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Identifying early osteoclastic resorptive lesions in feline teeth: a model for understanding the origin of multiple idiopathic root resorption

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Background and Objective: Domestic cats commonly suffer from external osteoclastic tooth resorption, a disease with many similarities to human multiple idiopathic root resorption. In both diseases, it is unclear whether anatomical features of the tooth surface are associated with a predisposition for resorptive lesions. The aim of the present study was to investigate the origin and progression of early feline osteoclastic resorptive lesions in teeth exhibiting no clinical signs of disease.

Material and Methods: The entire surfaces of 138 teeth from 13 adult cats were analysed using back-scattered electron microscopy. The distribution of lesions was assessed by tooth type, location and between individuals.

Results: Seventy-three (53%) teeth showed at least one resorptive lesion. Eleven (85%) cats had lesions, and there was a significant association between increasing age and incidence of resorptive lesions. The highest frequency occurred in mandibular molars (82%). On average, there were 3.5 lesions per tooth. Fifty-two (38%) teeth featured resorptive lesions at the cemento–enamel junction. Twenty-three per cent of teeth with resorptive lesions showed evidence of repair of lesions that was limited to the root surface. There was no evidence of repair of resorptive lesions at the cemento–enamel junction.

Conclusion: Resorption is prevalent without evidence of clinical disease, and occured at younger ages than previously reported. It can initiate anywhere on the root surface, but lack of repair of lesions at the cemento–enamel junction indicates that mechanisms of replacement are absent or compromised in this region. Whereas resorption of the root may undergo repair, resorption at the cervix may progress to clinically evident lesions.

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Tooth root resorption by osteoclasts is a normal process required for the shedding of deciduous teeth in mammals (1). Resorption of the cementum and dentine of human permanent tooth roots is also normal, frequently occurring apically and at the origin of secondary cementum deposition (i.e. more cementum is replaced than is resorbed) (2). Resorption of permanent teeth may also occur where periodontal ligament tissues are compressed during mechanical (e.g. orthodontic or accidental intrusion) tooth movement (3–6). In the case of resorption induced by orthodontics, the net replacement of lost cementum and dentine by new cementum is generally deficient (i.e. less cementum is replaced than is resorbed). However, in general, pathological resorption of teeth in the absence of another pathology is not common.In humans, multiple idiopathic root resorption by osteoclasts is a rare, but serious, condition associated with external resorption at the cemento–enamel junction or root surface, which progresses to involve root dentine, usually leading to significant loss of tooth structure (7–12). Domestic cats are unusual in that resorption of permanent teeth by osteo(odonto)clasts is a very common problem: epidemiological reports suggest that up to 72.5% of animals suffer from the condition, and the incidence of disease increases with age, yet the mechanism causing the disease remains unknown (13–23). Feline osteoclastic resorptive lesions affect multiple teeth in the same individual and are associated with plaque accumulation, inflammation, ankylosis and a variable reparative response, although loss of teeth is inevitable (24–30). Because of the apparent similarities in the pathogenesis of resorption in both species, and because feline osteoclastic resorptive lesions are more common than multiple idiopathic root resorption, feline osteoclastic resorptive lesions provide a valuable model system for understanding the origin and progression of multiple idiopathic root resorption (31).

Previous studies of feline osteoclastic resorptive lesions have characterized the pathogenesis of advanced lesions (24,28,30,32,33), while analyses of early lesions have been limited (34,35). One question that remains under debate is whether there is any relationship between where a lesion initiates on the tooth and whether it will become clinically significant. Many early studies of feline osteoclastic resorptive lesions described lesions as starting at the cemento–enamel junction (i.e. 'neck' or 'cervical' lesions) (25–27), while more recent analyses suggest that resorptive lesions may initiate anywhere on the tooth surface (34,35).

To understand how the progression of resorption may vary depending on the region of the tooth affected, it is critical to examine teeth at the initiation of disease. Therefore, the aims of this study were to use scanning electron microscopy to identify and describe early lesions in cats with no apparent signs of clinical disease and with radiographically normal dentitions. The objectives were as follows: (i) to identify the sites of early resorptive lesions; (ii) to establish the incidence of resorptive lesions and association with tooth type; (iii) to describe the structure of mineralized tissues of the tooth associated with resorptive lesions; and (iv) to identify evidence of repair of lesions.

Material and methods

Samples

In total, 138 teeth from the dentitions of 13 cats were examined (Table 1). Mandibles and maxillae were obtained from a number of different veterinary sources. All cats were killed for reasons unrelated to this research. Details of age and gender were known for some individuals, although neuter status, general health and dietary history were not known for most cats. Intact left and right upper and lower jaws were removed using standard dissection tools. Jaws were examined for evidence of clinical disease and were radiographed using a digital dental radiography unit (RVG™; Trophy, Marne la Vallee France). Dentitions with evidence of alveolar bone loss, fractured teeth, ankylosis, resorptive lesions, or periapical abscesses were excluded from the analysis. All teeth included in the study were from cats with permanent dentitions, where all teeth showed fully closed root apices. Jaws were fixed in formaldehyde [4% formaldehyde (from paraformaldehyde) in phosphate-buffered saline, pH 7.4] for 1 wk at 4°C, washed in distilled H_2O and stored in 70% ethanol at room temperature.

Preparation of samples for surface analysis

Intact jaws were digested with an alkaline bacterial enzyme detergent (2% Tergazyme™; Alconox Inc., White Plains, NY, USA) in distilled H_2O, at 50°C in a shaker at 50 rpm until no soft tissue could be detected visually (approximately 6 wk). Samples were rinsed for 1–2 h in running tap water, followed by several rinses in distilled H_2O, and then air-dried. Teeth were carefully removed from alveolar bone using bone-cutting forceps, where alveolar bone surrounding the tooth was cut away until the tooth root was fully exposed and could be removed from the bone. Throughout this process, jaws and teeth were examined under a dissecting microscope to assess any signs of pathology or damage during processing. Any damaged teeth

Table 1. Normal teeth surveyed for evidence of resorptive lesions

Cat	Maxillae	Mandible	Age (years)	Number of teeth examined
A	Left	Left	3	13
B	Left	Right	3	11
C	Right	Right	3	14
D	Left	Right	3	12
E	Right	Right	8	8
F	Left	Left	Adult	11
G	Right	Right	Adult	15
H	Left	Left	1	12
I	Right	Right	Adult	15
J	Left	—	Adult	2
K	Right	Right	2	15
L	Left	Left	Adult	3
M	——	Left	Adult	7
Total				138

were excluded from the analysis. To remove all remaining soft tissue and fat, loose teeth were treated with NaCl (< 3% available chlorine) at room temperature for 2 wk. Samples were thoroughly washed in distilled water, air dried, mounted by their crown tips on aluminium rivets using conductive carbon putty (Leit-C Plast™; Agar Scientific, Stansted, UK) and coated with carbon by evaporation.

