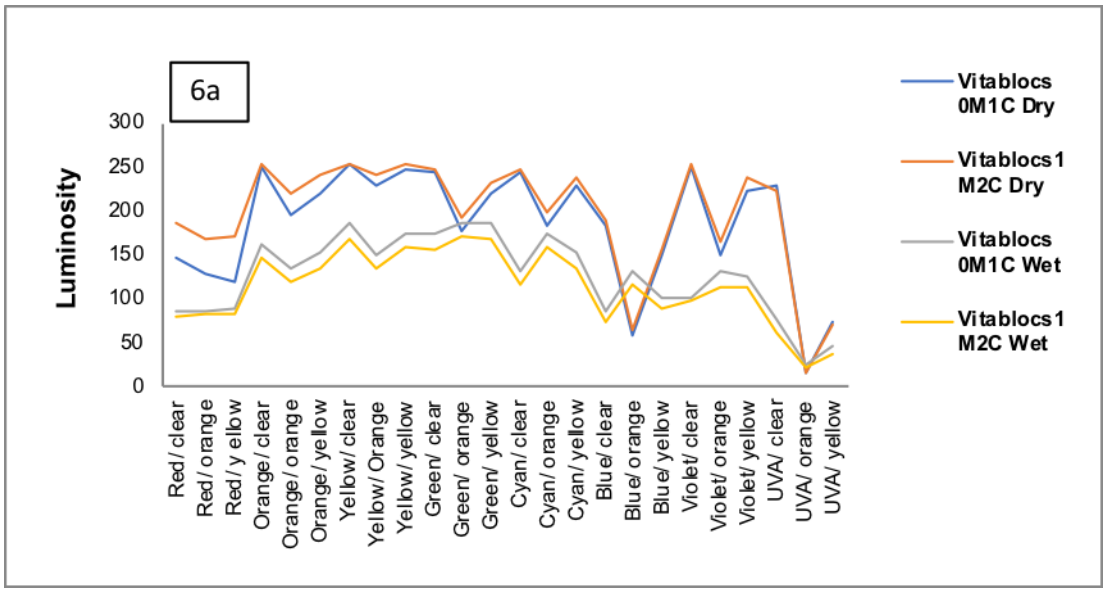


Figure 3. 5. Distribution of peak emission according to materials shade. a: Dry and wet samples of materials with dentine and enamel shades, b: Dry and wet samples of materials with opaque and translucent shades.

red, orange and yellow light (670± nm to 585± nm). This is in contrast to illumination with light ranging from 535± nm to 405± nm (that is, from green to UV-A/Violet), where the emission pattern exhibited some unique features in that, with an orange filter, the wet samples showed greater emission in comparison to dry samples. For whole teeth samples this reverse pattern of emission among wet and dry samples was seen only with UV-A + an orange filter and UV-A with a blue + orange filter combination (Figure 3.6a and b). We observed, however, that 3MFiltek Supreme XTE™ composite material exhibited the highest level of fluorescence emission when the samples were wet but exhibited the second lowest level of fluorescence emission when dry, in comparison to rest of the restorative materials.

3.3.5. Permanent vs deciduous teeth

Among the deciduous and permanent teeth, the peak fluorescence emission did not differ significantly from one tooth to another, although we observed a slight variation between posterior teeth and anterior teeth. Under all of the excitation light and filter combinations used in this study, the wet samples of teeth showed significantly reduced emission spectra in comparison to dry tooth samples with the exception of the UV-A light + orange filter and blue light + orange filter combinations.



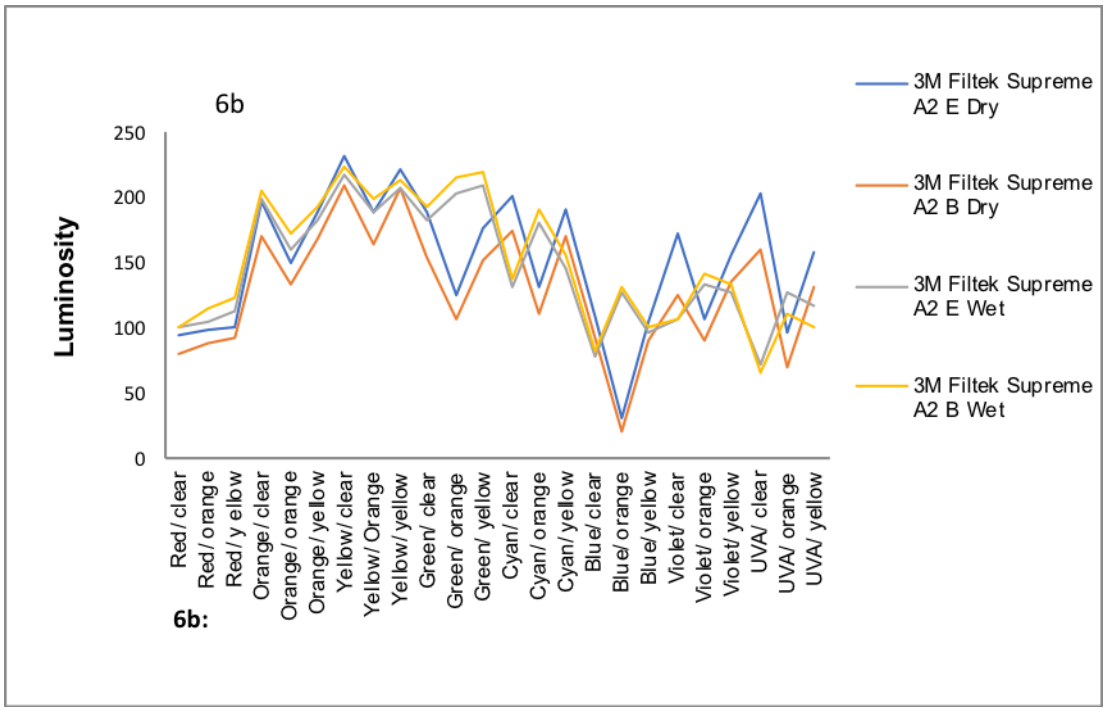


Figure 3. 6. Fluorescence emission of dry and wet samples of Vitablocs and 3M Filtek Supreme when excited with red (670 nm), orange (635 nm), yellow (585 nm), green (535 nm), cyan (470 nm), royal blue (450 nm), blue-violet (430 nm) and violet/UV-A (405 nm) light and viewed under clear, orange and yellow filters between posterior teeth and anterior teeth.

3.4. Discussion

The results of this study show the usefulness of fluorescence for identification of different types of tooth-coloured restorative materials. Fluorescence emissions have a longer wavelength than the excitation light source, which allows filters to be used to remove the excitation component reflecting from the sample surface, leaving only the fluorescence component to pass to the camera.

Natural human teeth exhibit fluorescence when illuminated under both broadband and narrow band light, with varying intensities depending on the excitation wavelengths. Ultraviolet light elicits green fluorescence while blue light elicits yellow fluorescence from healthy tooth enamel. Alterations in these patterns can be used to detect missing or decalcified tooth structure (in the case of dental caries), as well as the presence of a restorative material (51, 70, 167). Human enamel exhibits three distinct luminescence peaks in the regions of 350–360, 405–410 and 440–450 nm (31, 32). While fluorescence using excitation with ultraviolet light has been the most extensively examined, other wavelengths (including visible green and red) have also been used (1, 4, 6, 17, 36, 168). The present study indicates that there are variations in fluorescence patterns with all the wavelengths we used to compare restorative materials and natural tooth structure, but the greatest differences occur for excitation at 405 nm, which is on the boundary of the UV-A and visible (violet) light spectra. Thus, the first hypothesis of this study was confirmed, since the best wavelength for discrimination of tooth-coloured restorative materials, including composite resins, ceramics and hybrid restorative materials was found to be 405 nm. This aligns with results of previous studies analysing composite resin materials (49, 70) which showed that optimal excitation wavelengths for the composite resin materials used in their studies were in the range of 365–380 nm and 398 ± 5 nm, respectively.

Fluorescence assessments of teeth and restorative materials are facilitated when filters are used. These remove specific wavelengths of light, blocking shorter wavelength excitation light reflected from the sample surface, but allowing longer wavelength fluorescence emissions to pass. The use of filters enables selective detection of the various colours (169). In the present study, the combinations of excitation light and three different filters were assessed. The orange filter gave the maximum variance in emission patterns for any given excitation light source. This filter blocks ultraviolet, violet, blue and cyan light. By making some materials appear darker and others brighter, such filters aid in identifying different types of tooth-coloured restorative materials. Similar benefits but of lesser scale were seen with the yellow filter. The hybrid restorative materials included in this study exhibited bright pink fluorescence when illuminated with UV-A/violet light and viewed through the yellow filter.

Fluorescence properties of materials in the mouth can be affected by the ingress of moisture as well as by degradation over time. Some studies have reported that the fluorescence properties of composite resin materials can alter as the material ages (35, 170). When the fluorescence properties of dry and hydrated samples were compared in the present study, storage in distilled water for eight weeks at room temperature gave lower emissions. This difference could be due to several factors including greater scatter of fluorescence emissions (due to refraction by water) and quenching of fluorescence (from dissolved atmospheric oxygen in the water). There was no evidence of drift in the properties of materials when kept in

the dry state over 60 days after having been previously immersed in water, which indicates that the effects caused by sorption of water are reversible.

3.5. Conclusion

The present study suggests that fluorescence-based photography may be a useful adjunct for recognising types of tooth-coloured restorative materials. A fluorescence technique could be employed in routine dental examination as well as for forensic identification purposes. Certain restorative materials such as VITAEnamic, ormocers and glass-ionomer cements exhibit unique emission patterns, which makes their presence readily apparent. The intensity of fluorescence is influenced by hydration, since in general wet samples have less intense peak fluorescence emissions than those in the dry state. Further studies are needed to assess the accuracy of the fluorescence-based inspection approaches for detecting tooth-coloured restorative materials in the clinical setting.

Chapter 4 Fluorescent emission among the varied shades of tooth-coloured restorative materials

Additional material for this study is attached as Appendix A

This chapter has been accepted for peer review publication in the International Journal of Esthetic Dentistry (Appendix)

This chapter compared the fluorescence behaviour of varied shades of tooth-coloured restorative materials: among the shades of a particular brand type and between the shade groups of different brand types, and between different material and brand types of a particular shade.

4.1. Introduction

Tooth-coloured restorations are intended to replicate the optical properties of natural tooth structure, including the absorption, transmission, reflection and scatter of light (171). Likewise, these restorations attempt to mimic the fluorescence traits of natural tooth structure (172). Fluorescence is a photoluminescence phenomenon, where absorption of light is followed by the emission of light of a longer wavelength. Excitation of sound natural teeth with long wavelength ultraviolet (UV) light elicits visible blue emissions (36, 173), with the major contributors to this being the organic components, particularly amino acids such as tryptophan, which is why dentine fluoresces more intensely than enamel (18).

To attempt to replicate the inherent fluorescence properties of natural tooth structure, a range of additives are included in aesthetic restorative materials such as oxides of rare earth elements (europium, cerium and ytterbium) in the glass filler particles (133). Several studies have demonstrated the fluorescence behaviour of tooth-coloured restorative materials under UV light (47, 49, 62, 130, 171). The current range of tooth-coloured restorative materials includes ceramics, resin composites, glass ionomer cements, ormocers, and hybrid

materials such as polymer-infiltrated ceramics (for example, Vita Enamic™). When illuminated with monochromatic (laser) or near-monochromatic light, the fluorescence emitted varies according to the material type and, as a result, it is possible to differentiate tooth-coloured restorations from natural tooth structure (42, 70, 174, 175). This is important because when tooth-coloured restorations have been fabricated with a careful choice of layered materials, with correct translucency and optimal shade, they can be very difficult to detect by visual examination alone. Their ability to blend into the natural tooth structure makes it difficult to distinguish where the restoration margins are located — a point of great relevance when replacing restorations and when charting their presence. The use of non-invasive, non-damaging methods to reliably detect tooth-coloured restorations would be of value in everyday clinical dental practice as well as in forensic odontology (69, 174, 175).

The fluorescence properties of a particular tooth-coloured restorative material is likely to vary according to the shade of the material, as different tints will be used to create the final colour of the material. As this aspect has not been examined in any previous studies, the present study was designed to explore the fluorescent behaviour of different shades of selected contemporary tooth-coloured restorative materials when illuminated with violet light (405 nm wavelength).

4.2. Materials and methods

4.2.1. Materials

A total of 15 commercial direct tooth-coloured restorative materials was used in the study. They comprised resin composites Filtek™ Z250, Filtek™ Supreme XT, Filtek™ Bulk Fill Posterior (3M-ESPE Dental products, St. Paul, MN), G-aenial™ Posterior, Gradia®Direct Anterior Universal , Gradia®Direct X (GC Corp, Tokyo, Japan), TPH Spectra LV (Dentsply DeTrey, Konstanz, Germany), and Herculite™ Ultra (Kerr Corporation, 717 West Collins, Orange, CA 92867); glass ionomer cements (Fuji IX GP, Fuji II LC, GC Corp.), ormocers (Admira Fusion™, Voco, Cuxhaven, Germany) and ceramics (VITABLOCS® Mark II , VitaEnamic® (VITA Zahnfabrik, Bad Säckingen, Germany), and IPS EMAX (Ivoclar Vivadent AG, Liechtenstein).

4.2.2. Sample preparation

From the 15 materials, three samples were prepared for each shade of each material, giving some 111 differing shade sample sets in total (Table 4.1). For the ceramic materials, samples were prepared by slicing ceramic blocks with a precision diamond saw (IsoMet™ 1000, Buehler, Lake Bluff, IL, USA). For the remaining materials, samples of 10 mm diameter and 5 mm thickness were fabricated, in a standardised manner. Each direct restorative material was packed into a rigid plastic mould and then covered with a glass slide to obtain a

flat surface. For light-cured materials, samples were light cured for 60 seconds using a 470 nm LED curing light (Mini LED, Acteon Satelec, Merignac, France) in pulsed mode at a power density of 1250 mW/cm2. Once the samples had been retrieved from the mould, they were further cured for another 60 seconds. All samples were coded and stored in airtight containers at room temperature for 20 days.

Table 4. 1. List of materials used in the study

Group	Brand Name	Filler Type	Shade	Shade Type	Batch No	Manufacturer
1	Aura	Nanohybrid	E1	Enamel	141998	SDI Limited,
2			E3	Enamel	141998	Bayswater,
3			Db	Dentine	141998	Victoria,
4			DC2	Dentine	141998	Australia
5			DC3	Dentine	141998	
6			DC4	Dentine	141998	
7			DC5	Dentine	141998	
8			DC6	Dentine	141998	
9			DC7	Dentine	141998	
10	3M Filtek™ Z250	Microhybrid	A1	Enamel	N851947	3M ESPE Dental
11			A2	Enamel	N878736	Products, St
12			A3	Enamel	N875042	Paul, MN,
13			A3.5	Enamel	N876955	USA
14			B1	Enamel	N888384	
15			B2	Enamel	N846375	
16			B3	Enamel	N878050	
17	3M Filtek™ One Bulk Fill Restorative	Nanocomposite	A1	Bulk fill	N882564	3M ESPE Dental Products, St
18			A2	Bulk Fill	N880789	Paul, MN,
19			A3	Bulk Fill	N885211	USA
20			B1	Bulk Fill	N888384	
21			C2	Bulk Fill	N869835	
22	3M Filtek™ Supreme XTE	Nanofilled	A1	Enamel	N867907	3M ESPE Dental Products, St
23			A2	Enamel	N710711	Paul, MN,
24			A3	Enamel	N849778	USA

25			B2	Enamel	N862298	
26			D2	Enamel	N844004	
27	Herculite™ Ultra	Nanohybrid	A1	Enamel	6183867	Kerr Corporation,
28			A2	Enamel	6176290	Orange, CA,
29			A3	Enamel	6296114	USA
30			A3.5	Enamel	6189914	
31			A4	Enamel	6043798	
32			B1	Enamel	5876571	
33			B2	Enamel	5918661	
34			B4	Enamel	5472575	
35			C1	Enamel	5594037	
36			C2	Enamel	5994275	
37			C3	Enamel	5599410	
38			C4	Enamel	5375625	
39			D2	Enamel	5876575	
40			D3	Enamel	6000816	
41			D4	Enamel	5880856	
42	Dentsply TPH Spectra® LV	Nano-hybrid composite with pre-polymerized fillers	A1	Universal	1608112	DENTSPLY Caulk, Milford, DE, USA
43			A2	Universal	160918	
44			A3	Universal	161004	
45			A3.5	Universal	161005	
46			A4	Universal	1505182	
47			B1	Universal	1609293	
48			B2	Universal	1606303	
49			B3	Universal	170429	
50			C1	Universal	160519	
51			C2	Universal	1504271	
52			C3	Universal	150124	
53			C4	Universal	160705	
54			D3	Universal	1607271	
55			Db	Universal	1602041	
56			LG	Universal	1611142	
57			L	Universal	160820	
58			LB	Universal	161016	
59	G- aenial™ Posterior	Micro filled hybrid resin	P-A1	Bulk fill	1612061	GC Europe, Leuven,
60			P-A2	Bulk fill	1703171	Belgium
61			P-A3	Bulk fill	1704272	
62			P-A3.5	Bulk fill	1704111	
63	Gradia® Direct	Microhybrid	A1	Enamel	1512091	GC Europe, Leuven,

	Anterior					Belgium
64			A2	Enamel	1703293	
65			A3	Enamel	1606071	
66			A3.5	Enamel	1509172	
67			A4	Enamel	1703301	
68			B1	Enamel	1601141	
69			B2	Enamel	1509151	
70			B3	Enamel	1701121	
71			C3	Enamel	17202061	
72	Gradia® Direct X	Nanohybrid	A1	Enamel	1611241	GC Europe, Leuven,
73			A2	Enamel	1611252	Belgium
74			A3	Enamel	1705012	
75			A3.5	Enamel	1610212	
76			B1	Enamel	1702061	
77			B2	Enamel	1501191	
78			C2	Enamel	1703101	
79	Admira Fusion	Nanohybrid Ormocer	A1	Bulk Fill	1610380	VOCO
80			A2	Bulk Fill	1601121	GmbH,
81			A3	Bulk Fill	1712214	Cuxhaven,
82			A3.5	Bulk Fill	1707169	Germany
83			A4	Bulk Fill	1734371	
84			B1	Bulk Fill	1731442	
85			B2	Bulk Fill	1734362	
86			B3	Bulk Fill	1739336	
87			C2	Bulk Fill	1731307	
88			D3	Bulk Fill	1726469	
89	Fuji IX	Glass ionomer Cement	A2	Bulk Fill	1703141	GC Europe, Leuven,
90			A3	Bulk Fill	1703091	Belgium
91			A3.5	Bulk Fill	1703031	
92			B2	Bulk Fill	1703041	
93			B3	Bulk Fill	1704041	
96	Fuji II	Glass Ionomer Cement	A1	Bulk Fill	1601121	
97			A4	Bulk Fill	1705231	
98			B2	Bulk Fill	1705251	
99			B3	Bulk Fill	1705231	
100			C2	Bulk Fill	1705161	
101			B4	Bulk Fill	1705191	
102			D2	Bulk Fill	1705151	
103	VITABLOCS ® Mark II	Feldspar Ceramics	3M3C			VITA Zahnfabrik,
104			TM2C			Bad

105			OM1C	Säckingen, Germany
106	IPS EMAX	Lithium disilicate glass ceramic	HTA1	Ivoclar Vivadent AG, Schaan, Liechtenstein
107			HTA2	
108			LTA1	
109			LTA2	
110	VitaEnamic®	Hybrid Ceramic	2M2T	VITA Zahnfabrik, Bad Säckingen, Germany
111			2M2HT	

To record their fluorescence emissions, samples were photographed in a dark environment, using a customised digital single-lens reflex camera (Canon model EOS Rebel T2i/EOS 550D, Tokyo, Japan) under 405 nm illumination (spectral band width ±10 nm) in a standardised manner, as described previously (130). The camera settings were: aperture F2.8, shutter speed of 1/30 second, sensor sensitivity ISO 400, image colour depth 24 bit (8 bits for each colour channel, giving 16.7 million colours), image size of 18 megapixels. The camera was set to use the Adobe RGB 1998 colour space. The white balance was set to white fluorescence light (temperature 4000K) with white balance correction of zero. The samples were placed at a constant distance of 200 mm from the lens. The excitation light was directed at an angle of 90◦ to the sample surface. The images were recorded through a long pass filter, which removed the excitation light. This ensured that data for luminosity could only be from fluorescence emissions.

4.2.3. Analysis

Images were analysed using the histogram feature of Adobe Photoshop™ Creative Cloud 2018 software. Within Photoshop™, the histogram tool was used for analysis of the pixel data. The colour coordinates were recorded for red (R), green (G) and blue (B) colour channels, and for luminosity. The histogram values ranged on a scale of 0–255 for each colour coordinate, since the images were recorded in 8-bit colour.

The mean values of the red, green, blue and luminosity channels were compared between shades of the same material. Data for luminosity were used to compare the same shade for differing materials. The average of 3 samples was used for data analysis. The statistical significance of the differences was determined using one-way ANOVA or the Kruskal Wallis test as appropriate, with post-hoc Tukey or Wilcoxon tests. The threshold for significance was set at the 5% level.

4.3. Results

4.3.1. Comparison of different shades of the same material

Within the range of direct tooth-coloured restorative materials studied, all showed similar patterns in the change in luminosity across the shades of any given material. The fluorescence luminosity decreased steadily from A1 to A4 shades, and then increased dramatically for the B1 shade, which had the highest values

(Figure 1). In general, blue colour channel values were greater than green or red values, with red being the lowest overall (Figure 1).

Within any one material, there were significant variations in fluorescence luminosity amongst different shades ($p < 0.05$, Table 4.2,). The greatest magnitude of difference within the one material was seen for composite resins, followed by ormocers, and then glass ionomer materials. In a similar way, there were also significant variations among the red, green and blue colour channel values for all materials, with the exception of Fuji IX, where the difference failed to reach the threshold for significance ($P = 0.056$), and for Vita Enamic® ($P = 0.112$), for which only three shades were evaluated.

4.3.2. Comparison between material types

For the same labelled shade, there were statistically significant differences between different classes of material ($p < 0.001$ for all comparisons), with the single exception of ceramic versus hybrid ceramic materials ($p = 0.50$) where again only few shades were evaluated.

Table 4. 2. P values from Kruskal Wallis tests comparing variations between shades

Material	Red colour	Green colour	Blue colour	Luminosity
Filtek™Z250	0.005	0.004	0.004	0.004
Aura	0.014	0.001	0.018	0.001
Herculite™Ultra	0.008	<0.0001	0.002	<0.0002
TPH Spectra	<0.0002	0.0001	<0.0002	0.0001
GC Fuji IX	0.006	0.005	0.006	0.005
GC Fuji II	0.015	0.004	0.003	0.003
G-aenial® P	0.001	0.019	0.015	0.016
Gradia®DirectA	0.002	0.003	0.002	0.002
Gradia®DirectX	0.004	0.004	0.004	0.004
Filtek™	0.024	0.009	0.011	0.010
Filtek™ Supreme XTE	0.029	0.020	0.009	0.013
Admira Fusion	0.002	0.002	0.004	0.002

4.4. Discussion

This study provides insight into the variations in fluorescence properties between materials, and within the same brand of material, according to shade. Variations in the colour and intensity of fluorescence emissions were found, with the greatest variation being found within particular materials for composite resins, and then reducing in order for ormocers, glass ionomer materials, ceramics and hybrid ceramics. The large range of materials and shades that was tested extends the information gained from past studies (49, 62, 171, 176), and documents the nature of the variations in terms of the intensity and colour of fluorescence under violet light excitation.

The colour of a tooth as perceived by human eye is the result of interaction of ambient light with the chromatic, geometric and optical features of the dental

tissues, (37) including reflection, scattering, and fluorescence. In the present study, a long pass filter was used so that only fluorescence emissions would be analysed. Differences in fluorescence elicited by 405 nm light can be very useful for identifying tooth-coloured restorations (130). It can reveal differences in the substrate (that is, natural tooth structure versus restorative material). Such differences would not be evident under natural ambient light (22, 171).

Because ageing of resin based composite materials may notably change their fluorescence properties (35), standardised storage time and conditions were maintained in the present study. Further work is needed to assess how fluorescence properties may be affected by adverse environmental conditions, such as elevated temperatures (as would occur during a fire).

Many past studies that have assessed the fluorescence properties of resin-based composite materials have used spectral analysis (35, 49, 54, 171). While this is an elegant method, it cannot be used easily in the field or clinical setting, unlike the approach of using violet light fluorescence (405 nm $\pm$ 10 nm) which is deployed easily (130) and provides quantitative data from image analysis.

Fluorescence emissions of dental restorative materials arise mostly from their most superficial layer (177). This is why enamel shades and bulk fill/universal restorative materials were chosen for the present study. It is important, however, to note that while dental restorative materials fluoresce from their surface, natural

tooth structure shows the opposite pattern, since dentine fluoresces under violet light more intensely than enamel.

When the fluorescence emissions of restorations are accurately matched to the natural tooth colour, their clinical recognition can be challenging. From clinical, epidemiological and forensic perspectives it is desirable that tooth-coloured restorative materials fluoresce either more dimly or brightly than the adjacent tooth structure when particular light sources are used, to allow them to be identified readily.

The present study reveals considerable variations of fluorescence properties within the same labelled shade for different restorative materials. Some materials fluoresce more brightly than tooth structure, whilst others fluoresce less brightly (130). The fluorophores added to the fillers of these materials provide the fluorescence properties. The composition and concentrations of these fluorophores is not disclosed by manufacturers. As demonstrated in the present study there can be significant variation in fluorescence between materials from the same supplier, in some cases but not in others. The three Filtek materials Bulk Fill Filtek ™, Filtek Z250 ™ and Filtek™ Supreme XTE differ in their fluorescence properties, even though they use fundamentally the same filler system. Bulk Fill Filtek™ has an additional component comprising 100 nm particles of ytterbium trifluoride which are included as a radio-opacifier. In contrast to this, three composite resins from the same manufacturer (GC)

(Gradia® Direct, Gradia® Direct X and G-aenial® Posterior) that have known variations in filler composition showed no statistically significant variations in fluorescent emissions between them (Table 4.3). According to the manufacturer, the main difference between Gradia® Direct and Gradia® Direct X is nanoparticles of a lanthanide compound added to Gradia® Direct X as a radio-opacifier, while G-aenial® Posterior has an additional fluoro-aluminosilicate included as a radio-opacifier. Together, these results highlight the importance of the choice of fluorophores in determining the fluorescence properties. Colouring agents vary in concentration between different shades of the same material, and it can be concluded that some colouring agents must also affect fluorescence. Finally, it is possible that the fluorophores are the filler particles themselves. Fillers may vary between materials of the same category by factors such as size and shape, as well as composition.

Table 4. 3. Table representing the p values, showing the variation between two material types

Material	Filtek Z250	Aura	Herculite Ultra	TPH Spectra	G-aenial	Gradia Anterior	Gradia Direct X	Filtek™	Filtek Supreme XTE
Filtek Z250		0.79	0.013*	0.00*	0.00*	0.00*	0.00*	0.00*	0.045*
Aura			0.28	0.00*	0.00*	0.00*	0.00*	0.09	0.301
Herculite Ultra				0.00*	0.00*	0.00*	0.00*	0.085	0.75
TPH Spectra					0.00*	0.00*	0.00*	0.002	0.00*
G-aenial						0.62	0.91	0.00*	0.00*
Gradia Anterior							0.39	0.00*	0.00*
Gradia Direct X								0.00*	0.00*
Filtek™									0.038

*p value significant at 0.05

4.5. Conclusion

The fluorescence emissions vary considerably amongst different shades of the same material, and between different materials that are labelled as having the same shade. Under violet light, the greatest emissions occur for the lightest shade (for example, B1), reducing from A1 to A4, and from B1 to B3.

Figures for this study are attached as Appendix A

Chapter 5 Comparison of three diagnostic methods in identification of tooth-coloured restorative materials

Additional material for this study is attached as Appendix B

This chapter is published as a peer reviewed article (Appendix F):

Kiran R, Chapman J, Tennant M, Forrest A, Walsh LJ. Detection of tooth-coloured restorative materials for forensic purposes based on their optical properties: an in vitro comparative study. J Forensic Sci 019;64:254-9. https://doi.org/10.1111/1556-4029.13851

This chapter compared the sensitivity and specificity of identifying contemporary tooth-coloured restorative materials using the FAIR method, or DiFOTI with the DIAGNOcam™ , and conventional visual and tactile examination.

5.1. Introduction

The availability of aesthetic restorative materials with colour and optical properties close to those of human teeth, as well as novel techniques such as layering of materials and the use of tints and opaque stains allows dentists to produce restorations that mimic the optical properties of natural tooth structure (178, 179). Using such materials helps meet the aesthetic demands of patients for restorations which cannot be detected with the unaided eye, but at the same time raises the challenge of how clinicians can reliably detect and record the presence and extent of such restorations. Accurate charting of restorations is important both in living patients (for their ongoing clinical care), and for epidemiological purposes, as well as in deceased persons who have not been identified (Table 5.1).

The traditional method of detecting tooth-coloured restorations is visual inspection, supplemented in some cases with radiographs. Visual inspection has poor sensitivity, meaning that the examiner cannot distinguish the restoration from the surrounding normal tooth structure (43, 69, 82, 87).

Table 5. 1. Advantages of accurately detecting tooth-coloured restorations and differentiating them from natural tooth structure

Clinical patient care	**Forensic odontology**	**Epidemiology**
Improved treatment planning (pattern of past restoration, recognition of sites for wall lesions of dental caries)	Deceased victim identification based on comparison of their teeth with dental records, especially when other methods of victim identification are not possible.	Correct scoring of restored deciduous and teeth
Identification of excess material near margins and overhangs		
Selective removal of restorations during restoration replacement.		

Optical methods based on the interaction of light with hard tissues may aid in distinguishing restorations from natural tooth structure. Optical methods in current clinical use include light-induced fluorescence, fibre-optic trans-illumination (FOTI) and digital imaging fibre-optic trans-illumination (DiFOTI), as in the DIAGNOcam™. The interaction between the light and the tissue is both wavelength and time/rate dependent. In the case of fluorescence, light of a longer wavelength than the illuminating light is produced.

With both FOTI and DiFOTI, patterns of reflection, scattering, transmission, and absorption of light are important for discriminating between internal structural features of teeth, including the identification of restorations (93). FOTI typically uses broad-spectrum high-intensity white light, which gives strong scattering

effects, while DiFOTI systems such as the DIAGNOcam™ use coherent near-infrared light from a diode laser. Compared with visible light, near infrared light penetrates better through sound tooth structure and gives less scatter (94–96). While DiFOTI has been used to detect lesions of dental caries on approximal enamel surfaces, it is feasible to apply the same principles to the detection of tooth-coloured restorations, because these transmit light differently to natural tooth structure.

Since most tooth-coloured restorative materials exhibit distinctive fluorescence properties when compared to natural tooth structure upon illumination with violet light, **Fluorescence Aided Identification of Restorations** (FAIR) should be possible. While several studies have examined the fluorescence properties of resin composite restorative materials (49, 70, 78, 92, 180), little is known regarding the relative performance of FAIR versus conventional inspection or DiFOTI, for the specific purpose of identifying tooth-coloured restorations. Accordingly, the aim of the present study was to compare the diagnostic reliability and validity of DiFOTI and FAIR with conventional visual and tactile examination for identifying tooth-coloured restorations. The study was performed under defined laboratory conditions with consistent lighting and in the absence of confounding factors such as saliva or dental plaque biofilms on the teeth. For FAIR, the approach used was illumination with 405 nm wavelength violet light, accompanied by viewing the sample through a long pass filter which permits light of greater than 520 nm in wavelength to pass (130).

The study tested two hypotheses: (1) that differences in light transmission through natural tooth structure versus various tooth-coloured restorations would aid in their identification using DiFOTI; and (2) that FAIR using 405 nm violet light would enhance the identification of tooth-coloured restorative materials, to give more reliable identification than either conventional examination or DiFOTI.

5.2. Materials and Methods

5.2.1. Extracted teeth

The study was conducted with human ethics approval (Central Queensland University approval number H15/03-035). A total of 90 extracted human permanent teeth were collected following extraction procedures, and stored in 1% thymol solution (Sigma Aldrich, Australia) before being cleaned of any surface deposits by mechanical debridement using hand scalers, and then sterilised by exposure to gamma radiation (20 kGy).

5.2.2. Preparation of tooth models

The extracted teeth were used to fabricate three sets of models for both maxillary and mandibular arches. The teeth were mounted in anatomical order in typodonts, which had a soft silicone material to replicate normal gingival soft tissues [model D95SDP-TRM.670, Nissin Dental products Inc, Kyoto, Japan]. Once populated with teeth, the six models were stored in distilled water until used, to ensure hydration.

To simulate clinical conditions, each set of maxillary and mandibular models was placed into a phantom head dental mannequin [Columbia Dentoform, Long Island City, NY, USA], and the teeth polished for 60 seconds using 1200 rpm with a rubber cup and dental prophylaxis paste [ClinPro™ prophy paste, 3M ESPE, Minneapolis, MN, USA] in a low speed handpiece (Model Ti-Max X25L, NSK, Kanuma, Japan). The teeth were then rinsed thoroughly with distilled water.

Cavity preparations were performed according to existing carious lesions that were present in the extracted teeth (n = 33 teeth). Either Class I or Class II (Black's classification) cavities with dimensions of 3.0 mm width and 2.0 mm depth were prepared in the caries-free teeth (n = 22) using water-cooled diamond burs in a high-speed dental handpiece (NSK S-Max M600L).

The selection of the shade of each material corresponded to that of the individual teeth, with shade matching being performed under diffuse natural light. Restorations were then placed according to the standard clinical protocols for each material type. The tooth-coloured restorative materials listed in Table 5.2 were then used to restore the 55 mounted teeth. The materials comprised tooth-coloured resin composites, ceramics and hybrid restorative materials such as ormocers, Vita Enamic™ and resin reinforced glass-ionomer cements.

Table 5. 2. Restorative materials used

Brand Name	Shade	Batch

			Numbers
Vitabloc	Vita	1M2C	33450
Vita Enamic	Vita	2M2T	52210
Filtek Supreme XTE	3M Espe	A2E	N710711
3M Filtek Supreme XTE	3M Espe	A2 E/B	N767183
Gradia	GC	A3.5	1509172
Fuji VIII	GC	A2	1504021
Fuji IX	GC	A2	1511101
Amaris	Voco	O1	1452078
Admira Fusion	Voco	A2	1525583
Admira	Voco	OA2	1527294
Grandio	Voco	A2	1445171
Grandioso	Voco	A2	1512560
Herculite Ultra	Kerr	A2D	5582394
Herculite XRV	Kerr	E	5580988
Herculite XRV	Kerr	D/E	5580988
Spectrum TPH	Dentsply Sirona	A3	151020

Manufacturer details: Vita Zahnfabrik, Bad Sackingen, Germany; 3M ESPE, Minneapolis, MN, USA; GC Corp, Tokyo, Japan; Voco, Cuxhaven, Germany; Kerr, Uxbridge, UK Dentsply Sirona, York, PA, USA

Once polymerisation was completed, the direct tooth-coloured restorations were polished using impregnated rubber polishers (Identoflex, Kerr, Orange, CA, USA; and Sof-Lex™ Finishing/Polishing Discs and Strips, 3M ESPE, Minneapolis, MN, USA). The restored teeth were then stored in distilled water until needed and between examinations. The dental clinician who placed the restorations was not involved in the subsequent phases of the study involving detection of restorations.

5.2.3. Examination protocol

The models containing unrestored and restored teeth were mounted in a phantom head dental mannequin. The teeth were kept in a moistened state during examination by periodic rehydration with water. Four clinicians (two dentists and two oral health therapists) served as the examiners. They were provided with written instructions for the examination protocol, and each received training in the use of the DIAGNOcam™ and the FAIR method. No time limit was placed on examinations. The examiners were instructed not to discuss the findings among themselves for the duration of the study.

Each examiner charted all teeth for filled and unfilled surfaces using each of the three diagnostic methods: (1) conventional visual and tactile examination; (2) DIAGNOcam™; and (3) FAIR. The examinations were conducted using the three techniques in the same order on every occasion. The examiners repeated the charting using the same protocol after four weeks, to calculate intra-examiner

variability. In total, each examiner produced 18 sets of records (3 sets of models x 3 methods x 2 repetitions).

Conventional visual and tactile examination was performed using a double-ended sharp explorer, mouth mirror, and three-way triplex air/water syringe, using white (daylight colour temperature) light from a simulator-mounted dental LED lamp (white HB-LED dental light A-dec, Newberg, Oregon) with an intensity of 30,000 lux. DiFOTI examination was undertaken using the DIAGNOcam™ (KaVo, Biberach, Germany) according to the manufacturer's recommended method for transillumination for dental caries diagnosis. Examination using FAIR was conducted with the aid of a mouth mirror and light from a light emitting diode (LED) array (9 LEDs), which had an emission wavelength of 405 ± 10 nm (model SEFL33UV-405, SE Electronics, Shanghai, China). The LED was attached to the handle of the dental unit light (Figure 5.1) so that the examiner could adjust the incident light in a similar manner to a conventional dental lamp. The examiners wore orange-coloured protective glasses which filtered out the violet incident fluorescent light and any reflections but allowed fluorescence emissions to pass through (long pass filter cutoff $\lambda \geq 520$ nm). When using FAIR, other light sources were turned off in the examination room. This method was adopted as it could be applied clinically without changes to the regular dental surgery setup.

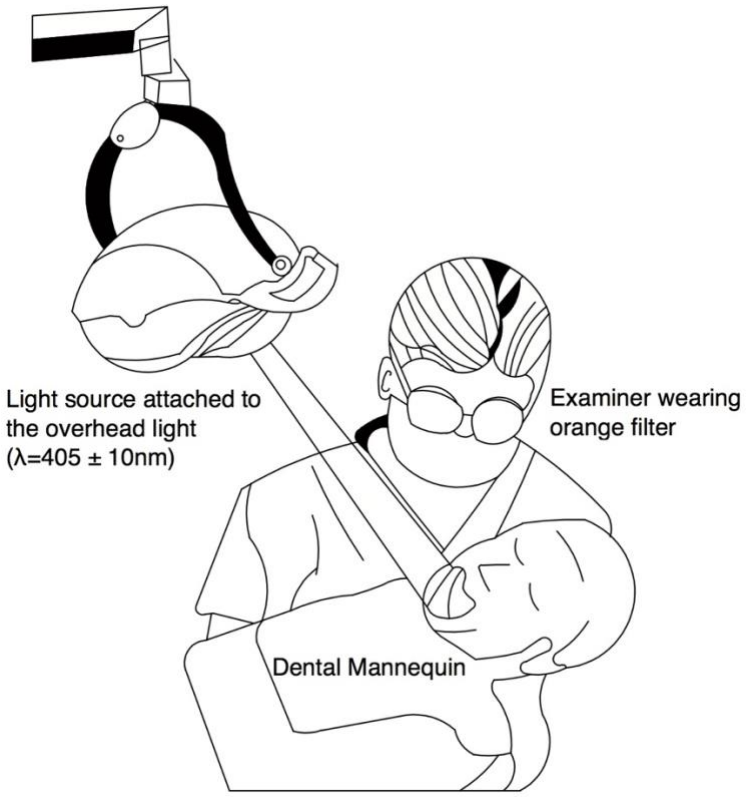


Figure 5. 1. Clinical simulation setup for FAIR

5.2.4. Statistical analysis

Statistical analyses were performed using R version 3.3.1, Microsoft Excel®, or SPSS version 19 software, with the threshold set at the 5% significance level. For each method, the sensitivity and the specificity were calculated with a 95% confidence interval, using Pearson's chi-square test for association. The positive and negative likelihood ratio was calculated to assess the efficiency of each method, but since this analysis resulted in multiple values, only the highest values are presented in the results. The variations between the methods and the intra-/inter-examiner variations were evaluated using a two-way ANOVA, and statistical significance was determined by the Tukey post-hoc test. Receiver operating characteristic (ROC) curves were determined. These curves reflect

how well a given test can distinguish between a true condition and a predicted condition, since the greater the area under the curve, the better the test. Finally, the average sensitivity and specificity of all three methods were determined, and the inter- and intra- examiner agreement evaluated using the kappa statistic.

5.3. Results

The sensitivity of detecting tooth-coloured restorations using FAIR was 95% (confidence interval 92%–97%), which was significantly higher than both conventional examination (71%) and DiFOTI (82%). The DiFOTI method using the DIAGNOcam™ detected 10% more restorations than did the conventional examination and gave slightly higher true positive predictive values. The specificity was higher with FAIR (97%) than conventional examination (82%) and DiFOTI (82%). Summary data on the sensitivity and specificity of all three methods are presented in (Figure 5.2). A representative image showing FAIR is presented in Figure 5.3, which shows two set of models when illuminated under white light and under violet (405 nm) light.

