

Assessing the Appearance and Fluorescence of Resin-Infiltrated White Spot Lesions With Caries Detection Devices

K Markowitz • K Carey

Clinical Relevance

Fluorescent camera caries detectors can be used to assess the effectiveness of resin infiltration in improving the optic properties of white spot lesions. Clinicians can use a fluorescent camera to demonstrate early lesions to patients.

SUMMARY

Objective: This *in vitro* study examined the effectiveness of caries detector devices in assessing the ability of resin infiltration (RI) (Icon, DMG-Hamburg, Hamburg, Germany) to improve the optical properties of enamel white spot lesions (WSLs).

Methods and Materials: Ten caries-free third molars were used. Photographs, a subjective visual assessment of the photographs, fluorescent camera (FC) images using the Spectra (Air Techniques, Melville, NY, USA), and laser fluorescent (LF) readings using the DIAGNOdent (KaVo, Biberach, Germany) were obtained from each tooth's buccal surface. Specimens were coated with nail polish leaving a rectan-

gular window on the buccal surface and placed in pH 4.5 lactic acid gel for two weeks to create a WSL. The WSLs were analyzed by the same methods. RI was applied to half of each WSL; final photographs were then taken, and caries detector assessments were conducted. FC images were converted to grayscale, and the fluorescent image's brightness intensity was measured using ImageJ. Data were analyzed with analysis of variance and Tukey-Kramer honestly significant difference test. Significance was set at $\alpha=0.05$.

Results: Subjective assessment of the photographs showed that RI improved the appearance of the WSLs so that they resembled intact enamel. Mean FC-brightness intensities for intact, demineralized, and demineralized RI-treated areas were 159.6 ± 9.2 , 123.4 ± 7.2 , and 160.9 ± 11.5 , respectively. There were no significant differences in fluorescent intensity between the intact and RI areas ($p=0.58$). The demineralized areas had significantly lower fluorescent intensity than both the RI-treated and intact areas ($p<0.001$). LF values did not differ signifi-

*Kenneth Markowitz, DDS, MSD, Departments of Oral Biology and Restorative Dentistry, Rutgers School of Dental Medicine, Newark, NJ, USA

Kevin Carey, DMD, Department of Community Health, Rutgers School of Dental Medicine, Newark, NJ,

*Corresponding author: 185 S. Orange Ave, Newark, NJ 07103, USA; e-mail: markowkj@sdm.rutgers.edu

DOI: 10.2341/16-153-L

cantly between intact, demineralized, or RI-treated areas.

Conclusions: This study demonstrates the ability of RI to restore artificial WSLs to the esthetics and fluorescence of intact enamel. The FC can be used to assess the optical properties of WSLs and the impact of RI on these properties.

INTRODUCTION

The enamel white spot lesion (WSL) is an early manifestation of caries. When exposed to bacterially derived organic acids, subsurface enamel mineral loss occurs and results in porous enamel that appears white owing to its capacity to scatter light.¹ If the process of acid generation and enamel demineralization proceeds unchecked, the enamel surface will give way, resulting in a cavitated lesion.

WSLs can form on any plaque-covered enamel surface. Teeth with orthodontic brackets generally accumulate plaque and are frequently found to have WSLs when the orthodontic treatment is completed.² Typically, these lesions are arrested when access to the tooth surface is improved by removal of the orthodontic hardware, which allows for saliva exposure and more effective tooth brushing.³ Fluoride and various other agents can facilitate WSL remineralization.⁴ When WSLs regain mineral, the surface experiences a reduction in porosity, blocking the diffusion of mineral constituents into the subsurface areas of the lesion. Consequently, the use of remineralizing therapies does not ensure complete lesion resolution and restoration of the tooth's esthetics.^{5,6}

Resin infiltration (RI) is a professionally applied, microinvasive, treatment for WSLs where a low-viscosity resin-containing triethylene glycol dimethacrylate is applied to the acid-etched surface of the lesion and penetrates the WSL's porous enamel.^{7,8} After it penetrates the WSL, the resin is polymerized by light curing. By infiltrating the enamel surface and filling most of the porosities in the WSL, this procedure protects the enamel from further demineralization⁹ and improves the mechanical properties of the acid-damaged enamel surface.¹⁰ This procedure has been used to treat both interproximal lesions and WSLs on facial smooth surfaces.¹¹ Since the resins used in RI have indexes of refraction that match enamel, this procedure reduces the light scattering of the WSL, thereby improving the color and esthetics of the treated tooth.¹²⁻¹⁴ To date, assessments of the RI treatment's impact on the

appearance and optical properties of teeth with WSLs have been largely limited to the visual evaluation of treated teeth.¹⁵

