Ameloblastoma with Prominent Ossification in the Mandible of a Dog
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ABSTRACT. A rare case of ameloblastoma with prominent stromal ossification in an 8-year-old female dog was studied. A bony mass recurred rapidly in the right mandible at the first molar region. Histopathologic examination revealed the lesion to be an atypical variant of ameloblastoma. Epithelial cells showed marked cell atypia, and mitotic figures were rather common. The collagenous stroma was abundant, with prominent formation of bone trabecular rimmed by active osteoblasts. The tumor was highly proliferative and aggressive, and thought to be malignant in nature.—KEY WORDS: ameloblastoma, canine, stromal ossification.


Only a few odontogenic tumors have been reported in domestic animals. Ameloblastoma is rarely observed in dogs and cats, and most are microscopically benign and lack cytologic atypia [2, 7, 11]. Microscopic evidence of malignancy is occasionally encountered in human ameloblastoma, and was called odontogenic carcinoma or malignant ameloblastoma, which was thought to develop by malignant transformation of an ameloblastoma [4, 8]. In human ameloblastomas, prominent stromal ossification has been rarely observed, except for periostial bone formation on the surface of the affected bone [6, 9].

The present paper reports a case of ameloblastoma characterized by microscopic evidence of malignancy and extensive stromal bone formation. To our knowledge, this type of ameloblastoma has not been documented previously in dogs.

An 8-year-old, female mixed breed dog was referred to a veterinary clinic in April, 1994, because of hard swelling of the right mandible of two months' duration. Physical examination revealed diffuse swelling with an externally projecting mass (2 × 2 × 2 cm) in the right mandible including the first molar tooth. The mass was tender on palpation and was well demarcated from the adjacent soft tissue. In May 1994, an additional mass (2 × 2 × 2 cm) was found on the lingual side of the mandibular swelling. Both projecting masses were resected surgically, and anticancer therapy was applied using Vincristine. Despite the anticancer therapy, the tumor rapidly recurred within 20 days. A small piece of biopsy sample was obtained from the mass, and diagnosed as osteosarcoma with prominent ossification. The mass continued to grow more rapidly, and a right hemimandibulectomy was performed under general anesthesia in July 1994. Results of laboratory test, including serum calcium and alkaline phosphatase levels, were within normal limits. Radiographic examination showed a radiopaque mass, closely associated with the molar, suggestive of a fibro-osseous lesion of the jaw with scalloping of the margins. The regional lymph nodes were not palpable. She had no clinical history of any diseases of the head and neck region. The postoperative history was uneventful, and no signs of recurrence have been noted 11 months later.

The excised mandible was fixed in 10% buffered formalin. After fixation, tissue blocks from the mass were decalcified, hydrated and embedded in paraffin in an usual manner. Sections at 5 μm in thickness were stained with hematoxylin-eosin (HE). Selected sections from the tumor were also stained with periodic acid-Schiff (PAS), alcian blue, Masson trichrome and Congo red. For immunohistochemical studies, the labelled strept-avidin-biotin (LSAB) method was applied on deparaffinized sections using a commercial kit (DAKO Corp., Santa Barbara, CA). The primary antibodies used were: anti-keratin (polyclonal, DAKO Corp.), anti-cytokeratin AE1 and AE3 (monoclonal, Signet Labs. Inc., Dedham, MS), anti-epithelial membrane antigen (EMA) (monoclonal, DAKO Corp.), anti-vimentin (monoclonal, DAKO Corp.) and anti-proliferating cell nuclear antigen (PCNA) (monoclonal, DAKO Corp.). Negative and substituted serum controls and positive tissue controls were also employed. The specimens were counterstained with Mayer's hematoxylin.

Grossly, a large mass (4.2 × 5.0 × 3.0 cm) with external mass (3 × 2 × 2 cm) involving the first molar was present in the submental region of the right mandible, and the buccal cortical bone was expanded by the tumor growth, resulting in an asymmetrical jaw (Fig. 1). A transverse cut of the mass revealed extensive destruction of the mandible and replacement by fleshy, firm and faintly lobulated tumor growth surrounding the first molar.

