When Things are Not to Your Liking

The proper response to a perceived misstep by NYSDA is to air your grievance. Dropping out accomplishes nothing and may be harmful to the profession.

Toward the end of last year, a *NYSDJ* DentAd, emailed to subscribing members, promoted an orthodontic course, also being promoted at the Greater New York Dental Meeting. Since this advertisement was directed at general dentists so they could learn the intricate secrets of orthodontics through a self-taught course, several orthodontists in New York State expressed their concern over NYSDA accepting and distributing such an ad.

Maybe it was the name of the product, "Orthodontics in a Box," that caused their ire. Maybe it was the fact that it was aimed at general dentists. After all, it was perceived as increasing competition in a very competitive world, with orthodontists being badly hurt. As an orthodontist, I know of what I speak. However, I didn't see anything wrong with the ad; this is a free country, and this person had something to sell. NYSDA received some non-dues income from the advertiser, which can help keep our dues down. It was dentally related, so it was appropriate for the audience.

I'm not going to get into the philosophical debate about non-orthodontists doing orthodontics. That's a losing battle. I spent two and a half years of my life in residency to learn the science and art of orthodontics. I know I can treat an orthodontic case better than a dentist who learned about orthodontics from a course called "Orthodontics in a Box." However, general dentists can do any aspect of dentistry they wish, as long as it is up to the standard of care of a specialist. If it fails that test, the patient has the right to sue the treating dentist for malpractice.

No, that is not what I'm concerned about here. Rather, it is the fact that some orthodontists took such umbrage with that ad—and other DentAds they disagreed with—that they threatened to quit the tripartite all together. One said he had been thinking a long time about quitting, so I guess this ad was the last straw. These disgruntled members said they saw the ad as a betrayal by NYSDA. After all, they argued, isn't NYSDA supposed to protect all its members? And in this case, hadn't NYSDA fallen short of that protection?

I can see their point, but I think they are overreacting just a bit by quitting or threatening to quit organized dentistry.

Several years ago, a friend of mine actually did quit organized dentistry because he had an adverse judgment against him in peer review. He thought he was wasting his time and money belonging to an organization that didn't support him in his time of need. I spoke with him several times about this, explaining why the process he went through was important and how it really had protected him. Yes, he had to refund the cost of treatment to a dissatisfied patient when he felt he had done nothing wrong, had provided excellent care and that it was the patient who had erred by not following his post-treatment instructions. He wondered why he would have to refund the entire treatment fee.

What he didn't realize at the time was that that was all he would ever have to pay the patient. Because he went through peer review, the patient could not sue him. This saved him untold thousands of dollars, as well as the stress and heartache a lawsuit can bring—all because he was a member of NYSDA and had this benefit. That was worth his yearly dues payment. Eventually, he saw the error of his ways and rejoined organized dentistry.

I have to think the orthodontists who have threatened to quit are just reacting to a situation

they don't like and where they see themselves as aggrieved parties. However, to quit organized dentistry because it ran an ad they didn't appreciate is short-sighted. After all, "Orthodontics in a Box" will never be the challenge to orthodontists and the health of their practices that Invisalign already is. Talk about cookbook orthodontics. Take two impressions, send them off and get the treatment plan back looking all nice and neat. Then just slap the aligners into the patient's mouth and off you go. What could be simpler than that?

Or more dangerous. For if you don't know how teeth move, where you can move teeth, what their stability will be when treatment is finished, you really aren't practicing orthodontics. What you are practicing is "expandodontics." It will always look good on the computer screen, but will it actually work and be beneficial in the patient's mouth? That's where the extra education comes in. You have to know these things implicitly or you will get into a lot of trouble and you won't be treating the patient up to the standard of care.

If you object to an ad, don't quit over it. Write a letter to the editor; contact NYSDA; discuss it with those in charge. Only in that way will you be able to exert any influence over what is actually accepted as advertisements. All specialties are in the same boat. I'm sure endodontists would object to "Endodontics in a Box," or oral surgeons to "Third Molar Extractions in a Box" or periodontists to "Perio Surgery in a Box." Object if you must, but don't just get up, take your ball and go home.

My advice to orthodontists who want to resign from organized dentistry is don't. Organized dentistry is our only voice in the cacophony of voices out there trying to be heard. If every dentist who disagreed with something that organized dentistry did quit, there would be no organized dentistry. What you see as an affront to your sensibilities is only a minor inconvenience when you look at the big picture. When you disagree with something, let your voice be heard. There is always someone who will listen and take what you have to say to heart.


D.D.S.

NYSDA Directory

OFFICERS

Joel M. Friedman, President	525 E. 68th St., New York, NY 10065
John J. Liang, President Elect	2813 Genesee St., Utica, NY 13501
David J. Miller, Vice President	467 Newbridge Rd., East Meadow, NY 11554
Mark J. Weinberger, Treasurer	78 Southbury Road, Clifton Park, NY 12065
Robert M. Peskin, Speaker of the House	601 Franklin Ave., #225, Garden City, NY 11530
Mark J. Feldman, Executive Director	20 Corporate Woods Blvd., Albany, NY 12211
Steven Gounardes, ADA Trustee	351 87th St., Brooklyn, NY 11209

BOARD OF TRUSTEES

P. Deborah Weisfuse, Immediate Past President	12 E. 41st St., #1100, New York, NY 10017
NY – Edward J. Miller Jr	121 E. 60th St., #7A, New York, NY 10022
2 - Craig S. Ratner	7030 Hylan Blvd., Staten Island, NY 10307
3 – Lawrence J. Busino	2 Executive Park Dr., Albany, NY 12203
4 – Frederick W. Wetzel	1556 Union St., Schenectady, NY 12309
5 – William H. Karp	Bldg. 6, 4500 Pewter Lane, Manlius, NY 13104
6 - Scott J. Farrell	39 Leroy St., Binghamton, NY 13905
7 - Robert J. Buhite II	564 E. Ridge Rd., #201, Rochester, NY 14621
8 – Jeffrey A. Baumler	2145 Lancelot Drive, Niagara Falls, NY 14304
9 – Stuart H. Coleton	7 Stone Rd., Chappaqua, NY 10514
N—Michael S. Shreck	1300 Union Turnpike, #201, New Hyde Park, NY 11040
Q – Joseph R. Caruso	40-29 Utopia Pky., Auburndale, NY 11358
S – Paul R. Leary	80 Maple Ave., #206, Smithtown, NY 11787
B - Richard P. Herman	2 Lockwood Lane, New Windsor, NY 12553

COUNCIL CHAIRPERSONS

Awards	**Lawrence E. Volland** 115 Professional Parkway, Lockport, NY 14094
Dental Benefit Programs	**Eugene G. Porcelli** 601 Franklin Ave., #211, Garden City, NY 11530
Dental Education & Licensure	**Rekha C. Gehani** 35-40 82nd St., #1F, Jackson Heights, NY 11372
Dental Health Planning & Hospital Dentistry	**Carl H. Tegtmeier** 359 E. Main St., #2C, Mt. Kisco, NY 10549
Dental Practice	**Bijan Anvar** 15 Rocky Rd., Larchmont, NY 10538
Ethics	**Richard B. Serchuk** 5 Valentines Ln., Old Brookville, NY 11545
Governmental Affairs	**Prabha Krishnan** 110-45 Queens Blvd., #108, Forest Hills, NY 11375
Membership & Communications	**Jay Skolnick** 39 W. Main St., Webster, NY 14580
Nominations	**P. Deborah Weisfuse** 12 E. 41st St., #1100, New York, NY 10017
Peer Review & Quality Assurance	**Jeffrey M. Galler** 18 Copper Beech Lane, Lawrence, NY 11559
Professional Liability Insurance	**Roland C. Emmanuele** 4 Hinchcliffe Dr., Newburgh, NY 12550

OFFICE

Suite 602, 20 Corporate Woods Blvd., Albany, NY 12211
(518) 465-0044 (800) 255-2100

Mark J. Feldman	Executive Director
Lance Plunkett	General Counsel
Beth M. Wanek	Associate Executive Director
Michael J. Herrmann	Assistant Executive Director Finance-Administration
Judith L. Shub	Assistant Executive Director Health Affairs
Joshua Poupore	Assistant Executive Director Marketing and Communication
Laura B. Leon	Assistant Executive Director
Mary Grates Stoll	Managing Editor

Read, Learn and Earn
CE
CONTINUING EDUCATION

Septic Arthritis of the Shoulder in a Dental Patient

A Case Report and Review

Elana Dolin, D.M.D.; Leigh D. Perlmutter, D.M.D.; Stuart L. Segelnick, D.D.S., M.S.;

Mea A. Weinberg, D.M.D., R.Ph., M.S.D.; Robert Schoor, D.D.S.

ABSTRACT

Septic arthritis of the glenohumoral joint is rare following dental procedures, comprising approximately 3% of all joint infections. Septic arthritis following bacteremia from dental procedures is uncommon and generally occurs in prosthetic joints. Predisposing causes may include immunocompromising diseases such as diabetes, HIV infection, renal failure and intravenous drug abuse. We report a rare case of unilateral glenohumoral joint septic arthritis in a 60-year-old male patient (without a prosthetic joint) secondary to a dental procedure. The insidious nature of the presentation is highlighted.

Septic arthritis infections, though rare, require a high level of clinical suspicion. Vague symptoms of shoulder pain may mask the initial diagnosis, as was the case in our patient. Incision and drainage via surgical intervention are often required, followed by parenteral antibiotics.

Pneumococcal septic arthritis, or acute pyogenic arthritis, is diagnosed when *Streptococcus pneumoniae* is isolated from the synovial fluid or purulent joint fluid.[1] An imaging study showing sacroiliitis—inflammation of one or both of the sacroiliac joints which connect the lower spine and pelvis—or a Gram staining showing the presence of the bacteria is necessary to confirm this diagnosis.[2] In some cases there may be a high sensitivity of elevated polymorphonuclear leukocytes in the joint fluid. Elevation of erythrocyte sedimentation rate (ESR) and C-reactive protein (CRP) has also been documented, but both are considered nonspecific indicators of the infection.[3]

Most cases of septic arthritis occur in the weight-bearing joints of the lower extremities, with involvement of the glenohumeral joint occurring in only 3% of patients.[2] However, it has been reported to have occurred in as many as 12% of cases, which would place it as the third most common site following the knees and hips.[4]

Septic arthritis following bacteremia from dental procedures is uncommon and generally occurs in patients with prosthetic joints.[5-8] It is documented that septic arthritis is 15-times more common in prosthetic joints than in native joints.[9] Previous reports have shown that septic arthritis in a normal joint following a dental procedure has occurred in both the hand and knee.[5,7,8]

This report describes a case of septic arthritis of the shoulder in a patient following a dental procedure. It is important to be aware of this potential complication and to be able to advise the patient on how to receive the proper care.

Case Report

A 60-year-old male presented to New York University College of Dentistry for dental treatment. His last dental exam was 15 years prior. The patient denied any history of dental infections. Periodontal probing was performed, and appropriate radiographs were taken as part of the comprehensive examination. The patient was diagnosed with generalized severe chronic periodontitis.

The patient's past medical history consisted of prior tobacco use (1ppd for 6 years) and a history of a duodenal ulcer 15 years ago. The patient indicated that he drank rarely.

Two days after the periodontal examination, the patient presented for a full-mouth debridement. At this appointment, the patient said he had discomfort in his left shoulder. One of the first signs of septic arthritis is an inability to use a limb. He said he thought he had torn a ligament in his left shoulder. The evening after the full-mouth debridement, the patient reportedly developed severe sweats.

Two days later, the patient presented to a hospital emergency department with pain and heat emanating from his left shoulder. Radiographs were taken and read as negative, and he was prescribed hydrocodone/acetaminophen for pain relief. He had no relief from the narcotic analgesic and presented to an emergency department at a nearby hospital two days later with the chief complaint of "I have sharp throbbing pain down to the bone" on his left shoulder. At that time, the patient said he had developed a fever four days ago, which coincided with an increase in the pain level in his left shoulder.

Examination revealed a decreased range of motion; and the shoulder was tender to palpation. He had a body temperature of 38.8 degrees C (101.8 degrees F). A computed tomography (CT) scan was taken that revealed myositis and a septic joint (Figure 1). Blood lab results showed an elevated sedimentation rate of 107 (reference range 0-20 mm/hr), an elevated white blood cell count of 12.7 K/uL (reference range 3.4- 11.2 K/uL) and an elevated CRP of 42.85 mg/dL (reference range <1.00 mg/dL). Additionally, the examination revealed a heart murmur. The patient's blood glucose was elevated, ranging from 152 mg/dL to 178 mg/dL (reference range 74-118mg/dL), and HgbA1c was 6.7%. The remainder of the examination was normal, with no evidence of other infectious sources.

Aspiration of the joint with a large needle revealed frank pus. The presence of pus in the joint, in addition to the patient being febrile and having limited and painful range of motion, were indications for an arthroscopic incision and drainage procedure. The final diagnosis given was uncomplicated Type 2 diabetes, a new heart murmur and a septic left shoulder joint.

Results of the arthroscopic surgery revealed Grade II changes involving about 40% of the humeral head and isolated Grade III changes involving 10% of the humeral head. Extensive synovitis in the rotator interval and fraying on the undersurface of the rotator cuff were observed. The anterior labrum had frayed as well. The surgeons established an anterior rotator into a portal, through which bacterial cultures were taken. Cultures were also taken through the posterior portal. It was noted that there was suppuration via the trocar before the arthroscopy was performed in the rotator interval. A culture was taken and all scar and syno-

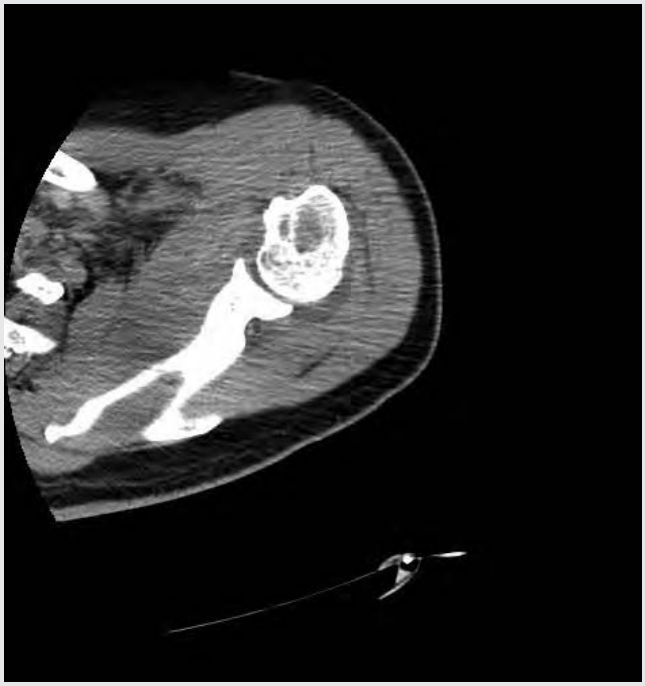


Figure 1. Glenohumeral joint effusion.

vitis was debrided. Antibiotic (gentamycin 80mL/2mL NaCl) impregnated solution was flushed through the glenohumeral joint.

