Variations in the configuration of pulp chambers subjacent to protostylids

Kei Sakakibara1), Yoshiro Kawano2), Hiroaki Yamamoto2), Masatoshi Iwahori3), Yukako Sugiura4) and Motonobu Miyao5)

1) Department of Prosthodontics, Division of Oral functional Science and Rehabilitation, Asahi University School of Dentistry, Mizuho, Japan
2) Department of Oral Anatomy and Forensic Dentistry, Asahi University School of Dentistry, Mizuho, Japan
3) Department of Oral Anatomy and Forensic Dentistry, Asahi University School of Dentistry, Mizuho, Japan

Abstract

Purpose: This study aimed to clarify the positional relationship between the crown contour and pulp chamber of protostylids using three-dimensional reconstructed images.

Methods: Fourteen molars with protostylids from Japanese subjects were subjected to micro-computed tomography. The external surface configurations of the teeth and pulp chambers were reconstructed. Hard tissue thicknesses in appointed buccal areas were measured on the reconstructed images.

Results: Well-developed protostylids exhibited pulp-prominences above or at the cervical line level. Those that were moderately developed exhibited bulges of the pulp chamber subjacent to the protostylids. Ten of the 14 teeth had prominences in the crown pulp above or at the cervical line level. In addition, 13 teeth exhibited pulp chamber bulges surrounding the lower tooth trunk. No significant differences were apparent in the buccal horizontal thickness of the hard tissue between the protostylids with pulp chamber prominences and the protostylids without pulp chamber prominences at the cervical line level.

Conclusion: Pulp chamber configurations subjacent to protostylids vary based on the development of the traits of the protostylids. Minimum possible taper should be applied during standard vital tooth preparations, as reduced residual dentin thickness is predicted in well- and moderately developed protostylids.

Keywords: dental restorations, Japanese population, micro-computed tomography, protostylids, pulp chamber configuration, pulp variation

Introduction

Human teeth exhibit various morphological features and forms. The term ‘paramolar tubercle’ was first defined as an additional cusp formation on the buccal surface of the second and third molars [1]. The manifestations of paramolar tubercles vary from a mere prominence on the buccal surface to a well-developed lobulated cusp that is similar to a fused supernumerary tooth in appearance. These anomalous tooth contours can pose a problem during ideal dental preparation processes, such as while developing designs for crown preparation and in dental banding during orthodontic treatment [2]. Moreover, they are often associated with rudimentary or fully formed roots and contain their own pulp and root canals [1,3]. Hence, paramolar tubercles are potentially clinically relevant as they have an influence on treatment modalities and associated problems in dental disciplines [4-6].

Advancements in three-dimensional (3D) imaging devices have led to a better understanding of a tooth’s internal and external structures without causing destructive effects [7-9]. From an anthropological point of view, many researchers have investigated the morphology of the dentino-enamel junction of paramolar tubercles using 3D-reconstructed images [10,11]. However, there are few reports on the pulp chamber configurations of teeth with anomalous tubercles, even though this is additional useful information that is required in dental practice.

A protostylid is a prominence or ridge of the enamel that ascends from the gingival ends of the buccal groove, extends mesio-occlusally, and is limited to the mesial part of the buccal surface in the lower molars [12]. Owing to its low global prevalence rate, little information is available regarding both the morphological characteristics of protostylids and their relationship with pulp chambers. However, several studies have reported higher incidence rates of protostylids in some racial and/or ethnic groups, such as in Japanese and Pima Indian populations (26.8%, 31%, respectively), which are considerably higher than in other racial groups [13]. Therefore, during dental practice, the presence of protostylids cannot be disregarded in these races and/or ethnic groups.

The amount of remaining dentin between the prepared surface and pulp is a critical factor in determining the severity and incidence of pulpal lesions [14]. However, in standard dental preparation techniques, only an estimate can be made regarding the mesiodistal proximity of a pulp chamber to the outer tooth surface using conventional dental radiography. There is currently no way of knowing the exact thickness of the residual dentin either before or following crown preparation. In prosthetic dentistry, knowledge of the entire circumferential pulp morphology is a prerequisite for vital tooth preparation for full-coverage crowns. However, little information is available on the 3D pulp chamber anatomy, and particularly, on the detailed morphology of the pulp subjacent to anomalous protuberances.