Caries detector devices are used to identify caries at various stages of development including WSLs.¹⁶ Quantitative light fluorescence (QLF) (Inspektor Research Systems BV, Amsterdam, The Netherlands) uses a wavelength of light that induces dentin to fluoresce. WSLs scatter the fluorescent light and appear dark in QLF images. The device's software can calculate lesion volume and mineral loss.¹ A laser fluorescence (LF) device, the DIAGNOdent (KaVo, Biberach, Germany), excites bacterially derived pigment to fluoresce. The values on the device's numerical display indicate the intensity of this fluorescence in lesions.¹⁷ Fluorescent camera (FC)—based caries detection systems, such as the Vista-Proof (Dürr Dental AG, Bietigheim-Bissingen Germany) and the Spectra Caries Detection Aid (Air Techniques, Melville, NY, USA) measure alterations in the tooth's fluorescence as a means of detecting and assessing the severity of WSLs. In this determination, FCs function in a manner like the QLF—identifying WSLs as areas of reduced fluorescence observed as dark areas on fluorescent images. Like the LF device, the FC devices can detect and assess the severity of dentin caries by measuring the light-induced fluorescence of bacterial pigments.^{18,19} The FC can present fluorescent images of the tooth, or the device's Visix software can generate false color images of the tooth based on the fluorescent measurements that indicate areas of enamel demineralization and dentin caries as well as numerical values that reflect the severity of these changes.^{18,20}

Caries detectors that measure the optical properties of tooth structure can be used in studies comparing the effectiveness of various WSL treatments.^{3,21} Caries detectors that produce fluorescent images of teeth should be able to visualize and measure changes in the optical properties of WSLs brought about by RI since this treatment reduces light scattering within the lesion. QLF and spectrophotometer readings have been used in *in vitro* studies to evaluate how RI affects the optical properties of WSLs.^{12,22,23} Caries detectors have advantages to visual examination since they allow quantitation of subtle changes in the optical properties of WSLs.

In this *in vitro* study, we examined the effect of RI treatment on the FC and LF devices' assessment of the fluorescence of artificial WSLs created in human teeth by exposure of enamel to a lactic acid gel. Since RI reduces the light scattering of the WSL, we

hypothesized that treatment would restore the fluorescence of treated areas of WSLs to values that were equivalent to that of intact enamel. A subjective assessment of the visual appearance of the surfaces was also conducted to ensure that changes in the caries detector readings caused by demineralization and RI were reflected in the clinically important parameter of WSLs: the esthetics of the affected tooth surface. This study evaluated the potential contribution that the FC and LF devices can make to studies evaluating the impact of RI and other treatments on the appearance and optical properties of teeth with WSLs.

METHODS AND MATERIALS

Tooth Selection and Baseline Measurements

This *in vitro* study was performed using extracted third molars collected at the Rutgers School of Dental Medicine's Oral Surgery clinic. Consent to donate third molars for research projects was obtained from adult patients between the ages of 18 and 30 years. The university's Institutional Review Board approved the tooth collection procedure (Protocol number 0120050074).

Following extraction, teeth were debrided of adherent soft tissue and visually inspected. Ten caries-free and restoration-free teeth with intact buccal surfaces that were free of white areas or other discolorations were used in this study. Teeth were stored in 1% phenol solution and used within 1 month of collection. The buccal surfaces of the 10 selected teeth were photographed at eight-power magnification (DP12 Microscope Digital Camera System, Olympus, Tokyo, Japan).

LF readings from each tooth's buccal surface were taken with the DIAGNOdent model 2095 using the instrument's B-tip, as recommended by the manufacturer for smooth surface examinations. The instrument was calibrated per manufacturer's instructions. The B-tip lightly contacted the enamel surface and was gently rocked. The highest reading (instrument's maximum reading = 99) was recorded for each area examined. For each tooth, LF measurements were obtained for four areas near the tooth's height of contour located mesial and distal to the buccal groove. This area would be the future site of the demineralized lesion. Using Microsoft Power Point, LF readings obtained from the various buccal sites were superimposed, at the time that the readings were performed, onto the photograph of those tooth surfaces, creating an LF map of each tooth's buccal surface. In superimposing these

readings on the images of the tooth surface, care was taken to insert the readings on the sites examined with the LF device. This procedure was used in a previous study to create LF maps of cut and intact occlusal surfaces.²⁴

Fluorescent images from the buccal surfaces of the teeth were obtained using the Spectra Caries Detection Aid. The FC handpiece with an 8-mm spacer was placed over the buccal surface and the images obtained per the manufacturer's instructions. In order to collect data concerning subtle changes in the tooth structure's fluorescent intensity, the images provided by the FC, instead of the false color images and numerical readings generated by the device's software, were used in the subsequent data analysis. To obtain quantitative fluorescent values; the stored FC images were imported into the public domain image analysis program ImageJ (National institute of Health, Bethesda, MD, USA) digitized, and converted to grayscale; the brightness intensity values of selected areas were then recorded.²⁵ The brightness intensity values of the fluorescent images ranged from 0 to 244 arbitrary units. In preliminary experiments, where we varied the duration of the demineralizing treatment, we observed that these fluorescent intensities correlated better with the visual severity of the buccal surface WSL than did the numerical values obtained with the instrument's software. We also noticed that the instrument's software failed to detect areas of mild demineralization that were detectable on the fluorescent images and observed on visual examination. The buccal areas selected for analysis corresponded to the areas where LF measurements were previously obtained. ImageJ was also used to convert FC images into line graph maps of the tooth surfaces where fluorescent intensity values of all imaged sites are displayed.

