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## ORIGINAL ARTICLE

# A histological and radiographic study of pulpal calcification in periodontally involved teeth in a Taiwanese population



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#### **KEYWORDS**

age related; dental pulp; histological examination; pulpal calcification; radiographic examination; systemic disease

Abstract Background/purpose: The prevalence of pulpal calcifications was widely studied in the past. The purposes of this study were to investigate the incidence of pulpal calcifications of periodontally involved teeth in a Taiwan Chinese population using radiographic and histological examinations, and to find out any association of pulpal calcification with systemic disease and dental conditions.

Materials and methods: A total of 197 teeth freshly extracted because of severe periodontal destruction were collected and prepared for histological and radiographic studies of the incidence of pulpal tissue calcifications. The occurrences of calcifications were recorded based on the different types of classifications proposed by Seltzer (1972). The number of examined teeth with pulpal calcifications was calculated, and they were statistically analyzed with the Chi-square test.

Results: The patient population ranged in age from 16 years to 85 years. Of them, 165 (84%) were male and 32 (17%) were female. The results show that the incidence of pulpal calcifications of periodontally involved teeth was 62% in histological and 30% in radiographic examinations. The occurrences were slightly higher than that reported in some previous studies and significantly different between the two examined methods. No significant association of pulpal calcification with age and systemic disease was found. Moreover, molars were observed to have more pulpal calcifications than bicuspids and incisors statistically (P < 0.001).

Conclusion: The true incidence of pulpal calcifications of periodontally involved teeth is likely to be higher in histological examination because pulpal calcifications with a small diameter may not be seen on radiographs. The prevalence of pulpal calcifications was found to increase

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significantly in molar teeth, and the results indicated that localized calcifications occur mostly in the radicular area of the pulp tissue.

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#### Introduction

Pulpal calcifications are fairly common in human dental pulps. They may occur in any one tooth or all teeth, including deciduous or permanent, unerupted or impacted, and healthy or diseased teeth, and in tooth-like structures of dermoid cysts. <sup>2,3</sup> Calcification may occur in the dental pulp in diffuse forms or as discrete calcified stones that may exist "freely" in the pulp tissue or become "attached" or "embedded" into dentin. <sup>4,5</sup> Calcification of tissues has been of interest for well over a century, but the factors related to dystrophic changes leading to calcifications are not absolutely known. <sup>6</sup> Pulp canal calcifications usually pose a challenge in both endodontic diagnosis and treatment. <sup>7</sup>

Earlier literatures report that calcified changes frequently occur because of dental caries, tooth abrasion, periodontal disease, pulp inflammation, and traumatic injury. 2,8,9 Cahn<sup>10</sup> first hypothesized that calcification within the pulp was a pathological process of deposition of calcium salts within the tissue. 4,11 Orban, 12 and Stenvik and Mjor<sup>13</sup> asserted that epithelial rests trapped in the pulp tissue initiated cellular activity, resulting in the formation of calcifications. Hill<sup>14</sup> expressed the opinion that the formation of pulpal calcifications was associated with certain stellate cells of the pulp, which are responsible for building an irregular dentin around calcific deposits. Calcifications might be caused by infolding of odontoblasts during the tooth development period, resulting in the formation of islands of dentin. 15 Anderson 16 suggested that the formation of pulp stones could be due to hypercalcemia of foreign bodies, such as dead cell or bacteria. Seltzer et al<sup>5</sup> in their study suggested that the etiology of pulpal calcifications was hemorrhage.

