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Patho-morphological study of the supplemental groove

AWAZAWA, Y.*, HAYASHI, K.**, KIBA, H.**, AWAZAWA, I.***, TOBARI, H.**

*Nihon University Research Institute of Sciences, 4-8-24, Kudan-minami, Chiyoda-Ku, Tokyo (Japan).
Present address: Nihon University School of Dentistry at Matsudo, Matsudo (Japan).
**Department of Pathology, Nihon University School of Dentistry at Matsudo, Matsudo (Japan).
***Private dental clinic, Okabe-Cho, Ohsato-Gun, Saitama-Ken (Japan).

SUMMARY

The following results have been obtained in consequence of patho-morphological examination regarding the supplemental groove.

1. Light microscopic observation of cross-sectioned supplemental grooves revealed that most of them were shallow in the form of plate or bowl. Some of the supplemental grooves had contents not described in the past and the structure of the contents was not clear under a light microscope. The contents were found in 22% of the supplemental grooves examined.

2. The contents in supplemental grooves which were confirmed under a light microscope were found to consist of enamel itself when examined by means of an electron microscope. Microhardness measurements of this enamel showed less than one third the values of normal enamel. By means of microradiography, it was established that radiolucency of this enamel was, for the most part, much higher than normal enamel.

3. It was ascertained that enamel with low hardness and high radiolucency constitutes the contents of supplemental grooves. Judging from its tissue properties, the contents were believed to be susceptible to attack by caries. This view was supported by the results of an investigation of caries sites in supplemental grooves.

KEY WORDS:

Groove contents - Primary caries - Supplemental groove - Light and electron microscopy

RÉSUMÉ

L'examen morpho-pathologique de sillons surnuméraires a donné les résultats suivants:

1. En coupes transversales observées en microscopie classique, les sillons surnuméraires apparaissent peu profonds, en forme d'assiette ou de coupe. Dans certains, on trouve un contenu, non encore décrit dans la littérature et dont la structure n'apparaît pas nettement en microscopie classique. On trouve ce contenu dans 22% des sillons surnuméraires étudiés.

2. Etudié en microscopie électronique à transmission, le contenu des sillons surnuméraires apparaît fait d'émail. Les mesures de dureté indiquent pour cet émail des valeurs inférieures d'un tiers à celles de l'émail normal. La microradiographie montre que cet émail est, en général, beaucoup moins radiodense que l'émail normal.

3. Etant donné que le contenu des sillons surnuméraires s'est révélé être un émail de faible dureté et de faible radiodensité, on peut considérer que, par ses propriétés tissulaires, cet émail est sensible à l'attaque carieuse. Cette hypothèse est corroborée par les résultats de l'étude des sites carieux dans les sillons surnuméraires.

MOTS-CLÉS:

Contenu du sillon - Caries initiales - Sillon surnuméraire - Microscopie classique - Microscopie électronique à transmission

INTRODUCTION

Two types of grooves, developmental grooves and supplemental grooves, are distributed on the occlusal surface of tooth enamel. According to Black's description (1894, 1977), the developmental groove appears as a deep cut in enamel so that it is usually distinguishable with the naked eye.

The developmental groove is found either as a linear depression between adjoining cusps or in the valley between a cusp and a marginal ridge. A developmental groove is interconnected with other developmental grooves covering a wide range of area and is therefore the main groove on the occlusal surface.

On the other hand, the supplemental groove is the fine groove branching out from the developmental groove. It is so shallow that its shape is not easy to distinguish macroscopically. The shape of the supplemental groove remains indistinct even under slight attrition and compared with the developmental groove the supplemental groove is short and very limited in area of coverage.

Black (1894, 1977) and Diamond (1929) mention that parts of the development groove are found as abnormally deep depressions that are called fissured grooves or fissures. As well, a deep pit may be seen at the union of two developmental grooves. These morphological characteristics bring about a tendancy for caries-susceptibility to appear in the developmental grooves (Black, 1894, 1977; Diamond, 1929).