The patient's blood cultures grew Gram-positive cocci in pairs and short chains, suggestive of enterococci or pneumococci. The presumed causative organism was *Streptococcus pneumoniae* sensitive to ceftriaxone. The patient was given IV ceftriaxone and IV vancomycin. After five days of intravenous antibiotics, the patient was discharged with instructions to take ceftriaxone 2g/50mL IV 1/day x 40 days.

Discussion

Septic arthritis is an uncommon complication following dental procedures, especially in normal joints via hematogenous seeding of a joint.[1] The responsible bacterial organism in this case report was *Streptococcus pneumoniae,* a Gram-positive facultative anaerobe often found in the nasopharynx of healthy individuals.[10] Approximately 70% of cases of septic arthritis are isolated through blood or joint culture. *Staphylococcus aureus* is the most common organism isolated and accounts for 44% of septic arthritis. Other common organisms include *Kingella kingae* (14%), *Streptococcus pyogenes* (10%) and *Streptococcus pneumoniae* (10%). It should be noted that each risk (age) group has its own characteristic infective microorganisms, including bacteria, fungi and viruses. However, bacteria are most important because of their invasive, destructive nature.

The knee is the most common site involved, followed by the hip and shoulder. Septic arthritis has also been identified in the ankle, elbow and wrist, but these sites are relatively uncommon.[11-13] The majority of patients diagnosed with septic arthritis have at least one serious medical condition. Comorbidities present in adult septic arthritis of the shoulder include diabetes mellitus, rheumatoid arthritis, HIV infection, alcoholism, osteoarthritis, prosthetic joints, coronary disease, IV drug use, corticosteroid use, and multiple myeloma or monoclonal gammopathy.[1,3] In this case, there was an incidental finding of Type II diabetes.

Alcohol increases the risk of aspiration and upper airway bacterial colonization, decreases pulmonary macrophage phagocytosis and alters surfactant biochemistry. Alcohol is implicated as a risk factor in up to 70% of cases of pneumococcal bacteremia.[14]

Ross et al. reported on 2,407 cases of septic arthritis. Of these, 6% were caused by *Streptococcus pneumoniae*. After reviewing 190 cases of pneumococcal septic arthritis, it was noted that one-half of the patients reviewed did not have an underlying focus of pneumococcal disease, such as pneumonia or meningitis. It was presumed that septic arthritis arose from joint seeding during transient bacteremia, with a mucous membrane source. Mortality was 19% among adults, with pneumococcal bacteremia being the strongest predictor of mortality.[1]

The temporal relationship of the infection to the dental examination and probing, the lack of other infections, trauma, arthritis in the joint and negative blood cultures suggest a transient streptococcal bacteremia following a procedure involving the oral/pharyngeal region as etiology of the infection.

Septic arthritis following dental procedures is rare and exhibits a rapidly destructive infectious process with notable systemic symptoms. It occurs through the blood from a distant septic focus. The suggested treatment is prompt surgical drainage and irrigation, as well as intravenous antibiotic administration to reduce morbidity.[5] It is important to be aware of this potential complication after dental treatment, as early diagnosis and treatment are critical to infection resolution. A differential diagnosis of septic arthritis includes patients with upper extremity joint complaints especially after dental treatment.

Drs. Dolin, Perlmutter, Segelnick and Weinberg have dedicated their paper to the memory of their coauthor Dr. Robert Schoor, their mentor and inspiration, who died in November 2012. Queries about this article can be sent to Dr. Segelnick at EperioDr@aol.com.

REFERENCES

1. Ross JJ, Saltzman CL, Carling P, Shapiro DS. Pneumococcal septic arthritis: review of 190 cases. Clin Infect Dis 2003 Feb 1;36(3):319-27. Epub 2003 Jan 13. Review.
2. Lossos IS, Yossepowitch O, Kandel L, Yardeni D, Arber N. Septic arthritis of the glenohumeral joint. A report of 11 cases and review of the literature. Medicine (Baltimore) 1998 May;77(3):177-87. Review.
3. Mehta P, Schnall SB, Zalavras CG. Septic arthritis of the shoulder, elbow, and wrist. Clin Orthop Relat Res 2006 Oct;451:42-5.
4. Le Dantec L, Maury F, Flipo RM, Laskri S, Cortet B, Duquesnoy B, Delcambre B. Peripheral pyogenic arthritis. A study of one hundred seventy-nine cases. Rev Rheum Engl Ed 1996 Feb;63(2):103-10.
5. Payne LZ, Wolfe, SW. Septic arthritis in a normal interphalangeal joint following a dental procedure. Journal of Hand Surgery 1994 Sep;19(5):815-6.
6. Thyne GM, Ferguson JW. Antibiotic prophylaxis during dental treatment in patients with prosthetic joints. J Bone Joint Surg 1991 Mar;73(2):191-4.
7. Marshall DA, Moussa KM, Sanders PA. Streptococcal arthritis and erysipelas complicating dental extractions. Br. J Rheumatology 1993 Jan;32(1):83-4.
8. Patrick MR, Lewis D. Short of a length: Streptococcus sanguis knee infection from dental source. Br J Rheumatology 1992 Aug;31(8):569.
9. Kaandorp CL, Schaardenburg D, Krijnen P, Habbema JD, van de Laar MA. Risk factors for septic arthritis in patients with joint disease: a prospective study. Arthritis Rheum 1995 Dec;38(12):1819-25.
10. Muscoso M, Garcia E, Lopez R. Pneumococcal biofilms. International Microbiology 2009 Jun;12(2):77-85.
11. Smith JW, Piercy EA. Infectious arthritis. Clin Infect Dis 1995 Feb;20(2):225-30.
12. Moumile K, Merckx J, Glorion C, Pouliquen JC, Berche P, Ferroni A. Bacterial aetiology of acute osteoarticular infections in children. Acta Paediatr 2005 Apr;94(4):419-22.
13. Krogstad P. Septic arthritis. In: R.D. Feigin, J.D Cherry, G.J. Demmler- Harrison, S.L. Kaplan (Eds), 6th ed., Textbook of Pediatric Infectious Diseases. Philadelphia: Saunders, 2009.
14. Hook EW III, Horton CA, Schaberg DR. Failure of intensive care unit support to influence mortality from pneumococcal bacteremia. JAMA 1983 Feb 25;249(8):1055-7.

Concrescence

Assessment of Case by Periapical Radiography, Cone Beam Computed Tomography and Micro-computed Tomography

Frederico Sampaio Neves, D.D.S., M.Sc., Ph.D.; Karla Rovaris, D.D.S., M.Sc.;
Matheus Lima Oliveira, D.D.S., M.Sc., Ph.D.; Pedro Duarte Novaes, D.D.S., M.Sc., Ph.D.;
Deborah Queiroz de Freitas, D.D.S., M.Sc., Ph.D.

ABSTRACT

The aim of this article was to describe imaging aspects of concrescence analyzed by three imaging modalities. A second molar joined together with a third molar was imaged using digital periapical radiography, cone beam computed tomography (CBCT) and micro-computed tomography (Micro-CT). On periapical radiograph, the mesial root of the third molar is superimposed on the distal root of the second molar. On CBCT images, a large cementum union between bulbous roots was detected, confirming the diagnosis of concrescence. On micro-CT images, the cementum union appeared limited to the apical third of the roots. In conclusion, both computed tomography modalities allowed for the diagnosis of concrescence. However, only micro-CT provided the real extension of the cementum union.

Concrescence is a rare dental anomaly in which the cementum overlying the roots of at least two teeth joins together. The involved teeth present independent pulp chambers and root canal systems. This anomaly may occur during or after root formation, which is known as true or acquired concrescence, respectively.[1] Concrescence is more frequently noted in maxillary molars. It can affect normal, supernumerary, erupted and impacted teeth.[1-3] The incidence is 3.7% in the deciduous dentition and 0.8% in the permanent dentition.[4]

A clear understanding of the imaging features of concrescence is of great importance due to the rare occurrence of this dental anomaly. Therefore, the aim of this article is to describe imaging aspects of concrescence analyzed by digital periapical radiography, cone beam computed tomography (CBCT) and micro-computed tomography (micro-CT).

Report

In an evaluation of the teeth bank of a dental school, a maxillary second molar was found joined to the third molar. On clinical examination, the union was restricted to the roots, which were covered by a uniform layer of cement. There was no evidence of caries.

Both teeth were X-rayed using a digital periapical radiographic system *(VistaScan, Dürr Dental, Beitigheim-Bissingen, Germany)*, and scanned using CBCT *(Picasso Trio, VATECH, Yongin, North Korea)* and micro-CT *(SkyScan 1174v2, SkyScan N.V., Kon-*

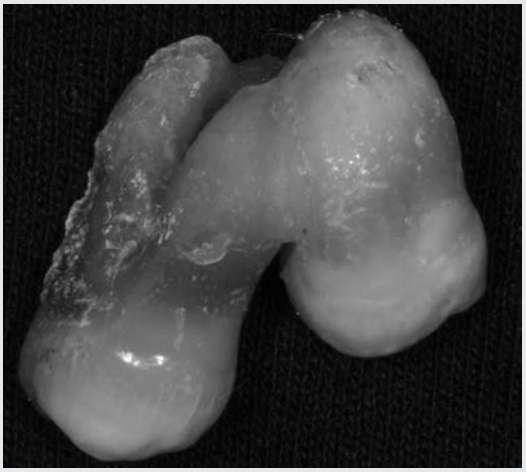


Figure 1. Clinical aspect of concrescence.

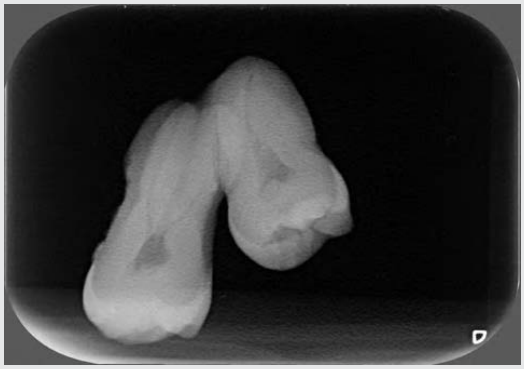


Figure 2. Digital periapical radiography showing concrescence.

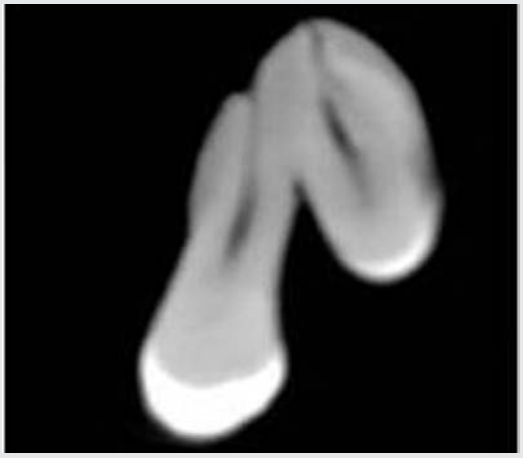


Figure 3. Coronal slice of CBCT showing concrescence.

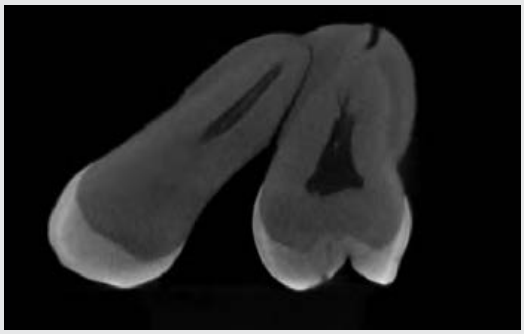


Figure 4. Coronal slice of micro-CT showing concrescence.

tich, Belgium) systems. On periapical radiograph, the mesial root of the third molar is superimposed on the distal root of the second molar. Additionally, it is possible to observe the root fusion of the third molar roots and the hypercementosis in all root teeth (Figure 2). On CBCT images, a large cementum union between bulbous roots was detected, confirming the diagnosis of hypercementosis and concrescence (Figure 3). On micro-CT images, the cementum union appeared limited to the apical third of the roots; and the remainder of both teeth was completely separated (Figure 4).

Discussion

Specific nomenclature has been used to describe abnormal events during tooth development that manifest as odontogenic anomalies in conjoined teeth. Germination appears to be two teeth developed from one as a result of incomplete splitting of the dental follicle. It begins at the incisal edge and stops before cleavage is complete.[5] Regardless of changes observed in the crown, a single root with only one root canal is present. Clinically, germinated teeth present a bifid crown with a well-defined groove or an incisal notch delineating two crowns. In contrast, fusion is a dental anomaly characterized by the union of two normally separated tooth germs during odontogenesis.[6] The extent and location of the union depends upon the tooth developmental stage at the time of the fusion. In most cases, it results in a large tooth with independent pulp chambers and root canals.

Concrescence may occur during or after rhizogenesis. Although the exact etiology is unknown, it is thought to be a result of trauma or crowding of adjacent teeth, such that the interdental bone resorbs, favoring the deposition of cementum between adjacent tooth roots.[7] It may also be a response to an inflammatory condition, such as dental caries or periodontal inflammation.[8] In the case presented here we believe the cementum deposition resulted from periodontal inflammation, since neither tooth had caries.

Concrescence is nearly impossible to detect clinically because the crowns of affected teeth, when erupted, appear normal, as there is no enamel involvement. The superimposition of teeth and the amount of cementum may compromise the radiographic detection of concrescence and contribute to an inaccurate diagnosis. It has been suggested that in some cases, concrescence can be identified radiographically when exposed at different parameters and angles, or when there is a substantial amount of cementum deposition.[1,2] In this particular case, the detection of concrescence could be confirmed only after CBCT and micro-CT scans.

According to Mader,[8] concrescence may happen from one small site to a solid cementum mass along the entire extent of the root surfaces. In the case presented here, CBCT images showed the union of two teeth. But micro-CT showed that only the apical

third was involved.