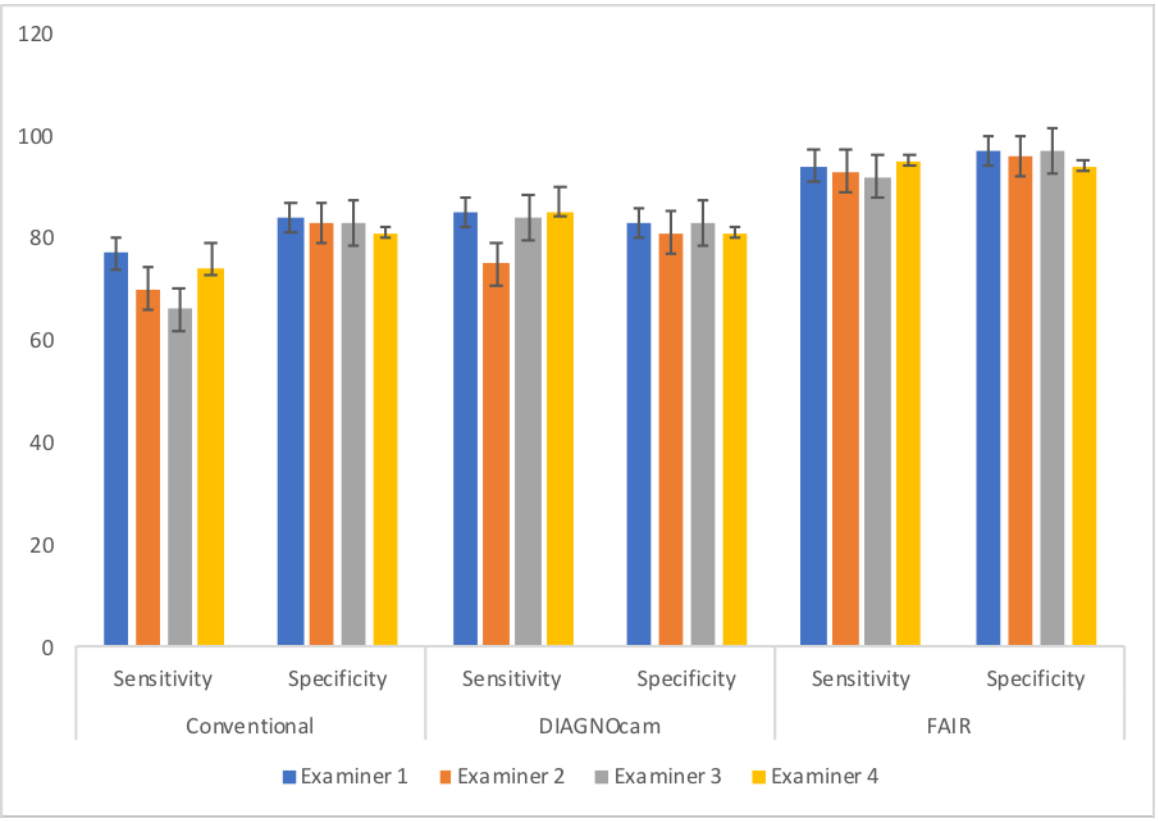


Figure 5. 2. Summary of the diagnostic performance of FAIR and DIAGNOcam™ versus conventional examination for identifying tooth-coloured restorations.

The positive likelihood ratio of detecting tooth-coloured restorations was 33 for FAIR, 4.81 for conventional examination and 4.84 for DiFOTI. The corresponding negative likelihood ratios (for identifying unfilled surfaces correctly) were 0.08, 0.2 and 0.41, respectively. Based on the classification table for sensitivity and specificity (Table 3), FAIR was the best and most reliable method for detecting tooth coloured restorations.

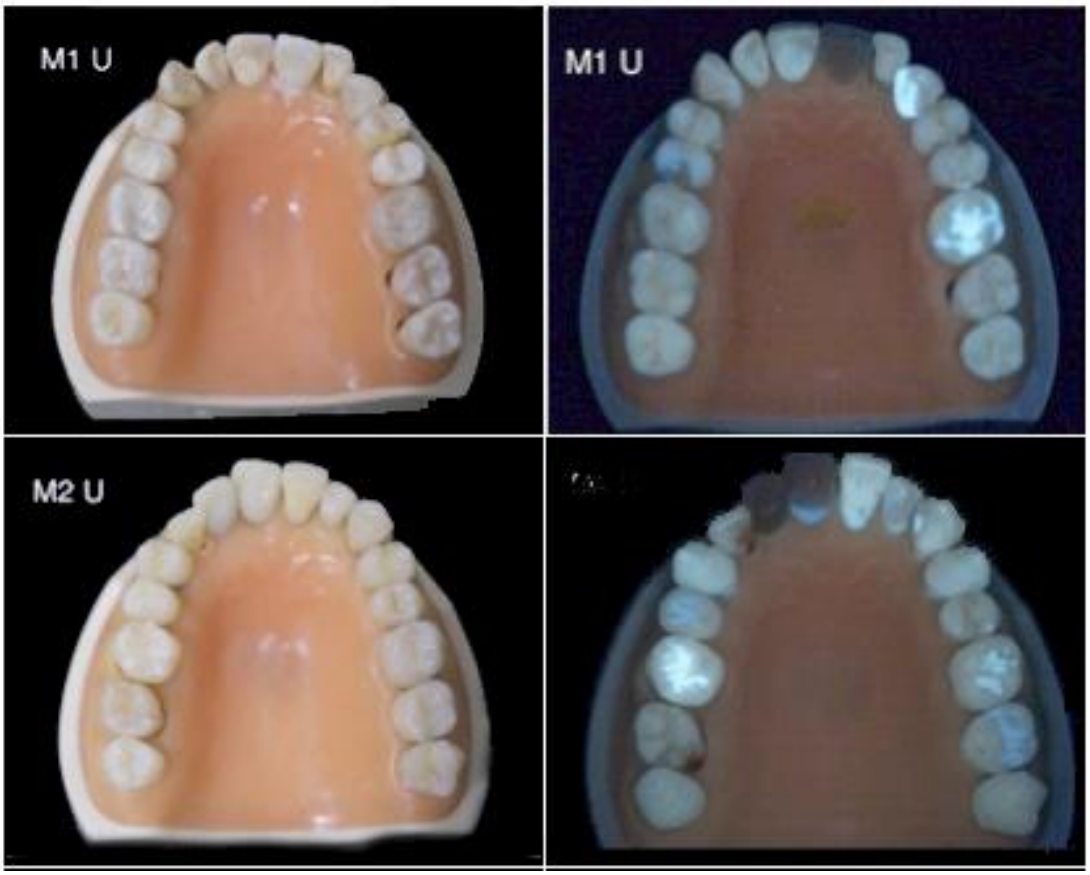


Figure 5. 3. Occlusal view of 2 different models when each is examined under white light and under violet 405 nm light. M1= Model1, M2= Model 2, U= Upper jaw, L=Lower jaw.

Table 5. 3. Positive likelihood ratio (PLR) and negative likelihood ratio (NLR)

PLR	NLR	Results
>10	<0.1	Large/Better or useful test
5-10	0.1-0.2	Moderate/Often useful test
2-5	0.20-0.5	Small/Sometimes useful test
1-2	0.50-1.0	Very small/Rarely useful test

Any test with a PLR of more than 10 and an NLR of less than 0.1 is considered as the better/more reliable diagnostic test. A test with a PLR value less than 2 and a NLR more than 0.5 is considered the least reliable test.

FAIR was found to be the superior and most reliable method for detecting tooth-coloured restorations. This was in keeping with the receiver operating characteristic (ROC) curves, which showed that FAIR had the greatest area under the curve.

In terms of variation between and within examiners, using a two-way ANOVA test, no statistically significant inter/intra-examiner variations for identifying filled surfaces ($p = 0.381$) were observed. However, there were significant differences between the three methods in detecting filled surfaces ($P < 1.27e^{-09}$) (Figure 5.4) . The Tukey post-hoc test revealed significant differences between FAIR and conventional examinations, and between DiFOTI and FAIR. However, there was no significant difference between the conventional examination method and DiFOTI.

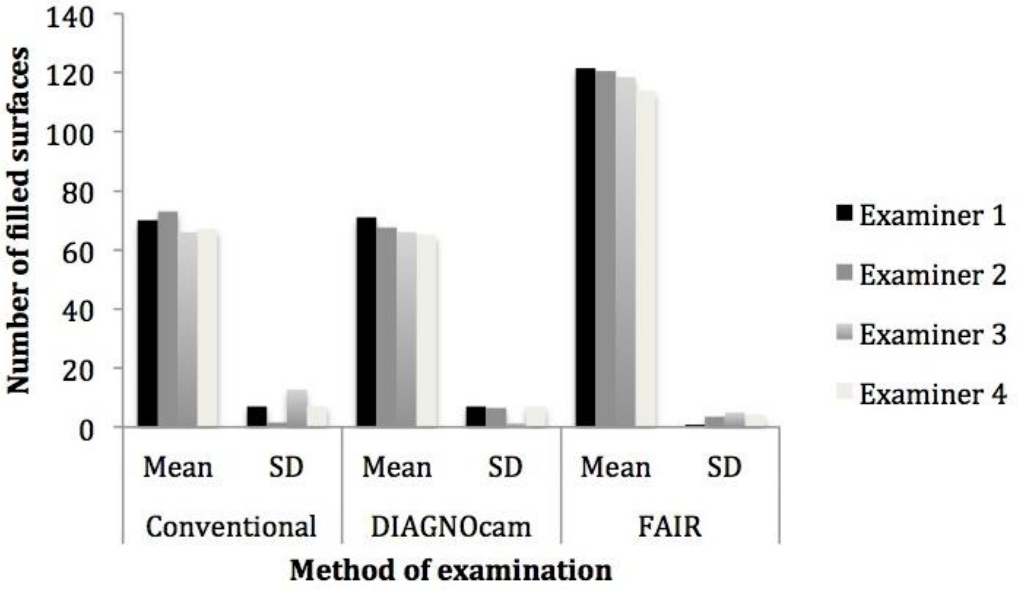


Figure 5. 4. Comparative analysis of the three methods for identifying filled surfaces

The intra-examiner agreement for FAIR (mean kappa 0.85) was significantly higher than that for DiFOTI or for conventional examinations (mean kappa 0.56 and 0.54 respectively). Inter-examiner agreement was greatest for FAIR (kappa 0.82), and less for DiFOTI (0.56) and for conventional examination (0.54).

5.4. Discussion

This study provides several insights into the performance differences of methods used to identify tooth-coloured restorations. FAIR showed improved sensitivity when compared with both conventional examination and with DiFOTI. Likewise, the specificity for FAIR was superior to both other methods. FAIR gave significantly fewer false positive results (6.6) compared to DiFOTI (15.5) or

conventional examination (27.1). It is possible that the estimation of specificity could be excessive because of the inclusion of many sound surfaces in the sample material where the total number of surfaces inspected was 450, out of which number of restored surfaces were 131 and unrestored surfaces were 319 (181). Nevertheless, the favourable results for FAIR indicate its potential for use in clinical practice to aid identification of tooth- coloured restorations.

Currently available tooth-coloured restorative materials fluoresce with varied intensities based on their composition, making them appear different to the adjacent tooth structure. Restorative materials do not exhibit metamerism under all wavelengths of light (49, 62, 182). In the present study using FAIR, the restorative materials that were most frequently missed were 3M Filtek Supreme XTE™ (which fluoresces similarly to tooth structure) and VOCO Admira™. FAIR may be useful for showing the extent of the restoration when removing old restorations in cases of repair, to prevent unnecessary removal of the tooth structure. FAIR can facilitate the discrimination of two different brands of materials used in restoring the same tooth as these fluoresce differently (Fig. 5.5).

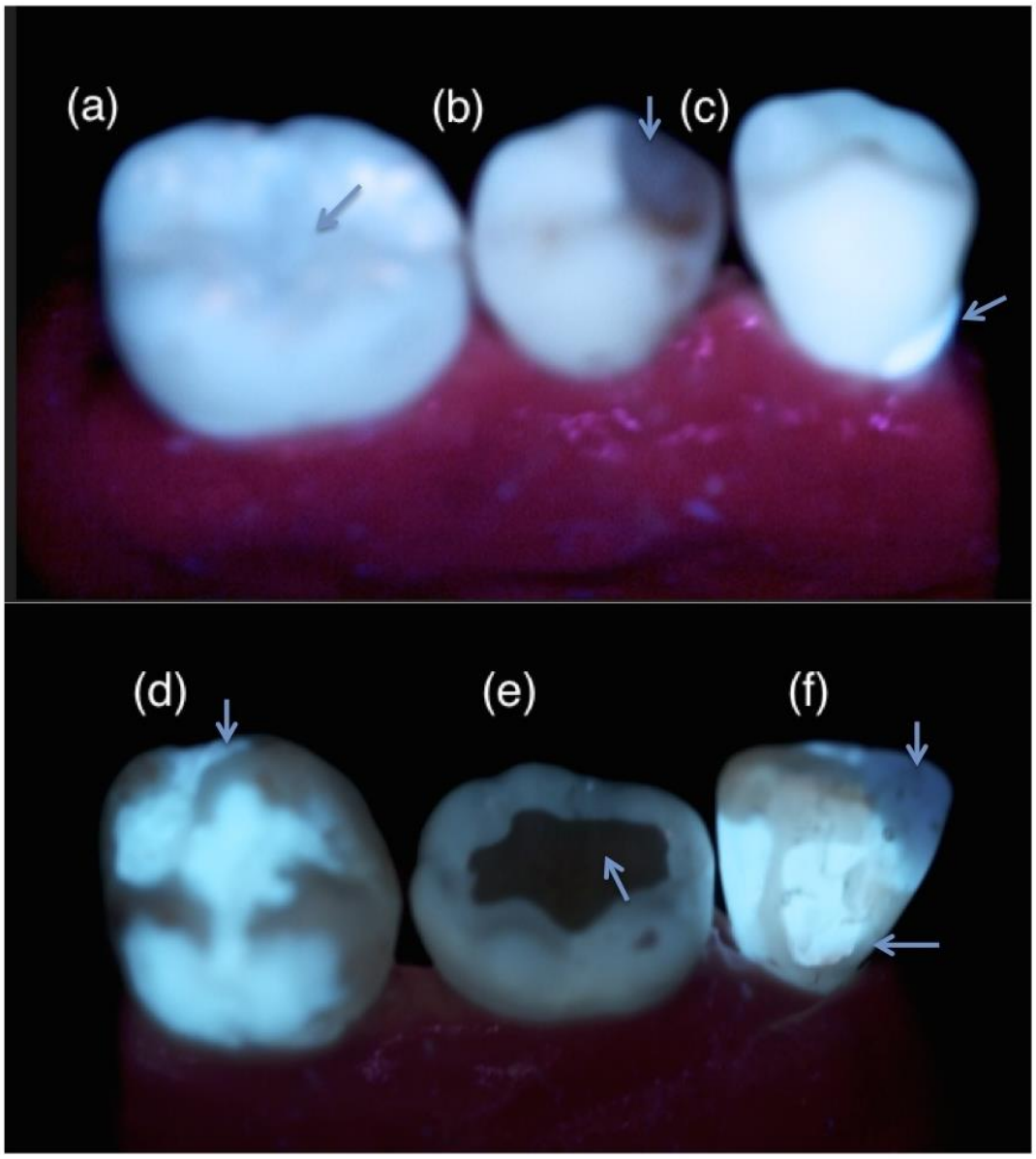


Figure 5. 5. Fluorescence emissions from different restorative materials when illuminated with UV-A light. 5a 3M Filtek Supreme XTE™; 5b Voco Admira™; 5c Admira Fusion; 5d Admira Fusion; 5e. Vita Enamic™ , 5f Admira Fusion (bright fluorescence) and Herculite Ultra (blue fluorescence).

DiFOTI was better than conventional examination for detecting filled surfaces (both for sensitivity and specificity). DIFOTI is based on differential patterns of light scatter (93, 183). Scattering events are wavelength sensitive (184). The DIAGNOcam™ employs near-infrared (NIR) light since enamel is transparent, while dentine scatters more strongly due to presence of water (94). Restorations scatter light and appear dark, and the extent of this varies according to the material composition (Figure 5.6). When a restorative material scatters light in a manner similar to dentine it is harder to detect. Most ceramic-based restorations such as inlays and onlays absorb near-infrared light and appear darker, making them easy to identify. Full crown restorations in ceramic-based materials obscure the normal pattern of translucent enamel and underlying darker dentine. This can be subtle and therefore may be overlooked.

In the present study, the positive likelihood and negative likelihood ratios were greatest for FAIR, making it the best of the three diagnostic methods used for detecting the tooth-coloured restorations. However, use of magnification and/or an LED headlight when examining or removing the restorations might affect the difference in results between the conventional and FAIR method and therefore further studies comparing magnification with headlight and FAIR in vivo are needed. Even though the time needed for examination was not recorded in this study, all four examiners stated that using fluorescence made examinations seem quicker and easier to undertake.

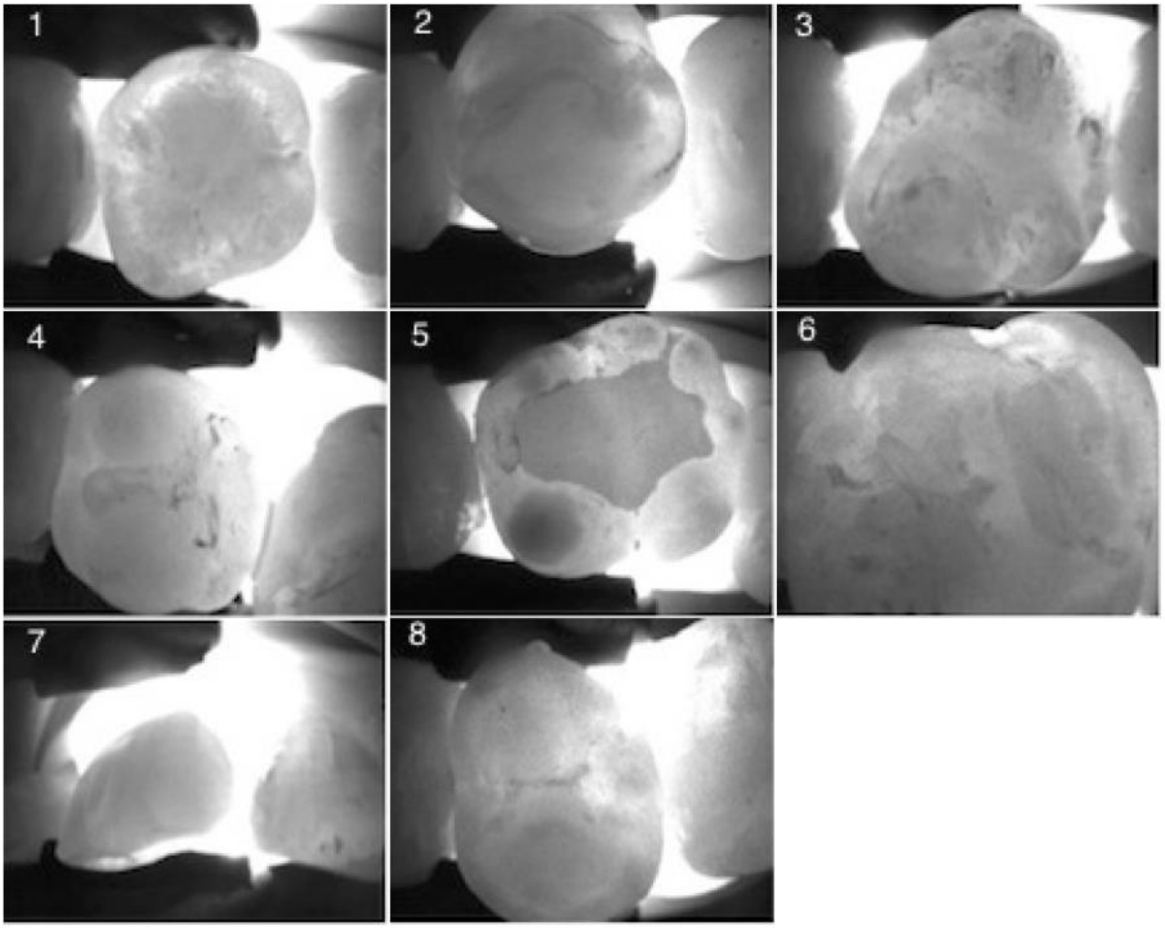


Figure 5. 6. DIAGNOcam™ images of teeth restored with different tooth-coloured restorative materials. 1 Natural tooth showing the translucency of enamel to near infrared light; 2 Occlusal restoration with Voco Amaris™; 3. Mesio-occlusal restoration with Voco Admira™; 4. Disto-occlusal restoration with Grandio™; 5. Occlusal restoration with Vita Enamic™ 6. Occlusal restoration with Herculite Ultra™; 7. Full crown using Vitabloc™; 8. Distal restoration with Voco Amaris™.

5.5. Conclusion

For detecting tooth-coloured restorations, FAIR was superior to DiFOTI and conventional examination methods. This method is simple to use and low in cost. DiFOTI improves the identification of occlusal and interproximal restorations in posterior teeth over conventional visual and tactile examination but is a more complex approach to use.

Chapter 6 Guided selective removal of composite resins

This chapter is published as a peer reviewed article (Appendix G)

Kiran R, Chapman J, Tennant M, Forrest A, Walsh LJ. Fluorescence-aided selective removal of resin-based composite restorative materials: An in vitro comparative study. J Esthet and Restor Dent. 2019 Oct 16. DOI: 10.1111/jerd.12536

This chapter compared the accuracy of the FAIR method in guided selective removal of resin based composite restorative materials with conventional white light illumination. It also assessed the amount of loss of tooth structure, restorative material remnants and time needed to effectively remove the restoration.

6.1. Introduction

Since their introduction in 1968, resin-based composites (RBC) have been the material of choice for conservative anterior aesthetic restorations (99). With progressive improvements over time in their physical properties, RBC have now become a common material used to restore posterior teeth (100, 101, 185, 186). In many dental practices, RBC have replaced amalgam restorations as the standard material choice for posterior intra-coronal restorations (102, 185, 186). Acceptable survival rates have been reported, with an annual failure rate of 1% to 3% (103).

A range of situations will arise where an RBC restoration requires replacement or repair. Replacement of failing restorations constitutes the majority of operative dentistry procedures in clinical practice (104-106). Reasons for replacing RBC restorations include secondary caries, marginal discolouration, wear, and bulk fracture (106–109, 187). When replacing an existing restoration, there is a high probability that a significant amount of tooth structure will be lost. Unnecessary removal of healthy tooth structure weakens the tooth and reduces its long-term outlook. A particular challenge when repairing or replacing RBC is that it is

difficult to differentiate the restorative material from tooth structure, as they can have a very similar colour. The same issue arises with cements for on-lays and crowns that are based on RBC. Thus, methods that facilitate the selective removal of RBC would be useful for clinical practice.

One approach is to exploit differences in the fluorescence properties of tooth structure versus restorations. Fluorescence diagnosis has been used to discriminate between tooth structure and tooth-coloured restorative materials (78). The choice of suitable wavelengths for fluorescence diagnosis includes ultraviolet (UV) light and visible light (36, 168). Natural healthy tooth structure emits strong fluorescence emissions when illuminated with violet light (405–410 nm) and blue light (440–450 nm) (31, 32). The primary fluorophores responsible for these emissions are organic components of the hard tissue (26). Different brands of RBC vary considerably in their fluorescence properties (130). The resin and filler components of tooth-coloured RBC typically do not fluoresce, rather particular organic fluorophores and rare earth oxides are added to the material (133).

When applying violet light (405 nm) and viewing teeth under a long pass filter, natural tooth structure will have a blue-green fluorescence, while individual tooth-coloured RBC may show stronger or weaker emissions of the same colour, or emissions of different colours (130). This allows for fluorescence-aided identification of restorations (FAIR). This method is based on the principle that

teeth and restorations may match under white light but will differ in appearance when viewed under specific wavelengths of light (37, 49).

Recent studies have demonstrated that FAIR using 405 nm light improves the ability of clinicians to identify RBC restorations, compared with conventional visual and tactile examination under white light (174, 188). Following the same logic, the present study was designed to compare the FAIR method with conventional white light illumination for the selective removal of tooth-coloured RBC restorations. The study assessed the extent of loss of tooth structure, the amount of residual material, and the difference in the time taken to remove the restorations with both methods.

6.2. Materials and methods

The study was conducted with human ethics approval (approval number H15/03-035).

6.2.1. Teeth

A total of 60 extracted human permanent posterior teeth devoid of any pathology were stored in a solution of 0.1% thymol solution (Sigma Aldrich, St Louis, MO, USA) at room temperature. After removal of all surface deposits by mechanical debridement using hand scalers, the teeth were sterilised by subjecting them to gamma radiation (20 kGy). The teeth were then embedded in a poly-siloxane based silicone material (Lab-putty, Coltene Whaledent, Altstätten, Switzerland) to prepare 15 sets of models, each with 4 teeth.

6.2.2. Cavity preparations

Occlusal (GV Black Class 1) cavities with a depth of 2 mm were prepared in the molar and premolar teeth (2 mm or 1.5 mm in width, respectively), using water-cooled diamond straight fissure burs in a high-speed dental handpiece (model S-Max M600L, NSK, Kanuma, Japan). The cavity designs varied from tooth to tooth, to provide complexity. The cavities were rinsed thoroughly and then dried with compressed air. Each of the 15 models were coded for purposes of identification. A digital 3-dimensional scan of each model was performed using an intraoral scanner (TRIOS®3, 3Shape, 1060 Copenhagen K, Denmark), and saved as STL (stereolithography) files. Prior to use, the scanner was calibrated as per the manufacturer's instructions.

6.2.3. Restorations

The cavities were etched using 35% phosphoric acid (K-etchant Gel, Kuraray Medical, Okayama, Japan) for 30 seconds, rinsed for 60 seconds and dried with compressed air. A bonding agent (Scotchbond™ Universal Adhesive, 3M ESPE, St Paul, MN, USA) was then applied as per the manufacturer's instructions and cured with an LED curing light (1250 mW/cm$_2$) for 30 seconds.

Three different brands of tooth-coloured restorative material were used: Admira® Fusion (Voco Gmbh, Cuxhaven, Germany) (10 shades), GRADIA® DIRECT X (GC Corp., Tokyo, Japan) (7 shades), and TPH Spectra® LV (Dentsply DeTrey,

Konstanz, Germany) (13 shades). Details of the materials are given in Table 1, which also lists the fluorescence luminosity for each shade of each material. To ensure correct shade selection, a custom-made shade guide was fabricated for each brand of material, using all the available shades. Each brand was used in restoring 20 teeth, choosing the shade to correspond exactly to the individual teeth. The cavities were restored using the oblique layering technique. The models were then stored in distilled water at room temperature.

6.2.4. Removal of restorations

Five general dentists were tasked with removing the restorations from the teeth, with each being assigned 12 teeth (six teeth for the conventional method, and six teeth for the FAIR method). In the conventional method, the operators used the white light of the dental unit (30,000 lux) (HB-LED dental light, A-dec, Newburg, OR). For the FAIR method, operators illuminated the samples with an array of nine light emitting diodes (emission wavelength of 405 ± 10 nm) (model SEFL33UV-405, SE Electronics, Shanghai, China). The LED array was attached to the dental unit light. The operators used orange eyeglasses as a long pass filter.

Table 6. 1. Restorative materials

No.	Brand Name	Material type	Shade	Luminosity values
1	Admira Fusion	Nanohybrid Ormocer	A1	225.75
			A2	225.15
			A3	213.92
			A3.5	207.79
			A4	191.3
			B1	224.53
			B2	225.91
			B3	218.86
			C2	208.63
			D3	218.75
2	GRADIA® DIRECT X	Nano filled Composite	A1	226.89
			A2	221.26
			A3	238.49
			A3.5	202.37
			B1	228.93
			B2	220.85
			C2	206.61
3	Dentsply TPH Spectra® LV	Nano filled Composite resin	A1	177.13
			A2	168.02
			A3	159.67
			A3.5	155.51
			A4	111.48
			B1	185.76
			B2	144.03
			B3	136.97
			C1	152.01
			C2	142.17
			C3	125.91
			C4	109.88
			D3	138.27

Luminosity values are data for fluorescence luminosity when restorations are illumination by 405 nm light from Chapter 4 of this thesis (Study 2).

The dentists were provided with a double-ended sharp explorer, a conventional mouth mirror, and a three-way triple air/water syringe for use with both methods. They were instructed to remove the restorations using water-cooled straight fissure diamond burs in a high-speed dental handpiece. The total time needed to remove the restoration in each tooth was recorded, in seconds.

6.2.5. Post-operative scanning

After completing an optical three-dimensional scan of each model, 3D Tool version 13.2 CAD software was used to compare the cavity preparations prior to restoration and following restoration removal. Inter-cuspal cavity width measurements were recorded. To quantify tooth structure that was lost during restoration removal, the linear distance between the cavity walls at their external marginal line angles was measured. All measurements were made to the nearest 0.01 mm (Figure 6.1). In total, four measurements were taken for each tooth at specific sites in both the pre- and post-operative scans. The mean difference was calculated.

6.2.6. Statistical analysis

A student's t Test was used to evaluate the amount of tooth structure lost, the quantity of composite material remnants, and the time taken to remove the restorations. Inter-examiner variation was determined for each method by ANOVA. The level of significance was set at α = 0.05. The mean difference in

inter-cuspal cavity width measurements for CM was -0.502 (21.48%) and for FAIRM was -0.0492 (1.7%).

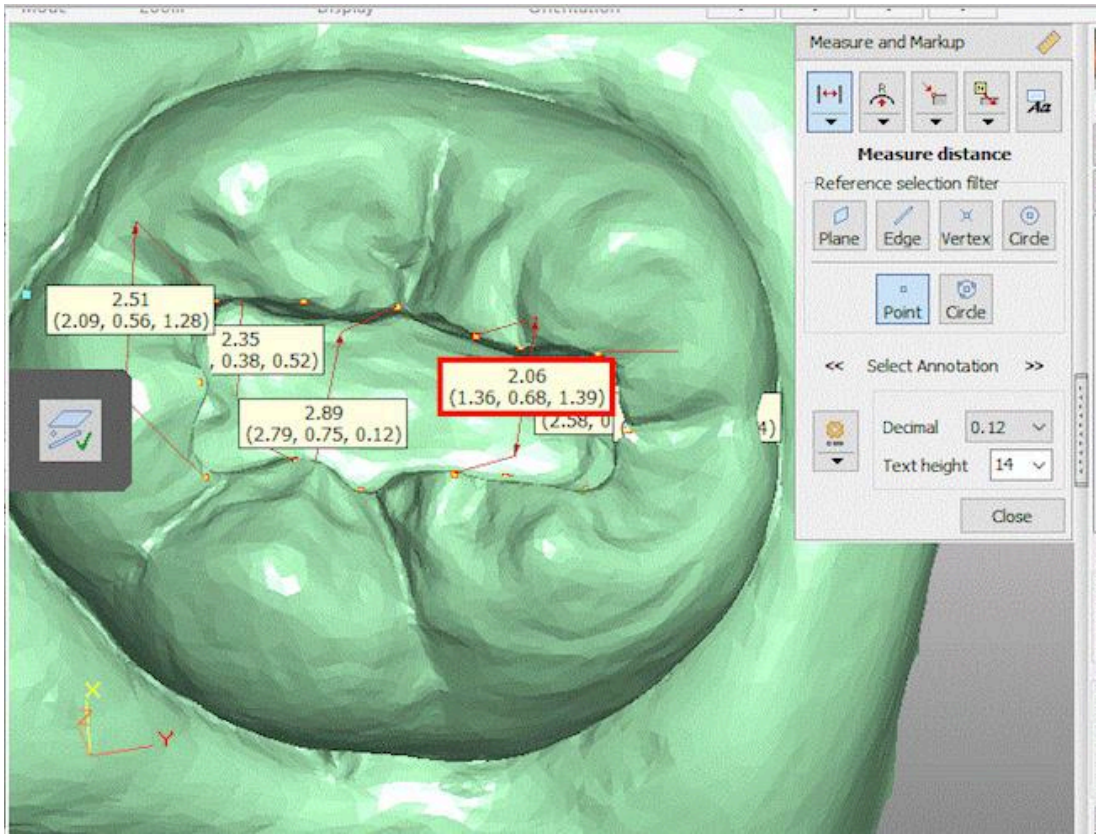


Figure 6. 1. Example of inter-cuspal width measurements

6.3. Results

Using the FAIR method, there was a small increase in inter-cuspal width of the cavities, but this did not reach the threshold for statistical significance (p = 0.17). The inter-operator variance in the mean difference of inter-cuspal width of the cavities between pre-test and post-test models was not significant (p= 0.30). In contrast, with the conventional method, there was a considerable increase in the

inter-cuspal width of the cavities (p = 0.0025). In addition, there was significant variation among the operators (p = 0.03) (Figure 6.2).

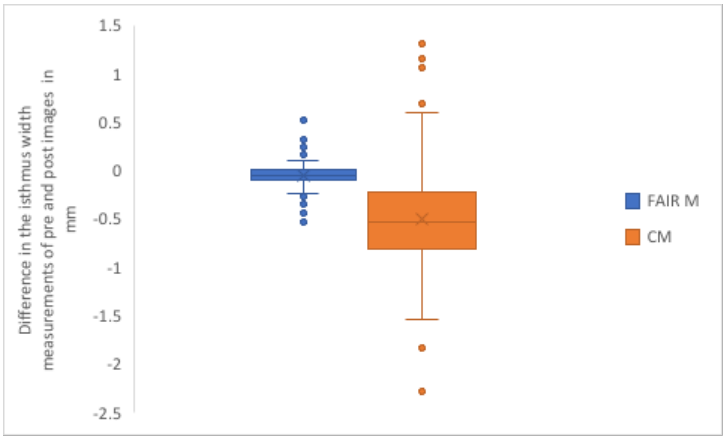


Figure 6. 2. Box plots comparing FAIR and the conventional method for inter-cuspal width measurements

Less time was required to remove the restoration when using the FAIR method, compared to the conventional method (p < 0.0001). Across all 5 dentists, the average time with FAIR was 100.23 seconds, versus 165.13 seconds with the traditional approach (Figure 6.3).

6.4. Discussion

This study demonstrates several advantages for fluorescence-aided removal of resin-based restorations, over conventional methods, including less operating time, less variation between operators, less remaining material, and most importantly, less gain in the inter-cuspal width of the cavity (Figure 6.4).

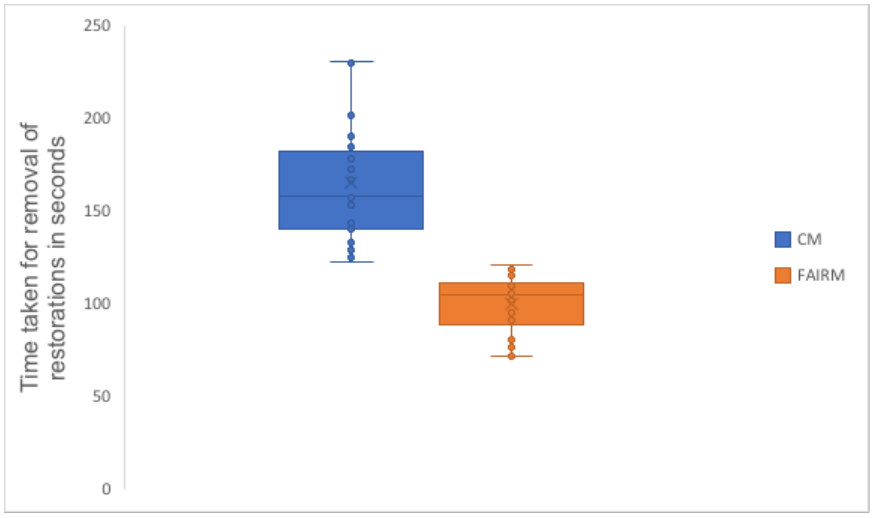


Figure 6. 3. Time taken for removal of restorations. Left side (blue) = conventional method, right side (red) = FAIR method.

Together, these indicate that the FAIR method facilitates the selective removal of tooth-coloured intra-coronal restorations.

The present study used 3-dimensional digital scans to compare cavities before and after restoration removal. The accuracy of these digital impressions varies according to the total arch length of the impression. Single tooth and quadrant impressions have demonstrated precision of 20 to 35 μm, while scans of complete dental arches have variations of 50 to 80 μm (189). This is the reason why, in the present study, models had 4 teeth. The scanner used has been shown to have a high accuracy for scans of quadrants (190).

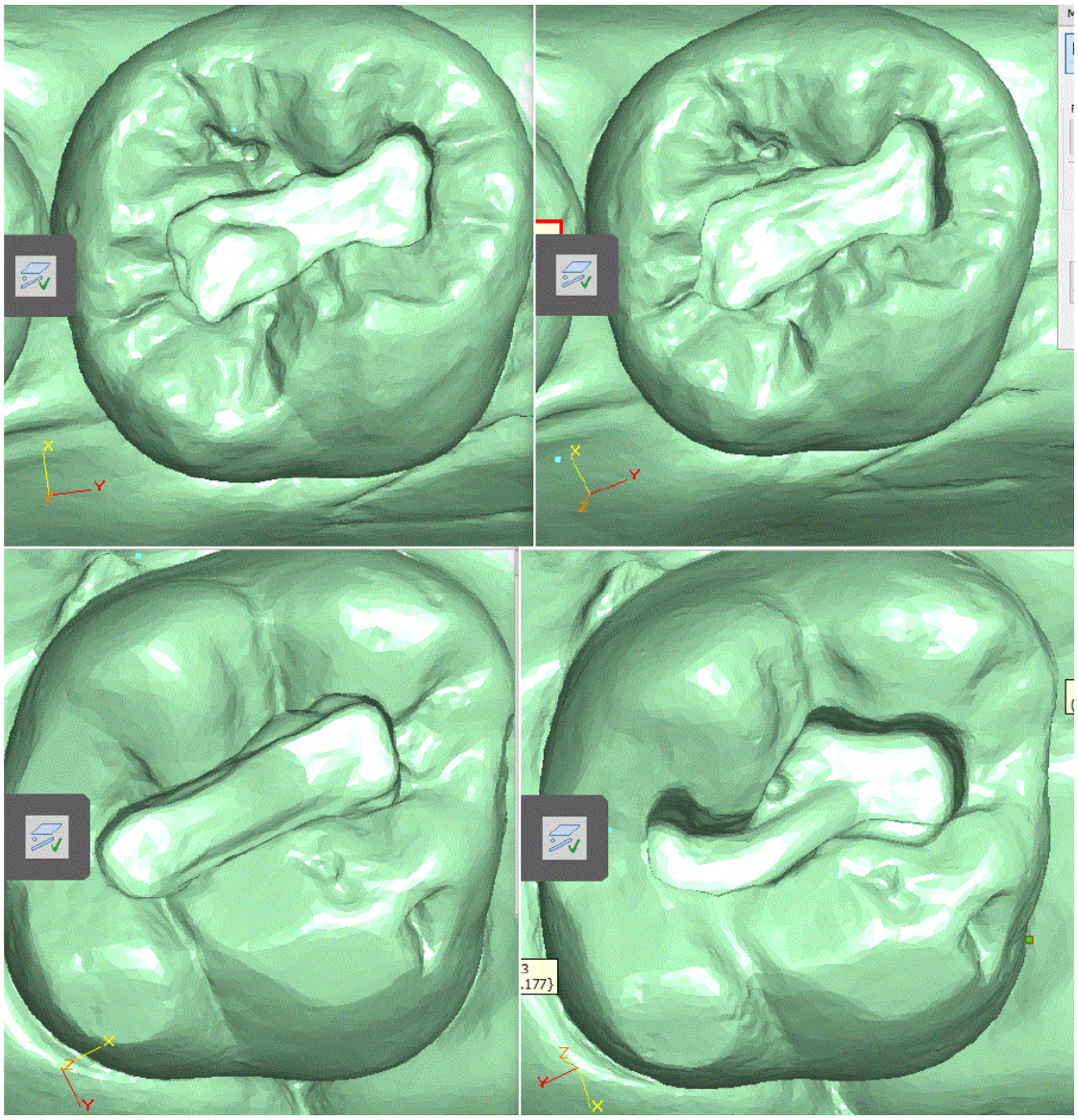
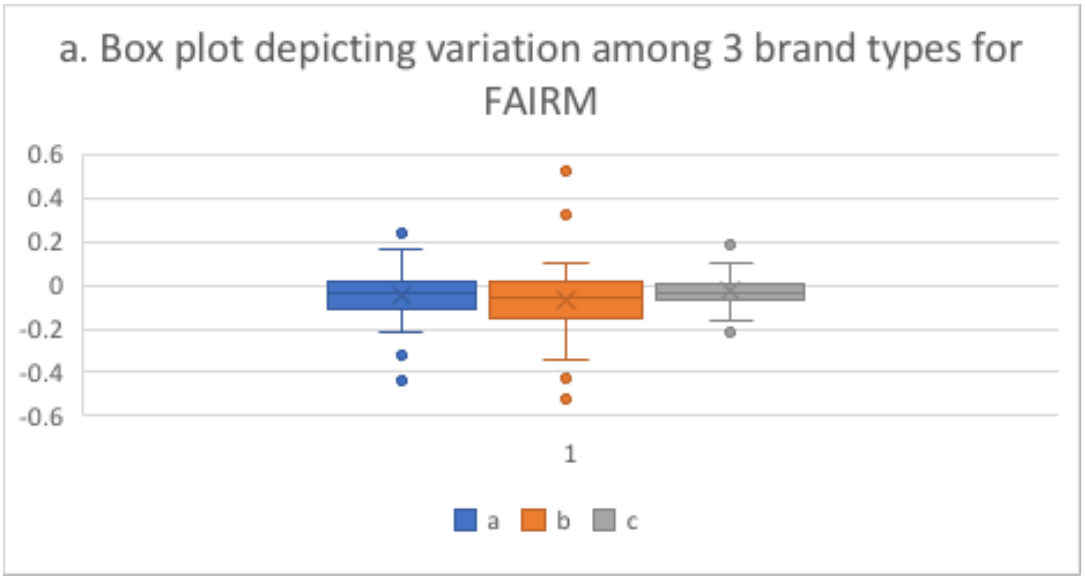


Figure 6. 4. Scans of teeth showing baseline cavities (left) and cavity preparations after restoration removal (right), using the a. the FAIR method (upper image pair) or b. the conventional method (lower image pair)

The principle of the FAIR method exploits the difference in fluorescence between natural tooth structure and RBC materials. There are variations between brands of RBC, and also between shades of the same material (as shown in Table 1). It is important to note that the majority of resin-based restorative composite materials fluoresce more brightly than tooth substance (49, 130). In the present study, two resin-based composites (GRADIA® DIRECT X and Dentsply TPH Spectra® LV) and an ormocer (Admira Fusion) were used, and no variation in performance of FAIR method among the three types of material was noted (Figure 6.5). Their luminosity values ranged from 111.48 to 228.93. The FAIR method should be applicable to the removal of other resin-based restorations.

