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Evaluation of the clinical efficacy of quantitative light-induced fluorescence technology in diagnosing cracked teeth

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ABSTRACT

Background: This retrospective study evaluated the clinical efficacy of quantitative light-induced fluorescence (QLF) technology for crack detection and the diagnosis of cracked teeth and assessed the possibility of a quantitative evaluation of cracks using QLF technology.
Methods: Patients who were clinically diagnosed with cracked teeth over a 1-year period were included. The QLF images of the corresponding symptomatic cracked teeth and asymptomatic contralateral teeth with crack lines were taken with Qraypen C (AIOBIO, Seoul, Korea). Fluorescence loss (ΔF), maximum fluorescence loss (ΔFmax), red fluorescence (ΔR), and maximum red fluorescence (ΔRmax) of the crack line were analyzed. The correlation between these parameters and sex, age, tooth position (1st premolar, 2nd premolar, 1st molar, 2nd molar), spontaneous pain (+/-), percussion test (+/-), cold test (++/+/-), and bite test (+/-) were statistically analyzed.
Results: A total of 66 patients were included. Twenty-four patients had asymptomatic contralateral teeth with apparent crack lines; thus, 90 teeth were analyzed. The crack lines in 84 teeth observed as red fluorescent lines on the QLF images showed ΔR values higher than the cut-off value set by the analysis program used. The patient's age and the |ΔF| and ΔR values were positively correlated. However, there was no statistically significant difference in the QLF parameters between the same patient's symptomatic tooth and the contralateral tooth.
Conclusions: QLF technology is a useful assistive diagnostic device for diagnosing cracked teeth.

1. Introduction

According to the 2020 version of the American Association of Endodontists Glossary of Endodontic Terms, a crack refers to “a thin surface disruption of enamel and dentin, and possibly cementum, of unknown depth or extension” [1]. Cameron first introduced the term “cracked tooth syndrome” [2] and defined it as an “incomplete fracture of a vital posterior tooth that involves the dentine and occasionally extends into the pulp” [3].

The causes of cracked tooth syndrome include excessive tooth preparation, inadequate restoration design (e.g., insufficient cuspal protection), stress concentration, excessive masticatory force, physical trauma, dental caries, parafunction, and balancing side interference [3]. Cracked tooth syndrome can occur at various ages, but it is most common in people in their 40 s and 50 s, and there is no significant difference in its incidence according to sex [4]. A cracked tooth can cause various symptoms depending on its depth. In the initial stage, pulp has vitality and sharp pain can occur during mastication or when eating cold food [5]. In addition, when eating fibrous food, a symptom called “rebound pain” can occur when pressure on the tooth is released [6]. As the crack progresses, it may progress to the pulp, causing pulpal necrosis or vertical fracture of the tooth [7]. Therefore, it is important to diagnose cracked teeth early and prevent the progression of cracks.

However, the symptoms of cracked teeth are sporadic and may be

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difficult to reproduce, and they are not easily observed through radiographs and visual inspection [8,9]. Techniques to observe cracked teeth and diagnose crack lines by visual inspection more easily include dental microscopy with shadow-free co-axial illumination [10], trans-illumination [11], and staining techniques using methylene blue, iodine, and gentian violet [12].

Quantitative light-induced fluorescence (QLF) technology detects early carious lesions by irradiating teeth with visible blue light at a wavelength of 405 nm. Green autofluorescence occurs when sound teeth are irradiated with this wavelength of visible light. However, in carious lesions, the fluorescence is lost as light is scattered, and the area appears dark. Loss of fluorescence (ΔF) can be measured by comparing sound areas with those in which fluorescence disappears. In addition, red fluorescence (ΔR) caused by bacterial metabolites such as porphyrin can be quantified by comparison with normal areas as an indicator of bacterial deposition at the lesion site [13].

Bacteria can inhabit crack lines, and Ricucci et al. [14] reported that bacterial colonization was always observed in histological analyses of crack lines and the surrounding dentinal tubules. Therefore, through QLF technology, the presence and location of cracked teeth can be confirmed by detecting the red fluorescence of residual porphyrin secreted by microorganisms in cracks [13]. In a case report by Jun et al. [15] and a case series by Son et al. [16], crack lines appearing as red fluorescence using QLF were observed extending to the pulp cavity during root canal treatment. Thus, the QLF technology can confirm the depth of crack lines and may have advantages in predicting the prognosis of cracked teeth. Compared with the ability of radiography and visual inspection, QLF was reported to have a higher crack line detection ability [17].

