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Sialolithiasis: mineralogical composition, crystalline structure, calculus site, and epidemiological features

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Abstract

The purpose of this paper was to describe the characteristics of salivary calculi and their relationship to epidemiological factors, through a cross-sectional study. We analysed 100 calculi obtained in 2017–2021. Patient data including age, time since onset of symptoms, gland involved, and site of location in the salivary system were studied. The calculi were studied to determine their morphological features using scanning electron microscopy and energy dispersive plain radiographic analysis. Most of the calculi had formed in the submandibular gland (SG) (82%). The mean age of patients at onset was 45.83 years; patients presenting parotid gland (PG) stones were somewhat older ($p = 0.031$). The mean time since the onset of symptoms was longer in PG calculi ($p = 0.038$). The most common lithiasis site was the main duct (74%), followed by the hilum (22%). Hilar stones were the largest ($p < 0.05$) and heaviest ($p = 0.028$). Octacalcium phosphate (OCP) was the most common crystalline phase (Cp) founded, followed by hydroxyapatite (HA) and whitlockite (WH). Specifically, OCP had a higher presence in PG calculi ($p = 0.029$) and WH was the most common phase in SG calculi ($p = 0.017$). The most prevalent site of lithiasis was the main duct, and the largest and heaviest calculi were found in the SG. PG stones were associated with a longer history of symptoms and older age. OCP was the most frequent Cp of the calculi studied, and the main Cp in PG stones. WH was the predominant Cp in SG stones. The Cp of the calculi was not influenced by location, patient age, or time of symptoms.

Keywords: Salivary stones; Lithiasis; Sialendoscopy; Crystallography; Phosphates

Introduction

Sialolithiasis is considered one of the primary causes of chronic obstructive sialadenitis,[1] and postmortem studies of this disease have reported a prevalence of 1.2%[2] and an incidence of 2.9–5.5 cases/100,000 of the population.[3]

A wide majority of these calculi (80–90%) involve the submandibular gland (SG).[4] Detailed understanding of how these salivary stones form is lacking. It is likely that microscopic concretions, known as sialomicroliths,[5] accumulate during normal activity of the salivary gland, favouring the creation of inflammatory foci,[6] which can be either primary or caused by retrograde migration of oral bacteria.[7] Sialomicroliths are microscopic mineral masses that contain calcium and phosphorus as well as organic material and necrotic cell residues.[8,9]

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It appears that sialomicroliths tend to develop more in the SG,[10] and their size depends on their location in the salivary ductal system.

Submandibular gland calculi are made of 70–80% inorganic material, as compared to 50% in parotid gland (PG) stones.[11] Their inorganic component appears to be proportional to the size of the stone and they tend to be composed of calcium and phosphate in different forms.

It is therefore necessary to describe the composition and morphology of salivary calculi and their relationship to epidemiological factors that may predispose patients to the disease, thus aiding prevention and early diagnosis.

Material and methods

Sialolith retrieval

We analysed salivary calculi retrieved in the ENT and Cervicofacial Surgery Department of the Fundación Jiménez Díaz University Hospital and Villalba General University Hospital. Calculi were obtained via sialendoscopy, direct extraction, or spontaneous extrusion. Patient data were recorded, including gender, age, time of symptoms, gland involved, lithiasis site within the main salivary duct, and the method used to obtain the sialolith.

The study was approved by the Fundación Jiménez Díaz University Hospital Ethics Committee (FJD-SAC-16-01), and all patients were informed of the study and signed a specific informed consent form permitting subsequent analysis of their data.

Morphology and composition (chemical and mineral)

Once the calculi were retrieved, the samples were sent to the Geological Techniques Unit (Research Support Centre for the Earth Sciences and Archeometry) of Universidad Complutense de Madrid for study. Prior to analysis, all stones were immersed in hydrogen peroxide for 48 hours to eliminate any residual blood or organic material that could alter the results of the analysis. Subsequently, the stones were washed with the Millipore Milli-Q system and left to air-dry. Once dry, the stones were weighed using a precision scale and measured with a calliper. The stones were described using a stereoscopic microscope and a JEOL JSM-820 (JEOL Ltd) scanning electron microscope was used. The microscope was outfitted with a detector of dispersive plain radiograph diffraction for chemical analysis.