Surface analysis using back-scattered scanning electron microscopy

Teeth were examined using a JEOL 5410LV™ scanning electron microscope (JEOL, Welwyn Garden City, UK) with an accelerating voltage set at 15 or 20 kv. Images were recorded using digital image-acquisition software (PRINTERFACE™; GW Electronics, Norcross, GA, USA). A calibration standard (Planotech™; Agar, Stansted, UK) with intervals of 10 μm was used to calculate field width.

Microscopic examination and image analysis: survey of resorptive lesions

The buccal or labial (in the case of incisors), lingual/palatal, mesial and distal surfaces of all teeth were examined. The root surfaces of teeth were systematically surveyed for evidence of resorptive lesions from the cemento–enamel junction to the root apex at 350–500× magnification. Teeth were rotated and tilted to allow examination of all surfaces. Images of 1024 × 512 pixel resolution were recorded of all areas of resorptive lesions and the associated tooth surface. The location of resorptive lesions was recorded as occurring on the crown, the cemento–enamel junction, the cervical root (i.e. the cervical third of the root), the mid-root (i.e. the mid-third of the root), or at the apical root (i.e. the apical third of the root) (Fig. 1).

Resorptive lesions were measured from images using NIH IMAGE (National Institutes of Health, Bethesda, MD, USA) using distance calculated from the calibration standard. The maximum diameter of circular resorption lacunae, and the maximum length and width of noncircular areas of resorption, were measured. The depth of resorption lacunae could not be measured.

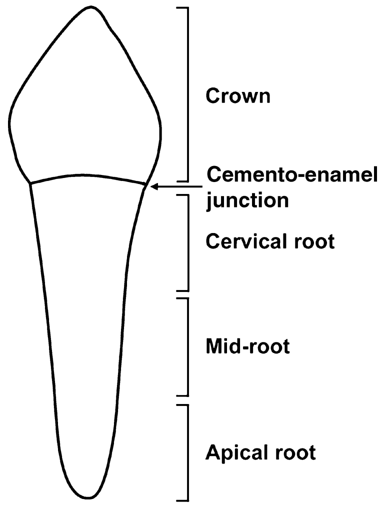

Fig. 1. Locations on the tooth analysed for evidence of resorptive lesions.

Definition of early osteoclastic resorption

Sites of resorptive lesions were defined as being focal or conjoined circular depressions with clear, sharp boundaries, with or without evidence of reparative tissue. These features have been described by other scanning electron microscopy studies of *in vivo* bone and tooth resorption by osteoclasts (2). For the purposes of this study, early osteoclastic activity was defined as the presence of focal lesions representing sites of osteoclast attachment, or larger areas of resorption related to multiple sites of attachment or tracking of resorbing osteoclasts on the tooth surface. For the purposes of analysis, lesions were classified as focal (20–50 μm in diameter, Fig. 2A), medium sized (50–100 μm in maximum dimension, Fig. 2B), or large (> 100 μm in diameter in any dimension, Fig. 2C).

Statistical analysis

Differences in the incidence, distribution and size of lesions between different teeth and different locations on the tooth were assessed using chi-square tests, as were differences in the characteristics of the root surface adjacent to regions of resorptive lesions. The association between age and incidence of resorptive lesions was assessed using Spearman's rank correlation. The Kruskal–Wallis test was used to determine if the average number of lesions was significantly different between tooth types.

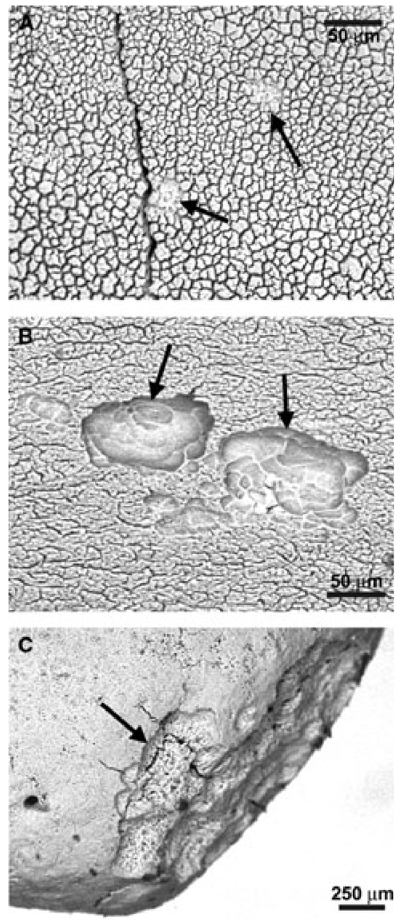

Fig. 2. Examples of differently sized lesions on the root surface. Resorptive lesions were defined by the presence of focal or conjoined circular depressions with clear, sharp boundaries. (A) 'Focal' lesion (20–50 μm in diameter) located at the proximal root of a right upper premolar. (B) 'Medium' lesion (50–100 μm in diameter) located at the mid-root of a left upper incisor. (C) 'Large' lesion (> 100 μm in diameter) located at the apical root of a left upper molar.

Results

Distribution of resorptive lesions among different teeth

In total, 73 (53%) teeth, including 39 (53%) maxillary teeth and 34 (53%)

mandibular teeth, showed evidence of at least one resorptive lesion. Lesions ranged in size from single resorption pits of 10–20 μm in diameter, to complex lesions as large as 2 mm in diameter. The frequency of resorptive lesions varied between tooth types (30–82%), although there was no significant association between tooth type and frequency of resorptive lesions. The lowest frequency of resorptive lesions occurred in mandibular incisors (30%), and the highest in mandibular molars (82%).