As observed in demineralized tooth structure, light is scattered, and transmission is obstructed by the crack line in the tooth, resulting in decreased fluorescence. Therefore, the crack line shows a lower ΔF value than the surrounding tooth structure on the QLF image, and this value seems to be correlated with the depth of the crack [18]. They analyzed 96 extracted teeth suspected of having cracks using QLF performed with a digital camera and reported a strong correlation between the presence and depth of an enamel crack and the maximum degree of fluorescence loss (ΔFmax). Therefore, it may be possible to use QLF to detect cracks and quantify their depth and progression.

To our best knowledge, clinical study that evaluated application of QLF technology on diagnosing cracked teeth is scarce. This study aimed to confirm the clinical efficacy of QLF technology for crack detection and the diagnosis of cracked teeth and to assess the possibility of a quantitative evaluation of cracks using QLF technology by examining the relationship between the clinical symptoms of teeth with crack lines and QLF parameters, fluorescence loss and red fluorescence. The null hypothesis was there would be no correlation between the QLF parameters driven by QLF images of cracked teeth and the clinical symptoms of patients with cracked teeth.

2. Materials and methods

2.1. Patient selection

This retrospective study was conducted after receiving approval from the Gangnam Severance Dental Hospital (IRB No.: 3–2021–0422). Patients who visited the Department of Conservative Dentistry at the Gangnam Severance Dental Hospital from January 2021 to December 2021 had their teeth inspected using a Qraypen C (AIOBIO, Seoul, Korea) and had dental images taken. Of these patients, those who met all the following criteria were included: discomfort upon biting, one or more crack lines observed visually on the tooth in the relevant area, and a clinical diagnosis of cracked teeth. Cases in which the diagnosis was unclear or patients felt masticatory pain due to other causes, such as periodontitis, apical periodontitis, and temporomandibular disorder, were excluded.

2.2. Data collection

Data on patients' age and sex were collected, and the teeth were divided based on type (first premolars, second premolars, first molars, and second molars). In addition, the presence of spontaneous pain and the results of the percussion, cold, and bite tests of each tooth were recorded. The result of the percussion test was recorded as +/- after tapping teeth with a mirror handle or the back of a pincette. In the bite test, teeth were divided into +/- after biting into a cotton roll. If there was any discomfort or pain, including the "rebound pain" that occurs right after the release of bite force, the result was recorded as "+," and if there was no discomfort at all, the result was "-." In the cold test, the teeth were evaluated using an ice stick or a cotton swab that was cooled with a refrigerant spray. If response was normal, the result was recorded as "+," and if there was increased sensitivity or pain, it was recorded as "++." The result was recorded as "-" if there was no response.

The images of the target tooth with the crack line were then obtained using a Qraypen C, the pen-shaped intraoral imaging device that can obtain white light and fluorescent image at the same time. Fluorescent images are obtained by irradiating light with wavelength of 405 nm $\pm$ 5 and energy power density of 270 $\pm$ 20% mW. To obtain consistent image quality to the highest extent possible, a W-Block (AIOBIO) was installed, and a Qraypen C was positioned according to the manufacturer's instructions before taking images. At this time, the contralateral tooth of the target tooth was photographed and clinically tested using the same methods as described above. For comparison purposes, patients who felt no symptoms and those with visual crack lines who showed normal reactions to the clinical tests were included in the analysis.

2.3. QLF image analysis

The images were first automatically saved in Q-Ray software version 1.45 (AIOBIO) automatically and transferred to a secure network-attached storage. A specially developed program was used to quantitatively analyze cracks in natural teeth from in vivo fluorescence images (captured with Qraypen C at 405 nm excitation with 1280 $\times$ 720-pixel resolution)—CrazeLineWizard01 v1.02 (Inspektor Research Systems B. V., The Netherlands) (Fig. 1). First, the user manually outlined the crack using a band with a width of 72 pixels. This curve-shaped band was then automatically transformed into a stretched image with the width of the curved band (72 pixels) and with a length equaling the flattened curve length.

An original sound surface was then approximated by the program for each pixel in the image by linear interpolation of the relative colors of the pixels at either side, the far left and right borders of the straightened band with the crack in the center line of the band. The band border lines with thickness of one pixel and length of the crack thus formed the effective field of reference for reconstruction of the sound surface in the band. By using the pixels of the original image and the reconstructed one, the program then calculated the following parameters: ΔF (mean value of the fluorescence change for all points where ΔFpix $<-3\%$), ΔFmax (the maximum value of the fluorescence loss), ΔR (mean value of the fluorescence change for all points where ΔRpix $>5\%$), ΔRmax (the maximum value of ΔR). All parameters were recorded for each tooth. When two or more separate crack lines existed due to restoration, each crack line was individually analyzed, and the highest value was selected among the parameters derived from each crack line. A single clinically experienced post-graduated resident analyzed all images.