In using both caries detectors, measurements were taken in triplicate to ensure reproducibility. All photos and caries detector measurements were taken in dim ambient light and from moist tooth surfaces to simulate intraoral conditions. Following water rinsing, the teeth were exposed to a brief air current to remove excess fluid and leave the tooth surface moist; care was taken to avoid accumulations of fluid on the teeth that would cause reflections. A training and calibration exercise was held where both examiners (KM and KC) obtained FC images and LF measurements from the buccal surfaces of 10 teeth. By following uniform procedures for positioning the handpieces of the two instruments, the LF values

obtained by the two examiners were within ± 2 units, and the FC fluorescent intensity values were $\pm 5\%$.

Creation of Artificial White Spot Lesions

Following baseline measurements, the 10 teeth were coated with waterproof nail polish except for a 1×4 mm window on the buccal surface of each tooth. The teeth were then placed into separate containers with 25 mL of a gel containing 0.1 M lactic acid and 1.5 mM CaPO_4 adjusted to pH 4.5 with sodium hydroxide and thickened with ethyl cellulose (Natrosol, Ashland Aqualon Inc, Parlin, NJ, USA).²⁶ The containers holding the demineralizing teeth were agitated twice daily, and the gel was changed after 7 days. This demineralization treatment was carried out for 14 days at room temperature. Following acid exposure, the teeth were rinsed with deionized water and inspected under eight-power magnification to ensure that the surface of the acid-exposed area was intact and the tooth structure was white when viewed wet. The nail varnish was then removed by agitating the teeth in acetone. After we created the artificial WSLs, the buccal surfaces of the teeth were photographed and evaluated with the LF and FC devices using methods identical to those used to obtain the baseline measurements.

Following the experiment, four of the 10 teeth used were sectioned perpendicular to the long axis of the tooth using a low-speed saw (Isomet, Buehler LTD., Lake Bluff, IL, USA) with diamond blade and water lubrication. Tooth slices 0.25-mm thick were cut through the WSLs and polished with 600-grit silica carbide paper. The areas of the sections containing the WSLs were examined at 50-power magnification. The WSLs were observed to extend to $150 \pm 8.2 \mu\text{m}$ below the enamel surface.

Application of the Resin Infiltration Treatment and Final Measurements

The mesial or distal half of each tooth's WSL was randomly selected to receive the RI treatment (Icon, DMG-Hamburg, Hamburg, Germany) following the manufacturer's instructions for smooth-surface lesion application. The procedure involved drying the buccal surfaces with air from an air-water syringe and applying the Icon kit's HCl etch. After removing the etchant by copious rinsing and drying with an air-water syringe, the kit's ethanol drying treatment was applied and the tooth surface dried thoroughly. This was followed by application of the RI material and light curing for 30 seconds using an ESPE Elipar light curing unit (3M, St Paul, MN, USA). The etchant, ethanol drying agent, and RI material were

applied using the syringes and applicator tips provided in the Icon kit that are intended to be used for treating facial smooth-surface lesions. As recommended by the manufacturer, the RI material was applied and light cured twice. Care was taken to limit application of the etchant and RI to the WSL and to avoid applying excess material. Following RI application, the teeth were stored overnight in a humid atmosphere and then a final set of photographs, FC images, and LF measurements were made.

Subjective Assessment of WSLs

As an independent method of assessing the severity of the WSLs and the impact of RI, photographs of the teeth used in this study were rated for the presence and intensity of visible WSLs by a panel of five dentists (two orthodontists and three restorative dentists) that did not include the authors. Each evaluator was shown photographs of the teeth used in this study on a flat computer screen. The evaluators were trained to rate the photos using a 0 to 10 visual analog scale (VAS) in which 0 represented no visible WSL and 10 represented an obvious WSL. Following this training the examiners' ratings were found to agree with the ratings of the investigators to within ± 1 . This type of assessment utilizing photographs and a VAS scale has been used in a clinical study of the effect of RI on WSL esthetics.¹⁵ The photos showed the buccal surfaces prior to demineralization, following the creation of the WSL, and following RI of the mesial or distal half of the WSL. In addition to these 30 photos, each evaluator was shown 10 duplicate images so that the reproducibility of the assessment could be determined. The photos were shown in random order and cropped to show the mesial or distal half of the tooth's buccal surface; hence, separate images showed demineralized or RI-treated areas.

Data Analysis

LF, FC brightness intensity readings, and subjective VAS ratings are reported as mean \pm standard deviation. A repeat measure analysis of variance (ANOVA) with pairwise Tukey-Kramer test was performed using JMP version 11 statistical software (SAS institute, Cary, NC, USA) to determine if significant differences in these parameters were measured from intact, demineralized, and RI enamel.

