# Chapter

# Frequency and Distribution of Pulpal Calcifications in Teeth Involved in Jaw Tumors

María Teresa Hernandez-Solis, Edith Lara-Carrillo, Víctor Hugo Toral-Rizo, Ronell Eduardo Bologna-Molina, José Edgar Garduño-Mejía, Norma Guadalupe Ibáñez-Mancera, Wael Hegazy-Hassan and Ana Miriam Santillán-Reyes

## Abstract

Pulp calcifications are idiopathic mineralized masses associated with irritation, age, trauma, and systemic or genetic diseases. The objective of this work was to examine frequency and distribution of pulp calcifications in teeth involved in jaw tumors, analyzing their relationship with age, sex, location, size, and diagnosis of the lesion in a sample of 21 teeth associated with tumors of the jaws. Imaging analysis included CT scans, periapical X-rays, and orthopantomography of the clinical record; histological analysis included pulp tissue fixed in 10% buffered formalin for 24 h, with hematoxylin and eosin staining, and examined under light microscope. A chi-square test was applied to associate calcifications with all variables. The tumor lesions were from patients aged 17-66 years. Calcifications were observed in 38.1% of cases on image and histologically in 76.2%; 56% were nodular and 68.8% were distributed in chamber and root canal. The male sex presented a higher frequency of pulp calcifications, estimating a statistically significant difference with respect to women (p = 0.004); there was no statistical significance with the other variables. In conclusion, the pulp tissue of teeth affected by maxillary tumors presents a percentage of pulp calcifications similar to the tissue where the periradicular tissue is intact.

Keywords: pulpal calcifications, jaw tumors, pulpal histology, dental pulp, pulp stones

# 1. Introduction

Pulpal calcifications are masses of mineralized tissue that can develop freely in any area of the pulp tissue or be found adhered at the interface of the dentin and pulp of healthy or diseased, erupted, or unerupted teeth [1]. Morphologically, they can be nodular, oval, needle-like, or irregularly shaped and are predominantly composed of minerals such as hydroxyapatite, aluminum, copper, iron, potassium, lead, and zinc [2–4].

Previously published studies have reported a prevalence of 8–90%, and they can be located both in the pulp chamber and in the root canal; they can be single or multiple, and in terms of size, they can be tiny below 200 µm or so large that they can obliterate the pulp chamber or root canal [5–7].

According to their topographic location, Satheeshkumar et al. [8] classified them as follows: single pulp calcification in the pulp chamber (Type I), multiple pulp calcifications present in the pulp chamber (Type IA), single pulp calcification present in the root canal (Type II), multiple pulp calcifications present in the root canal (Type IIA), multiple pulp calcifications present in pulp chamber and root canal (Type IIB), and continuous calcifications extending from the pulp chamber to the root canal (Type III).

The etiological factors of this process are still unknown; however, they are associated with different factors, such as trauma, pulp degeneration, orthodontic treatment, periodontal disease, caries, operative procedures, pulp inflammation, systemic diseases, or genetic conditions [9–13]. This process used to be associated with age; however, it has been observed that they are also found in young patients and in teeth that have not yet erupted [14].

Investigations regarding pulpal calcifications have been carried out over time with different methodologies based on imaging studies that include various techniques, such as periapical X-ray, panoramic X-ray, bitewing X-ray, cone beam tomography, or micro-CT and histological studies. These methodologies have been performed on teeth present in the mouth, extracted dental organs, or prehistoric skeletal remains [15–17].

Tumor lesions are abnormal tissue that grows in both the maxilla and mandible; they are relatively rare and affect soft and hard tissues and may extend to the facial region. Most of them are benign, but they tend to be aggressive and grow exponentially, displacing bone tables and thus the teeth [18].

The aim of this work was to show the frequency and distribution of pulp calcifications of teeth involved in tumor lesions of the jaws by means of imaging and histological analysis, analyzing whether there is any relationship between pulp calcifications and age, sex, location, size, and diagnosis of the lesion.

### 2. Materials and methods

For this purpose, an analytical, descriptive, and cross-sectional study was designed with nonprobabilistic sampling by convenience. The sample consisted of 21 caries-free teeth with intact enamel involved in eight tumor lesions in patients between 17 and 66 years old, which were removed by the therapeutic indication of the maxillofacial surgeon. All donors signed the informed consent, in which it was mentioned that the teeth would be used for scientific research and the tissue obtained from the surgical maneuver would be processed for histopathological diagnostic purposes. In addition, this project was approved and registered by the Research Ethics Committee of the Autonomous University of the State of Mexico (2021/P11).