In the present study, considerable effort was made to ensure that the restorations were optimally fabricated to make them difficult to detect by visual examination, matching optical traits such as shade and translucency to mimic the nuances of the adjacent tooth structure. While the current investigation focused on the removal of restorations, the same issues apply for removal of resin-based materials such as resin cements under onlays and crowns, resin fissure sealants, and bonding resins used under orthodontic bands. FAIR may have value for these clinical situations, since it can provide a clear demarcation between the material and adjacent tooth structure. Using FAIR helps to prevent the unnecessary removal of healthy tooth structure during replacement of defective restorations, and thereby supports preservation of tooth structure, a core principle of minimal intervention dentistry (191).



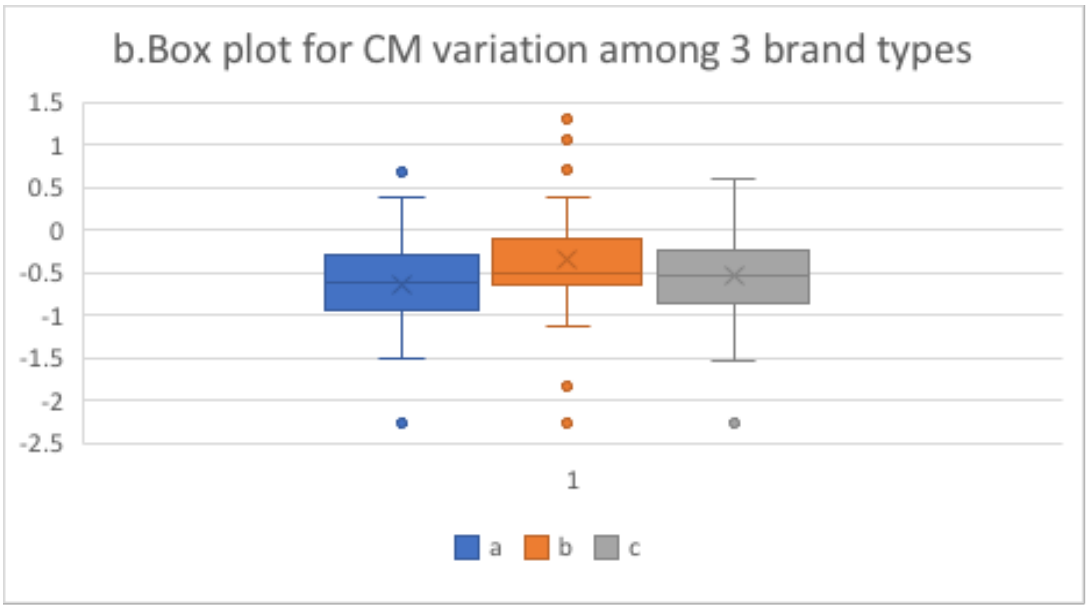


Figure 6. 5. Variations in changes in inter-cuspal width measurements between restorative materials. Upper panel A = FAIR, Lower panel – Conventional method. Material types are as follows: a = Admira Fusion, b = GRADIA® DIRECT X, c = Dentsply TPH Spectra® LV.

6.5. Conclusion

The FAIR method is a simple and less invasive technique which facilitates the selective removal of tooth-coloured resin-based composite materials, with minimal additional tooth substance loss, and lower amounts of remnant material at the end of restoration removal. It reduces the time required for complete removal of the restoration.

Chapter 7 Effect of heat on restorative materials

Additional material (colour plates and figures) for this study are attached as Appendix C

This chapter is published as a peer reviewed article (Appendix H)

Kiran R, Chapman J, Tennant M, Forrest A, Walsh LJ. Effect of Heat on the Fluorescence Properties of Tooth-Colored Restorative Materials and Their Forensic Implications. Journal of forensic sciences. 2019 Nov;64(6):1698-706

This chapter evaluated the effect of temperature on the fluorescence properties of selected contemporary tooth-coloured restorative materials, when exposed to 200 ◦C, 500 ◦C, 900 ◦C and 1200 ◦C using a fluorescence based DSLR camera.

7.1. Introduction

Fire is one of the most significant causes of morbidity and mortality across the world (192). Following a fire, the identification process of deceased individuals can be a challenging and complex endeavour, especially in cases where there are multiple fatalities. The bodies of victims of fires from house fires, automobile fires and aircraft crashes may be partly or completely carbonised (193, 194).

The principal means of identifying deceased persons are fingerprints, DNA, and dental comparisons (75), however fingerprint and DNA evidence may not be available when the soft tissues are totally decomposed or carbonised (74). On the other hand, the distinctive structure, composition and location of teeth may still provide useful information even under conditions when the soft tissues have decomposed (195). The information in dental records regarding the presence and distribution of teeth as well as the details of restored, non-restored, missing and decayed surfaces of teeth can be individuating. Along with these dental variables, additional information on the type or product brand of restorative materials used for dental restorations could add additional features to enhance the level of certainty in identification work when appropriate ante-mortem records exist (76, 77).

Methods that can be used for identification of specific brands of restorative materials include light-based fluorescence, X-ray fluorescence (XRF), scanning electron microscopy (SEM), and energy dispersive X-ray spectroscopy (EDS) (77). Light-induced fluorescence can enhance the gross recognition of the presence of restorative materials in teeth (78). In natural healthy tooth structure, dental enamel emits strong green-yellow fluorescence when illuminated by violet light (405–410 nm) and blue light (440–450 nm) (32). The key fluorophores responsible include amino acids in dentine, and inorganic minerals (14). To mimic the fluorescence properties of natural tooth structure, manufacturers of restorative dental materials include organic fluorophores and inorganic rare earth oxides into restorative materials (77).

The inorganic components of tooth-coloured restorative materials are mostly found in the glass or ceramic fillers which provide much of the high-strength and low-wear attributes of modern dental materials. These inorganic fillers have high melting points. The importance of temperature in the manufacture and the response of various components of dental restorative materials to heat is summarised in Table 7.1.

Table 7. 1. Sintering or fusion temperatures of common dental restorative materials

Material	Temperature range in ◦C	Comment
Glass Ionomer Cement	1100–1500	The glass powder particles are manufactured by fusing the raw materials at 1100–1500◦C
Composite Resins	300–500	Organic component of the resin evaporates
	1090	Inorganic components can withstand temperatures over 1090 ◦C
Dental porcelains		Dental porcelain materials are classified according to their fusing temperature
• High fusing	1300	
• Medium fusing	1101–1300	
• Low fusing	850–1100	
• Ultra-low fusing	<850	
Pressable glass ceramics IPS Empress™	1180	Pressing temperature
Zirconia core ceramics	1350	The coping is fired for 6 hours to sinter the yttrium-stabilised zirconia core coping.

The objective of the present study was to evaluate the effect of heat on the fluorescence behaviour of contemporary tooth-coloured restorative materials and natural tooth structure when subjected to a range of temperatures. The materials used comprised resin-modified glass ionomer cements, composite resins, ormocers (organically modified ceramics) and hybrid ceramics, each of which contain both resin and ceramic components. The study was conducted with two hypotheses. The first was that the ceramic component would show less

degradation when heated to temperatures above 1000 ◦C. The second was that tooth-coloured restorative materials which contain inorganic fluorophores would retain fluorescence properties after exposure to a temperature of 900 ◦C.

7.2. Materials and Methods

Extracted teeth

A total of 132 extracted human permanent teeth (premolars and molars) devoid of any pathology were selected for the study. Following extraction, the teeth were stored in a solution of 0.1% thymol (Sigma Aldrich, St Louis, MO), and sterilised by exposure to gamma radiation (20 kGy).

7.2.1. Sample preparation

Any surface deposits on the roots or crowns of the teeth were removed by mechanical debridement using hand scalers. The teeth were then divided randomly into groups of 12, with each group used for a particular restorative material type (Table 7.2). Twelve unrestored teeth were retained as untreated controls, while 24 teeth were prepared for full coverage crown restorations, and 96 teeth were prepared for occlusal (GC Black Class I) cavity preparations with dimensions of 2.0 mm width and 2.0 mm depth in each tooth. All tooth preparations were undertaken using water-cooled diamond burs in a high-speed dental handpiece (S-Max M600L, NSK, Kanuma, Japan).

Table 7. 2. Restorative materials used in the study

Material Type	Commercial Name	Manufacturer	Shade
Ormocer	Admira Fusion	Voco	A2
Composite Resin	Herculite™ Ultra	Kerr	A1 E
Composite Resin	Gradia® Direct X	GC	A1
Composite Resin	Dentsply TPH Spectra®	Dentsply Sirona	A2
Composite Resin	3M Filtek™ supreme XTE	3M ESPE	A2 E
Glass-Ionomer cement	Fuji II	GC	A1
Glass-Ionomer cement	Fuji VIII	GC	A2
Glass-Ionomer cement	Fuji IX	GC	A2
Hybrid ceramic	Vita Enamic®	Vita	2M2T
Feldspar Ceramic	Vitablocs®	Vita	1M2C

Manufacturer details: Vita Zahnfabrik, Bad Sackingen, Germany; 3M ESPE, Minneapolis, MN, USA; GC Corp, Tokyo, Japan; Voco, Cuxhaven, Germany; Kerr, Uxbridge, UK Dentsply Sirona, York, PA, USA.

For teeth with Class 1 cavities that were restored with resin-based restorative materials, the following standard clinical protocol was used. The cavity was

etched with 34% phosphoric acid (Scotchbond™ Universal Etchant, 3M-Espe, St Paul, MN) for 20 seconds. After the etchant had been removed by water spray, a dentine bonding agent (Scotchbond™ Universal adhesive, 3M-Espe) was applied and left for 20 seconds, then thinned by compressed air for 0.5 seconds and polymerised for 10 seconds using an LED curing light (Mini-LED, Acteon Satelec, Merignac, France). The restorative material was placed using a multiple layering technique, and each increment was polymerised for 20 seconds.

For teeth with Class 1 cavities that were restored with glass ionomer cement-based restorative materials, the following standard clinical protocol was used. The cavity was conditioned with 20% polyacrylic acid solution (GC Cavity Conditioner™, GC Corp, Tokyo, Japan) for 10 seconds. The conditioner was then removed by rinsing with water for 10 seconds and the cavity was air-dried. The relevant material was then mixed using a mechanical mixer (Ultramat 2 Amalgamator, SDI Limited, Bayswater, Victoria 3153, Australia) and applied directly into the cavity. For light-cured materials, a 20 second exposure to a curing light was used. Self-curing materials were allowed to set fully over 4 minutes, as per the manufacturer's instructions. Once set, a surface coating (Fuji Coat™, GC Corp) was applied to all glass ionomer materials, and this was light cured for 20 seconds.

For teeth prepared for full crown restorations, the enamel was etched with phosphoric acid gel for 30 seconds. After the etchant had been removed by

water spray, the bonding agent (Scotchbond™ Universal adhesive, 3M-Espe) was applied and left for 20 seconds, then thinned by compressed air for 5 seconds and polymerised for 10 seconds using an LED curing light (Mini-LED, Acteon Satelec, Merignac, France). A second coat of bonding agent was applied in the same manner. The full crown restoration was manufactured for each tooth using a CAD-CAM milling machine (inLab MC XL, Sirona Dental Systems GmbH, Bensheim, Germany). For Vita Enamic®, crowns were made both with and without a clear glaze being applied to the final crown (Optiglaze™, colour clear HV, GC Corp., Tokyo, Japan), so that the influence of the glaze on the response to heating could be assessed.

The inner surface of each ceramic full crown was etched with phosphoric acid for 15 seconds, then rinsed with water for 60 seconds, and air dried with compressed air for 20 seconds. A silane-coupling agent was applied (RelyX™ Ceramic primer, 3M-Espe), followed by the bonding agent (Scotchbond™ Universal adhesive, 3M-Espe), which was air thinned for 5 seconds before being polymerised. A luting cement (RelyX™ veneer cement, A1 shade, 3M-Espe) was then applied to the fitting surface of the restoration, and the crown was seated onto the tooth. Excess cement was removed with a probe. Once seated fully, all margins were light-cured for 30 seconds.

The restored teeth were then set in individual custom trays fabricated using heat-stable dental investment material for ease of manipulation. As controls, three

discs of 1 cm diameter and 2 mm thickness for each restorative material were prepared using a microscope glass slide and acetate strip. Each disc sample of material was cured with a LED light curing lamp for 20 seconds. For VitaEnamic™ and Vitablocs™, samples of material that were 2 mm in thickness were prepared using a diamond saw (IsoMet™ 1000, Buehler, Lake Bluff, IL, USA).

7.2.2. Methodology

All samples and restorations were photographed prior to exposure to heat and then again after heat treatment. Samples were imaged with white light (WL), and also with violet light (VL) (405 nm), the latter in conjunction with yellow or orange filters to block reflected violet light. Samples were photographed with a digital single-lens reflex camera (EOS Rebel T2i/EOS550D, Canon, Tokyo, Japan) fitted with a macro lens (focal length 60mm and f/2.8) in a dark environment. The illumination was perpendicular to the sample. All the images were taken under standardised conditions with a fixed camera to a sample distance of 200 mm and a dark background. The same camera settings (ISO 400, aperture F2.8, shutter speed 1/30 second) were used throughout the study. Images were recorded using 8 bits per colour channel (16.8 million colours), with an image size of 18 megapixels. The camera was set to use the Adobe RGB1998 colour space, and the white balance control was set to white fluorescent light, with white balance correction set to zero.

The discs and restored teeth for each material type were randomly divided into four sub-groups of three for each temperature (200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C). Each tooth was placed in a 30 mL glazed porcelain crucible with a lid, while discs were placed in a custom-made tray fabricated by using heat-stable dental investment material (Bellavest®, Herbst GmbH, Bremen, Germany). Heat treatment was undertaken using a silicon carbide muffle chamber furnace (ModuTemp™, XRF Scientific, Osborne Park, Western Australia), with an increasing rate of 10 ◦C per minute and then a holding time of 30 minutes. After cooling on the bench, samples were photographed using the same arrangement as for the baseline images.

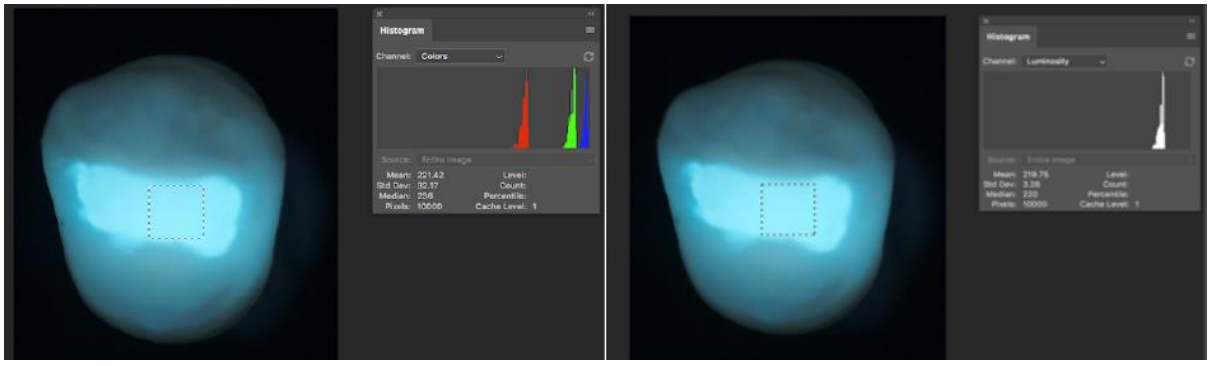


Figure 7. 1. Example of image analysis. A violet light fluorescence image of an occlusal restoration is shown, with the sample selection area (a square of 10,000 pixels in area). The histogram analysis tool shows data for red, green and blue (left) and luminosity (right).

7.2.3. Statistical analysis

Adobe Photoshop™ Creative Cloud software was used for digital image analysis. The histogram tool was used to record the average 8-bit red, green, blue and

luminosity (RBGL) values for a fixed sample area of 10,000 pixels, taken from the baseline and post-test images (Figure 7.1). All colour data was recorded in levels from 0 to 255. In a digital colour image, each pixel is a triad of red, green and blue, each of which has a possible brightness value from 0 (black) to 255 (maximum colour). Likewise, luminosity (brightness) also varies from 0 to 255. The average RGBL values were recorded for the sample area for each sample, and then the mean of three replicate samples for each material calculated, and used to compare baseline and post-treatment values, using repeated measures ANOVA, with Tukey post-hoc tests. The significance threshold was set at $p < 0.05$.

7.3. Results

Overall, there was significant variation in the changes seen for colour and fluorescence between the different materials tested (Table 7.3).

Table 7. 3. Summary of differences between materials

Comparison	p value
White light - luminosity	0.031
White light - red channel	0.037
White light - green channel	0.026
White light - blue channel	0.160 (NS)
Violet with orange filter - luminosity	< 0.001
Violet with orange filter - red channel	<0.001
Violet with orange filter - green channel	<0.001
Violet with orange filter - blue channel	<0.001
Violet with yellow filter - luminosity	< 0.001
Violet with yellow filter - red channel	<0.001
Violet with yellow filter - green channel	<0.001
Violet with yellow filter - blue channel	0.004

NS = not significant

7.3.1. Changes observed at 200 ºC

Under white light, natural tooth structure turned slightly brownish in colour. In a similar way, Vita Enamic crowns and composite resin (CR) and ormocer restorations showed brown discolouration. Glass ionomer cement [GIC]) restorations appeared a more chalky white colour (Figure 7.2). In contrast, changes to the VitaBloc ceramic full crowns were minimal, and not statistically significant.

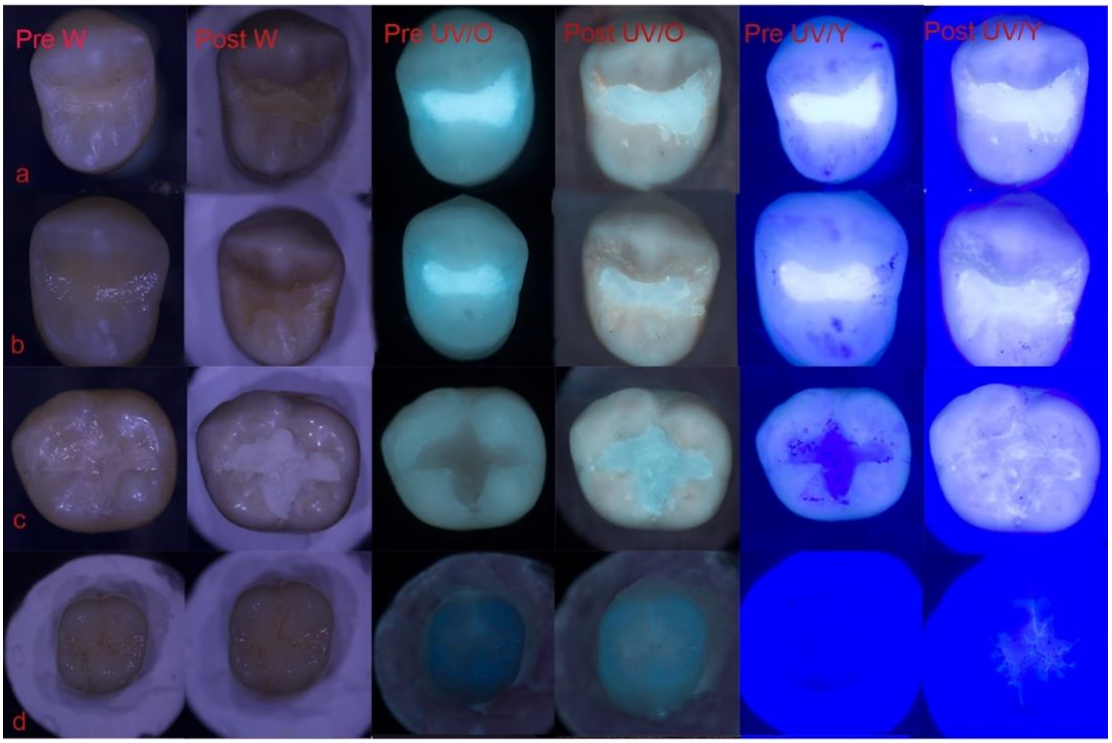


Figure 7. 2. Images of teeth samples comparing the baseline images(pre) to post heat treatment at 200 ◦C, when excited with white light (W), UV-A light with orange filter (UV/O) and UV-A light with yellow filter for a. Admira Fusion, b. Dentsply TPH Spectra®, c. Fuji II GIC and d. VitaEnamic®.

Under violet light, the natural bright blue-green fluorescence of natural tooth structure was reduced. Likewise, the fluorescence of composite/ormocer restorations was reduced. The two exceptions were 3M Filtek Supreme XTE composite resin and Fuji II LC resin modified glass ionomer cement, both of which showed increased fluorescence (both $P < 0.05$) (Figure 7.3).

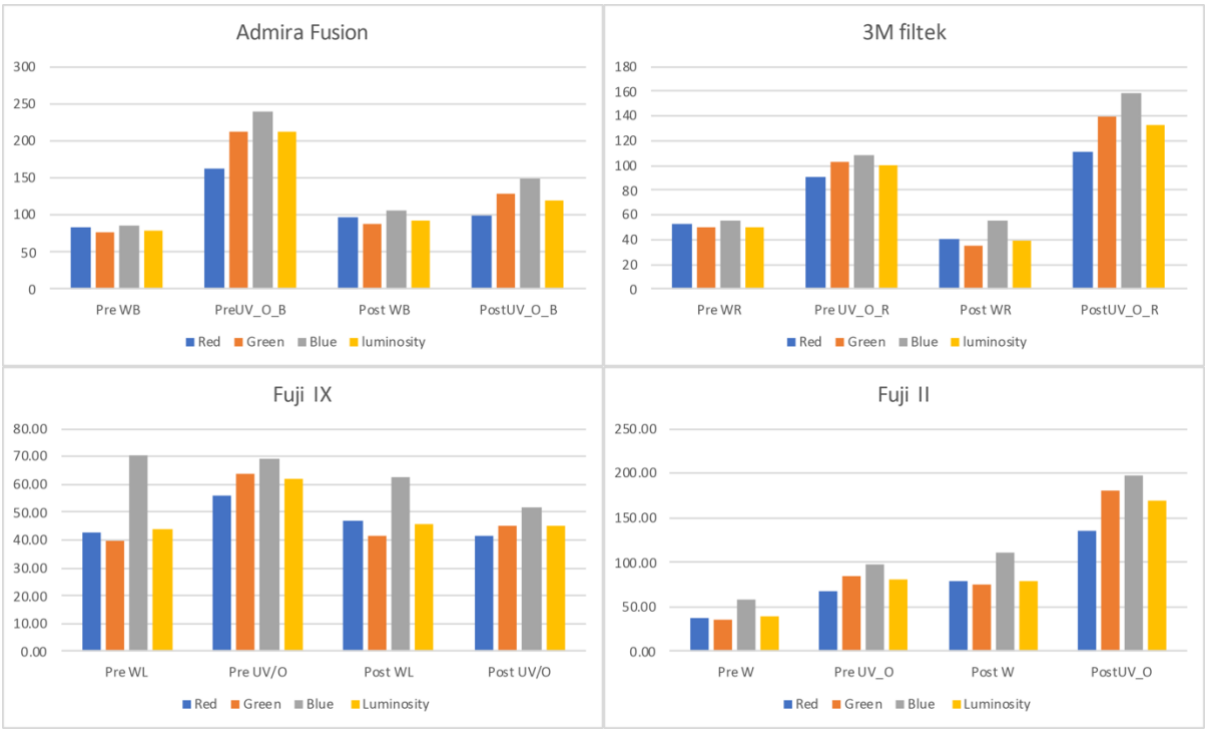


Figure 7. 3. Bar graph comparing the Red, Green, Blue and Luminosity colour channel values for Admira Fusion, 3M Filtek Supreme XTE, Fuji II and Fuji IX materials before and after undergoing heat treatment at 200 ◦C.

Crowns made using Vita Enamic exhibited enhanced fluorescence after 200 ◦C heat treatment, which was due entirely to the glazing material, as this did not occur in unglazed crowns. VitaBlocs did not show any changes in fluorescence.

7.3.2. Changes observed at 500 ºC

Under white light, the crowns of the teeth appeared increasingly dark in colour, with fissures stained black. The discolouration of composite resin and ormocer restorations was more pronounced than at the lower temperature, and these now appeared grey-black in colour, and were quite distinct compared to adjacent tooth structure. This change was seen both visually and through reduced values for RGB and luminosity. There was shrinkage at the margins, with visible cracks

evident. All glass ionomer cement restorations appeared blackish-grey in colour under WL, with some cracks at the margins. Vita Enamic crowns now were a more chalky-white in colour and had undergone fracture. In contrast, there were mild visible changes with VitaBloc ceramic full crowns at this higher temperature, with the material starting to become brown.

Under violet light with an orange filter, natural teeth had reduced blue fluorescence and now appeared brownish in colour. In contrast, using the yellow filter, natural tooth structure now showed a pink colour. Using the orange filter, composite resin, ormocer and GIC materials showed a total loss of their original fluorescence. In contrast, using the yellow filter, these materials retained some blue fluorescence, which allowed their margins to be distinguished clearly from adjacent tooth structure. The Vita Enamic crown exhibited no fluorescence, and the original fluorescence from the glaze was also lost (Figure 7.4). There was no significant change in the fluorescence of VitaBlocs at 500 ◦C.

7.3.3. Changes observed at 900 ºC

Under white light, natural tooth structure appeared as charcoal (greyish-black) in colour. Composite materials had now become chalky white (Gradia X) or white, frosty, translucent and vitreous or glass-like in appearance (Herculite Ultra, TPH Spectra® , and ormocers), and had a corresponding increase in their values for RGB and luminosity, compared to baseline. GIC restorations also appeared increasingly chalky white, with more obvious marginal gaps at this higher

temperature then had been seen at lower temperatures. Glazed Vita Enamic crowns lost their glossy finish and showed fractures in the restoration. VitaBloc crowns developed a ground-glass appearance, with an increase in values for RGB and luminosity.

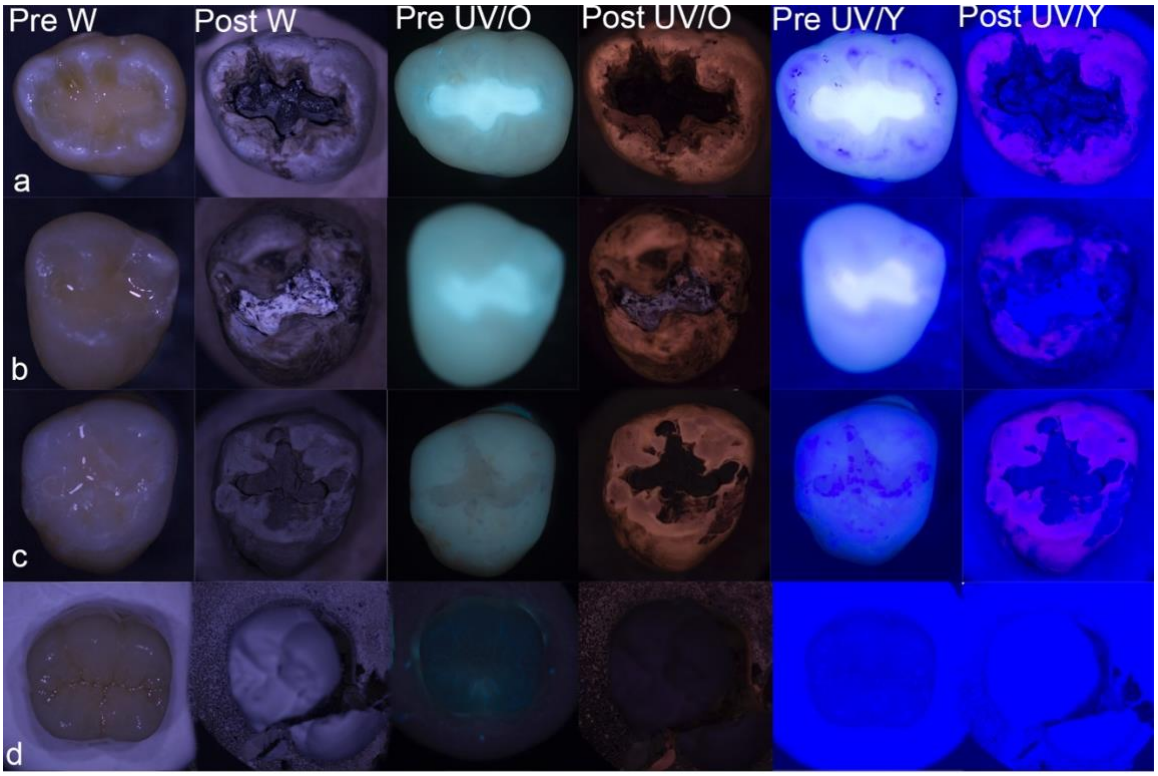


Figure 7. 4. Images of teeth samples comparing the baseline images(pre) to post heat treatment at 500 ◦C, when excited with white light (W), UV-A light with orange filter (UV/O) and UV-A light with yellow filter for a. Admira Fusion, b. Dentsply TPH Spectra®, c. Fuji II GIC and d. VitaEnamic®.

Under violet light with an orange filter, enhanced pink-red fluorescence was seen for GIC restorations, and TPH Spectra composite resin (Figures 7.5 and 7.6). There was almost no fluorescence from natural tooth structure or from other

composite resin or ormocer restorations, making these now indistinguishable from tooth structure. The same overall patterns were seen for violet light with a yellow filter, except that this removed the red fluorescence signals. There were no changes in the fluorescence of VitaBloc crowns.

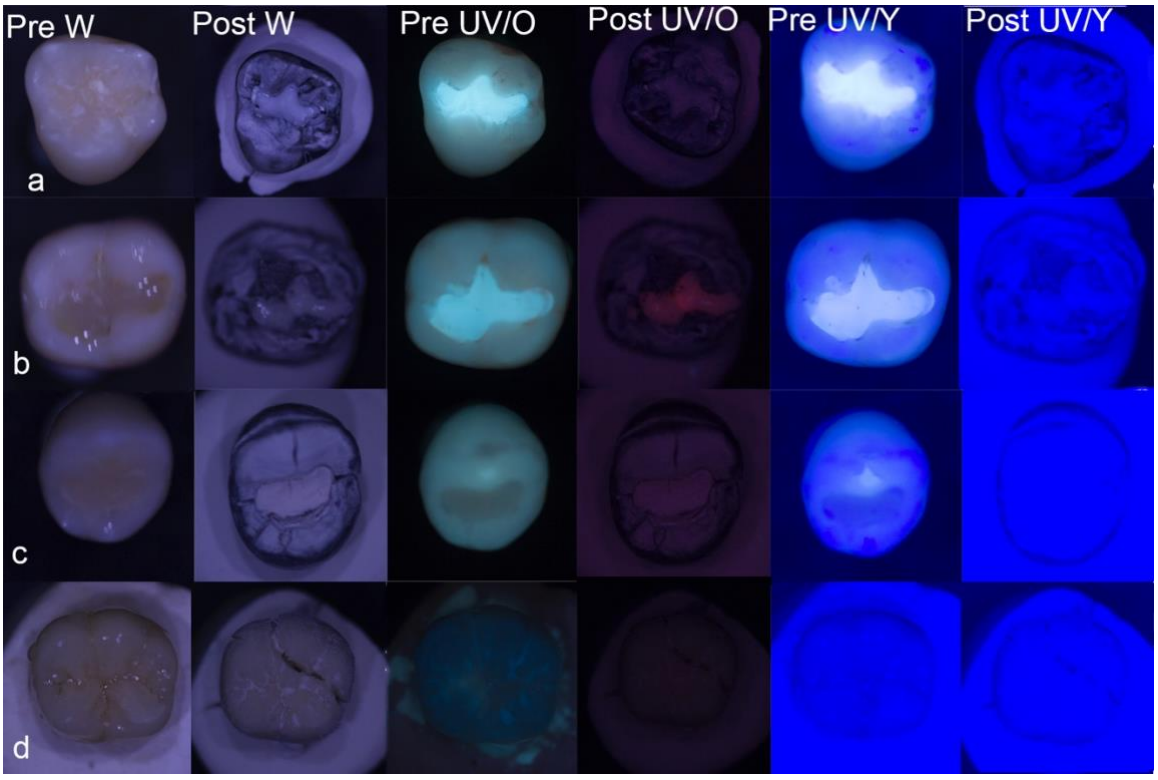


Figure 7. 5. Images of teeth samples comparing the baseline images(pre) to post heat treatment at 900 ◦C, when excited with white light (W), UV-A light with orange filter (UV/O) and UV-A light with yellow filter for a. Admira Fusion, b. Dentsply TPH Spectra®, c. Fuji II GIC and d. VitaEnamic®.

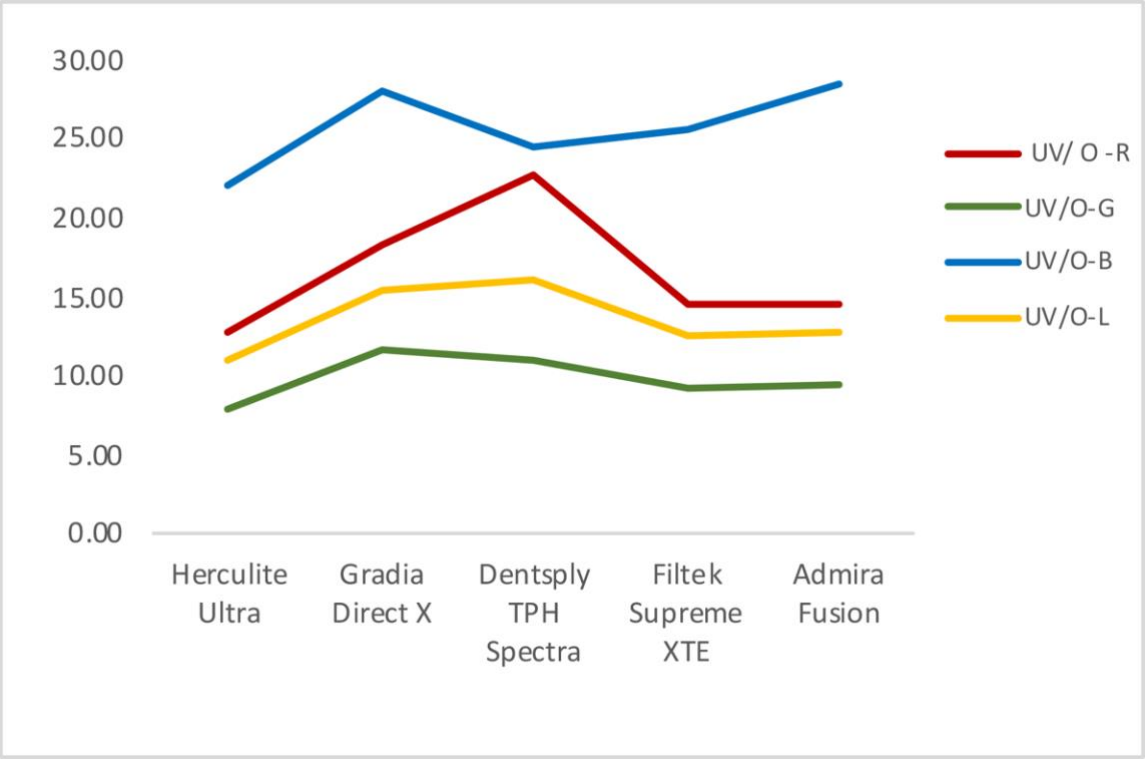


Figure 7. 6. Line graph depicting the Red, Green Blue and Luminosity colour channel values for resin-based composite and ormocer materials following 900 ◦C heat treatment when excited with UV-A light and viewed under orange filter (UV/O).

7.3.4. Changes observed at 1200 ºC

Under white light, crowns of natural teeth were charcoal (greyish-black) in colour, but the roots had now become chalky white. The process of vitrification was pronounced in the composite resin and ormocer materials, which had become translucent glass-like masses or chalky white in colour (Figure 7). There was a corresponding increase in the R, B, G colour channel and luminosity values. Likewise, GIC restorative materials appeared chalky white, and showed marginal gaps. Vita Enamic crowns developed a glossy glass-like appearance. VitaBloc

crowns showed a melted surface, and a reduction on the R, B, G and luminosity values compared to 900 ◦C.

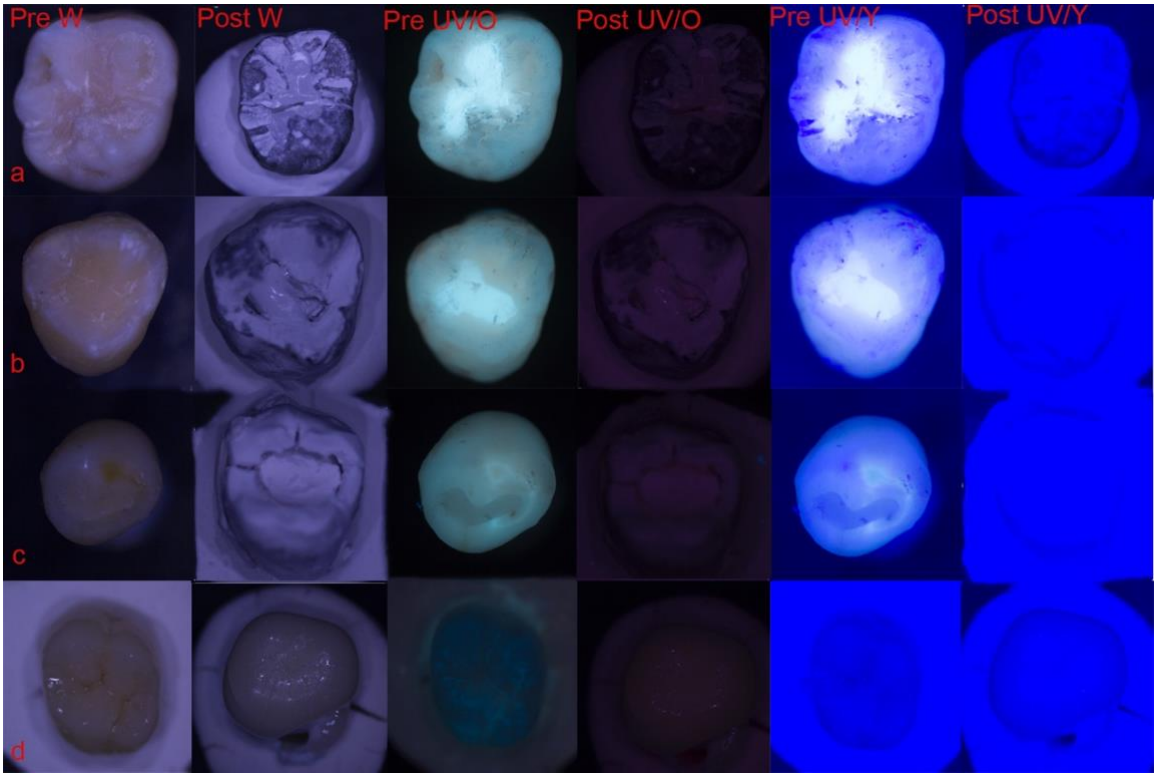


Figure 7. 7. Images of teeth samples comparing the baseline images(pre) to post heat treatment at 1200 ◦C, when excited with white light (W), UV-A light with orange filter (UV/O) and UV-A light with yellow filter for a. Admira Fusion, b. Dentsply TPH Spectra®, c. Fuji II GIC and d. VitaEnamic®.