Micro-CT, also known as high resolution computed tomography, was developed for 3D imaging of small objects. This is a non-destructive technique capable of reconstructing samples on a micrometric scale. Its application in biology involves different structures from the human body,[9] including extensive studies of teeth and bone. This technology is also used in endodontics and has shown precision in depicting the finest anatomy of the root canal, which could not be achieved by previous imaging modalities.[10]

Clinicians should be aware of this dental anomaly and consider concrescence when performing an extraction of a difficult upper molar to avoid complications such as fractures of the tuberosity and floor of maxillary sinus or injury to the inferior alveolar nerve. Radiographs should be taken prior to extractions to detect possible challenges and to establish a treatment plan that provides the best possible outcome for the patient.

Conclusions

Three-dimensional exams were of paramount importance in the diagnosis of concrescence. Micro-CT was the only imaging modality that provided the real extension of the cementum union. The most accurate diagnosis of concrescence should be done to assess the relationship of the teeth with anatomical structures, particularly the maxillary sinus and mandibular canal, and to provide a higher operative safety.

Queries about this article can be sent to Dr. Neves at fredsampaio@yahoo.com.br.

REFERENCES

1. Gunduz K, Sumer M, Sumer AP, Gunhan O. Concrescence of a mandibular third molar and a supernumerary fourth molar: report of a rare case. Br Dental J 2006;200:141–2.
2. Romito LM. Concrescence: report of a rare case. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2004;97:325–7.
3. Foran D, Komabayashi T, Lin LM. Concrescence of permanent maxillary second and third molars: case report of non-surgical root canal treatment. J Oral Science 2012;54:133–6.
4. Schuurs AH, van Loveren C. Double teeth: review of the literature. ASDC J Dental Child 2000;67:313–25.
5. Hernandez-Guisado JM, Torres-Lagares D, Infante-Cossio P, Gutierrez-Perez JL. Dental germination: report of case. Med Oral 2000;7:231–6.
6. Kaffe I, Littner MM, Begleiter A, Buchner A. Fusion of permanent molars. Quintessence Int Dental Dig 1982;13:1237–9.
7. Linn EK. Concrescence: a case report. Gen Dent 1998;46:388–9.
8. Mader CL. Concrescence of teeth: a potential treatment hazard. Gen Dent 1984;32,52–5.
9. Mizutani R, Takeuchi A, Uesugi K, Ohyama M, Takekoshi S, Osamura RY, Suzuki Y. Three-dimensional microtomographic imaging of human brain cortex. Brain Res 2008;1199:53–61.
10. Yamada M, Ide Y, Matsunaga S, Kato H, Nakagawa K. Three-dimensional analysis of mesiobuccal root canal of Japanese maxillary first molar using Micro-CT. The Bull Tokyo Dental Coll 2011;52:77–84.

Dental Management of Florid Cemento-Osseous Dysplasia

Raid S. Sadda, D.D.S., M.S., MFDRCS; Joan Phelan, D.D.S.

ABSTRACT

Cemento-osseous dysplasia encompasses several different clinical and radiographic presentations, including periapical, focal and florid cemento-osseous dysplasia (FCOD). FCOD is usually asymptomatic and discovered only fortuitously. No treatment is required unless the lesion is secondarily infected. Oral hygiene advice should be emphasized for patients with FCOD to prevent caries and periodontal diseases and to maintain natural teeth. Osseointegration of implants would likely not be successful in these patients because the bone is abnormal and not well-vascularized.

Cemento-osseous dysplasia encompasses several different clinical and radiographic presentations, including periapical, focal and florid cemento-osseous dysplasia (FCOD). FCOD, the most extensive form of this condition, was first described by Melrose et al. in 1976. It presents with extensive involvement in one quadrant or involvement of multiple quadrants. The etiology is unknown.[1,2] Since these lesions occur only in tooth-bearing areas of the jaws, periodontal ligament involvement in the pathogenesis has been suggested.

Histopathological examination of this disorder reveals a fibro-osseous lesion that is composed of cellular fibrous connective tissue and mineralized components that vary from trabeculate-like osteoid to rounded mineralized material that has been called cementoid material. As the lesion matures, it becomes more mineralized and less vascular. The condition is most commonly diagnosed in black women, between 30 and 50 years of age. It has occasionally been reported in men, Caucasian and Asian patients.

The radiographic presentation of this condition is pathognomonic. Ideally, the diagnosis is established on the basis of the radiographic finding. Classic FCOD presents as multiple, diffuse, lobular or irregular-shaped radiopaque masses in the tooth-bearing or edentulous areas of the maxilla and/or mandible. Waldron[3] commented that the histopathologic finding of a fibro-osseous lesion would not be diagnostic of this condition without consistent clinical and radiographic findings. Although an initial radiolucent phase has been described, with progressive radiopacity in a manner similar to periapical cemento-osseous dysplasia, the radiopaque phase is more commonly recognized. The condition is generally asymptomatic, with no enlargement of the bone; teeth in the area are vital unless they have been devitalized due to other causes.

When this condition is identified, preventative management for caries and periodontal disease to maintain natural teeth and prevent the need for extractions should be considered.

FCOD is a condition that should be recognized by dentists on the basis of its radiographic appearance. Periapical radiographs may suggest this condition, but a pantomographic radiograph is usually more helpful in confirming the diagnosis. Although biopsy and histopathologic examination of the tissue may be helpful in confirming the diagnosis, they are generally not necessary, and the avascular nature of the tissue may result in delayed healing and increase the risk of infection following the biopsy. Tissue-borne removable prosthesis is generally contraindicated in patients with FCOD because the mineralized masses remain when alveolar bone atrophies and perforation of the mucosa may lead to osteomyelitis. FCOD can make the mandible more susceptible to osteomyelitis.[13]

Ideally, this condition should be diagnosed when the patient has natural teeth and preventative measures to maintain natural teeth can be encouraged.

Case Report

A 52-year-old black woman was referred to the Department of Oral and Maxillofacial Surgery at New York University College of Dentistry with severe pain in the lower right first molar. The patient had no significant medical history and was not taking medication. Clinical examination revealed a vertical fracture of the mandibular right first molar. There was no bone expansion.

Pantomographic radiograph revealed multiple radiolucent/radiopaque areas in the right and left quadrants of the mandible and a suggestion of the possibility of two inferior alveolar canals at the left mandible (Figure 1). Radiolucent areas were found at the furcation and periapical areas of the mandibular molars. A cone beam radiograph was taken and confirmed the presence of only one canal.

Extraction of the fractured tooth was performed. Postoperative instruction was given, and the extraction site healed without complication (Figure 2).

Discussion

FCOD is usually asymptomatic and discovered only fortuitously. No treatment is required unless the lesion is secondarily infected.

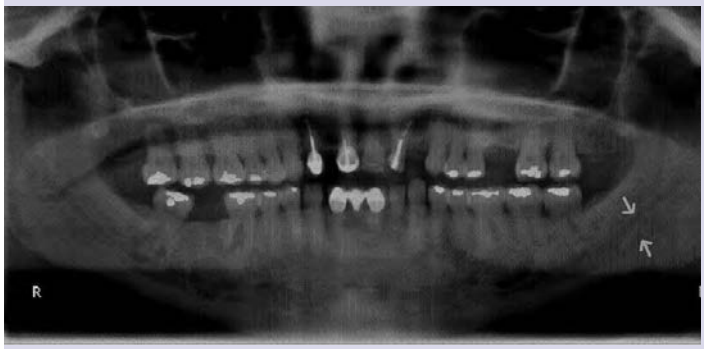


Figure 1. Panoramic X-ray revealed multiple radiolucent /radiopaque areas in right and left quadrants of mandible and suggestion of possibility of two inferior alveolar canals at left mandible.

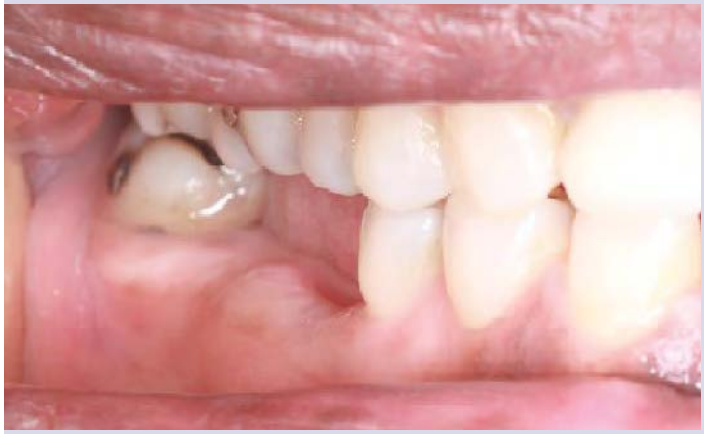


Figure 2. Four weeks after extraction of mandibular right first molar.

Occasionally, simple bone cysts occur associated with this lesion, and surgical exploration may be necessary to establish the diagnosis.[12]

When this condition is identified, preventative management for caries and periodontal disease to maintain natural teeth and prevent the need for extractions should be considered. Oral hygiene advice should be dispensed to prevent periodontal diseases. Osseointegration of implants would likely not be successful in these patients because the bone is abnormal and not well-vascularized. However, no studies have explored osseointegration in patients with this condition.[8]

Tissue-borne partial dentures and complete dentures are problematic because of the inconsistent resorption of alveolar bone. The tissues underneath tissue-borne prostheses would be prone to trauma and potential secondary infection, which could lead to more complicated treatment in the future.

FCOD lesions are not treated unless the bone becomes secondarily infected and osteomyelitis develops. Treatment of the complicating osteomyelitis consists of debridement, drainage and antibiotic therapy.[7] Clinically, FCOD consists of asymptomatic lesions; biopsy and surgery are not necessary.[9] If a patient presents with symptoms in the sclerotic phase,

it becomes a management problem because an inflammatory component has been added and the disease may transform into osteomyelitis.[14] Simple bone cysts have been reported to occur in association with FCOD; therefore, timely diagnosis and well-planned treatment are important to obtain a good prognosis when a rare co-occurrence of two or more bone lesions affect the jaw.[12]

Rarely has FCOD been reported to cause expansion and rupture of cortical plates. Surgical treatment through removal of the masses has been reported to be effective.[10] The association of osteosarcoma and FCOD has been reported. The relationship between these two conditions is not clear and could represent a collision between a benign and malignant lesion.[11]

Conclusion

The cause of FCOD is unknown. It typically affects middle-age black women. However, cases have been reported in men and in Asian and Caucasian individuals. A familial pattern was demonstrated in a few reports. A CT scan could be useful in distinguishing FCOD from other lesions. Oral hygiene advice should be emphasized for patients with FCOD to prevent caries and periodontal diseases and to maintain natural teeth. Surgical intervention should be avoided unless the complication of osteomyelitis occurs. Treatment of the patient with osteomyelitis in FCOD is complex and may require hospitalization.

Queries about this article can be sent to Dr. Sadda at rs211@nyu.edu.

REFERENCES

1. Higuchi Y, et al. Clinical and histopathological studies of the cementoma: periapical cemental dysplasia and massive cemental lesion. J Oral Maxfac Surg 1986;32:1884-94.
2. Muroki T, et al. A case of the multiple cemental dysplasia in the maxilla and mandible. J Oral Maxfac Surg 1989;35:1254-60.
3. Waldron CA. Fibro-osseous lesions of the jaws. J Oral Maxfac Surg 1993;51:828-835.
4. Alsufyani NA, Lam EWN. Dentomaxfac Radio 2011;40:141. The British Institute of Radiology.
5. Ariji et al. Florid cemento-osseous dysplasia. Radiographic study with special emphasis on CT. Oral Surg, Oral Med Oral Path Sept 1994,391-396.
6. Gold L, Cleveland DB. Surgical pathology: fibro-osseous diseases: considerations in diagnosis and management. Oral and Maxfac Surg. Clinics of North America Nov 1997: 648-652.
7. Sapp JP, et al. Contemporary oral and maxfac path. Mosby Year Book 1997:92-93.
8. Misch C. Contemporary Implant Dentistry. 2nd ed. 1999, p. 61.
9. Beylouni I, et-al. Fibro-cemento osseous dysplasia. Report of a case documented with CT and 3D imaging. Oral Surg Oral Med Oral Path Oral Radiology Endo 1998;85:707-11.
10. Martini ZM, Pereira J, Carvalho Jr DE, Soares HA. Surgical management of an infected aggressive florid-cemento-osseous dysplasia. Report of a case. Minerva Stomatologica Oct 2006;55:515-21.
11. Lopes MA, et al. Clinico-path conference: case report. Head and Neck Path 2010;4:329-333.
12. Rao KA, et al. Co-occurrence of florid cemento-osseous dysplasia and simple bone cyst: a case report. J Oral Maxillofac Res 2011(July-Sept.);2(3):e5.
13. Singer SR, et al. Florid cemento-osseous dysplasia and chronic diffuse osteomyelitis. JADA July 2005;136:927.
14. Neville BW, Damm DD, Allen CM, et al., eds. Oral and Maxillofacial Pathology. 2nd ed. Philadelphia, PA: Saunders; 2002:533-588.

Calcifying Odontogenic Cyst

Dilemma in Classification

Adesh S. Manchanda, B.D.S., M.D.S.; Ramandeep S. Narang, B.D.S., M.D.S.

ABSTRACT

Calcifying odontogenic cyst (COC) shows extreme diversity in its clinical and histopathological features, as well as in its biological behavior. Because of this diversity, there has been confusion and disagreement on the terminology and classification of this lesion. Attempts to classify COC can be divided into two concepts: monistic and dualistic. We present a case of COC with coexisting histopathologic features of a cyst and a neoplasm, thus posing a dilemma in the terminology used to categorize and classify it.

The term calcifying odontogenic cyst has been commonly used since the first description by Gorlin et al.[1] in 1962. Although it was recognized as a distinct pathologic entity at first, the COC shows extreme diversity in its clinical and histopathological features, as well as in its biological behavior. Because of this diversity, there has been confusion and disagreement on the terminology and classification of this lesion.[2]

Attempts to classify COC can be divided into two concepts. The first concept is the "monistic," in which all COCs are neoplastic in nature, even though the majority are cystic in architecture and appear to be non-neoplastic. The second is the "dualistic" concept that posits that COC contains two entities: a cyst and a neoplasm.[3] Radiographically, the lesion appears as a well-defined, uni- or multilocular, often quite large radiolucency that may or may not contain varying amounts of radiopaque material; some lesions are associated with unerupted teeth. Histologically, the lining epithelium of the cyst often exhibits extensive luminal and intramural proliferation, sometimes resembling an ameloblastoma; numerous daughter cysts are often found as well. Moreover, the COC is frequently found in association with, or exhibits areas histologically similar to, various odontogenic tumors, including complex and compound odontomas and ameloblastoma.[2]

We report a case of COC whose diversity in histopathology poses a dilemma in the terminology used to categorize and classify it.