Therefore, the purpose of this study was to clarify the configurations of the pulp chambers subjacent to protostylids and to assess their spatial relationship to tooth contours by measuring the thickness of hard tissue in selected positions.

Materials and Methods

Study samples

The research protocol was approved by the Human Subjects Committee at Asahi University (approval no. 30035, 2019). The study sample included 14 molars (first, second, and third molars; Ps1, Ps2, Ps3…Ps14) with protostylids that were housed at the Department of Oral Anatomy in Asahi University. The age and sex of the individuals that they belonged to and the reasons for extraction were unknown. They were all from Japanese subjects and had various extents of protostylytis traits with minimum attrition and negligible caries. Each of the teeth were numbered and macroscopically categorized into three types based on Sakai’s scoring method (Sakai T et al., Aichi-Gakuin Daigaku Shigakukai Shi 7, 265-314, 1970): Type ++++, a well-developed protrusion forming an independent cusp; Type ++, a distinct protuberance with a tip; and Type +, a tubercle distinguishable from the buccal surface with an indistinct tip and border.

Micro-computed tomography (micro-CT) and 3D reconstruction

In order to produce 3D reconstructed images of the tooth contours and pulp chambers, each tooth was subjected to micro-CT scans (ScanXmate-RR090SS, Comscantecno, Yokohama, Japan) at an isometric voxel resolution of 16 μm (90 kV, 90 μA, 0.1 mm copper filter, 0.18 rotation step, 360 degrees of rotation, 2 frame averaging). Using 3D reconstruction software (Image-pro, Media Cybernetics, Rockville, MD, USA), 3D images of the molars were reconstructed from approximately a thousand image slices. The reconstructed images of the right molars were transformed into mirror images using a software application (Photoshop 2019, San Jose, CA, USA), and all the specimens were regarded as left molars.

Correspondence to Dr. Yoshiro Kawano, Department of Oral Anatomy and Forensic Dentistry, Asahi University School of Dentistry, 1851 Hozumi, Mizuho, Gifu 501-0296, Japan
Fax: +81-58-329-1407 E-mail: kawano@dent.asahi-u.ac.jp

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The dental hard tissue and pulp chambers were segmented by the thresholding of the different gray-scale intensities, and the configurations of the external surface and pulp chambers of the teeth were reconstructed. To facilitate the identification of each part and the assessment of the spatial relationship, different colors were applied to the tooth contours (green and pulp chambers (red)). For a detailed observation, the tooth contours were rendered half-transparent, and the topographic relationships between the contours of protostylos with the pulp chambers were observed from various aspects. The shapes of the pulp chambers in the tooth trunk and those of the supernumerary root canals were also focused on.

Morphometric analysis
As illustrated in the schematic drawing presented in Fig. 1, morphometric measurements of the dimensions were performed on the reconstructed 3D images in the different appointed positions: Distance 1, the distance from the buccal cusp to the corresponding pulp horn; Distance 2, the distance from the protostylid peak to the corresponding pulp prominence; Distance 3, the shortest horizontal distance from the prominence of the pulp chamber subjacent to the protostylid to the buccal tooth surface; and Distance 4, the shortest distance in the tooth trunk. The shortest distance at the cervical line level (Distance 3) was substituted for Distance 3 in the Type + teeth (Fig. 1).

Statistical analysis
Using a statistical software application (JMP 14.2, SAS, Cary, NC, USA), comparisons of the morphometric data associated with the thickness of the hard tissue among the different positions were performed via a Wilcoxon signed-rank test in the case of individual teeth and a Kruskal-Wallis test among the different positions of the morphometric data associated with the thickness of the hard tissue among the different positions of the teeth.

Results
Macroscopic observations
Figure 2 presents the occlusal and buccal views of the Type +++ protostylids. They had a well-developed tubercle forming independent cusps and a well-defined sulcus (Fig. 2). The height of the cusps of the protostylids varied from reaching up to two thirds of the height of the mesiobuccal cusp (Fig. 2a, b, d) to reaching the level of the buccal marginal ridge (Fig. 2c, e, f). Ps14 exhibited a paramolar tubercle in the middle of the buccal surface and a superimery root between the anterior and posterior roots (Fig. 2f). Ps8 exhibited a superimery root coalesced with the anterior root (Fig. 2c, arrowhead). Cementum was developed on the root surfaces of Ps2, Ps8, and Ps14 (Fig. 2a, c, f). The Type +++ protostylids exhibited less developed, less distinct tubercles and grooves than the Type +++ ones (Fig. 3).