Bödecker (1926, a, b), Hyatt (1923, a, c, 1927) also agree that of all the enamel parts the developmental groove is the most susceptible to carious attack and state that there is a relationship between developmental grooves and caries.

It is considered, on the other hand, that supplemental grooves show little or no susceptibility to caries because they are much shallower than developmental grooves so that food debris do not accumulate or stagnate inside (Black, 1894, 1977; Diamond, 1929). Later researches have not shown any interest in the possible relationship between supplemental grooves and caries and therefore no patho-morphological study of the supplemental groove is available. It was the purpose of the present study to investigate the supplemental groove using light microscopy, microradiography, microhardness tests and transmission electron microscopy.

MATERIAL AND METHODS

A total of 166 human premolars and molars were used. All of them were permanent teeth with relatively slight attrition. Table I shows details of each tooth.

By means of serial grinding, supplemental grooves were sectioned transversely and replicas were obtained from each sectioned surface of the supplemental groove.

TABLE I.
Number of extracted teeth and number of supplemental grooves observed.
Nombre de dents extraites et de sillons surnuméraires observés.

Tooth		Number of extracted teeth	Number of supplemental grooves observed
Premolars	$\underline{4\vert}$	25	44
	$\overline{4\vert}$	21	33
	$\underline{5\vert}$	26	38
	$\overline{5\vert}$	27	40
Molars	$\underline{6\vert}$	16	28
	$\overline{6\vert}$	16	30
	$\underline{7\vert}$	18	25
	$\overline{7\vert}$	17	30

Each sectioned surface was first etched with 0.05% hydrochloric acid for ten seconds and then several replicas were obtained from it to be used in light and electron microscopy. For the purpose of obtaining replicas, acetylcellulose film and methyl acetate were used. The latter was employed as the solvent of the former.

For electron microscopic observation, a two-step replica method was employed. The obtained replicas were first shadowed with chromium, and then carbon was evaporated on to the surface. Furthermore, microhardness values were measured using the teeth from which the replicas had been obtained and for this, a Akashi Vickers type microhardness tester was used. The measurement was made on these teeth embedded in resin in the routine procedure.

Measurement was made on two spots: a) the center of the contents seen inside the supplemental groove and b) a spot on the enamel 300 microns away from the center of the contents, opposite to the center of the contents. The load weight was fixed at 25 g for the contents and 100 g for the enamel, with the length of time set at 20 seconds for both spots.

For light microscopy of supplemental grooves, the teeth used for the microhardness measurement were employed further to prepare ground sections and the ground sections were stained with hematoxylin and eosin. On the other hand, replicas from cross-sectioned supplemental grooves were used for the size measurement of the groove contents. For this, a stereoscopic microscope with an ocular micrometer was used. Also, by means of Soken Sofron SRO-M40 type microradiography device, contact microradiography of supplemental grooves was conducted. Additionally there were prepared ground sections of 12 teeth in which small caries in supplemental grooves were detected under the stereoscopic microscope and were observed with light microscopy after staining with hematoxylin and eosin.

RESULTS

1. Light microscopy of the supplemental groove.

A light microscopic observation of both the replicas and ground sections of teeth revealed slight differences in depth among cross-sectioned supplemental grooves. This observation proved the supplemental groove to be a shallow depression in the enamel and sometimes the supplemental grooves were so entirely filled with contents that no space was left (Fig. 1). Contents were observed in 22% of the supplemental grooves (Tab. II).

The location of the contents in the supplemental groove was not fixed, but in most cases they were restricted to one place or to several spots of the groove. Even in places where the contents spread most widely, they occupied only one third of the entire length of the groove.

In the ground sections stained with hematoxylin and eosin, portions of the contents did not show any affinity for the dye-stuff, others showed either a strong or a weak affinity for hematoxylin or eosin (Fig. 2).

TABLE II.
Number of supplemental grooves with contents in premolars and molars.
Nombre de sillons surnuméraires avec un contenu dans les prémolaires et les molaires.