Under violet light with either filters used, there was no difference between composite restorations and adjacent tooth structure. The GIC materials retained their light pink fluorescence. There were no other significant changes compared to 900 ◦C.

7.3.5. Disc samples

At 200 ◦C, the composite resins turned light brown in colour, and started to show a shift in fluorescence to a brownish hue, and greater fluorescence under both filters. At 500 ◦C, the composite resins and ormocers had developed distinctive patterns. The ormocers, Herculite Ultra and Filtek Supreme XTE had developed a bluish-black colour, while Gradia X had developed a mosaic-like pattern of a white background with brown lines, and TPH Spectra showed a mosaic pattern of a dark brown background with scattered white chips. At 900 ◦C, the appearance of areas of ash-white material or vitrification became apparent in all composite resin and ormocer materials, and this progressed such that by 1200 ◦C ormocers and composites all had developed a glass-like appearance.

For the glass ionomer materials, at 200 ◦C Fuji II appeared whiter while Fuji VIII and Fuji IX appeared darker than baseline. At 500 ◦C, all GIC materials darkened, and at 900 ◦C and 1200 ◦C all GIC samples appeared ash white in colour. Across all temperatures, Fuji II was lighter than Fuji VIII and IX. There were significant changes in the pattern of fluorescence with increasing temperature, when the long pass orange filter was used. By 500 ◦C the fluorescence in red had increased, giving a brownish colour emission, and at 900 ◦C and 1200 ◦C these materials showed a light pink emission. These pink signals were not seen when the shorter cut-off (yellow) filter was used.

For the ceramic materials, Vita Enamic discs underwent the same pattern of changes as seen for the crowns of the same material. At 500 ◦C, the material became chalky white in colour, then at 900 ◦C darkened, and at 1200 ◦C developed a molten glass appearance. VitaBloc discs melted at 1200 ◦C.

7.3.6. Teeth versus disc samples

In general, there was little or no variation in the pre-test recordings among the same material type for disc samples versus restorations, when viewed under white light. However, restored teeth samples showed statistically significant variations in baseline readings among the same material type for disc samples versus restorations, when viewed under violet light with a long pass (orange) filter. This variation was observed mostly amongst composite resin materials, which were sufficiently translucent that underlying fluorescence from natural tooth structure would be present.

There were some unique patterns of results seen when heating the discs samples versus the restored teeth. The most notable was the much stronger red fluorescence observed in restorations of Dentsply TPH Spectra in teeth CC at 900 ◦C than in the disc samples, which in contrast had only a weak pink emission.

7.4. Discussion

This study assessed the effect of heat on the fluorescence behaviour of tooth-coloured restorative materials and natural tooth structures. The results demonstrate the importance of material composition as a variable for the response to heating. The original hypothesis that the inorganic component of materials would show the greatest resistance to degradation was confirmed. VitaBloc crowns maintained their form, colour, and fluorescence properties until the point was reached where the material melted. In other materials, the organic component degraded at lower temperatures, causing marginal gaps to develop, and the material to become brown. Eventually, with increasing temperatures, this component degraded, leaving behind the inorganic part, which eventually melted or vitrified with increasing temperature.

Most restorative materials tested in this study lost their signature room temperature fluorescence properties at 500 ◦C, however there were some useful new patterns that emerged, such as Dentsply TPH Spectra®, which exhibited some red fluorescence at 900 ◦C, and the glass ionomer materials with their pink-red fluorescence (attributed to strontium-based glass components) that was maintained at high temperatures. Such findings indicate that identification of such patterns could be useful for detecting particular types of materials. According to the literature (196) and to product information provided by manufacturers, the inclusion of strontium is specific to glass ionomer cements from one

manufacturer (GC Corporation), so the identification of material types and brands based on such patterns could be of potential value.

Because of the location of teeth in the body, they gain a degree of protection from the effects of fires from the soft tissues around the jaws and the tongue and alveolar bones (136, 197). The posterior teeth in particular may be spared some of the influence of heat. The results of the present study show that the effects of heat on restorative materials occur as fundamental effects of heat per se, with only a small influence for the material being present within a tooth, as seen from the comparisons of discs versus restorations. Having said that, it must be stated that composite resin materials, which are designed to be translucent, will permit some light to pass through them, and thus some differences from fluorescence patterns at baseline for a disc of the material, compared to a filling of the same material inside a tooth, are not unexpected.

The effects of temperature on teeth and restorations within those teeth will be influenced by a range of factors, including whether there is direct exposure to flames, the maximum temperatures achieved, the duration of the heating event, the presence of any materials (in addition to the soft tissues) interposed between the teeth and the fire, and temperature changes that are due to water or other substances used to quench the fire (137). Adding to this, the carbonised body may be found at a stage where the soft tissue putrefaction has occurred, meaning that there are few bodily remains, other than bones and teeth (193).

Thus, in cases of long duration fires with temperatures above 700 ◦C, dental analysis may be the only reliable method of identification of victims, since other primary and secondary means of identification have been destroyed (143, 145). This is why, in the present study, the behaviour of various tooth-coloured dental materials was tested within the range of temperatures commonly encountered during fires in which victims die.

The present findings are consistent with the known properties of dental materials. At 200 ◦C, conventional glass-ionomer cements lose their water content, while at 500 ◦C, composite resin materials begin to decompose due to volatilisation of their organic component (140). At 900 ◦C, the inorganic fillers of tooth-coloured materials (such as the composite resins and ormocers used in the present study) would be expected to be stable (74).

VitaEnamic™ is a hybrid ceramic material that consists of a ceramic structure filled with polymer material. The ceramic component comprises a fine-structure feldspar ceramic enriched with aluminium oxide. A major change was seen at 500 ◦C, where the material appeared as chalky white and brittle, due to decomposition of the resin-based polymer material. At 1200 ◦C, the ceramic part that remained formed into a molten vitrified mass.

In a fire, the upper anterior teeth of a person are affected the most, while the posterior teeth may remain partially protected (146). Posterior teeth with multiple roots tend to remain in place within their alveolar bone sockets (147). The separation of the enamel from dentine, a phenomenon referred to as 'popping off', may occur at around 450 ◦C to 500 ◦C, when teeth are heated for 30 to 60 minutes (148, 149). This phenomenon is due to the difference in the water content of enamel (3–4% by weight) and dentine (10% in weight), where due to increased shrinkage of dentine, the resultant forces causes separation of enamel and dentine (198). This is the reason why, in the present study, posterior teeth were used, and they were mounted in dental investment material and placed in ceramic containers. Placing teeth samples in ceramic containers with lids was designed to reduce this "popping off" effect. However, in some fire incidents (such as bush fires), due to the prolonged effect of heat, it may be that only a few roots are recovered, with few or no tooth crowns (199).

Although several studies have assessed the macroscopic and microscopic changes that occur in tooth-coloured restorative materials with elevated temperatures, there has been to date little examination of the fluorescence behaviour of tooth-coloured materials, or on the changes which occur to contemporary hybrid restorative materials. Tooth-coloured restorative materials when illuminated with violet (405 nm) light and viewed under a long pass (orange) filter exhibit varied emission patterns, which enables an examiner to readily distinguish them from natural tooth structure (174).

The different fluorescence properties of dental materials are attributed primarily to the inorganic fillers used. Contemporary resin-based composite restorative materials comprise both an organic matrix and inorganic filler component. The organic matrix generally consists of a combination of crosslinked di-methacrylate monomers, such as Bis-GMA, UDMA or decanidol dimethacrylate (D3MA). Fillers such as silicate glass, fumed oxides/mixed oxides constitute the inorganic component. The filler content, size and shape influence the colour difference values and translucency of the resin-based composite materials. In a fire, the inorganic filler, including various inorganic oxide pigments, will be the most heat-stable parts of a dental restoration. However, at 900 ◦C, these will tend to melt together into an opaque vitrified mass.

7.5. Conclusion

Exposure to heat caused colour changes in all the tooth-coloured restorative materials tested, at all the temperatures tested. These colour changes reflected the differences in the composition of the materials, as different materials decompose at different temperatures. At 200 ◦C the resin-based composite materials still fluoresced. Resin-based restorative materials exhibited major changes at 500 ◦C, due to volatilisation of resin and hence loss of fluorescence. Materials with inorganic fluorescent agents fluoresced at 900 ◦C as these fillers are still present at 900 ◦C, while at 1200 ◦C the inorganic fillers fused to form vitrified masses. None of the materials tested in this study exhibited fluorescence

after treatment at 1200 ◦C. In natural or man-made fire accidents where teeth and bones are the only remains (if well preserved or stabilised for forensic examination), tooth-coloured restorative materials may provide some forensic evidence towards carbonised victim identification.

Chapter 8 General Discussion

In addressing the overarching question, "*Can tooth-coloured restorations be identified accurately under UVA-violet light illumination?*", a series of smaller questions were considered and examined. The experiments that tested the hypotheses that followed have addressed several gaps in the knowledge of the fluorescence properties of contemporary tooth-coloured restorative materials, particularly the hybrid restorative materials.

8.1. Studies 1 & 2 (Chapters 3 and 4)

While there has been considerable past work on the fluorescence properties of tooth structure and of older types of tooth-coloured restorative materials, the present work has filled some important gaps in the literature. Firstly, it has used 405 nm light from LEDs in various arrays, which are low cost and simple to fabricate, and more practicable than other types of light sources in the UVA or visible violet range.

Secondly, the present work has used a range of contemporary tooth-coloured materials, many of which have not been examined in previous studies, such as glass ionomer cements, organically modified ceramics (ormocers) and polymer infiltrated ceramics.

Tooth-coloured restorative materials have evolved considerably since their introduction, and with improvements in technology, newer materials with better

physical properties and superior optical properties have been developed. With the rising demand for aesthetic restorative materials, newer materials with different fluorophores are now in common usage. Despite attempts to develop materials that mimic natural tooth structure, it is clear that the fluorophores used at the present time do not overcome the issue of illuminant metameric failure. Hence, restorations can be detected using violet light-induced fluorescence, despite being imperceptible under white light.

Thirdly, the present work shows the influence of water on the fluorescence properties of tooth-coloured restorative materials, an aspect that has not been examined previously in the literature. After storage in water at room temperature, the fluorescence intensity was reduced. However, there was no evidence of drift in the fluorescence properties of materials when kept in the dry state over 60 days, after having been previously immersed in water. This indicates that the effects caused by sorption of water are reversible.

The work described in Chapter 3 was the first study to consider a broad range of materials, with most types of tooth-coloured restorative materials included. Violet light elicits green fluorescence from healthy tooth enamel, while blue light elicits yellow fluorescence. Alterations in these patterns can be used to detect missing or decalcified tooth structure (in the case of dental caries), as well as the presence of a restorative material. The results suggest that the greatest difference in fluorescence patterns occurs for excitation at 405 nm, when used in

combination with an orange high pass (520 nm) filter. This appears to be the best suited for identifying restorative materials when they match the shade of the adjacent tooth structure.

Finally, the present work extends the understanding of differences in fluorescence properties due to variations in shade for the same material, an aspect that has to date received limited attention. Future studies could explore other materials using a similar methodology, to generate a database that could assist in identifying the material type and brand, for forensic purposes.

8.2. Study 3 (Chapter 5)

Many past studies of methods to identify restorations for forensic purposes have focused on more elaborate laboratory methods, such as SEM with EDS. Such methods are indeed elegant and informative, but are time consuming, and require specialised equipment and considerable technical skill. As well, such methods are not applicable to clinical situations, or to forensic odontology in the event of a mass disaster.

The positive results of study 1 led to the proposal of the fluorescence aided identification of restorations (FAIR) technique, using 405 nm light. This is a progression or refinement of the body of past work on fluorescence identification methods. As shown in studies 3 and 4, the FAIR method is superior to examination using white light, and also surpasses DiFOTI for the detection of

restorations. This result has application to clinical practice and to forensic inspections.

8.3. Study 4 (Chapter 6)

The FAIR method has further application for the selective removal of composite resin materials from teeth when these restorations are being replaced. This material is used widely and is the direct placement material of choice for both anterior and posterior restorations in most clinical situations. Since repair and replacements of composite resins is an everyday task of dentists, a better method for clearly differentiating the margins of the restorative material and tooth structure is a useful development. This technique can reduce unnecessary and accidental removal of healthy tooth structure, and thereby ensure optimisation of the structural integrity of the remaining tooth structure. The same method could theoretically be used to detect residues of other resin materials, such as resin cements under veneers, onlays and crowns, fissure sealants or orthodontic bonding resins. This could be a direction for future work.

The FAIR method can also be employed for selective removal of ceramic and hybrid materials, as these materials can be also be clearly differentiated from tooth structure. The basic principle is that the restorative material differs in its fluorescence properties from tooth structure. A general pattern was that most composite resins and ormocers fluoresce more intensely than tooth structure, while ceramics and hybrid ceramics fluoresce less, and therefore appear dark.

Regardless, they can still be differentiated from normal tooth structure. Further studies could explore this aspect, taking the current work and extending it into the clinical setting.

8.4. Study 5 (Chapter 7)

This part of the thesis adds to existing literature on the thermal degradation of various dental materials and extends that by showing how the fluorescence properties are affected by the temperature at which the sample was treated. The forensic literature attests to the significance of dental restorations in identification of cremains. Past literature has explored techniques such as SEM with EDS and XRF as ways of identifying the brand or type of restorations. These methods identify charred restorative materials based on their elemental composition. The present study assessed the value of a simpler approach using fluorescence. A key aspect of the work was that the temperatures employed were relevant to particular types of fires, and that the materials tested were commonly used contemporary tooth-coloured restorative materials.

The results from this part of the work showed that all the tested restorative materials, irrespective of their material type, contain some type of glass or ceramic fillers as an inorganic component. These glass and ceramic fillers have high melting points. The consequence of this is that the ceramic component of resin materials showed less degradation than the resin component when the materials were heated to temperatures above 1000 ◦C.

Further, it was found that tooth-coloured restorative materials which contain inorganic fluorophores would retain some of their fluorescence properties after heating. Although most materials lost their fluorescence properties at 500 ◦C, Dentsply TPH Spectra® exhibited some red fluorescence after heating to 900 ◦C, and the glass ionomer materials from GC Corporation retained their pink-red fluorescence (attributed to strontium-based glass components), even at high temperatures. These particular materials could potentially be identifiable because of these properties after a fire.

Chapter 9 Practical considerations and Conclusion

9.1. Practical Considerations

9.1.1. Study 1 (Chapter 3)

In clinical practice and in forensic examinations of deceased persons, by illuminating teeth with 405 nm violet light, it is possible to identify the presence of most types of tooth-coloured restorative material, because of the difference in their fluorescence emissions compared to natural tooth structure.

9.1.2. Study 2 (Chapter 4)

Within a given tooth-coloured restorative material, the lighter shades (for example, shade B1) show stronger fluorescence emissions than the darker shades. This effect, which is attributed to the addition of greater amounts of fluorophores, may help to create a more lifelike effect when these light shades are used to replace dental enamel in a layered restoration.

9.1.3. Study 3 (Chapter 5)

The FAIR technique assists dental clinicians in detecting the presence of tooth-coloured restorations in teeth, and is superior to conventional clinical examination and to DiFOTI. This technique may be useful for dental charting in dental patients as part of everyday clinical practice, as well as in the forensic examination of deceased persons.

9.1.5. Study 4 (Chapter 6)

The FAIR technique assists dental clinicians when they are removing tooth-coloured materials from teeth, since it allows them to distinguish between these materials and natural tooth structure. The FAIR technique may have value in clinical situations where tooth-coloured materials need to be removed, replaced

or repaired. The FAIR technique could be used for guided removal of restorative materials, as well as resin cements, fissure sealants, and bonding resins used under orthodontic brackets and orthodontic bands.

9.1.4. Study 5 (Chapter 7)

When performing a forensic examination of the teeth from persons who have died in a fire event, the response of certain materials to high temperatures may help to identify these in cremains. The way that restorative materials respond when exposed to high temperatures varies according to their composition. Resin-based restorative materials still fluoresced at 200 ◦C, and at 500 ◦C underwent major colour changes due to volatilisation of resin. Materials containing inorganic fluorophores still fluoresced at 900 ◦C. GIC materials from GC Corporation retained red fluorescence across most elevated temperatures that were tested. Identification of such patterns may help in identifying the material type and brand that is present. Such information could potentially assist in victim identification in cases of fire accidents.

9.2. Conclusion

This thesis has explored the overarching question, “Can tooth-coloured restorations be identified accurately under UVA-violet light illumination?”, and has demonstrated, in a simulation laboratory environment, a range of practical implications for clinical dentistry and forensic odontology from the use of violet light-induced fluorescence.

A number of areas for further work have been identified and discussed in the thesis. These include how the FAIR technique would alter the effectiveness of dental examination and charting, and of replacing tooth-coloured restorations in real world clinical practice settings, where results may be different than those obtained in a simulation laboratory setting. The promising finding that FAIR allows the operative dentistry procedure to be completed in a shorter time, and with less removal of natural tooth structure, indicates that there are benefits both for practitioners as well as for dental patients.

Finally, the work in this thesis has applied the fluorescence identification technique in several ways, such as through an illuminating ring on a clinical camera, or through a LED array that is used to replace a normal dental operating light. Such methods could be adopted readily in dental clinical practice and in forensic odontology.

There may be other ways to apply the same concept of fluorescence inspection, for example, in an intra-oral camera or in a dental handpiece, with the ability to choose between white LED illumination and 405 nm LED illumination. Using an intraoral camera would have the advantage of documentation, by recording images under both white and violet light. Having white and violet light sources built into a dental handpiece would simplify the approach for restorative dentistry. It would be worthwhile exploring how such devices could be used to improve dental clinical practice and forensic examination in real world situations.

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Appendices

Appendix A. Figures of Line graphs and samples, and Statistical analysis tables for study 2- Chapter 4

Appendix A (Figures and Tables for study 2—Chapter 4)

Table representing the p values, showing the variation between two material types

Material	Comparison materials	Mean Difference	Wilcoxon Signed-rank test	P value
3M Filtek™Z250	Aura	-9.389	485.00	0.79
	Herculite™ Ultra	-23.964	524.00	0.013*
	TPH Spectra® LV	-90.456	40.00	0.00*
	G- aenial™	-156.125*	0.00	0.00*
	Gradia®Direct anterior	-152.417*	0.00	0.00*
	Gradia®Direct X	-157.500*	0.00	0.00*
	Filtek™	-51.500	109.50	0.00*
	Filtek™ Supreme XTE	-144.800*	184.00	0.045*
Aura	HerculiteTM Ultra	-14.575	875.00	0.28
	TPH Spectra® LV	-81.067	347.00	0.00*
	G- aenialTM	-146.736*	6.00	0.00*
	Gradia®Direct anterior	-143.028*	21.00	0.00*
	Gradia®Direct X	-148.111*	11.50	0.00*
	FiltekTM	-42.111	207.50	0.09
	FiltekTM Supreme XTE	-135.411	299.50	0.301
Herculite™ Ultra	TPH Spectra® LV	-66.492	771.0	0.00*
	G- aenialTM	-132.161	18.50	0.00*
	Gradia®Direct anterior	-128.452*	53.50	0.00*
	Gradia®Direct X	-133.536*	32.50	0.00*
	FiltekTM	-27.536	414.50	0.085
	FiltekTM Supreme XTE	-120.836	534.00	0.75

TPH Spectra® LV	G- aenial™	-65.669	91.00	0.00*
	Gradia®Direct anterior	-61.961	248.50	0.00*
	Gradia®Direct X	-67.044	150.00	0.00*
	Filtek™	38.956	370.50	0.002*
	Filtek™ Supreme XTE	-54.344	198.00	0.00*
G- aenial™ Posterior	Gradia®Direct anterior	3.708	263.50	0.62
	Gradia®Direct X	-1.375	219.5	0.91
	Filtek™	104.625	6.00	0.00*
	Filtek™ Supreme XTE	11.325	16.00	0.00*
Gradia®Direct Anterior	Gradia®Direct X	-5.083	441.5	0.39*
	Filtek™	100.917	16.00	0.00*
	Filtek™ Supreme XTE	7.617	37.00	0.00*
Gradia®Direct X	Filtek™	106.000	8.50	0.00*

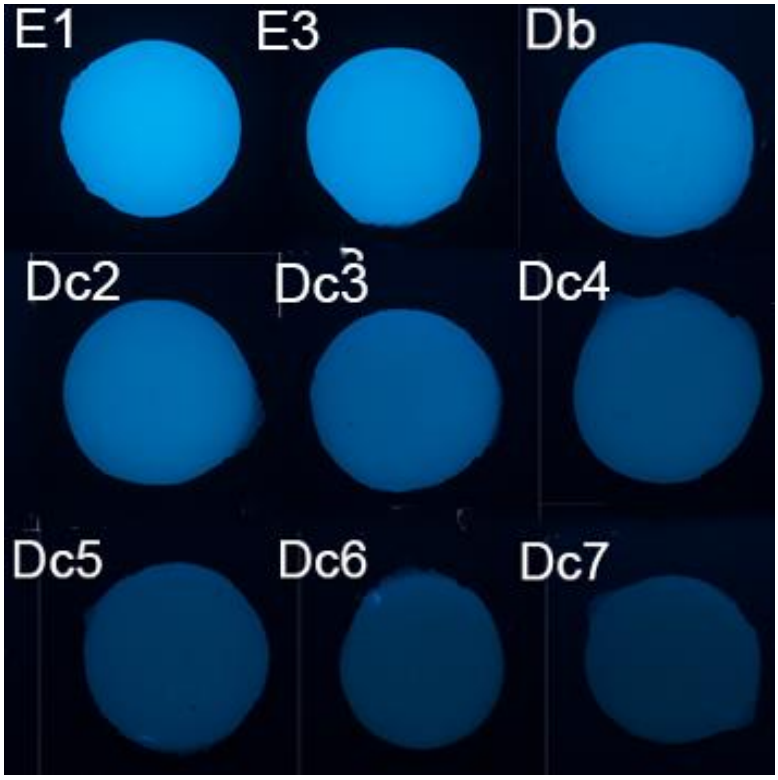


Figure A1.1. A Violet light fluorescence image showing all the shades of Aura (resin-based composite) included in the study. The fluorescence emission is in descending order from Enamel shades to dentine shades.

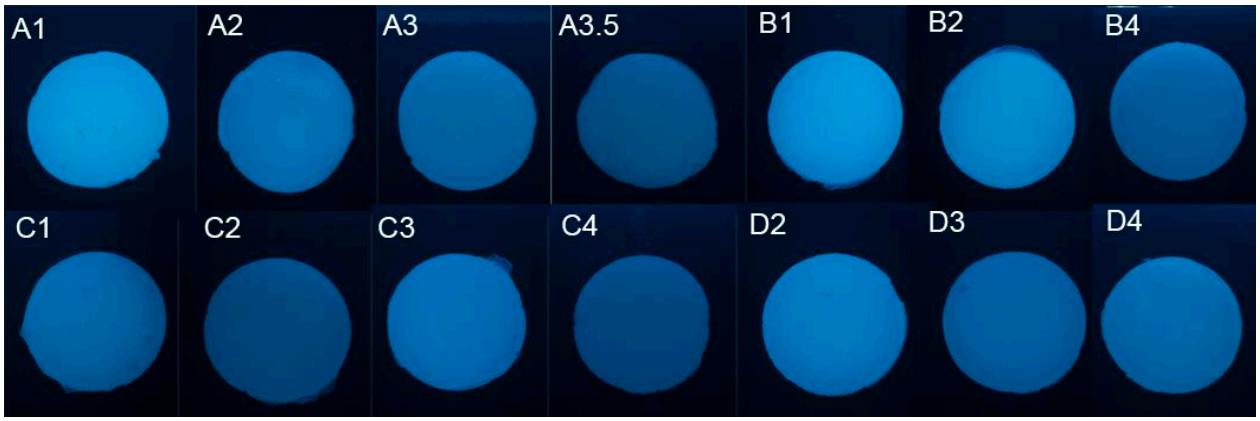
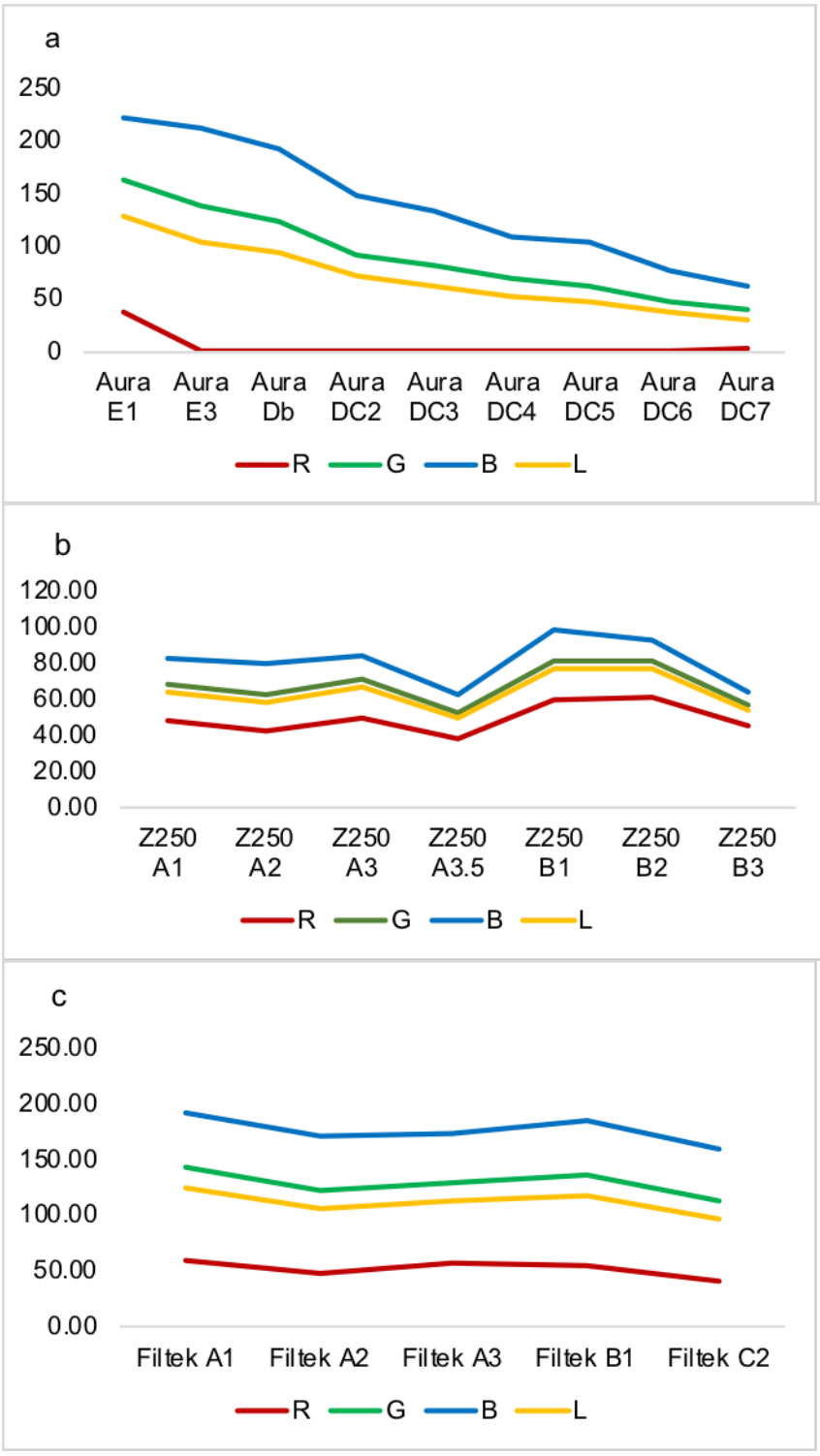
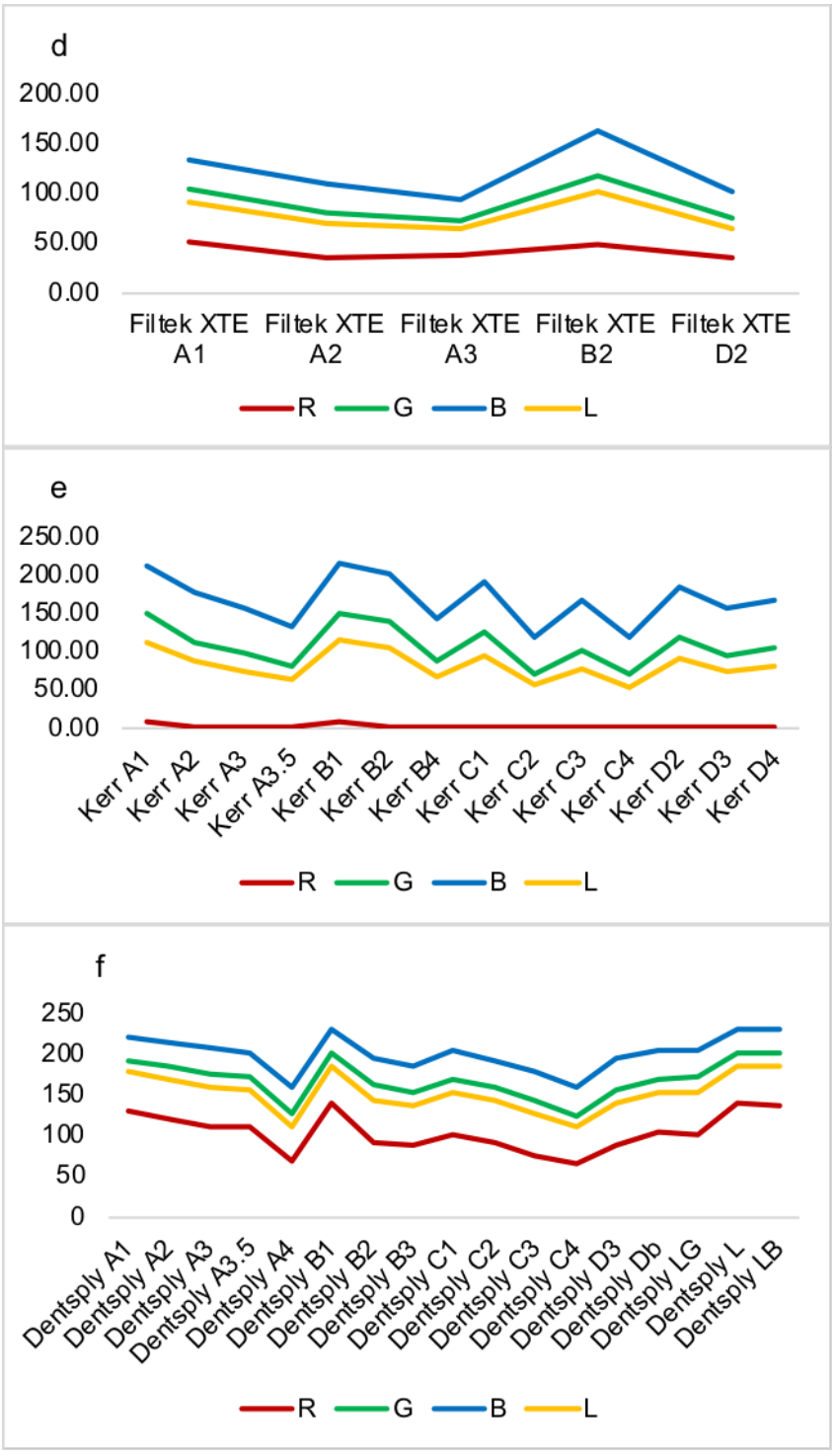


Figure A1.2. A Violet light fluorescence image showing all the shades of Kerr (resin-based composite) included in the study. A1, B1 and D2 the lightest shades showing the maximum fluorescence emission.

Line diagrams for study 2—Chapter 4

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Kerr A2
Kerr A3
Kerr A3.5
Kerr B1
Kerr B2
Kerr B4
Kerr C1
Kerr C2
Kerr C3
Kerr C4
Kerr D2
Kerr D3
Kerr D4
R
G
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f
250
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100
50
0
Dentsply A1
Dentsply A2
Dentsply A3
Dentsply A3.5
Dentsply A4
Dentsply B1
Dentsply B2
Dentsply B3
Dentsply C1
Dentsply C2
Dentsply C3
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Dentsply D3
Dentsply Db
Dentsply LG
Dentsply L
Dentsply LB
R
G
B
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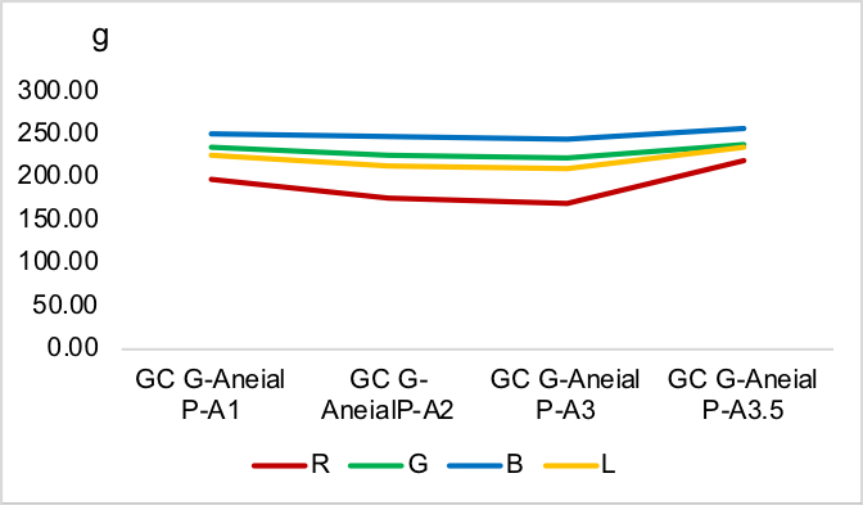
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GC G-Aneial P-A1
GC G-AneialP-A2
GC G-Aneial P-A3
GC G-Aneial P-A3.5
R
G
B
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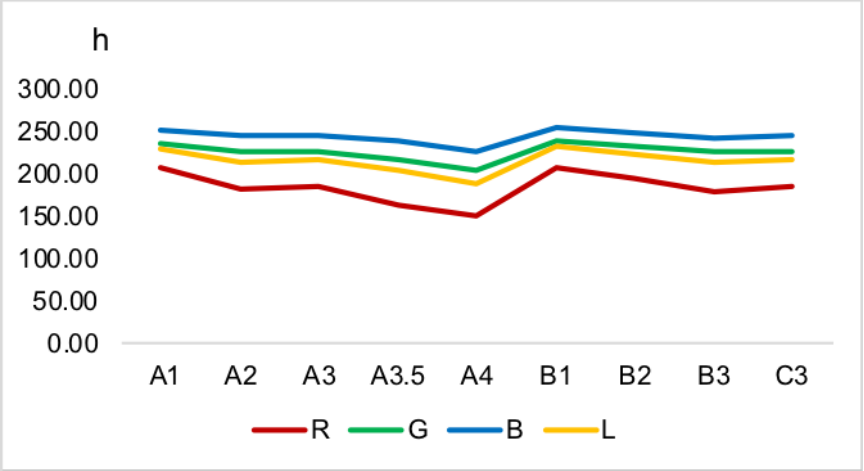
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G
B
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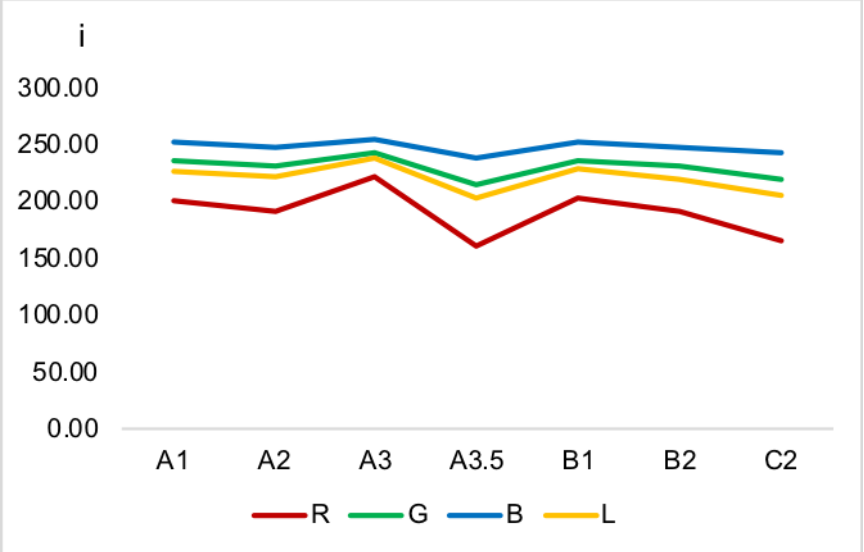
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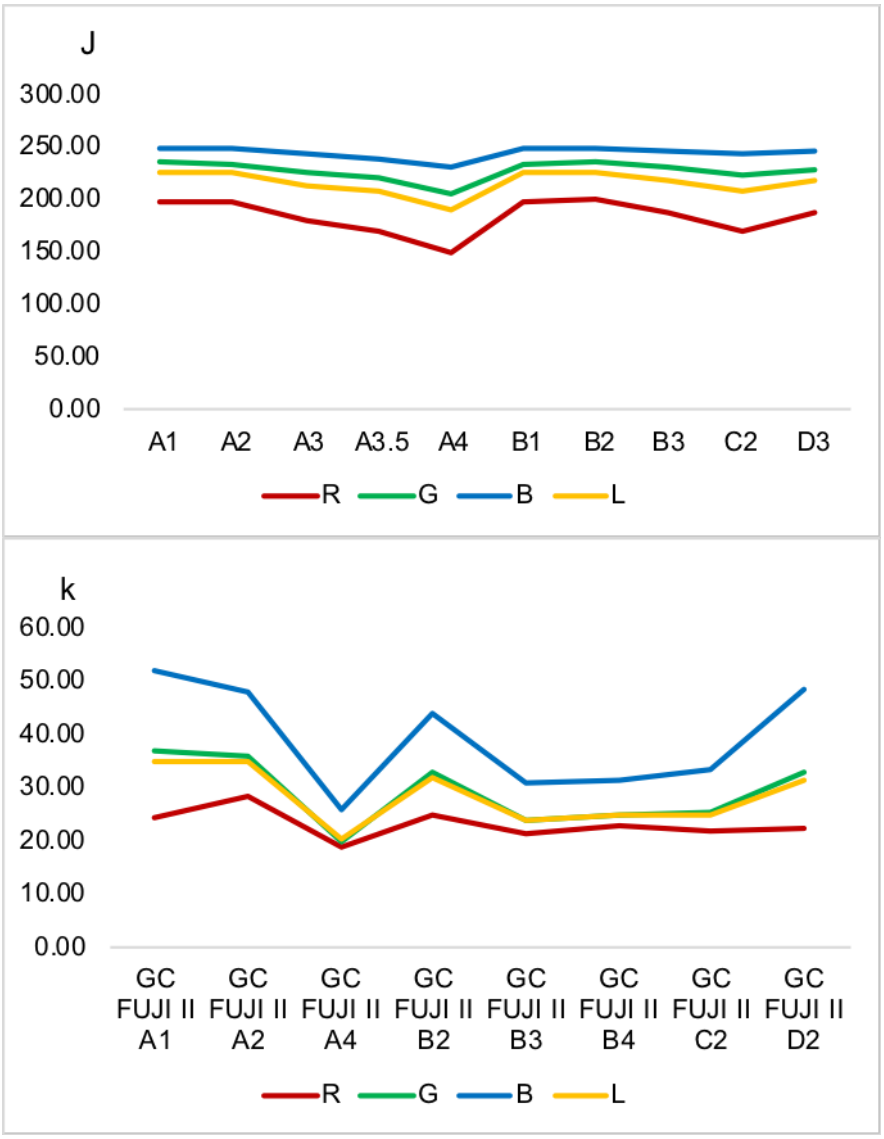
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D3
R
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k
60.00
50.00
40.00
30.00
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GC FUJI II A1
GC FUJI II A2
GC FUJI II A4
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R
G
B
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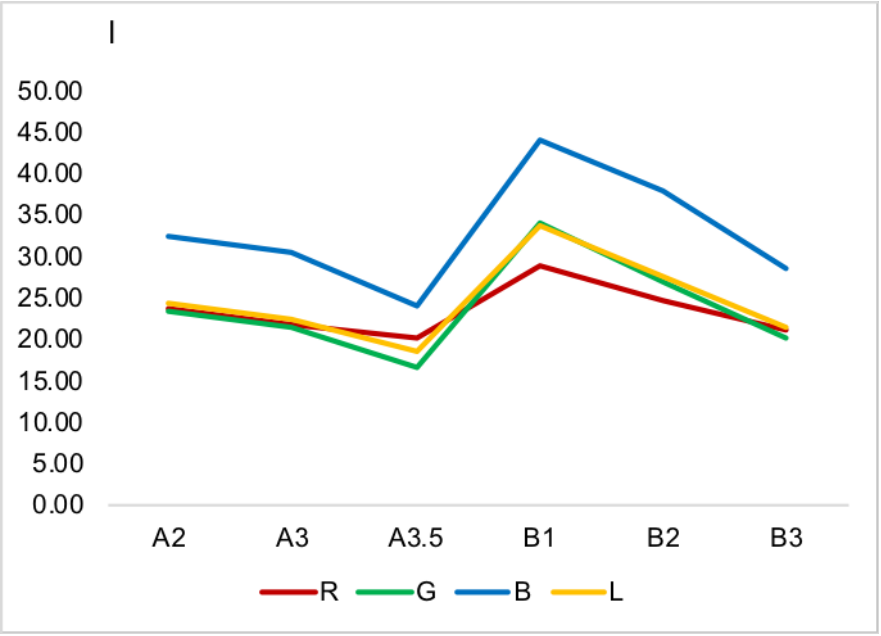