Among teeth with resorptive lesions, the average number of areas of resorptive lesions per tooth was calculated for each tooth type. On average, 3.5 lesions per tooth were examined, for both maxillary and mandibular teeth. The maxillary incisors had the lowest number of lesions per tooth (2.0) and the maxillary canine had the highest number of lesions per tooth (6.5), although there was no significant difference in the average number of resorptive lesions between tooth types.

Distribution of lesions among individual cats and association with age

Among the 13 cats examined, 11 (85%) had evidence of at least one tooth with one area of resorptive lesions (Table 2). Three individuals had resorptive lesions on all teeth examined. Two individuals had no evidence of resorptive lesions, although few teeth were examined in these two cats. Six cats with evidence of resorptive lesions were under 4 years of age. Among the seven cats of known age, the frequency of resorptive lesions correlated significantly with increasing age ($R = 0.9$, $p < 0.0001$; Table 2).

Size and distribution of lesions

Focal (20–50 μm), medium (50–100 μm) and large (> 100 μm) lesions were present on all tooth surfaces (Table 3). Resorptive lesions were most frequently focal in the vicinity of the cemento–enamel junction (39, 75%, $p < 0.01$). Lesions were most frequently medium sized at the cervical third of the root (67%), the mid-root (75%) and the apical root (69%). The highest incidence of resorptive lesions was on the buccal/labial surfaces of teeth (53%), followed by the distal (45%) and the mesial (39%) surfaces. The lowest frequency of resorptive lesions was on the lingual/palatal surfaces of the teeth (37%). However, there was no significant relationship between the aspect of the tooth and the incidence of resorptive lesions.

Table 2. Frequency of teeth with resorptive lesions among individual cats

Cat	Age (years)	Number of teeth with resorptive lesions	Total teeth examined	Frequency
E	8	8	8	1.0
F	Adult	11	11	1.0
M	Adult	7	7	1.0
G	Adult	14	15	0.93
D	3	10	12	0.83
B	3	8	11	0.73
C	3	7	14	0.50
A	3	4	13	0.31
K	2	2	15	0.13
H	1	1	12	0.08
I	Adult	1	15	0.07
J	Adult	0	2	0.0
L	Adult	0	3	0.0
Total		73	138	0.53

Frequency = number of teeth with resorptive lesions/number of teeth examined.
The association between age (where known) and frequency of resorptive lesions was significant ($R = 0.9$, $p < 0.0001$).

Fifty-two teeth (38%), including 31 (42%) maxillary teeth and 21 (33%) mandibular teeth, featured resorptive lesions involving the cemento–enamel junction. Of these, 25 (18%) (15 maxillary and 10 mandibular teeth) featured resorptive lesions exclusively involving the cemento–enamel junction, with no evidence of other resorption of the root surface.

There were three distinct patterns of resorption at the cemento–enamel junction (Table 4; Figs. 3A–C): resorption exclusively of the junctional enamel (within 250 μm coronal of the cemento–enamel junction or enamel margin, 37%; Fig. 3A), resorption at the cemento–enamel junction or enamel margin, involving both the junctional enamel and the root surface (50%; Fig. 3B) and resorption of the root surface (within 250 μm apical of the cemento–enamel junction or enamel margin, 13%; Fig. 3C). In cases where multiple types of resorptive lesions of the cemento–enamel junction were observed, the predominant type (i.e. majority of the resorbed area) was recorded. Cases of resorption were significantly associated with both the enamel and root surface of the cemento–enamel junction margin ($p < 0.01$).

Resorption of the root surface occurred at the cervical, mid-root and apical root surfaces. Resorption involving the proximal root occurred in 33 (69%) cases. Twenty-eight (58%) cases featured the mid-root, and 35 (73%) cases featured apical resorptive lesions. In no case was there a significant difference in the number of lesions between different locations on the root.

The structure of the tooth surface adjacent to resorptive lesions

Features of the tooth surface immediately adjacent to resorptive lesions were studied at the cemento–enamel junction and on the root surface. The nonresorbed cemento–enamel junction adjacent to areas of resorptive activity was studied in 43/52 teeth (the nonresorbed cemento–enamel junction in the remainder could not be studied as the resorptive lesions were too extensive).

Table 3. Size of resorptive lesions at the cemento–enamel junction (CEJ), cervical root, mid-root and apical root surfaces

Size of resorptive lesions lacunae	CEJ	Cervical root	Mid-root	Apical root
Focal (20–50 μm)	39 (0.75)*	20 (0.61)	18 (0.64)	11 (0.31)
Medium (50–100 μm)	22 (0.42)	22 (0.67)	21 (0.75)	24 (0.69)
Large (> 100 μm)	17 (0.33)	21 (0.64)	12 (0.43)	19 (0.54)
Total teeth with resorptive lesions	52	33	28	35

*$p < 0.01$.
Values represent the total number of teeth with resorptive lesions and frequency (in brackets). Resorptive lesions at the CEJ were significantly associated with focal lesions ($p < 0.01$).

Table 4. Distribution of types of resorptive lesions at the cemento–enamel junction (CEJ)

Type of resorptive lesions	Number
Junctional enamel only	19 (0.37)
CEJ margin (involving both enamel and root surface)	26 (0.50)*
Root surface only	7 (0.13)
Total teeth with resorptive lesions involving the CEJ	52

*$p < 0.01$.
Values represent the total number of teeth with resorptive lesions and the frequency (in parenthesis).
Resorptive lesions were significantly associated with both the enamel and the root surface of the CEJ margin ($p < 0.01$).

The most frequently observed cemento–enamel junction structure associated with resorptive lesions was bare dentine with exposed tubules. This was identified in 21 (49%) teeth (Fig. 4A). Fibrillar cementum at the cemento–enamel junction was associated with resorptive lesions in 14 (33%) teeth (Fig. 4B). A 'trough' between the enamel edge and fibrillar cementum was present in eight (19%) teeth (Fig. 4C). There was no significant relationship between type of cemento–enamel junction structure and frequency of resorptive lesions.