Tooth		Number of supplemental grooves with contents	
Premolars	$\underline{4\|}$	6/44 (13.6%)	33/155 (21.3%)
	$\overline{4\|}$	6/33 (18.2%)	
	$\underline{5\|}$	10/38 (26.3%)	
	$\overline{5\|}$	11/40 (27.5%)	
Molars	$\underline{6\|}$	8/28 (28.6%)	26/113 (23.0%)
	$\overline{6\|}$	8/30 (26.7%)	
	$\underline{7\|}$	3/25 (12.0%)	
	$\overline{7\|}$	7/30 (23.3%)	
Total			59/268 (22.0%)

Although the tissue structure of the contents could not be established, wherever there were contents, they densely occupied the entire depth of the supplemental groove (Figs. 1-3).

2. Microhardness of the supplemental groove

Since the size of the contents was extremely limited in the transversely sectioned supplemental groove, microhardness measurements were done only at the center of the respective contents (Tab. III). The average value of the microhardness of the contents was 91, whereas that of the enamel around the groove wall was 292. That is, microhardness of the contents was less than one third of the microhardness of enamel.

TABLE III.
Microhardness (Vickers hardness) values of contents in supplemental grooves.
Coefficients de dureté (Vickers) du contenu des sillons surnuméraires.

Tooth		Number of supplemental grooves	M ± SD	
Premolars	$\underline{4\|}$	6	85.9 ± 37.7	90.5 ± 34.2
	$\overline{4\|}$	6		
	$\underline{5\|}$	10		
	$\overline{5\|}$	11		
Molars	$\underline{6\|}$	8	93.0 ± 32.4	
	$\overline{6\|}$	8		
	$\underline{7\|}$	3		
	$\overline{7\|}$	7		

3. Microradiography of the supplemental groove.

Fig. 3 shows the microradiographic aspect of a cross-sectioned supplemental groove. Throughout the contents, there are parts where radiolucency is about the same as dentin or higher but in the upper half of the contents, radiolucency is about the same as that of enamel.

4. Electron microscopy of the supplemental groove

Examined with TEM, a relatively large number of supplemental grooves had a groove wall consisting of almost normal enamel (Figs. 4, 7, 9). In some cases however the groove wall was composed of rodless enamel. On the other hand, contents of the supplemental groove were made up of a mixture of enamel with rods and rodless enamel (Figs. 4-10). A structural continuity was found between the part with enamel rods and the rodless part, and it has been established that the rodless part was enamel.