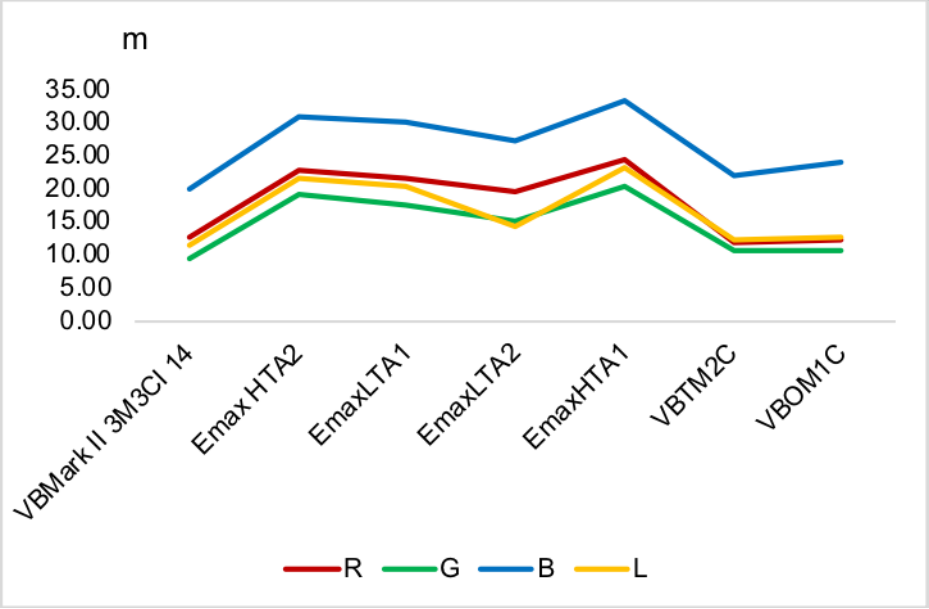

Figure 1. Distribution of colour coordinates *R G B* and fluorescence emission *L* values among the shades of each brand type of tooth coloured restorative materials. a. Aura, b. Filtek™ Z250, c. Filtek™, d. Filtek™ Supreme XTE, e. Herculite™ Ultra, f. TPH Spectra® LV, g. G-aenial®, h. Gradia® Direct Anterior, I. Gradia®Direct X, J. Admira Fusion, K. Fuji II, l. Fuji IX, m. Ceramics.


a
35.00
30.00
25.00
20.00
15.00
10.00
5.00
0.00
VBMark II 3M3CI 14
Emax HTA2
EmaxLTA1
EmaxLTA2
EmaxHTA1
VBTM2C
VE2M2T
VE2M2HT
VBOM1C
R
G
B
L
b
30.00
25.00
20.00
15.00
10.00
5.00
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VBMark II 3M3CI 14
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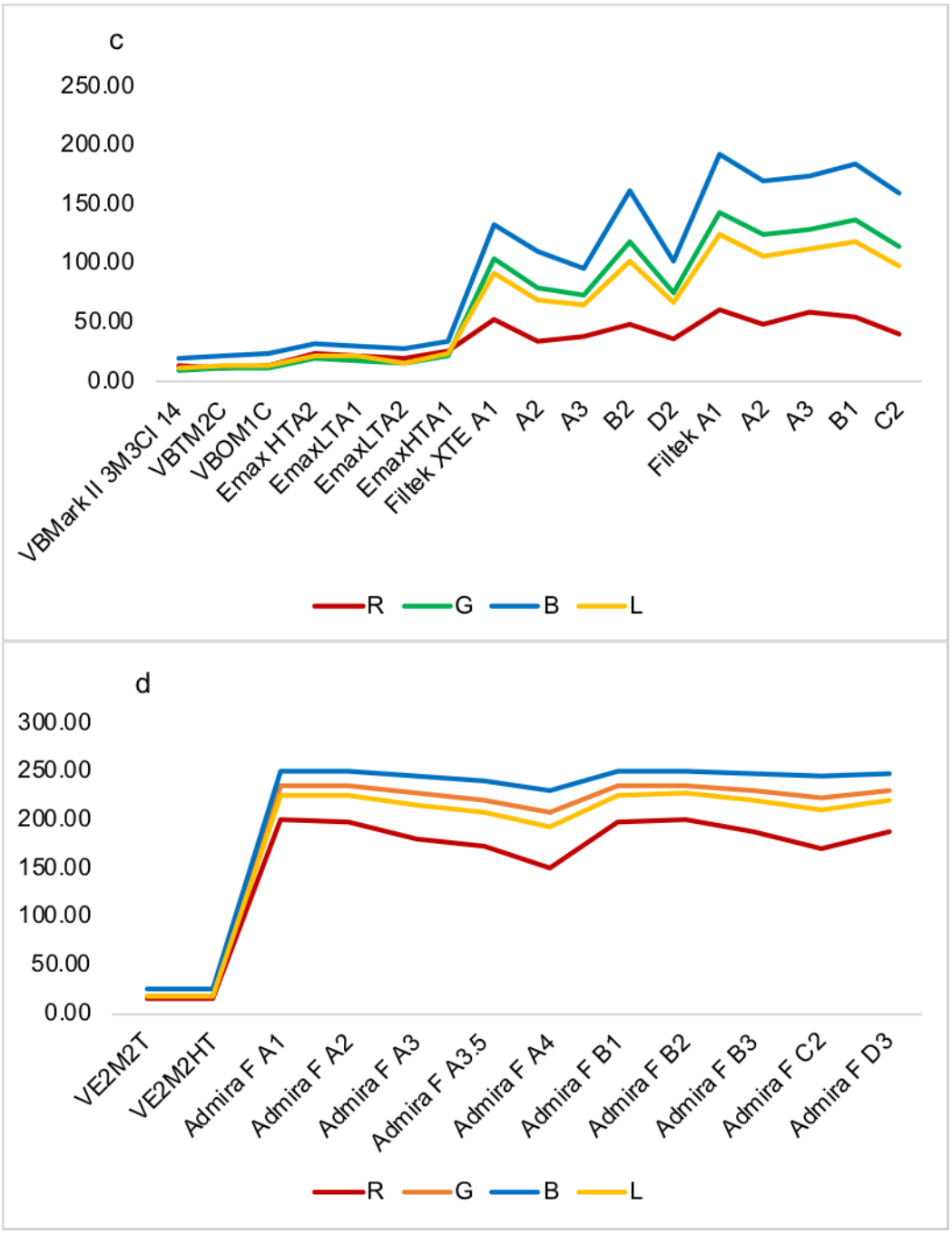
VE2M2HT
R
G
B
L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200.00
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VBTM2C
VBOM1C
Emax HTA2
EmaxLTA1
EmaxLTA2
EmaxHTA1
Filtek XTE A1
A2
A3
B2
D2
Filtek A1
A2
A3
B1
C2
R
G
B
L
d
300.00
250.00
200.00
150.00
100.00
50.00
0.00
VE2M2T
VE2M2HT
Admira F A1
Admira F A2
Admira F A3
Admira F A3.5
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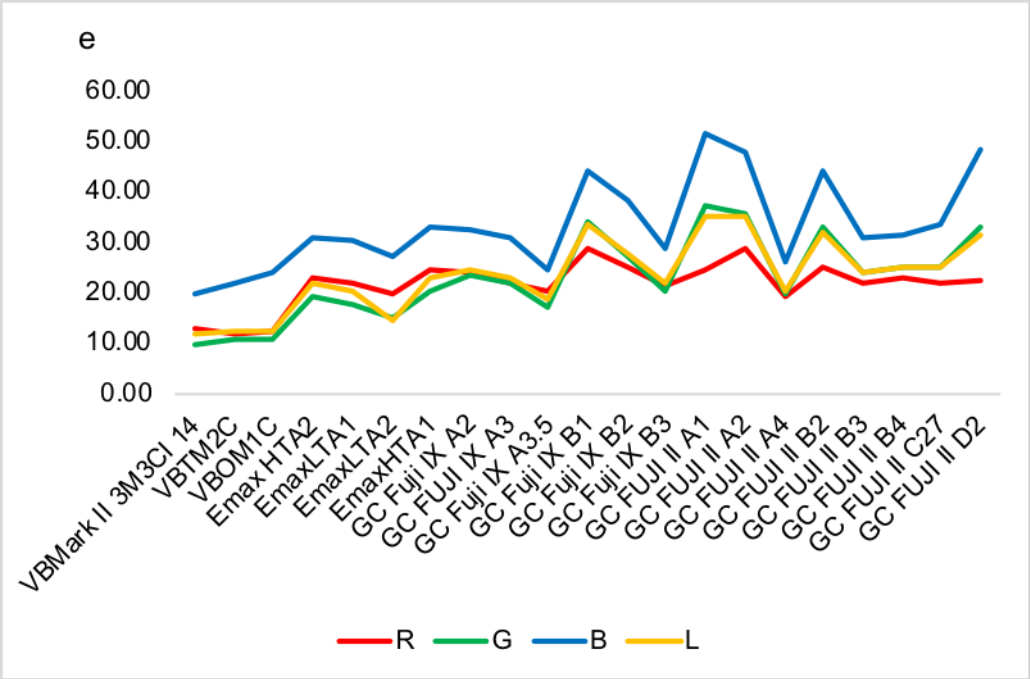
Admira F B2
Admira F B3
Admira F C2
Admira F D3
R
G
B
L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50.00
40.00
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10.00
0.00
VBMark II 3M3CI 14
VBTM2C
VBOM1C
Emax HTA2
EmaxLTA1
EmaxLTA2
EmaxHTA1
GC Fuji IX A2
GC FUJI IX A3
GC Fuji IX A3.5
GC Fuji IX B1
GC Fuji IX B2
GC Fuji IX B3
GC FUJI II A1
GC FUJI II A2
GC FUJI II A4
GC FUJI II B2
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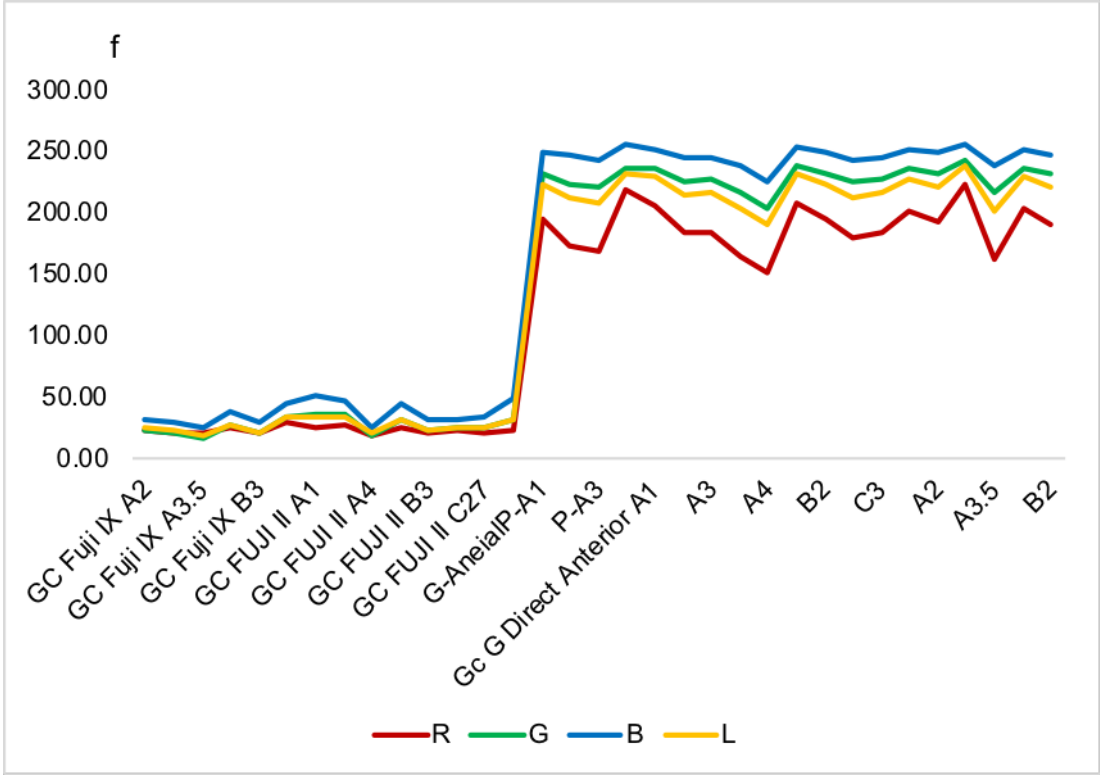
GC FUJI II B4
GC FUJI II C27
GC FUJI II D2
R
G
B
L


f
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200.00
150.00
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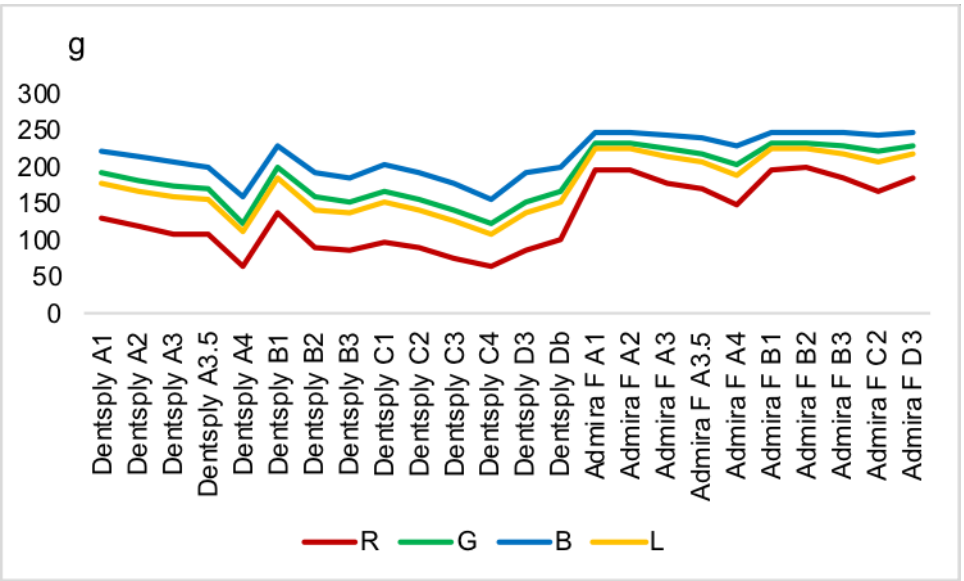
50.00
0.00
GC Fuji IX A2
GC Fuji IX A3.5
GC Fuji IX B3
GC FUJI II A1
GC FUJI II A4
GC FUJI II B3
GC FUJI II C27
G-AneialP-A1
P-A3
Gc G Direct Anterior A1
A3
A4
B2
C3
A2
A3.5
B2
R
G
B
L



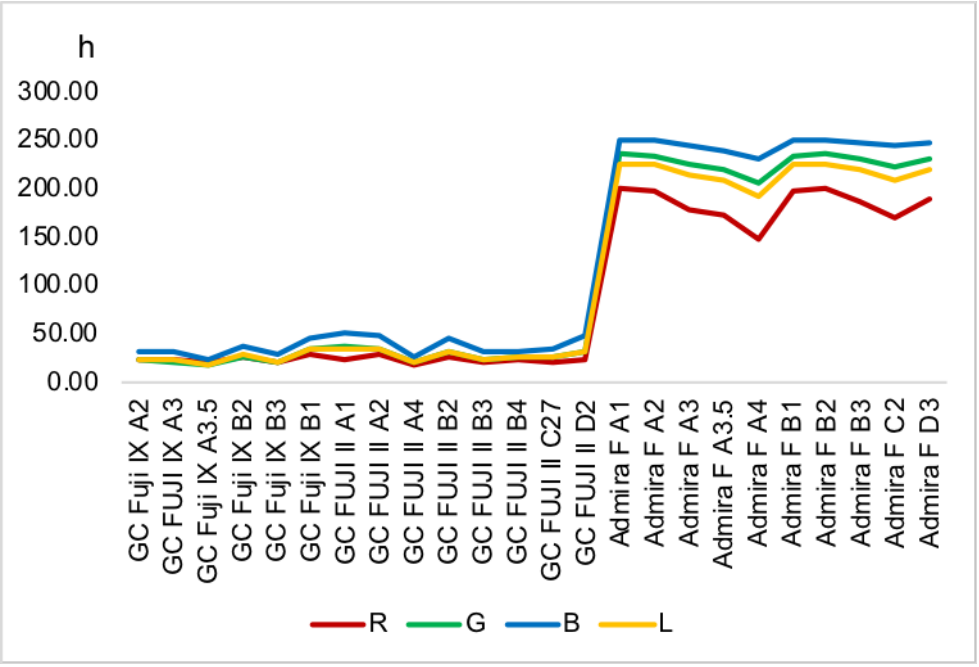


Figure 2. Comparison of colour coordinates *R G B* and *L* (fluorescence emission) values among the shades of different material types

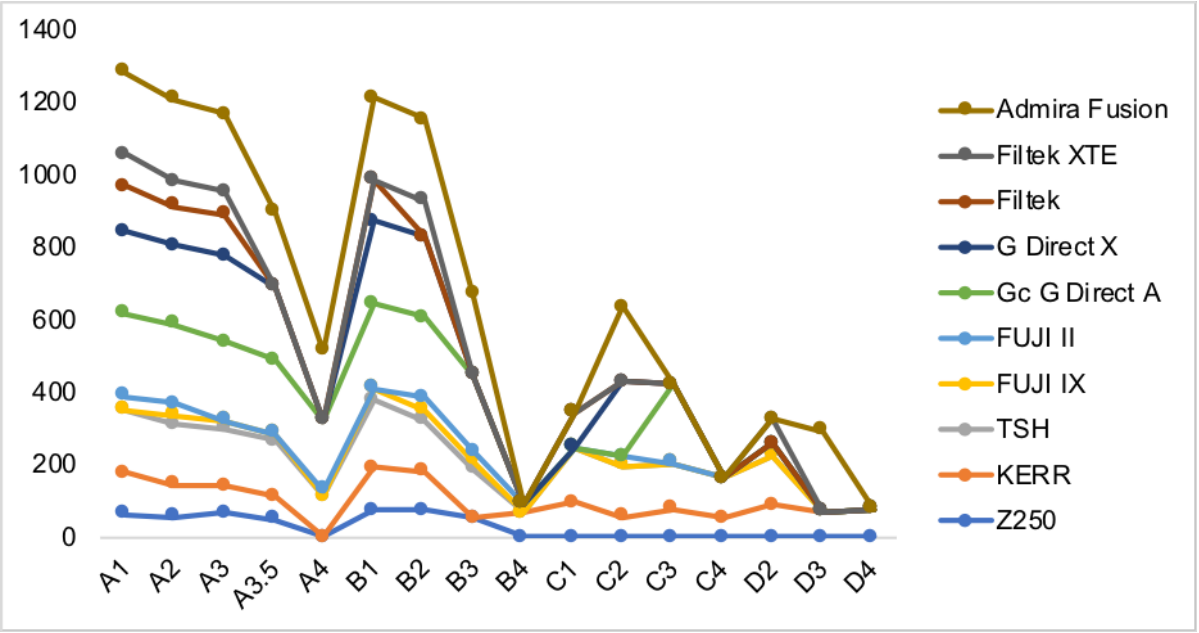


Figure 3. Distribution of Luminosity values across the shades for direct tooth coloured materials

Appendix B: Colour plates and data graphs of study 5- chapter 7

Appendix B

In this appendix images of teeth samples and disc samples before and after heat treatment for each material type tested in the study 3 are included, also data and graphs of study 3 are included.

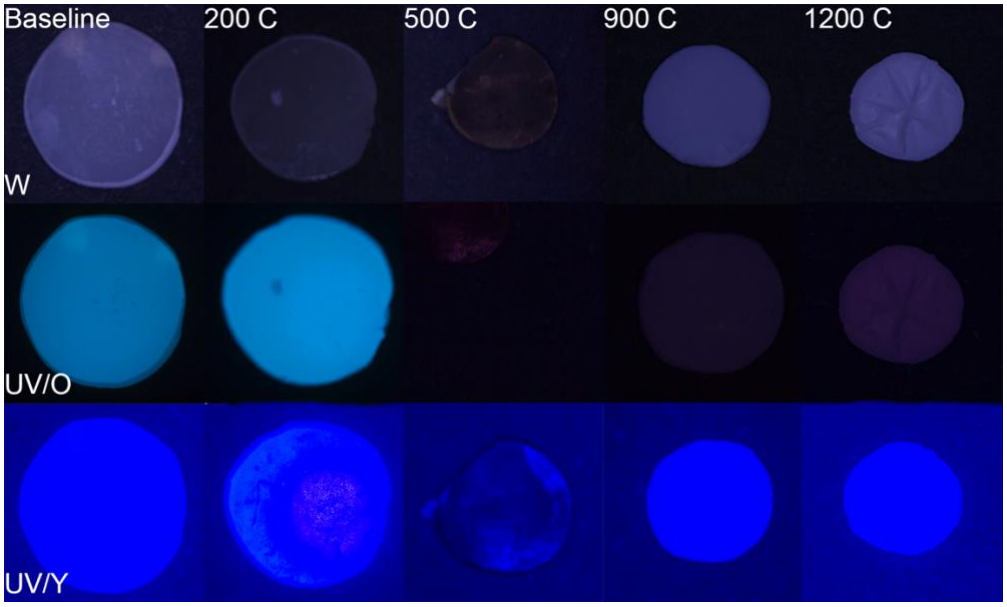


Figure 1. Image of disc samples comparing the baseline images (Pre-heat treatment) to post-heat treatment at 200 °C, 500 °C, 900 °C and 1200 °C, when excited with white light (W), UV-A light with orange filter (UV/O) and UV-A light with yellow filter for Herculite™ Ultra.

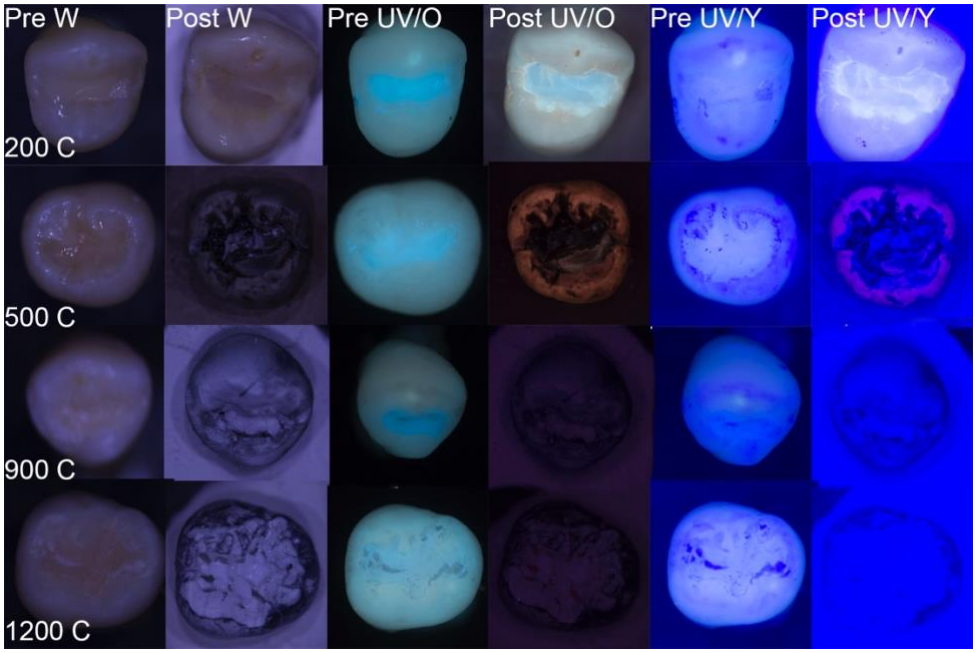


Figure 2. Image of teeth samples comparing the baseline images (Pre-heat treatment) to post-heat treatment at 200 °C, 500 °C, 900 °C and 1200 °C, when excited with white light (W), UV-A light with orange filter (UV/O) and UV-A light with yellow filter for Herculite™ Ultra

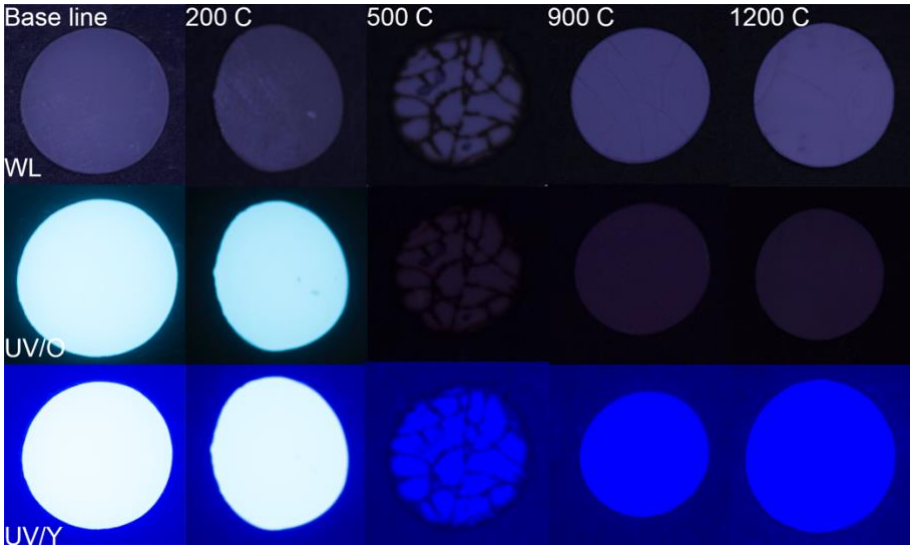


Figure 3. Image of disc samples comparing the baseline images (Pre-heat treatment) to post-heat treatment at 200 °C, 500 °C, 900 °C and 1200 °C, when

excited with white light (W), UV-A light with orange filter (UV/O) and UV-A light with yellow filter for Gradia® Direct X.

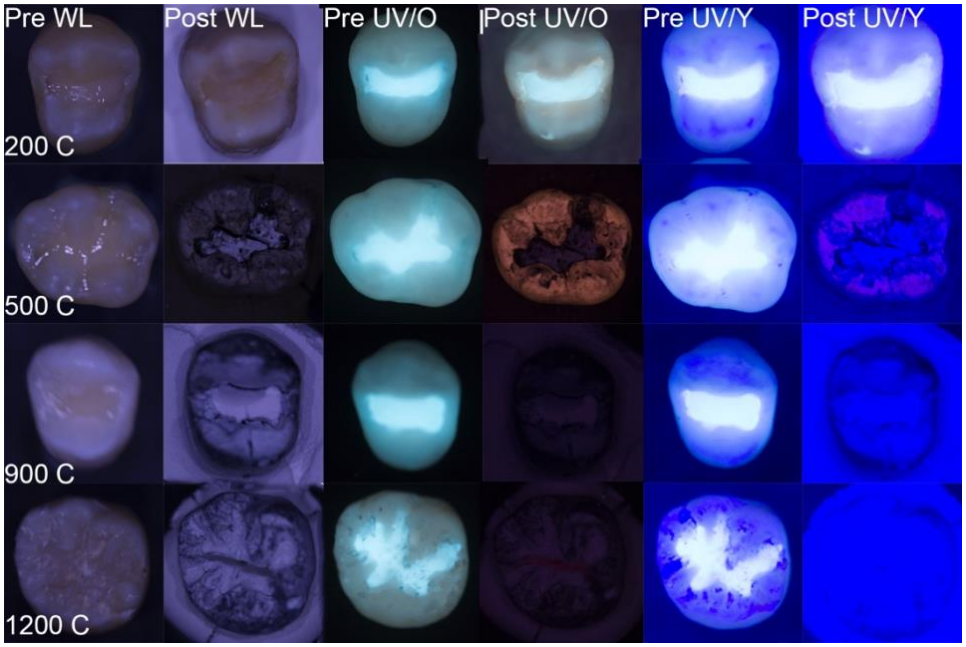


Figure 4. Image of teeth samples comparing the baseline images (Pre-heat treatment) to post-heat treatment at 200 °C, 500 °C, 900 °C and 1200 °C, when excited with white light (W), UV-A light with orange filter (UV/O) and UV-A light with yellow filter for Gradia® Direct X.

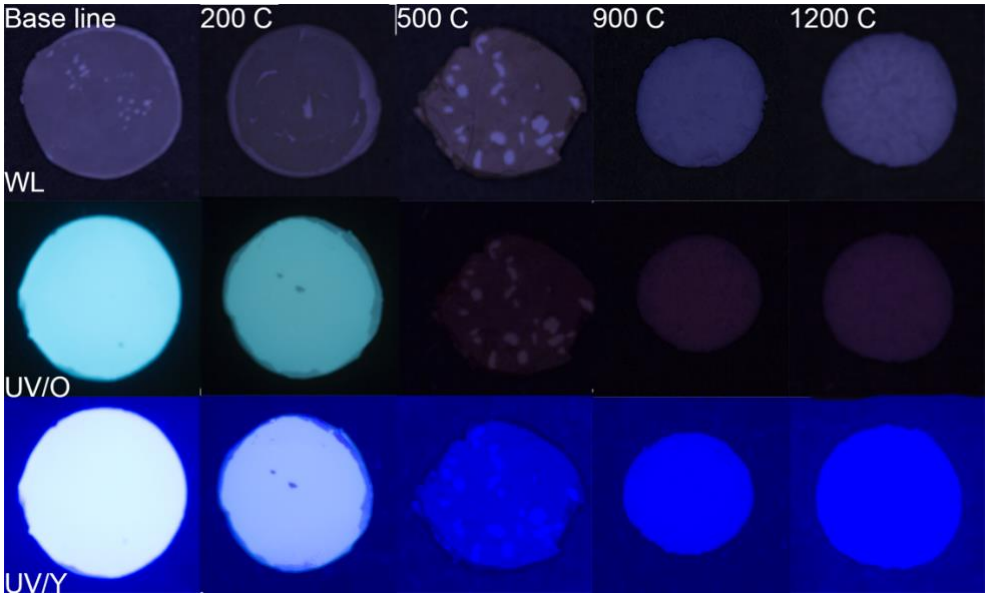


Figure 5. Image of disc samples comparing the baseline images (Pre-heat treatment) to post-heat treatment at 200 °C, 500 °C, 900 °C and 1200 °C, when excited with white light (W), UV-A light with orange filter (UV/O) and UV-A light with yellow filter for Dentsply TPH Spectra®.

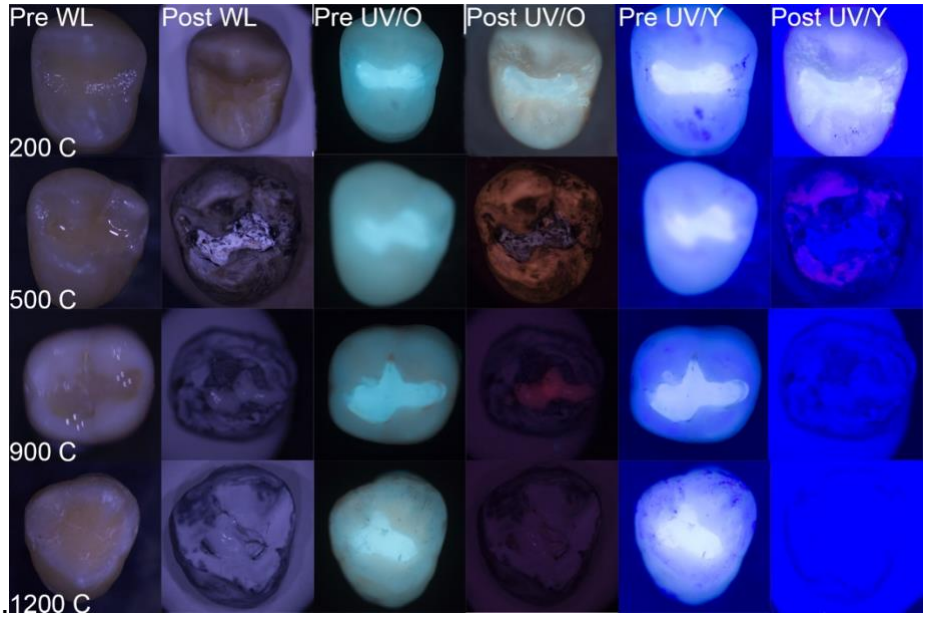


Figure 6. Image of teeth samples comparing the baseline images (Pre-heat treatment) to post-heat treatment at 200 °C, 500 °C, 900 °C and 1200 °C, when excited with white light (W), UV-A light with orange filter (UV/O) and UV-A light with yellow filter for Dentsply TPH Spectra®.

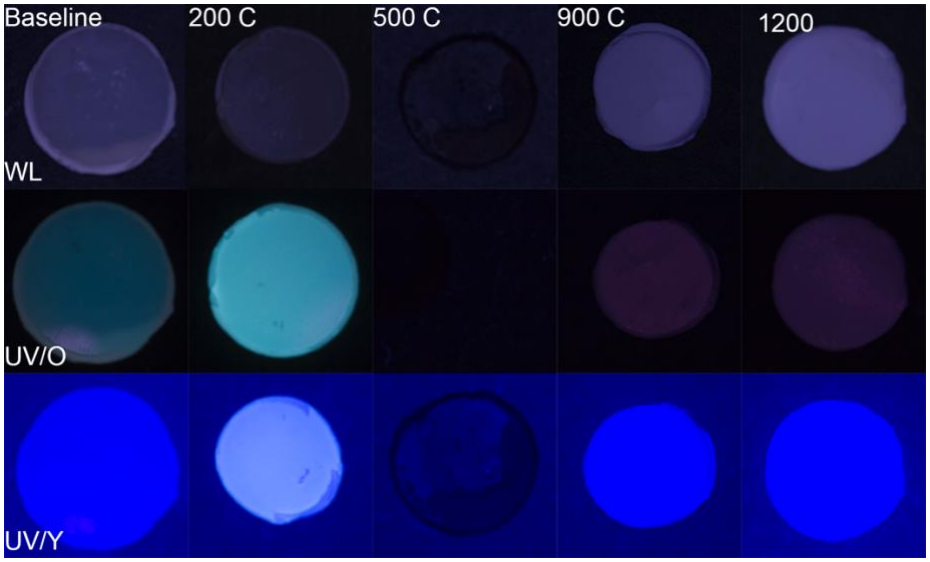


Figure 7. Image of disc samples comparing the baseline images (Pre-heat treatment) to post-heat treatment at 200 °C, 500 °C, 900 °C and 1200 °C, when excited with white light (W), UV-A light with orange filter (UV/O) and UV-A light with yellow filter for 3M Filtek™ supreme XTE.

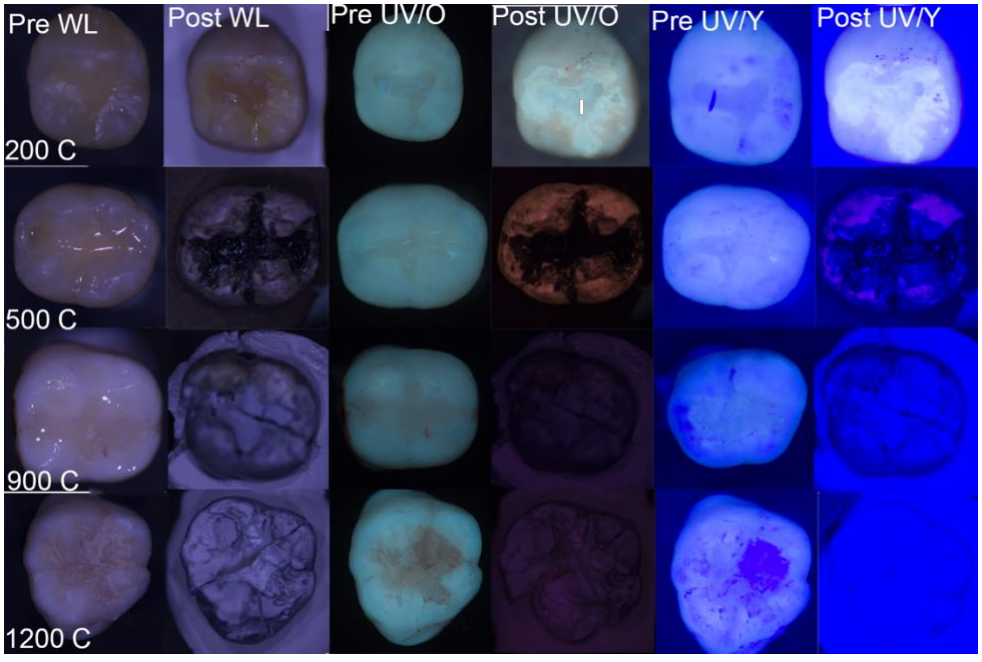


Figure 8. Image of teeth samples comparing the baseline images (Pre-heat treatment) to post-heat treatment at 200 °C, 500 °C, 900 °C and 1200 °C, when

excited with white light (W), UV-A light with orange filter (UV/O) and UV-A light with yellow filter for 3M Filtek™ supreme XTE.

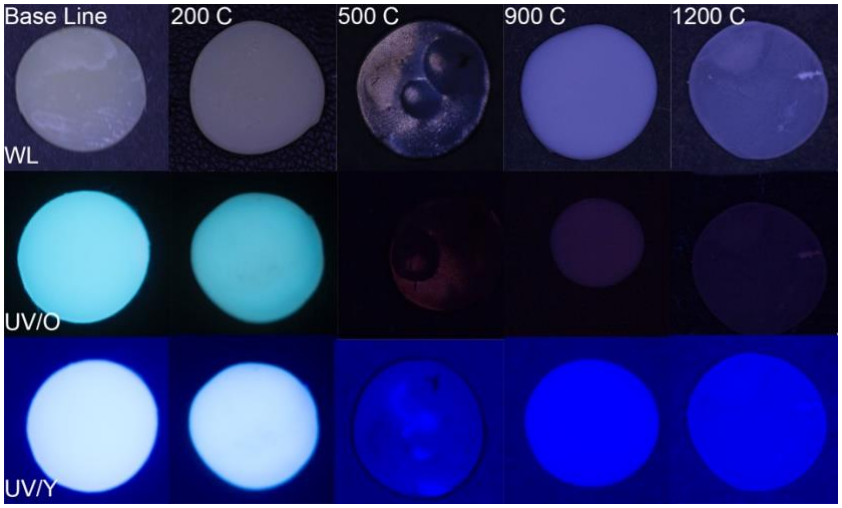


Figure 9. Image of disc samples comparing the baseline images (Pre-heat treatment) to post-heat treatment at 200 °C, 500 °C, 900 °C and 1200 °C, when excited with white light (W), UV-A light with orange filter (UV/O) and UV-A light with yellow filter for 3M Admira Fusion.

