Cytology of oral masses

Leslie Sharkey DVM, PhD, DACVP

Introduction
Oral cytology, as for cytology of other anatomic regions, has the potential to be a quick, relatively non-invasive diagnostic method for screening lesions. As the value of routine dental care for dogs and cats is increasingly appreciated, that potential can be further developed with additional research on optimal use of cytology in the oral cavity. Understanding the strengths and weaknesses specific to cytological evaluation of oral pathology needs to be determined. Currently, there is very little information beyond case reports and relatively small case series focusing on individual tumors or disease entities, demonstrating a need for additional research in this important area of small animal practice.

Sample collection
There are several challenges to collecting adequate samples from oral lesions. One is of course, the location of the lesion and the animal’s tolerance for manipulation of and in the oral cavity. It may be necessary and preferable in some cases to obtain samples under sedation or general anesthesia for better visualization of the lesion, easier collection of samples, and for the safety and comfort of the patient and clinic staff.

Once the lesion has been visualized, collection of adequately cellular and representative samples is the next priority. Touch imprints can be cellular in some cases, however most of the time only superficial processes will be represented, so other components of the lesion may be missed. For example, some proliferative lesions may have ulcerated surfaces, in which case the superficial inflammation and ulceration will be observed in the cytology, however the proliferative component will not be represented. For this reason, it may be preferable to also collect cytology samples by aspiration or the “woodpecker” technique in which a needle without syringe is inserted into the lesion with a quick chopping motion in several directions. A syringe with air in it may then be attached for expulsion of the needle contents onto a glass slide. Several smears should be prepared if possible, in order to increase the chances of obtaining a maximally diagnostic sample in which cells are spread out enough for good staining and visualization but not disrupted by the smearing process. If the lesion is to be surgically removed for diagnostic or therapeutic purposes, it is possible to make impression smears of the cut surface of the lesion. Being careful to not disturb the tissue that is required for histopathologic evaluation, a section of the removed tissue can be blotted on a paper towel to remove excess blood or fluid, and then touched to the surface of a glass slide. For lesions that are poorly exfoliative, for example those that contain a lot of connective tissue cells, it may be necessary to gently scrape the surface with a scalpel blade to obtain cells for cytology.

In our experience, cytology smears prepared from swabs are generally of very poor quality, usually having few cells that are often broken.

Inflammatory conditions of the oral cavity
Normal cytology of the oral cavity consists primarily of superficial squamous epithelial cells and large numbers of commensal oral bacteria of mixed morphology. Simonsiella is a specific oral
bacteria in which rod shaped bacteria arrange in stacks that resemble “caterpillers.” Small amounts of blood or neutrophils may be normal. Inflammatory disease of the oral cavity is common in small animals, with a variety of lesions that may be chronic or recurrent as well as generalized or localized. Grossly, lesions are usually classified as ulcerative, vesiculobullous, or proliferative. One excellent recent review classifies inflammatory diseases etiologically, focusing on those caused by dental disease, infectious conditions (often feline viral), idiopathic inflammatory disease, mucosal and cutaneous immune-mediated, reactive, and neoplasia associated (Lommer 2013). Calcinoses circumscripta is an inflammatory lesion of uncertain etiology possibly related to previous trauma that is characterized by deposition of ectopic dystrophic mineral in soft tissues, sometimes near the tongue. Cytology samples consist of abundant refractile mineralized material with variable numbers of macrophages and fibroblasts. If treatment is necessary, surgical excision is typically curative.

Despite the frequency and importance of oral inflammatory lesions in dogs and cats, our approach cytologically has been very generic and descriptive up to this point. The severity and cellular composition of the inflammatory response is described, as well as the presence or absence of infectious agents and their morphology. It is very important to distinguish the presence of commensal organisms from those that might be opportunistically present in compromised tissue. Typically this is done by assessing the accompanying inflammation, evaluating for intracellular organisms that are more likely to be pathogenic, and looking for a population of organisms that appears to predominate, which may suggest pathogenicity as well. In most cases, correlation of cytologic findings to a distinct clinical entity is limited due to a minimal research data base supporting interpretation.

**Neoplastic and hyperplastic conditions of the oral cavity**

A large range of tumors can occur in the oral cavity, with many having similar cytologic appearance to their extra-oral counterparts. However, biological behavior may be different (e.g. osteosarcoma, melanoma). The following section will describe the appearance of some common tumors, including those lesions unique to the oral cavity. It should be noted that oral pathology is a distinct subspecialty in human pathology that is currently not as well developed in veterinary medicine. The classification of oral neoplasia beyond the “classic” common lesions is slowly evolving as the literature expands.