In 1965, Rubach and Mitchell<sup>17</sup> studied the microscopic variations of calcific alterations in the pulp and concluded that calcifications were generally not related to periodontal bone loss. Holcomb and Gregory<sup>18</sup> and Patterson and Mitchell<sup>19</sup> analyzed the incidence of calcific metamorphosis using radiographic techniques, and found a significant correlation between calcification and trauma. Although many studies have reported that the presence of mineralization is unrelated to age, 3,6,11,15 some studies purported to show that the incidence of mineralized bodies in the pulp increases with age.<sup>5,20,21</sup> A high incidence of calcified masses in pulps with carious lesions or in restored teeth was also reported.<sup>22</sup> Other possible factors such as microorganisms as well as various local or systemic diseases, including arteriolosclerosis, renal lithiasis, gout, osteitis deformans, hypercementosis, and torus palatinus, have been considered as the possible causes of pulpal calcifications. 23-25

The prevalence or occurrence of pulpal calcifications in human teeth has widely been studied and reported to range from a low of 8% to a high of 90% (Table 1).  $^{26-32}$  Many previous studies on pulpal calcifications in the dental pulp have been based on the radiographic method.  $^{33-37}$  The true prevalence is believed to be higher in histological findings because calcified masses with a diameter smaller than 200  $\mu m$  cannot be identified easily on radiographs.  $^{35}$  It has been estimated that only about 20–25% of pulpal calcifications can be detected by radiographic observations.  $^{2,3,6,36,37}$  Furthermore, the limited number of histological sections through each examined tooth during observations may result in under-reporting.  $^{15}$ 

	Previous f pulpa	s studies on th	e prev	alence (or	
Methodology	Year	Investigators	No. of teeth	Prevalence (%)	
	4000	a. c. a. a. 1. 74			
Histological	1933	Stafne & Szabo <sup>24</sup>	200	46	
studies	1934	Hill <sup>14</sup>	132	66-90	
	1934	William <sup>15</sup>	164	87	
	1959	James et al <sup>26</sup>	159	52-57	
	1965	Langeland & Langeland <sup>27</sup>	155	19	
	1968	Sayegh & Reed <sup>20</sup>	591	8-90	
	1968	Sundell et al <sup>22</sup>	470	11-18	
	1970	Stenvik & Mjor <sup>13</sup>	95	25-50	
	1983	Moss-Salentijn & Klyvert <sup>21</sup>	175	25	
	1986	Yaacob & Hamid <sup>29</sup>	120	7	
	1988	Baghdady et al <sup>2</sup>	515	19	
	1993	Arys et al <sup>31</sup>	42	78	
	1997	Hillmann & Geurtsen <sup>32</sup>	332	3–19	
	2016	Present study	197	62	
Radiological	1934	William <sup>15</sup>	164	14-15	
studies	1967	Holcomb & Gregory <sup>18</sup>	881	4	
	1982	Tamse et al <sup>28</sup>	1380	8-57	
	1983	Moss-Salentijn & Klyvert <sup>21</sup>	175	25	
	1988	Baghdady et al <sup>2</sup> 6228 19		19	
	1990		120	6-25	
	1998			22	
		Darwazeh <sup>3</sup>			
	2002	Ranjitkar et al <sup>36</sup>	217	10	
	2003		121	4	
	2016	Present study	197	31	
<sup>a</sup> Based on t	he stuc	ly of Goga et al <sup>6</sup> in 2	.800		

The purpose of this study was to: (1) investigate and compare the occurrence of pulpal calcifications of periodontally involved teeth using histological and radiographic techniques in a Taiwan Chinese population; (2) find out the incidence of the different types of pulpal calcifications at different locations of dental pulp tissues; and (3) evaluate the association between the occurrence of pulpal calcifications and some related factors such as age, gender, tooth type, and systemic condition.

#### Materials and methods

A total of 197 (174 patients in the age range of 16-85 years) human teeth with severe periodontal involvement but no previous dental treatment were collected for this study. The consent form was explained, and signed forms were obtained from the patients who agreed to have their teeth extracted. The teeth were then extracted because of extreme mobility and severe periodontal breakdown. All the specimens included permanent, single-rooted, and multirooted teeth. Detailed information concerning the patient's medical and dental histories, and tooth conditions and locations were clinically inspected and recorded for further analysis. The vitality status of the experimental teeth were first verified using a vitality scanner (SybronEndo; Kerr, Orange, CA, USA), and the teeth were collected immediately after extraction. The specimens were fixed with 15% aqueous buffered formalin for a minimum of 5 days. During the fixation period, all teeth were accessed with a No. 4 diamond round bur through the incisal or occlusal surface to expose the pulp tissue and improve tissue fixation.