The root surface adjacent to areas of resorptive lesions was studied in all 33 teeth with resorptive lesions at the cervical root, in 22/28 teeth with resorptive lesions at the mid-root and in 33/35 teeth with resorptive lesions at the apical root (Table 5). The root surfaces of six teeth at the mid-root and of two teeth at the root apex could

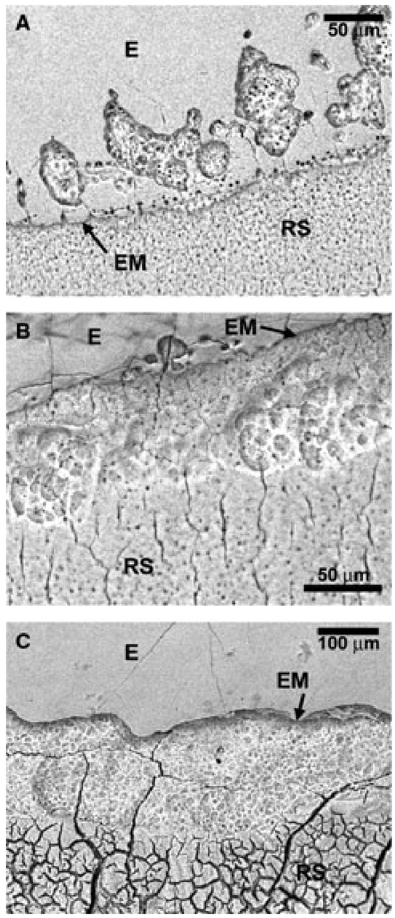

Fig. 3. Patterns of resorptive lesions at the root surface/enamel junction (E, enamel; EM, enamel margin; RS, root surface). (A) Resorptive lesions of the junctional enamel only (within 250 μm coronal of the enamel margin) of a right upper premolar. (B) Resorptive lesions exclusively involving the root surface (within 250 μm apical of the enamel margin) of a left lower incisor. (C) Resorptive lesions at the cemento–enamel junction involving both junctional enamel and the root surface of a left lower incisor.

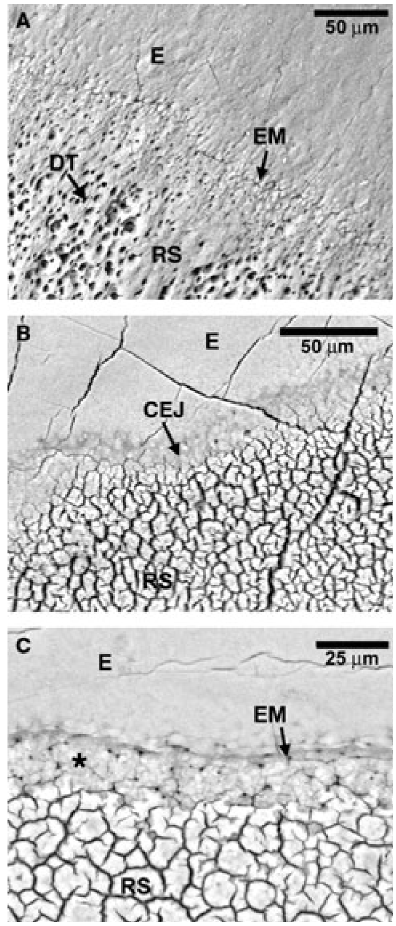

Fig. 4. Structure at the cemento–enamel junction surface associated with resorptive lesions (CEJ, cemento–enamel junction; DT, dentine tubules; E, enamel; EM, enamel margin; RS, root surface). (A) Enamel margin overlapping a porous root surface showing exposed dentine tubules on a right lower incisor. (B) Enamel continuous with extrinsic fibre cementum on the root surface of a left lower premolar. (C) Enamel associated with a porous 'trough' feature (indicated by an asterisk) between the enamel margin and extrinsic fibre cementum on the root of a left upper premolar.

not be assessed because of damage or extensive resorption of these areas. In general, three patterns of root surface structure were observed to be associated with resorption: a surface covered with fibrillar cementum (Fig. 5A), a surface with fibrillar cementum and evidence of resorptive lesions and repair (Fig. 5B) and a surface with exposed dentine tubules (Fig. 5C).

At the cervical root, 20 (61%) teeth had fibrillar cementum with

Table 5. Characteristics of the root surface adjacent to regions of resorptive lesions at the cervical root, mid-root and apical root

Root surface	Cervical root	Mid-root	Apical root
Fibrillar cementum only	5 (0.15)	3 (0.14)	3 (0.09)
Fibrillar cementum with resorptive lesions and repair	20 (0.61)*	19 (0.86)*	30 (0.91)*
Exposed dentine tubules	8 (0.24)	0 (0.0)	0 (0.0)
Total teeth with resorptive lesions	33	22	33

*$p < 0.0001$.
Values represent the total number of teeth with resorptive lesions and the frequency (in parenthesis).
All locations were significantly associated with fibrillar cementum with resorptive lesions and repair.

cementocyte lacunae (cellular cementum) associated with the repair of resorptive lesions. Eight (24%) teeth featured exposed dentine tubules in areas of resorbed fibrillar cementum and five (15%) had fibrillar cementum without associated repair or exposed dentine tubules. At the mid-root, 19 (86%) teeth had fibrillar cementum associated with the repair of resorptive lesions and cementocyte lacunae adjacent to sites of resorptive lesions. Three (14%) teeth featured fibrillar cementum without any association with the repair of resorptive lesions, tubules or afibrillar areas. At the apical root, 30 (91%) teeth had reparative fibrillar cementum associated with resorptive lesions, and three (9%) teeth had fibrillar cementum that was not associated with the repair of resorptive lesions. The cervical root, mid-root and apical root were all significantly associated with fibrillar cementum and the repair of resorptive lesions ($p < 0.0001$).

Repair of resorptive lesions

Seventeen of 73 teeth (23%) with resorptive lesions on the root surface had evidence of repair of resorption lacunae (Table 6). However, signs of repair were never detected in the 52 teeth with resorptive lesions at the cemento–enamel junction ($p < 0.01$). At the cervical root, repair was present in 7/33 (21%) teeth with resorptive lesions, mid-root repair was present in 3/28 (11%) teeth and, at the apical root, repair was present in 7/35 (20%) teeth. On the root surface, most teeth had repair of medium-size or large lesions (14/17 teeth, 82%), whereas three teeth (3/17, 18%) had repair of focal or medium-size lesions. Repair of the root featured intrinsic fibre matrix filling in areas of resorptive lesions or overlapping the edges of lacunae (Fig. 5D). Teeth of all types featured such repair.