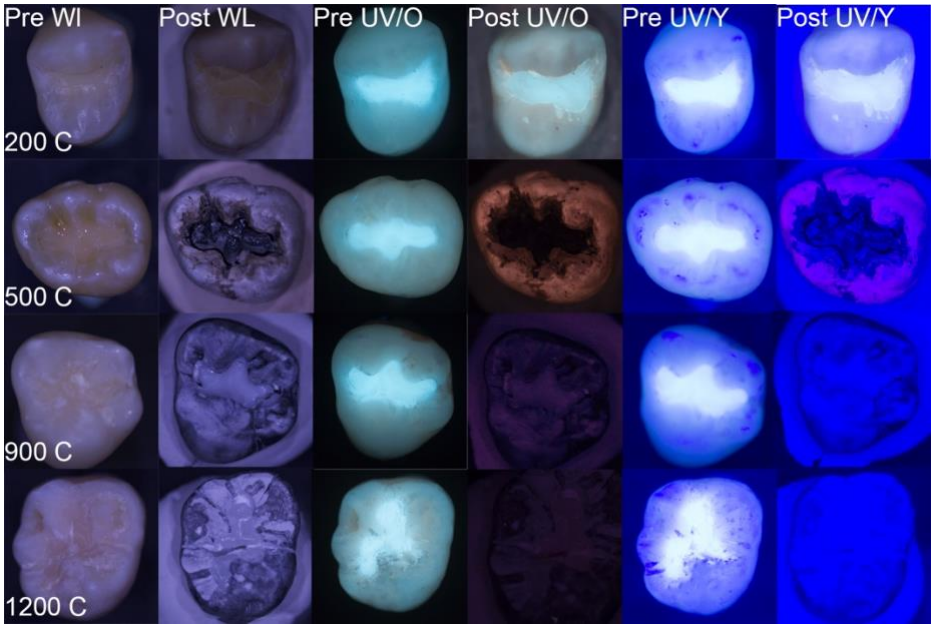


Figure 10. Image of teeth samples comparing the baseline images (Pre-heat treatment) to post-heat treatment at 200 °C, 500 °C, 900 °C and 1200 °C, when excited with white light (W), UV-A light with orange filter (UV/O) and UV-A light with yellow filter for Admira Fusion.

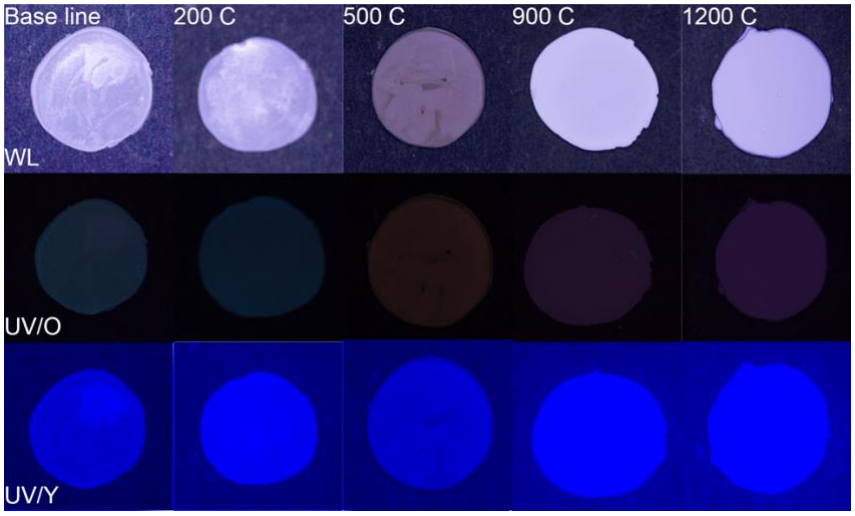


Figure 11. Image of disc samples comparing the baseline images (Pre-heat treatment) to post-heat treatment at 200 °C, 500 °C, 900 °C and 1200 °C, when excited with white light (W), UV-A light with orange filter (UV/O) and UV-A light with yellow filter for 3M GC Fuji II.

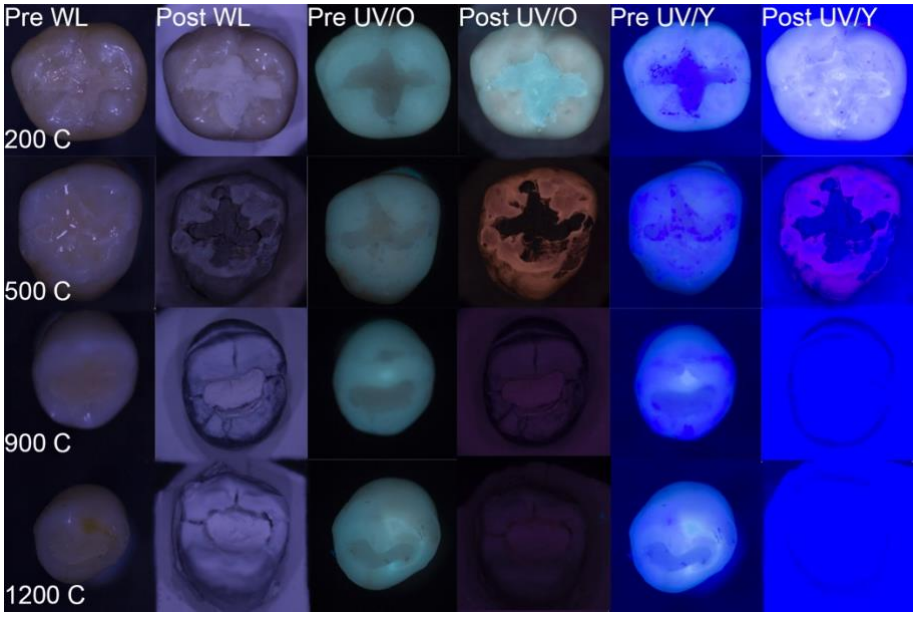


Figure 12. Image of teeth samples comparing the baseline images (Pre-heat treatment) to post-heat treatment at 200 °C, 500 °C, 900 °C and 1200 °C, when excited with white light (W), UV-A light with orange filter (UV/O) and UV-A light with yellow filter forGC Fuji II.

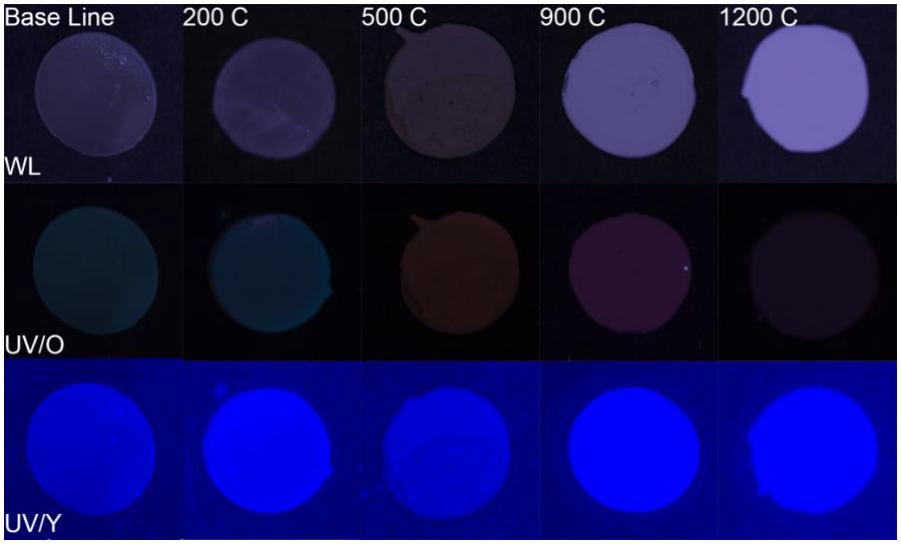


Figure 13. Image of disc samples comparing the baseline images (Pre-heat treatment) to post-heat treatment at 200 °C, 500 °C, 900 °C and 1200 °C, when

excited with white light (W), UV-A light with orange filter (UV/O) and UV-A light with yellow filter for GC Fuji VIII.

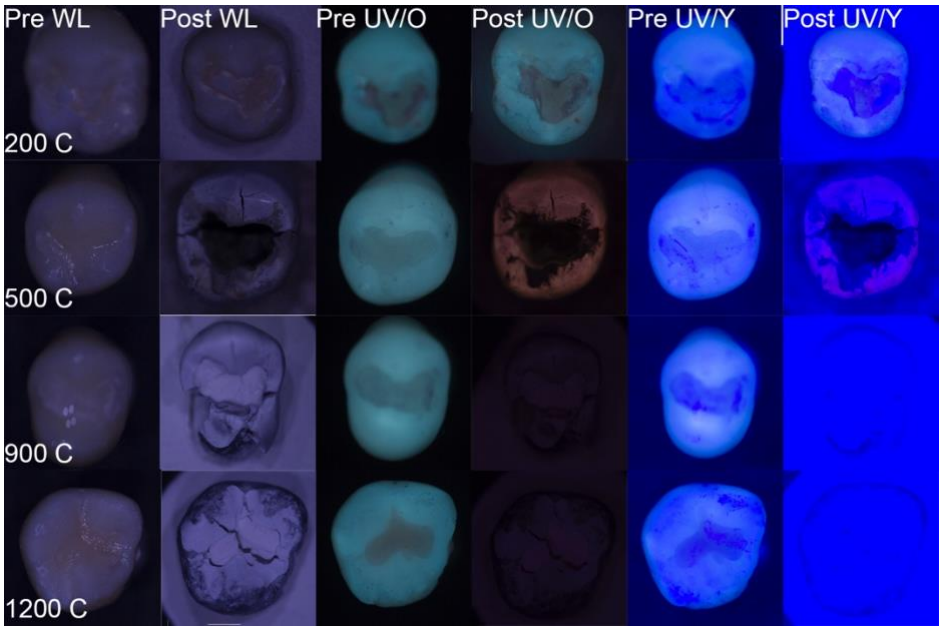


Figure 14. Image of teeth samples comparing the baseline images (Pre-heat treatment) to post-heat treatment at 200 °C, 500 °C, 900 °C and 1200 °C, when excited with white light (W), UV-A light with orange filter (UV/O) and UV-A light with yellow filter for GC Fuji VIII.

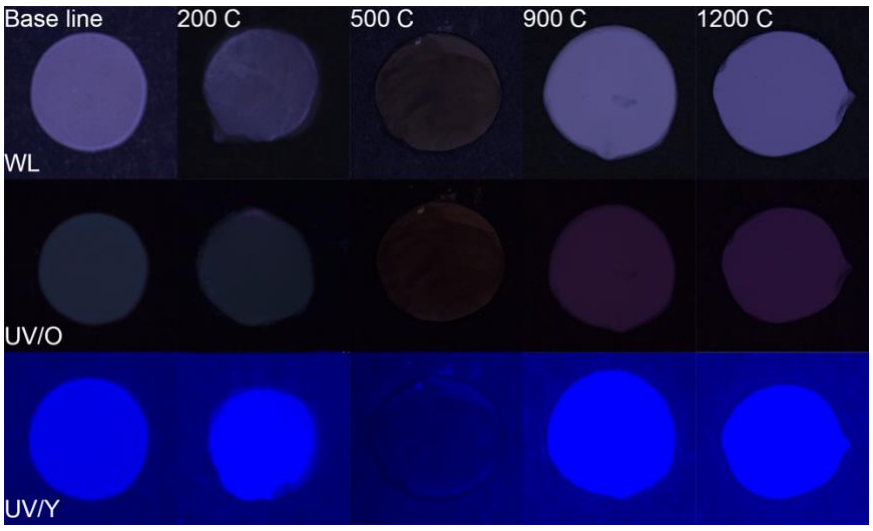


Figure 15. Image of disc samples comparing the baseline images (Pre-heat treatment) to post-heat treatment at 200 °C, 500 °C, 900 °C and 1200 °C, when excited with white light (W), UV-A light with orange filter (UV/O) and UV-A light with yellow filter for GC Fuji IX.

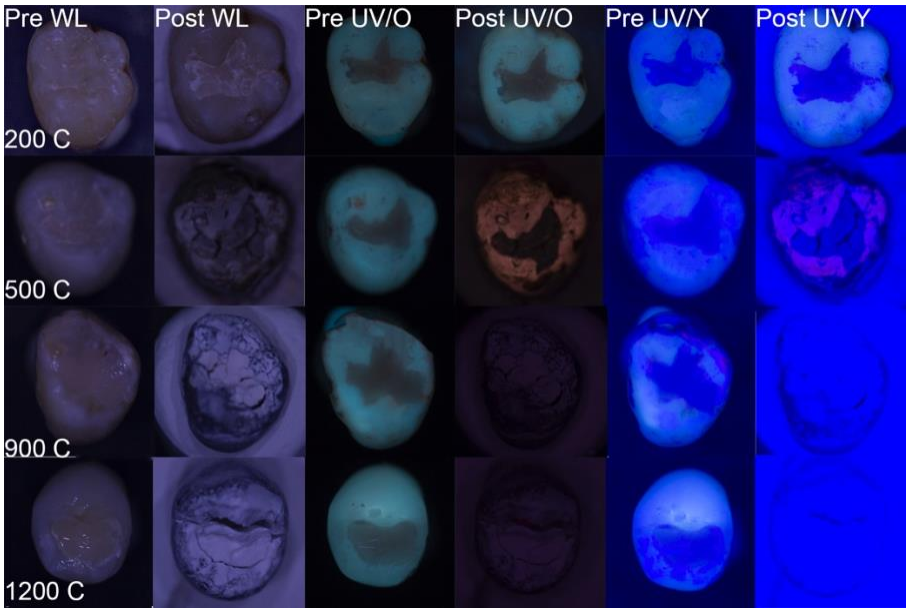


Figure 16. Image of teeth samples comparing the baseline images (Pre-heat treatment) to post-heat treatment at 200 °C, 500 °C, 900 °C and 1200 °C, when excited with white light (W), UV-A light with orange filter (UV/O) and UV-A light with yellow filter for GC Fuji IX.

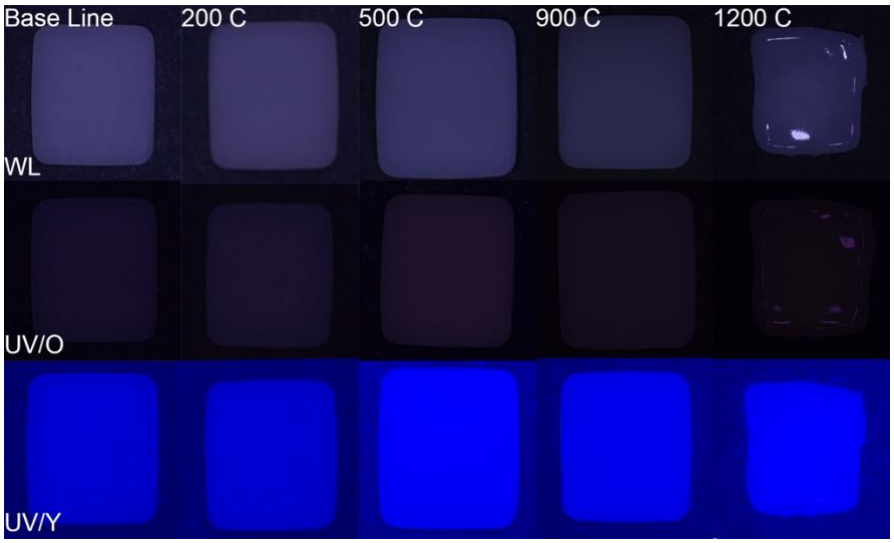


Figure 17. Image of disc samples comparing the baseline images (Pre-heat treatment) to post-heat treatment at 200 °C, 500 °C, 900 °C and 1200 °C, when excited with white light (W), UV-A light with orange filter (UV/O) and UV-A light with yellow filter for VITA BLOC.

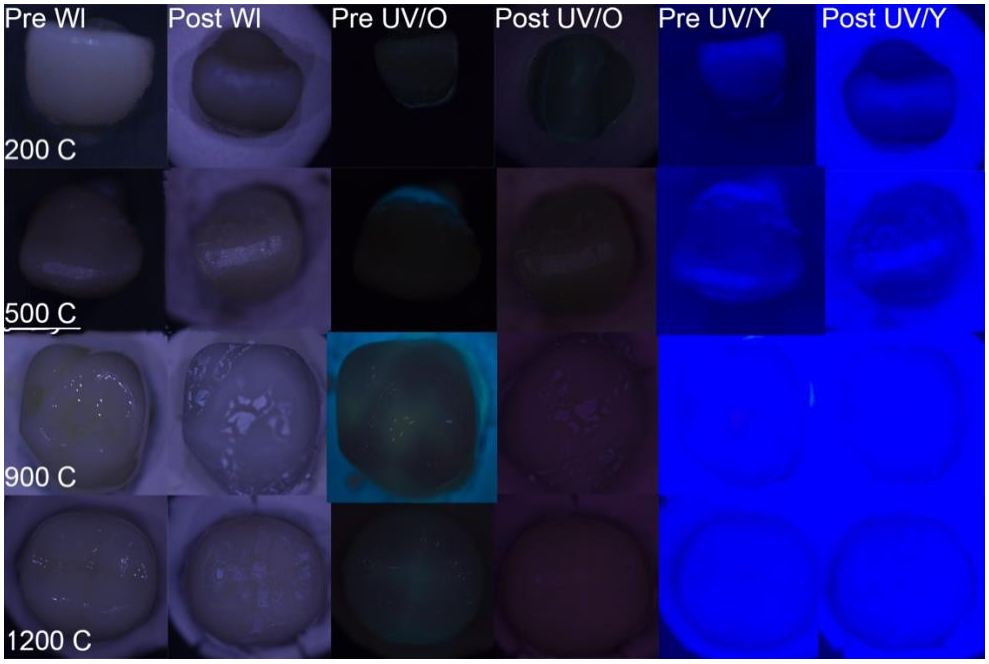


Figure 18. Image of teeth samples comparing the baseline images (Pre-heat treatment) to post-heat treatment at 200 °C, 500 °C, 900 °C and 1200 °C, when excited with white light (W), UV-A light with orange filter (UV/O) and UV-A light with yellow filter for VITABLOC.

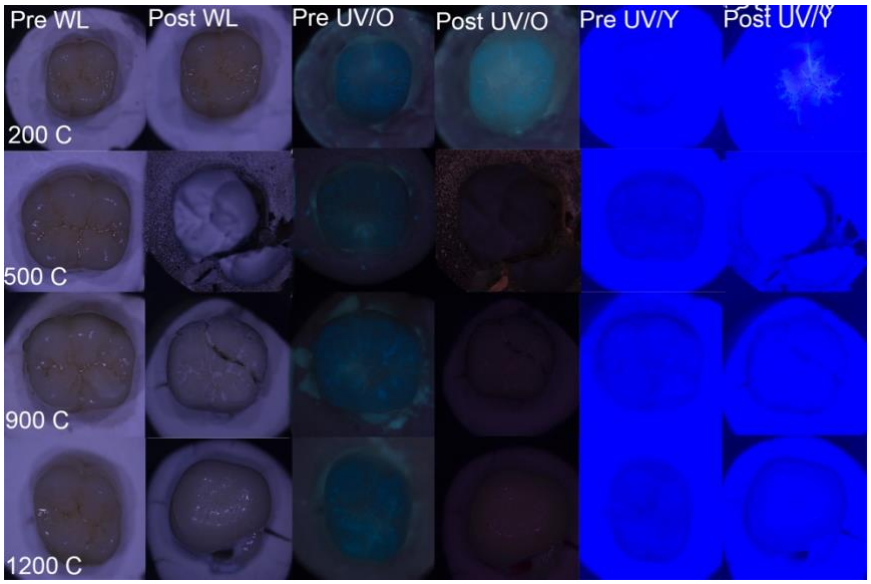


Figure 19. Image of teeth samples comparing the baseline images (Pre-heat treatment) to post-heat treatment at 200 °C, 500 °C, 900 °C and 1200 °C, when excited with white light (W), UV-A light with orange filter (UV/O) and UV-A light with yellow filter for VitaEnamic®.

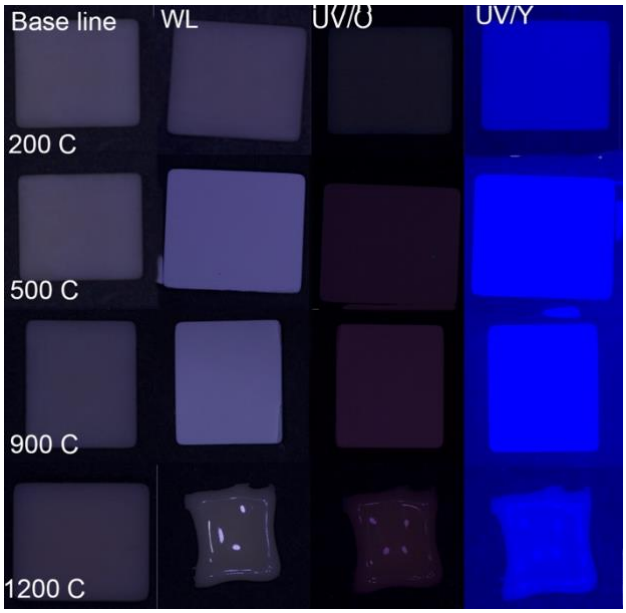


Figure 20. Image of disc samples comparing the baseline images (Pre-heat treatment) to post-heat treatment at 200 °C, 500 °C, 900 °C and 1200 °C, when excited with white light (W), UV-A light with orange filter (UV/O) and UV-A light with yellow filter for VitaEnamic®.

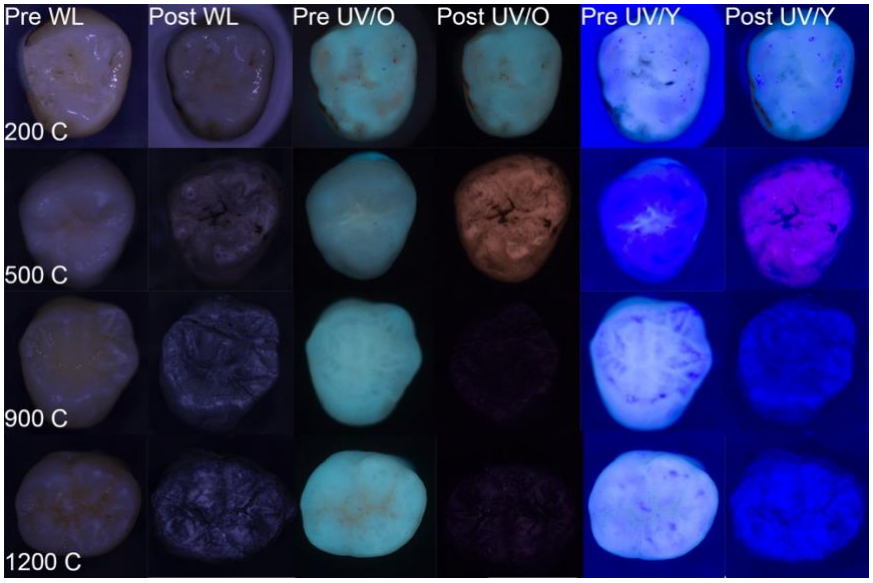


Figure 21. Image of unrestored teeth samples comparing the baseline images (Pre- heat treatment) to post-heat treatment at 200 °C, 500 °C, 900 °C and 1200

°C, when excited with white light (W), UV-A light with orange filter (UV/O) and UV-A light with yellow filter.

Data plots for all the materials subjected to heat test.

1. Ormocer: Admira Fusion®

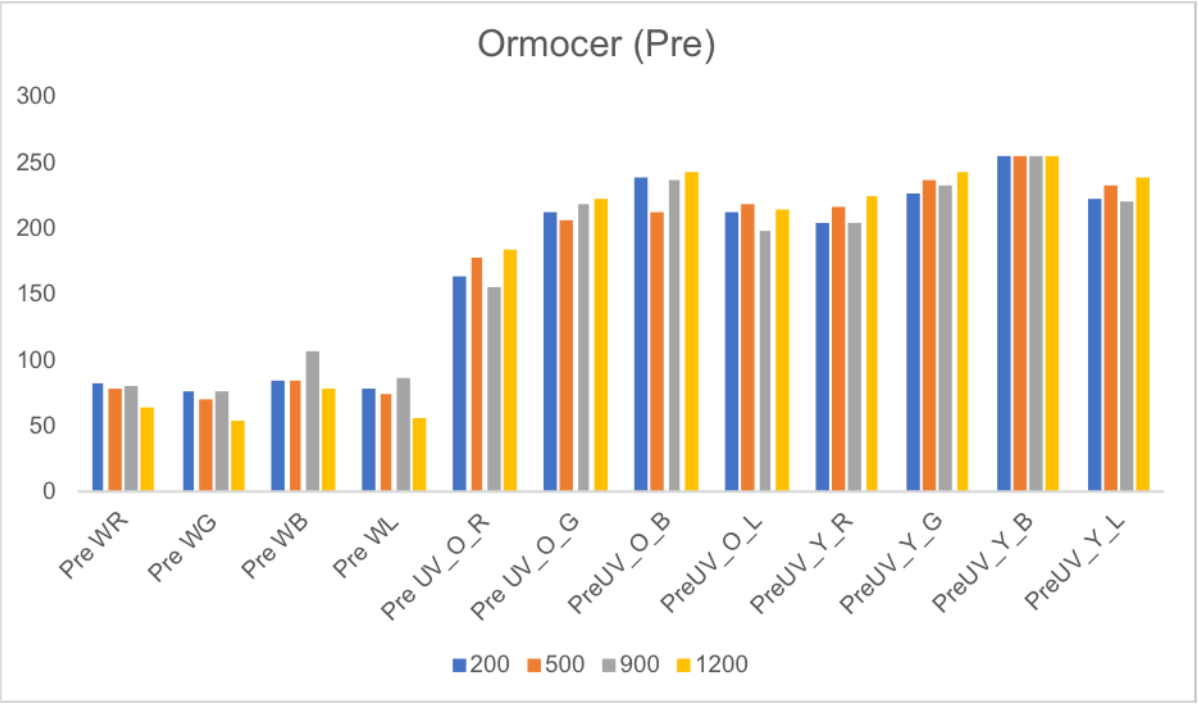


Figure 1.1. Comparing the baseline readings of all restored teeth samples for Admira Fusion (Ormocer) before subjecting to the heat tests.

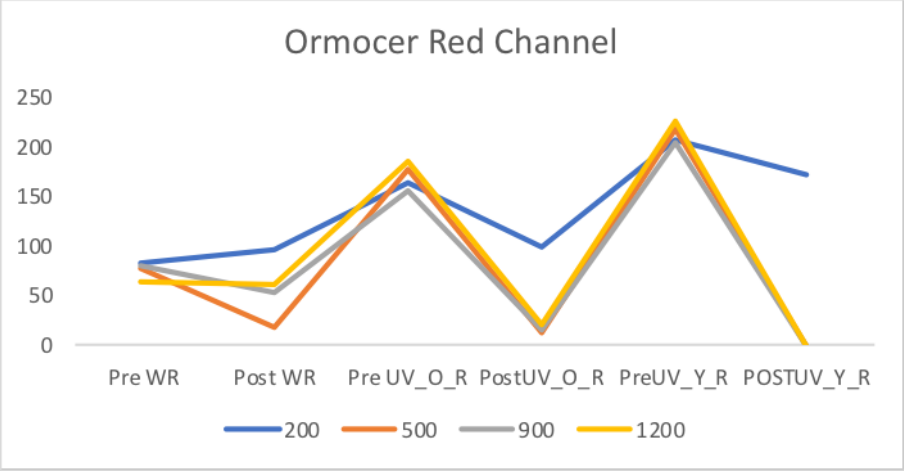


Figure 1.2. Comparing the distribution of red colour channel values of Ormocer (Admira Fusion®) material baseline readings (Pre-) to post-heat treatments readings at 200 ◦ C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

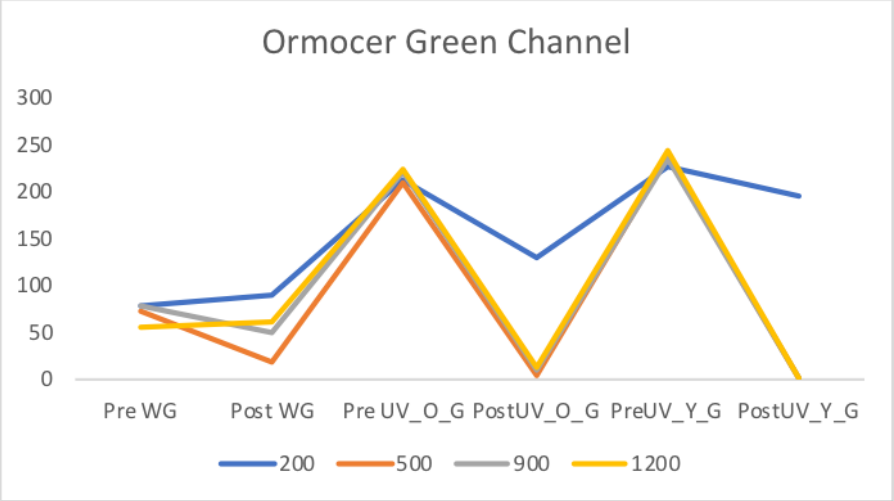


Figure 1.3. Comparing the distribution of green colour channel values of Ormocer (Admira Fusion®) material baseline readings (Pre) to post heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

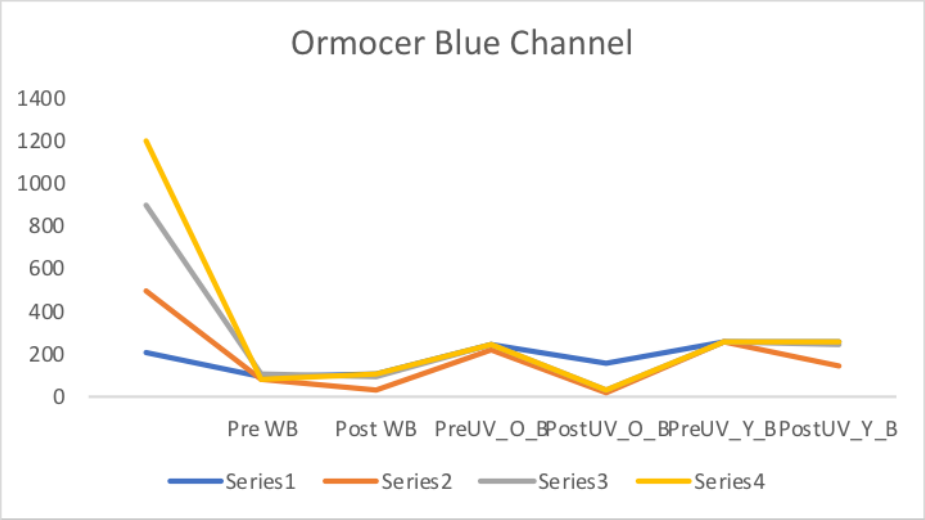


Figure 1.4. Comparing the distribution of blue colour channel values of Ormocer (Admira Fusion®) material baseline readings (Pre-) to post-heat treatments readings at 200 ◦ C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

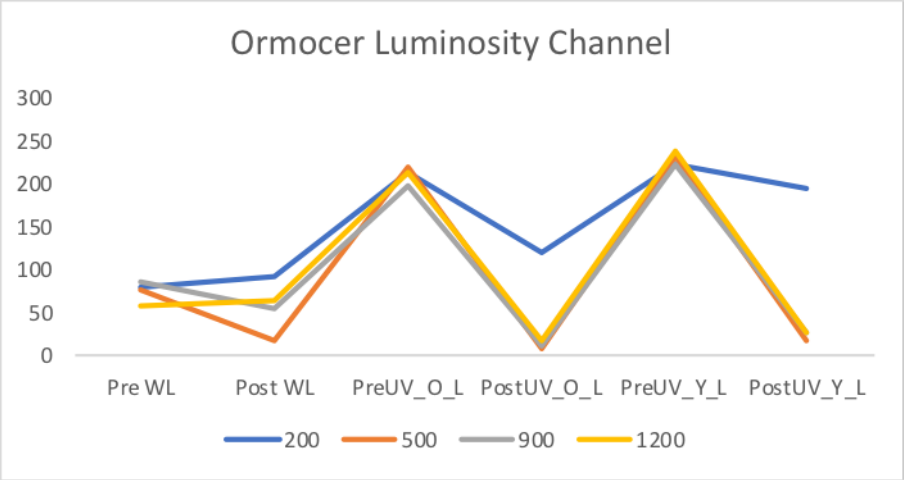


Figure 1.5. Comparing the distribution of luminosity colour channel values of Ormocer (Admira Fusion®) material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

2.Herculite™ Ultra (resin-based composite)

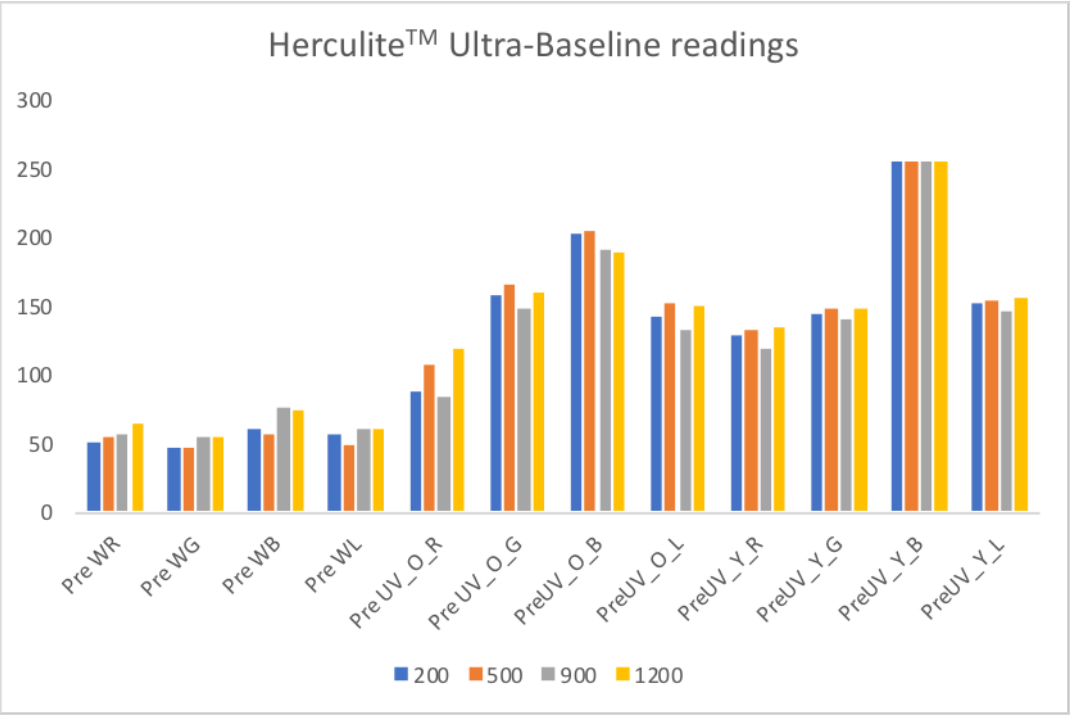


Figure 2.1. Comparing the baseline readings of all restored teeth samples for Herculite™ Ultra (resin-based composite) before subjecting to the heat tests

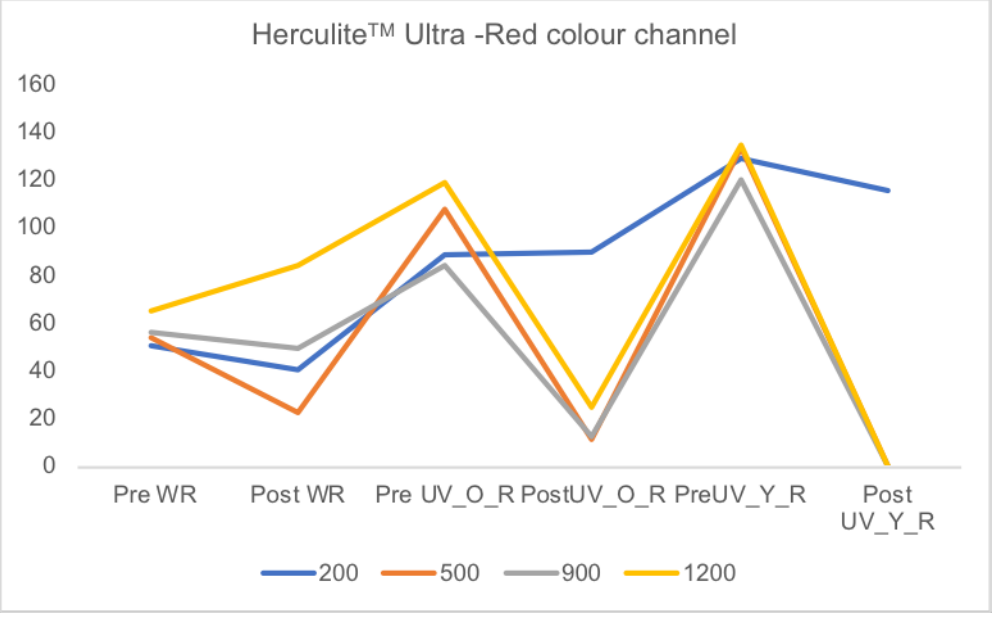


Figure 2.2. Comparing the distribution of red colour channel values of Herculite™ Ultra resin-based composite material baseline readings (Pre-) to post-heat

treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405nm) light with orange filter and yellow filter.

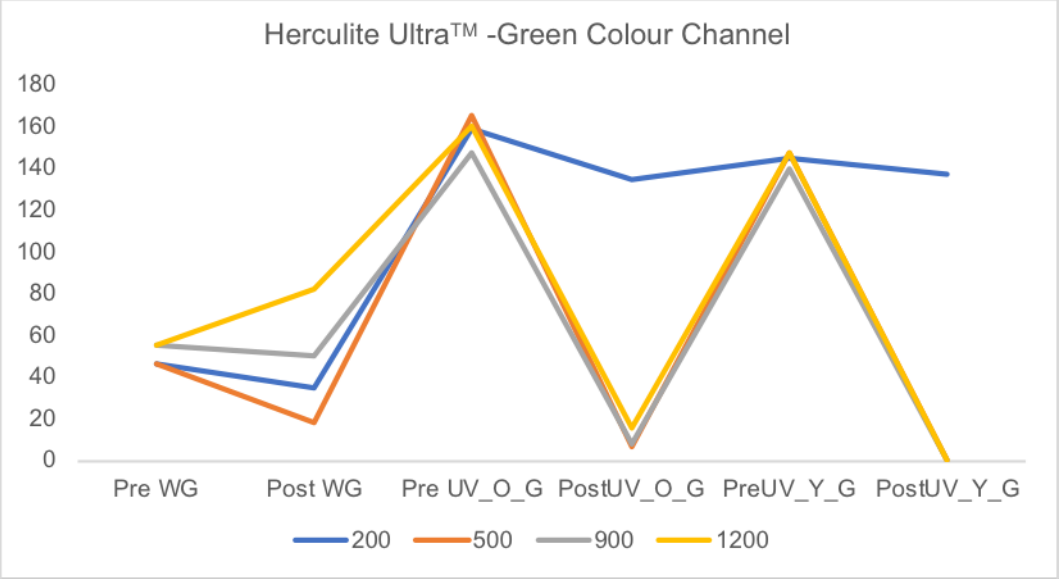


Figure 2.3. Comparing the distribution of green colour channel values of Herculite™ Ultra resin-based composite material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405nm) light with orange filter and yellow filter.

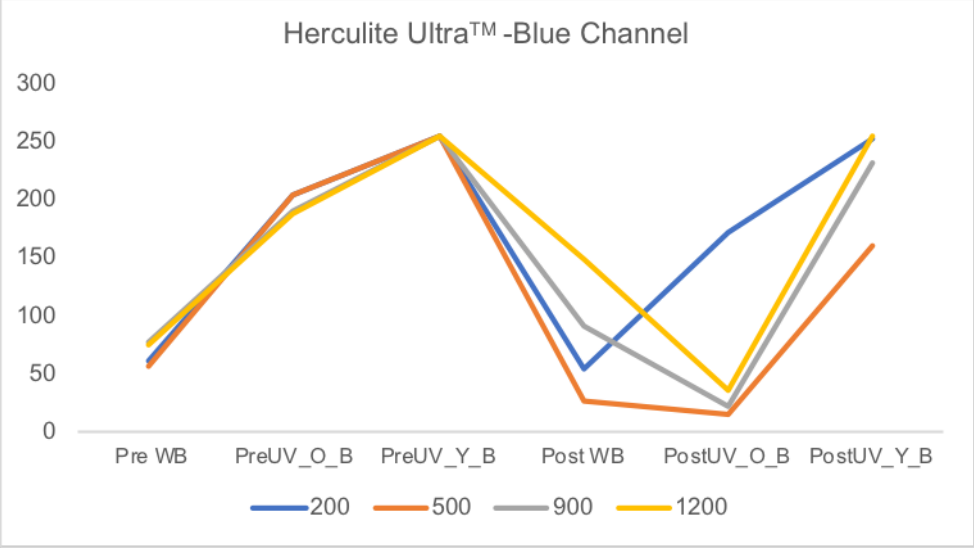


Figure 2.4. Comparing the distribution of blue colour channel values of Herculite™ Ultra resin-based composite material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405nm) light with orange filter and yellow filter.