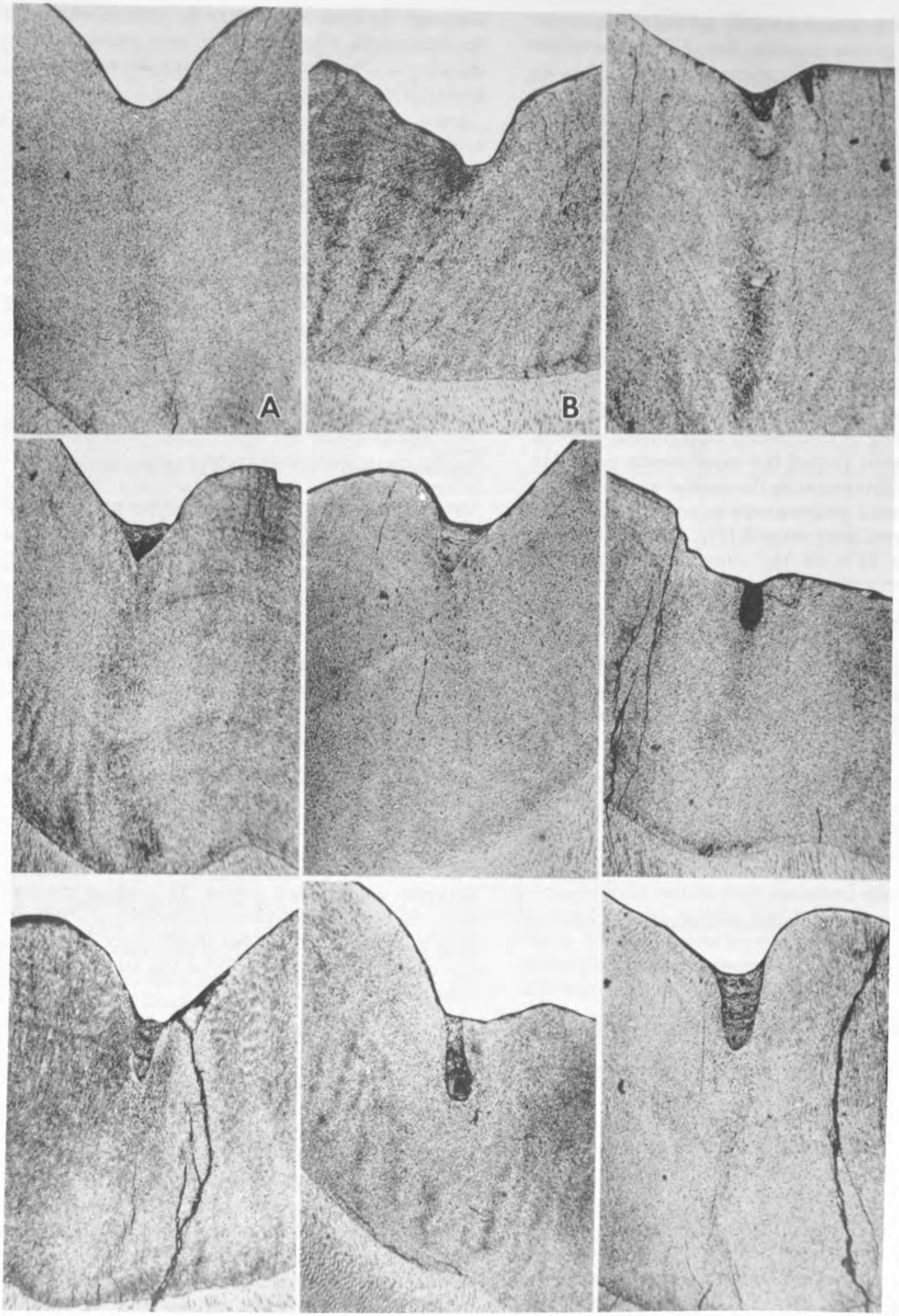
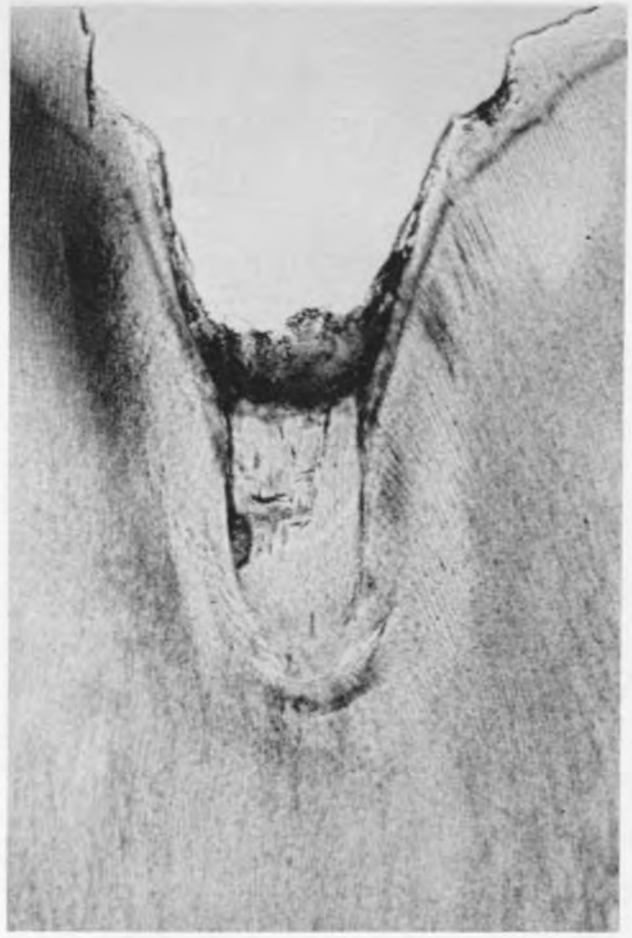


Fig. 1: Replicas from cros sectioned supplemental grooves, without content (A,B) or with content.

