## Original

# Efficacy of Three Caries-staining Agents

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Abstract: To avoid excessive excavation of carious dentin, we developed a caries-detecting agent composed of polypropylene glycol to stain caries-affected dentin. The purpose of this study was to evaluate the new agent by examining the characteristics of the dentin surface after removal of carious dentin. Eight extracted human teeth with dentin caries were used. The teeth were longitudinally sectioned through the center of the caries and micro Vickers hardness (MVH) was measured from the pulpal wall to the caries cavity every 200  $\mu$ m. Carious dentin was then removed using a steel bur following Caries Check (Nishika, Yamaguchi, Japan) staining. After removal of the stained dentin, the MVH of the dentin was determined by observing the indentation on the dentin; the DIAGNOdent value (D-value) was measured using DIAGNOdent (Kavo, Biberach, Germany). Then a new caries-staining agent (Discover RED, Phoenix Dental, Inc., Fenton, MI, USA) was applied in the cavity, the stained dentin was removed, and the MVH and D-value were measured. Finally, such procedures were repeated using Caries Detector (Kuraray, Tokyo, Japan). After the final measurements, the microstructure of the dentin cavity wall was observed by scanning electron microscopy. The differences in the MVH and D-value between the Caries Check and the other two methods were significant. Caries Check may be the most useful modality for the more precise excavation of carious dentin and preservation of sclerotic dentin.

Key words: carious dentin, caries detector, polypropylene glycol.

The removal of carious dentin is the most important clinical procedure for minimal intervention. However, dentists excavate carious dentin based on their tactile sensitivity because no consistently reliable standard for the removal of carious dentin has been established. The detailed mechanical and histological differences between the outer and inner layers of carious dentin have been reported in many papers.<sup>1~4)</sup> It was concluded that only the outer layer should be removed because the inner layer was not infected and may be recalcified by dental treatment. In addition, propylene glycol containing red dye has been introduced clinically as a caries-staining agent because it stains the outer layer selectively.<sup>5,6)</sup> In

some papers, however, it was claimed that the dentin was possibly excessively excavated when the dentin stained by this caries-staining agent was removed completely.<sup>7~9)</sup> To prevent excessive excavation into the inner layer of the carious dentin, a new caries-staining agent composed of polypropylene glycol has been introduced.<sup>10,11)</sup> In this study, the efficacy of the three commercial caries-staining agents was investigated by measuring the micro Vickers hardness (MVH) of the dentin and the DIAGNOdent value, which represents caries-associated fluorescence.

#### **Materials and Methods**

Three commercially available caries-staining agents

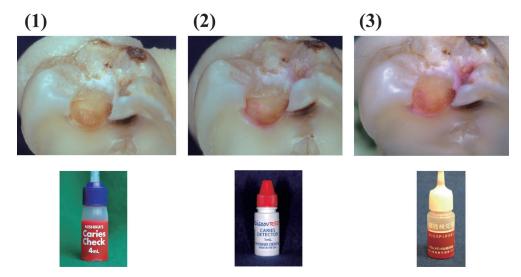


Fig. 1 The same cavity stained by three solutions. (1) The cavity after removal of carious dentin stained by Caries Check. (2) The cavity after removal of carious dentin stained by Discover RED. (3) The cavity after removal of carious dentin stained by Caries Detector.

Table 1	The commercial	caries	detectors	tested.
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Product	Manufacturer
Caries Check	Phoenix Dental, Inc, Michigan USA
DiscovRED	Nishika, Yamaguchi, Japan
Caries Detector	Kuraray Co., Ltd., Tokyo, Japan

listed in Table 1 were employed. We decided to stain and remove carious dentin in the order of Caries Check, Discover RED, and Caries Detector, because staining depth in the same cavity increased in the order of Caries Check, Discover RED, and Caries Detector (Fig. 1). The sequence methodology is shown in Fig. 2. Eight extracted human teeth with carious dentin were sectioned through the center of the caries along the long axis and the section surface was polished on a wet silicon carbide paper with a grit number of 1500. The MVH of the dentin was measured from the pulp chamber to the caries every 200 um (MVK Hardness Tester, Akashi, Kanagawa, Japan). The carious dentin was then removed according to the caries-staining agent (Caries Check, Nishika, Yamaguchi, Japan) using a steel round bur mounted on a low-speed dental cutting machine with water as coolant. Staining and excavating the stained dentin was repeated until the dentin cavity wall was no longer stained. After the removal of stained dentin was finished, the hardness of the dentin was determined by observing the indentation

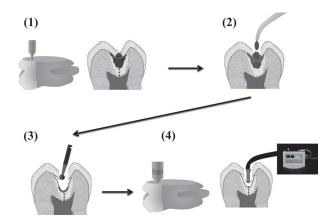


Fig. 2 The sequence methodology. (1) The micro Vickers hardness of the dentin was measured from the pulp chamber to the caries every 200  $\mu$ m. (2) A drop of each of the three solutions was added. (3) The stained dentin was removed. (4) The micro Vickers hardness of the dentin was determined by observing the indentation on the dentin and the D-value was measured by using the DIAGNOdent caries detection system.

on the dentin and the D-value was measured by using the LASER fluorescence method of the DIAGNOdent caries detection system (Kavo, Biberach, Germany). Then another caries-staining agent (Discover RED, Phoenix Dental, Inc., Fenton, MI, USA) was applied in the cavity according to the manufacturer's instructions and the stained dentin was removed. After the Discover RED staining and cutting was finished, the MVH was determined and the D-value was measured. Finally, these procedures were repeated using a third commercial caries-staining agent (Caries Detector, Kuraray Dental, Tokyo, Japan). After finishing the measurements, for the microstructural observation of the dentin cavity wall by scanning electron microscopy, the smear layer on the cavity wall was removed using 0.5 mol/L neutralized EDTA (pH 7.4) for 60 s followed by rinsing and drying. Next, the teeth were dehydrated in graduated alcohol solutions and vacuum ion spattered with palladium and platinum.

