

“True” Versus “Bay” Apical Cysts: Clinical, Radiographic, Histopathologic, and Histobacteriologic Features



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ABSTRACT

Introduction: This study compared the main clinical, radiographic, and histologic features of true and bay apical cysts. **Methods:** The study material comprised 95 biopsy specimens of apical periodontitis lesions obtained attached to the root tip of both untreated and root canal-treated teeth. Clinical and radiographic data were recorded. Specimens were obtained by extraction or periradicular surgery and were meticulously processed for histopathologic and histobacteriologic methods. All cases diagnosed as apical cysts ($n = 23$) were divided into the true and bay types, which were then compared for tooth location, patient's sex, lesion size, severity of clinical symptoms, presence of a sinus tract, previous abscess episodes, and prevalence of bacteria in the main root canal lumen and ramifications, on the outer root surface, and within the cyst cavity. **Results:** Eleven specimens were classified as true (48%) and 12 (52%) as bay cysts. Bacteria were found in all specimens, regardless of the histopathologic diagnosis. Planktonic bacteria were observed in the main root canal in all true cysts and in 11 of 12 (92%) bay cyst cases. Biofilms were detected in the main canal in 10 cases from each diagnostic group and were frequently observed in ramifications. Extraradicular biofilms occurred in a few specimens only. Bacteria were visualized within the cavity of both true (4/11, 36%) and bay (6/12, 50%) cyst specimens. The severity of histologic inflammation was always high. There were no significant differences between true and bay cysts for all the clinical, radiographic, histopathologic, and histobacteriologic parameters assessed. **Conclusions:** Except for the morphologic relationship of the cyst cavity with the root canal space, true and bay cysts exhibited no other significant differences in the various parameters evaluated. The 2 cyst types were always associated with an intraradicular infection and sometimes with an extraradicular infection. Findings question the need to differentiate true and bay cysts and do not support the assumption that true cysts are self-sustainable entities not maintained by infection. (*J Endod* 2020;46:1217–1227.)

KEY WORDS

Apical periodontitis; bay apical cyst; biofilm; endodontic infection; true apical cyst

In response to root canal infection, the periradicular tissues mount an immune reaction that may give rise to bone resorption and granuloma formation¹. With the passage of time, the lesion may become epithelialized as the epithelial cell rests of Malassez start to proliferate in the granuloma, and, ultimately, a cavity lined by an epithelium is formed, which characterizes the apical cyst. The lumen of the apical cyst cavity is usually lined by a stratified squamous epithelium, although in about 8% of the apical cysts the cavity may be partially or predominantly lined by ciliated columnar cells of respiratory origin^{2,3}. Four theories have tried to explain the genesis of the apical cyst cavity, including the breakdown theory⁴, the abscess theory^{5,6}, the immunologic theory⁷, and the trapped connective tissue theory⁸, but none of them has been clearly demonstrated to be true.

Numerous studies have evaluated the prevalence of apical cysts among periradicular lesions. Apical granuloma is the most common histopathologic form of apical periodontitis in the large majority of studies, and the prevalence of cysts ranges from 6%–55%^{9–16}. Cysts and granulomas cannot be distinguished by radiographic examination alone^{9,13,14}, although large lesions are more likely to be cysts^{17,18}. Although some studies have suggested that cysts can be differentiated from granulomas by

SIGNIFICANCE

This study found no differences between true and bay cysts concerning clinical and histopathologic manifestations. Both types always exhibited intraradicular and sometimes extraradicular infection. Findings do not support the assumption that true cysts are self-sustainable entities not associated with infection.

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other approaches, including polyacrylamide gel electrophoresis of the periapical fluid, tomography, and ultrasound real-time imaging^{18–21}, a definitive diagnostic differentiation can only be attained by histopathology, especially using a serial sectioning approach to include the entire lesion. Most of the studies that evaluated the cyst prevalence did not perform serial sectioning, and this may have compromised the results and help explain the wide range reported. The few studies that used serial sectioning or serial step sectioning showed a cyst prevalence of 15%–32% of the apical periodontitis lesions^{3,10,22}.

Depending on the relationship of the cyst cavity with the root canal via the apical foramen, the apical cyst has been classified as a “true” or “bay” (also known as a “pocket”) cyst^{22,23}. The lumen of the bay cyst cavity communicates directly with the root canal system through the apical foramen with the root apex protruding into the cavity, whereas the true cyst has a completely independent cavity with no continuity or connection to the root canal. Simon²² used the serial sectioning approach to evaluate apical periodontitis lesions in untreated teeth that remained attached to the root apices after extraction and reported that true and bay cysts occurred in similar prevalences (ie, 9% of the lesions). Therefore, true and bay cysts each corresponded to 50% of the apical cysts. Another study using serial sections or serial step sections of lesions adhered to the apices reported that 9% were true cysts and 6% were bay cysts¹⁰. Of the apical cysts, 61.5% were true, and 38.5% were bay cysts. The study by Ricucci et al³ was another one that evaluated serial sections from whole lesion specimens attached to the apices of untreated extracted teeth. Although 42% of the lesions showed an epithelium, the frequency of cysts was 32%. True cysts occurred in 16% of the lesions, whereas bay cysts corresponded to 18%. Of the apical cysts, 50% were true, and 56% were bay cysts (1 lesion contained the 2 types).

It was proposed in a study of untreated teeth²² and was reinforced by only 1 case report²⁴ that the true cyst, assumed to be a self-sustaining lesion, cannot heal after nonsurgical root canal treatment, whereas bay cysts, especially the smaller lesions, can^{10,22}. The rationale provided was that the bay cyst is open to the root canal and then amenable to intracanal infection control, whereas the true cyst represents a disease entity no more dependent on the root canal infection and then not responsive to root canal treatment^{22,23}. This theory has been strongly questioned because it does not exhibit biological plausibility^{8,25}. Siqueira²⁶ raised the point that

the bay cyst may be more prone to become infected by bacteria advancing directly from the canal into the cyst cavity, which might impair healing even after proper intracanal antimicrobial treatment. Indeed, an infected bay cyst has been reported as the cause of nonsurgical treatment failure²⁷.