Discussion

Domestic cats are the only animals known to suffer routinely from osteoclastic activity in the oral area, leading to a significant excess of resorption and loss of teeth (13–23). Feline osteoclastic resorptive lesions have also been detected in other cat species (36–38). In humans, multiple idiopathic root resorption is rare, but serious (7–12), and understanding the factors associated with feline disease may elucidate mechanisms of the disease progression in humans. To date, the factors associated with the initiation and progression of feline osteoclastic resorptive lesions remain unknown. In particular, it is not known whether there are specific locations on teeth that are predisposed to resorption, and whether this is associated with morphological features of the tooth. Previous studies of early feline osteoclastic resorptive lesions have been used in section histology and radiography in an attempt to describe the origin of lesions (34,35). One limitation of radiography is that it is not sufficiently sensitive to detect very small resorptive lesions. Histological analysis using paraffin sectioning of teeth has been demonstrated to be sensitive enough to detect very small areas of resorption (34). However, this

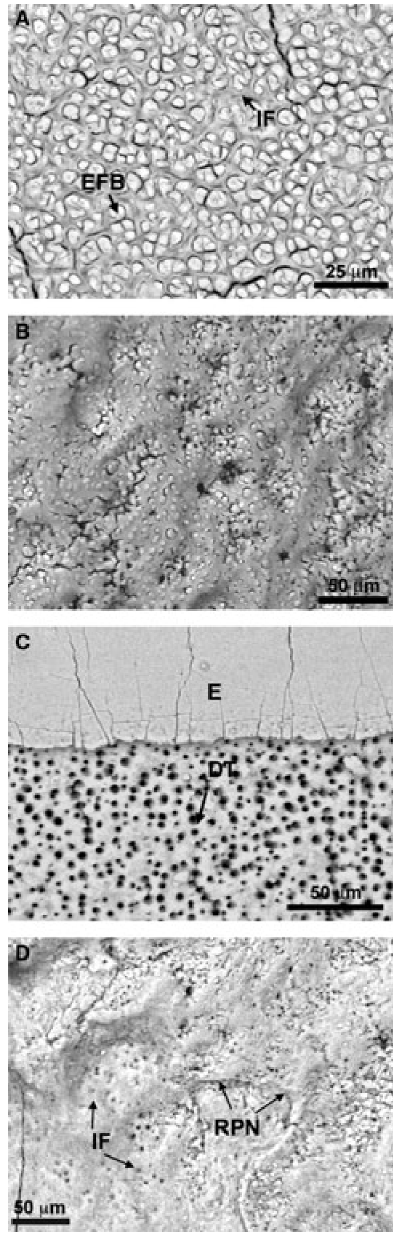

Fig. 5. Features of the nonresorbed root surface in regions commonly subject to resorption (DT, dentine tubule; E, enamel; EFB, extrinsic fibre bundle; IF, intrinsic fibre cementum; RPN, resorption). (A) Fibrillar cementum, including extrinsic and intrinsic fibres on the surface of a left lower molar. (B) Resorptive lesions and repair of the root surface of a right upper premolar associated with fibrillar cementum. (C) Dentine tubules exposed at the cervical root surface on a left upper premolar. (D) Repair of a resorptive lesion on a right lower premolar showing the formation of a woven bone-like or intrinsic fibre matrix filling in areas of resorption.

Table 6. Frequency of repair of resorptive lesions at the cemento–enamel junction (CEJ), and at the cervical root, mid-root and apical root

Location	Number of teeth with resorptive lesions	Frequency of repair
CEJ	52	0*
Cervical root	33	7 (0.21)
Mid-root	28	3 (0.11)
Apical root	35	7 (0.20)
Total	73	17 (0.23)

*$p < 0.01$.
Values represent the total number of teeth with resorptive lesions and the frequency (in parenthesis).
The CEJ is significantly associated with the nonrepair of lesions compared with the cervical root, the mid-root and the apical root.

approach requires the demineralization of teeth, thus losing enamel and destroying the cemento–enamel junction, and is limited to examination of only a small fraction of the entire surface of the tooth.

In our recent analysis of the normal morphology and mineralization of feline teeth we demonstrated that scanning electron microscopy is ideally suited for high-resolution analysis of the entire surface of feline teeth (39). Other analyses of advanced feline osteoclastic resorptive lesions have also demonstrated the power of scanning electron microscopy for describing resorptive lesions (32,33). In our recent study, we showed that there are features of the feline cemento–enamel junction and cervical root that may be associated with an increased rate of destruction by osteoclasts, including significantly reduced levels of mineralization of enamel and dentine, and thinner enamel and cementum (39). Previous studies have shown that the rate of osteoclastic resorption of calcified tissues is inversely proportional to mineral density (40). To investigate further the relationship between these features and early resorptive lesions, the present study was undertaken to analyse the distribution and characteristics of initiating lesions that could never be detectable by radiography.

In this study we showed that approximately half of all clinically normal teeth surveyed showed evidence of resorptive lesions that could not be detected radiographically, indicating that resorptive lesions are prevalent in 'normal' dentitions. The frequency of teeth affected increased in an anterior to posterior (mesial to distal) direction, and the mandibular carnassial (first molar) was found, in this study, to be the most commonly involved tooth. This confirms clinical studies showing that premolars and molars are predominantly affected by feline osteoclastic resorptive lesions (13,14,16,21,27). Among different classes of teeth studied, the average number of resorptive lesions was highest in canines, consistent with an earlier radiographic study of the localization of early feline osteoclastic resorptive lesions, which also showed that most lesions occurred in canines (35). In multiple idiopathic root resorption, some studies show that lesions are associated with anterior (incisor) teeth (7,8,10,41), whereas others show that posterior (premolar and molar) teeth are affected (42,43). In general, the average number of teeth per cat affected with resorptive lesions (3.5) was within the range reported in epidemiological studies of advanced feline osteoclastic resorptive lesions (1.6–3.9 teeth per cat) (14,18,21). The frequency of resorptive lesions among individuals (85%) was higher than that generally reported (20–72.5%) by epidemiological studies of the prevalence of advanced feline osteoclastic resorptive lesions; however, this almost certainly reflects the limitations of clinical techniques for identifying early lesions (13–23,25).