Fig. 1: Répliques de coupes transversales de sillons surnuméraires sans contenu (A, B) ou avec.

2a.



2b.

Fig. 2: Cross-sectioned supplemental grooves, without content (a) or with content (b). Part of the contents do not show any affinity for the dyestuff, part of them show strong or weak affinity for eosin or hematoxylin (ground sections, hematoxylin-eosin stain).

Fig. 2: Coupes transversales de sillons surnuméraires, sans contenu (a) ou avec contenu (b). Parfois, le contenu du sillon ne présente aucune affinité tinctoriale, parfois, une affinité plus ou moins prononcée pour l'éosine ou l'hématoxyline (coupes par usure).

3a.

3b.

Fig. 3: Microradiography of a transversaly sectioned supplemental groove. Radiolucency of the contents is sometimes the same or higher than dentin and sometimes about the same as enamel (b is an enlargement of a).

Fig. 3: Microradiographie d'une coupe transversale de sillon surnuméraire. La radiodensité du contenu est parfois celle de la dentine ou inférieure, parfois proche de celle de l'émail (b est un agrandissement de a).

4.

Fig. 4: Supplemental groove. The enamel structure of the content is evident in areas a, b, c, d. The enamel structure of the groove adjoining the contents assumes a normal aspect. Arrows: boundary between the supplemental groove and its content. E: enamel. C: Content. (TEM, replica).

Fig. 4: Sillon surnuméraire. La structure du contenu est, à l'évidence, de l'émail, dans les zones a, b, c, d. L'émail autour du contenu a une structure normale. Flèches: frontière entre le sillon et son contenu. E: émail. C: Contenu. (MET, réplique).

5.

Fig. 5: Enlargement of areas a and b of Fig. 4.

Fig. 5: Agrandissement des zones a et b de la Fig. 4.

6.

Fig. 6: Enlarged aspect of parts centering around c and d respectively observed in the lower half of Fig. 4.

Fig. 6: Agrandissement des zones centrées autour de c et d visibles dans la moitié inférieure de la Fig. 4.

