Histologic characteristics of the gingiva associated with the primary and permanent teeth of children

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Abstract

The severity of the gingival inflammatory response to dental plaque increases with age, and it has been suggested that this phenomenon may be related to histological characteristics of the gingiva. The objective of this study was to compare the histological characteristics of the gingival tissues of primary teeth with that of permanent teeth in children. Prior to extraction, children were subjected to a period of thorough oral hygiene. Histological sections prepared from gingival biopsies were examined using the light microscope. One biopsy from each of seven primary and seven permanent teeth of 14 children, whose mean ages were 11.0 ±0.9 and 12.9 ±0.9 years respectively, was obtained. All sections exhibited clear signs of inflammation. Apical migration of the junctional epithelium onto the root surface was associated only with the primary teeth. Compared with the permanent teeth, the primary teeth were associated with a thicker junctional epithelium ($P < 0.05$), higher numbers of leukocytes in the connective tissue adjacent to the apical end of the junctional epithelium ($P < 0.05$), and a higher density of collagen fibers in the suboral epithelial connective tissue ($P < 0.01$). No significant differences were noted in the width of the free gingiva, thickness of the oral epithelium, or its keratinized layer. In conclusion, this study indicates significant differences in the microanatomy of the gingival tissues between primary and permanent teeth in children. (Pediatr Dent 16:206–10, 1994)

Introduction

Clinical and histological studies have indicated that the severity of the gingival inflammatory response to dental plaque increases with age.[1-3] This phenomenon may be related to several factors including the maturity of the bacterial plaque,[4-6] the innate and learned immune response,[7-11] and the histomorphology of the tissues.[12-16]

Differences reported in the histomorphology of the gingivae associated with the human primary and the permanent dentitions include: a thinner stratum corneum associated with the gingiva of the permanent dentition, greater vascularity of the gingival connective tissue in the primary dentition, and a less organized pattern of orientation of the collagen fibers of the gingival connective tissue.[12-15] However, in these studies, the degree of gingival inflammation was poorly defined, and no comparisons between the gingiva of the primary and permanent dentitions in children were made. Matsson and Attström[16] compared the gingival tissues adjacent to the deciduous teeth in juvenile dogs with that of the permanent teeth of adult dogs. The teeth had been subjected to meticulous cleaning in order to achieve clinically healthy gingiva. The gingivae of the juvenile dogs displayed a thicker stratum corneum and junctional epithelium than the adult dogs. No differences were found in the density of collagen fibers or in the degree of vascularity of the gingivae. It is reasonable to assume that thickness of the stratum corneum and the junctional epithelium could affect their permeability and thus, provide a partial explanation for the difference in susceptibility of the juvenile and adult dentitions to plaque-induced inflammation. Consequently, the objective of this study was to compare the histological characteristics of the gingival tissues associated with the human primary teeth with those associated with the permanent teeth.

Methods and materials

Children referred to the department of oral and maxillofacial surgery for extraction prior to orthodontic treatment were included in this study. All the children were healthy and were not on any medication. None of the primary teeth used in the study were close to shedding, and all the permanent teeth used had reached occlusion. After confirming a patient's suitability for inclusion in the study, informed consent was obtained from the child and parent. In order to obtain clinically healthy gingivae, all the children were subjected to a thorough prophylaxis using a rubber cup and prophylaxis paste 14 and 7 days prior to extraction. In addition, the children were instructed on brushing techniques and requested to brush thoroughly at least twice a day.

Only one tooth from each patient was obtained for study. The sample consisted of seven primary and seven permanent teeth. Immediately prior to extraction, the gingival and plaque indices (GI & PII) were recorded.[17] In order to retain the relationship between the marginal gingiva and the tooth, a reverse bevel incision was made approximately 1–2 mm apical to the free gingival margin onto the underlying bone or the crown of the underlying permanent successor. The teeth with their gingivae were removed in one piece and placed in 4%