Accurate histopathologic diagnosis of apical periodontitis lesions is reliant on serial sectioning evaluation of a biopsy specimen that represents the entire lesion. Histopathology of a limited number of sections may be confusing and may make the operator erroneously classify an epithelialized granuloma as a cyst or a bay cyst as a true cyst. In addition, a proper differentiation between true and bay cysts can only be made if the lesion specimen remains attached to the root apex obtained by periradicular surgery or extraction because the continuity of the cyst cavity with the root canal is essential information. Comparisons of the histopathologic features of the 2 conditions are limited in the literature, and no study has so far evaluated the histobacteriology of true and bay cysts.

The present study used meticulous serial sectioning and histopathologic/histobacteriologic evaluations to compare the main features of true and bay cysts that might justify the alleged different biological behavior between them after root canal treatment.

MATERIALS AND METHODS

Clinical Specimens

The study material was composed of 95 human biopsies of apical periodontitis lesions obtained attached to the root tip of both untreated and root canal-treated teeth. The specimens were obtained by endodontic specialists or oral surgeons through periradicular surgery or extraction in private dental practices and dental schools and sent consecutively over a period of 12 years to a single histologic laboratory. At the time of treatment, the patients were presented with risks, benefits, treatments, and options and had given consent for examination of their teeth. The patients' mean age was 37.3 years (range, 15–82 years). The protocol for this retrospective study was approved by the institutional review board.

Clinical and radiographic data were available for all teeth. Symptoms were categorized as follows:

1. absent (asymptomatic), when the patient reported no pain episodes and the tooth was comfortable with normal response to vertical/lateral percussion and periapical palpation;

2. mild, when the patient reported no episodes of spontaneous pain and no self-medication with analgesics, but the tooth was slightly tender to chewing and pressure;
3. moderate, when the patient declared episodes of spontaneous pain and self-medication with analgesics, which succeeded in resolving pain, and exhibited tenderness to percussion/palpation; and
4. severe, when there was excruciating pain, not resolved with analgesics, associated with a painful response to percussion/palpation.

Information on the occurrence of acute abscess episodes related to the affected tooth any time before surgery/extraction was also available. These cases had been diagnosed by the clinician based on the development of swelling and redness of the skin associated with pain that prompted the patient to seek professional aid. Some patients with abscesses had been treated with antibiotics.

The radiographic lesion size was determined as the mean diameter of the periapical radiolucency and categorized as small if they were ≤ 5 mm and large if they were > 5 mm. Teeth with periodontal pockets communicating with the periapical lesion or teeth with vertical fractures were excluded from the study.

To be included in the study, the specimen obtained by extraction or surgery should have consisted of the entire apical periodontitis lesion still adhered to the root apex. Periradicular surgery was performed as follows. After elevation of a full-thickness periosteal flap, the buccal bone covering the lesion was carefully removed until the pathologic tissue and the root tip were exposed. The root tip was first resected approximately 3 mm short of the apex with a fissure bur. Subsequently the soft tissue was carefully enucleated from the bone crypt with smooth microelevators, in an attempt to obtain the resected root tip and the surrounding pathologic soft tissue in one piece.

The teeth were processed for light microscopy. From this pool, only specimens in which a cyst cavity was detected in the histologic sections (23 cases) were selected for the present study.

Histopathologic and Histobacteriologic Analyses

Specimens were fixed in 10% buffered formalin for at least 48 hours. Demineralization was performed in a solution of 22.5% (vol/vol) formic acid and 10% (wt/vol) sodium citrate for a period of 3–4 weeks. The end point was determined radiographically. For the

specimens obtained by extraction, the apical 4-to 5-mm segment of the root was separated with a sharp razor blade. At the end of the demineralization process, specimens were washed in running water for 24 hours and dehydrated in ascending grades of ethanol (50%–100%). After clearing in xylene, they were infiltrated and embedded in paraffin. Next, the biopsies were oriented parallel to the long axis of the main root canal in the apical third in order to obtain sections with the main canal and periapical tissue in direct continuity. Serial sections were taken with the microtome set at 4–5 μ m. Every fifth slide was stained with hematoxylin-eosin for screening purposes in order to locate the areas with the most severe reactions. Additional slides were stained as needed. Selected slides were stained with Masson trichrome to identify collagen and with the Taylor modification of the Brown-Brenn stain for the presence of bacteria²⁸.

Apical periodontitis lesions were classified according to agreed histomorphologic criteria into granulomas, abscesses, and cysts^{29,30}. The diagnosis of a cyst was made when a distinct cavity lined by epithelium and filled with semisolid material was observed. Depending on the relationship between the epithelial lining of the cyst cavity and the root and between the cyst cavity and the root canal space, cystic lesions were differentiated into “true” or “bay” cysts²². The lesions classified as true cysts were characterized by the presence of a cavity bordered by an epithelial wall that was not continuous with the canal lumen in any of the serial histologic sections. The lesions classified as bay cysts showed a cystic space surrounded by an epithelial wall that joined the external root surface forming a “sac,” isolating the foramen from the rest of the lesion. The bay cyst cavity had a direct opening into the canal lumen.

In the 23 cases diagnosed as apical cysts (true or bay), the following aspects were specifically looked for in the histopathologic and histobacteriologic analyses (Table 1):

1. The presence, location, and arrangement of bacteria (planktonic or biofilm structures) in the apical portion of the root canal system, including the main canal lumen and walls and apical ramifications (intraradicular infection). The parameter used for biofilm classification was as defined elsewhere as follows: “populations of microorganisms that are concentrated at an interface and typically surrounded by an extracellular polymeric substance matrix”³¹.
2. The presence of bacterial aggregations adhering to the outer apical root surface (extraradicular biofilm) eventually

presenting patterns of calcification (calculuslike structures)

3. The presence of bacteria/bacterial aggregations within the cyst lumen. If present, occasional bacterial cells or aggregations observed on the peripheral collagenous surface of the periapical lesion and not surrounded by an inflammatory infiltrate were regarded as contaminants, possibly resulting from the passage of the specimen through the socket during extraction; as such, they were excluded from evaluation.
4. The intensity of histologic inflammation was ranked as none; mild, when limited aggregations of exclusively chronic inflammatory cells were seen in the cyst lumen and/or a few chronic inflammatory cells infiltrated the subepithelial connective tissue; moderate, when discrete accumulations of chronic inflammatory cells occurred in the cyst lumen and epithelial walls with some occasional polymorphonuclear leukocytes (PMNs); or severe, when large accumulations of acute and chronic inflammatory cells with a prevalence of PMNs were evident in the cyst lumen intermixed with necrotic debris or cholesterol crystals or in the cyst walls, heavily infiltrating the epithelium.