Microscopically, infiltrative tumor growth was observed around the first molar region. The tumor was composed of proliferating epithelial cells, forming ramifying and irregular cell nests with prominent stromal collagenization (Fig. 2-1). The tumor cells appeared to be prefuctioning ameloblasts or basal cells with peripheral palisading (Fig. 2-2). Nuclei of epithelial cells were oval in shape and showed marked hyperchromasia and pleomorphism with frequent mitotic figures (Fig. 2-2). The centers of epithelial cell nests tended to be loosely arranged and various amounts of fibrilar material was deposited, in which granular cells were observed in close association with the matrix (Fig. 2-2). By Congo red staining, the substance showed green birefringence under polarized light. The stroma was abundant and was rich in collagen. Some areas were highly
cellular, consisting of pleomorphic spindle cells arranged in a haphazard fashion. Trabecular bone with a rim of osteoblasts was frequently formed, and new bone trabeculae fused to each other, forming large bone tissue (Figs. 3–1 & 2). Neither inflammatory reaction nor formation of granulation tissue was seen in any part of the tumor. PAS, alcian blue and Masson trichrome stain revealed no differentiating cytoplasmic features.

Immunohistochemically, a positive reaction for keratin, cytokeratin AE1, cytokeratin AE3 and EMA was consistently evident in the epithelial tumor cells. Vimentin was positive for the stromal area. Immunostaining for proliferating cell nuclear antigen (PCNA) frequently labelled nuclei (45% of labeling index) in the odontogenic epithelial cell nests.

Based on the clinical, gross and light microscopic findings, the present case was diagnosed as ameloblastoma with malignancy. The diagnosis was based on the following characteristics: repeated and extremely rapid recurrence and invasion into surrounding tissues suggesting malignancy; irregular cell nests with odontogenic epithelium. Although some canine ameloblastomas are considered to be locally aggressive and recur easily, there have been few reports of highly aggressive and rapidly recurring ones like the present case. This case was thought to be a malignant variant of odontogenic epithelial neoplasm which was classified as ameloblastoma because the epithelial component was very cellular with nuclei varying in size and shape and with prominent mitotic activity. Microscopic evidence of malignancy is occasionally encountered in human primary epithelial neoplasm in the jaw [8], but there have been almost no reports in domestic animals. Malignant ameloblastoma may develop due to malignant transformation of an ameloblastoma, which is thought to arise from rests of Malassez, reduced enamel epithelium that envelopes impacted teeth or gingival epithelium [4, 8].

The other characteristic feature in the present case was prominent stromal ossification. The inductive stroma was rather abundant, with prominent formation of bone

![Fig. 1. Gross appearance of the tumor. The jaw bone is swollen due to the expansive growth of the tumor mass (arrow).](image)

![Fig. 2-1. Mandibular ameloblastoma of a dog. Irregular patterns of odontogenic epithelium with prominent stromal collagenization. Epithelial cells show marked cell atypia. H & E x 375.](image)

![Fig. 2-2. The epithelial cells are palisading in the periphery of cell nests (arrows). The center of cell nests are loosely arranged and fibrillar material (arrow heads) is seen. H & E x 375.](image)
trabeculae rimmed by osteoblasts. Differentiation of mesenchymal cells into active osteoblasts with formation of new trabecular bone was frequently found in the dense collagenous tissue. Stromal osteoid and bone formation may be an epithelial secondary inductive effect [6, 9].

The differential diagnosis includes acanthomatous epulis, calcifying epithelial odontogenic tumor and keratinizing ameloblastoma. The acanthomatous epulis has cords and solid sheets of epithelial cells, and always infiltrate locally into bone [1, 10]. The cells of acanthomatous epulis are characterized by the prominent intercellular bridges and lack of cellular or nuclear pleomorphism and anaplasia which is encountered in the present ameloblastoma [1]. Calcifying epithelial odontogenic tumors (CEOT) are rare tumors, which are characterized by polyhedral epithelial cells, often of dental type, and also by amyloid deposition in the epithelium and stroma. Calcification of amyloid globules, sometimes in large amounts, are essential lesions in CEOT and are usually in the form of Liesegang rings which are concentrically laminated rings [7, 11]. The present case lacks calcification, especially Liesegang rings form of calcification, in the amyloid deposition, and shows prominent stromal ossification instead of it. Keratinizing ameloblastoma is one variant of ameloblastoma, and is composed of epithelial cell islands with central keratinization and calcification. Amyloid is present between neoplastic epithelial cells [3].

The present case showed positive reaction for cytokeratins AE1 and AE3 in tumor cells. The expression of such cytokeratins has been demonstrated in different types of human ameloblastoma, and the expression pattern of the intermediate contents differ from those found in the basal cells of the oral mucosa [5]. These authors concluded that ameloblastomas are of odontogenic origin and are not direct derivatives of basal cells of the oral epithelium [5]. In addition, the present case, containing numerous PCNA-positive cells, appeared to correlate well with rapid clinical growth and histological malignancy.

REFERENCES