The mineral composition of the stones was studied using plain radiograph powder diffraction (Bruker D8 ADVANCE diffractometer) with a Cu Kα2 wavelength at an angle interval of 2° to 65° 2θ, a step size of 0.02°, and scan step time of 1 s. For this study, samples were dehydrated for 24 hours at 45 °C with a stove and then hand-ground using an agate mortar and pestle to prevent contamination. Semiquantitative analysis of the crystalline and amorphous structures was performed using the XPowder software program.[12,13]

Statistical analysis

First, a histogram for each of the study variables found that none followed a normal distribution; as a result, we decided to use nonparametric statistical methods and take the median rather than the mean as the measure of central tendency. Data for the variables age and time since onset of symptoms were grouped to facilitate analysis. Data on patient age were divided into 10-year groups, and times since the onset of symptoms were grouped by three-month periods.

Statistical analysis of crystalline phase (Cp) was performed using the Mann-Whitney U test to study this variable and its relationship with the gland involved and the sex of patients. To avoid loss of statistical power, the Kruskall-Wallis test with Bonferroni correction was used to analyse the site of the calculi within the salivary duct, the time since the onset of symptoms, and patient age.

Correlations between the study variables were analysed with the Spearman correlation coefficient to establish associations between two nonparametric, continuous quantitative samples.

Results

One hundred calculi obtained from 95 patients were analysed (51 male, 44 female). Five patients developed simultaneous calculi in different salivary glands or in two sites within the same salivary gland, with no remarkable differences in the characteristics of these stones. Patient sex did not influence the gland involved ($p = 0.297$).

Most calculi were obtained through sialendoscopy, using either a simple or combined approach (90%, n = 90). The remaining calculi were obtained by means of spontaneous extrusion (9%, n = 9) or submandibulectomy (1%, n = 1).

Epidemiological data

Most of the stones originated in the SG (82%, n = 82), and 18% had formed in the PG (18%, n = 18).

The mean (range) patient age at the onset of symptoms was 45.83 (6–88) years. Patients were grouped by age into either child (<18 years) or adult (≥18 years) patients: four calculi were studied in children, and their mean (range) age was 10 (6–14) years; 96 calculi were retrieved from adult patients, who had a mean (range) age of 47.72 (20–88) years. The age of patients with stones in the PG was significantly older (mean 53.7 years) than patients with SG stones (mean 44.1 years) ($p = 0.031$).

The median length of symptoms of sialadenitis was 24 months. The time since the onset of symptoms was significantly higher among patients with calculi in the PG (median 53.8 months) than those with stones in the SG (median 37.9 months) ($p = 0.038$).

All epidemiological data is detailed in Supplemental Table 1 (online only).

Sialolith location in the salivary ductal system

Overall, the majority of calculi were found in the ducts (74%, n = 74) followed by the hilar region (22%, n = 22) and the intraglandular ducts (4%, n = 4). A similar result was obtained when stratifying by gland, and no statistically significant differences were found. Among calculi located in the SG, 73.2% were in the main duct and 24.4% were in the hilar region. Of PG stones, 77.8% were located in the main duct and 11.1% were in the hilar region, respectively.

Size and weight

Mean size was 5.44 mm and the median size was 4.45 mm (range 0.75–65.6 mm). The mean calculus weight was 119.84 mg; the median (range) weight was 48.4 (1–1196.3) mg.

A significant relationship ($p < 0.001$) was found between the size and weight of the calculi, with strong correlation (0.828), indicating that larger calculi were heavier.