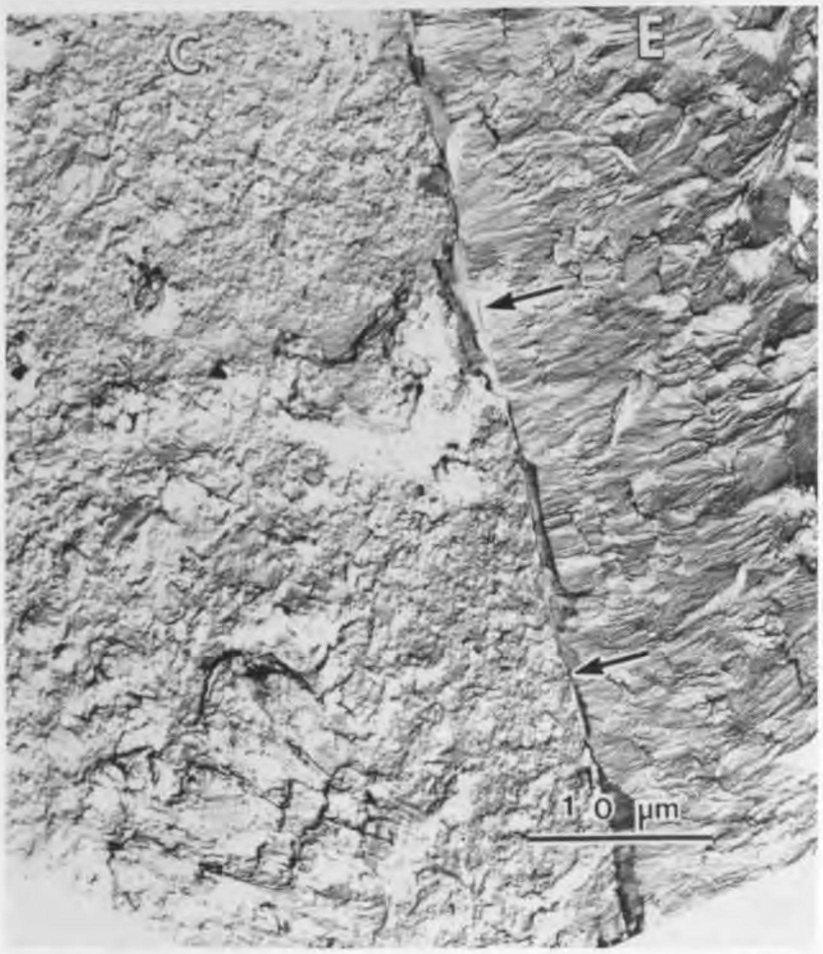


Fig. 7: Supplemental groove. Most part of groove content of rodless enamel. Enamel rods can be seen in the lower part of the content. Arrows: boundary between groove and its content. E: enamel. C: content. (TEM, replica).

Fig. 7: Sillon surnuméraire. La plus grande partie du contenu consiste en émail aprismatique. On voit de l'émail prismatique dans la partie inférieure du sillon. Flèches: frontière entre le sillon et son contenu. E: émail. C: Contenu. (MET, réplique).

Fig. 8: Enlargement of the lower part of Fig. 7 showing the rodless enamel to be continuous with enamel rods. Arrows: boundary between groove and its content. E: enamel. C: content. (TEM, replica).

Fig. 8: Agrandissement de la partie inférieure de la Fig. 7. Il existe une continuité entre l'émail aprismatique et l'émail prismatique. Flèches: frontière entre le sillon et son contenu. E: Email. C: Contenu. (MET, réplique).

These observations revealed that the part which had been regarded as the groove content composed of foreign bodies actually consisted of poorly mineralized enamel.

This part constitutes the base of the supplemental groove.

5. Size measurement of contents in the supplemental groove

The mean value of the length of the contents in the cross-sectioned supplemental groove was 241 microns, while that of the maximum width of the contents was 228 microns (Tab. IV). Actually, the values of the length and maximum width of the contents varied widely among supplemental grooves.

6. Light microscopy of primary caries in the supplemental groove.

As regards primary caries in the supplemental groove, it was noticed in every case that caries had first attacked part of the contents adjacent to the oral cavity and in some cases carious lesions had expanded further (Fig. 11). Following this stage, carious infiltration was noted on the groove wall adjoining the contents but in contrast caries were not found in the supplemental grooves without contents.

◀ Fig. 9: Supplemental groove. The content consists of both rodless enamel and enamel with rods. Arrows: boundary between groove and its content. E: Enamel. C: Content. (TEM, replica).

Fig. 9: Sillon surnuméraire. Son contenu est fait d'émail prismatique et d'émail aprismatique. Flèches: frontière entre le sillon et son contenu. E: Email. C: Contenu. (MET, réplique).

Fig. 10: Partial magnification of Fig. 9. A structural continuity is observed between rodless enamel and enamel with rods in the groove content. (TEM, replica).

Fig. 10: Agrandissement de la Fig. 9 montrant la continuité qui existe entre l'émail prismatique et l'émail aprismatique du contenu du sillon. (MET, réplique).

11a

Fig. 11: Primary caries in cross-sectioned supplemental groove.
a: early caries affect the part of the groove content directly adjoining the oral cavity.

Fig. 11: Caries initiales dans des sillons surnuméraires en coupe transversale.
a: au début, la carie atteint la partie du contenu du sillon adjacente à la cavité orale.

11b.

11d.

11c.

b: carie begins its attack toward the inside of the content.
c: finally, carie spreads over the entire content.
d: the groove content breaks down under carious attack.
b: la carie commence son attaque vers l'intérieur du contenu du sillon.
c: la carie s'étend sur toute l'étendue du contenu du sillon.
d: le contenu du sillon est détruit sous l'attaque carieuse.