All the specimens were first accepted the digital periapical radiographic examination with the Sirona Dental X-ray system (Siemens, Bernsheim, Germany) in buccolingual and mesiodistal direction using the paralleling technique along with film holders (Rinn Corporation, Elgin, IL, USA). A storage phosphor plate film was processed using a ScanX Duo digital imaging system (Air Techniques, Melville, NY, USA) following the manufacturer's instructions. Two endodontists examined the radiographic images on the monitor to detect the occurrence of calcifications.

The teeth were then notched along the long axis in the buccolingual direction with tapered fissure diamond bur

under water coolant, and then split with a sharp rongeur or chisel. All the pulp tissues were completely removed from the tooth segments using a DG-16 endodontic explorer (Hu-Friedy, Chicago, IL, USA). The pulp tissues were then dehydrated in ascending concentrations of ethyl alcohol and embedded in paraffin. Five representative histological sections cut in a plane perpendicular to the long axis of the pulp were sectioned at 5u, and the sections were stained with hematoxylin and eosin for histological examination. All sections were examined under a light microscope to detect the presence of calcified structures in the fixed pulp tissue by the two endodontists. The occurrence of any tissue calcifications were confirmed if it was present in at least two sections from a given pulp tissue. Different types of calcifications, such as diffuse and localized (including pulp stone and denticles), were recorded according to the classification of Seltzer<sup>1</sup> in 1972.

The frequency distribution of teeth with pulpal calcifications was calculated. The data were evaluated by Chisquare analysis. Differences were considered significant when P < 0.05.

#### Results

A total of 226 teeth were collected for this study. Of the patients, 187 (83%) were male and 39 (17%) female. Owing to technical difficulties involving the procedures of tooth splitting and pulp tissue removal, 197 specimens were included for histological examination. The specimens consisted of 53 single-rooted teeth (incisors and cuspids) and 144 multirooted teeth (40 bicuspids and 104 molars; Table 2).

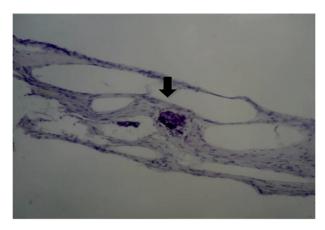
Histological hematoxylin and eosin examination revealed the occurrence of pulpal tissue calcifications in 122 (62%) out of 197 specimens. Of these teeth with calcifications, 29 (24%) were incisors and cuspids, 24 (20%) bicuspids, and 69 (56%) molars (Table 2). Calcifications could be subdivided into two different types, localized (Figure 1) and diffuse calcifications, based on their distribution and structures (Figure 2). Localized pulpal calcifications were seen in 71 teeth (15 incisors and cuspids, 17 bicuspids, and 39 molars) and diffuse calcifications in 51 teeth (14 incisors and cuspids, 7 bicuspids, and 30 molars). In addition, localized pulpal calcifications were found in the coronal region of 39 (54.9%) teeth and in the radicular area of 62

Teeth	No. of teeth	Teeth with PC <sup>a</sup>	Histolog	ogical findings of PC (%) <sup>b</sup> Localized		Radiographic findings of PC (%)	
			Diffuse			Yes	No
				Coronal	Apical		
Incisor & cuspid	53 (27)	29 (24)	14 (48)	8	15	23 (38)	30 (22)
Bicuspid	40 (20)	24 (20)	7 (29)	12	16	17 (28)	23 (18)
Molar	104 (53)	69 (56)	30 (43)	19	31	20 (34)	84 (60)
Total	197	122 (62)	51 (42)	71	(58) <sup>c</sup>	60 (30)	137 (70)
Chi-square test			P = 0.341			P = 0.001	

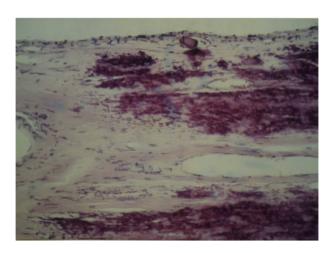
Data are presented as n or n (%).