Overall, significant differences were found for stone size ($p = 0.001$) and weight ($p = 0.019$) depending on the location of these stones in the salivary system (Table 1). When Bonferroni correction was applied, we confirmed that stones in the hilar region were larger than those of the salivary ($p = 0.004$) and intraglandular ducts ($p = 0.016$). As for weight, Bonferroni correction only confirmed that stones in the hilar region had a significantly greater weight than those of the salivary ducts ($p = 0.028$).

When stratified by weight and size and by gland involved and site of the lithiasis, the only significant differences revealed were the size of calculi found in the SG, as calculi of the hilar region had a significantly larger median size than all other locations ($p = 0.016$) (Supplemental Table 2, online only).

Morphology and mineral composition

The morphological study confirmed the presence of microstructures in which carbon and sulphur were detected in the interior and calcium phosphate was present on the surface. These formed concentric laminated layers with a botryoidal surface (Fig. 1A).

The most common Cp found were octacalcium phosphate (OCP) (Fig. 2), hydroxyapatite (HA) (Fig. 1B), whitlockite (WH) (Fig. 3), brushite (BT), and amorphous variants (AM). The median percentage values for these compositions were 36.18% OCP, 34.15% HA, 0% WH, 0.0% BT, and 12.80% AM.

OCP was the most commonly found Cp overall. Of note, this phase was more present in PG calculi (median 52% and mean of 644.03 in the PG vs a median of 32% and mean of 47.53 in the SG) ($p = 0.029$).

Additionally, WH was significantly more present in the SG ($p = 0.017$) (Table 2), with a median of 10.5% and a mean of 53.51% for the SG, compared to a median of 0% and an mean of 36.78% for PG calculi.

No significant relationship was found between the Cp identified and the site of the lithiasis within the salivary duct at the time of retrieval (Supplemental Table 3, online only). A p value of 0.046 was found in the case of OCP which, after applying Bonferroni correction to adjust the statistical significance, resulted in a p value of 0.056. Similarly, when each Cp was stratified according to gland and site, no statistically significant relationship was found.

Patient sex (Supplemental Table 4, online only) and age did not significantly influence the median composition of the stones studied.

The time since symptom onset did not determine the Cp of the calculi. An initial analysis revealed that the median presence of HA varied depending on the time since the onset of symptoms ($p = 0.042$). However, this association was no longer statistically significant once Bonferroni correction was applied ($p = 0.6$).

Table 1
Median sialolith size and weight according to specific location (Kruskal-Wallis test for independent samples).

Variable	Site	Median	p value
Size (mm)	Salivary ducts	4.2	0.001
	Hilar region	7	
	Intraglandular ducts	2.75	
Weight (mg)	Salivary ducts	44.6	0.019
	Hilar region	130.5	
	Intraglandular ducts	22.1	

Discussion

Our series (n = 100) is one of the largest in the published literature, and this study provides a detailed analysis of these salivary calculi and the statistical relationship between their crystalline phase, the gland involved, and lithiasis site, in addition to epidemiological factors such as patient sex, age, and time since the onset of symptoms.

Limitations of our study include that samples were obtained from two hospital centres and that it mostly draws on data from patients who underwent sialendoscopy. Despite this limitation, the characteristics of this population resemble those published previously, that is, absence of a sex-based difference,[14,15] lower than 5% presence of stones in patients of paediatric age,[14,16] mean patient age of 47.7 years,[17] and a mean time since the onset of symptoms of 24 months.[14]

In our series, most of the calculi originated in the SG (82%). Overall, the predominant sialolith site was the main duct (74%), followed by the hilar region (22%). When we stratified our data by the gland involved, as in earlier series,[14,18] we found that most PG calculi (77.8%) had formed in the main duct. However, the most common site of SG stones was the main duct (73.2%), thus contrasting with the findings of other publications,[14,18] in which the most prevalent site of these calculi was the hilar region.