The Type + teeth displayed protostylids with indistinct borders, but the contours of the protostylids were discernible (Fig. 4). Ps10 and Ps11 had protuberances on the buccal surface of the distobuccal cusp and Ps11 had a supernumerary root between the medial and distal roots (Fig. 4b, c). Moderately developed cementum was noted on Ps7 (Fig. 4a).

Reconstructed 3D tooth contours and pulp chambers
The reconstructed images highlighting the topologic features subjacent to the protostylids clearly demonstrate the 3D anatomy of the pulp chamber and its relationship with the outer surface (Fig. 5 for control, Fig. 6 for Type +++, Fig. 7 for Type ++, and Fig. 8 for Type +). The control tooth exhibited rounded bulges of pulp chamber in the tooth trunk below the cervical line and smooth lateral pulp chamber surfaces from the marginal ridge to the bulges (Fig. 5).

The Type + teeth had pointed pulp horn-like prominences subjacent to the protostylids (Fig. 6a-f). The tips of the pulp prominences were located higher than or around the cervical line level. All the teeth that were categorized as Type +++ exhibited similar structural and positional relationships. The Type + protostylids exhibited bulges in the pulp chamber corresponding to the protostylid protuberances (Fig. 7). The vertical positions of the bulges of the pulp chambers were nearly at the cervical line level or above it. The Type + teeth did not exhibit any anomalies in the corresponding pulp chamber area (Fig. 8).

Rounded bulges in the tooth trunk
Most of the teeth including the control molar had large rounded bulges of the pulp chamber in the tooth trunk below the cervical line (Figs. 5-8). The convexities of these bulges varied from being shaped as a pointed horn to a

**Fig. 1** Diagram presenting the distances measured. Distance 1, distance from the pulp prominence to the corresponding buccal cusp; Distance 2, distance from the pulp prominence to the corresponding protostylid peak; Distance 3, the shortest distance from the prominence to the buccal tooth surface; and Distance 4, the shortest distance from the pulp chamber to the root trunk surface.

**Fig. 2** Occlusal and buccal views of the Type +++ protostylids. Ps8 has a rudimentary root on the anterior root (c, arrowhead). Ps14 has a protrusion in the middle of the buccal surface (f, arrow) and an extra root between the anterior and posterior roots (f, arrowhead). Well-developed cementum on the root surfaces and rounded root apexes are discernible in Ps2, Ps8, and Ps14 (a, c, f). Scale bar, 4 mm.
rounded mound. Ps5, Ps8, Ps12, and Ps14 exhibited pointed prominences below the cervical line around the tooth trunk pulp chamber, and their rudimentary roots are on the anterior and posterior roots (d, arrowheads). Ps4 and Ps5 exhibit well-developed cementum on their root surfaces (b, c). Scale bar, 4 mm

Distances from the pulp to the external surface
The measured distances in each tooth are plotted in Fig. 9. In the Type +++ protostylids, the distances from the protostylid peak to the pulp chamber (Distance 2) ranged from 4.39 mm to 5.62 mm (Mean, 5.16 ± 0.43 mm), with no significant differences from those between the protostylids and buccal cusp (Distance 1), which ranged from 4.60 mm to 5.91 mm (Mean 5.00 ± 0.48 mm) in each tooth (P = 0.47) (Fig. 9a).

In the Type ++ protostylids, these distances ranged from 4.70 mm to 5.71 mm (mean, 5.17 ± 0.43 mm), and in the mediobuccal cusp, they...
ranged from 4.55 mm to 5.70 mm (mean, 5.13 ± 0.58 mm). Similarly, there were no significant differences between them ($P = 0.77$) (Fig. 9b).