The frequency of teeth affected with resorptive lesions increased significantly with age, which has also been reported in epidemiological studies of feline osteoclastic resorptive lesions (14,16–18,21,23). In our study, root resorptive lesions were present in a cat as young as 1 year of age. Among 3-year-old cats, the frequency of teeth affected with resorptive lesions ranged from 31 to 83%. By contrast, epidemiological studies have previously suggested that resorptive lesions occur infrequently in cats under the age of 4 years (14,17,21). The results of the present study, which used the most sensitive observation technique possible to detect early, and from the clinical point of view, certainly insignificant, lesions shows that resorptive lesions frequently occur in very young cats. However, early resorptive lesions may not be clinically evident until the cat is older and the lesion has progressed to a size that is detectable by visual examination or radiography. A similar situation may be occurring in cases of multiple idiopathic root resorption, with lesions initiating much earlier than can be detected clinically, and it is only when a patient develops symptoms that they seek dental treatment.

The size of small focal lesions observed in this study (10–40 μm), which represents the site of attachment of a single osteoclast, were within the size range of focal lesions described in other scanning electron microscopy studies of feline osteoclastic resorptive lesions (20–40 μm) (32,33,44). Focal lesions were also within the size range reported in studies of deciduous tooth root resorption in the cat (20 μm) (45,46). Resorption lacunae in cat teeth are similar in size to those described in human teeth (1). However, an *in vitro* study has demonstrated that individual feline osteoclasts may be larger than human osteoclasts, as the minimum diameter of resorptive lesions by feline osteoclasts was approximately 40 μm (47).

The buccal surfaces of teeth were most frequently affected, as has been described in epidemiological studies of feline osteoclastic resorptive lesions (16,21,25,27,29). However, no significant relationship was established between the side of the tooth and the frequency of resorptive lesions. In multiple idiopathic root resorption, a side bias to lesions has also not been reported. The reason for the buccal side bias of feline osteoclastic resorptive lesions is not well understood, but it has been suggested that feline osteoclastic resorptive lesions may be a sequel of periodontal disease, and a buccal side bias in periodontal inflammation and plaque accumulation has been described in feline periodontitis (48). Self-cleansing of lingual tooth surfaces by tongue movement is the most likely explanation.

This study also described the distribution of early resorptive lesions on the entire surface of the tooth root. Many radiographic, clinical and histological studies over the years have emphasized the cemento–enamel junction origin of feline osteoclastic resorptive lesions, hence the term 'neck' lesions (24,25,28,37). More recently, it has been shown that resorption can also originate at the root surface (34,35). The pattern of resorptive lesions at the cemento–enamel junction was distinct when it was compared with sites of resorptive lesions elsewhere on the root. At the cemento–enamel junction, resorption was significantly associated with focal lesions. The cemento–enamel junction structure was also significantly associated with resorptive lesions of the enamel margin and was most frequently associated with exposed dentine tubules.

The relationship of the cemento–enamel junction with a predisposition to resorptive lesions is unclear. The exposure of dentine tubules might be taken to suggest loss of cementum or overlying enamel associated with damage or periodontitis. Resorption of tooth surfaces (both enamel and dentine) is prevented by the presence of an intact reduced enamel epithelium on the developing tooth or of cementum. Chinks in reduced enamel epithelium allow access of connective tissue cells to a matrix that can be sought by pre-osteoclasts, just as gaps in the coverage by osteoblasts or bone lining cells is a necessary preliminary to the resorption of bone. Organized breakdown of postmaturational ameloblasts during tooth formation occurs in the Equidae, where enamel is routinely resorbed to permit a firm bond to a reparative bone-like tissue, coronal cementum (49). Future studies should target the postmaturational enamel organ organization in the cervical region of feline teeth. We further note that dentine coverage by any form of cementum may indeed be lacking during development. In addition it should be remembered that dentine tubules (odontoblast processes) normally extend to the enamel-dentine junction, which they penetrate as enamel tubules (50). Less information is available for the root, but it is possible also that during development, dentine tubules penetrate to the cemento-dentine junction and remain 'exposed' unless covered by cementum, which may be deficient in its inception at the cervix.

There is evidence in humans that root resorption may also be associated with damage to, or deficiency of, cementum (1,12,51,52). However, in this study, we observed fibrillar, reparative cementum on the roots of affected teeth, suggesting the presence of a vital periodontium. Clearly, the environment of the cemento–enamel junction differs from the environment around the root surface and therefore factors associated with the initiation of resorption and the control of repair are likely to differ between locations on the root.

The frequent presence of resorptive lesions on enamel was an intriguing and novel finding of this study. These have not been reported in other studies of feline osteoclastic resorptive lesions, and in other species have only been described in studies of resorption of deciduous enamel during primary tooth shedding (53). However, as just noted in horses, enamel is normally partially resorbed prior to the formation of cementum on the crown surface (1,49). In humans, resorption of enamel is observed in unerupted teeth and can be seen on areas that lose the protection of the enamel epithelium as a result of trauma or tumours (1). To date, it is unknown if multiple idiopathic root resorption originates at the enamel surface of the cemento–enamel junction. Further studies examining the physico-chemical properties of feline enamel, and a detailed examination of the development of the cemento–enamel junction, may help to provide answers to the question of the physical role of enamel in feline tooth resorption.

Examination of gingival crevicular fluid for the presence of factors (such as cytokine levels and acidity) known to stimulate osteoclast activity would also provide a better understanding of how the microenvironment of the cemento–enamel junction may support the activation and survival of osteoclasts. We have previously shown that the cytokines interleukin-1 and interleukin-6 are upregulated in feline osteoclastic resorptive lesions and may be involved in stimulating osteoclast activity (54). Recently it has been revealed that feline osteoclasts are significantly activated in hypoxic conditions (55), and local hypoxia in the tooth microenvironment may play a role in the pathogenesis of feline osteoclastic resorptive lesions (56). In our previous morphological study of the normal cat tooth, we describe that enamel is significantly thinner at the cemento–enamel junction, and both enamel and dentine are significantly less mineralized than elsewhere on the tooth (39). While this does not imply that the cemento–enamel junction is predisposed to resorptive lesions, it does suggest that if osteoclasts are activated in this region, enamel and dentine may be at risk of relatively rapid dissolution by these cells.