2.4. Statistical analysis

For the statistical analysis, SAS version 9.4 (SAS Institute, Cary, NC, USA) was used. The Shapiro-Wilk test was used to test for the normality of data distribution at the 95% confidence level. To investigate the correlations between ΔF, ΔFmax, ΔR, and ΔRmax and patients' sex, age,

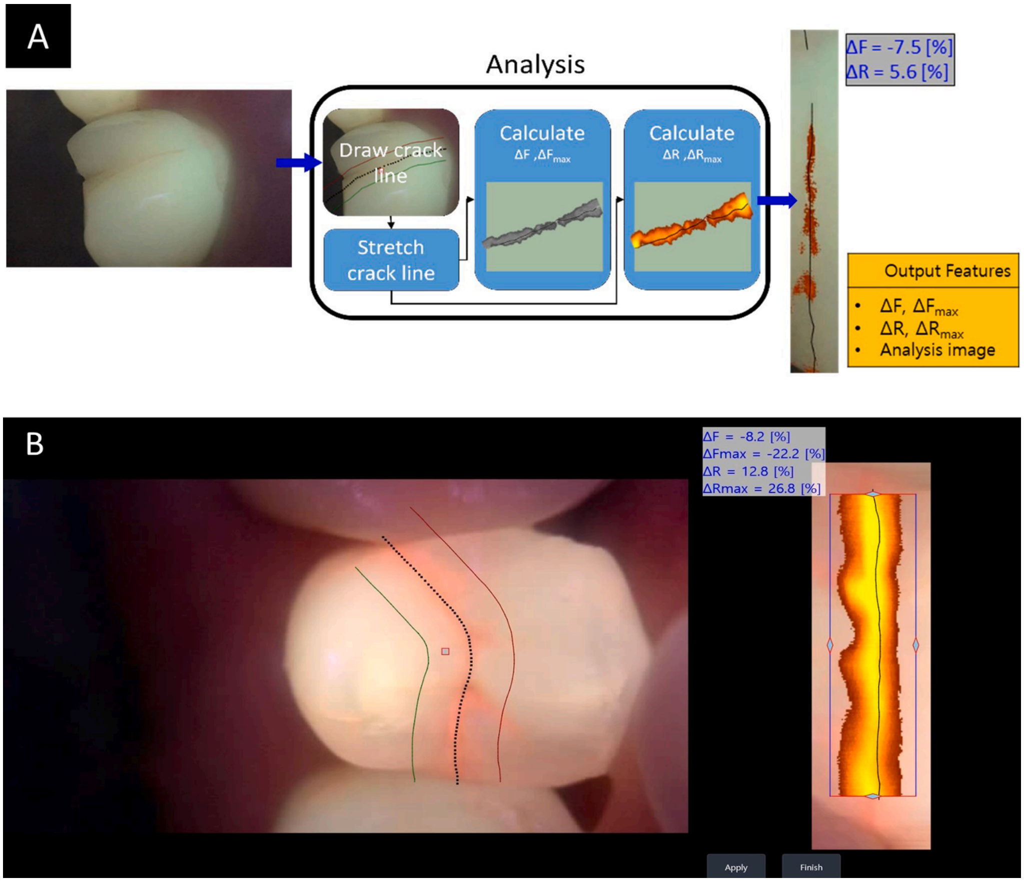

Fig. 1. Prototype program (CrazeLineWizard01 v1.02) used for crack line analysis. (A) Schematic of analysis process. (B) A representative image of a cracked tooth and the results of the analysis.

tooth position, spontaneous pain, percussion, bite, and cold test results, the independent two-sample *t*-test, analysis of variance (ANOVA), and Bonferroni post-hoc tests were used at the 95% confidence level. Pearson correlation coefficients were obtained for age at the 95% confidence level as a continuous variable. To evaluate the significance of differences in the QLF parameters depending on the patients' subjective symptoms, the paired *t*-test was performed at the 95% confidence level.

3. Results

Sixty-six teeth in 66 patients complaining of pain in the relevant tooth were selected and included in the analysis. The average age of the patients was 54.79 years. Of the patients included, 36.36% ($N = 24$) had asymptomatic teeth with crack lines on the contralateral side, and these contralateral teeth were also included in the analysis; thus, a total of 90 teeth were included (Table 1).

Table 1
Distribution of the specimen.