### Results

The results of the MVH and D-value measurements are listed in Tables 2 and 3. After removing the dentin stained by Caries Check, the cavity wall was still stained

Table 2 Results of micro Vickers hardness measurements.

Product	Micro Vickers Hardness (MVH)		
Caries Check	34.7±5.5		
DiscovRED	42.3±5.5 <b>*</b>		
Caries Detector	42.3±3.4		

Values connected with a line is not significantly different. (Student's *t*-test, n=8)

by Discover RED in all eight of the specimens. The differences in both the MVH and D-value between the Caries Check and the Discover RED were statistically significant (p<0.05, Student's *t*-test). After the removal of the dentin stained by Discover RED, the dentin cavity wall was still stained by Caries Detector in six of the eight specimens. The difference between the Caries Check and Discover RED was insignificant in both the MVH and D-value. In the scanning electron microscopy observation, debris in the dentin tubules was not observed in the specimens of Caries Detector and Discover RED. The dentin tubules were filled with debris after removing the dentin that was stained by Caries Check (Fig. 3).

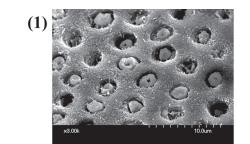
#### Discussion

Fusayama et al. defined the two layers distinguished in carious dentin.<sup>6)</sup> In addition, they suggested that the outer

Table 3 Results of DIAGNOdent value (D-value) measure-

ments.		
Product	D-value	
Caries Check	18.0±5.2	
DiscovRED	13.3±5.3	ale
Caries Detector	11.3±2.7	*

Values connected with a line is not significantly different. (Student's *t*-test, n=8)



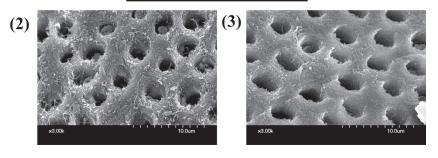


Fig. 3 Scanning electron microscopy observation of the cavity wall. (1) Carious dentin was removed based on Caries Check staining. The dentin tubules were filled with debris. (2) Carious dentin was removed based on Discover RED staining. A few dentin tubules were filled with debris. (3) Carious dentin was removed based on Caries Detector staining. The dentin tubules were completely open. layer should be removed selectively because the inner caries layer may be recalcified. Kuboki et al. reported that the outer layer of the caries layer was not re-calcified because the cross-linkage of the dental collagen was irreversibly destroyed.<sup>12)</sup> The clinical application of a caries-staining agent has an advantage because dentists are able to detect the outer layer of the caries dentin easily with the naked eye. The idea of a caries-staining agent and the initial product was introduced by Terashima et al.<sup>5)</sup> They demonstrated that the inner carious lesion was stained by propylene glycol colored with basic fuchsine. The detailed mechanism of the caries staining has not been explained clearly. Therefore, it is impossible to conclude that Caries Detector stained the caries-infected dentin exactly. As suggested by Yip et al., the dentin was stained because the colored alcohol penetrated the dentin that was decalcified and the density was consequently decreased.<sup>17)</sup> Therefore, it is possible to speculate that the dentin region stained by the caries-staining agent was determined by the penetration efficacy of the staining agent. The over staining by two caries-staining agents tested in this study was due to their penetrating effect into relatively high-density dentin.

Lucci suggested the criteria of the D-value and clinical requirement of the dentin. He proposed that dentin indicating a D-value lower than 13 should be observed without any clinical restorative treatment. In addition, the caries should be excavated when the cavity indicated a D-value over 20.14) The dentin cavity wall after the removal of dentin that was stained by two of the commercial caries-staining agents tested in this study was diagnosed as nearly sound dentin by its D-value measurement. It has been recommended that the cariesaffected dentin should not be excavated because the dentin tubules are filled with fine crystals of tricalcium phosphate that prevent caries invasion. Such dentin tubules filled with crystals interfere with the fluid flow through the tubules, resulting in the interruption of dentin sensitivity. In our previous report, it was revealed that dentin adhesive exhibited higher efficacy on the cariesaffected dentin rather than on the sound dentin.<sup> $15 \sim 17$ </sup>)

The dentin cavity wall of the Caries Check specimen was considered to be caries-affected dentin because the D-value was as high as 18.0±5.2 and the dentin tubules were filled with debris.

As demonstrated in this examination, it is possible to conclude that dentin might be excessively excavated when a dentist cuts the dentin according to the staining with two of the caries-staining agents tested in this study. The dentin cavity wall of the Caries Check specimen was considered to be caries-affected dentin. From the clinical point of view, two of the three commercial caries-staining agents tested require some improvement to limit the invasion into the dentin.

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