Case Report

A 16-year-old male reported to SGRD Institute of Dental Sciences with a chief complaint of swelling in the left mandibular posterior region. On evaluation, there was an asymmetry involving the left side of the body of the mandible. Intraoral examination revealed buccal as well as palatal cortical expansion. Radiographic examination disclosed a unilocular, well-circumscribed, round radiolucency extending from tooth #17 to tooth #19, with #17 and #18 impacted. The differential diagnosis included dentigerous cyst and unicystic ameloblastoma. A biopsy was performed, and a cystic lesion with a thickened cyst wall in some areas and impacted second and third molars were observed.

Microscopic examination showed a cystic cavity lined by a non-keratinized odontogenic epithelium of 4 to 10 cells in thickness that exhibited isolated as well as clusters of fusiform ghost cells (Figure 1), some of which were calcified. There were areas of non-proliferating epithelial lining (Figure 2) and areas where the epithelial lining showed intramural as well as intraluminal

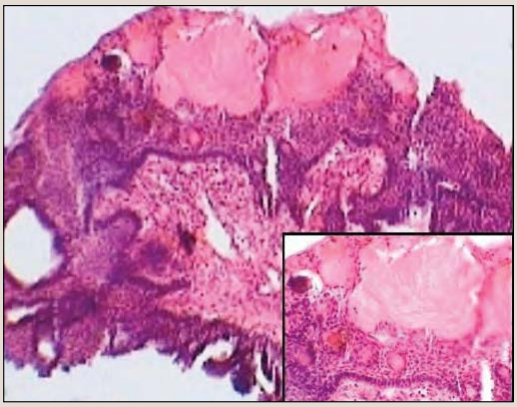


Figure 1. Cystic cavity lined by a non-keratinized odontogenic epithelium exhibiting ghost cells (H & E, X4). Inset showing isolated and clusters of fusiform ghost cells (H & E, X10).

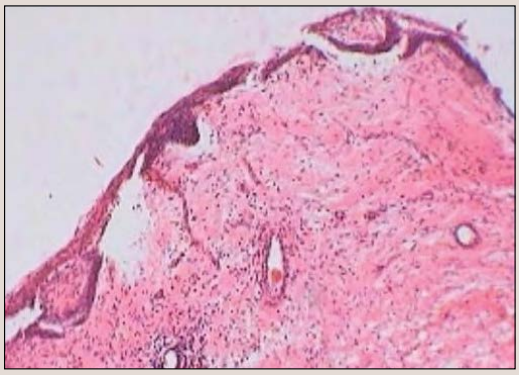


Figure 2. Non-proliferative epithelial lining (H & E, X10).

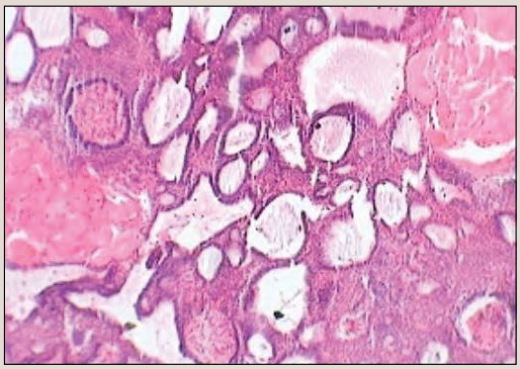


Figure 3. Epithelial lining showed proliferations in plexiform pattern (H & E, X10).

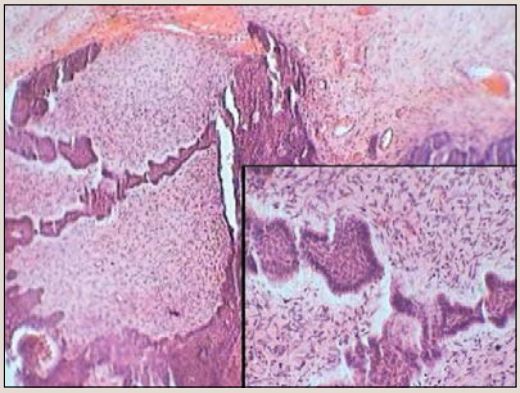


Figure 4. Connective tissue stroma varying from mature (right) to immature (left) areas with presence of ameloblastic fibroma (H & E, X4). Inset showing islands of ameloblastic fibroma (H & E, X10).

proliferations of classic SMA tissue in a plexiform pattern (Figure 3), with histological features of ameloblastoma. The transformed ameloblastomatous portion showed little or no ghost cells. The connective tissue stroma varied from mature to immature, with the presence of odontogenic islands. Few of these odontogenic islands resembled an ameloblastic fibroma in a cellular ectomesenchymal tissue mimicking a dental papilla (Figure 4).

Discussion

It is presumed that the COC originates from odontogenic epithelium.[1] However, there has been much discussion as to the possible histogenesis of the lesion. Shear[4] raised the question of whether those COCs, which also have features of other odontogenic tumors, develop these secondarily, or whether the COCs are themselves secondary phenomena in pre-existing odontogenic tumors. The latter theory was supported by Fejerskov and Krogh.[5] However, Abrams and Howell[6] pointed out that the combined occurrence of the COC with another odontogenic lesion could be expected because of the multipotentiality of the odontogenic epithelium. They believe that a COC in association with an odontoma results from the processes of differentiation and degeneration of the odontogenic epithelium. Furthermore, Praetorius et al.[7] believed that what appears to be an associated odontogenic tumor is simply an integral part of the entire lesion developing from the wall of COC. The accepted classification of COC as given by Praetorius is highlighted in Table 1.

TABLE 1
Suggested Classification of COCs[3]

Type 1. Non-neoplastic (simple cystic) Variant (CGCOC[a]) a. with nonproliferative epithelial lining b. with nonproliferative (or proliferative) epithelial lining associated with odontomas[b] c. with proliferative epithelial lining d. with unicystic, plexiform ameloblastomatous proliferation of epithelial lining[c]
Type 2. Neoplastic Variants A. Benign type (CGCOT[d]): a. cystic subtype (cystic CGCOT) α) SMA ex epithelial cyst lining[e] b. solid subtype (solid CGCOT) α) Peripheral ameloblastoma-like[f] β) SMA-like[g] B. malignant type (malignant CGCOT or OGCC[h]): a. cystic subtype b. solid subtype
a Calcifying ghost cell odontogenic cyst. b Also classified as compound (or complex) cystic ghost cell odontomas. c Does not completely fulfill the histopathologic criteria of early ameloblastoma as suggested by Vickers and Gorlin. d Calcifying ghost cell odontogenic tumor. e With histopathologic features of early ameloblastoma as suggested by Vickers and Gorlin. f Resembling a peripheral amelobastoma, hence termed peripheral epithelial odontogenic ghost cell tumor. g Often called central epithelial odontogenic ghost cell tumor. h Odontogenic ghost cell carcinoma.

Although the true nature of the lesion-cyst or tumor remains controversial, its behavior is, nevertheless, benign. The classification is contentious; some authors consider the lesion to be a special type of odontogenic cyst, while WHO classifies the lesion under benign neoplasms and other tumors related to the odontogenic apparatus. This follows the hypothesis that the formation of dysplastic dentine results from abnormal induction changes occurring in the mesenchyme as a result of the influence of odontogenic epithelium. This interpretation is further supported by the fact that the cyst is often encountered together with other odontogenic tumors.[8]

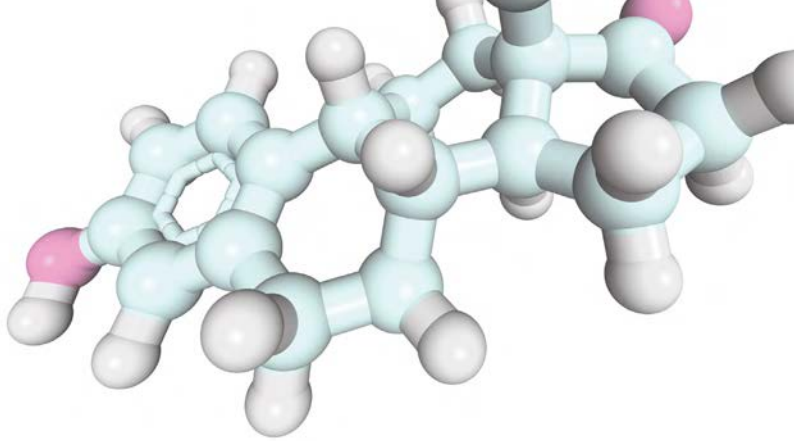
In the case presented here, the cystic lining in some areas resembles a non-neoplastic (simple cystic) variant with a non-proliferative epithelial lining Type 1a.[7] In other areas, it resembles a benign type of neoplastic variant with SMA ex-epithelial cyst lining (Type 2Aaα).[7] Furthermore, the connective tissue stroma in the case presented here exhibits islands resembling ameloblastic fibroma, which have not been categorized in the present classification. Thus, our case showed non-neoplastic areas, benign neoplastic areas and areas resembling ameloblastic fibroma. In this case, it is difficult to interpret whether the epithelial changes have developed in a pre-existing cyst wall or whether cystic degeneration has taken place in the center of a proliferating odontogenic island. Such diverse, multifaceted histopathology poses a challenge in classifying and categorizing the lesion.

It remains very difficult to determine whether the lesion is truly cystic (non-neoplastic), neoplastic or a probable hybrid variety. However, multiple co-existent lesions associated with COC have not been addressed in any of the previous classifications. Further extensive and systematic analysis of such cases is required to resolve this problem.

Queries about this article can be sent to Dr. Manchanda at adesh_manchanda@yahoo.com.

REFERENCES

1. Gorlin RG, Pindborg JJ, Clausen FP, Vickers RA. The calcifying odontogenic cyst—a possible analogue of the cutaneous calcifying epithelium of Malherbe. Oral Surg Oral Med Oral Pathol 1962;15:1235-43.
2. Toida M. So-called calcifying odontogenic cyst: review and discussion on the terminology and classification. J Oral Pathol Med 1998, 27:49-52.
3. Philpsen HP, Reichart PA. Odontogenic Tumors and Allied Lesions. 1st ed. Hanover Park, IL: Quintessence Publishing, 2004, pp.155-170.
4. Shear M, Speight P. Calcifying odontogenic cyst (Calcifying cystic odontogenic tumour). In: Cyst of the Oral and Maxillofacial Region. 4th ed. New York, NY: Blackwell Munksgaard, 2007, pp. 100–107.
5. Fejerskov O, Krogh J. The calcifying ghost cell odontogenic tumour or the calcifying odontogenic cyst. J Oral Pathol 1972;1:273-87.
6. Abrams AM, Howell FV. The calcifying odontogenic cyst. Report of four cases. Oral Surg Oral Med Oral Pathol 1968; 25:594-606.
7. Praetorius F, Hjorting-Hansen E, Gorlin RJ, Vickers RA. Calcifying odontogenic cyst. Range, variations and neoplastic potential. Acta Odontol Scand 1981;39:227-40.
8. Lello GE, Makek M. Calcifying odontogenic cyst. Int J Oral Maxillofac Surg 1986;15:637-44.



The Relationship between Estrogen, Estrogen Receptors and Periodontal Disease in Adult Women

A Review of the Literature

Laurence F. Shapiro D.D.S.; Katherine Freeman, Dr. P.H.

ABSTRACT

The literature supports the fact that estrogen plays an important role in skeletal maintenance and remodeling. Estrogen, acting through estrogen receptors in the cells of the periodontal ligament, has a regulatory interaction on bone dynamics through a complex set of basic multicellular units (BMUs). Deficiency of estrogen results in an increased number of BMUs and enhanced bone turnover. The impact of the changes in estrogen deficiency on bone dynamics is primarily mediated through osteoclasts, with greater interdiction of estrogen's actions on trabecular bone than on cortical bone. The purpose of this manuscript is to review the literature for evidence to support an association between estrogen and periodontal disease in adult women, as well as bone mineral density, and to help clarify the mechanism of action.

We found in our review of all pertinent databases, including Cochrane, that there are few peer-reviewed clinical studies that examine the relationship between estrogen deficiency and periodontal disease, and bone mineral density (BMD) and periodontal disease. Thus, future research is needed to investigate these associations so that at-risk patients can be identified earlier to avoid functional and esthetic sequellae of periodontal disease.

Periodontal disease is seen more frequently in postmenopausal women whose menopausal status has been altered either surgically or as a side effect of chemotherapy. Oral bone loss may also be an indicator of periodontal disease.[1,2] However, the specific nature of the relationship between estrogen levels and periodontal disease is still being investigated.

A systematic review of 64 published papers between 1990 and 2008 was conducted. It suggested that women who were post-antineoplastic therapy had a higher plaque index, increased gingival inflammation and bleeding upon probing compared to healthy controls. With regard to periodontal disease, the weighted prevalence of severe gingivitis was 20.3% higher compared to the healthy control group.[1] (Most chemotherapeutic agents affect estrogen levels by causing the death of ovarian cells.) This is a result of the cytostatic effect of chemotherapy agents. These

drugs prevent cell division of cancer cells and also prevent cellular mitosis. However, normal cells are also killed, especially sensitive reproductive cells in the ovaries. This is known as "chemotherapy induced ovarian failure," which, in turn, causes a large decrease in estrogen that affects changes in bone mineral density. These changes are mediated by estrogen receptor alpha (ERα) and estrogen receptor beta (ERβ.)

In pre-clinical studies, investigators have shown that oral mucosa and salivary glands are sensitive to estrogen. Estrogen receptors are found in all layers of the gingival epithelium, buccal mucosa and salivary glands. Estrogen receptors exist as two subtypes: estrogen receptor alpha (ERα) and (ERβ).[3]

Some research articles have shown that ERα was completely undetected in oral tissues, whereas ERβ was expressed in high levels in oral epithelium and salivary glands.[4,5] The differential expression of ERβ in these tissues may account for the conflicting results in estrogen receptor expression in earlier studies. In this study, the results show that a tissue-specific subtype distribution is also observed in oral tissues with ERβ but not ERα. ERα is usually expressed in classic target tissues, such as breast tissue. The conflicting results in some of the studies may have had to do with the specificity of the testing methods used. It has been concluded that there is a tissue-specific expression of the ERβ subtype. "The identification of ERβ in these tissues has significant clinical importance and suggests a direct role for estrogen in the physiology of the oral mucosa and salivary gland function."[4] Forty percent of these cells demonstrate ERβ immunoreactivity.

In a study by Pan, Zhang et al.,[4] both estrogen receptors, ERα and ERβ, have been detected in periodontal ligament cells (PDLCs). And both receptors have been shown to stimulate the bone formation capacity of cultured PDLCs by increasing alkaline phosphatase (ALP) activity, osteocalcin distribution and the formation of mineralized nodules.[6,7] These compounds are markers for bone formation.