Statistical Analysis

Statistical analysis was performed to evaluate if there were differences between true and bay cysts regarding tooth location (maxillary or mandibular); patient’s sex; lesion size (small or large); severity of clinical symptoms (severe or not); presence of a sinus tract; history of a previous acute abscess; and prevalence of planktonic bacteria or bacterial biofilms in the main canal lumen, biofilms in ramifications, extraradicular biofilm, and bacteria in the cyst lumen. The Fisher exact test was used for all analyses with the level of significance set at 5% ($P < .05$).

RESULTS

Twenty-three cystic lesions were obtained from 21 patients (11 men and 10 women) without contributory medical conditions. These patients ranged in age from 20–70 years (mean age = 40 years). Cyst specimens were collected from maxillary and mandibular treated and untreated teeth. Of the 23 specimens, 2 true cysts from different teeth belonged to the same patient, and 2 bay cysts from different teeth were from another patient. Twenty specimens were obtained from tooth extraction and 3 from apical surgery.

Eleven specimens were identified as true cysts (48%) and 12 (52%) as bay cysts. The mean age for patients with true cysts was 37 years (men = 34 years and women = 43 years), and for bay cysts, it was 43 years (men = 39 years and women = 46 years). Treated teeth with true cysts were followed up for 5–10 years. One treated tooth with a bay cyst was followed up for 4 years, whereas another tooth with a bay cyst had showed persistent exudation and symptoms that could not be resolved after several sessions of nonsurgical treatment and required periradicular surgery.

The frequency of true cysts according to the tooth type was as follows: maxillary premolars (5), maxillary molars (3), maxillary incisors (1), mandibular premolars (1) and mandibular molars (1). For bay cyst, frequency was as follows: maxillary molars (6), maxillary incisors (3), mandibular premolars (2) and maxillary premolars (1).

Bacteria were found in all cases examined, regardless of the histopathologic diagnosis (Figs. 1–5). Planktonic bacteria were observed in the main root canal in all true cyst cases and in 11 of 12 (92%) bay cyst cases. Biofilms were detected in the main canal in 10 cases from each of the diagnostic groups. These appeared to be very thick, often filling the entire canal lumen in the apical portion (Figs. 1A, C, and D and 2A, C, and D) and faced with a severe concentration of PMNs (Fig. 1C and D). Biofilms were frequently observed in ramifications from both treated and untreated teeth (Figs. 1B, 4B and D). Ramifications were sometimes clogged by dense biofilms exhibiting different bacterial morphotypes, with the bacterial cells prevailing over the extracellular matrix component (Fig. 4D).

Extraradicular biofilms formed on the external apical surface were detected only in a few specimens from both types of cysts. None of them showed signs of calcification. These biofilms exhibited varying proportions of cells and extracellular substance, and in 1 instance the biofilm was observed between the layers of cementum that were detached from the radicular surface (Fig. 4E).

Bacterial cells occurred in the lumen of true cysts in 4 of 11 (36%) specimens (Figs. 2A and B and 3A and B) and in 6 of 12 (50%) bay cyst specimens (Figs. 4B, C, and F and 5B and C). These bacteria were arranged in aggregations of varying sizes intermixed with necrotic debris and inflammatory cells, apparently free in the cyst lumen (Figs. 3A and B; 4B, C, and F; and 5B and C). In 2 cases (1 bay and 1 true cyst), they were present in the cyst lumen in the form of typical actinomycotic colonies, with intertwining branching

TABLE 1 - Clinical, Radiographic, Histopathologic, and Histobacteriologic Findings Associated with True and Bay Apical Cysts

	True cyst			Bay cyst			P value*
	Total, N (%)	Untreated, n (%)	Treated, n (%)	Total, n (%)	Untreated, n (%)	Treated, n (%)	
Specimens	11	7	4	12	10	2	
Sex [†]							.2
Male	7 (70)	4 (67)	3 (75)	4 (36)	3 (33)	1 (50)	
Female	3 (30)	2 (33)	1 (25)	7 (64)	6 (67)	1 (50)	
Tooth location							1.0
Maxillary	9 (82)	5 (71)	4 (100)	10 (83)	8 (80)	2 (100)	
Mandibular	2 (18)	2 (29)	0 (0)	2 (17)	2 (20)	0 (0)	
Lesion size							.68
Small (≤ 5 mm)	5 (45)	4 (57)	1 (25)	7 (58)	7 (70)	0 (0)	
Large (> 5 mm)	6 (55)	3 (43)	3 (75)	5 (42)	3 (30)	2 (100)	
Clinical symptoms							.4
None	2 (18)	1 (14)	1 (25)	2 (17)	2 (20)	0 (0)	
Mild	0 (0)	0 (0)	0 (0)	3 (25)	2 (20)	1 (50)	
Moderate	6 (55)	4 (57)	2 (50)	1 (8)	1 (10)	0 (0)	
Severe	3 (27)	2 (29)	1 (25)	6 (50)	5 (50)	1 (50)	
Previous acute abscess							.4
Yes	6 (55)	4 (57)	2 (50)	9 (75)	8 (80)	1 (50)	
No	5 (45)	3 (43)	2 (50)	3 (25)	2 (20)	1 (50)	
Sinus tract							.6
Yes	1 (9)	1 (14)	0 (0)	3 (25)	2 (20)	1 (50)	
No	10 (91)	6 (86)	4 (100)	9 (75)	8 (80)	1 (50)	
Planktonic bacteria							1.0
Main canal	11 (100)	7 (100)	4 (100)	11 (92)	10 (100)	1 (50)	
Bacterial biofilm							
Main canal	10 (91)	6 (86)	4 (100)	10 (83)	10 (100)	0 (0)	1.0
Ramifications	10 (91)	7 (100)	3 (75)	9 (75)	7 (70)	2 (100)	.59
Extraradicular	1 (9)	1 (14)	0 (0)	3 (25)	2 (20)	1 (50)	.59
Bacteria in the cyst cavity	4 (36)	3 (43)	1 (25)	6 (50)	5 (50)	1 (50)	.68

*Fisher's exact test.