In conducting the subjective visual assessments of the tooth surfaces, each examiner rated 10 duplicate photographs. A weighted kappa coefficient was

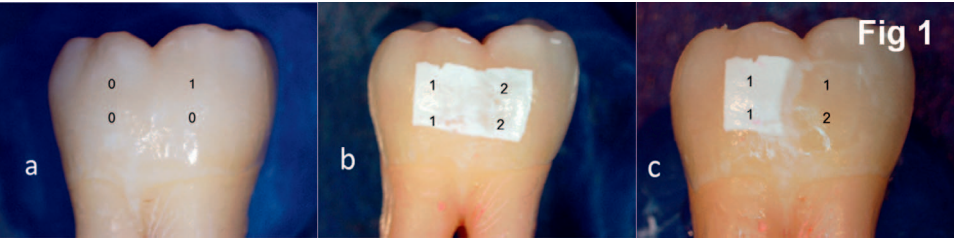


Figure 1. Photographs with superimposed LF readings of the buccal surface of an extracted third molar (A): before demineralization, (B): after demineralization, and (C): after RI of the right half of the demineralized area. The low LF values recorded from the intact tooth tissue were not affected by demineralization or RI treatment.

calculated for intraobserver and interobserver agreement using SAS version 9.4 (SAS institute).

Statistical significance was set at $\alpha=0.05$. Based on preliminary experiments, a minimum sample size of eight teeth was necessary to achieve a statistical power of 0.8 in resolving a fluorescent intensity difference of 20% as measured by the FC, with standard deviations of approximately $\pm 10\%$ of the intensity value.

RESULTS

Effect of Demineralization and RI on Appearance of the Tooth Structure and LF Readings

Photographs showing the buccal surface of a tooth with LF readings obtained from the areas over which the numbers are superimposed are shown in Figure 1A. Intact tooth structure has LF readings close to zero. A rectangular area of the same tooth’s buccal surface was exposed to a pH 4.5 lactic acid gel. This resulted in the white area of demineralization shown in Figure 1B. Despite the marked change in appearance of the tooth surface following acid treatment, LF measurements in the demineralized areas were still close to zero. When half of the demineralized area was treated with RI, there was a marked change in the appearance of the demineralized enamel so that it resembled intact tooth structure (Figure 1C). LF readings obtained from the RI-treated half of the demineralized area continued to be close to zero. As shown in Table 1, intact teeth had a mean LF reading of 1.07 ± 0.9 . Following demineralization and RI, the mean LF

readings were 1.25 ± 0.9 and 0.9 ± 0.9 , respectively. Both demineralization and RI failed to cause significant changes in the LF readings obtained from the buccal surface of the 10 teeth used in this study ($p=0.69$ and $p=0.67$, respectively).

Effect of Demineralization and RI on Fluorescent Intensity of the Tooth Structure Measured With the FC

FC images of the same tooth shown in Figure 1 are shown in Figure 2. Prior to demineralization, the buccal surface presents a uniform fluorescent intensity (Figure 2A). Following acid treatment of a rectangular portion of the buccal enamel, reduced fluorescence was observed in the acid-exposed area (Figure 2B). Following RI (Figure 2C), the treated portion of the demineralized area showed increased fluorescent intensity and resembled the intact areas of the same tooth.

Figure 3 shows fluorescent brightness intensity readings measured from the buccal surface of one tooth; mesial-distal locations are on the X-axis, gingival-occlusal locations are plotted on the Y-axis, and the Z-axis values for each location on the lines represent image brightness indicating the fluorescent intensity. The intact tooth shown in Figure 3A has a peak fluorescent intensity in the center of the crown near the tooth’s height of contour. Following demineralization of a rectangular window, the fluorescent intensity of this demineralized area was reduced (Figure 3B.). Following application of RI to the right portion of the demineralized area, the fluorescent intensity of the treated portion was observed to increase (Figure 3C). The fluorescent intensities measured in the RI-treated area were less uniform than the intensities measured in the same area prior to demineralization.

The mean fluorescent intensity values measured by the FC from the 10 teeth used in this study obtained prior to and following demineralization and from RI-treated areas are shown in Figure 4. Intact enamel had a fluorescent intensity of 159.6 ± 9.2 . Demineralization caused a drop in fluorescent intensity to 123.4 ± 7.2 , a 22.7% reduction that

Table 1: Effect of Demineralization and Resin Infiltration on LF Measurements (Mean \pm SD) Obtained from the Buccal Surfaces of Teeth In Vitro (N=10)			
	Intact	Demineralized	RI-Treated Areas
LF readings	1.07 ± 0.9	$1.25 \pm 0.9^*$	$0.9 \pm 0.9^*$
Abbreviation: LF, laser fluorescence; RI, resin infiltration; SD, standard deviation.			
* Not statistically different compared with initial measurement ($p>0.05$).			