			Total N	Symptomatic tooth N	Contralateral tooth N
Sex	Male		41	29	12
	Female		49	37	12
Tooth position	Maxilla	1st premolar	3	2	1
		2nd premolar	2	2	0
		1st molar	22	17	5
		2nd molar	27	20	7
	Mandible	1st premolar	0	0	0
		2nd premolar	0	0	0
		1st molar	12	9	3
		2nd molar	24	16	8
Spontaneous pain	+		7	7	0
	–		83	59	24
Percussion test	+		18	18	0
	–		70	48	24
Bite test	+		32	32	0
	–		56	34	24
Cold test	++		13	13	0
	+		68	46	24
	–		7	7	0

3.1. Efficacy for crack detection

All images were obtained as a pair of white light and fluorescent images. Thus the efficacy of the crack detection of QLF technology was analyzed by comparing the white light images as controls. As a result, In QLF images, most crack lines visible with the white light images were observable as a dark or red fluorescent line. Fig. 2 shows representative examples of QLF images showing relatively low, intermediate, and high parameters. A total of 89 teeth showed a higher |ΔF| value and 84 teeth showed a higher ΔR value than the cut-off, $\Delta F < -3\%$ and $\Delta R > 5\%$. Therefore, QLF technology showed efficacy for crack line detection with the high ΔR sensitivity of 0.96, that obtained by the QLF device and software used. In some cases, crack lines were observed on the QLF image, confirming the existence of additional crack lines that were not observed upon visual inspection (Fig. 3). In addition, an example of a QLF image of an asymptomatic contralateral tooth with crack lines was obtained, as represented in Fig. 4.

3.2. Relationship between clinical symptoms and QLF parameters

All groups satisfied normality as determined by the normality test. For all teeth ($N = 90$), Table 2 shows a correlation analysis between QLF image analysis parameters (|ΔF|, |ΔFmax|, ΔR, ΔRmax) and sex, tooth position, spontaneous pain, percussion test, bite test, cold test, and age. The independent two-sample *t*-test revealed that teeth with pain upon percussion had significantly higher |ΔF|, |ΔFmax|, and ΔR values ($p < 0.05$), and teeth with pain upon biting had significantly higher |ΔF| and |ΔFmax| values ($p < 0.05$). For investigating the correlation between the responses to the cold test and ΔF, ΔFmax, ΔR, and ΔRmax, the ANOVA test was performed. And a significant difference between the cold responses and the |ΔF|value was found($p < 0.05$). However, no significant difference was observed between the groups according to the Bonferroni correction as a post hoc analysis. Pearson correlation coefficient between |ΔFmax|, ΔR values, and age ($p < 0.05$) were positive and statistically significant ($p < 0.05$) and a similar tendency was observed in |ΔF| ($p = 0.062$) and ΔRmax ($p = 0.067$), although it was not statistically significant.

However, there was no statistically significant difference in the QLF parameters between the symptomatic tooth and the contralateral tooth in the same patient (Table 3, $n = 24$).

4. Discussion

When the QLF device irradiates teeth with blue light with a 405 nm wavelength, the demineralized teeth and the crack lines of the teeth interfere with light transmission and induce light scatter, resulting in darker fluorescence. Jun et al. [18] showed that the higher the fluorescence loss in the crack line within the enamel and the deeper the crack line, the higher the absolute value of the maximum fluorescence loss (|ΔFmax|).

Through the degree of red fluorescence emitted by porphyrin, it is possible to know whether bacteria are deposited and activated; thus, the degree of lesion progression can be predicted [19]. Hence, longer lasting crack lines in which the bacterial colonization progressed can be shown emit a stronger red fluorescence.

It has previously been reported that crack lines can be directly and easily observed using QLF in several case reports [15,16]. In this study, 86 of the 90 teeth demonstrated crack lines with a distinct red fluorescence from which the red fluorescence value (ΔR) was derived. Among the six teeth with a lower ΔR value than the cut-off, four teeth showed a craze line located in the superficial level of enamel, and two teeth showed unclear red fluorescence. Oh et al. [17] evaluated the ability of QLF technology to diagnose cracked teeth. When two QLF parameters, ΔFmax and ΔRmax, were used, they demonstrated good area under the curve levels (0.83 and 0.82, respectively) and optimal sensitivity (0.85 and 0.75, respectively). In the present study, red fluorescence was used as the parameter because the color red is visually more distinct and is therefore more useful for diagnosis than fluorescence loss, and the sensitivity was 0.93. The difference in the results between the two studies was probably due to the different criteria that Oh et al. applied to