As in other published series,[14,17] patients with calculi formed in the PG were older on average than those patients in whom the SG was the site of formation (53.7 years PG

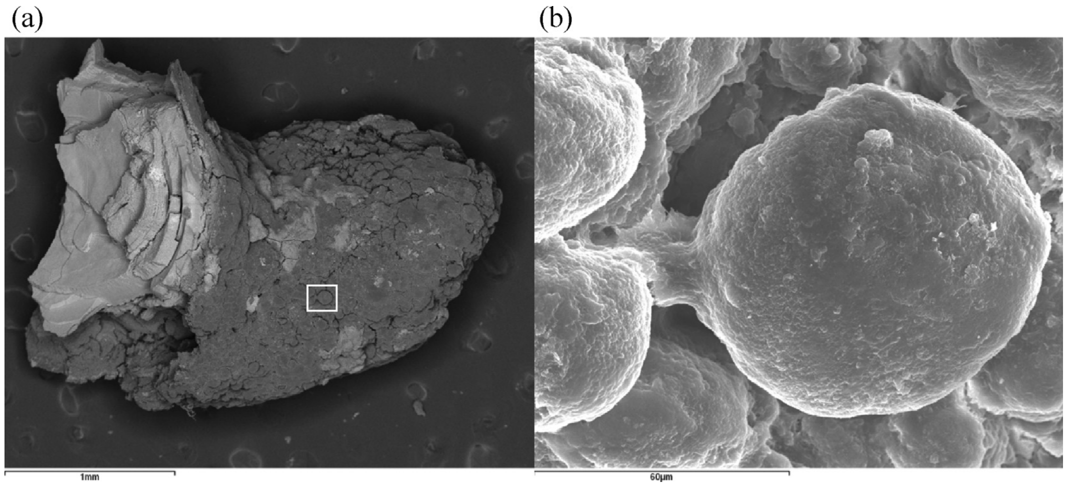

Fig. 1. A–B. Scanning electron microscopic image of a ductal submandibular sialolith. On the left (original magnification × 40), concentric laminated layers with a botryoidal surface are evident. On the right, a detailed view of hydroxyapatite (magnification × 200).

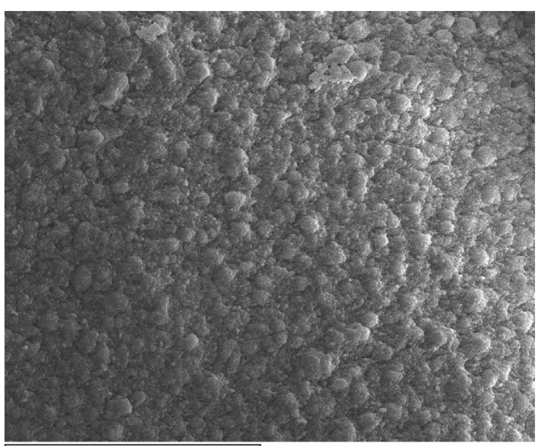

Fig. 2. Scanning electron microscopic image (original magnification × 1000) of the surface of the inner nucleus of a submaxillary salivary duct stone in the hilar region, composed of microscopic mineral masses compatible with octacalcium phosphate.

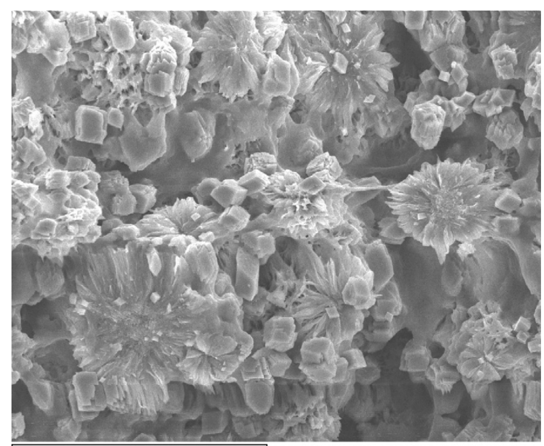

Fig. 3. Scanning electron microscopic image (original magnification × 1000) of the internal nucleus of a submaxillary salivary duct stone. Rhombohedral crystals having a mineral composition compatible with whitlockite and radial aggregates with a crystalising structure compatible with hydroxyapatite.