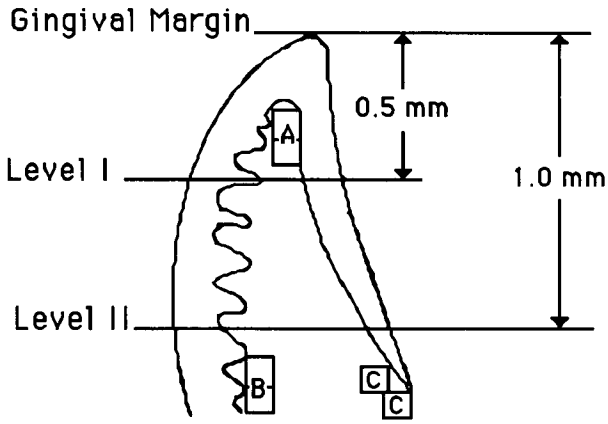

Fig 1. Diagramatic representation of the microscopic field. A = subcrevicular connective tissue area, B = suboral ephitelial connective tissue area, C = connective tissue area adjacent and apical to the apical end of the junctional epithelium.

neutral formalin. After a period of 1–3 weeks, the teeth were demineralized in a 10% solution of EDTA, pH 7.4, and embedded in water soluble resin (Historesin®, LKB, Bromma, Sweden). Sections, 1–2 μm, were cut and stained with hematoxylin and eosin.

From each tooth, the best midbuccal section was selected for light microscopy and the following measurements were made by one of the authors (LM) at two defined levels (Fig 1):

1. Thickness of junctional epithelium at levels I and II
2. Thickness of oral epithelium at levels I and II (the thickness of the oral epithelium was assessed at and between the rete-pegs nearest to levels I and II)
3. Width of gingiva at level II, measured from the junctional epithelium-enamel interface to the external surface of the oral epithelium
4. Thickness of stratum corneum at levels I and II.

In addition, the total number of cells and the number of leukocytes (neutrophilic granulocytes, macrophages, lymphocytes, and plasma cells) in a 0.02-mm^2 area were counted at three different zones (Fig 1): 1) The subcrevicular connective tissue (SCCT), above level I; 2)The suboral epithelial connective tissue (SECT), below level II; 3) The connective tissue adjacent and apical to the apical end of the junctional epithelium (JECT).

In these same areas, the density of the collagen bundles and the blood vessels were measured using an ocular grid. The densities were expressed as the percentage of grid intersections superimposed on the collagen bundles or the blood vessels.[16]

Measurements of the width of the free gingiva, the junctional epithelium, and the oral epithelium were made at a magnification of 200x (objective 20x, ocular 10x). Measurement of the width of the keratinized layer of the oral epithelium and assessment of the cell numbers and the density of the collagen bundles and the blood vessels were made at 1000x magnification (objective 100x, ocular 10x). At the latter magnification the grid covered an area of 0.01 mm^2 and comprised 100 squares and 121 cross-points. Each unit of analysis included two adjacent 0.1x0.1-mm squares (Fig 1).

Statistical comparisons were made using the Mann-Whitney nonparametric test. Differences at the 5% level of probability were considered statistically significant.

Results

Clinical findings

The mean age of the children at the time of extractions was 11.0 ± 0.9 years for those who contributed the primary teeth and 12.9 ± 0.9 years for those who contributed the permanent teeth. The group of primary teeth consisted of three maxillary canines, one maxillary molar and three mandibular molars. The group of permanent teeth included four maxillary and three mandibular premolars.

Immediately prior to extraction, zero PII were recorded on all teeth except for one primary and one permanent tooth, which had a PII score of 1. However, five primary and four permanent teeth had clinical signs of gingivitis (GI = 1). The remaining GI scores were zero.

Histologic findings

All sections exhibited clear signs of inflammation within the SCCT. The inflammatory cell infiltrate was usually clearly defined and often adjacent to areas of dense collagen bundles. Beneath the SECT, only solitary inflammatory cells were seen. No differences in the size or density of the inflammatory cell infiltrate in the connective tissue were noted between the two groups of teeth.

Four of six samples from the primary teeth had an unbroken tooth-epithelium interface. Apical migration

Table 1. Width of the junctional epithelium in mm

Type of Teeth	*Level I*		*Level II*	
	Mean	*SD*	*Mean*	*SD*
Primary	0.32	0.14	0.15	0.06
Permanent	0.18	0.06	0.14	0.15
P•	< 0.05		NS	

• Mann-Whitney.

