Original

Scanning electron microscopic study of dentinal pulpal walls in relation to age and tooth area

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Abstract: A study was conducted to observe the surface morphological changes of human dentinal pulpal walls in specific areas of the tooth at various ages. Thirty-two extracted human non-carious teeth with single root canals were used. The teeth were divided longitudinally in the bucco-lingual plane and prepared for scanning electron microscopy (SEM). The specimens were divided into two groups (younger and older) according to age. Four central sites of the dentinal pulpal wall, including coronal, cervical, mid-root and apical sections, were selected as specific locations. Under the conditions of this study, six basic types of SEM appearance at dentinal pulpal wall surfaces were identified on the basis of calcospherite shape and mineralization. The appearance of the calcospherites varied according to tooth age and location along the dentinal pulpal walls. (J. Oral Sci. 50, 199-203, 2008)

Keywords: aging; dentinal pulpal wall; scanning electron microscopy.

Introduction

It is well known that tooth morphology can alter secondary to environmental conditions within the oral cavity. Such morphologic changes may take place in the dental pulp cavity, which is surrounded by the dentin. Initially the pulpal wall is formed by primary dentin. After the tooth has erupted, various forms of stimuli such as attrition, abrasion, occlusion, chemical irritation, and restoration lead to continuous deposition of dentin on the pulpal walls. Therefore, the pulp cavity gradually becomes smaller through environmental effects and aging. There have been several studies of dentin formation and calcification in relation to these conditions (1-3). Different types of dentin tissues have been described: primary, secondary, and tertiary dentin (4). Primary dentin is the original tubular dentin largely formed prior to eruption of a tooth. Secondary dentin is the same circumpulpal dentin as primary dentin, but formed after completion of root formation. Tertiary dentin is found only in dentin that has been subjected to trauma or irritation. Thus, detailed knowledge about these structures is essential in order to understand the progressive changes in the dentin pulp cavity, and therefore the efficacy of intracanal medications and root canal enlargement methods in current use. However, there are still insufficient scientific data on the changes in histological and physiological characteristics of dentin in relation to age.

The purpose of this study was to observe the surface morphological changes in human dentinal pulpal walls in specific areas of the tooth at various ages using SEM.

Materials and Methods

A total of 32 extracted human teeth that had no caries or restorations from patients aged between 9 and 62 years

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were selected for this study. The reasons for extraction included orthodontic problems (15 teeth), impaction (1 tooth) and periodontal disease (16 teeth). The teeth were grooved longitudinally in the bucco-lingual plane with a high-speed turbine. The guiding grooves were as near as possible to the dental pulp cavity but avoided penetrating the pulpal walls. These teeth were further fractured longitudinally by hand. Remnants of pulp tissue were carefully removed with a pair of tweezers and the specimens were immersed in 3% sodium hypochlorite for 24 h. Each specimen was thoroughly washed in distilled water, ultrasonically cleaned, and dehydrated through a graded series of ethanol solutions. Samples for SEM were sputtercoated with gold in an argon atmosphere within a Polaron coating unit, followed by observation by SEM (JSM-T200 JEOL, Tokyo, Japan) at 5 to 15 kV. The specimens were marked at the cementum-enamel junction, cervical rootmid root, and mid root-apical root levels; then four sites on the dentinal pulpal wall, including coronal, cervical, midroot and apical sections, were selected as specific locations for SEM photomicrography (Fig. 1). Two standard magnifications ranging from ×35 to ×1,000 were generally utilized. Chi-squared analysis (P < 0.05) was performed to determine any statistically significant differences in type appearance between teeth from younger and older individuals.

Results

On the basis of typical SEM photomicrographs of the dentinal pulpal wall, six basic appearances were categorized as follows (Fig. 2).

Type 1. Partially fused calcospherites.

Many calcospherites locally coalesced in a dome-like shape were commonly observed at the buccal and lingual surfaces of dentinal pulpal walls, or forming a cogwheel shape at the proximal surfaces, especially in teeth from younger individuals.

Type 2. Almost or completely fused calcospherites.

Calcospherites fully coalesced in a flattened shape were mostly found at the proximal surfaces of dentinal pulpal walls in teeth from younger individuals. These seemed to be scarcely present or decreased in number in older teeth.

Type 3. Network-like mineralized appearance.

Many clusters consisting of mineralized fibers were crossed and interlaced together like a mesh. The clusters varied in size from fine to large and rolling. This appearance was mostly evident on the proximal surfaces of dentinal pulpal walls in the apical-root or mid-root location, especially in teeth from older individuals.

Type 4. Ridge-like mineralized appearance.

Many clusters consisting of mineralized fibers projected to the pulpal surface. In some cases, they resembled bamboo shoots and were mostly found on the proximal surfaces of dentinal pulpal walls in the apical-root location, especially in older teeth.

Type 5. Spherically mineralized appearance.