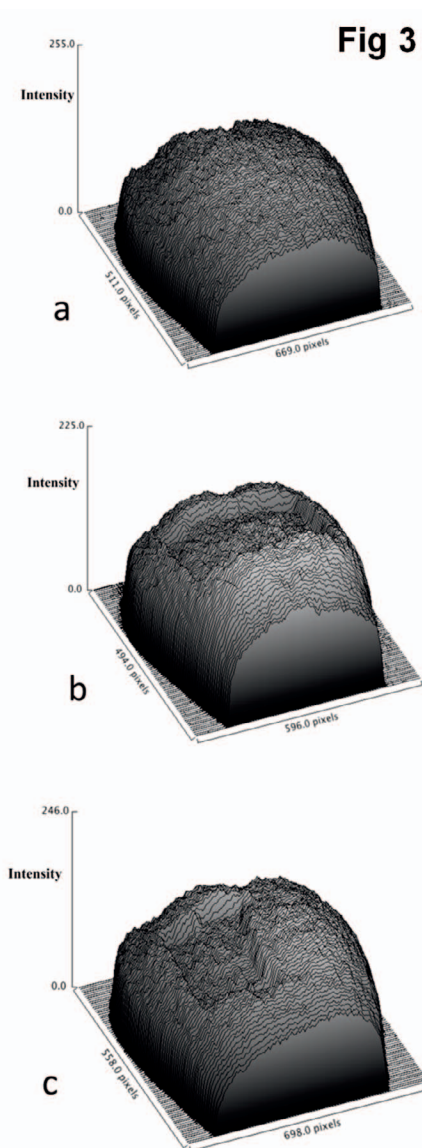
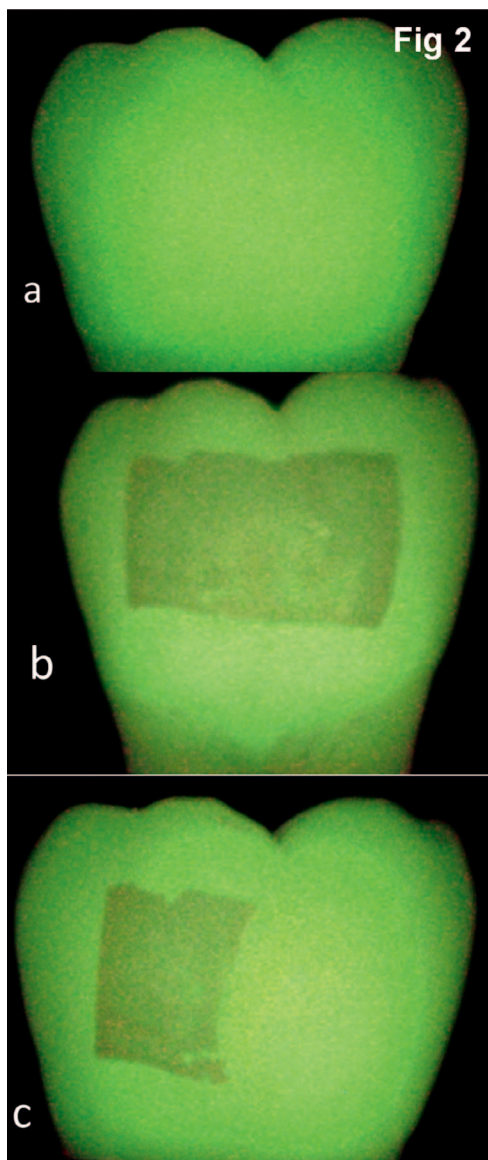


Figure 2. FC images of the same extracted third molar shown in Figure 1 (A): Before demineralization, (B): after demineralization, and (C): after RI treatment of the right half of the demineralized area. Demineralized enamel appears darker than intact areas due to the scattering of light from the fluorescent dentin.

Figure 3. Brightness intensity plots generated from the FC images shown in Figure 2. These images were converted to three-dimensional brightness intensity maps to highlight the effects of demineralization and RI on (A): the intact tooth, (B): demineralized area, and (C): demineralized area where the right half of the demineralized enamel was resin infiltrated.

was statistically significant ($p < 0.001$). Following RI of half of each demineralized area, the mean fluorescent intensity increased to 160.9 ± 11.5 , a value that was significantly higher than the value for untreated demineralized areas ($p < 0.001$) and not significantly different from the values measured from the intact tooth surfaces ($p = 0.58$).

Subjective Assessment of Demineralized and RI Surfaces

Demineralized enamel surfaces had a distinct chalky white appearance and were readily distinguished by the evaluators and rated highly on the WSL severity scale used in this study. The mean subjective VAS scores for intact, demineralized, and RI surfaces were 1.81 ± 1.32 , 9.77 ± 0.58 , and 3.77 ± 1.74 ,

respectively (Table 2). Each of these values was significantly different from the other values ($p < 0.001$). Weighted kappa coefficients for intra-observer and interexaminer agreement were 0.81 and 0.73, respectively, indicating good agreement.