- <sup>a</sup> PC = pulpal calcification.
- <sup>b</sup> The distinction between diffuse and localized calcification is based on the classification by Seltzer<sup>1</sup> in 1972.
- <sup>c</sup> Seventy-one teeth were found to have localized calcification, with 30 of them being identified in both coronal and apical areas.

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**Figure 1** Localized dystrophic calcification (arrow) and acute inflammatory cell infiltrate and hemorrhage (H&E stains,  $40\times$ ). H&E = hematoxylin and eosin.



**Figure 2** Diffuse dystrophic calcification in dental pulp. High magnification shows needle-like cylinder type of calcified material, which appeared parallel to collagen fibers of the pulp tissue (H&E stains,  $40\times$ ). H&E = hematoxylin and eosin.

(87.3%) teeth. Localized calcifications were found both in the coronal and in the radicular regions in 30 cases (Table 2). Under radiographic assessment, calcifications in the pulp canal were found in 60 (30%) tooth specimens, of which 23 (38%) were incisors and cuspids, 17 (28%) bicuspids, and 20 (34%) molars (Table 2 and Figure 3).

The frequency of pulpal calcifications was demonstrated to be higher in molars than in premolars or in incisors and cuspids, based on both histological and radiographic findings. In the meantime, distribution of the study patients having pulpal calcifications according to age groups is shown in Table 3, and the relationship between the prevalence of pulpal calcifications and systemic diseases, as analyzed statistically, is shown in Table 4. The frequency of pulpal calcifications increased in elderly patients with periodontal disease. In the meantime, the frequency of pulpal calcifications was higher in patients with hypertension, nasopharyngeal carcinoma, cardiovascular disease, diabetes mellitus, and rheumatoid arthritis than in those without these symptoms.



**Figure 3** Radiographic examination revealing dystrophic calcification in coronal pulp corn of dental pulp.

Table 3	Distribution of PC by patient age.				
Age (y)	No. of patients	No. of patients with PC	Patients with PC (%)		
<20	4	3	75		
21-40	67	44	66		
41-60	20	13	65		
>60	106	62	58		
Total	197	122	62 (average)		
Chi-squar	e test		P = 0.729		
PC = pulpal calcification.					

Table 4 Distribu	tion of PC	by systemic disea	se.	
Systemic disease	No. of patients	No. of patients with PC	Patients with PC (%)	
Hypertension	37	23	75	
Nasopharynx carcinoma	22	19	86	
Cardiovascular disease	20	8	40	
Tuberculosis	18	10	56	
Diabetes mellitus	7	6	86	
Rheumatoid arthritis	4	3	75	
Normal	89	53	60	
Total	197	122	62	
Chi-square test			P = 0.059	
PC = pulpal calcification.				

#### **Discussion**

Calcified structures in the pulp have been described and studied for many years. Such calcifications are commonly found in the dental pulps of diseased, healthy, and even unerupted teeth. There is very little disagreement in the