Table 2. Thickness of the oral epithelium in mm at (1) and between (2) the rete pegs

Type of Teeth	Level I				Level II			
	1		2		1		2	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Primary	0.34	0.07	0.20	0.12	0.36	0.06	0.16	0.06
Permanent	0.32	0.09	0.21	0.12	0.38	0.11	0.19	0.07
P*	NS		NS		NS		NS	

* Mann-Whitney.

Table 3. Thickness of the keratinized layer of the oral epithelium in mm

Type of Teeth	Level I		Level II	
	Mean	SD	Mean	SD
Primary	0.027	0.033	0.017	0.006
Permanent	0.020	0.009	0.019	0.009
P*	NS		NS	

* Mann-Whitney.

of the junctional epithelium onto the root surface was noted in all four samples. The distance from the cementoenamel junction to the most apical part of the epithelium varied from 0.2 to 1.2 mm. Apical migration of the junctional epithelium was not seen in the permanent teeth.

In most cases, the coronal 0.5 mm of the junctional epithelium showed an irregular interface with the connective tissue, often with rete peg-like projections. This was noted in both the primary and the permanent teeth.

In areas with an inflammatory cell infiltrate the collagen was fragmented and separated by the cells and exudate. However, in the connective tissue on the oral side, the collagen fiber bundles appeared dense, often extending into the connective tissue papillae of the oral epithelium. The appearance of the collagen was similar in both groups of teeth.

The average width of the free gingiva was greater in the primary teeth (1.48 ± 0.29 mm) than in the permanent teeth (1.19 ± 0.21 mm) however, the difference was not statistically significant. At level I, the width of the junctional epithelium was significantly larger in the primary teeth than in the permanent teeth, whereas at level II no such difference was found (Table 1). No significant differences were found in the thickness of the oral epithelium (Table 2) or in the keratinized layer of the oral epithelium (Table 3).

The total number of cells in the SCCT was significantly higher in the permanent tooth group (Table 4).

Table 4. Total number of cells and number of leukocytes at the subcrevicular connective tissue (SCCT), the junctional epithelium connective tissue (JECT), and at the subepithelial connective tissue (SECT)

	SCCT				JECT				SECT			
	Total		Leukocytes		Total		Leukocytes		Total		Leukocytes	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Primary	60.9	12.1	22.0	8.7	51.9	12.3	12.8	4.6	25.4	8.9	5.9	2.3
Permanent	77.8	14.4	21.9	11.3	43.0	9.8	7.7	2.7	31.7	8.8	8.6	3.0
P*	< 0.05		NS		NS		< 0.05		NS		NS	

* Mann-Whitney.

Table 5. Density of collagen fibers and blood vessels* at the subcrevicular connective tissue (SCCT), the junctional epithelium connective tissue (JECT), and at the subepithelial connective tissue (SECT)

	SCCT				JECT				SECT			
	Collagen		Vessels		Collagen		Vessels		Collagen		Vessels	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Primary	22.7	11.9	3.5	0.9	36.0	13.0	1.8	1.3	58.3	5.9	2.1	1.9
Permanent	20.6	9.4	4.2	2.2	36.8	13.7	2.8	3.5	47.7	6.4	1.9	1.1
P†	NS		NS		NS		NS		< 0.01		NS	

* Expressed as the percentage of grid intersections (N = 121) superimposing collagen bundles or blood vessels.

† Mann-Whitney.

No difference in the number of leukocytes was noted in this area. In the JECT the number of cells classified as leukocytes was significantly higher in the group of primary teeth. In the SECT no statistically significant difference in the number of cells was found.

No statistically significant differences in the density of collagen and blood vessels were noted between the two groups, except in the SECT where a significantly greater density of collagen bundles was found in the primary teeth (Table 5).

Discussion

The objective of the pre-extraction phase of plaque control was to achieve clinical gingival health. However, the measures taken in this study did not achieve this goal as both minimal amounts of plaque and mild inflammation were present at the time of the extraction. It has been shown that during gingival inflammation, the density of collagen decreases and the relative proportion of blood vessels in the tissues increases.[16,18] However, since the degree of clinical inflammation was minimal and the difference between the permanent and primary teeth was not significant, it was felt that comparisons between the two were still relevant.