11e.

11f.

f: groove content does not break down under carious attack.
e, f: following the carious changes, carie in the content infiltrates into the wall of the supplemental groove (coupe par usure, hématoxyline-éosine).

f: le contenu du sillon n'est pas détruit sous l'effet de l'attaque carieuse.
e, f: la carie du contenu du sillon s'infiltre dans la paroi du sillon surnuméraire (coupe par usure, hématoxyline-éosine).

TABLE IV.
Measurement values (nm) of the contents in supplemental grooves
Mesures du contenu des sillons surnuméraires (en nm).

Tooth		Number supplemental grooves with contents	M ± SD	
			Lenght of the contents	Maximum width of the contents
Premolars	4/ (upper)	6	299.3 ± 274.2	230.0 ± 164.4
	/4 (lower)	6		
	5/ (upper)	10		
	/5 (lower)	11		
Molars	6/ (upper)	8	209.4 ± 176.3	227.3 ± 179.5
	/6 (lower)	8		
	7/ (upper)	3		
	/7 (lower)	7		
Total		59	240.6 ± 219.0	228.1 ± 173.6

DISCUSSION

Anatomically, how is the supplemental groove distinguished from the developmental groove? Black (1894, 1977) and Diamond (1929), who are considered to have initiated the field of dental anatomy in the United States and whose approach has been maintained to date, make the distribution between the two as follows:

In the process of tooth formation, teeth have four to five calcification origins which spread over the dentin and enamel. An area formed around one calcification origin is called a developmental lobe and a tooth consists of four developmental lobes, with some exceptions.

According to the growth of the tooth, adjoining developmental lobes coalesce with one another, and as the result of this coalescence, a linear depression is left on the enamel between the developmental lobes. This is called a developmental groove. On the occlusal surface of the lower first molar, for instance, five developmental grooves are found, namely mesial, distal, buccal, distobuccal and lingual.

The more perfect the coalescence is between the developmental lobes, the shallower and less distinct a developmental groove becomes. If the coalescence between the developmental lobes is imperfect, a developmental groove created between the developmental lobes deepens, forming a fissure. In the dentin, in contrast, there is no trace of coalescence between the developmental lobes. Besides these, within the area of each developmental lobe, a linear depression is often found, branching out from the developmental groove. This is called the supplemental groove. The above-mentioned is the basic view of the developmental groove and supplemental groove held by Black (1894, 1977) and Diamond (1929).

According to Black (1894, 1977), the supplemental groove is a groove attached to the developmental groove and is a very shallow and linear depression in the enamel. Usually, the base of this groove is semicircular and smooth. These characteristics lead to the conclusion that the supplemental groove is quite different from the developmental groove. Supplemental grooves are often described as mere wrinkles on the enamel surface and regarded as part of those wrinkles, which, however, is wrong. The location and shape of the supplemental groove are always fixed, and such a groove contributes in part to the formation of the typical shape of a tooth. Black believes that the supplemental groove is usually shallow and forms no fissure.

Diamond (1929), on the other hand, holds that supplemental grooves are seen as plural fine grooves branching out from the developmental groove and are found on the occlusal surface of premolars and molars or on the lingual surface of the upper anterior teeth, and that supplemental grooves often disappear as a result of wear of the enamel surface.

The morphological view of the developmental groove and of the supplemental groove held by Ono (1919) almost exactly meets that of Black. Ono believes that as far as the supplemental groove is concerned, a linear depression is formed within an area of a developmental lobe. There are triangular grooves that appear on the molar cusp and some unusual grooves are found on the lingual ridge of the anterior tooth. These are also called supplemental grooves. Usually, the supplemental groove is shallower than the developmental groove, but it is also not unusual to find supplemental grooves which are as deep as developmental grooves. According to Ono (1919) the supplemental grooves are usually connected with a developmental groove, and this is particularly true of a triangular groove.