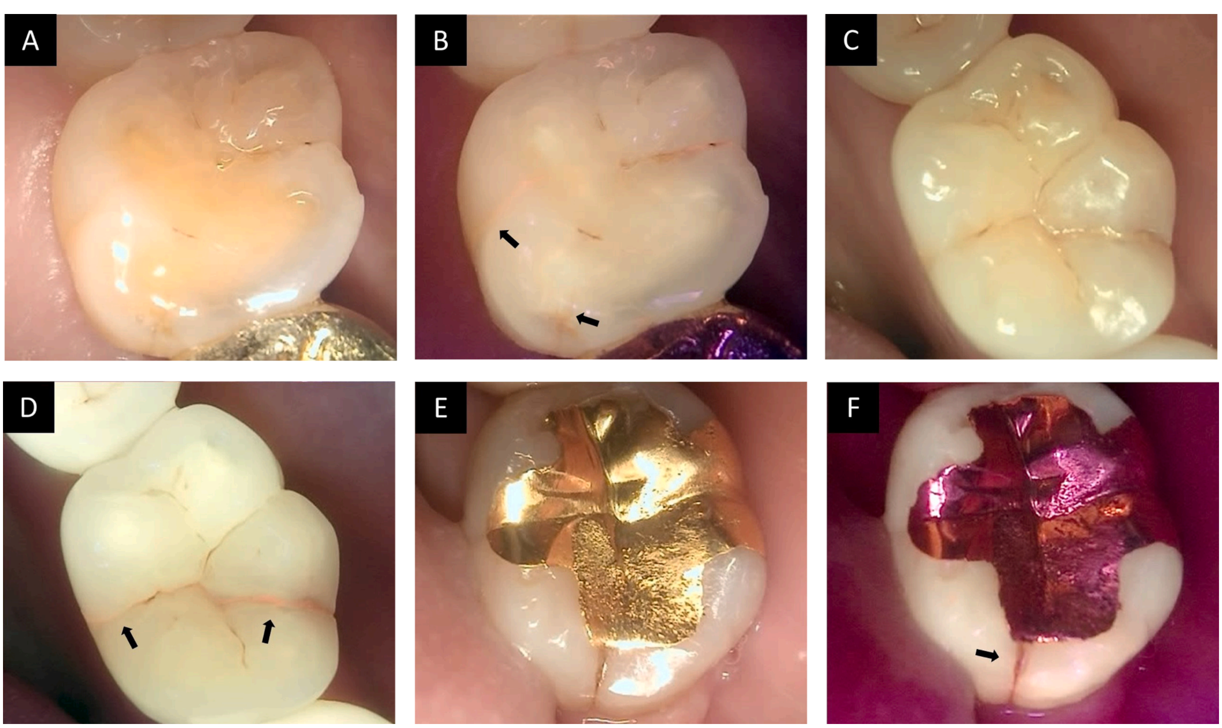

Fig. 2. Representative QLF images showing relatively low, intermediate, and high parameters. Arrows indicate crack lines, and QLF parameters of each tooth are as follows: (A), (B) ΔF= −7.5, ΔFmax= −15.1, ΔR= 8.7, ΔRmax= 14.8; (C), (D) ΔF= −10.8, ΔFmax= −29.6, ΔR= 12.7, ΔRmax= 35.7; (E), (F) ΔF= −16.7, ΔFmax= −43.7, ΔR= 18.2, ΔRmax= 46.0.