[†]One patient contributed 2 true cysts and another patient contributed 2 bay cysts.

filamentous bacteria surrounded by a severe accumulation of PMNs (Fig. 2A and B).

The severity of inflammation was high in all specimens, so it was not subjected to statistical analysis. All kinds of inflammatory cells were observed in the cyst lumen together with varying amounts of amorphous necrotic debris. The epithelial lining of all but 4 radicular cysts was constituted by a typical stratified squamous epithelium. One true cyst and 3 bay cysts in 4 maxillary (one third, one second, and two first) molars showed the characteristics of a respiratory epithelium, with ciliated epithelial cells and goblet cells in limited portions of the cyst walls. The epithelial lining was in general thick and irregular, with extensive proliferation and the tendency to form arcading structures (Figs. 1A; 2A; 3A; 4A; and 5A, B, and D) with entrapped islands of highly vascularized connective tissue (Fig. 5D and E). The strands of epithelium and the connective tissue islands were heavily infiltrated by acute and chronic inflammatory cells (Fig. 5E and F).

There were no significant differences between true and bay cysts for all the clinical, radiographic, histopathologic, and

histobacteriologic parameters evaluated ($P > .05$). Data are shown in Table 1.

DISCUSSION

This study compared the prevalence and several features of true and bay cysts diagnosed histopathologically using a meticulous research protocol. All apical periodontitis lesions included in this study were obtained attached to the teeth and therefore maintaining their natural morphologic relationship with the root apex. This strict criterion was not used in the large majority of studies evaluating the prevalence of the different types of apical periodontitis lesions because it is more difficult to meet. This justifies the smaller number of cases evaluated herein. Serial sectioning from 1 side to the other of the apical periodontitis lesion was also performed in this study to report on the relationship of the lesion to the apical foramina. These 2 approaches are essential for an accurate histopathologic diagnosis of cysts and their classification as true or bay types. Findings from the present study showed that, apart from the morphologic relationship

between the cyst cavity and the root canal space, there were no other significant differences between true and bay cysts in terms of clinical, radiographic, histopathologic, and histobacteriologic features. These findings question the need to subdivide the classification of apical cysts.

The frequency of cysts observed in this study was 24% (23/95) of the apical periodontitis lesions examined. When compared with other studies that also evaluated serial or serial step sections of lesions attached to the apex, this figure is higher than the 15% and 17% reported by Nair et al¹⁰ and Simon²², respectively, but is lower than the 32% reported by Ricucci et al³ (Table 2). As for the prevalence of the 2 types of apical cysts, this study is in agreement with two others that shown that each type occurs in approximately half of the cases with a cyst diagnosis^{3,22}. The true cyst corresponded to 12% of all lesions, which is within the range previously reported of 9% to 16% of all lesions. Corresponding figures for the bay cyst were 13% in this study, in a previously reported range of 6% to 18% of all lesions^{3,10,22} (Table 2).