ALP activity was much higher in the estrogen-treated periodontal ligament stem cells (PDLSCs) than in the control group.[6] Additionally, in the estrogen-treated groups, the ALP activity was stimulated in a dose-dependent manner. The higher expression of ERα and ERβ in PDLSCs as compared to PDLCs indicates a potential involvement of ERα and ERβ in the process of estrogen-induced osteogenic differentiation of PDLSCs.[8]

Evidence suggests that insufficient estrogen levels are a major cause of osteopenia and postmenopausal osteoporosis. Metabolic diseases that may also affect the levels of estrogen include hyperparathyroidism, hypopituitarism, Cushing's disease, adrenal insufficiency, rheumatoid arthritis and malignant disease. Several studies suggest that osteoporosis is a risk factor for periodontal disease and tooth loss.[2-5,7] As estrogen plays an important role in maintaining normal bone turnover, the genes for ERα and ERβ have been considered as possible candidate genes that might influence bone mass and osteoporotic risk.[4-6] Cells of the periodontal ligament are capable of producing all of the structures of the attachment apparatus. They can differentiate into osteoblasts (bone formation), cementoblasts (cementum formation) and fibroblasts (collagen formation).[8-10] These cells also modulate the production of osteoclasts, which induce the breakdown of bone.[5,8] Estrogen plays a regulatory role in maintaining the balance between osteoblast production and osteoclast production. This role of estrogen is vital to maintaining normal bone mass and bone density. Thus, a relationship between estrogen levels and periodontal disease is plausible.

The role of estrogen is vital to maintaining normal bone mass and bone density. Thus, a relationship between estrogen levels and periodontal disease is plausible.

During menopause, the onset of ovarian deficiency (resulting in a decrease in estrogen levels) affects bone mass density (BMD). Normal loss of BMD is 0.7 percent per year during menopause. This may be explained by the inhibition of the down-regulating effects of osteoclasts, resulting in more bone breakdown than bone apposition.[8,11] Other effects include loss of keratin, thinning of gingival tissues, redness, soreness and decreased salivary gland function. Estrogen can also modulate the pathogenicity of periodontal pathogens, such as P. Gingivalis and P. Intermedia; and it is reasonable to expect that its reduction may imply more severe periodontitis.

In a study involving ovariectomized rats with induced periodontitis, by Ainbinder, Prado, et al.,[13] the conclusion drawn was that estrogen deficiency could not be considered a risk factor for periodontal disease. The literature shows that the effects of estrogen deficiency on bone characteristics such as size, mass and density are site-dependent. This study could not relate the absence of ovarian hormones with periodontal alterations. A consensus has not been reached regarding the relationship between ovariectomy and periodontal disease.[12]

A study by Bin Zhang, Ying Li, et al.,[14] utilizing ovariectomized rats with induced periodontitis and a SHAM control, explored the effect of estrogen on the potential for osteogenic differentiation of periodontal ligament stem cells. The results showed there was a lower expression of estrogen receptors (ERα and ERβ) in the ovariectomized group than the sham group. Treatment with 17β estradiol significantly increased osteogenic differentiation of PDLSC in both groups in vitro. The results seem to indicate that estrogen plays an important role in maintaining osteogenic differentiation of periodontal stem cells, which act through ERα and ERβ.[13]

Evidence suggests that "estrogen deficiency leads to impaired osteogenic differentiation of periodontal stem cells in rats."[14-17]

Stossi et al.[4] demonstrated that both ERα and ERβ transcriptionally up-regulated bone morphogenic protein-6 (BMP-6), a key factor in bone formation. This supports the findings of the present study by Feng Pan, Zhang, et al.[4] that both ERα and ERβ may function in the osteogenic differentiation of periodontal stem cells. This difference may be explained by the fact that in the different types of cells, the varied expression patterns of ERα and ERβ are under the control of specific mechanisms.

Investigators have also considered the role of a variety of inflammatory cytokines and growth factors and the effect of estrogen on the gingival fibroblast. Cytokines, such as interleukins and interferons, are substances that are produced by specific cells of the immune system and have an immune modulating effect and are critical to the functioning of the immune system.[15]

In a human study by Shu, Guan, et al.,[16] it was shown that estrogen deficiency can provoke an imbalance in the remodeling sequence of periodontal tissues. The study was designed to explore the modulatory effect of estrogen on bone-resorbing cytokines such as TNF-alpha, IL-1B, IL-6 and RANKL and the anti-resorptive factor osteoprotegerin (OPG) in periodontal ligament cells.

Some of the conclusions from this study are as follows:

- 17-β estradiol (E2) may not alter the ability of human periodontal ligament cells to produce pro-inflammatory cytokines, but it may modify the stimulatory effect of lipopolysaccharides (LPS) on pro-inflammatory cells.[16]
- Periodontitis is a chronic inflammatory disease characterized by gingival inflammation and alveolar bone resorption. It is generally accepted that much of the periodontal tissue destruction observed is host-mediated through inflammatory cytokines produced by local tissues and immune cells in response to bacterial flora and its products or metabolites, especially lipopolysaccharides.
- Estrogen alters the expression of inflammatory cytokines, thereby exerting its bone sparing effects. These estrogen deficiency-induced changes on inflammatory cytokines may have significant impact on bone resorption in periodontal tissues.[16]

"The inhibitory effect of E2 on LPS-induced tumor necrosis factor alpha (TNF-α), IL-B and IL-6 synthesis can at least in part explain how E2 down regulates osteoclastogenesis, for all of these cytokines are thought to play important roles in osteoclastogenesis and may cause inflammatory resorption of alveolar bone by various mechanisms."[18]

In a study by Wattanaroonwong et al.,[18] it was shown that periodontal fibroblasts may promote osteoclastogenesis more strongly than gingival fibroblasts. 17-β estradiol (E2) inhibits the PLF-induced formation of osteoclast-like cells. Thus, estrogen has an inhibitory effect on osteoclast formation.[19] Since the discovery of RANKL, and its decoy receptor osteoprotegerin (OPG), it has been believed that the ratio between RANKL and OPG determines osteoclast differentiation and activation and that these two factors have emerged as essential mediators in the modulation of bone resorption.[18]

RANKL/RANK signaling regulates the formation of multinucleated osteoclasts from their precursors, as well as their activation and survival in normal bone remodeling and in a variety of pathologic conditions. OPG protects the skeleton from excessive bone resorption by binding to RANKL and preventing it from binding to its receptor, RANK. Thus, RANKL/RANK ratio is an important determinant of bone mass and skeletal integrity.[16] E2 increased OPG expression and attenuated the RANKL-inducing effects of lipopolysaccharides (LPS) on RANK. The LPS-induced decrease of OPG/RANKL ratio in human periodontal ligament cells was completely reversed by E2. This observation suggests that E2 may influence the progression of periodontal disease by altering the ratio of OPG/RANKL in human periodontal ligament cells. It was suggested in the study by Shu et al.[16] that estrogen may modulate OPG expression in a manner that is independent of TNF-alpha, IL-B and IL-6.

There is an important association between loss of bone mass and periodontal disease.[28]

Estrogen may suppress osteoclastogenesis by modulating the synthesis of cytokines produced locally by human PDLCs. There is now convincing evidence that estrogen, acting through ERα, stimulates osteoclast apoptosis[16] and, conversely, suppresses osteoblast and osteocyte apoptosis. Therefore, estrogen deficiency is associated with an increase in the lifespan of osteoclasts, as well as a concomitant decrease in the osteoblastic lifespan.[16] Estrogen deficiency up regulates osteoblastogenesis and expands the number of T cells. Both osteoblasts and T cells produce RANKL, which leads to enhanced osteoclast development.[11,17-20]

During the postmenopausal process, women lose 30% to 50% of the trabecular bone and 25% to 35% of the cortical bone mass that was present during the peak bone mass years between ages 20 to 30. (Normal bone loss averages 0.7% per year.)[21] Postmenopausal women with osteoporosis and concurrent periodontitis may show a loss of dentoalveolar bone height and decreased BMD of the alveolar crestal and subcrestal bone.[21]

Methotrexate in particular has an effect on osteoblast production and results in a decrease in mineralized surfaces and, therefore, affects bone mass density and matrix mineralization.[22,23] Methotrexate is a cytostatic drug and, thus, lowers estrogen production. Methotrexate has both direct and indirect effects on bone. The direct effects are due to the cytotoxic effects of methotrexate on osteoblastic activity and the proliferation of osteoblasts.[24] The indirect effects are caused by chemotherapy-induced ovarian failure.

Clinical research by Atkinson et al.[25] demonstrated a reduction in bone mineral content between six and nine months after MTX treatment. It is unknown if the bone mass remains depressed indefinitely, slowly recovers to normal levels or recovers to osteopenic levels below the normal range. It is known that one course of methotrexate therapy will induce osteopenia and depress bone formation 14 days following treatment.[25] Friedlander, et al.[26,27] also reported significant reduction (27%) in cancellous bone volume after one course of methotrexate.

There is an important association between loss of bone mass and periodontal disease.[28] Kribbs showed there was a significant correlation between skeletal bone mass measurements and the number of remaining teeth.[28] Yoshihara et al. and Klemetti et al. showed that the BMD of the mandible is affected by the mineral status of the skeleton and by any disease that causes generalized bone loss.[29] Ward and Manson were able to find an association between the periodontal disease index and alveolar bone loss.[31] Groen et al. assessed the relationship between osteoporosis or low bone density and clinical attachment loss. Toothlessness and severe periodontal disease were found among 38 patients ages 43 to 73 who exhibited clinical and radiographic signs of advanced osteoporosis.[32] Most studies showed a correlation between reduced bone mineral density and increased severity of periodontal disease.

Numerous studies on the effect of MTX were noted involving use of the chemotherapeutic agent in treating rheumatoid arthritis. The effects of low doses of MTX are long lasting and are present for over 170 days after cessation of therapy.[23-26]

In a study by Brennan, et al.,[33] it was shown that estrogen enhancement improves matrix mineralization by MC3T3 cells in vitro. Furthermore, this study found a significant reduction in the level of mineralization when cells are treated with a combination of estrogen and Fulvestrant (estrogen antagonist). The expression of osteocalcin and osteopontin proteins (markers of osteoblast differentiation and mineralization) was monitored by using immunohistochemistry.[32]

The findings from the Women's Health Initiative (W.H.I.) on 42,171 postmenopausal women showed the overall risk of tooth loss was 24% lower in current hormone replacement therapy (HRT) users when compared to nonusers. Furthermore, the results showed that estrogen may promote tooth retention by strengthening the periodontal attachment surrounding the teeth, without increasing oral bone height or decreasing oral bone porosity. Tagutchi et al.[34] and Grossi[35] further showed that women who were not treated with estrogen replacement therapy (ERT) were twice as likely as their ERT counterparts and three-times more likely than premenopausal women to exhibit severe attachment loss. The individual percentages of women affected by severe attachment loss were 18.6%, 11.9% and 6.3% for non ERT, ERT and premenopausal women, respectively.

Also, severe alveolar bone loss (ABL) was detected in 34%, 20.3% and 9.7% of the non-ERT, postmenopausal ERT, and premenopausal women, respectively. The authors, therefore, concluded that ERT appears to have a protective effect on the severity of periodontal disease and the periodontium.[35] The duration of estrogen use was significantly associated with the number of remaining teeth.

Discussion

At this time, a thorough review of all data bases revealed there are few studies that provide evidence of the direct association between estrogen deficiency and periodontal disease and between BMD and periodontal disease. Furthermore, there are no prospective observational studies to assess estrogen as a risk factor for periodontal disease that account for confounding factors such as age, socioeconomic status, nutrition, smoking and health status.

While the current concepts of inflammatory disease and tissue reaction to locally produced cytokines (TNF-α and interleukins) are regarded as modifying factors, the clinical significance of other mechanisms such as estrogen and estrogen receptors to account for changes in bone volume and density should be considered, especially in situations where causative inflammatory agents are not clinically evident.

There are many factors involved in the progression of periodontal disease in adult women. Clinicians have found that "HRT

(hormone replacement therapy) can improve the clinical outcome of periodontal disease and may serve as an effective adjunct treatment for preserving periodontal bone mass."[4]

Limitations

The review of the literature was limited to articles published in English only. Additionally, the studies were primarily qualitative. Other than W.H.I, no study was longitudinal, to evaluate possible casual relationships.

Despite these limitations, there is evidence to suggest an association between estrogen and periodontal disease, because estrogen receptors are found in the cells of the periodontal ligament, gingiva, salivary glands and jaw bone. Estrogen has an effect on these tissues and exerts its effect locally. These effects include maturation of gingival connective tissue, osteoblastic differentiation and mineralization. Estrogen deficiency will alter skeletal remodeling and, thus, affect bone mass density and bone volume.

Conclusion

Estrogen, acting through ERα andERβ located in the cells of the periodontal ligament, may have a significant impact on the periodontium. Estrogen receptors have a regulatory effect on both the maturation of gingival epithelium and on the osteoblastic differentiation of periodontal ligament cells. ERβ may play an important role in bone formation

Estrogen deficiency may result in osteoporosis, which is considered to be a risk factor for periodontal disease, loss of BMD and tooth loss. Both disease processes share common risk factors, are inflammatory in nature and are bone-resorptive entities.

Further research is needed to better establish the associations between periodontal disease, estrogen and bone mineral density, so that at-risk patients can be identified earlier to avoid the functional and esthetic sequellae of periodontal disease.

The authors thank student doctor Samantha Robles, B.S., for her assistance in proofreading their manuscript. Queries about this article can be sent to Dr. Shapiro at laurshap@nova.edu.