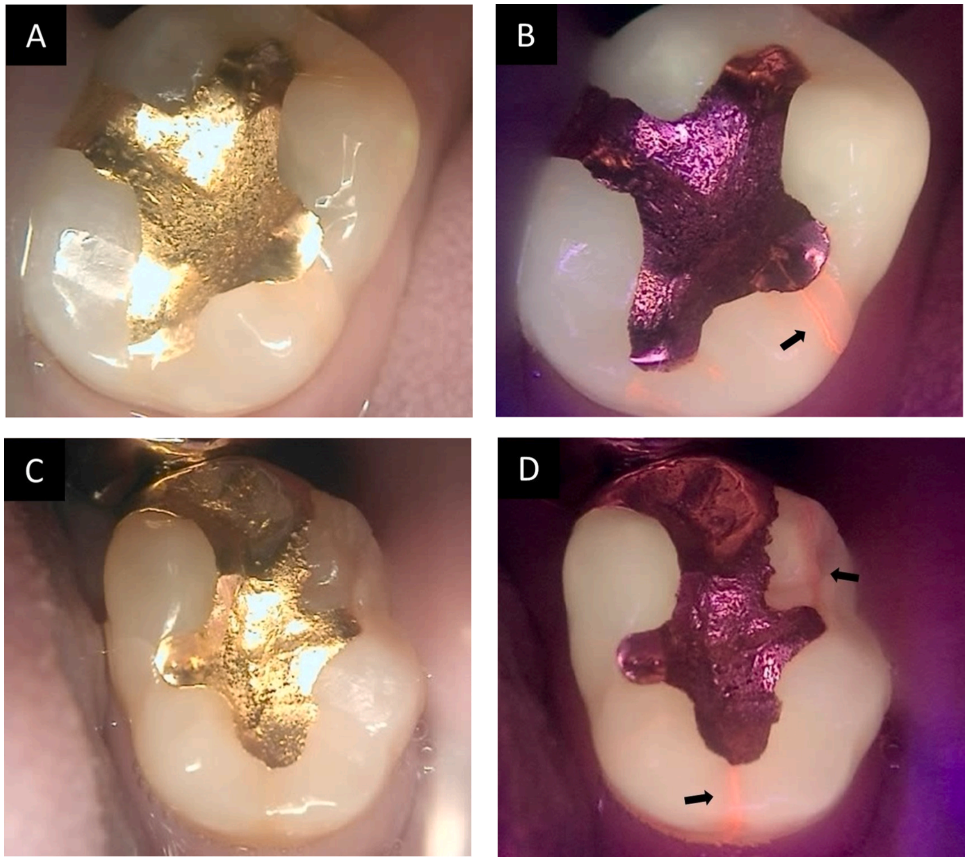

Fig. 3. Representative image of additional crack lines observed in QLF image. (A) Distinct crack line was observed on the mesial marginal ridge. (B) Additional crack lines were confirmed on buccal and distobuccal cusps of the tooth, indicated by arrows.

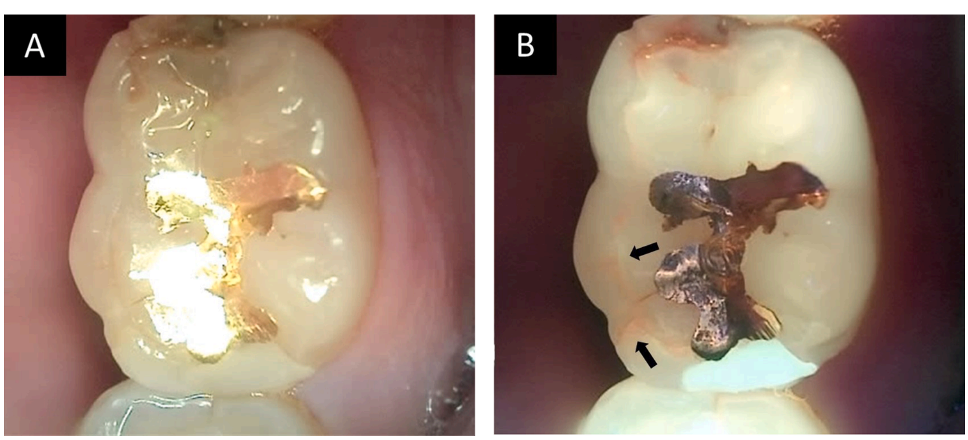

Fig. 4. Representative image of symptomatic cracked tooth in the area of chief complaint and asymptomatic contralateral tooth with crack line. (A) Left mandibular second molar that had pain on mastication, cold test, and bite test. (B) Crack line indicated by arrow was observed on lingual surface. (C) Asymptomatic contralateral tooth with crack line on distal and mesiobuccal cusps. (D) Arrows indicate the red fluorescence in the crack lines.

the crack lines observed on the QLF images and the difference in the cut-off setting of the software used. However, it can be concluded that both studies demonstrated the diagnostic ability of QLF. In addition, in several cases, crack lines that were not clearly observed with the naked eye were incidentally identified on the QLF image. Therefore, QLF can be useful for diagnosing cracked teeth by observing and detecting crack lines in a harmless manner, unlike radiation in a clinical environment.

In this study, patient age had statistical correlation with some QLF parameters. Therefore, the null hypothesis was partially rejected. There was a positive correlation between the patient's age and the $|\Delta F|$ and ΔR values. In other words, older patients demonstrated a higher average fluorescence loss and red fluorescence in their crack lines. Regardless of the symptoms, this can be interpreted to be caused by the progression of previously existing crack lines and the colonization of bacteria.