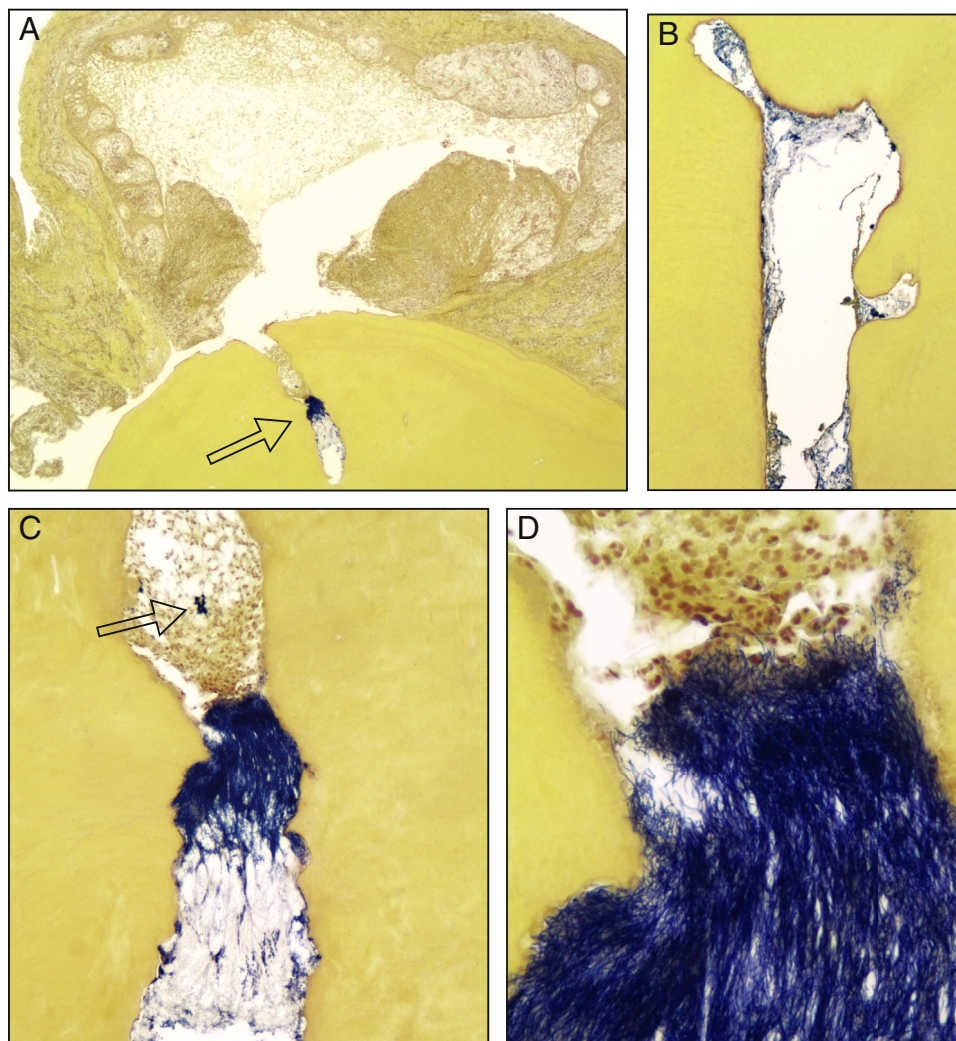


FIGURE 1 – A bay cyst. (A) A section cut through the main canal and foramen. The cyst lumen is continuous with the root canal space (Taylor modified Brown and Brenn, original magnification $\times 16$). (B) An area of the canal located approximately 5 mm coronal to the foramen (not shown in A). Biofilms with low bacterial density in the main canal and ramifications (original magnification $\times 50$). (C) Detail of the area of the apical canal indicated by the arrow in A. A thick bacterial biofilm fills the entire lumen and is faced with an accumulation of inflammatory cells. Note the “floc” displaced apically in the inflammatory tissue (arrow) (original magnification $\times 100$). (D) A high-power view of the biofilm showing a dense aggregation of filamentous forms (original magnification $\times 400$).

No previous study had compared the histobacteriology of true and bay cysts. No distinct pattern of infection was observed between the 2 types. Intraradicular bacteria were found in all cases, regardless of the associated cyst type and the root canal status (untreated or treated). Planktonic bacteria were observed in the main canal in all teeth, except for 1 bay cyst specimen, in which bacteria occurred as a biofilm in ramifications and in the extraradicular space. Bacteria organized in biofilm structures were observed in the main apical canal in the large majority of specimens, except for 1 true cyst and 2 bay cyst cases. In response to bacteria present in the root canal and sometimes inside the lesion, severe inflammation was observed in both cyst types. The present findings do not give support to the

assumption that true cysts can be an independent entity in the absence of concomitant bacterial infection because infection occurred in all cases.

Extraradicular bacteria were found in many cases, either inside the cyst lumen or in fewer cases as a biofilm adhered to the outer root surface near the exits of apical foramina. The histobacteriologic method permits better distinction of bacterial contaminants when compared with other methods of bacterial detection, such as culture and molecular methods, because it provides information on bacterial spatial location in the lesion and the association with inflammation. Specimens with contaminants were excluded from the study. Extraradicular bacteria were found in 5 specimens of true cysts, 4 within the cyst

cavity and 1 as a biofilm adhered to the outer root surface. Of these 5 cases, 4 had a history of previous acute abscess, and 1 had a sinus tract. These may have been the most possible explanations for the bacteria detected in the extraradicular space, particularly in the cyst lumen. After resolution of the abscess, bacteria may have persisted in the periradicular tissues and maintained a chronic infectious process. Even though the case with a sinus tract had not had a previous report of acute abscess, this might have occurred with subclinical symptoms to justify the fistula. Cases with a sinus tract have been shown to harbor an extraradicular infection in about 83% of the teeth³². Bacteria present within the cyst cavity are beyond the reach of nonsurgical root canal procedures and are

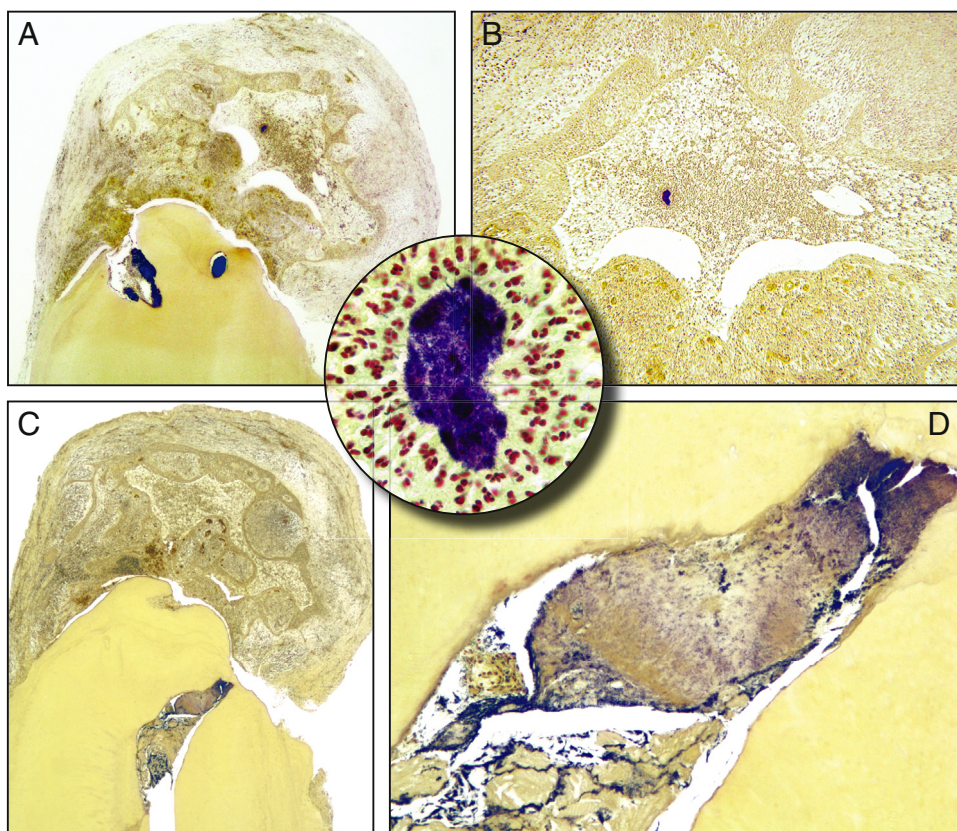


FIGURE 2 – A true cyst. (A) A section cut through 2 ramifications filled with bacterial biofilms. The cyst lumen is separated from the root canal space (this anatomic feature is maintained in all sections of the series) (Taylor modified Brown and Brenn, original magnification $\times 16$). (B) Detail of the cyst cavity whose lumen is filled with cells and debris. A high-power view of the blue spot at the center of the cyst lumen reveals an actinomycoticlike colony surrounded by a severe concentration of PMNs (original magnification $\times 50$, inset $\times 630$). (C) A section cut approximately 150 sections from that in A disclosing another large ramification clogged with a thick bacterial biofilm (original magnification $\times 16$). (D) Middle magnification from C (original magnification $\times 100$).

located in an area that makes it difficult for the host defenses to eliminate, given the type and consistency of the cyst lumen content.