REFERENCES

1. Hong CH, Napenas JJ, Hodgson BD, et al. A systematic review of dental disease in patients undergoing cancer therapy. Support Care Cancer 2010;18:1007-1021.
2. Meurman JH, Tarkkila L, Tiltinen A. The menopause and oral health. Maturitas May 2009;63(1):56-62.
3. Valimaa H, Savolainen S, Soukka T, Silvoniemi P, Makela S, Kujari H, Gustafsson JA, Laine M. Estrogen receptor beta is the predominant estrogen receptor subtype in human oral epithelium and salivary glands. J Endocrinology 2004;180:55-56.
4. Pan F, Zhang R, Wang G, Ding Y. Estrogen receptors are involved in the osteogenic differentiation of periodontal ligament stem cells. Biosci Rep 2011;31:117-124.
5. Nebel D, Bratthall G, Ekblad E, Norderyd O, Nilsson BO. Oestrogen regulates D.N.A. synthesis in human gingival epithelial cells displaying strong estrogen receptor beta immunoreactivity. J Periodontal Res Oct 201146(5):622-8.
6. Tang X, Meng H, Zhang L, Hou J, Zhang F. Up-regulation of estrogen receptor-beta expression during osteogenic differentiation of human periodontal ligament cells. J Periodont Res 2008;43:311-321.
7. Liang L, Yu JF, Wang G, Ding Y. Effect of estrogen receptor beta on the osteoblastic differentiation function of human periodontal ligament cells. Archives of Oral Biology June 2008;53(6):553-557.
8. Cao M, Shu L, Li J, Su J, Zhang W, Wang Q, Guo T, Ding Y. The expression of estrogen receptors and the effects of estrogen on human periodontal ligament cells. Methods find Exp. Clin Pharmacol June2007;29(5):329-35.
9. Mamalis A, Markopulou C, Lagou A, Vrostos I. Estrogen regulates proliferation osteoblastic differentiation, collagen synthesis and periostin gene expression in human periodontal ligament cells through estrogen receptor beta. Arch Oral Biol May 2011;56(5):446-55.
10. Friedlander AH. The physiology, medical management and oral implications of menopause. J Am Dent Assoc Jan 2002;133(1):73-78.
11. Santen RJ. Effect of endocrine therapies on bone in breast cancer patients. J Clin Endocrinology & Metabolism. February 2011;96(2): 308-319.
12. Johnsson D, Wahlin A, Idvall I, Johnsson I, Bratthall G, Nilsson B-O. Differential effects of estrogen on DNA synthesis in human periodontal ligament and breast cancer cells. J Periodontal Res 2005(5):401-406.
13. Ainbinder AL, De Almeida Prado M, Spaulding M, Balducci I, Carvalho YR, Fernandes de Rocha R. Estrogen deficiency and periodontal condition in rats. A radiographic and macroscopic study. Braz Dent J 2006;17(3):201-207.
14. Zhang B, Li Y, Zhou Q, Ding Y. Estrogen deficiency leads to impaired osteogenic differentiation of periodontal ligament stem cells in rats. Tohoku J Experimental Med 2011;223(3):177-186.
15. Seymour RA. Effects of medications on the periodontal tissues in health and disease. Periodontology 2000 2006;40:120-129.
16. Shu L, Guan SM, Fu SM, Guo T, Cao M, Ding Y. Estrogen modulates cytokine expression in human periodontal ligament cells. J Dent Res 2008;87(2):142-147.
17. Zhang B, Li Y, Zhou Q, Ding Y. Estrogen deficiency leads to impaired osteogenic differentiation of periodontal ligament stem cells in rats. Tohoku J Experimental Med. 2011;223(3):177-86.
18. Wattanaroonwong N, Schoenmaker T, DeVries TJ, Everts V. Oestrogen inhibits osteoclast formation induced by periodontal ligament fibroblasts. Arch Oral Biol Mar 2011;56(3):212-9.
19. Hirb. A, Morgan EA, Uluckan O, Weilbaecher K. Skeletal complications of breast cancer therapies. Clin Cancer Res Oct 2006;162:6309s-6314s.
20. Hershman DL, McMahon DJ, Crew KD, Shao T, et al. Prevention of bone loss by Zolendronic Acid in premenopausal women undergoing adjuvant chemotherapy persists up to one year following discontinuing treatment. J Clinical Endocrinology & Metabolism. Feb 2010;95(2):559-566.
21. Greep NC, Guiliano AE, Hansen NM, Taketani T, Wang HJ, Singer FR. The effects of adjuvant chemotherapy on bone density in postmenopausal women with early breast cancer. Am J Med 2003:114:653-659.
22. Bruning PF, Pit MJ, DeJong-Bakker M, Van Den Ende A, Hart A, Van Enk A. Bone mineral density after adjuvant chemotherapy for postmenopausal breast cancer. Br J Cancer 1990;61:308-310.
23. Headley JA, Theriault RL, LeBlanc AD, Vassilopoulou–Sellin R, Hortobagyi GN. Pilot Study of Bone Mineral Density in Breast Cancer Patients Treated with Adjuvant Chemotherapy. Cancer Invest 1998;16:6-11.
24. Scheven BA, van der Veen. MJ, Damen CA, et al. Effects of methotrexate on human osteoblasts in vitro: modulation by 1,25 dihydroxy Vit.D. J Bone Miner Res 1995;10:874-80.
25. Atkinson SA, Fraher L, Gundberg CM, Andrew Pai M, Barr RD. Mineral homeostasis and bone mass in children treated for acute lymphoblastic leukemia. J Pediatr 1989;114:793-800.
26. Wheeler DL,VanderGriend TJ, Wronski.GJ, et al. The short and long-term effects of methotrexate on the rat skeleton. Bone Feb 1995;16(2):215-22.
27. Friedlander GE, Tross RB, Douganis AC, Kirkwood JM, Baron R. Effects of chemotherapeutic agents on bone. JBJS 1984;66A:602-607.
28. Kribbs PJ, Smith DE, Chestnut CH. Oral findings in osteoporosis Part II: relationship between residual ridge and alveolar bone J Prosth Dent 1983;50:719-724.
29. Yoshihara A, Seida Y, Hanada N, Miyazaki H. The relationship between bone mineral density and the number of remaining teeth in community dwelling older adults. J Oral Rehabil 2005;32:735-740.
30. Klemetti E, Vainio P, Lassila V, Alhava E. Cortical bone mineral density in the mandible and osteoporosis status in postmenopausal women. Scand J Dent Res 1993;101:219-223.
31. Ward VJ, Manson JD. Alveolar bone loss in periodontal disease and metacarpal index. J Periodontol 1973; 44:763-769.
32. Groen JJ, Menczel J, Shapiro S. Chronic destructive periodontal disease in patients with presenile osteoporosis. J Perio 1968;39:19-23.
33. Brennan O, O'Brien FJ, McNamara L. Estrogen plus estrogen receptor antagonists alter mineral production by osteoblasts in vitro. Hormone Metab Res Jan 2012;44(1):47-53.
34. Tagutchi A, Senada M, Suei Y, et al. Effect of estrogen use on tooth retention and oral bone height and oral bone porosity in Japanese postmenopausal women. Menopause 2004;11:556-562.
35. Grossi SG. Effect of estrogen supplementation on periodontal disease. Compend Contin Educ Dent Suppl 1998;22:S 30-6.

Read, Learn and Earn
CE
CONTINUING EDUCATION

An Unusual Affliction of the Tongue

Amarpreet Sabharwal, B.D.S., M.S.; Michael Hatton, D.D.S., M.S.; Alfredo Aguirre, D.D.S., M.S.

ABSTRACT

A dramatic presentation of a large ulcer on the dorsal tongue of a recently hospitalized patient is presented. The lesion was found to be nosocomial in origin, and consistent with traumatic ulcerative granuloma with stromal eosinophilia (TUGSE). A review of the current pathogenic mechanisms, differential diagnosis and management of TUGSE is included.

An ulcer is a soft tissue defect characterized by full-thickness loss of overlying epithelium, extending beyond the basement membrane. Most oral ulcers are painful due to exposure of nerve endings to the oral environment. Although ulcers in general have overlapping clinical and histopathological features, there are subtle clues that may guide the practitioner toward a definitive diagnosis. Oral ulcers may vary in shape, number, location, age of onset and duration. These features, coupled with possible etiological factors, can be used to obtain a sound initial clinical impression.[1-4] This issue has been explored comprehensively, and efforts have been made to develop clinically based diagnostic tools for the recognition and classification of oral ulcers.[5]

Traumatic ulcerations of the oral mucosa are the most common type of ulcers.[2] They usually result from an intimate relationship between dental hard tissues and mucosal surfaces. The extensive mobility of soft tissues such as the tongue and, to a lesser extent, labial and buccal mucosa, may result in a higher risk for traumatic events. Exposure to sharp foodstuffs, faulty toothbrushing habits and chemical insults are also important factors.[5,6]

Traumatic ulcerative granuloma with stromal eosinophilia (TUGSE) is a unique self-limiting lesion of putative traumatic genesis.[3,7] It may however, present with ominous clinical features. Because of its clinical appearance and unusual biological behavior, TUGSE has to be differentiated from serious conditions, such as malignancies, deep fungal and bacterial infections, and chronic ulcerative immune-mediated conditions.[3,8,9]

We present in this paper an unusual case of lingual TUGSE that was associated with trauma induced by tracheal intubation. In addition, we discuss its pathogenic mechanism and management.

Case Report

A 60-year-old Caucasian female presented to the Advanced Education in General Dentistry (AEGD) Clinic at the University at Buffalo School of Dental Medicine. Her chief complaint was "My tooth is loose and I have not been able to taste food well for a while." Her past medical history included hypertension, rheumatoid arthritis and an allergy to penicillin. Medications included naproxen 275 mg, lisinopril 20 mg, lovastatin 10 mg and chon-

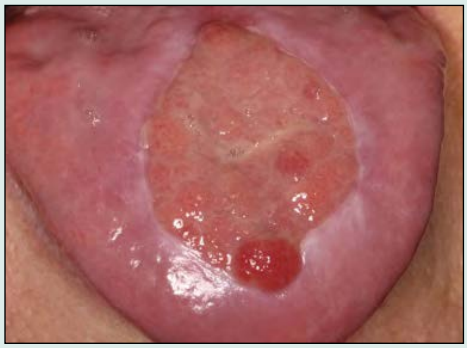


Figure 1. Initial presentation of lesion on dorsum of tongue.

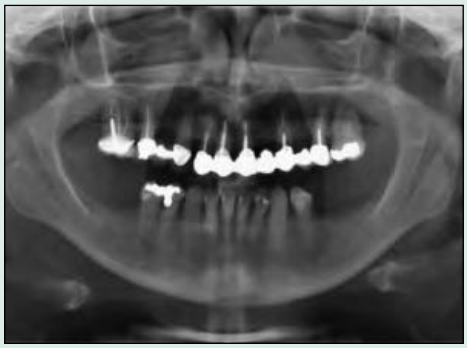


Figure 2. Panoramic film exhibiting bilateral radiopacities in carotid area.

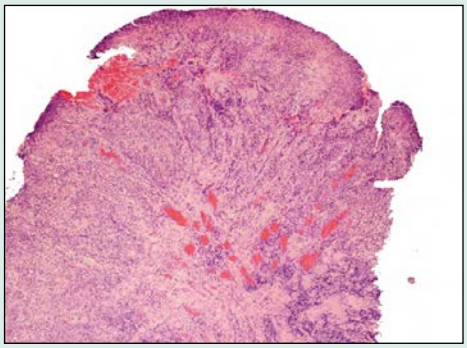


Figure 3. Necrosis of surface epithelium covered by fibrin mesh is seen in upper field of this photomicrograph. In addition, superficial and deep inflammatory cell infiltrates and hyperemic blood capillaries are observed. (Hematoxylin-eosin stain; magnification 40x.)

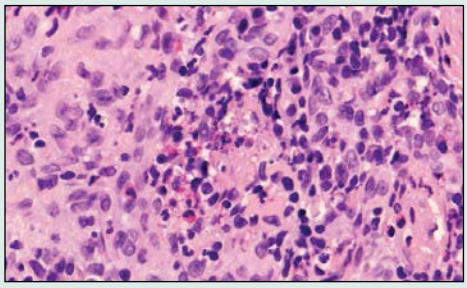


Figure 4. High magnification showing granulation tissue with numerous interspersed, conspicuous eosinophils, neutrophils and lymphocytes amid proliferating blood capillaries. (Hematoxylin-eosin stain; magnification 200x.)

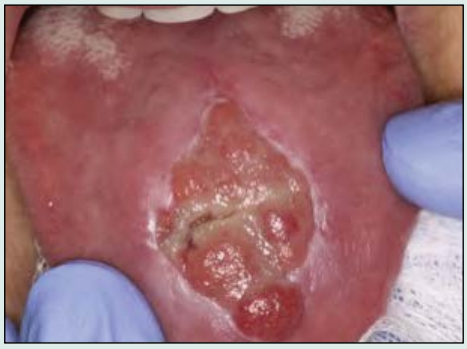


Figure 5. One-week post biopsy exhibiting normal healing of biopsy site with persistence of lesion.

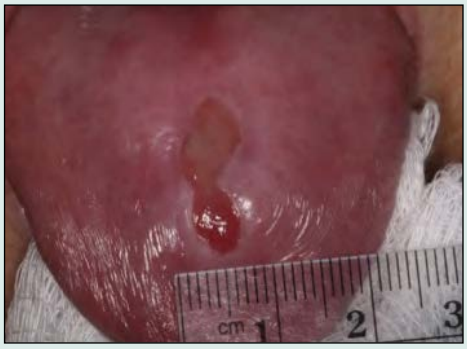


Figure 6. One-month post biopsy and three weeks of topical applications of compounded steroid/antifungal medication.

droitin sulfate 300 mg daily. The AEGD resident noted gross alteration of the dorsum of the tongue. An oral and maxillofacial surgeon and oral and maxillofacial pathologist were asked to consult on the patient.

Intraoral examination revealed a large ulceration on the anterior two-thirds of the dorsal tongue, measuring 2.7 cm x 1.8 cm (Figure 1). The lesion was indurated, exophytic, erythematous, tender on palpation and showed extensive loss of filliform and fungiform papillae. No other oral mucosal abnormalities were noted. The neck was clinically normal to examination. A detailed conversation with the patient revealed a significant nosocomial event that occurred three months prior to our initial oral examination. The patient had been admitted to a local hospital because of pneumonitis. Our communication with the patient's physician disclosed that she had been intubated with an oral tube for one week during her hospital stay. After resolution of the pulmonary infection, the patient was discharged uneventfully from the hospital. While recuperating at home, she noticed that her tongue was sore and felt swollen. As part of the initial physical examination, a panoramic film was also obtained. It revealed bilateral radiopacities in the area of the carotid arteries (Figure 2).

To further characterize the bilateral radiopacities seen on panoramic image, we requested a Doppler ultrasound imaging study on carotid arteries, results of which revealed mild bilateral plaques at the level of the carotid bulbs. No evidence of hemodynamically significant stenosis was found (<40% blockage), and no bruits were noted on auscultation of the carotid arteries. Our initial clinical impression was a traumatic ulcer due to a prolonged oral intubation. A two-week follow-up showed no resolution of the lesion, and an incisional biopsy was obtained. The patient was also referred for serum laboratory studies to rule out nutritional deficiencies. Serum levels of iron, folic acid, vitamin B12, niacin, riboflavin and pyridoxine were all within normal limits.