Immediately after surgical excision, the tumor lesion was immersed in 10% buffered formalin for immersion fixation and transferred to the oral pathology laboratory at the School of Dentistry of the Autonomous University of the State of Mexico for histological processing. The teeth were separated from the tumor lesion with a scalpel blade no. 15 in soft tissue and flexible diamond disc (Plexoflex Fine Grain. DFS-Diamon, Riedenburg, Germany) when the alveolar bone was involved.

The teeth separated from the tumor lesion were kept in 10% buffered formalin while pulp tissue was obtained; this process was performed by marking with a flexible diamond disc (Plexoflex Fine Grain. DFS-Diamon. Riedenburg, Germany) and abundant irrigation along the longitudinal axis of the uniradicular teeth. For the multiradicular teeth, two marks were made, one in the crown and the other in the root zone, removing the hard tissues and obtaining the pulp tissue from the chamber and root canal.

Immediately after the pulp tissue was obtained, it was placed again in 10% buffered formalin for 24 h and then dehydrated in a series of alcohols to be included in a paraffin block and stained with hematoxylin and eosin. The slides were obtained with 5 µm thick tissue sections.

To perform the histological analysis and topographical location with the classification of Satheeshkumary et al. [8], the slides were observed using a Leica Microsystems DM750 microscope at a magnification of 10x. The analysis was performed by two examiners from the Endodontics and Oral Pathology areas. Imaging was performed with Motic, VM 3.0, Digital Slide Scanning System using the programs Phatomation PMA.start (Pathomation BV, Berchem, Belgium) and ImageJ-FIJI.

For the imaging analysis, panoramic radiographs, periapical radiographs, and tomographies were collected and included in the clinical records, which were reviewed to determine the absence or presence of calcifications.

Subsequently, the statistical analysis of the data was carried out using SPSS package version 26 (IBM Corp. Armonk, N.Y. USA). The descriptive statistical data of variables were obtained, and Pearson's chi-square test was performed to associate the variables (age, sex, diagnosis, location, and size of the lesion) with the presence of pulpal calcifications.

# 3. Results

The sample consisted of 21 teeth, seven incisors (33.3%), two canines (9.5%), five premolars (23.8%), and seven molars (33.3%). They were from eight tumor lesions with histopathological diagnosis: two ameloblastomas (25%), two keratocysts (25%), one central giant cell lesion (12.5%), one peripheral giant cell lesion (12.5%), one calcifying epithelial odontogenic tumor (12.5%), one calcifying epitheli

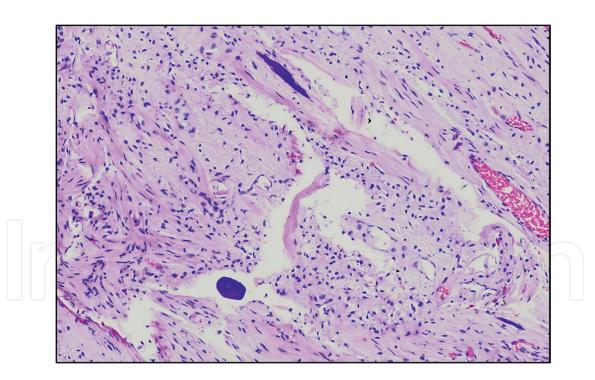
The donor patients were four women (50%) and four men (50%), ranging in age from 17 to 66 years, with a mean age of 34.75 years and a standard deviation of 20.76 years.

The location of the tumor lesions was 50% in the maxilla and 50% in the mandible, with a minimum size of 2 cm and a maximum of 15 cm, a mean of 7.43 cm, and a standard deviation of 4.04 cm. All teeth were analyzed together with their imaging studies included in the clinical records; 10 teeth were reviewed with tomography (46.6%), eight with panoramic radiographs (38.1%), and three with periapical radiographs (14.3%), observing calcifications in the imaging analysis in 38.1% of the cases (**Figure 1**). Sagittal view of a CT scan showing pulp calcification in the coronal area (white arrow) of a developing third molar associated with a keratocyst). Histological examination with hematoxylin and eosin revealed that pulp calcifications were present in 76.2% of the analyzed teeth. With respect to shape, 56.25% were only



#### Figure 1.

Sagittal view of a CT scan showing pulp calcification in the coronal area (white arrow) of a developing third molar associated with a keratocyst.