DISCUSSION

RI treatment is used to halt the progression of WSLs and improve the appearance of teeth with WSLs.¹³ Facial WSLs can be directly observed, but visual examination is difficult to quantify in research studies comparing RI with other methods of WSL treatment.^{5,15} Several methods of caries detection have been applied to evaluate the effect of preventive therapies on facial WSLs. The QLF method has been used to monitor the remineralization of WSLs *in*

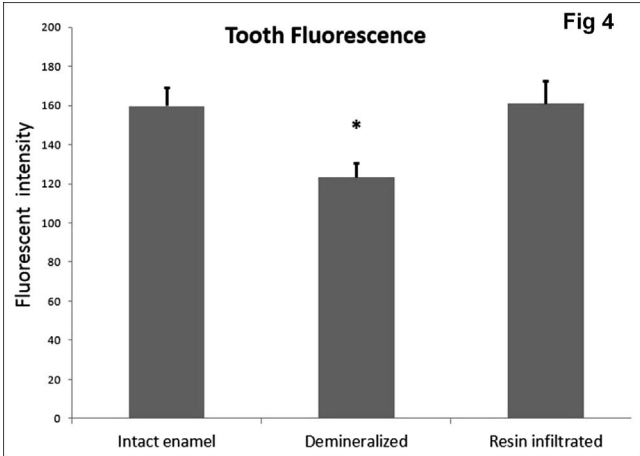


Figure 4. Mean (\pm standard deviation) fluorescent intensity readings measured by the FC device from the buccal surface of extracted molars (N=10). Left bar represents intact surfaces, middle bar demineralized surfaces and right bar RI surfaces. The demineralized surfaces (*) had significantly lower brightness intensities than the other two groups ($p < 0.01$).

vitro and *in vivo* following the removal of fixed orthodontic appliances.³ In a recent study utilizing the QLF in examining the efficacy of a dentifrice-delivered anti-caries agent, a specially designed apparatus was used to position the patient when measurements were taken from facial WSLs on the anterior teeth. This positioning device was required to ensure the reproducibility of the QLF readings taken at periodic examinations during the treatment period.²⁷

The FC and LF devices are simple to use and have been reported to have excellent reliability.^{28,29} Since RI alters the optic properties of WSLs, our aim was to characterize the effects of RI on LF and FC readings obtained from teeth with artificial WSLs. The WSLs examined in this study were induced by exposing the enamel to a pH 4.5 lactic acid gel. These WSLs were visible when the tooth surfaces were wet, indicating subsurface mineral loss, and were observed to extend approximately 150 μ m into the enamel. This is a typical depth for artificial WSLs on third molars.^{26,30}

FC images of the intact buccal surfaces of the extracted third molars used in this study showed characteristic green fluorescence indicative of caries-free tooth structure. In order to quantitatively assess the impact of demineralization and RI on tooth fluorescence, we measured the brightness intensity of FC images rather than the false color images generated by the FC's Visix software. This analysis was accomplished using public domain software and could be used in a variety of clinical research

Table 2: Subjective Assessment of WSL Severity by a Panel of Five Dentists.	
Enamel Condition	Subjective WSL VAS Score (Mean \pm SD)
Intact	1.81 \pm 1.32
Demineralized	9.77 \pm 0.58*
Resin infiltrated	3.77 \pm 1.74*
Abbreviations: SD, standard deviation; VAS, visual analog scale; WSL, white spot lesion.	
* Significantly different from the VAS score of intact enamel ($p < 0.05$).	

settings. Following demineralization, we observed a mean 22.7% reduction in the fluorescent brightness intensity of the tooth surface. This value is within the range of QLF-measured fluorescent intensity reductions reported in studies where the fluorescent intensity of natural and artificial WSLs was compared with intact tooth structure.³¹ This agreement between the QLF and FC was expected since both devices measure excited dentin fluorescence that is partially blocked by light scattering within WSLs.¹

Following RI, the fluorescent brightness intensity of the treated demineralized area was restored to values that were not statistically different from those of sound enamel. This supports our hypothesis concerning the effect of RI on WSL fluorescence and agrees with fluorescence measurement results obtained using the QLF.²³ The reason for the small variations in fluorescent intensity observed in the line graph plot of RI enamel (Figure 3C) is not known. Possibly some areas with residual light scattering exist within the RI area.

The finding of this study is consistent with the observed actions of RI on the visual appearance of WSLs as demonstrated by our subjective assessment. In our subjective WSL assessment, the examiners assigned the RI areas significantly higher values than the sound surfaces. These VAS values were well below those given to the demineralized surfaces. It is possible that the smooth and more reflective surface of the RI area or traces of the peripheral aspects of the demineralized area, as observed in Figure 1, affected the examiner rating of the RI surfaces.

Enamel areas with caries-associated WSLs have been reported as having LF readings that are higher than those of sound enamel but lower than those of cavitated lesions.²¹ In the current study, LF readings obtained from intact buccal surfaces were close to zero. The LF readings did not significantly change following creation of the WSL or RI of the demineralized enamel, remaining at the low end of the

instrument's range. Since the LF device has been used in clinical studies to assess remineralization, this result was unexpected.^{21,32} The LF device measures laser excited fluorescence from bacterial pigments such as porphyrin.^{17,24} In naturally occurring WSLs, bacterial pigment absorption by the demineralized enamel has been implicated as being responsible for the LF readings obtained from these lesions.²¹ The results of this study and our investigations indicate that LF does not measure demineralization directly.