We observed repair of dentine in approximately half of all teeth affected with feline osteoclastic resorptive lesions, most commonly in early lesions and in younger cats. Evidence of repair of lesions with bone-like tissue has also been reported in several cases of multiple idiopathic root resorption (11,57). The observation that cellular intrinsic fibre cementum is associated with root-resorptive lesions in feline osteoclastic resorptive lesions indicates that repair occurs in response to the stimuli associated with the activation of resorption. In contrast to the situation on the root, resorptive lesions at the cemento–enamel junction were never associated with repair. This implies that the cells involved in repair are not located at the cemento–enamel junction and are not stimulated to initiate a repair response at the cemento–enamel junction, or that they are unable to form reparative matrix owing to the physical nature of the environment. A similar mechanism may underlie the lack of repair of lesions reported in many cases of multiple idiopathic root resorption. Our study thus indicates that resorptive lesions at the cemento–enamel junction are probably progressive, whereas resorptive lesions of the root may undergo repair.

In conclusion, this study has shown that resorptive lesions are prevalent among cats without evidence of clinical disease, and at younger ages than has been previously reported in epidemiological studies. Furthermore, resorptive lesions may initiate anywhere on the surface of cat teeth. However, the likelihood of resorptive lesions progressing to clinically detectable feline osteoclastic resorptive lesions may depend on the location on the tooth where resorptive lesions occur. Although lesions were most frequently detected on the root, this was also most frequently associated with repair. By contrast, resorptive lesions at the cemento–enamel junction were not associated with a reparative response. This indicates that while lesions on the root undergo repair, lesions at the cemento–enamel junction may progress to feline osteoclastic resorptive lesions. The relationship between anatomical features of affected human teeth with the origin and progression of resorptive lesions requires further investigation.

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References

1. Jones SJ, Boyde A. The resorption of dentine and cementum in vivo and in vitro. In: Davidovich Z, ed. *The Biological Mechanisms of Tooth Eruption and Root Resorption*. Birmingham: EBSCO Media, 1988:335–354.
2. Jones SJ. *Morphological and experimental observations on bony tissues using the scanning electron microscope*. London: University of London, 1973. 225pp. PhD Thesis.
3. Kvam E. Scanning electron microscopy of human premolars following experimental tooth movement. *Trans Eur Orthod Soc* 1972;381–391.
4. Rygh P. Orthodontic root resorption studied by electron microscopy. *Angle Orthod* 1977;**47**:1–16.
5. Han G, Huang S, Von den Hoff JW, Zeng X, Kuijpers-Jagtman AM. Root resorption after orthodontic intrusion and extrusion: an intraindividual study. *Angle Orthod* 2005;**75**:912–918.
6. Casa MA, Faltin RM, Faltin K, Arana-Chavez VE. Root resorption on torqued human premolars shown by tartrate-resistant acid phosphatase histochemistry and transmission electron microscopy. *Angle Orthod* 2006;**76**:1015–1021.
7. Kerr DA, Courtney RM, Burkes EJ. Multiple idiopathic root resorption. *Oral Surg Oral Med Oral Pathol* 1970;**29**:552–565.
8. Hopkins R, Adams D. Multiple idiopathic resorption of the teeth. *Br Dent J* 1979;**146**:309–312.
9. George DI Jr, Miller RL. Idiopathic resorption of teeth. A report of three cases. *Am J Orthod* 1986;**89**:13–20.
10. Postlethwaite KR, Hamilton M. Multiple idiopathic external root resorption. *Oral Surg Oral Med Oral Pathol* 1989;**68**:640–643.
11. Beertsen W, Piscaer M, Van Winkelhoff AJ, Everts V. Generalized cervical root resorption associated with periodontal disease. *J Clin Periodontol* 2001;**28**:1067–1073.
12. Coyle M, Toner M, Barry H. Multiple teeth showing invasive cervical resorption – an entity with little known histologic features. *J Oral Pathol Med* 2006;**35**:55–57.
13. Schlup D. Epidemiological and morphological – studies on the cats jaw 1. Epidemiological – studies. *Kleintier Praxis* 1982;**27**:87–94.
14. Coles S. The prevalence of buccal cervical root resorptions in Australian cats. *J Vet Dent* 1990;**7**:14–16.
15. Crossley DA. Survey of feline dental problems encountered in a small animal practice in NW England. *Br Vet Dent Assoc J* 1991;**2**:3–6.
16. van Wessum R, Harvey CE, Hennet P. Feline dental resorptive lesions. Prevalence patterns. *Vet Clin North Am Small Anim Pract* 1992;**22**:1405–1416.
17. Gengler W, Dubielzig R, Ramer J. Physical examination and radiographic analysis to detect dental and mandibular bone resorption in cats: a study of 81 cases from necropsy. *J Vet Dent* 1995;**12**:97–100.
18. Lund EM, Bohacek LK, Dahlke JL, King VL, Kramek BA, Logan EI. Prevalence and risk factors for odontoclastic resorptive lesions in cats. *J Am Vet Med Assoc* 1998;**212**:392–395.
19. Lommer MJ, Verstraete FJ. Prevalence of odontoclastic resorption lesions and periapical radiographic lucencies in cats: 265 cases (1995–1998). *J Am Vet Med Assoc* 2000;**217**:1866–1869.
20. Lommer MJ, Verstraete FJ. Radiographic patterns of periodontitis in cats: 147 cases (1998–1999). *J Am Vet Med Assoc* 2001;**218**:230–234.
21. Ingham KE, Gorrel C, Blackburn J, Farnsworth W. Prevalence of odontoclastic resorptive lesions in a population of clinically healthy cats. *J Small Anim Pract* 2001;**42**:439–443.
22. Reiter AM, Lewis JR, Okuda A. Update on the etiology of tooth resorption in domestic cats. *Vet Clin North Am Small Anim Pract* 2005;**35**:913–942.
23. Pettersson A, Mannerfelt T. Prevalence of dental resorptive lesions in Swedish cats. *J Vet Dent* 2003;**20**:140–142.
24. Hopewell-Smith A. The process of osteolysis and odontolysis, or so-called "absorption" of calcified tissues: a new and original investigation H. The evidences in the cat. *Dental Cosmos* 1930;**72**:1036–1048.
25. Schneck GW, Osborn JW. Neck lesions in the teeth of cats. *Vet Rec* 1976;**99**:100.
26. Schlup D, Stich H. Epidemiological and morphological investigations of teeth in the cat 2. Morphological investigations of the neck lesions. *Kleintier Praxis* 1982;**27**:179–188.
27. Reichart PA, Durr UM, Triadan H, Vickendey G. Periodontal disease in the domestic cat. A histopathologic study. *J Periodontal Res* 1984;**19**:67–75.
28. Okuda A, Harvey CE. Etiopathogenesis of feline dental resorptive lesions. *Vet Clin North Am Small Anim Pract* 1992;**22**:1385–1404.
29. Lyon KF. Subgingival odontoclastic resorptive lesions. Classification, treatment, and results in 58 cats. *Vet Clin North Am Small Anim Pract* 1992;**22**:1417–1432.
30. Ohba S, Kiba H, Kuwabara M, Yoshida H, Koide F, Takeishi M. Contact microradiographic analysis of feline tooth resorptive lesions. *J Vet Med Sci* 1993;**55**:329–332.
31. Burke FJ, Johnston N, Wiggs RB, Hall AF. An alternative hypothesis from veterinary science for the pathogenesis of noncarious cervical lesions. *Quintessence Int* 2000;**31**:475–482.
32. DeLaurier A, Boyde A, Horton MA, Price JS. A scanning electron microscopy study of idiopathic external tooth resorption in the cat. *J Periodontol* 2005;**76**:1106–1112.
33. Gauthier O, Boudigues S, Pilet P, Aguado E, Heymann D, Daculsi G. Scanning electron microscopic description of cellular activity and mineral changes in feline odontoclastic resorptive lesions. *J Vet Dent* 2001;**18**:171–176.
34. Gorrel C, Larsson A. Feline odontoclastic resorptive lesions: unveiling the early lesion. *J Small Anim Pract* 2002;**43**:482–488.
35. Harvey CE, Orsini P, McLahan C, Schuster C. Mapping of the radiographic