#### Figure 2.

Microphotograph of circular (nodule) and elongated pulp calcifications in the pulp tissue of a tooth associated with peripheral giant cell lesion (hematoxylin and eosin, 100× magnification, Motic VM 3.0. Digital Slide Scanning System).

nodules and 43.75% were nodules and needle-like in the pulp tissue itself (**Figure 2**). Microphotograph of circular (nodule) and elongated pulp calcifications in the pulp tissue of a tooth associated with peripheral giant cell lesion. Hematoxylin and eosin, 100× magnification, Motic VM 3.0. Digital Slide Scanning System).

Pulpal calcifications were distributed topographically according to the classification of Satheeshkumary et al. [8] as follows: 68.8% were in both the pulp chamber and root canal (Type IIB), 18.8% were multiple in the root canal (Type IIA) (**Figure 3**). Microphotograph of pulp tissue of a tooth involved in ameloblastoma, showing multiple calcifications of different sizes and shapes. Hematoxylin and eosin, 40× magnification, Motic VM 3.0. Digital Slide Scanning System), and 12.5% presented continuous calcifications from the crown to the root canal (Type III).

The presence of these calcifications was more frequent in males (87.5%) than in females (12.5%), showing a statistically significant difference with a value of p = 0.004.

In relation to age groups, more calcifications were observed in the 17- to 30-year-old group compared to the others, without showing statistically significant differences between groups (p = 0.028) (**Table 1**). It shows the frequency of pulpal calcifications in relation to age groups.

Pulp calcifications were found in 68.7% of mandibular teeth and 31.3% of maxillary teeth, but this difference was not statistically significant (p = 0.717).

Regarding the size of the tumor lesion and its relationship with calcifications, the 1 cm to 4 cm group had no calcified masses, while the 5 cm to 9 cm and 10 cm to 15 cm groups each had 8 teeth (50%); however, there was no significance between them (p = 0.131).

Regarding the histopathological diagnoses of the lesions and their relationship with calcifications, no significant association was found (p = 0.484), so the diagnosis



#### Figure 3.

Microphotograph of pulp tissue of a tooth involved in ameloblastoma, showing multiple calcifications of different sizes and shapes (hematoxylin and eosin, 40× magnification, Motic VM 3.0. Digital Slide Scanning System).

Age groups		17–30 years	31–59 years	60 + years	Total
Calcifications	Absent	2	2	1	5
-	Present	12	0	4	16
Total		14	2	5	21

Table 1.

Frequency of pulp calcifications in relation to age groups.

Diagnostic	Peripheral ossifying fibroma	Ameloblastoma	Peripheral giant cell lesion	Central giant cell lesion	Keratocyst	Total
Calcification _	1	3	0	0	1	5
	1	8	3	3	1	16
Total	2	11	3	3	2	21

Table 2.

Frequency of pulp calcifications in relation to histopathological diagnoses of tumor lesions.

does not determine the existence or absence of calcific precipitations (**Table 2**). It shows the frequency of pulp calcifications in relation to histopathological diagnoses of tumor lesions.

## 4. Discussion

The study of pulp calcifications has led to development research to determine the prevalence, distribution, and association with diseases such as caries, periodontal disease, or genetic conditions. However, in the literature review, no previous data were found on the study of pulp calcifications in teeth involved in maxillary tumors, so one of the objectives of this study was to publicize and disseminate the results, adding knowledge to this area.

Pulp calcifications can be detected through diagnostic means such as imaging and histological studies. Imaging studies are carried out by panoramic radiographs, periapical, bitewing, cone beam tomography, or micro-CT, which allow locating radiopaque nodules with a size greater than 200  $\mu$ m since below this size they are not detectable by these means. For histological studies, they allow observing calcifications of different sizes and locations, since the lamellae are observed under the microscope with different magnifications, allowing the localization of these in any path of the pulp tissue.