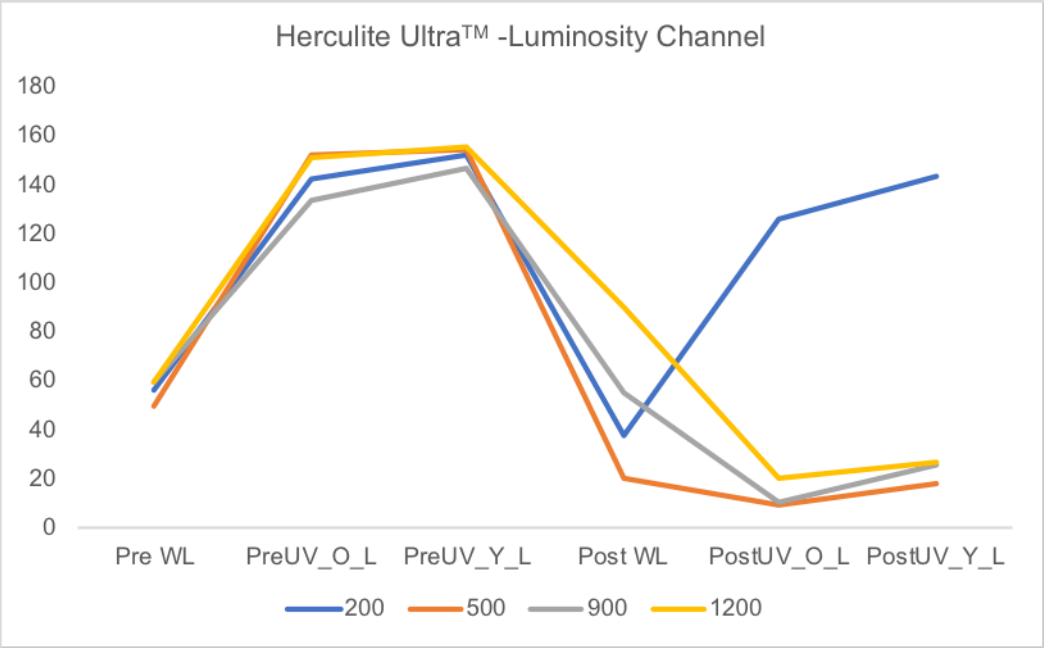


Figure 2.5. Comparing the distribution of blue colour channel values of Herculite™ Ultra resin-based composite material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405nm) light with orange filter and yellow filter.

3. Gradia Direct X: Composite resin

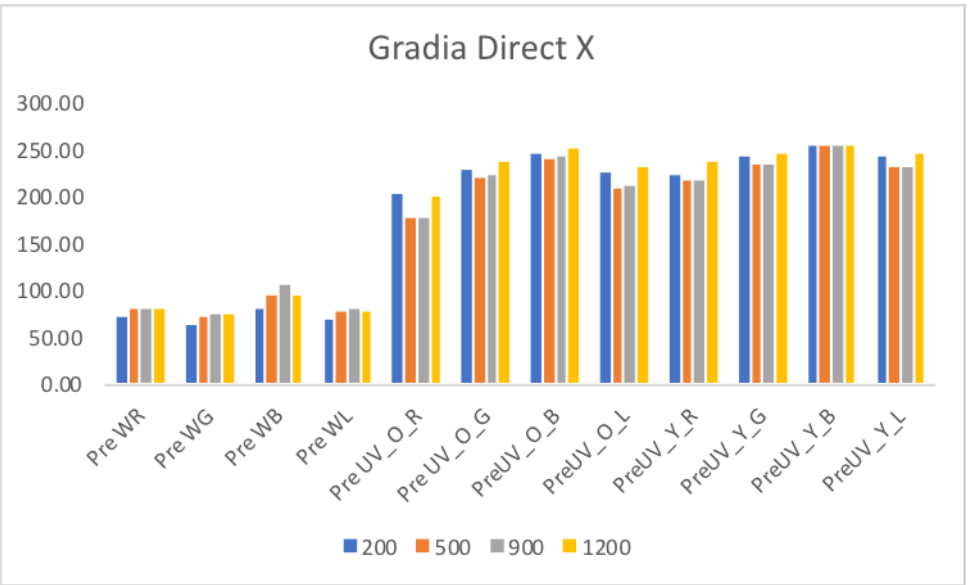


Figure 3.1. Comparing the baseline readings of all restored teeth samples for Gradia (resin-based composite) before subjecting to the heat tests

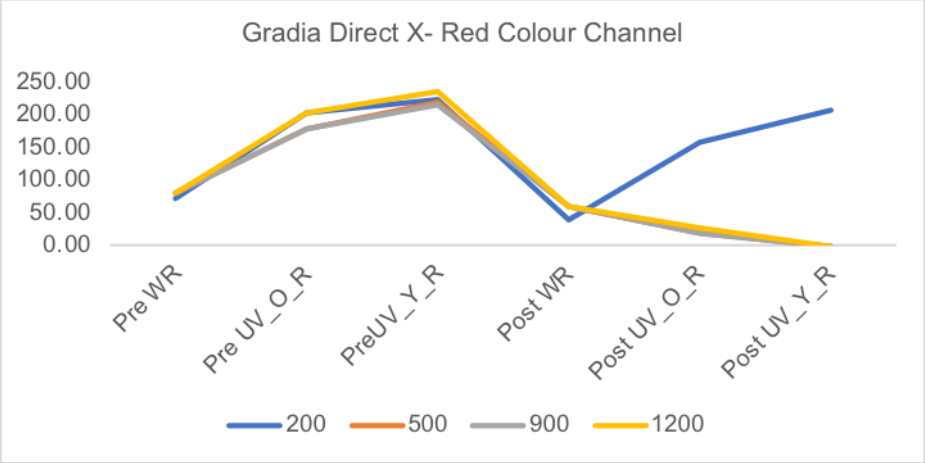


Figure 3.2. Comparing the distribution of red colour channel values of Gradia resin-based composite material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405nm) light with orange filter and yellow filter.

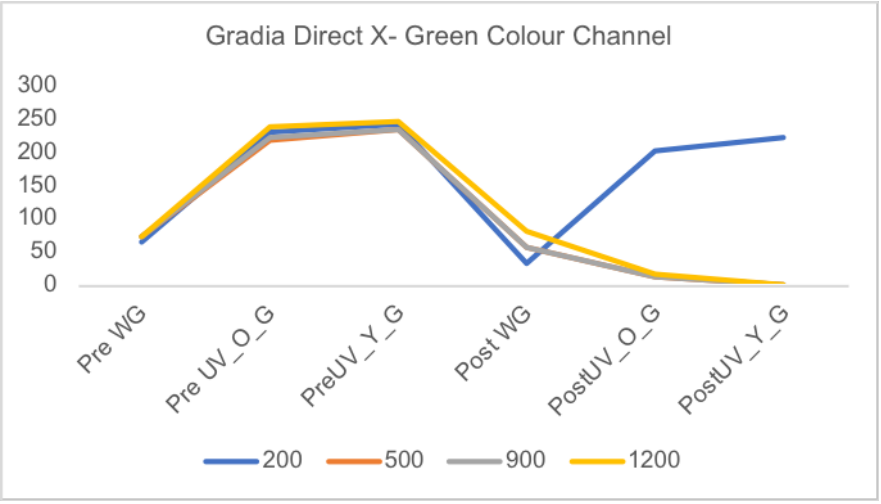


Figure 3.3. Comparing the distribution of green colour channel values of Gradia resin- based composite material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

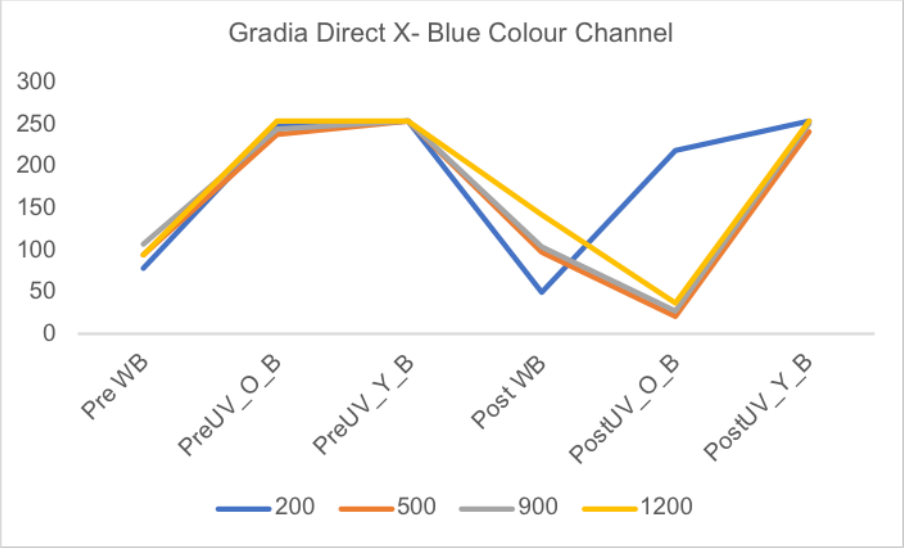


Figure 3.4. Comparing the distribution of blue colour channel values of Gradia resin-based composite material baseline readings (Pre-) to post-heat treatments readings at 200 ◦ C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

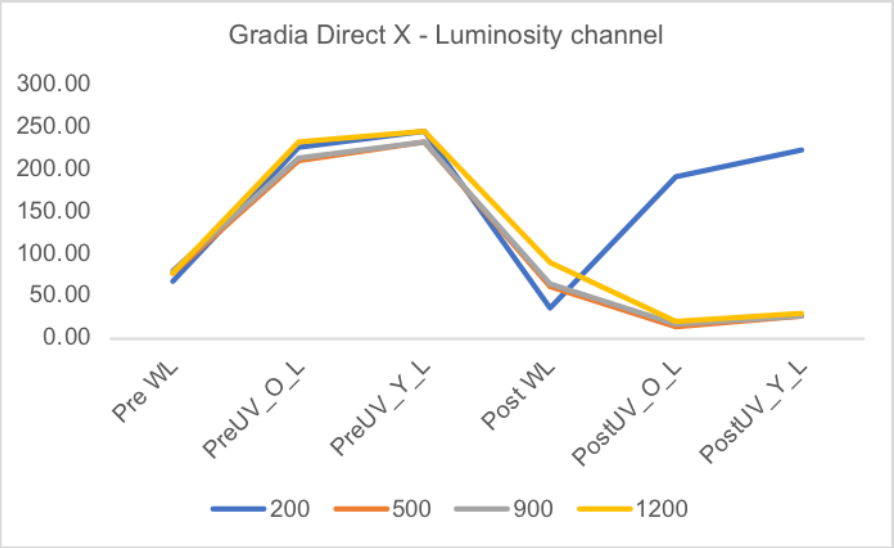


Figure 3.5. Comparing the distribution of luminosity channel values of Gradia resin-based composite material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

4. TPH Spectra Dentsply

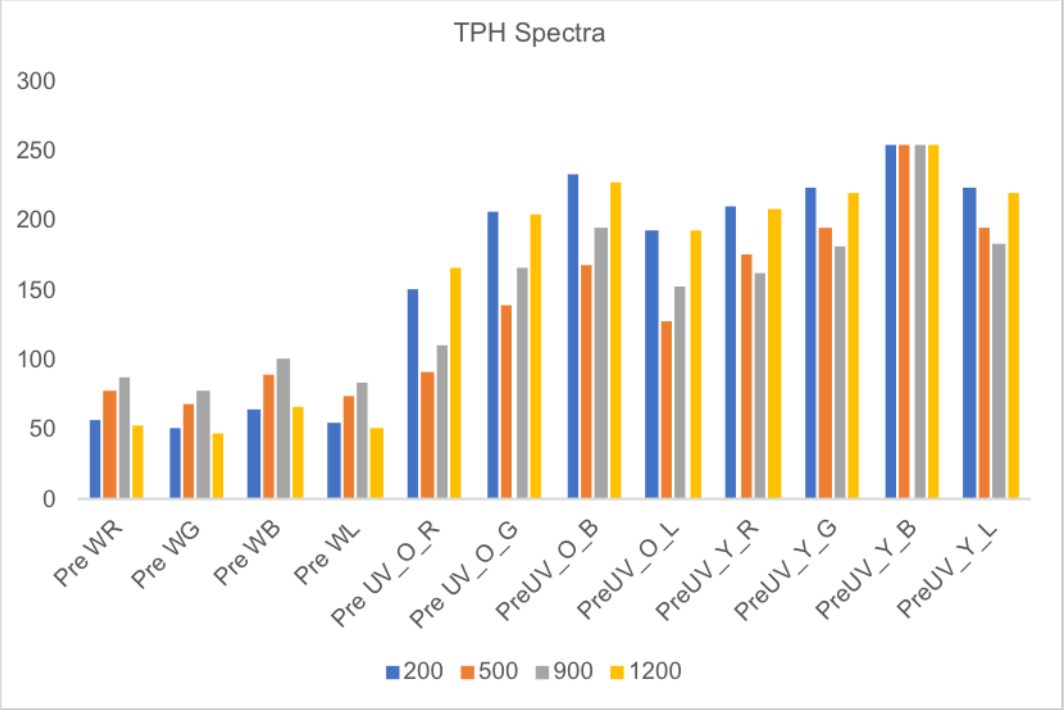


Figure 4.1. Comparing the baseline readings of all restored teeth samples for TPH Spectra (resin-based composite) before subjecting to the heat tests.

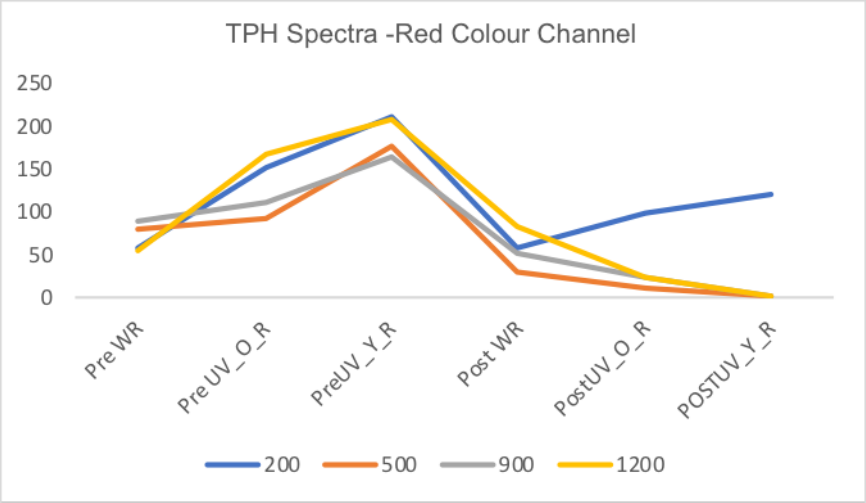


Figure 4.2. Comparing the distribution of red colour channel values of TPH Spectra resin- based composite material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

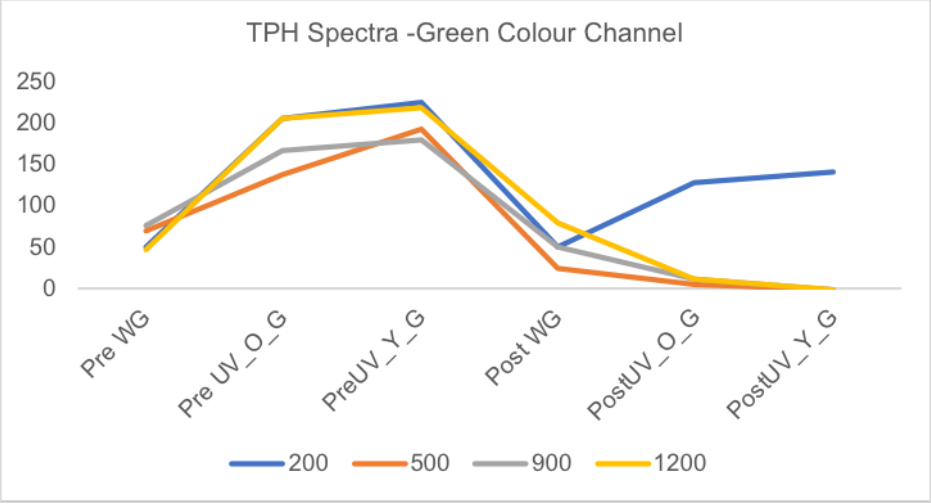


Figure 4.3. Comparing the distribution of green colour channel values of TPH Spectra resin-based composite material baseline readings (Pre-) to post-heat

treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

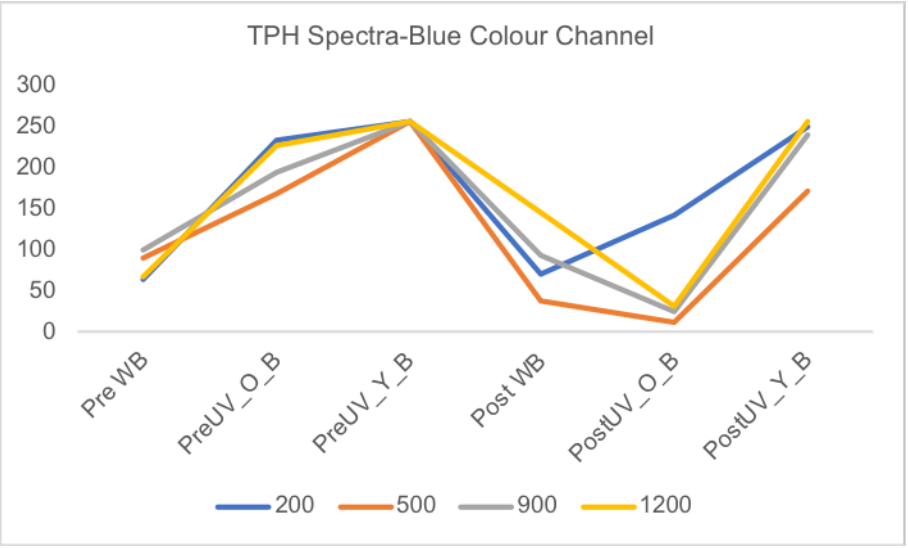


Figure 4.4. Comparing the distribution of blue colour channel values of TPH Spectra resin- based composite material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

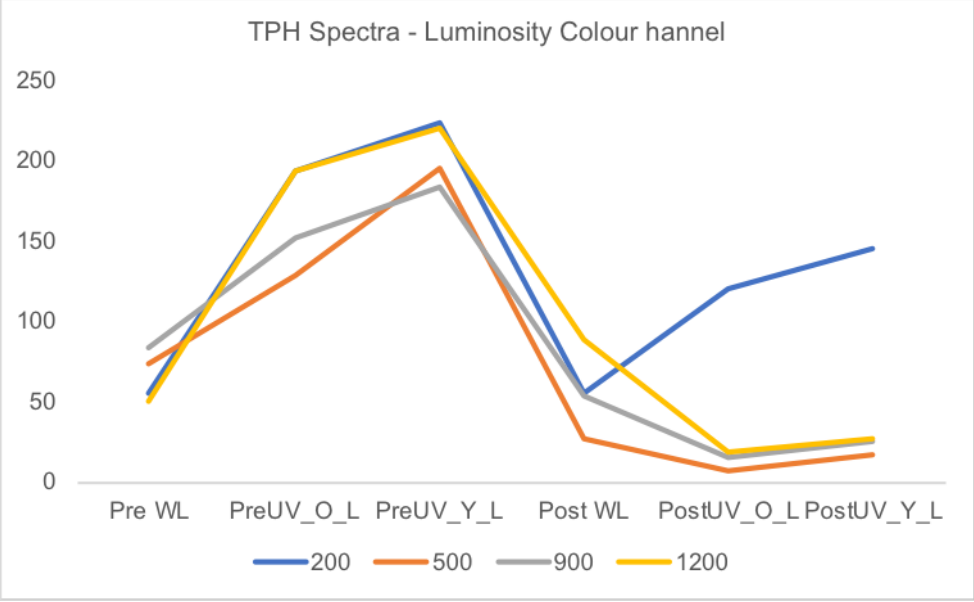


Figure 4.5. Comparing the distribution of luminosity channel values of TPH Spectra resin- based composite material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

5. 3M Filtek Supreme XTE: Resin-based composite

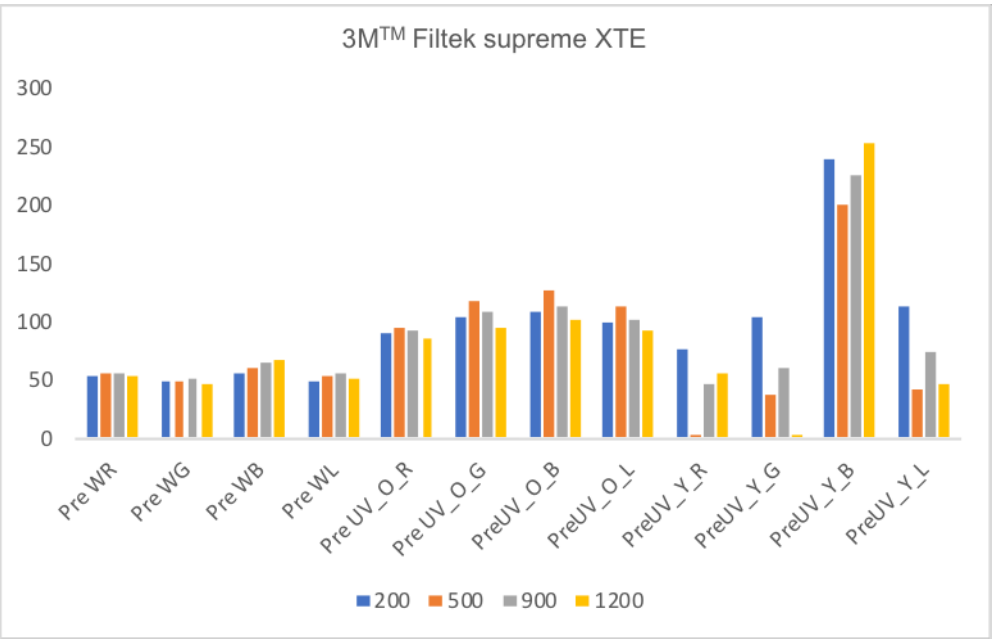


Figure 5.1. Comparing the baseline readings of all restored teeth samples for 3M Filtek Supreme XTE (Resin-based composite) before subjecting to the heat tests.

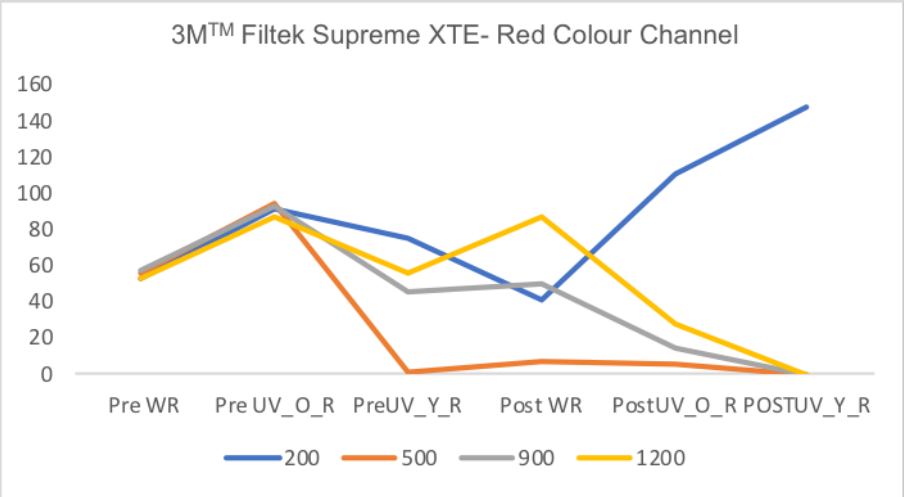


Figure 5.2. Comparing the distribution of red colour channel values of 3M Filtek Supreme XTE resin-based composite material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

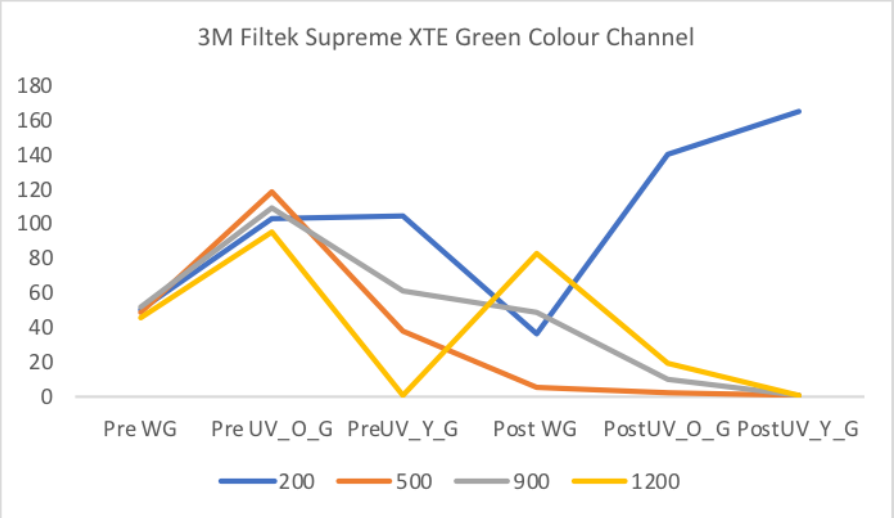


Figure 5.3. Comparing the distribution of green colour channel values of 3M Filtek Supreme XTE resin-based composite material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited

with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

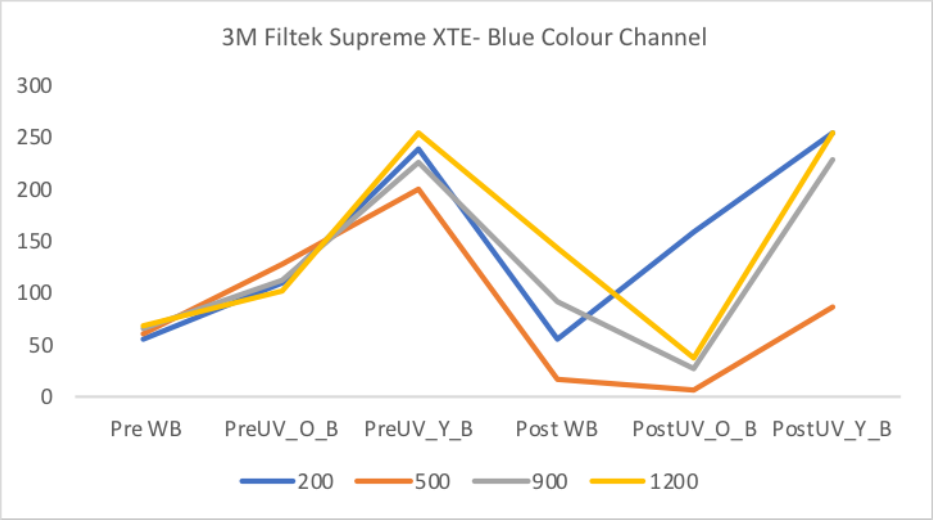


Figure 5.4. Comparing the distribution of blue colour channel values of 3M Filtek Supreme XTE resin-based composite material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

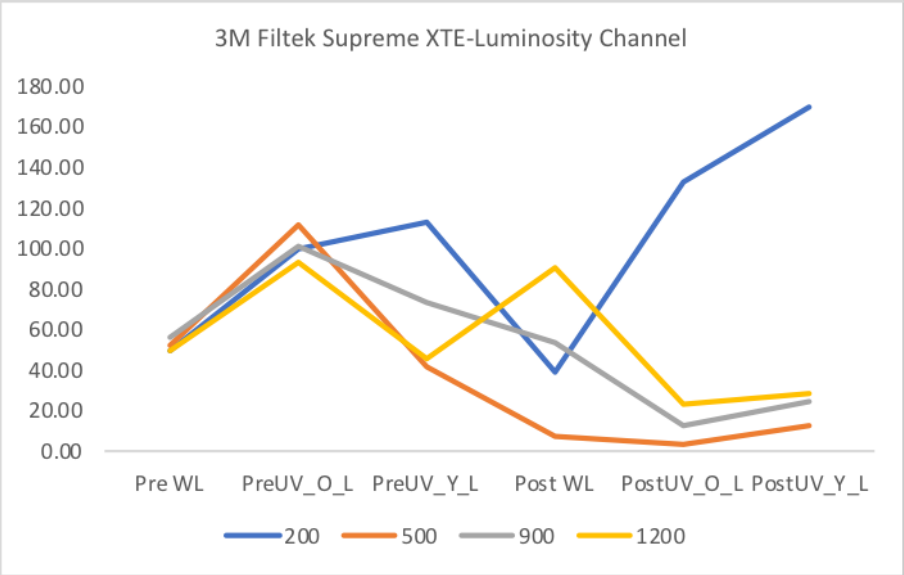


Figure 5.5. Comparing the distribution of luminosity channel values of 3M Filtek™ Supreme XTE resin-based composite material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

6. Fuji II: Glass Ionomer Cement

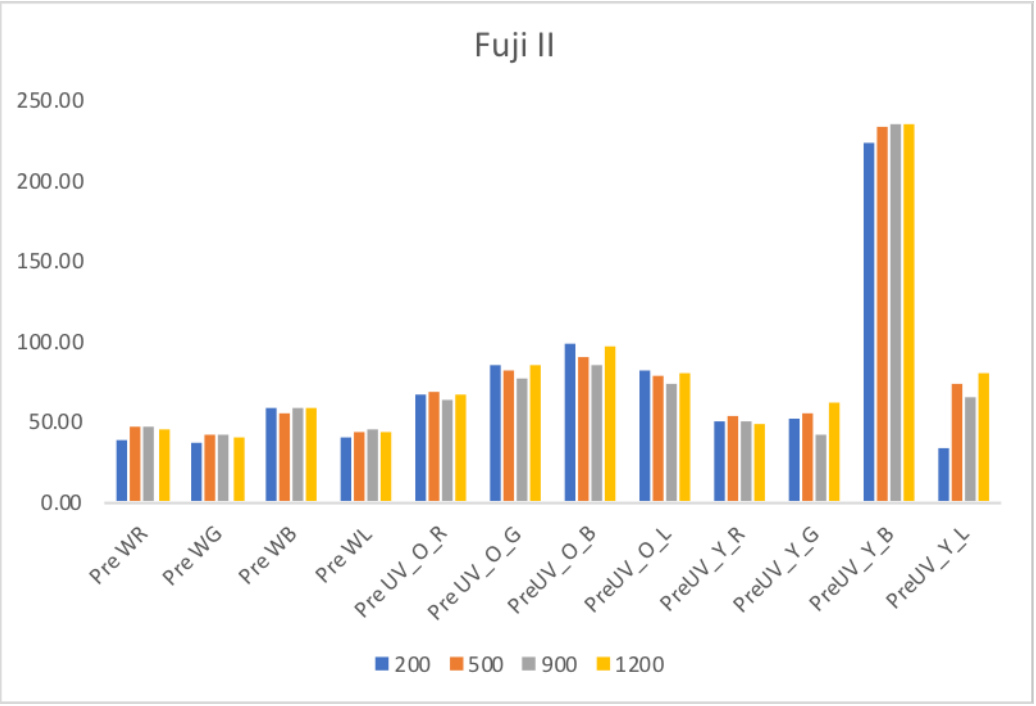


Figure 6.1. Comparing the baseline readings of all restored teeth samples for Fuji II (Glass-ionomer Cement) before subjecting to the heat tests.

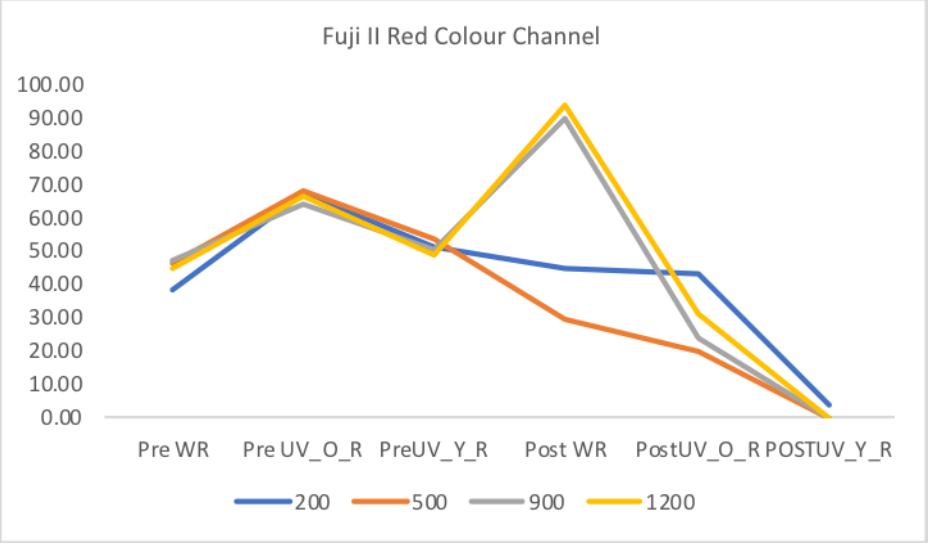


Figure 6.2. Comparing the distribution of red colour channel values of Fuji II (Glass-ionomer Cement) material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

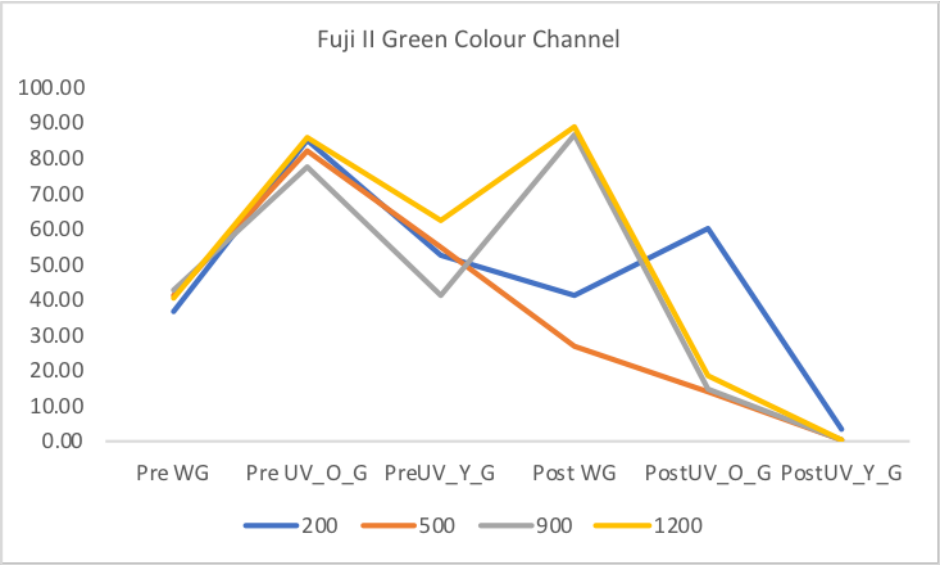


Figure 6.3. Comparing the distribution of green colour channel values of Fuji II (Glass-ionomer Cement) material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

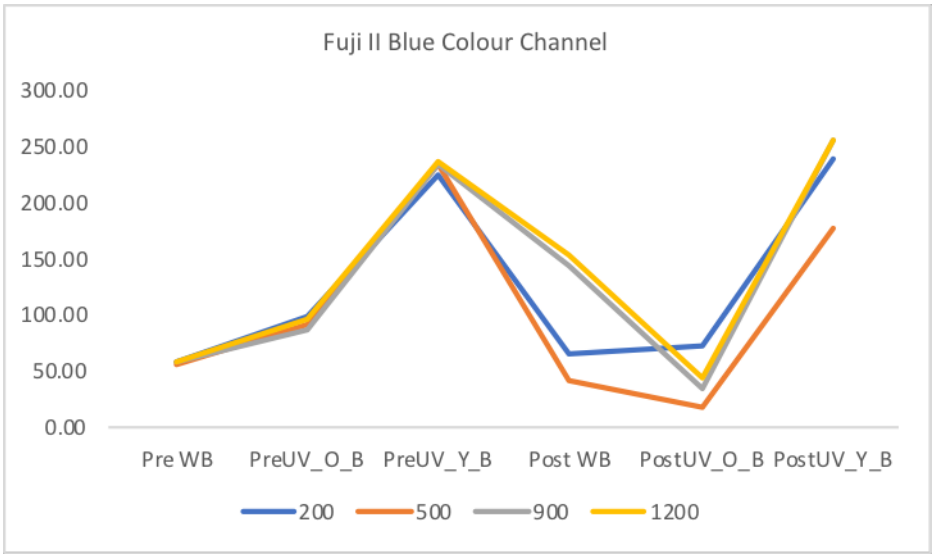


Figure 6.4. Comparing the distribution of blue colour channel values of Fuji II (Glass-ionomer Cement) material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

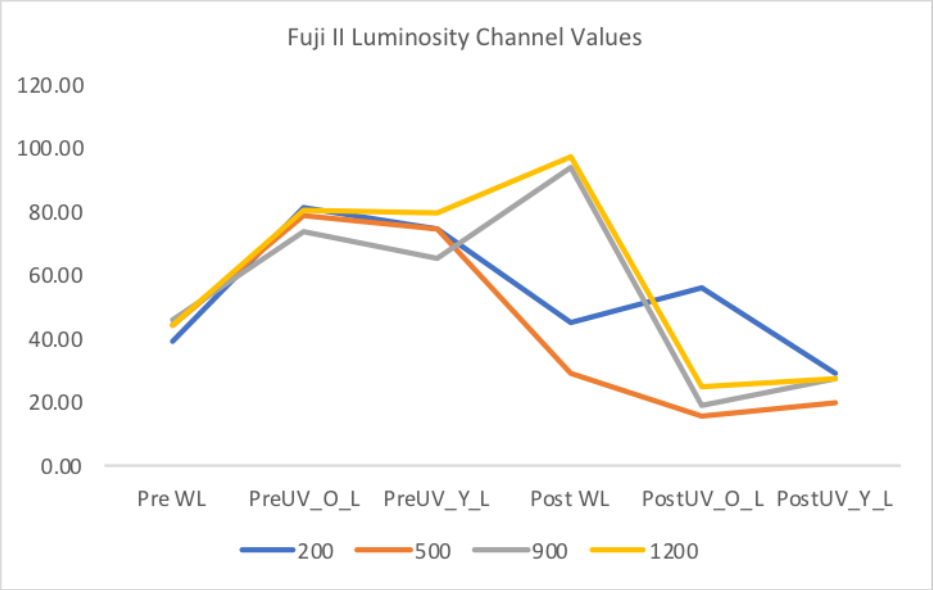


Figure 6.5. Comparing the distribution of luminosity values of Fuji II (Glass-ionomer Cement) material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

7. Fuji VIII: Glass-Ionomer cement

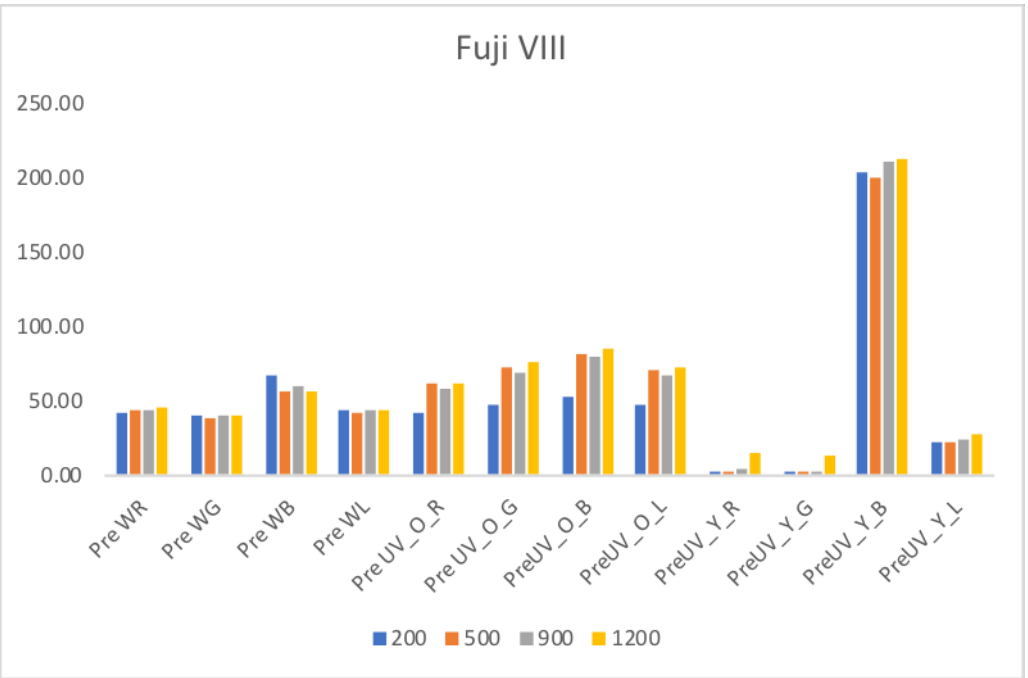


Figure 7.1. Comparing the baseline readings of all restored teeth samples for Fuji VIII (Glass-ionomer Cement) before subjecting to the heat tests.