When comparing the gingival status between primary and permanent teeth in children, pubertal status should be taken into consideration.[19] In this study, the pubertal status of the children was not examined. However extractions were done at the late mixed or early permanent dentition periods and therefore the difference between the ages in which both types of teeth were extracted was kept to a minimum. Nevertheless, one cannot eliminate the possibility that the differences seen may be related to hormonal changes at puberty.

In the present study, all primary teeth were extracted for orthodontic reasons — well before their expected exfoliation time — and therefore, none of the teeth had epithelium encroaching onto the resorbing root surface. In addition, none of the permanent teeth were extracted before they reached occlusion and therefore had a well-developed gingiva.

The conclusions than can be drawn from this study are somewhat limited since only one section per tooth was utilized, and since the statistically significant differences between the primary and permanent teeth were relatively small.

An inflammatory cell infiltrate was evident in all the samples, including those with no clinical signs of gingivitis. This inflammation was restricted to the connective tissue immediately adjacent to the junctional epithelium and was not present beneath the keratinized oral epithelium. This finding is consistent with previous findings in children and adolescents with primary and permanent dentitions and indicates that clinically healthy gingiva usually contains an inflammatory cell infiltrate adjacent to the junctional epithelium.[20–22]

The apical migration of the junctional epithelium of the primary teeth in this study is consistent with our previous findings.[20,21,23] It has been suggested that this phenomenon is related to a combination of several factors including passive eruption, the shedding process of the primary dentition and a response of the epithelium to inflammation.[21,24]

Various reports in the literature have suggested that there are differences in the thickness of the epithelium of the gingiva between the primary and permanent dentitions.[12,16] Earlier studies indicate that the free gingiva of the primary teeth is thicker than that of the permanent teeth.[20,21] A similar, but not statistically significant difference, was noted in the present study in which gingival inflammation was minimal. The thickness of the free gingiva may play a role in the presentation of the clinical signs of inflammation. The thicker the free gingiva, as noted in the primary dentition, the more it would mask the early histological signs of inflammation occurring adjacent to the junctional epithelium from clinical observation.

Our findings in children support the findings of Matsson & Attström in dogs,[16] who reported a thicker junctional epithelium in juvenile dogs with primary teeth compared with the junctional epithelium of the permanent teeth. This difference was probably not due to an inflammatory-induced epithelial proliferation since the intensity of the inflammatory reaction in the connective tissue in this area was similar in the two groups. The thickness of the junctional epithelium also may influence the development of gingival inflammation by affecting the permeability of the epithelial structures to bacterial toxins. The thicker junctional epithelium of the primary dentition would be less permeable and more resistant to the onset of inflammation.

We were unable to confirm the findings of Zappler,[12] who reported that the gingiva of primary teeth display a thinner epithelial layer and a less dense hornified layer than that of the permanent teeth. Nor we were able to confirm the reports of a greater vascularity and a less well-differentiated pattern of collagen fiber bundles in the gingivae of the primary teeth.[12–14] In fact, the density of collagen fibers in this study was higher in the gingivae of the primary teeth. It should be noted however, that a preliminary oral hygiene phase as carried out in this study, was not performed in previous reports and thus may account for these differences.

The presence of a significantly higher numbers of leukocytes adjacent to the apical end of the junctional epithelium of the primary dentition supports the hypothesis that the migration of the junctional epithelium in the primary dentition onto the resorbing root surface is an integral part of the shedding process and may be associated with the inflammatory cell infiltrate.[22]

Conclusions

This study confirms previous findings of a thicker junctional epithelium in the primary teeth than in the

permanent teeth and the presence of apical migration of the junctional epithelium onto the root surface.

The primary teeth were associated with a thicker junctional epithelium, higher numbers of leukocytes in the connective tissue adjacent to the apical end of the junctional epithelium, and a higher density of collagen fibers in the suboral epithelial connective tissue.

The earlier findings of a thinner, less keratinized oral epithelium, and of connective tissue with greater and less dense collagen fiber bundles could not be supported.

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