central point of feline dental resorptive lesions. *J Vet Dent* 2004;**21:**15–21.

36. Berger M, Schawalder P, Stich H, Lussi A. Neck lesions in big cats – examinations in the leopard (Panthera Pardus). *Kleintier Praxis* 1995;**40:**537–549.
37. Berger M, Schawalder P, Stich H, Lussi A. Feline dental resorptive lesions in captive and wild leopards and lions. *J Vet Dent* 1996;**13:**13–21.
38. Mendoza KA, Marretta SM, Klippert LS. Odontoclastic resorptive lesion of a mandibular right first molar in a cougar. *J Vet Dent* 2000;**17:**173–176.
39. DeLaurier A, Boyde A, Horton MA, Price JS. Analysis of the surface characteristics and mineralization status of feline teeth using scanning electron microscopy. *J Anat* 2006;**209:**655–669.
40. Jones SJ, Arora M, Boyde A. The rate of osteoclastic destruction of calcified tissues is inversely proportional to mineral density. *Calcif Tissue Int* 1995;**56:**554–558.
41. Liang H, Burkes EJ, Fredriksen NL. Multiple idiopathic cervical root resorption: systematic review and report of four cases. *Dentomaxillofac Radiol* 2003;**32:** 150–155.
42. Lydiatt DD, Hollins RR, Peterson G. Multiple idiopathic root resorption: diagnostic considerations. *Oral Surg Oral Med Oral Pathol* 1989;**67:**208–210.
43. Cholia S, Wilson P, Makdissi J. Multiple idiopathic external apical root resorption: a report of four cases. *Dentomaxillofac Radiol* 2005;**34:**240–246.
44. Ohba S, Kuwabara M, Kamata H, Yukawa M, Kiba H. Scanning electron microscopy of root resorption of feline teeth. *J Vet Med Sci* 2004;**66:**1579–1581.
45. Ten CateAR, Anderson RD. An ultrastructural study of tooth resorption in the kitten. *J Dent Res* 1986;**65:**1087–1093.
46. Sasaki T, Shimizu T, Watanabe C, Hiyoshi Y. Cellular roles in physiological root resorption of deciduous teeth in the cat. *J Dent Res* 1990;**69:**67–74.
47. Muzylak M, Flanagan AM, Ingham K, Gunn N, Price J, Horton MA. A feline assay using osteoclasts generated in vitro from peripheral blood for screening antiresorptive agents. *Res Vet Sci* 2002;**73:**283–290.
48. Mallonee DH, Harvey CE, Venner M, Hammond BF. Bacteriology of periodontal disease in the cat. *Arch Oral Biol* 1988;**33:**677–683.
49. Jones SJ, Boyde A. Coronal cementogenesis in the horse. *Arch Oral Biol* 1974;**19:** 605–614.
50. Boyde A. Enamel. In: Oksche A, Vollrath L, eds. *Handbook of Microscopic Anatomy*. Berlin: Springer-Verlag, 1989:309–473.
51. Malek S, Darendeliler MA, Swain MV. Physical properties of root cementum: part I. A new method for 3-dimensional evaluation. *Am J Orthod Dentofacial Orthop* 2001;**120:**198–208.
52. Bilgin E, Gurgan CA, Arpak MN, Bostanci HS, Guven K. Morphological changes in diseased cementum layers: a scanning electron microscopy study. *Calcif Tissue Int* 2004;**74:**476–485.
53. Boyde A, Lester KS. Electron microscopy of resorbing surfaces of dental hard tissues. *Z Zellforsch Mikrosk Anat* 1967;**83:** 538–548.
54. DeLaurier A, Allen S, deFlandre C, Horton MA, Price JS. Cytokine expression in feline osteoclastic resorptive lesions. *J Comp Pathol* 2002;**127:**169–177.
55. Muzylak M, Price JS, Horton MA. Hypoxia induces giant osteoclast formation and extensive bone resorption in the cat. *Calcif Tissue Int* 2006;**79:**301–309.
56. Muzylak M, Arnett TR, Price JS, Horton MA. The in vitro effect of pH on osteoclasts and bone resorption in the cat: implications for the pathogenesis of FORL. *J Cell Physiol* 2007;**213:**144–150.
57. Moody GH, Muir KF. Multiple idiopathic root resorption. A case report and discussion of pathogenesis. *J Clin Periodontol* 1991;**18:**577–580.