literature that the presence of such calcifications in the dental pulp most often represents a dystrophic or retrograde change and is, therefore, a pathological manifestation. Hence, it is not surprising that it is generally accepted that the occurrence of pulpal calcifications is related to disturbances due to age, pathological changes in dentinal structures, dental trauma, or systemic disease. Although the reported occurrence of calcifications in the pulp varies from as low as 8% to as high as 90.0%, the figure most often quoted in textbooks is 66-90.0%, as provided by Hill<sup>14</sup> (Table 1). The occurrences are varied according to different methods. The occurrence, based on radiographic methods, has been reported to be around 20-25%, 2,3,34,35 while histological examinations yield a higher prevalence. 6,13,20 In some previous studies, the occurrence of pulpal calcifications assessed by histological examinations may be underestimated due to some technical difficulties in maintaining the integrity of the pulp tissue or its preservation. In those cases, small foci of calcifications could be missed. In addition, many prevalence studies have identified pulpal calcifications using radiographs. The variation is likely to be different because calcifications with a diameter smaller than 200 µm are not easy to detect on radiographic images. The occurrence of pulpal calcifications in the present study is 62% in histological examination and 30% in radiographic findings, indicating a significant difference between the two assessed techniques (Table 2). This finding is obviously higher than the results of similar studies. 2,3,13,21,22,24,27,29,32

Some studies report that age was not associated with pulpal calcifications.<sup>3</sup> Our present study agrees with this finding (Table 2). The specimens include an age group younger than 20 years to older than 70 years of age but no increase in the occurrence of calcifications in the pulps with increasing age because the prevalence in all ages shows no obviously distinct and significantly different (P = 0.729; Table 3). With more than half of the specimens (106/197) obtained from the group of patients older than 60 years of age, the incidence is 58% that combining all the specimens (91/197) for the younger than 60 years of age group, and the occurrence of pulpal calcification is 42%. However, some investigators reported that the frequency of pulpal calcifications increases in elderly patients with periodontal disease. 5,15,17,20 This study also demonstrated a high incidence of pulpal calcifications in periodontally involved teeth, but no significant difference was found.

The tendency to a higher frequency of occurrence of pulpal calcifications in molar teeth observed in this study also agreed with the results of most of the previous studies. <sup>2,3,36,37</sup> This present study shows and confirms a higher prevalence of pulpal calcifications in molar teeth (56%) compared with that in bicuspids (20%) and incisors (24%; Table 2). This may be related to the fact that molars have a large pulp chamber and space in the dentition, and have more time and chances to be injured and damaged during mastication and biting after eruption. It may, therefore, have reasons to induce more pulpal response and result in precipitation of calcifications in the pulp tissue. <sup>4,7,17,22</sup>

The correlation between pulpal calcifications and medical compromise disease has also been investigated in this study. Stafne and Sazbo<sup>24</sup> and Edds et al<sup>25</sup> found a significant relation between pulpal calcifications and the presence of

arteriosclerosis, osteitis deformans, and cardiovascular disease<sup>36</sup>; the results of the present study do not show such a correlation, but the incidence of pulpal calcifications was higher only in patients with nasopharynx carcinoma, diabetes mellitus, hypertension, and arthritis. This interesting correlation may be focused on vascular response with stimulation and require further investigation (Table 4).

In conclusion, we agree with the opinion that pulpal calcifications most commonly occur in the human dental pulp tissue; certain findings suggest that the prevalence of pulpal calcifications in periodontally involved teeth in a group of Taiwan Chinese population is 62% in histological and 31% in radiographic examinations. Both the occurrences are shown to be higher than those in other populations such as Iraqi, Jordanian, Australian, and Turks. 2,3,36,37 It would appear that pulpal calcifications are obviously a pathological manifestation, and may develop in different patterns, numbers, and sizes due to local or systemic conditions. 4,6,11,12,17,22,24,25 In addition, pulpal calcifications in molar teeth are significantly more common than in bicuspids and incisors. It is, therefore, recommended to pay more attention during routine endodontic therapy because root canal calcification will be a big challenge to handle and negotiate. Moreover, we look up the incidence of pulpal calcification associated with tissue inflammation that was believed playing a key factor during the formation of pulpal calcification. Further investigations focusing on the correlation between traumatic injury and dental pulp tissue are needed.

### Conflicts of interest

The authors declare no conflicts of interest.

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