Regarding the prevalence of pulp calcifications, previous studies have shown a range from 9.6 to 95% [3, 16]. These percentages vary due to the methodologies used for each investigation; usually, imaging studies show a lower frequency than histological studies since the lamellae are seen under the microscope, identifying any size of calcification. This finding agrees with our results and those of Huang et al. [19] since both studies performed imaging and histological analyses and observed similar results. Huang et al. [19] showed the presence of calcifications of 62% in the histological area and 30% in the radiological area, while our results were 76.2% in the histological area and 38.1% in the imaging area.

Histological studies allow a greater morphological appreciation of pulp calcifications, and we agree with Milcent et al. [2] that pulp calcifications are morphologically heterogeneous since they are different from each other in shape and size.

Another important factor is that the prevalence of calcifications in pulp tissue may be different depending on the ethnic group. For example, in Mexican patients, they have been observed in 84% [20], Peruvians 83.58% [21], Taiwanese 83.3% [22], Iranians 76% [23], Argentines 55% [24], Jordanians 51.4% [25], Turks 38% [26], and Yemenis 3.99% [13].

Sener et al. [24] and Ranjitkar et al. [1] performed the analysis of pulp calcifications with bitewing radiographs, finding a prevalence of 38% and 46.1%, respectively. In the present study, periapical, panoramic, and tomography radiographs were examined analyzing the coronal and radicular areas, and the percentage of calcifications was similar at 38.1%.

The frequency of pulp calcifications in males was higher than in females, which agrees with Hsieh et al. [23]. The possible cause is that men present greater dental attrition in relation to women, and the pressure exerted by the tumor in the apical area allows for premature contact points, which provides a local irritant in the pulp tissue.

Another interesting fact is that pulpal calcifications have been related to advanced age [9]. In contrast, in this study, the age group 17–30 years old was the one with the highest percentage of calcifications.

Mandibular teeth had a higher percentage of pulp calcifications, which agrees with Olivares et al. [20]. However, there are also studies where the maxillary dental organs are the predominant ones without a significant difference [24].

Regarding the size of the lesion and the histopathological diagnosis, no similar antecedents were found in the literature since these variables have not been investigated in pulp tissue under these conditions. Therefore, the present study generates knowledge that can be used as a basis for future lines of research.

However, one of the limitations of the study is the small size of the sample, since being a histological study, the samples of the tumor lesions should present dental organs, and these should be kept in an adequate fixation medium for the preservation of the pulp tissue, which is not frequent to find or to obtain them in the correct preservation medium.

# 5. Conclusion

In conclusion, the pulp tissue of teeth affected by maxillary tumors presents a similar percentage of pulp calcifications to studies where the periradicular tissue does not present any alteration.

Another important fact is that there was no statistically significant relationship between pulp calcifications and tumor size or histopathological diagnosis.

The results of this study may be useful as a basis for future lines of research since they broaden the knowledge of pulp calcifications and histological characteristics of pulp tissue of teeth in the presence of maxillary tumors. For future studies, it would be convenient to analyze this finding in a larger sample.

# **Conflict of interest**

The authors declare no conflicts of interest.

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# Author details

María Teresa Hernandez-Solis<sup>1</sup>, Edith Lara-Carrillo<sup>2\*</sup>, Víctor Hugo Toral-Rizo<sup>3</sup>, Ronell Eduardo Bologna-Molina<sup>4</sup>, José Edgar Garduño-Mejía<sup>5</sup>, Norma Guadalupe Ibáñez-Mancera<sup>6</sup>, Wael Hegazy-Hassan<sup>2</sup> and Ana Miriam Santillán-Reyes<sup>2</sup>

1 Faculty of Dentistry, Doctoral Program in Health Sciences, Autonomous University of the State of Mexico, Toluca, Mexico

2 Center for Research and Advanced Studies in Dentistry "Dr. Keisaburo Miyata" Faculty of Dentistry, Autonomous University of the State of Mexico, Toluca, Mexico

3 Faculty of Dentistry, Orocentro Clinic, Autonomous University of the State of Mexico, Toluca, Mexico

4 Faculty of Dentistry, Center for Stomatological Molecular Pathology, Republic University, Montevideo, Uruguay

5 Department of Maxillofacial Surgery, Regional Hospital "Lic. Adolfo López Mateos", Toluca, Mexico

6 The Interdisciplinary Center of Health Sciences UST, Instituto Politécnico Nacional, Mexico City, Mexico

\*Address all correspondence to: laracaedith@hotmail.com

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