RI is an evolving technology with expanding indications, where various material or treatment procedure modifications influence the efficacy of the treatment.³³ Improved experimental methods and study designs are needed to determine the most effective method of WSL esthetic management.³⁴ Quantitative methods such as the FC can be used in clinical studies comparing RI with other methods of WSL esthetic management. In clinical practice, visual observation and patient satisfaction with esthetics are the main goals of facial surface WSL treatment. This is particularly true where RI is concerned since this treatment does not replace mineral lost during the demineralization. Clinicians can use the FC to illustrate areas of dentition with early carious lesions and highlight the need for minimally invasive intervention. The results of this study do not support the use of the LF device in studies of this nature. We plan to use the FC in clinical studies comparing the esthetic impact of RI with remineralization treatments. Maps of the tooth surface's fluorescent brightness intensity (Figure 3) can also be used in future studies to distinguish areas of a WSL that were treated with different RI formulations or procedures.

CONCLUSIONS

When viewed using the FC, artificial WSLs appear as dark areas of reduced fluorescent intensity. RI restores the fluorescent intensity of demineralized areas to values equivalent to those obtained from intact enamel. These changes in fluorescence accompany changes in appearance of the WSL, where RI improves the esthetics of demineralized enamel. In contrast, values obtained from the tooth structure with an LF caries detector were not significantly altered by WSL formation or resin infiltration. These results indicate that the FC caries detector can be used in studies comparing the ability of RI and other methods of WSL treatment to improve the appearance and optical properties of these lesions.

Acknowledgements

The authors thank Air Techniques for donating the Spectra Caries Detection Aid and for providing technical support. We thank DMG America for donating samples of the Icon resin infiltration system. We thank David Furgang and Shuying Jiang for assisting with the statistical analysis. We also thank the participants in the panel who performed the subjective evaluation of the white spot lesions. Neither of the authors have any conflict of interest pertinent to this research.

Regulatory Statement

This *in vitro* study was conducted in accordance with all the provisions of the local human subjects oversight committee guidelines and policies of the Rutgers Biomedical and Health Sciences and the university's Institutional Review Board. The approval code for this study is: 0120050074.

Conflict of Interest

The authors of this manuscript certify that they have no proprietary, financial, or other personal interest of any nature or kind in any product, service, and/or company that is presented in this article.

(Accepted 18 June 2017)

REFERENCES

1. van der Veen MH, & de Josselin de Jong E (2000) Application of quantitative light-induced fluorescence for assessing early caries lesions *Monograph in Oral Science* 17 144-162.
2. Ren Y, Jongsma MA, Mei L, van der Mei HC, & Busscher HJ (2014) Orthodontic treatment with fixed appliances and biofilm formation—A potential public health threat? *Clinical Oral Investigations* 18(7) 1711-1718.
3. Al-Khateeb S, Forsberg CM, de Josselin de Jong E, & Angmar-Mansson B (1998) A longitudinal laser fluorescence study of white spot lesions in orthodontic patients *American Journal of Orthodontics and Dentofacial Orthopedics* 113(6) 595-602.
4. Fontana M, Young DA, & Wolff MS (2009) Evidence-based caries, risk assessment, and treatment *Dental Clinics of North America* 53(1) 149-161
5. Bailey DL, Adams GG, Tsao CE, Hyslop A, Escobar K, Manton DJ, Reynolds EC, & Morgan MV (2009) Regression of post-orthodontic lesions by a remineralizing cream *Journal of Dental Research* 88(12) 1148-1153.
6. Bröchner A, Christensen C, Kristensen B, Tranaeus S, Karlsson L, Sonnesen L, & Twetmen S (2011) Treatment of post-orthodontic white spot lesions with casein phosphopeptide-stabilised amorphous calcium phosphate *Clinical Oral Investigations* 15(3) 369-373.
7. Phark JH, Duarte S Jr, Meyer-Lueckel H, & Paris S (2009) Caries infiltration with resins: A novel treatment option for interproximal caries *Compendium of Continuing Education in Dentistry* 30(Special No 3) 13-17.
8. Paris S, Schwendicke F, Keltsch J, Dorfer C, & Meyer-Lueckel H (2013) Masking of white spot lesions by resin infiltration *in vitro Journal of Dentistry* 41(Supplement 5) e28-34.