However, the fate of bacteria eventually persisting in a cyst cavity after elimination of the intraradicular component is not known.

There are no reports demonstrating that bacterial colonies located inside the cyst cavity may cause root canal treatment failure

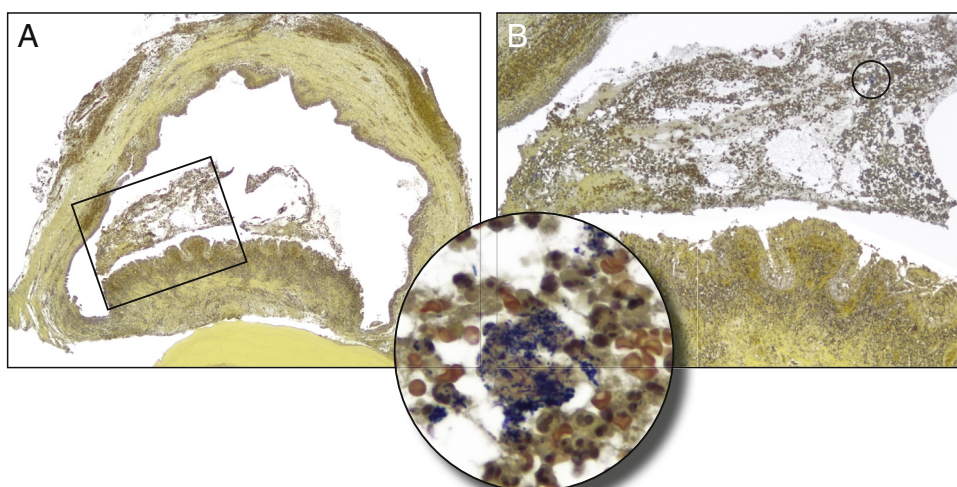


FIGURE 3 – A true cyst. (A) A large cyst cavity containing some debris (Taylor modified Brown and Brenn, original magnification $\times 16$). (B) Magnification of the rectangular area in A. Large numbers of bacteria colonize the necrotic debris in the lumen (original magnification $\times 100$, inset $\times 400$).