vs 44.1 years SG) ($p = 0.031$). Similarly, patients with PG calculi had a significantly longer history of symptoms (median 53.8 months) than those with SG calculi (median 37.9 months) ($p = 0.038$), a finding consistent with the report by McGurk et al.[17]

Overall, SG stones had a significantly larger median size ($p = 0.034$) and weight ($p = 0.004$) than PG calculi; other authors[14] have published similar findings. Sigismund et al,[14] in their retrospective study of 2,959 calculi studied using ultrasound scan, found that most SG calculi had a size of >5 mm, as compared with ≤5 mm among PG stones. These results closely match those from our series, in which the median weight was significantly larger in the submaxillary gland (5 mm vs 3.75 mm, $p = 0.034$). This difference in size was likely related to the larger diameter of these stones relative to the duct where they were retrieved (SG).[11] Hilar stones had the largest size ($p = 0.001$) and weight ($p = 0.028$). As expected, sialolith weight was directly proportional to calculus size ($p < 0.001$).

There are few reports containing a detailed analysis of the crystalline structure evaluated in terms of its relationship with epidemiological factors, and those that do exist tend to have a sample size of lower than 40.[19,20] Most studies[19–22] have found that HA is the most common Cp overall. In contrast, we found that the most common Cp among all calculi was OCP (median 36.18%), followed by HA, WH, BT, and AM. Octacalcium phosphate was found in a significantly higher proportion in PG calculi ($p = 0.029$), and WH was the most common phase in stones of the submaxillary gland ($p = 0.017$). The Cp of the calculi was not influenced by the site of the calculi of the stone in the salivary duct, patient age or, or the time since the onset of symptoms. Future studies might be interested in analysing the relationship between the organic and inorganic component found

Table 2
Median values, expressed as percentage of mineral composition, indicating the presence of different crystalline phases identified per gland involved (Mann-Whitney U test for independent samples). Median composition is in percentage.

Crystalline phase	Gland	Median composition	p value
Octacalcium phosphate	Submandibular	32	0.029
	Parotid	52	
Hydroxyapatite	Submandibular	35	0.757
	Parotid	29.5	
Whitlockite	Submandibular	10.5	0.017
	Parotid	0	
Brushite	Submandibular	0	0.756
	Parotid	0	
Amorphous	Submandibular	13	0.412
	Parotid	13	

in the residual organic component; this could help clarify the genesis and the growth of the stones.

The fact that a wide majority of calculi were retrieved with the use of sialendoscopy indicates the state of development and widespread use of this minimally invasive, conservative technique used in the salivary gland for the treatment of chronic obstructive sialadenitis.

Conclusions

The salivary gland calculi in our series presented with similar frequency in men and women, and the most common site was the SG. These masses are a rare finding in children and tend to manifest symptomatically in the fifth decade of life. The most prevalent location for lithiasis was the main duct, followed by the hilar region; the largest and heaviest calculi were found in the SG. Parotid gland stones were associated with a longer history of symptoms and older age.

The two most common crystalline phases observed were OCP and HA; and OCP was the most common crystalline structure in PG stones, while WH was the predominant phase in stones of the SG. The Cp of the calculi was not influenced by the site, patient age, or the time since the onset of symptoms.

Conflict of interest

We have no conflicts of interest.

Ethics statement/confirmation of patient permission

The study was approved by the Fundación Jiménez Díaz University Hospital Ethics Committee (FJD-SAC-16-01), and all patients were informed of the study and signed a specific informed consent form permitting subsequent analysis of their data.

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Appendix A. Supplementary material

Supplementary data to this article can be found online at https://doi.org/10.1016/j.bjoms.2022.08.005.

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