In the Type ++ specimens, the horizontal distances (Distance 3) measured from 2.34 mm to 3.48 mm (mean, 2.98 ± 0.33 mm), and in the Type + samples, they measured from 3.01 mm to 3.78 mm (mean, 3.31 ± 0.33 mm), which were similar or more than the distance measurements of the Type ++ samples at the cervical line level (mean, 2.98 ± 0.33 mm). There were no significant differences between the types ($P = 0.54$) (Fig. 9c).

The shortest distances in the buccal tooth trunk (Distance 4) measured from 1.95 mm to 2.48 mm (mean, 2.24 ± 0.16 mm), 1.99 mm to 2.44 mm (mean, 2.21 ± 0.16 mm), and 2.05 mm to 2.27 mm (mean, 2.16 ± 0.10 mm) in the Type +++, Type ++, and Type + specimens, respectively, with no significant differences among the types ($P = 0.60$) (Fig. 9d).

**Supernumerary roots**

Corresponding to the protostylids, four teeth possessed supernumerary roots or vestiges of roots that coalesced with the normal roots. Ps6 had rudimentary roots that coalesced with the anterior and posterior roots, respectively. The mesial one had its own independent root canal (Fig. 10a). Ps8 exhibited a supernumerary root that coalesced with the buccal side of the anterior root divided by a groove extending from the tooth trunk to the apex. The rudimentary root possessed its own root canal, which joined...
the pulp chamber on the mesiobuccal pulp floor (Fig. 10b). Ps11 and Ps14 had developed their own independent extra roots between the anterior and posterior roots (Fig. 10c, d).

Discussion

The results clearly demonstrate that the presence of anomalous pulp chamber morphologies is closely related to the degree of development of protostylist.

All the teeth categorized as Type +++ and Type ++ exhibited anomalous protrusions in the corresponding pulp chamber. Following their investigation into protostylids in 181 Japanese molars, Sakai et al. reported a 26.8% incidence in the first molars and 10.2% in the second molars (Sakai T et al., J Anthropol Soc Nippon 81, 25-45, 1973). They also reported the discrete incisions of the three protostylid types (1.8% for Type +++, 4.5% for Type ++, 20.5% for Type +) in the first molars. Hence, it can be concluded that 6.3% of Japanese individuals are estimated to have Type +++ or Type ++ protostylids and to exhibit anomalies in the configurations of the pulp chamber subjacent to them. The sample size was small consisting of a total of 14 teeth (6 for Type +++, 4 for Type ++ and 4 for Type +) and, therefore, may not be sufficient to generalize the trend involved in the relationship between the development of the trait and pulp configuration. However, considering the total incidence rate of protostylids in Japanese subjects, the fact that all the teeth that were categorized as Type +++, or Type ++ (a total of 10 in 14) exhibited anomalies in the pulp chamber, at the very least, presents information that necessitates the attention of clinicians. Although there are discrepancies in the incidences suggested by various researchers and given that most of the protostylids are categorized as Type +++, Type ++ and Type +, the prevalence of protostylids with abnormal pulp chambers in the Japanese population is not negligible when administering prosthetic restoration treatments.

Unexpectedly, the results showed that 13 of the 14 teeth had bulges surrounding the pulp chamber of the lower tooth trunk. Meanwhile, Ps1 did not exhibit any bulges in the tooth trunk. This report does not detail the reasons behind the formation of the bulges in the tooth trunks. Nevertheless, the possibility of encroachment of site-specific depositions of secondary irregular dentin into the pulp tissue being one of the reasons cannot be excluded. Studies have revealed that the production of secondary dentin increases with age throughout an individual’s entire lifespan and that a larger amount of secondary dentin is deposited on the floor of the pulp chamber and in the orifice of root canals than on the roof or side walls in multi-rooted teeth [15-17]. Accordingly, pulp chamber width decreases with an inflection point at 35-40 years of age, and the rate of the decrease becomes dramatic thereafter [18]. Moreover, from when an individual becomes approximately 50 years old, irregular secondary dentin apposition dramatically increases. Furthermore, the level of irregular secondary dentin apposition was found to be highest on the side walls of crown pulp chambers that were subjected to percussive contact, and in samples extracted from individuals over 50 years old, irregular secondary dentin might encroach into the crown pulp chamber as far as below the cervical line level from the side walls of the crown pulp chamber [15,19].