9. Paris S, & Meyer-Lueckel H (2010) Inhibition of caries progression by resin infiltration in situ *Caries Research* **44**(1) 47-54.
10. Paris S, Schwendicke F, Seddig S, Muller WD, Dorfer C, & Meyer-Lueckel H (2013) Microhardness and mineral loss of enamel lesions after infiltration with various resins: Influence of infiltrant composition and application frequency in vitro *Journal of Dentistry* **41**(6) 543-548.
11. Ekstrand K, Martignon S, Bakhshandeh A, & Ricketts DN (2012) The non-operative resin treatment of proximal caries lesions *Dentistry Update* **39**(9) 614-616, 618-620, 622.
12. Hallgren K, Akyalcin S, English J, Tufekci E, & Paravina RD (2016) Color properties of demineralized enamel surfaces treated with a resin infiltration system *Journal of Esthetic and Restorative Dentistry* **28**(5) 339-346.
13. Paris S, & Meyer-Lueckel H (2009) Masking of labial enamel white spot lesions by resin infiltration—A clinical report *Quintessence International* **40**(9) 713-718.
14. Rocha Gomes Torres C, Borges AB, Torres LM, Gomes IS, & de Oliveira RS (2011) Effect of caries infiltration technique and fluoride therapy on the colour masking of white spot lesions *Journal of Dentistry* **39**(3) 202-207.
15. Senestraro SV, Crowe JJ, Wang M, Vo A, Huang G, Ferracane J, & Covell DA Jr (2013) Minimally invasive resin infiltration of arrested white-spot lesions: A randomized clinical trial *Journal of the American Dental Association* **144**(9) 997-1005.
16. Pretty IA (2006) Caries detection and diagnosis: Novel technologies *Journal of Dentistry* **34**(10) 727-739.
17. Lussi A, Hibst R, & Paulus R (2004) DIAGNOdent: an optical method for caries detection *Journal of Dental Research* **83**(Special No C) C80-83.
18. Graye M, Markowitz K, Strickland M, Guzy G, Burke M, & Haupt M (2012) In vitro evaluation of the Spectra early caries detection system *Journal of Clinical Dentistry* **23**(1) 1-6.
19. Jablonski-Momeni A1, Heinzl-Gutenbrunner M, & Klein SM (2014) In vivo performance of the VistaProof fluorescence-based camera for detection of occlusal lesions *Clinical Oral Investigations* **18**(7) 1757-1762.
20. Achilleos EE, Rahiotis C, Kakaboura A, & Vougiouklakis G (2013) Evaluation of a new fluorescence-based device in the detection of incipient occlusal caries lesions *Lasers in Medicine and Science* **28**(1) 193-201.
21. Moriyama CM, Rodrigues JA, Lussi A, & Diniz MB (2014) Effectiveness of fluorescence-based methods to detect in situ demineralization and remineralization on smooth surfaces *Caries Research* **48**(6) 507-514.
22. Knosel M, Eckstein A, & Helms HJ (2013) Durability of esthetic improvement following Icon resin infiltration of multibracket-induced white spot lesions compared with no therapy over 6 months: A single-center, split-mouth, randomized clinical trial *American Journal of Orthodontics and Dentofacial Orthopedics* **144**(1) 86-96.
23. Yuan H, Li J, Chen L, Cheng L, Cannon RD, & Mei L. (2014) Esthetic comparison of white-spot lesion treatment modalities using spectrometry and fluorescence *Angle Orthodontist* **84**(2) 343-349.
24. Markowitz K, Stenvall RM, & Graye M (2012) The effect of distance and tooth structure on laser fluorescence caries detection *Operative Dentistry* **37**(2) 150-160.
25. Jensen EC (2013) Overview of live-cell imaging: Requirements and methods used *Anatomical Record (Hoboken)* **296**(1) 1-8.
26. Boyle EL, Higham SM, & Edgar WM (1998) The production of subsurface artificial caries lesions on third molar teeth *Caries Research* **32**(2) 154-158.
27. Yin W, Hu DY, Li X, Fan X, Zhang YP, Pretty IA, Mateo LR, Cummins D, & Ellwood RP (2013) The anti-caries efficacy of a dentifrice containing 1.5% arginine and 1450 ppm fluoride as sodium monofluorophosphate assessed using quantitative light-induced fluorescence (QLF) *Journal of Dentistry* **41**(Supplement 2) S22-S28.
28. Diniz MB, Boldieri T, Rodrigues JA, Santos-Pinto L, Lussi A, & Cordeiro RC (2012) The performance of conventional and fluorescence-based methods for occlusal caries detection: An in vivo study with histologic validation *Journal of the American Dental Association* **143**(4) 339-350.
29. Seremidi K, Lagouvardos P, & Kavvadia K (2012) Comparative in vitro validation of VistaProof and DIAGNOdent pen for occlusal caries detection in permanent teeth *Operative Dentistry* **37**(3) 234-245.
30. Belli R, Rahiotis C, Schubert EW, Baratieri LN, Petschelt A, & Lohbauer U (2011) Wear and morphology of infiltrated white spot lesions *Journal of Dentistry* **39**(5) 376-385.
31. Angmar-Mansson B, & ten Bosch JJ (2001) Quantitative light-induced fluorescence (QLF): A method for assessment of incipient caries lesions *Dentomaxillofacial Radiology* **30**(6) 298-307.
32. Demito CF, Rodrigues GV, Ramos AL, & Bowman SJ (2011) Efficacy of a fluoride varnish in preventing white-spot lesions as measured with laser fluorescence *Journal of Clinical Orthodontics* **45**(1) 25-29.
33. Meyer-Lueckel H, Chatzidakis A, Naumann M, Dorfer CE, & Paris S (2011) Influence of application time on penetration of an infiltrant into natural enamel caries *Journal of Dentistry* **39**(7) 465-469.
34. Sonesson M, Bergstrand F, Gizani S, & Twetman S (2017) Management of post-orthodontic white spot lesions: An updated systematic review *European Journal of Orthodontics* **39**(2) 116-121.