According to Wheeler (1940), the supplemental groove, like the developmental groove, is a linear depression on the enamel surface, but the supplemental groove, which has branched out from the developmental groove, is considered to be different from the developmental groove.

Zeisz and Nuckolls (1949) maintain that the supplemental groove lags behind the developmental groove in importance and that the difference between them is that the base of a supplemental groove is rounded U-shaped, and also, that the supplemental groove does not form a boundary between developmental lobes.

As can be seen, the researchers mentioned above hold approximately the same views regarding the developmental groove and the supplemental groove.

The developmental groove is frequently taken up as a subject of pathological or macro-morphological study, whereas the supplemental groove has seldom been studied in the past. The reason for the eager interest toward the developmental groove is that it is the part of enamel most susceptible to carious attack (Bödecker, 1926a,b; Hyatt. 1923a-c, 1927).

The supplemental groove, on the other hand, is very shallow and shows neither stagnation nor accumulation of food debris. It is therefore believed that it is seldom attacked by caries (Black, 1894, 1977; Bödecker, 1926a,b; Diamond, 1929; Hyatt, 1923a-c, 1927).

Fissures (fissured developmental grooves), which originally came under the category of developmental grooves, are remarkably deep depressions in the enamel and occupy a large portion of the distribution area of the developmental groove. It has also been established by the above-mentioned researchers that fissures contain foreign bodies consisting of food debris, germs, etc. In contrast, such contents have not been found in supplemental grooves.

In the present study, however, light microscopy allowed to show that some of the supplemental grooves were filled with contents. The frequency of their occurrence differed between premolars and molars, but 22% of the supplemental grooves observed had such contents. The contents detected with light microscopy were examined with electron microscopy and submitted to microhardness tests. It was thus confirmed that the contents of the supplemental grooves consisted of enamel whose hardness was less than one third of that of normal enamel. By microradiography, as well, it was established that the radiolucency of the contents was much higher than that of normal enamel, except for certain small parts.

Fissure contents which have hitherto been accepted as foreign bodies had been examined by a number of investigators using various methods: Awazawa (1966) used electron microscopy. Gwinnett (1966) made use of polarizing microscopy; Sobue and his collaborators (1974) used together polarizing microscopy, microradiography and X-ray crystallography.

Katada and his collaborators (1982) employed microhardness tests. The results obtained by these researchers show that the contents are made up of hypomineralized enamel.

Galil and Gwinnett (1975) made a histological examination of an unerupted tooth and made clear the origin of fissure contents as follows: In the tooth before eruption, developmental grooves were found in the state of fissures even after the completion of enamel formation, with the enamel organ still remaining in the groove. They observed alignment of ameloblasts on the groove wall, and considered that, under such circumstances, ameloblasts densely crowding in the groove would press against one another. This congestion and pressure, they said, would disturb the ameloblasts in the formation of enamel matrix and subsequently normal mineralization of the enamel would not be possible. Consequently, poorly mineralized enamel, which had been considered as fissure contents, would be produced in the groove. This is the case of the developmental groove.

Similarly, in our opinion, the enamel organ could be found also in the supplement groove during the period when tooth eruption has not yet started. Regarding the origin of the contents in the supplemental groove, we believe that it may be similar to that of the supplemental groove described by Galil and Gwinnett (1975).

It is generally believed that carries seldom attack supplemental grooves. According to Ono (1919), for instance, developmental grooves and supplemental grooves are in most cases connected with each other and therefore caries that attacked a developmental groove can easily spread to supplemental grooves or vice versa, although caries rarely attacks supplemental grooves.

The results obtained from the present study differ considerably from the conventional concept in that primary caries were seen to affect the contents of the supplemental grooves in many cases. However, there was no incidence of caries in supplemental grooves without contents. In other words, enamel with low hardness and high radiolucency which forms the contents of the supplemental groove proved to be more susceptible to carious attack than normal enamel.

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AWAZAWA, Y. et al.
Present address:
Nihon University School of Dentistry at Matsudo,
Matsudo (Japan).