Based on the results of these studies, it would appear that with advancing age, the irregular secondary dentin formation on the roof and surrounding upper lateral walls gradually squeezes the upper halves of the crown chamber and that the dentin on the walls surrounding the orifices of the root canals and the greater mass of dentin on the pulp floor together squeeze the lower halves of the pulp chamber. Consequently, pulp chamber bulges surrounding the tooth trunk are observed. There is no information regarding the exact ages of the patients during the extraction and the reasons for extraction of the teeth. However, from a macroscopic observation of the color and development of the cementum on the roots, it was presumed that the teeth that exhibited surrounding bulges in the lower tooth trunk were from comparatively elderly subjects. In contrast, based on the rounded pulp horn and morphology of the root apex, it was speculated that Ps1 was from an individual at quite a young age. A further study involving an intensive observation of a number of teeth from subjects with exact known ages is required in order to clarify the relation between the emergence of the bulges in the tooth and increasing age.

Bolk (1916) pointed out that paramolar tubercles are always attached to the mediodiabucal cusps and that their supernumerary roots are attached to the mesiobuccal roots [1]. Moreover, inconsistent tendencies are exhibited by the supernumerary roots that are involved with paramolar tubercles in the upper and lower molars. They tend to attach at the root in the upper molars, whereas they often develop in the lower molars even though the tubercle is not well developed or is entirely absent [1]. Regarding the location of the protuberances, in most of the teeth that were investigated, the location-related observations were consistent. Interestingly, however, Ps6 had two discrete protuberances on the buccal surface, and they were attached at the surface of the mesiobuccal and distobuccal cusps. Ps10 and Ps11 had indistinct tubercles on the buccal surface of the distobuccal cusps. On the other hand, four of the 14 teeth were associated with rudimentary or fully formed supernumerary roots. Two rudimentary roots of Ps6 were coalesced with the anterior and posterior roots. Ps11 and Ps14 developed independent supernumerary roots between the anterior and posterior roots regardless of the development of protostylistid. Although discrepancies between these findings and those of previous reports aroused interest in the associated odontogenesis process, an analysis of the location and developmental process of protostylistids and their roots is beyond the scope of this investigation. However, it can be clinically concluded that details regarding the root structure involved in protostylistid development cannot be determined from the degree of the development of the traits of protostylistids. These extra root elements lead to complications in the variation of root morphology and additional root canals and may result in unanticipated difficulties and prognoses associated with periodontal and endodontic treatment. To plan and administer optimal dental treatment, the supernumerary roots that are potentially involved with protostylistids must be taken into consideration. Further root morphology-related research requirements that are relevant to protostylistids must be extensively addressed.

Currently, there is no scientific consensus regarding the minimal thickness of dentin required to protect pulp vitality [20]. Practically, it is widely accepted that a thickness of 2 mm or more of remaining dentin is critical for the protection of the pulp following tooth preparation [14,21]. However, a previous study reported that analyzing the long-term outcomes of prosthetic treatment revealed that up to 15% of a sample of 672 teeth exhibited negative pulp vitality testing results 4-13 years following crown preparation [22]. Ideally, metal crowns should not be less than 0.5 mm in thickness. Porcelain-fused metal restorations, which are commonly used as crowns, require relatively deep tooth preparations ranging from approximately 1.0 to 1.5 mm on all surfaces apart from the occlusal surface. Although an equivalent amount of hard tissue must be removed, the shortest horizontal distance from the pulp to the buccal surface in Type +++ and Type ++ teeth is not significantly different from the corresponding distance at the cervical line level in Type + teeth. On the contrary, these values are more than or similar to those of normal mandibular molars [23,24]. Notwithstanding that no conclusions were drawn regarding the appropriate preparation design for crowns based on the findings, the fact that there are no significant differences in the thickness of the hard tissue between normal cusps and protostylistids allows for dentists to determine the outline of the pulp chambers by studying the contour of protostylistids. In cases in which pulpal prominences are expected to be present above the cervical line level, the minimum possible taper should be considered for pulp protection during tooth crown preparation, especially during complete-coverage restoration treatment that requires a deeper preparation into the dentin. The results can help practitioners to understand and anticipate the challenges that could affect dental treatment planning and outcomes for molars with protostylistids.

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Conflict of interest

The authors declare no conflict of interest in this study.
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