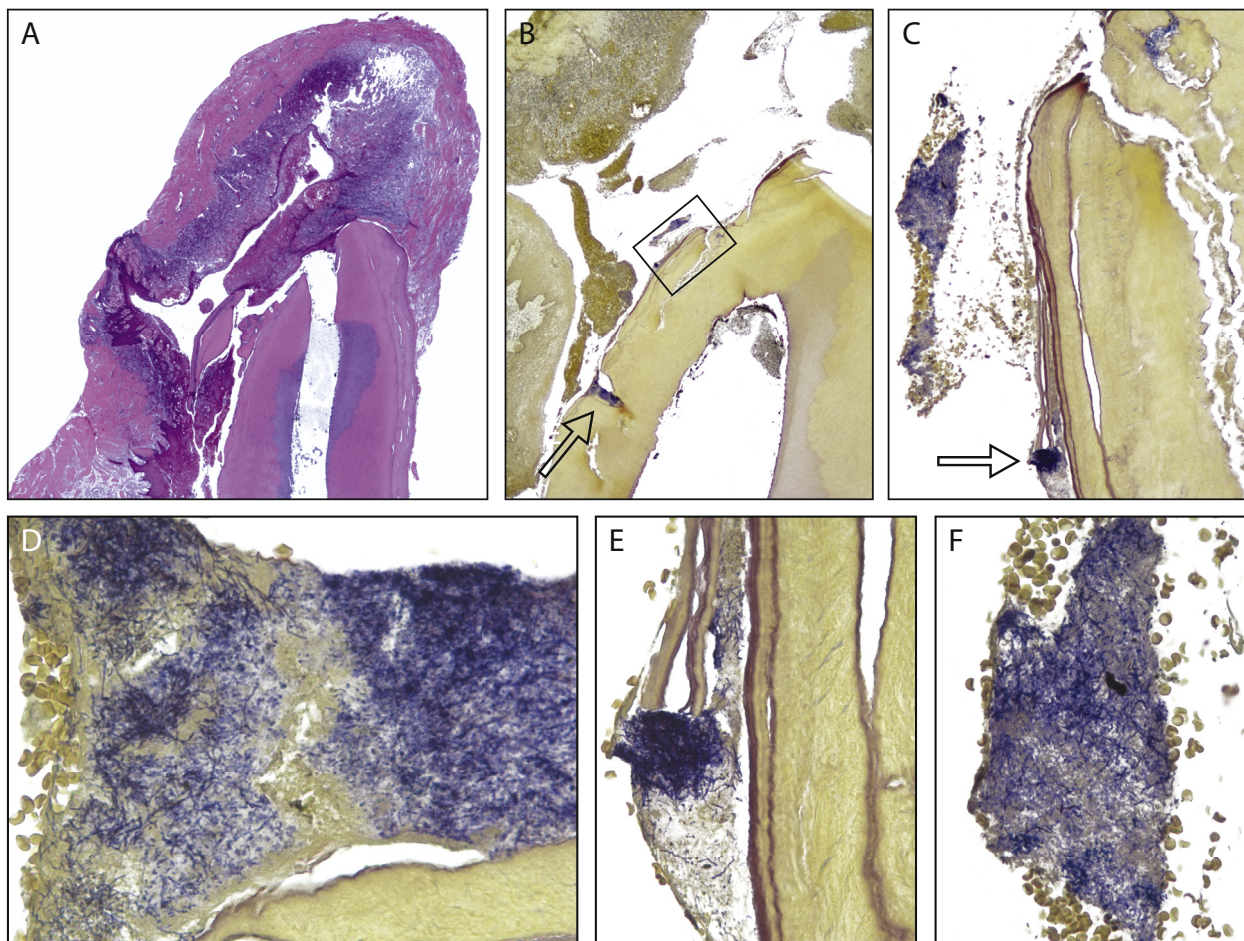


FIGURE 4 – A bay cyst. (A) A section encompassing the root canal, foramen, and cyst cavity. Overview (hematoxylin-eosin, original magnification $\times 8$). (B) A section taken 80 sections away passing through the canal but not including the main foramen. Several ramifications can be seen in the thickness of the left canal wall (Taylor modified Brown and Brenn, original magnification $\times 16$). (C) Magnification of the area demarcated by the rectangle in B (original magnification $\times 100$). (D) A high-power view of the exit of the lateral canal indicated by the arrow in B. Its lumen is occupied by a dense biofilm showing varying bacterial morphotypes (original magnification $\times 400$). (E) A high-power view of the area indicated by the arrow in C. Filaments prevail in a biofilm structure adhering to the radicular surface and showing varying bacterial concentrations (original magnification $\times 400$). (F) A high-power view of the elongated free colony in C (original magnification $\times 400$).

in the absence of a concomitant intraradicular infection.

The only case report that suggested that true cysts could be a self-sustaining entity independent from canal infection found no bacteria in the canal by culture and a correlative light and electron microscopic approach²⁴. However, bacteria may have passed unnoticed when using methods with low sensitivity for bacterial detection. A negative culture does not guarantee that the root canal is free of bacteria, especially because of the limitations of the culture technologies in detecting difficult-to-grow and as-yet-uncultivated bacteria and the inability to detect bacteria at low levels³³. In addition, the correlative light and electron microscopic approach used in the previous study can provide highly detailed information from some selected areas but has a very low sensitivity to detect bacteria. In that method, a serial step-

cutting approach is used in that stained sections are examined under the light microscope to locate sites for further sectioning and evaluation in a transmission electron microscope. Miniature pyramids are prepared at the selected sites that show bacteria in light microscopy or that are likely to harbor bacteria³⁴. The low sensitivity for bacterial detection is recognized by the authors themselves—“the extremely limited area that still can be covered by this means of investigation makes it rather easy for bacteria to go undetected”³⁴. This is because of the loss of biopsy material due to preparation for transmission electron microscopy and the very small area that can be examined under a transmission electron microscope. In turn, the histobacteriology approach used in this study has been very successful in revealing bacteria in the vast majority of cases with posttreatment apical periodontitis^{27,35,36},

including the lesions examined in the present investigation.

The assumption that the true cyst becomes a pathologic entity independent of the root canal system and is not affected by nonsurgical root canal treatment is only speculative and has neither scientific evidence support nor biological plausibility^{8,25}. Although this cyst type has no apparent communication with the root canal, this by no means can be interpreted as being a separate disease entity. The etiology is the same as the bay cyst (ie, infection of the root canal system that causes periradicular inflammation), and some locally released inflammatory mediators serve as growth factors for epithelium proliferation and cyst formation. There is no reason to believe that the epithelium lining of the true cyst can become self-sustainable, like neoplastic lesions^{8,25}. The present study lends substantial additional information to debunk