The histological diagnosis was "Non-specific ulcer with florid granulation tissue and features of traumatic ulcerative granuloma with stromal eosinophilia" (Figures 3 and 4). One week postoperatively, normal healing of the biopsy incision site was noted (Figure 5). However, the overall size and clinical appearance of the lesion remained unchanged. We then prescribed a compounded ointment of 0.05% clobetasol and 1% clotrimazole for topical application four times per day. Evaluation of the lesion was done at intervals of two weeks, one month (Figure 6) and two months (Figure 7). After two months, the lesion exhibited virtually complete resolution but displayed extensive loss of the filliform and fungiform papillae. The patient remains free of any mucosal lesions on multiple follow-up appointments.

Discussion

TUGSE is a self-limiting ulcerative lesion of the oral mucosa that frequently affects the tongue of adults. The term TUGSE was proposed by Elzay in 1983.[11] Using an experimental murine model, Bhaskar and Lilly[12] showed a relationship between trauma and TUGSE lesions. However, it is now well documented that not all TUGSE cases have a history of trauma.[11-14] It has also been suggested that microbial toxic products may result in an exaggerated eosinophil response.[11,15] Although, an ultra-structural investigation failed to show any microbial products, it revealed the presence of occluded blood vessels, which could lead to focal ischemia, thus magnifying the size of the ulcer.[15]

Demographic data from the two largest case series on TUGSE show a slight female predilection, with most lesions seen in the 5th to 7th decades of life, and lasting from a few days to several months. While the average age of presentation was 57 years, the lesions occurred over a very wide age range (6-92 years); the dorsum and lateral surfaces of the tongue were involved in about half of the cases. The buccal mucosa was the second most commonly involved site.[11,13] Interestingly, in the second largest case series published on TUGSE, only 7/38 cases (18%) revealed a history of trauma.[13]

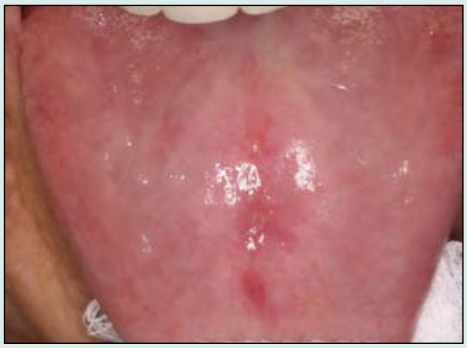


Figure 7. Two months post biopsy with continued use of topical medication showing only two small areas of superficial erosion.

Clinically, TUGSE can resemble a wide variety of pathologic entities (traumatic, infectious, neoplastic, autoimmune and reactive). Pain is typically associated with ulcerative lesions of inflammatory origin on oral mucosal surfaces. However, in the largest case series published so far in the English language literature, only 17% of the TUGSE lesions presented with pain.[11] The presence of indurated borders and the failure to heal mimic the clinical presentation of oral neoplasia. In addition, primary malignan-

Pain is typically associated with ulcerative lesions of inflammatory origin on oral mucosal surfaces.

cies from minor salivary glands, metastasis and hematological disorders may also present as ulcerative lesions on the tongue. Infectious diseases like histoplasmosis, blastomycosis, tuberculosis and oral manifestation of systemic diseases such as Wegener's granulomatosis, lupus erythematosus and sarcoidosis may also mimic TUGSE.[3,7-10]

Histologically, a typical TUGSE lesion presents with an area of ulceration covered by a fibrin mesh with interspersed neutrophils. The underlying connective tissue consists of granulation tissue with CD30 + T-lymphocytes, histiocytes, plasma cells and mast cells. Of significant concern is the predominance of CD 30+ T-lymphocytes, some of which have shown monoclonal TCRγ rearrangement, raising the possibility of the emergence of a CD 30+ lymphoproliferative disorder of T cells.[14,16,17] The most conspicuous microscopic finding is the presence of intact and degranulating eosinophils in the stroma.[3,11,13] Stromal eosinophilia in oral mucosal diseases is commonly seen in fungal and parasitic infections, vesiculobullous diseases and some rare disorders, such as Kimura's disease, angiolymphoid hyperplasia with eosinophilia and Langerhans cell histiocytosis.[18] The significance of eosinophilia in TUGSE remains a mystery. The studies of Elvoic et al.[19] have shown deficient tumor growth factor (TGF) levels in TUGSE compared to normal wound-healing processes. It is well known that T cells secrete eosinophil chemotactic (interleukin-1 and tumor necrosis factor) and maturation factors (interleukin-5). It is thus plausible that an increase in the indigenous T-cell population leads to active recruitment of eosinophils in TUGSE.[13]

Although TUGSE is a self-limiting disease, persistent cases may require intervention. In such cases, it has been documented in the literature that a biopsy may promote the restoration of the typical cytokine microenvironment associated with normal wound healing.[19,20-22] Thus, we believe this diagnostic procedure should be the first step in both establishing a diagnosis and in treating such lesions. When healing is not observed, despite a biopsy, intra-lesional infiltration of triamcinolone and/or topical corticosteroid application has been proposed as a treatment option.[10,23] We decided to compound an ultra-potent topical steroid (0.05 % clobetasol ointment) with 10 mg clotrimazole for daily q.i.d. topical applications. The results were highly satisfactory and led to complete resolution of this recalcitrant lesion.

Queries about this article can be sent to Dr. Hatton at hatton@buffalo.edu.

REFERENCES

1. Langlais RP, Miller CS, Nield-Gehrig JS. Color atlas of common oral diseases. 4th ed. Lippincott Williams & Wilkins 2009:18-9.
2. Porter SR, Leao JC. Review article: oral ulcers and relevance to systemic disorders. Aliment Pharmacol Ther 2005;21(4):295-306.
3. Segura S, Pujol RM. Eosinophilic ulcer of the oral mucosa: a distinct entity or a non-specific reactive pattern? Oral Diseases 2008;14(4):287-95.
4. Munoz-Corcuera M, Esparza-Gomez G, Gonzalez-Moles MA, Bascones-Martinez A. Oral ulcers: clinical aspects. A tool for dermatologists. Part II. Chronic ulcers. Clin Exp Dermatol 2009;34(4):456-61.
5. Compilato D, Cirilo N, Termine N, Kerr AR, Paderni C, Ciavarella D, Campisi G. Long-standing oral ulcers: proposal for a new 'S-C-D classification system.' J Oral Pathol Med 2009;38(3):241-53.
6. Neville BW, Damm DD, Allen CM, Bouquot JE. Oral and Maxillofacial Pathology. 3rd ed. Elsevier 2009:287-9.
7. Eleni G, Panagiotis S, Andreas K, Georgia A. Traumatic ulcerative granuloma with stromal eosinophilia: a lesion with alarming histopathologic presentation and benign clinical course. Am J Dermatopathol 2011;33(2):192-4.
8. Laskaris G. Color atlas of oral diseases. 3rd ed. Thieme 2003:64.
9. Gopalakrishnan R, Miloro M, Allen CM. Indurated ulceration of the tongue. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 1996;82(3):119-21.
10. Sklavounou A, Laskaris G. Eosinophilic ulcer of the oral mucosa. Oral Surg Oral Med Oral Pathol 1984;58(4):431-6.
11. Elzay RP. Traumatic ulcerative granuloma with stromal eosinophilia (Riga-Fede disease and traumatic eosinophilic granuloma). Oral Surg Oral Med Oral Pathol 1983;55(5):497-506.
12. Bhaskar SN, Lilly GE. Traumatic granuloma of the tongue (human and experimental). Oral Surg Oral Med Oral Pathol 1964;18(2):206-18.
13. El-Mofty SK, Swanson PE, Wick MR, Miller AS. Eosinophilic ulcer of the oral mucosa: report of 38 new cases with immunohistochemical observations. Oral Surg Oral Med Oral Pathol 1993;75(6):716-22.
14. Hirshberg A, Amariglio N, Akrish S, Yahalom R, Rosenbaum H, Okon E, Kaplan I. Traumatic ulcerative granuloma with stromal eosinophilia: a reactive lesion of the oral mucosa. Am J Clin Pathol 2006;126(4):522-9.
15. Tang TT, Glicklich M, Hodach AE, Oechler HW, McCreadie SR. Ulcerative eosinophilic granuloma of the tongue: a light electron-microscopy study. Am J Clin Pathol 1981;75(3):420-25.
16. Segura S, Romero D, Masarco Jr JM, Colomo L, Ferrando J, Estrach T. Eosinophilic ulcer of the oral mucosa: another histological simulator of CD30+ lymphoproliferative disorders. Br J Dermatol 2006;155(2):460-63.
17. Salisbury CL, Budnick SD, Li S. T-cell receptor gene rearrangement and CD30 immunoreactivity in traumatic ulcerative granuloma with stromal eosinophilia of the oral cavity. Am J Clin Pathol 2009;132(5):722-27.
18. Saraswathi TR, Nalinkumar S, Ranganathan K, Umadevi R, Elizabeth J. Eosinophils in health and disease: An overview. J Oral Maxillofac Pathol 2003;7(2):31-3.
19. Elvoic AE, Gallagher GT, Kabani S, Galli SJ, Weller PF, Wong DTW. Lack of TGF- and TGF- 1 synthesis by human eosinophils in chronic oral ulcers. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 1996;81(6):672-81.
20. Movassaghi K, Goodman ML, Keith D. Ulcerative eosinophilic granuloma: a report of five new cases. Br J Oral Maxillofac Surg 1996;34(1):115-7.
21. Chatzistamou I, Doussis-Anagnostopoulou I, Georgiou G, Gkilas H, Prodromidis G, Andrikopoulou M, Sklavounou A. Traumatic ulcerative granuloma with stromal eosinophilia: report of a case and literature review. J Oral Maxillofac Surg 2012;70(2):349-53.
22. Boffano P, Gallesio C, Campisi P, Roccia F. Traumatic ulcerative granuloma with stromal eosinophilia of the retromolar region. J Craniofac Surg 2009;20(6):2150-2.
23. Mezei MM, Tron VA, Stewart WD, Rivers JK. Eosinophilic ulcer of the oral mucosa. Am J Acad Dermatol 1995;33(5):734-40.

Effect of Irrigation with Tetraclean on Bacterial Leakage of Obturated Root Canals

Zahed Mohammadi, D.M.D., M.S.D.; Sousan Shalavi, D.M.D.; Luciano Giardino, M.D., D.D.S.; Flavio Palazzi, M.D., D.D.S.

ABSTRACT

The purpose of the study presented here was to evaluate the effect of Tetraclean, Hypoclean, Chlor-XTRA, 2% chlorhexidine and 6% sodium hypochlorite/17% EDTA as a final irrigant on bacterial leakage of the root canal. One hundred and fifty-five extracted human maxillary central incisors were randomly divided into five experimental groups of 25 teeth each and two control groups of 15 teeth each. The root canals in each group were irrigated with 2 ml of the relevant irrigant between each filing. The root canals in group 5 were irrigated with 5 ml of 17% EDTA at the end of root canal preparation. The teeth in each group were obturated with gutta-percha and AH-26 sealer. Positive control teeth were obturated with a single gutta-percha cone without sealer, and negative controls were obturated in the same way as experimental groups.

The coronal portion of each root was placed in contact with inoculum of *Enterococcus faecalis* in Brain Heart Infusion (BHI) culture media. Findings showed that the mean number of days for bacterial penetration in the Tetraclean group was greater than for other experimental groups. On the other hand, the Chlor-XTRA Vista group showed the fewest mean number of days for bacterial leakage.

The essential role of microorganisms in the initiation and perpetuation of pulpal and periradicular diseases has been well documented. Kakehashi et al.,[1] Möller et al.,[2] as well as Sundqvist have shown that pulpal and periradicular pathosis do not develop without the presence of bacterial contamination. Byström and Sundqvist[3] have shown that it is impossible to achieve complete removal or destruction of all bacteria using solely mechanical root canal instrumentation with normal saline as an irrigant.

The importance of removing the smear layer and the patency of dentinal tubules for the disinfecting effect of intracanal medications has been shown.[4] An ideal root canal irrigation solution, in addition to removing the smear layer, should be able to disinfect the root canal system—including its irregularities, such as dentinal tubules, fins, isthmi, and lateral and accessory canals—to penetrate into dentinal tubules and have substantivity. Furthermore, the irrigation solution should not irritate any antigenic reaction, and should be nontoxic and non-carcinogenic.[5]

Sodium hypochlorite (NaOCl) is the most common root canal irrigant. Despite its excellent antimicrobial and tissue dissolving abilities, NaOCl has major drawbacks. It has an unpleasant odor and taste. Because of its high surface tension, its penetration into dentinal tubules is not adequate. Therefore, it does not consistently disinfect the root canal system. Furthermore, it does not remove the smear layer from the dentin walls. In addition, when extruded into the periapical tissues, it is very toxic.[6]

In order to remove the smear layer, the canal should be irrigated with EDTA following irrigation with NaOCl. However, exposing dentin to EDTA for more than one minute makes dentin susceptible to erosion.[7]

Chlorhexidine (CHX) is a cationic biguanide that has a broad spectrum of antimicrobial activity. It has a wide antimi-

crobial spectrum and is effective against both Gram-positive and Gram-negative bacteria, as well as fungi.[8]

MTAD is an antibiotic-based root canal irrigant composed of doxycycline, citric acid and Tween 80 (as detergent).[9] Torabinejad et al.[9] demonstrated the effectiveness of MTAD as a final rinse to remove the smear layer with minimal erosive effects on the root canal walls. Tetraclean *(Ogna Laboratori Farmaceutici, Muggiò, Italy)*, like MTAD, is a mixture of an antibiotic, an acid and two detergents (propylene glycol and cetrimide).[10] However, the concentration of the antibiotic, doxycycline (50 mg mL-1) and the type of detergent (polypropylene glycol and cetrimide) differ from those of MTAD.[10]

Poggio et al.[11] demonstrated that a final rinse with Tetraclean is effective in removing the smear layer, leaving a high number of opened dentin tubules. Ghoddusi et al.[12] showed that it took longer for bacteria to penetrate when either EDTA or MTAD was used for smear layer removal. The effect of root canal irrigation with Tetraclean on the coronal leakage of obturated root canals has not yet been assessed. Therefore, the purpose of this study was to evaluate the effect of Tetraclean on coronal bacterial leakage of root canals obturated with gutta-percha and AH-26 sealer.

Materials and Methods

One hundred fifty-five extracted human maxillary central incisors were used. All teeth had closed apices, straight roots and single root canals. Soft and hard tissue remnants, as well as calculus, were removed using curettes. Teeth were stored in distilled water at room temperature until starting the experiments. To obtain a 14 mm- to 15 mm-long root, the crowns of all specimens were cut with a rotary, water-cooled diamond saw at 1000 rpm *(Isomet Plus precision saw, Buehler, IL)*.