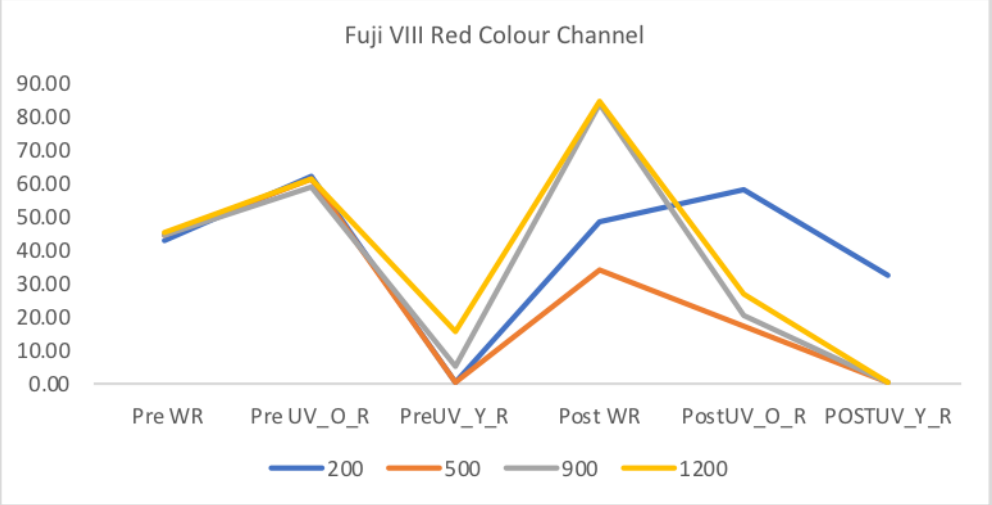


Figure 7.2. Comparing the distribution of red colour channel values of Fuji VIII (Glass-ionomer Cement) material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter

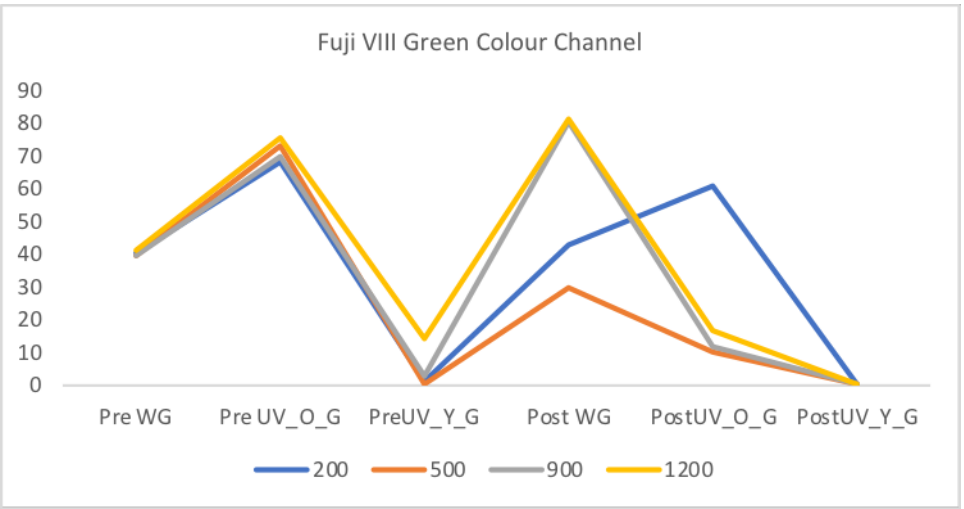


Figure 7.3. Comparing the distribution of green colour channel values of Fuji VIII (Glass-ionomer Cement) material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

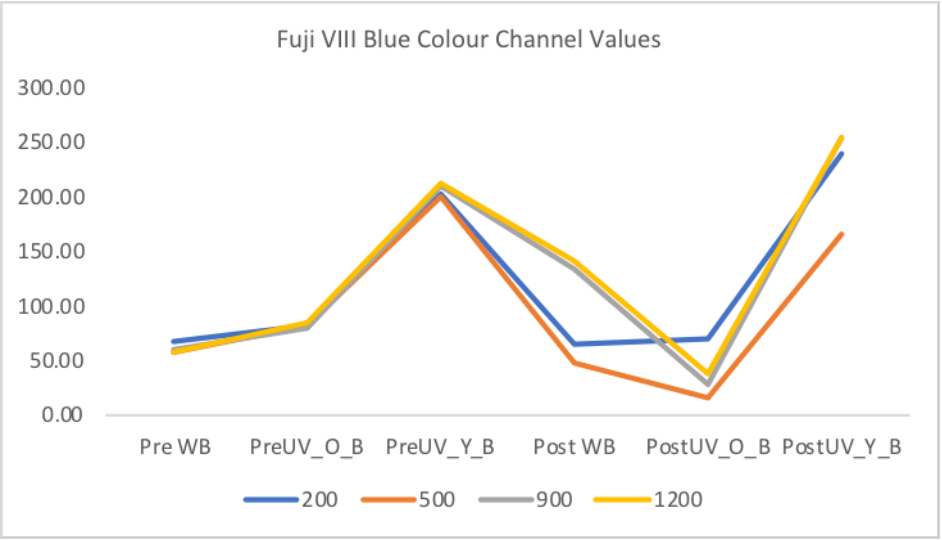


Figure 7.4. Comparing the distribution of blue colour channel values of Fuji VIII (Glass-ionomer Cement) material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter

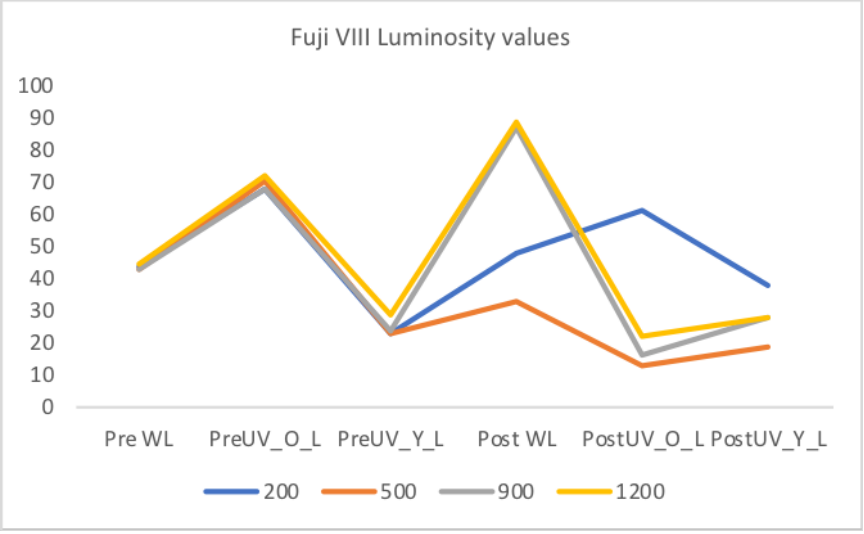


Figure 7.5. Comparing the distribution of luminosity values of Fuji VIII (Glass-ionomer Cement) material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

8. Fuji IX: Glass-Ionomer cement

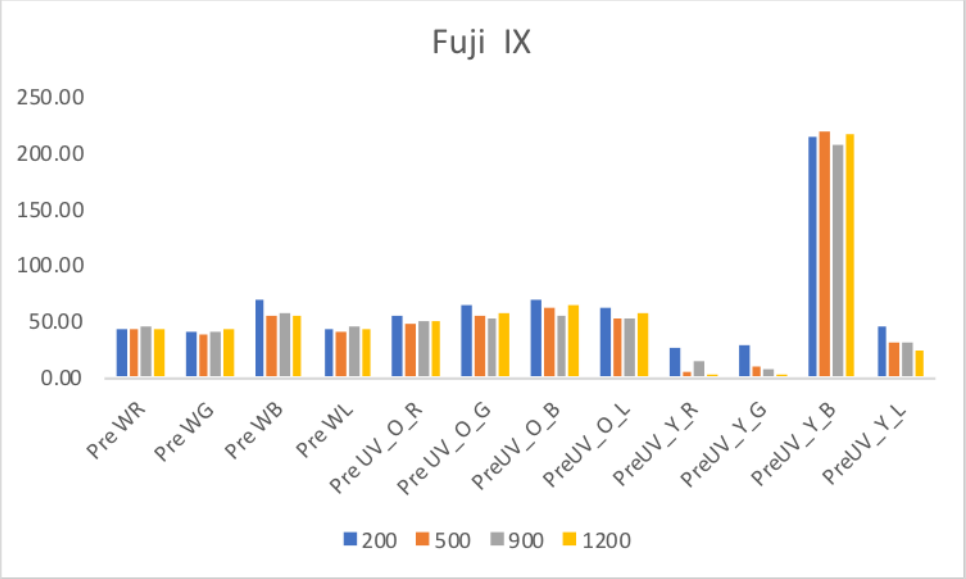


Figure 8.1. Comparing the baseline readings of all restored teeth samples for Fuji IX (Glass-ionomer Cement) before subjecting to the heat tests.

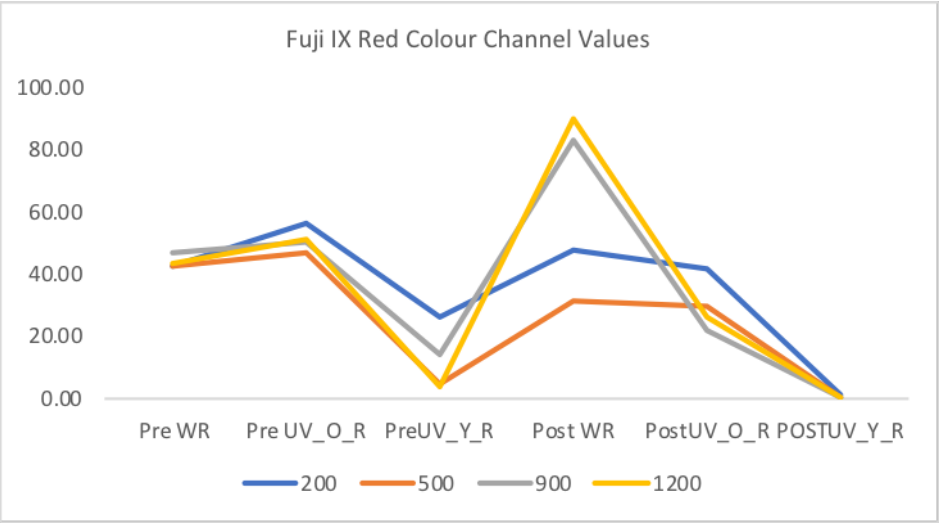


Figure 8.2. Comparing the distribution of red colour channel values of Fuji IX (Glass-ionomer Cement) material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

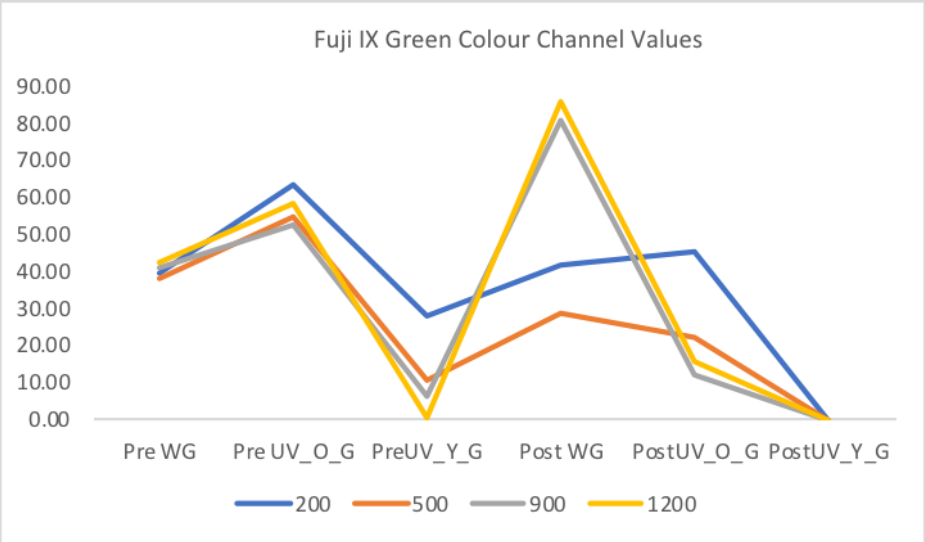


Figure 8.3. Comparing the distribution of green colour channel values of Fuji IX (Glass-ionomer Cement) material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

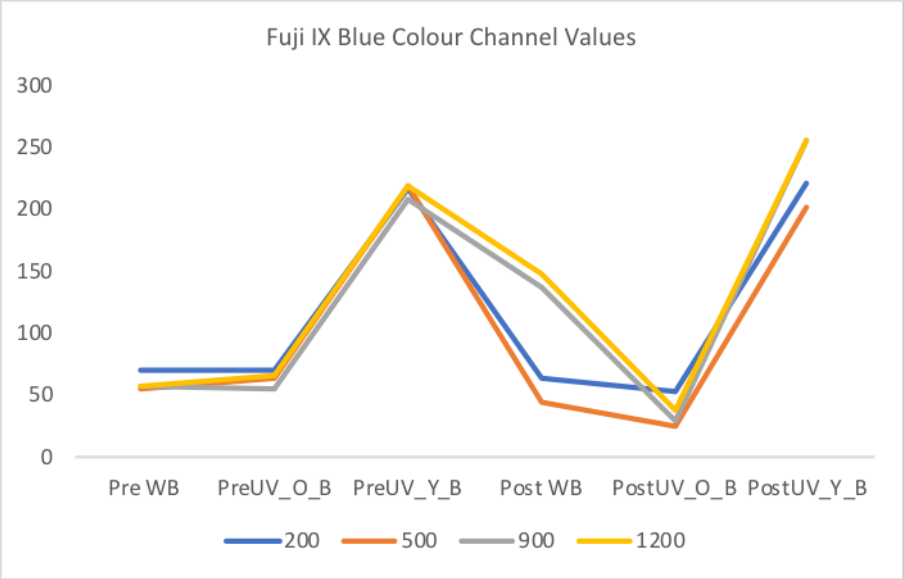


Figure 8.4. Comparing the distribution of blue colour channel values of Fuji IX (Glass-ionomer Cement) material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

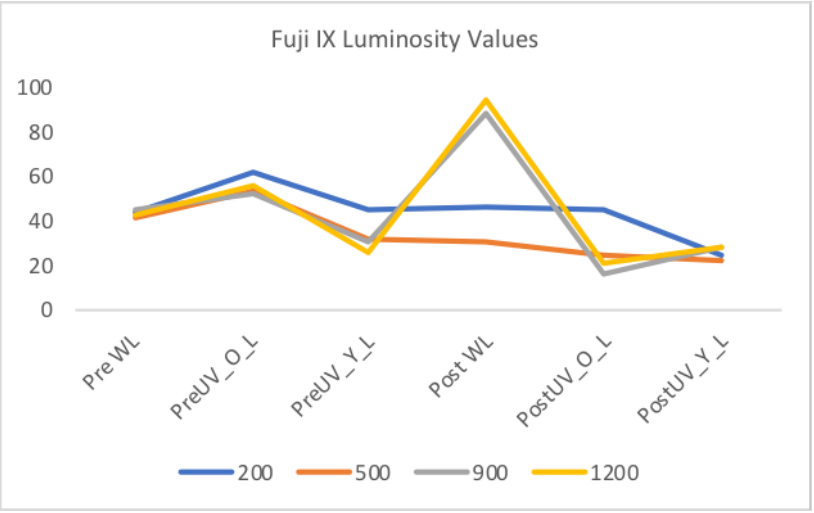


Figure 8.5. Comparing the distribution of luminosity values of Fuji IX (Glass-ionomer Cement) material baseline readings (Pre-) to post-heat treatments readings at 200 ◦ C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

9. Vita Enamic: (Hybrid ceramic)

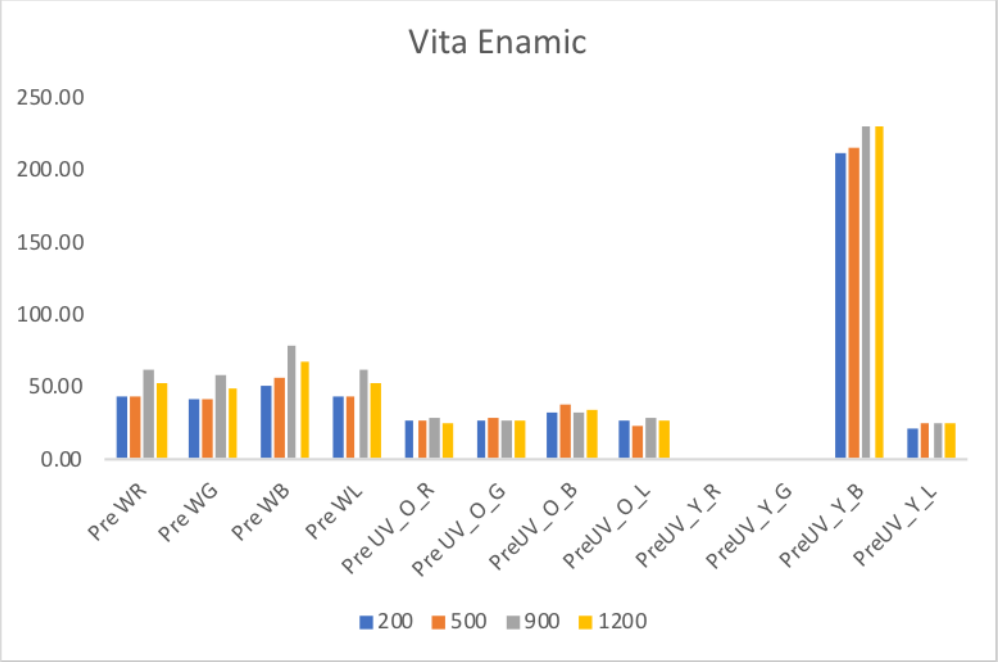


Figure 9.1. Comparing the baseline readings of all restored teeth samples for Vita Enamic before subjecting to the heat tests.

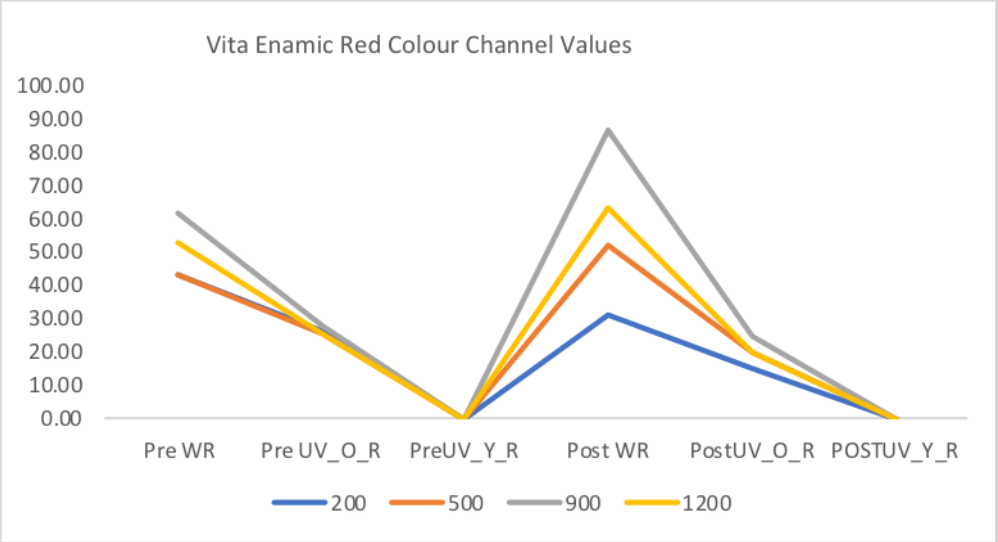


Figure 9.2. Comparing the distribution of red colour channel values of Vita Enamic® material baseline readings (Pre-) to post-heat treatments readings at

200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

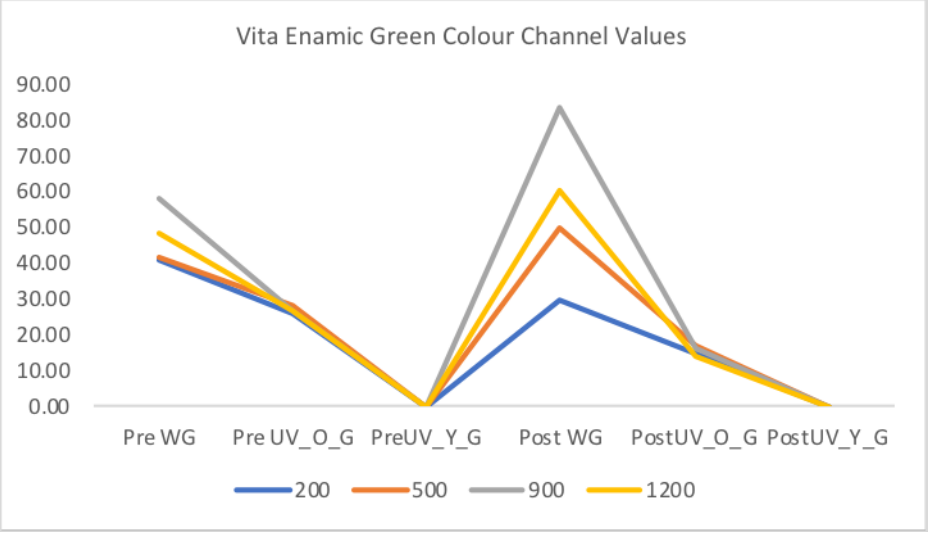


Figure 9.3. Comparing the distribution of green colour channel values of Vita Enamic® material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

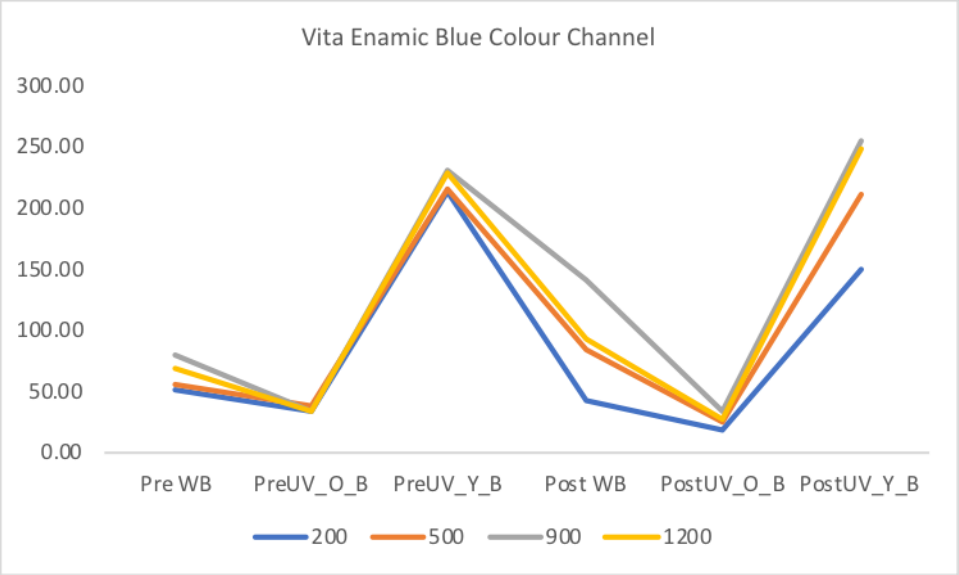


Figure 9.4. Comparing the distribution of blue colour channel values of Vita Enamic® material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

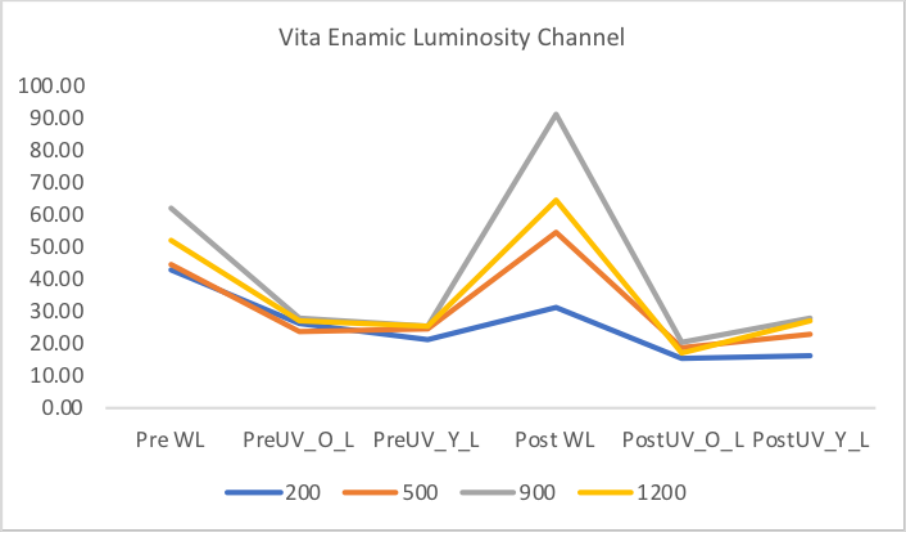


Figure 9.5. Comparing the distribution of luminosity channel values of Vita Enamic® material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

10. Vita Bloc: Feldspar ceramic

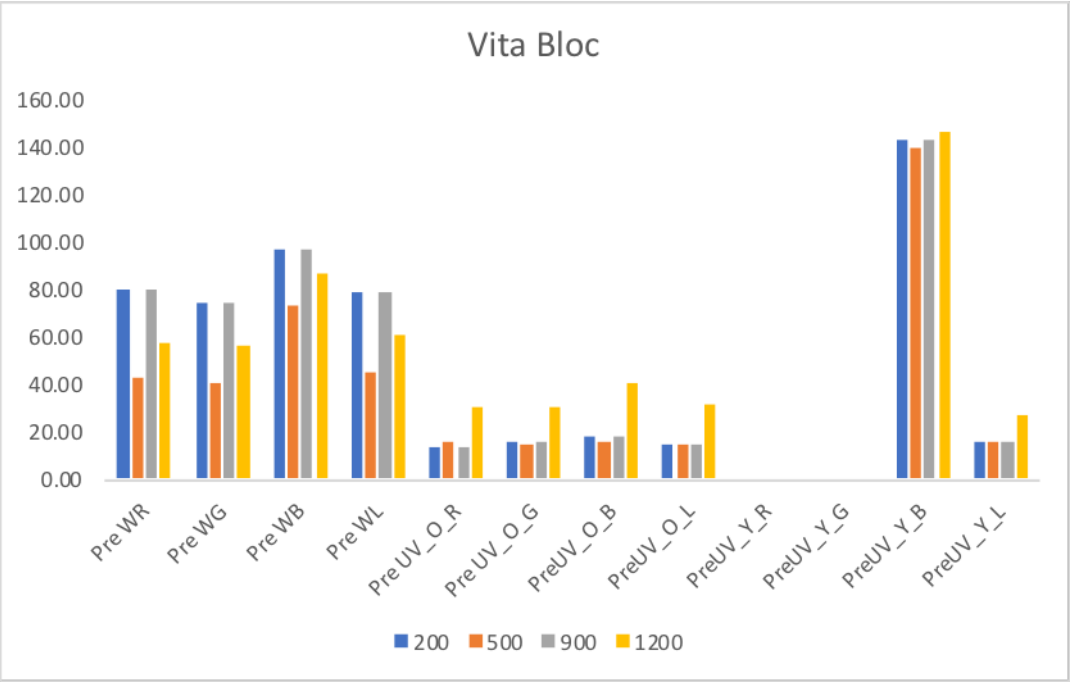


Figure 10.1. Comparing the baseline readings of all restored teeth samples for Vita Bloc before subjecting to the heat tests.

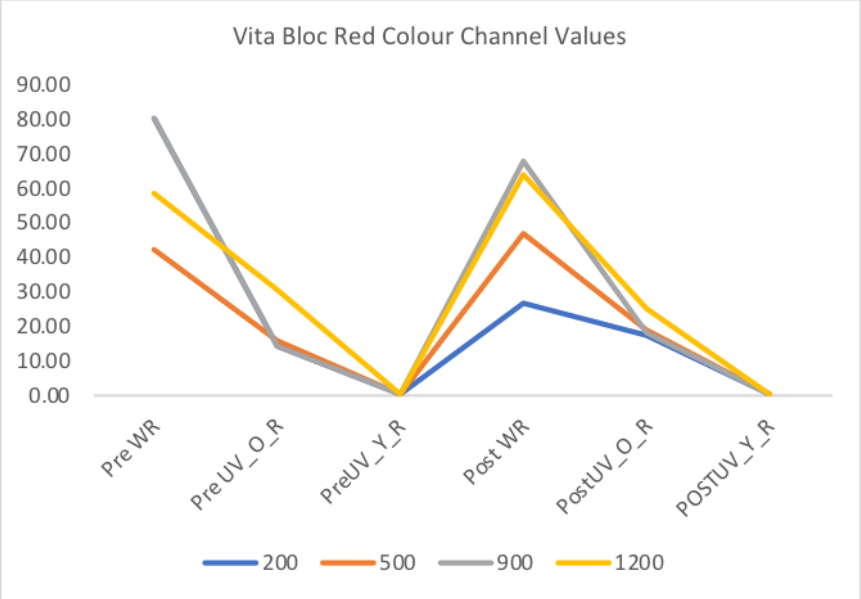


Figure 10.2. Comparing the distribution of red colour channel values of Vita Bloc material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

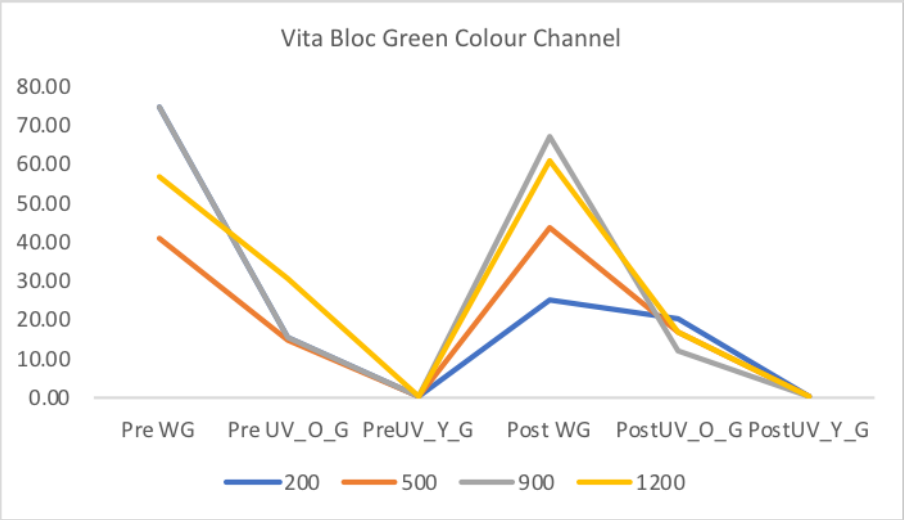


Figure 10.3. Comparing the distribution of green colour channel values of Vita Bloc material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

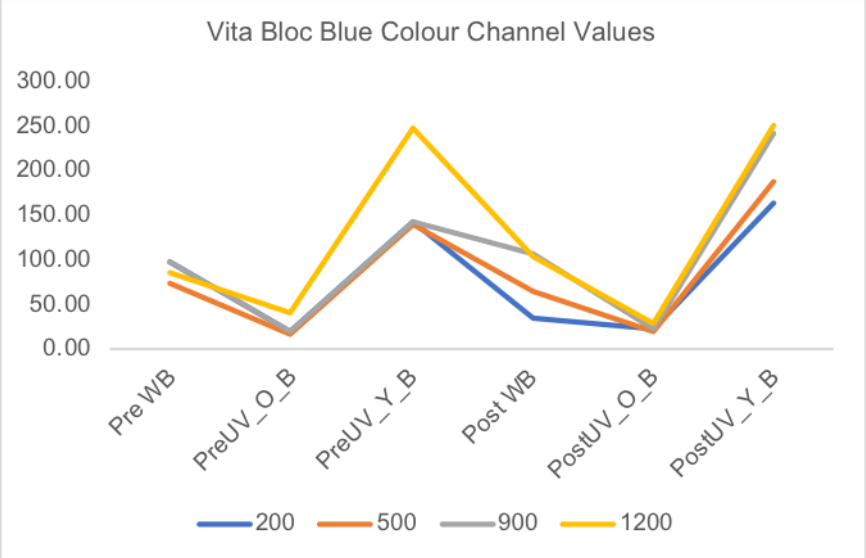


Figure 10.4. Comparing the distribution of blue colour channel values of Vita Bloc material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

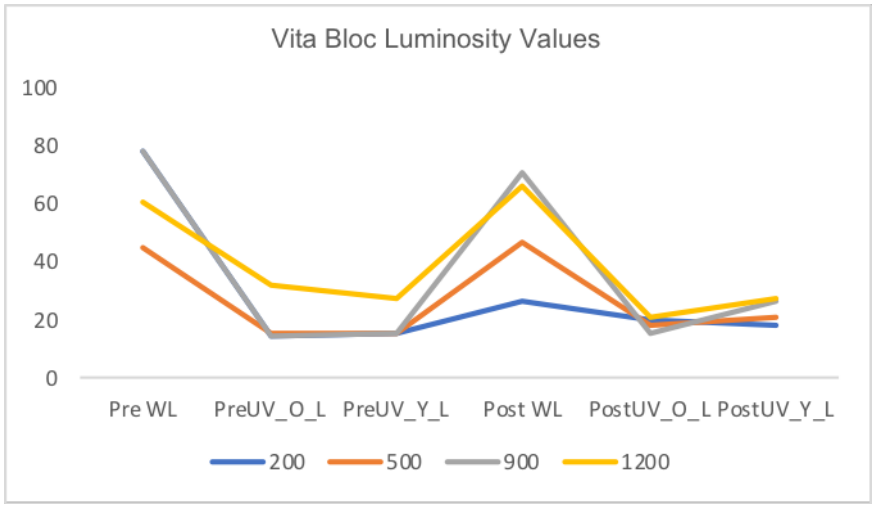


Figure 10.5. Comparing the distribution of luminosity values of Vita Bloc material baseline readings (Pre-) to post-heat treatments readings at 200 ◦C, 500 ◦C, 900 ◦C, 1200 ◦C, when excited with white light (W) without filter, UV-A (405 nm) light with orange filter and yellow filter.

Appendix C. Declaration of Co-authorship and contribution towards peer reviewed publication- Chapter 3

Declaration of Co-authorship and Contribution (Thesis)

Research Division

DECLARATION OF CO-AUTHORSHIP AND CONTRIBUTION

Full bibliographic reference to the item/publication, including authors, title, journal (vol/pages), year.	Kiran R, Chapman J, Forrest A, Tennant M, Walsh LJ. Forensic applications: Fluorescence properties of tooth-coloured restorative materials using a fluorescence DSLR camera. Forensic Sci Int. 2017;273:20-8. https://doi.org/10.1016/j.forsciint.2017.01.022
Status	Published

Nature of Candidate's Contribution, including percentage of total

In conducting the study, I was responsible for laboratory work, and collection of data. This publication was written by me. I formed the research question, collated the literature, analysed the data and interpreted the results (80%). I also won the funding for the study.

Nature of all Co-Authors' Contributions, including percentage of total

My co-authors, Professor Walsh, A/Professor Forrest, W/Professor Tennant and Dr Chapman, critically reviewed the manuscript with questions, comments and criticism. They assisted and provided guidance in editing and preparing the manuscript for submission (20%).

Has this paper been submitted for an award by another research degree candidate (Co-Author), either at CQUniversity or elsewhere? (if yes, give full details)

No

Candidate's Declaration

I declare that the publication above meets the requirements to be included in the thesis as outlined in the Research Higher Degree Theses Policy and Procedure

Date: 15/7/2019

Appendix D: Declaration of Co-authorship and contribution towards peer reviewed publication-Chapter 4

Declaration of Co-authorship and Contribution (Thesis)

Research Division

DECLARATION OF CO-AUTHORSHIP AND CONTRIBUTION

Full bibliographic reference to the item/publication, including authors, title, journal (vol/pages), year.	Kiran R, Chapman J, Forrest A, Tennant M, Walsh LJ. Direct tooth coloured restorative materials: A comparative analysis of the fluorescence properties among different shades. Int J of Esthet Dent
Status	Accepted on 26th November 2019

Nature of Candidate's Contribution, including percentage of total

In conducting the study, I was responsible for laboratory work, and collection of data. This publication was written by me. I formed the research question, collated the literature, analysed the data and interpreted the results (80%).

Nature of all Co-Authors' Contributions, including percentage of total

My co-authors, Professor Walsh, A/Professor Forrest, W/Professor Tennant and Dr Chapman, critically reviewed the manuscript with questions, comments and criticism. They assisted and provided guidance in editing and preparing the manuscript for submission (20%).

Has this paper been submitted for an award by another research degree candidate (Co-Author), either at CQUniversity or elsewhere? (if yes, give full details)

No

Candidate's Declaration

I declare that the publication above meets the requirements to be included in the thesis as outlined in the Research Higher Degree Theses Policy and Procedure

Date: 15/7/2019

Appendix E: Declaration of Co-authorship and contribution towards peer reviewed publication-Chapter 5

Declaration of Co-authorship and Contribution (Thesis)

Research Division

DECLARATION OF CO-AUTHORSHIP AND CONTRIBUTION

Full bibliographic reference to the item/publication, including authors, title, journal (vol/pages), year.	Kiran R, Chapman J, Tennant M, Forrest A, Walsh LJ. Detection of tooth-coloured restorative materials for forensic purposes based on their optical properties: an in vitro comparative study. J Forensic Sci 019;64:254-9. https://doi.org/10.1111/1556-4029.13851
Status	Published

Nature of Candidate's Contribution, including percentage of total

In conducting the study, I was responsible for laboratory work, and collection of data. This publication was written by me. I formed the research question, collated the literature, analysed the data and interpreted the results (85%). I also won the funding for the study.

Nature of all Co-Authors' Contributions, including percentage of total

My co-authors, Professor Walsh, A/Professor Forrest, W/Professor Tennant and Dr Chapman, critically reviewed the manuscript with questions, comments and criticism. They assisted and provided guidance in editing and preparing the manuscript for submission (15%).

Has this paper been submitted for an award by another research degree candidate (Co-Author), either at CQUniversity or elsewhere? (if yes, give full details)

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Candidate's Declaration

I declare that the publication above meets the requirements to be included in the thesis as outlined in the Research Higher Degree Theses Policy and Procedure

Date: 15/7/2019

Appendix F: Declaration of Co-authorship and contribution towards peer reviewed publication- Chapter 6

Declaration of Co-authorship and Contribution (Thesis)

Research Division

DECLARATION OF CO-AUTHORSHIP AND CONTRIBUTION

Full bibliographic reference to the item/publication, including authors, title, journal (vol/pages), year.	Kiran R, Chapman J, Tennant M, Forrest A, Walsh LJ. Fluorescence-aided selective removal of resin-based composite restorative materials: An in vitro comparative study. J Esthet and Restor Dent. 2019 Oct 16. DOI: 10.1111/jerd.12536
Status	Published

Nature of Candidate's Contribution, including percentage of total

In conducting the study, I was responsible for laboratory work, and collection of data. This publication was written by me. I formed the research question, collated the literature, analysed the data and interpreted the results (85%).

Nature of all Co-Authors' Contributions, including percentage of total

My co-authors, Professor Walsh, A/Professor Forrest, W/Professor Tennant and Dr Chapman, critically reviewed the manuscript with questions, comments and criticism. They assisted and provided guidance in editing and preparing the manuscript for submission (15%).

Has this paper been submitted for an award by another research degree candidate (Co-Author), either at CQ University or elsewhere? (if yes, give full details)

No

Candidate's Declaration

I declare that the publication above meets the requirements to be included in the thesis as outlined in the Research Higher Degree Theses Policy and Procedure

Date: 15/7/2019

Appendix G: Declaration of Co-authorship and contribution towards Peer reviewed publication - Chapter 7

Declaration of Co-authorship and Contribution (Thesis)

Research Division

DECLARATION OF CO-AUTHORSHIP AND CONTRIBUTION

Full bibliographic reference to the item/publication, including authors, title, journal (vol/pages), year.	Kiran R, Chapman J, Forrest A, Tennant M, Walsh LJ. Effect of heat on the fluorescence properties of tooth coloured restorative materials and their forensic implications. J Forensic Sci. 2019
Status	Accepted

Nature of Candidate's Contribution, including percentage of total

In conducting the study, I was responsible for laboratory work, and collection of data. This publication was written by me. I formed the research question, collated the literature, analysed the data and interpreted the results (85%).

Nature of all Co-Authors' Contributions, including percentage of total

My co-authors, Professor Walsh, A/Professor Forrest, W/Professor Tennant and Dr Chapman, critically reviewed the manuscript with questions, comments and criticism. They assisted and provided guidance in editing and preparing the manuscript for submission (15%).

Has this paper been submitted for an award by another research degree candidate (Co-Author), either at CQUniversity or elsewhere? (if yes, give full details)

No

Candidate's Declaration

I declare that the publication above meets the requirements to be included in the thesis as outlined in the Research Higher Degree Theses Policy and Procedure

Date: 15/07/2019