The pain caused by a crack can be explained for two reasons. The first is the change in fluid flow rate in the dentinal tubules due to pressure or temperature, known as the hydrodynamic theory [20]. The second is that the inflow of toxins produced by bacteria in the crack line and dentinal tubules can cause irritation of or inflammatory responses in the pulp tissue, inducing pain [14]. Therefore, it can be deduced that more

Table 2
Relationship between QLF image analysis parameters and variables.

		|ΔF| (%) Mean ± sd	p-value	|ΔFmax| (%) Mean ± sd	p-value	ΔR (%) Mean ± sd	p-value	ΔRmax (%) Mean ± sd	p-value
Sex	**Male**	7.854 ± 3.518	0.2195	16.868 ± 8.696	0.2254	9.432 ± 4.947	0.2834	18.166 ± 13.001	0.4268
	Female	8.757 ± 3.396		19.212 ± 9.372		10.451 ± 4.014		20.108 ± 10.068	
Tooth position	**1st premolar**	6.400 ± 0.361	0.7673	11.800 ± 1.833	0.6302	8.100 ± 2.007	0.8843	13.033 ± 4.860	0.8017
	2nd premolar	8.200 ± 0.000		19.150 ± 4.313		10.300 ± 3.536		19.450 ± 10.394	
	1st molar	8.224 ± 3.612		17.756 ± 8.672		9.850 ± 4.419		18.915 ± 10.728	
	2nd molar	8.547 ± 3.527		18.737 ± 9.707		10.176 ± 4.684		19.784 ± 12.348	
Spontaneous pain	+	9.457 ± 4.437	0.3794	20.386 ± 9.401	0.5003	10.871 ± 4.144	0.5881	19.986 ± 10.221	0.8559
	–	8.252 ± 3.385		17.955 ± 9.103		9.912 ± 4.508		19.159 ± 11.624	
Percussion	+	10.000 ± 3.193	0.0225	22.722 ± 7.326	0.0161	11.856 ± 2.965	0.0121	23.378 ± 9.298	0.0858
	–	7.932 ± 3.423		17.000 ± 9.177		9.519 ± 4.669		18.185 ± 11.780	
Bite	+	9.725 ± 4.032	0.0107	21.278 ± 9.720	0.0143	10.834 ± 5.237	0.1826	21.094 ± 13.774	0.2988
	–	7.584 ± 2.867		16.416 ± 8.322		9.519 ± 3.950		18.191 ± 9.961	
Cold	++	9.700 ± 3.913	0.0354	21.123 ± 10.382	0.1092	11.762 ± 4.201	0.0848	22.662 ± 11.777	0.1379
	+	7.817 ± 3.078		16.959 ± 8.531		9.369 ± 4.122		17.846 ± 10.009	
	–	10.817 ± 5.074		24.183 ± 10.503		13.217 ± 7.215		27.750 ± 21.962	
Age	**r (95% CI)**	0.197 (−0.011–0.388)	0.0622	0.235 (0.028–0.421)	0.0252	0.219 (0.011–0.407)	0.0378	0.193 (−0.015–0.384)	0.0677

Table 3
Difference of QLF parameters between symptomatic tooth and contralateral tooth.

	Symptomatic tooth Mean ± sd	Contralateral tooth Mean ± sd	Diff. mean ± sd	p-value
|ΔF| (%)	8.425 ± 3.607	7.571 ± 2.795	−0.854 ± 4.056	0.3129
|ΔFmax| (%)	18.067 ± 8.510	16.221 ± 7.770	−1.846 ± 10.724	0.4078
ΔR (%)	9.825 ± 4.795	9.271 ± 4.062	−0.554 ± 4.894	0.5844
ΔRmax (%)	18.200 ± 13.173	17.092 ± 9.092	−1.108 ± 13.622	0.6939

painful signs and symptoms can present as the crack progresses over time, beyond DEJ and across the dentin toward the pulp. Accordingly, it may be inferred that there may be a correlation between fluorescence loss known to be associated with the depth of the crack, the red fluorescence that increases with bacterial colonization and activity over time, and the signs and symptoms of the cracked tooth.

The statistical analysis gave some conflicting results in this study. When we analyzed 90 teeth, teeth showing pain on the percussion test demonstrated significantly higher |ΔF|, |ΔFmax|, and ΔR values. However, statistical significance was not confirmed when we analyzed 24 pairs of teeth even though the higher average of the QLF parameter was shown in the symptomatic teeth than in the asymptomatic contralateral teeth. Clinically, the findings from the same patient and tooth type would be important when QLF images are used to diagnose cracked teeth. In addition, we consider a limitation that subgroup analysis with factors that can affect symptoms and crack propagation, such as the age, gender, type of restorations, occlusion, and parafunction, was not done due to the lack of the number of samples because only 24 patients had a detectable crack line on the contralateral side. Although it is predicted that there would be a correlation between symptoms and the depth of the crack line, it is difficult to say that the progression of the crack and symptoms are necessarily proportional, as mentioned above. Therefore, even if a crack is present in the contralateral side of the same patient, the symptoms can be present or absent.