Many small spherical minerals were seen to coalesce. This feature was mostly found at the dentinal pulpal walls in the apical-root location, especially in older teeth.

Type 6. Structureless mineralized appearance.

Stone-like or crack-like appearances were mostly found at the dentinal pulpal walls in the apical location, especially in older teeth.

In teeth from younger individuals (Table 1, Fig. 3), the dentinal pulpal walls were fully occupied by fused calcospherites including types 1 and 2. Types 2 and 3 spread mostly on the proximal surfaces of the walls in the apical area and seemed gradually to replace type 1 (Table 3).

In teeth from older individuals (Table 2, Fig. 4), the appearance of dentinal pulpal walls was the same as in younger teeth, but the appearance on the proximal surfaces of the walls was replaced by types 3, 4, 5 and 6 in the mid-



Fig. 1 Selected specific points (*) for photomicrographs at each tooth location (×35). Dentinal pulpal walls of the teeth were divided into four sections including the coronal, cervical, mid-root and apical locations.

root and apical locations (P < 0.05). Especially, type 4 was represented in a high proportion of apical locations (Table 3).

Discussion

In this study, the dentinal pulpal wall of specimens from the crown to the root apex was fully exposed and observed in detail in various areas of the tooth using SEM. In order to show the mineralizing front, the organic elements were removed by immersion in sodium hypochlorite for 24 hours, ultrasonically cleaned, and washed with distilled water (5). These conditions allowed the appearance of the entire dentinal pulpal walls to be examined easily and clearly.



Fig. 2 Photomicrographs obtained by SEM (×1,000) – (a) partially fused calcospherites (type 1); (b) almost or completely fused calcospherites (type 2); (c) network-like mineralized appearance (type 3); (d) ridge-like mineralized appearance (type 4); (e) spherically mineralized appearance (type 5); (f) structureless mineralized appearance (type 6).

Case	Tooth	Aga		Tooth	location	
No.	1000	Age	Coronal	Cervical	Mid-root	Apical
1*	34	10	1+	1+,2	1,2+	1,2+
2*	44	10	1+,2	1,2+	1,2+	1,2+
3*	44	10	1 +	1+,2	1,2+	1,2+
4*	44	9	1 +	1+,2	1,2+	1,2+
5*	34	9	1+	1+	1+,2	1,2+
6*	34	10	1+	1+	1+,2	1,2+
7*	34	11	1+	1+,2	1,2+	1,2+
8*	44	11	1+	1,2+	1+,2	1,2+
9	44	12	1+,2	1+,2	1+,2	1,3+,4
10	44	12	1+,2	1+,2	1,2+	1,2+
11*	43	11	1+,2	1 +	1+,2	1+,2
12	18	19	1 +	1 +	1,2+	1,2,3+
13	44	13	1,2+	1+,2	1+,2	1,2,3+
14	35	14	1+	1+	1+,2	1,2,3+
15	45	14	1+	1+,2	1+,2	1,2,3+
16	35	13	1+	1+	12 + 3	1.2+

 Table 1 Distribution of various mineralized forms on the dentinal pulpal walls of teeth from younger individuals

Case No. *: Younger teeth with an open apex

1 to 6: The mineralized appearance from Type1 to Type 6

No.+: This type of mineralized appearance showing the most spread in the location.



Fig. 3 Occurrence of various mineralized forms on the dentinal pulpal walls of teeth from younger individuals.

Case	Teeth	Age	Looth location					
No.	1000		Coronal	Cervical	Mid-root	Apical		
1	42	46	1+,2	1+,2	1,2+,3	1,2,3,4+,6		
2	11	46	1+,2	1+,2	1,3+,4	1,4+,5,6		
3	11	52	1 +	1 +	1,3,6+	1,3,6+		
4	31	54	1 +	1 +	1,2+,5	5+,6		
5	42	54	1+,4	1+,3,4	1,3+,4	1,3,4+		
6	41	61	1+,2	1,2+	1+,3,4	1,3+,4,5,6		
7	44	61	1,3+	1+,3	1+,2,3	1,3,4+,6		
8	37	62	1+	1,2,3+	1,3,4,6+	1,2,3,4+,5,6		
9	34	43	1+	1+	1+	1,3,4+,6		
10	35	43	1+	1+	1+	1+,2,4		
11	44	43	1 +	1 +	1+5,6	1+,2,3,4,5,6		
12	35	48	1 +	1 +	1+	1,2,4+,6		
13	33	55	1 +	1 +	1+,2,3	1,3,4,6+		
14	45	52	1+,2	1+,6	1+	1,4,6+		
15	44	60	1+	1+	1+	1+,4,6		
16	45	55	1+	1+,2	1+,2	1,3+,4,6		

 Table 2 Distribution of various mineralized forms on the dentinal pulpal walls of teeth from older individuals

1 to 6: The mineralized appearance from Type1 to Type 6

No.+: This type of mineralized appearance showing the most spread in the location.