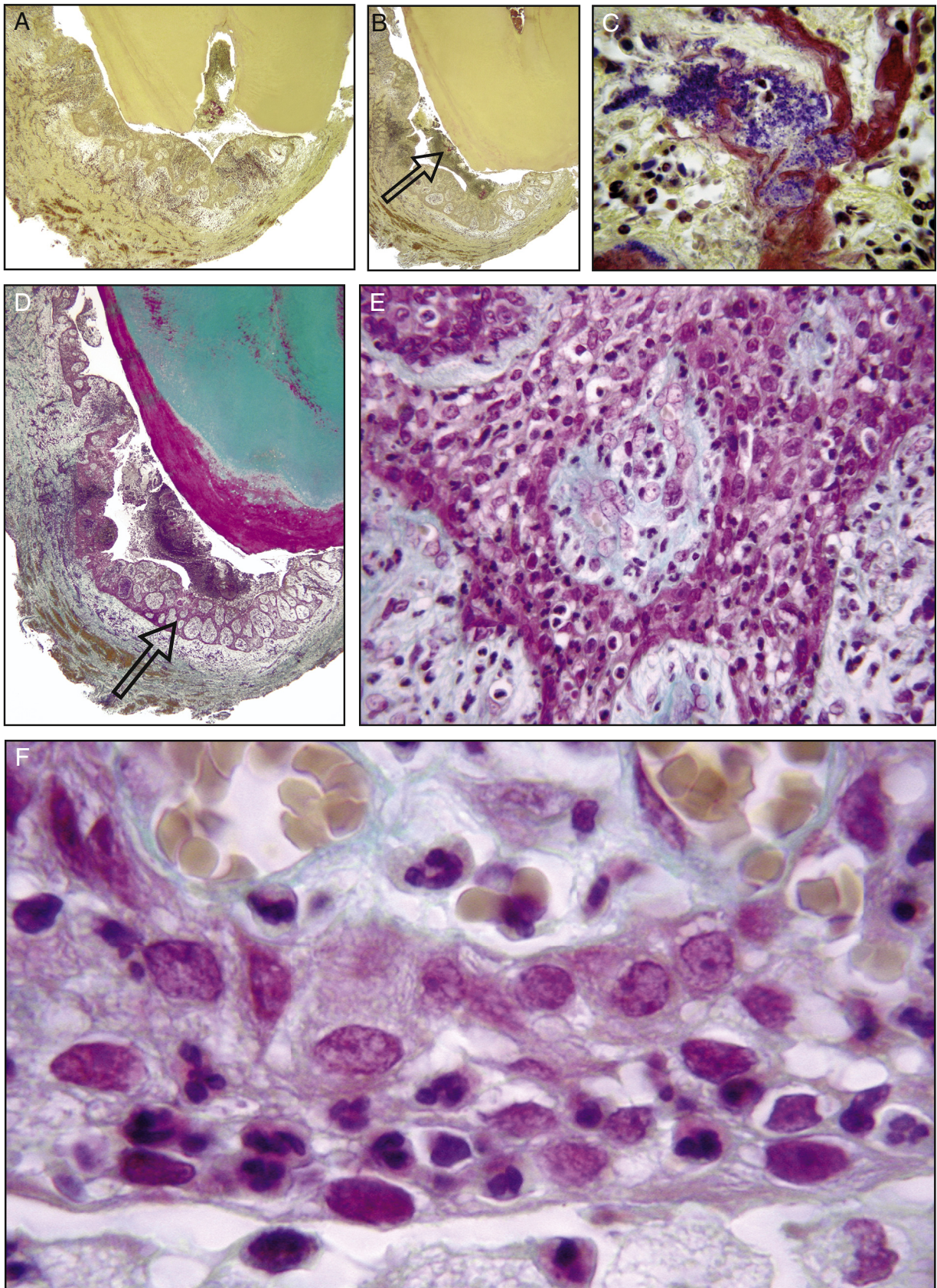


FIGURE 5 – A bay cyst. (A) A section cut through the apical canal and foramen (Taylor modified Brown and Brenn, original magnification $\times 25$). (B) A section taken approximately 80 sections away from that in A, not encompassing the apical foramen (original magnification $\times 25$). (C) A high-power view of the area of the cyst lumen indicated by the *arrow* in B. Bacterial aggregation intermixed with amorphous fuchsin-stained material and inflammatory cells (original magnification $\times 630$). (D) Another section of the series taken 40 sections after that in B. The cyst lumen is lined by a thick wall of arcading epithelium (Masson trichrome, original magnification $\times 25$). (E) A high-power view of epithelial strands indicated by the *arrow* in D. The epithelium surrounds cores of connective tissue infiltrated with inflammatory cells (original magnification $\times 400$). (F) The strands appear infiltrated by polymorphonuclear neutrophils (original magnification $\times 1000$).

TABLE 2 - The Prevalence of Apical Cysts and Their 2 Types in Studies That Used Histopathologic Serial or Serial Step Sectioning to Evaluate Lesions Attached to the Root Apices

Study	Cyst among all lesions, n (%)	True cyst among all lesions, n (%)	Bay cyst among all lesions, n (%)	True cyst among all cysts, n (%)	Bay cyst among all cysts, n (%)
Simon, 1980 ²²	6/35 (17)	3/35 (9)	3/35 (9)	3/6 (50)	3/6 (50)
Nair et al, 1996 ¹⁰	39/256 (15)	24/256 (9)	15/256 (6)	24/39 (61.5)	15/39 (38.5)
Ricucci et al, 2006 ³	16/50 (32)	8/50 (16)	9/50 (18)	8/16 (50)	9/16 (56)
This study	23/95 (24)	11/95 (12)	12/95 (13)	11/23 (48)	12/23 (52)

this theory because true and bay cysts showed no significant differences for all variables evaluated. In addition, the cysts that represented posttreatment apical periodontitis were of both types, and concurrent bacterial infection was always present inside the canal and sometimes outside. Therefore, it seems evident that failure of the endodontic treatment, including cases of true cysts, is primarily caused by persistent or secondary intraradicular bacterial infection with, in some cases, concurrent extraradicular bacterial infection.

A definitive answer as to whether cysts (all or only the “true” ones) can heal or not after nonsurgical root canal treatment would only be provided if cysts and other lesions could be distinguished without biopsy. A differentiation cannot be achieved by radiographs alone, and the effectiveness of the other methods has not been confirmed; only an old study using polyacrylamide gel electrophoresis of the root canal aspirates to differentiate granulomas and cysts suggested that many cysts healed after root canal treatment³⁷. Indirect evidence that cysts heal can be inferred from the fact that the success rate of endodontic treatment is higher than the cyst prevalence^{9,15,38,39}. Moreover, suggestive evidence is given by a study in which the nonsurgical root canal treatment of large cysticlike lesions with a fluid content containing cholesterol crystals resulted in

complete healing in 74% and incomplete healing in 9.5% of cases⁴⁰. Therefore, it seems reasonable to assume that apical cysts, regardless of their types, can heal provided the source of epithelial proliferation (ie, the root canal infection) is eliminated by treatment. If this is true, differentiation between granulomas and cysts or true and bay cysts would be of no clinical relevance.

A curious finding from this study was that from the 2 patients who contributed 2 specimens each, 1 had 2 true cysts associated with 2 different teeth and the other had 2 bay cysts from different teeth as well. This might suggest an individual predisposition to develop a true or a bay cyst, but the limited number of patients providing more than 1 specimen was too small to draw such a conclusion.

As typically found in apical cysts, a stratified squamous epithelium was found composing the lining of the cyst cavity in the large majority of specimens. However, 4 of 23 cysts (17%), all in the maxillary region, showed a ciliated columnar epithelium partially or completely lining the cyst wall. All involved teeth were maxillary molars, and the occurrence of a ciliated columnar epithelium in the lesions may be related to the proximity of the root apexes and lesions to the maxillary sinus floor².

In conclusion, the present findings revealed no significant differences between

true and bay cysts as to their prevalence, clinical, radiographic, histopathologic, and histobacteriologic manifestations. The 2 types of cysts were always associated with an intraradicular infection and sometimes with an extraradicular infection as well. **The fact that the 2 types of cysts only apparently differ from the morphologic relationship of the cavity to the root canal puts into question the real need to differentiate them and subdivide the apical cyst into categories.** Finally, the fact that all cases of true cysts from both untreated and treated teeth had an infectious component does not support the assumption that true cysts are self-sustainable entities not associated with infection.

CREDIT AUTHORSHIP CONTRIBUTION STATEMENT

Domenico Ricucci: Investigation, Methodology, Writing - review & editing, Visualization. **Isabela N. Rôças:** Data curation, Formal analysis, Validation. **Sandra Hernández:** Data curation, Formal analysis. **José F. Siqueira:** Conceptualization, Formal analysis, Writing - review & editing.

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