Pain on percussion, also known as mechanical allodynia, is generally understood to occur when the periodontal ligament is inflamed. However, irreversible pulpitis may also cause pain on percussion by activating mechanoreceptive neurons in pulp or periapical tissues, or central sensitization, which is caused by bacteria and their by-products or inflammatory mediators [21]. According to the prospective cohort study by Lee et al. [22], pain upon percussion was a significant predictor of pulp survival in cracked teeth with reversible pulpitis. Therefore, pain on percussion in cracked teeth can be considered mechanical allodynia that results from peripheral stimulation or central sensitization due to the influx of bacteria or toxins into the crack line, leading to pulpitis. Thus, it can be inferred that there is a correlation between pain upon percussion and the depth of the crack line and QLF parameters, which are known to be associated with the depth of the crack line and bacterial accumulation.

Teeth with pain upon biting showed significantly higher |ΔF| and |ΔFmax| values, but the ΔR and ΔRmax values did not show statistically significant differences. As mentioned above, pain during mastication is caused by the change in fluid movement in the dentinal tubules due to pressure. Therefore, it can be inferred that the deeper the crack line, which is related to ΔF and ΔFmax values, the greater the pain caused by fluid movement. On the other hand, a high ΔR is suggestive of a high amount of bacterial deposition and activity, no depth progression of the crack, which may mean that the crack line existed for a long time. According to Hilton et al. [23], only 45% of cracked teeth showed symptoms, and after 1 year of follow-up, the number of patients with a decrease in symptoms was twice as high as the number of patients who experienced an increase in symptoms, at 23% and 10%, respectively. In particular, a decrease in symptoms was more common when clinician did recommend treatment from baseline. Therefore, symptoms of a cracked tooth do not simply increase over time; sometimes, cracked teeth exhibit no symptoms, and symptoms can decrease or increase over time. Several mechanisms can explain a reduction in symptoms, such as tertiary dentin formation [24], relief of stress by the creation of additional crack lines [23], and the sealing effect of staining [25].

The limitation of this study is that QLF images cannot always be taken at a constant distance and angle in a clinical environment. In particular, errors may occur in the analysis depending on the angle of light. The most accurate analysis result can be obtained if the crack line is irradiated horizontally. However, clinically, this is difficult to achieve, and if the angle changes, the area of fluorescence loss is broader and the maximum fluorescence loss can be reduced. It is difficult to confirm the depth of the crack line nondestructively because only micro-CT and histological evaluation after extraction can be used. A limited number of previous studies investigated a correlation between QLF parameters and actual crack line depth, and no studies conducted a correlation between QLF parameters and symptoms of cracked teeth. Therefore, this study partially proved the relationship between QLF parameters and symptoms that can indirectly indicate the degree of crack progression.

In conclusion, visualization using QLF technology seems beneficial as a supplementary diagnostic technique. There was a positive correlation between the patient's age and the |ΔFmax| and ΔR values. There was no statistical difference when QLF parameters were compared between a symptomatic cracked tooth and an asymptomatic contralateral tooth in the same patient. Within the limitations of this study, there is a possibility of quantifying the progression of the crack line by QLF technology, but further research is needed to determine a method to analyze the depth of the crack line directly.

CRediT authorship contribution statement

Jong-In Lee: Conceptualization, Methodology, Software, Formal analysis, Investigation, Writing – original draft, Visualization. **Mi-Jeong Jeon:** Writing – review & editing, Supervision. **Elbert de Josselin de Jong:** Software. **Hoi-In Jung:** Conceptualization, Validation, Data curation, Writing – review & editing, Supervision. **Il Young Jung:** Validation, Writing – review & editing, Supervision. **Jeong-Won Park:** Validation, Supervision. **Su-Jung Shin:** Conceptualization, Validation, Resources, Data curation, Writing – review & editing, Supervision, Project administration, Funding acquisition.

Declaration of Competing Interest

The authors deny any conflicts of interest. However, we would like to clarify the fact that Inspektor Research Systems BV and AIOBIO provided the salary for the co-author, Elbert de Josselin de Jong (EdJdJ). AIOBIO is the manufacturer of the Qraypen-C used in this paper. EdJdJ did not have any additional role in the study design, data collection, analysis, decision to publish, or preparation of the manuscript. EdJdJ's involvement in this research was under the auspices of his status as adjunct professor at Yonsei University College of Dentistry supported by the BK21 FOUR Project. The specific role of EdJdJ was to provide his expertise regarding the fluorescence technology. EdJdJ holds several patents with respect to QLF technology.

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