Fig. 4 Occurrence of various mineralized forms on the dentinal pulpal walls of teeth from older individuals.

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Location	Group	Type 1	Type 2	Type 3	Type 4	Type 5	Type 6	
Coronal	Younger	15	1	0	0	0	0	NS ر
	Older	15	0	1	0	0	0	7
Cervial	Younger	14	2	0	0	0	0	NS ר
	Older	14	1	1	0	0	0	
Mid-root	Younger	8	8	0	0	0	0	$_{\rm NS}$
	Older	10	2	2	0	0	2	
Apical	Younger	1	11	4	0	0	0	* r
	Older	3	0	2	7	1	3	L

NS: Not significant

*: (P < 0.05) Significant difference

Several dental anatomy textbooks state that dentin is calcified through the formation, enlargement and ultimate fusion of many centers of calcification (6-8). These centers are named calcospherites. The calcospherites continue to grow uniformly in all directions until contact is made with other calcospherites (9). The resulting inhibition of growth leads to modification of the initially spherical calcospherite shape. Whittaker and Kneale (10) have reported a detailed study of the mineralizing front at the dentin-predentin interface in human teeth. They described the relationship of the various mineralization patterns to the position in the tooth, which they found to be of a general rather than a reliably specific nature. Mineralization of dentin continues with tooth aging (11). As the tooth matures, the peritubular dentin becomes more mineralized, which in some instances can result in complete obliteration of the dentinal tubules (12).

In younger teeth, the shape of calcospherites was described as being almost or completely fused (types 1 and 2). In older teeth, the shape of calcospherites in the coronal part of the pulp cavity had a similar appearance to that seen in younger teeth, but nearer to the mid-root and apical-root areas the appearance of calcospherites was replaced by types 3, 4, 5 and 6, with a less regular surface and fewer tubules.

The different appearances of calcospherites in relation to location on the tooth and age were considered to be related to the number of odontoblasts forming the tubular dentin and to dimensional variation of the pulp cavity.

Shellenberg et al. (13) concluded that a certain number of odontoblasts facing the walls of the greater concavity (buccal/lingual) have to crowd themselves more closely than the same number of cells facing the walls of the lesser concavity (mesial/distal). This anatomy-dependent regional difference leads to morphological differences between the bucco-lingual surfaces and proximal surfaces.

From a clinical viewpoint, it is of interest to clarify the morphological changes that occur with aging and location (14). Our present data suggest that apical dentin contains a lower number of tubules in which microbes can colonize, and subsequently release enzymes and degradation products into the periapical tissue. Hence, if the root canal system is well obturated, there are relatively few, if any, sites for remaining bacteria to occupy.

Acknowledgments

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References

- 1. Philippas GG, Applebaum E (1966) Age factor in secondary dentin formation. J Dent Res 45, 778-789
- 2. Nitzan DW, Michaeli Y, Weinreb M, Azaz B (1986) The effect of aging on tooth morphology: a study on impacted teeth. Oral Surg Oral Med Oral Pathol 61, 54-60
- Murray PE, Stanley HR, Matthews JB, Sloan AJ, Smith AJ (2002) Age-related odontometric changes of human teeth. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 93, 474-482
- Kuttler Y (1959) Classification of dentine into primary, secondary, and tertiary. Oral Surg Oral Med Oral Pathol 12, 996-1001
- 5. Tidmarsh BG (1980) Micromorphology of pulp chambers in human molar teeth. Int Endod J 13, 69-75
- 6. Bhaskar SN (1986) Orban's oral histology and embryology. 10th ed, CV Mosby, St Louis, 101-134
- 7. Provenza DV, Seibel W (1986) Oral histology: inheritance and development. 2nd ed, Lea & Febiger, Philadelphia, 264-290
- 8. Smith AJ (2002) Dentin formation and repair. In

Seltzer and Bender's dental pulp, Hargreaves KM, Goodis HE eds, Quintessence, Chicago, 41-62

- 9. Shellis RP (1983) Structural organization of calcospherites in normal and rachitic human dentine. Arch Oral Biol 28, 85-95
- Whittaker DK, Kneale MJ (1979) The dentinepredentine interface in human teeth. A scanning electron microscope study. Br Dent J 146, 43-46
- Vasiliadis L, Darling AI, Levers BGH (1983) The histology of sclerotic human root dentine. Arch Oral Biol 28, 693-700
- Tronstad L (1973) Ultrastructual observations on human coronal dentin. Scand J Dent Res 81, 101-111
- Schellenberg U, Krey G, Bosshardt D, Nair PNR (1992) Numerical density of dentinal tubules at the pulpal wall of human permanent premolars and third molars. J Endod 18, 104-109
- Thomas GJ, Whittaker DK, Embery G (1994) A comparative study of translucent apical dentine in vital and non-vital human teeth. Arch Oral Biol 39, 29-34