The specimens were randomly divided into five experimental groups of 25 teeth each and two control groups of 15 each. A K-file #15 *(Maillefer SA, Switzerland)* was placed into the root canal until it just protruded through the apex; working length was obtained by subtracting 1 mm from that measurement. The root canal of each root was cleaned and shaped to a size #45 master apical file using step-back technique and K-files and Gates Glidden drills *(Maillefer SA, Switzerland)*.

According to the root canal irrigation regimen, teeth were divided into five experimental groups as follows:

- Group 1: Chlor-XTRA Vista
- Group 2: Hypoclean
- Group 3: Tetraclean
- Group 4: 2% chlorhexidine
- Group 5: 6% sodium hypochlorite/17% EDTA

The root canals in each group were irrigated with 2 ml of the relevant irrigant between each filing. The root canals in Group 5 were irrigated with 5 ml of 17% EDTA *(AriaDent, Iran)* at the end of root canal preparation. This solution was left in the canal for five minutes, then followed by a 5 ml rinse of 5.25% NaOCl. The root canals were dried with paper points and obturated as follows: each canal was obturated with gutta-percha *(AriaDent, Tehran, Iran)* and AH-26 root canal sealer *(Dentsply, DeTrey, GmbH, Konstanz, Germany)* using cold lateral compaction technique.

In the positive and negative control groups (n=15), three root canals were prepared in the same way as Group 1, three as Group 2, three as Group 3, three as Group 4 and three as Group 5. All root canals in the positive control group were obturated with a single gutta-percha cone without root canal sealer. The root canals in the negative control group were obturated in the same way as the experimental groups.

In order to allow the sealer to set, all teeth were kept in 100% humidity at 37°C for 48 hours. The external surfaces of the specimens were covered with two layers of nail varnish except for the apical 2 mm of the root and apical foramen. All teeth in the negative control group were covered completely with two layers of nail varnish, including the apical foramen. The roots were inserted individually into a polypropylene tube with the root apex protruding through the end.

The coronal and middle portions of the specimens were sealed with cyanoacrylate glue to prevent leakage at the connection.

The system was sterilized using ethylene oxide gas and placed in a 5 ml sterile bottle containing 3 ml sterile BHI *(Merck, Darmstadt,*

Germany) to ensure that the apical portion of the root was placed in liquid. The polypropylene tubes were attached to the screw top of the bottle.

Using a sterile syringe and a 21-gauge needle, the upper chamber was filled with BHI containing *Enterococcus faecalis* adjusted to 0.5 McFarland scale. A fresh bacterial suspension of *E. faecalis*, which was prepared daily, was added to the access opening of each tooth until the chamber was nearly full. This procedure was performed every day throughout the experiment. Penetration of the root canal was recorded when turbidity was noted in the broth. Cultures were checked daily until the final test system became positive, at day 90. Cultures from the apical chamber were streaked onto blood agar culture plates and incubated under aerobic and anaerobic conditions. Microorganisms were identified by colony morphology Gram-stain. Data from observations of apical reservoirs were recorded daily. The statistical analysis between the experimental groups was performed using one- and two-way ANOVA and Tukey's Multiple Comparison Test.

Results

The control groups behaved as expected. All positive control teeth showed bacterial leakage within 24 to 48 hours, whereas none of the negative controls showed leakage. All experimental teeth showed bacterial leakage. The Tetraclean group showed the greatest delay in leakage (58.35±11.64 days) (Figure 1).

The differences between chlorhexidine and Tetraclean, chlorhexidine and NaOCl/EDTA, and Hypoclean and Chlor-XTRA Vista were not statistically significant ($p>0.05$). However, the differences between other groups, two by two, were statistically significant ($p<0.05$) (Table 1).

Discussion

The controls behaved as expected, which confirms the method of the study. The reason for using a bacterial leakage model is that this model may be closer to the clinical condition than other methods used to assess leakage, such as the dye penetration method and the fluid filtration method.[13] The reason for using a 90-day observation period was based on a study by Magura et al.[14] that suggested re-treatment of unsealed, obturated root canals that had been exposed to the oral cavity for this period.

The essence of removing the smear layer (SL) is controversial. On one side, studies focused on the removal of the SL for several reasons. The SL contains bacteria, their byproducts and necrotic tissues. Furthermore, the SL may act as a substrate for bacteria and may limit the penetration of root canal irrigants and medicaments into dentinal tubules. By creating a barrier between root filling materials and root canal walls, the SL compromises the formation of a satisfactory seal.[7] Clarke-Holke et al.[15] demonstrated that SL removal and root canal obturation using gutta-percha and AH-26 sealer reduced the leakage of bacteria through the root canal system. Furthermore, Cobankara et al.[16] showed that removal of the SL had a positive effect in reducing apical and coronal leakage for both AH-26 and Roekoseal root canal sealer. Economides et al.[17] and Khayat et al.[18] found that removal of the smear layer resulted in significant reduction of leakage in root

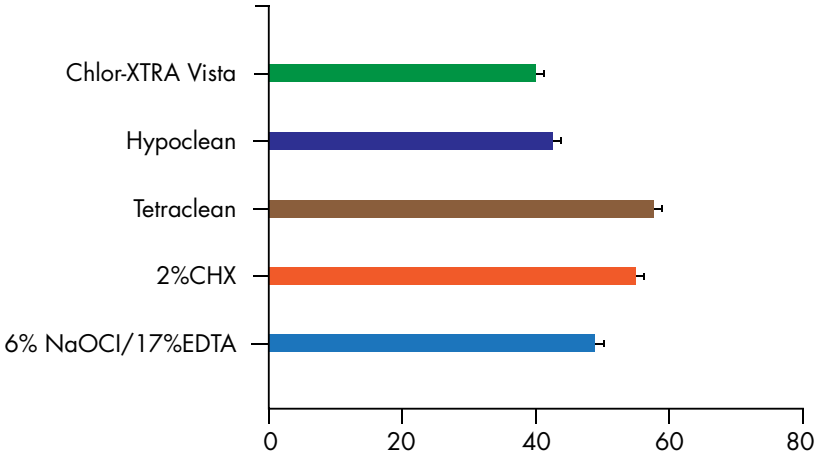


Figure 1. Number of Days for Bacterial Leakage in Different Experimental Groups

TABLE 1
Tukey's Multiple Comparison Test

	Mean Diff.	Significance (P<0.05)
6%NaOCl /17%EDTA vs 2%Chlorhexidine	-4.789	No
6%NaOCl /17%EDTA vs Tetraclean	-7.525	Yes
6%NaOCl /17%EDTA vs Hypoclean	6.783	Yes
6%NaOCl /17%EDTA vs Chlor-XTRA Vista	8.787	Yes
2%Chlorhexidine vs Tetraclean	-2.736	No
2%Chlorhexidine vs Hypoclean	11.57	Yes
2%Chlorhexidine vs Chlor-Xtra Vista	13.58	Yes
Tetraclean vs Hypoclean	14.31	Yes
Tetraclean vs Chlor-Xtra Vista	16.31	Yes
Hypoclean vs Chlor-Xtra Vista	2.004	No

The reduction in leakage in teeth without the smear layer is attributed to the improved mechanical locking of the sealer into patent tubules and better adhesion of sealers to the cleaned canal walls.

canals obturated with a resin-based sealer, but had no effect on leakage of a zinc oxide eugenol-based sealer.

On the other hand, other studies showed that the SL can block the dentinal tubules, preventing the exchange of bacteria and other irritants by altering permeability. Pashley[19] suggested that if the canals were inadequately disinfected, or if bacterial contamination occurred after canal preparation, the presence of a smear layer might stop bacterial invasion of the dentinal tubules. Bacteria remaining after canal preparation are sealed into the tubules by the smear layer and subsequent filling materials. Some studies provide evidence to support the hypothesis that the smear layer inhibits bacterial penetration.[20,21] Using *S. sanguis*, Chailertvanitkul et al.[22] found no statistically significant difference in leakage of the obturated canal when the smear layer was either removed or remained intact. Wimonchit et al.[23] and Saunders and Saunders[24] obtained the same results.

These conflicting results might be because of differences in types of sealers and obturation techniques, the means of producing a smear layer and the diversity of methodologies used to assess leakage under various laboratory conditions.

The findings of the study presented here show that all experimental samples in all groups demonstrated bacterial leakage. However, in the Tetraclean group, bacterial leakage occurred later than in other experimental groups. This can be attributed to the SL removal ability of Tetraclean. Poggio et al.[10] showed that Tetraclean was very effective in removing the smear layer from the root canal surface.

After the Tetraclean group, the 2% CHX group showed the latest leakage time. This good resistance of the CHX group can be attributed to its substantivity. The positively charged molecules of CHX can adsorb onto dentin and prevent microbial colonization on the dentin surface for some time beyond the actual medication period.[8] The substantivity of CHX for up to 12 weeks has been demonstrated.[25] It has been demonstrated

too that, due to its substantivity, CHX as an intracanal medicament/irrigant delays recontamination of the root canal system via the coronal route and does not adversely affect the apical seal of the root canal.[8]

Chlor-XTRA Vista and Hypoclean showed earlier bacterial leakage than other groups. Both of these solutions are NaOCl-based. The earlier bacterial leakage of Chlor-XTRA Vista and Hypoclean can be attributed to two issues. First, because they lack chelating agents, they have no SL removal ability. Second, Chlor-XTRA Vista and Hypoclean have little to no substantivity.[26]

The standard method for removing the SL is to use NaOCl and EDTA. The findings of this study showed there was no significant difference between 6% NaOCl/17% EDTA and 2% CHX. However, their mechanisms of action in delaying bacterial leakage are different. NaOCl/EDTA acts by removing the SL;[7] CHX delays bacterial leakage through its substantivity.[8]

The reduction in leakage in teeth without the smear layer is attributed to the improved mechanical locking of the sealer into patent tubules and better adhesion of sealers to the cleaned canal walls.

In conclusion, within the limitations of the present study, Tetraclean demonstrated the greatest effect on delaying bacterial leakage in root canals obturated with gutta-percha and AH-26 sealer.

Queries about this article can be sent to Dr. Mohammadi at zahed_mohammadi@yahoo.com.

REFERENCES

1. Kakehashi S, Stanley HR, Fitzgerald RJ. The effects of surgical exposures of dental pulps in germfree and conventional laboratory rats. Oral Surg Oral Med Oral Pathol 1965;20:340-9.
2. Moller AJ, Fabricius L, Dahlen G, Ohman AE, Heyden G. Influence on periapical tissues of indigenous oral bacteria and necrotic pulp tissue in monkeys. Scand J Den Res 1981; 89:475-84.
3. Bystrom A, Sundqvist G. Bacteriologic evaluation of efficacy of mechanical root canal instrumentation in endodontic therapy. Scand J Dent Res 1981;89:321-8.
4. Orstavik D, Haapasalo M. Disinfection by endodontic irrigants and dressing of experimentally infected dentinal tubules. Dent Traumatol 1990;6:142-9.
5. Siqueira JF, Machado AG, Silveira RM, Lopes HP, Uzeda M. Evaluation of the effectiveness of sodium hypochlorite used with three irrigation methods in the elimination of *Enterococcus faecalis* from root canal, in vitro. Int Endod J 1997;30:279-82.
6. Mohammadi Z. Sodium hypochlorite in endodontics: an update review. Int Dent J 2008;58: 329-41.
7. Violich DR, Chandler NP. The smear layer in endodontics-a review. Int Endod J 2010; 43:2-15.
8. Mohammadi Z, Abbott PV. The properties and applications of chlorhexidine in endodontics. Int Endod J 2009;42:288-302.
9. Torabinejad M, Khademi AA, Babagoli J, Cho Y, Johnson WB, Bozhilov K. A new solution for removal of the smear layer. J Endod 2003;29:170-5.
10. Mohammadi Z, Abbott PV. On the local applications of antibiotics and antibiotic-based agents in endodontics and dental traumatology. Int Endod J 2009;42:555-67.
11. Poggio C, Colombo M, Scribante A, Sforza D, Bianchi S. In vitro antibacterial activity of different endodontic irrigants. Dent Traumatol 2011 Nov 3 [Epub ahead of print].
12. Goddusi J, Rohani A, Rashed T, Ghaziani P, Akbari M. An evaluation of microbial leakage after using MTAD as a final irrigation. J Endod 2007; 33:173-6.
13. Verissimo DM, do Vale MS. Methodologies for assessment of apical and coronal leakage of endodontic filling materials: a critical review. J Oral Sci 2006;48:93-8.
14. Magura ME, Kafrawy AH, Brown CE, Newton CW. Human saliva coronal microleakage in obturated root canals: an in-vitro study. J Endod 1991;17:324-31.
15. Clarke-Holke D, Drake D, Walton R, Rivera E, Guthmiller JM. Bacterial penetration through canals of endodontically treated teeth in the presence or absence of the smear layer. J Dent 2003;31:275-81.
16. Cobankara FK, Adanir N, Belli S. Evaluation of the influence of smear layer on the apical and coronal sealing ability of two sealers. J Endod 2004;30:406-9.
17. Economides N, Liolios E, Kolokuris I, Beltes P. Long-term evaluation of the influence of smear layer removal on the sealing ability of different sealers. J Endod 1999;25:123-5.
18. Khayat A, Jahanbin A. The influence of smear layer on coronal leakage of Roth 801 and AH-26 root canal sealers. Aust Endod J 2005;31:66-8.
19. Pashley DH. Dentin-predentin complex and its permeability: physiologic overview. J Dent Res 1985:64:613-20.
20. Pashley DH, Michelich V, Kehl T. Dentin permeability: effects of smear layer removal. J Prosthet Dent 1981:46:531-7.
21. Safavi KE, Spangberg LSW, Costa NS, Sapounas G. An in vitro method for longitudinal evaluation of toxicity of endodontic sealers. J Endod 1989;15:484-6.
22. Chailertvanitkul P, Saunders WP, Mackenzie D. The effect of smear layer on microbial coronal leakage of gutta-percha root fillings. Int Endod J 1996; 29:242-8.
23. Wimonchit S, Timpawat S, Vongsavan N. A comparison of techniques for assessment of coronal dye leakage. J Endod 2002;28:1-4.
24. Saunders WP, Saunders EM. Influence of smear layer on the coronal leakage of thermafil and laterally condensed gutta-percha root fillings with a glass ionomer sealer. J Endod 1994;20:155-8.
25. Rosenthal S, Safavi K, Spangberg L. Chlorhexidine substantivity in root canal dentin. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2004; 98:488-92.
26. Mohammadi Z, Mombeinipour A, Giardino L, Shahriari S. Residual antibacterial activity of a new modified sodium hypochlorite-based endodontic irrigation solution. Med Oral Pathol Oral Cir Bucal 2011 Jul 1;16:e588-92.