*Squamous cell carcinoma (SCC)* is the most commonly diagnosed oral malignancy in cats. It is often aggressive, ulcerated, and associated with tooth loss and secondary bacterial infections that may be initially confused as the primary process. Bone-invasive variants can be associated with osteolysis, periosteal new bone formation, and osseous metaplasia of stroma (Martin), which can also present diagnostic challenges when interpreting radiographs or cytology samples. SCC is the second most common non-tonsillar oral tumor in dogs, with well differentiated tumors being rather common and sometimes difficult to distinguish from reactive epithelial hyperplasia, particularly in the presence of inflammation or infection. Other variants include basaloid, adenosquamous, and spindle-cell (Nemec), which may impact the cytologic appearance. The classic appearance includes some amount of suppurative or even septic suppurrative inflammation mixed with a robust population of squamous cells in various stages of differentiation. A key feature is “nuclear:cytoplasmic dysynchrony” in which large immature nuclei are observed in cells with abundant and partially to fully keratinized cytoplasm.
Melanoma is a common tumor in the oral cavity of dogs. While widely believed to have a universally negative clinical outcome, new evidence suggests that some well differentiated tumors, melanomas of the lip, and even a small subset of histologically malignant tumors may have a more indolent course (Bergin). Melanomas can have highly divergent cytologic appearances, from heavily pigmented well differentiated cells to poorly differentiated cells with features of either round cell, epithelial, or mesenchymal lineages, or even multiple lineages within a single tumor. Criteria of malignancy are often striking, with characteristically large and prominent nucleoli. While amelanotic tumors may have very sparse pigment, thorough examination of good quality samples will reveal at least a small amount of pigment in most cases. Because of the potentially nondescript appearance of malignant amelanotic melanomas, immunohistochemical staining of biopsy samples using a panel of markers may be required for definitive diagnosis, but even then, some spindloid variants can be difficult to confirm (Smedley).

Fibrosarcomas, osteosarcomas, and chondrosarcomas also occur in the oral cavity, with similar cytologic appearance as in other anatomic locations. As noted above, their diagnosis can be complicated by mesenchymal components to other neoplastic lesions or due to “mesenchymal” variants of other tumors such as melanomas. Granular cell tumors are tumors, frequently benign, of uncertain etiology that can occur on the tongues of dogs, but also in the thoracic cavity and central nervous system (Rallis). Cytology samples are characterized by numerous individualized cells with a low nuclear to cytoplasmic ratio and abundant pink granular cytoplasm and cytoplasmic vacuolization.

More specific oral growths include the epulides, which are defined as localized exophytic gingival growths that may be either reactive or neoplastic. While long known in veterinary medicine, the etiology and origin of epulides are not well characterized and various classification schemes have been used. This document will use the most recent scheme to present the various subtypes and their basic histologic features (Fiani). The cytology of these lesions has not been investigated but can be extrapolated from histologic characteristics. Given the histologic complexity of the lesions, there may be limitations to the use of cytology for diagnosis in some cases, however cytology offers the potential of a rapid and noninvasive screening tool once the advantages and disadvantages are elucidated. Although these odontogenic tumors are benign, they can cause local destruction and/or expansion of soft and hard tissue.

Canine acanthomatous ameloblastoma (CAA, formerly acanthomatous epulis) is an odontogenic tumor consisting of cords and islands of basal and squamous epithelial cells that invade a connective tissue stroma. These tumors are locally aggressive.

Peripheral odontogenic fibroma (POF, formerly fibromatous and/or ossifying epulis) is an odontogenic tumor consisting of a cellular mass of fibroblastic connective tissue and matrix divided from mucosal epithelium by normal fibrous connective tissue. Foci of odontogenic epithelium, bone, and collagenous matrix may be embedded in the lesions.

Focal fibrous hyperplasia (FFH) consists of reactive dense fibrous connective tissue that may contain dystrophic calcification but odontogenic epithelium is not present. This
lesion may develop in response to irritation secondary to plaque and calculus and is classified as a reactive lesion rather than an odontogenic tumor.

*Peripheral giant cell granulomas (formerly giant cell epulis)* consists of variable numbers of multinucleated giant cells mixed with spindle cells in a collagenous matrix that may also contain osteoid. Canine peripheral giant cell granulomas appear to have indolent biological behavior and rarely recur after excision, in contrast to cats, in which they are more common as well as more aggressive and likely to recur (De Brunn).

*Odontogenic tumors not discussed above* incorporate a complex group of lesions that are currently poorly characterized in veterinary pathology. One of the better known variants is the *amyloid-producing odontogenic tumor (APOT)*, which is associated with amyloid derived from enamel proteins secreted by ameloblasts in the tumor (Hirayama). These tumors also occur in cats (Delaney).

**New data and planned studies**

Given the increasing role of dental care in small animal practice, as well as the fact that oral tumors make up 5-7% of all malignancies in dogs and cats, a collaboration between the dental and pathology services at the University of Minnesota has formed to develop the use of cytology for the screening of oral lesions presenting through the dentistry service. This collaboration has recently been funded by the Foundation for Veterinary Dentistry. Preliminary data has shown good correlation between cytology and histopathology for both benign and neoplastic lesions, including peripheral odontogenic fibroma, and canine acanthomatous ameloblastoma. Updated results from this study will be presented.

**References**


