Proceedings of the 34th World Small Animal Veterinary Congress
WSAVA 2009
São Paulo, Brazil - 2009

Next WSAVA Congress:

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**CYTOLOGY OF SUBCUTANEOUS SWELLINGS, SKIN TUMOURS, AND SKIN LESIONS. PART I**

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Subcutaneous swellings, skin tumours, and skin lesions are extremely well suited for cytological examination via FNAB. Aspiration can be performed without difficulty, causes little or no pain except with processes on the feet or the nose, and even in case of the latter localizations, seldom requires anesthesia. An impression smear or scraping can easily be made of open lesions, but it is advisable to perform FNAB from the edges of the wound as well. Cytological examination can in many cases lead quickly to the correct diagnosis and be of decisive importance to the choice of therapy and to the prognosis.

The cytological examination of FNAB of skin tumours and subcutaneous swellings often makes histological examination unnecessary. Histological examination costs more time, is more invasive, and more expensive. However, it should be emphasized that follow-up histological examination almost always offers a solution in the event that cytological examination is insufficient, and some processes can only be determined by histological examination.

**Subcutaneous swellings**

Subcutaneous swellings can arise from structures of the subcutis as well as from other tissues located under the skin, such as salivary glands, lymph nodes, thyroid, mammary glands, cartilage, bone, etc. Mammary tumours and inflammations of the mammary glands will not be dealt with in this chapter. Cytology of the mammary gland of the cat and specially the dog is one of the most difficult areas of clinical cytology in companion animals.

**Hematoma**

Preparations that contain many erythrocytes are searched carefully for the presence of other cells. If only a few epithelial cells and/or mesenchymal cells are found and by examination under the 100x objective these are seen to have no malignancy criteria, then i) a blood vessel may have been penetrated, or ii) there may be a tumour of cells of vessel walls (e.g. hemangioma/hemangiosarcoma), or iii) the blood is from a hematoma. The latter can usually be confirmed by the fact that blood from a slightly older hematoma contains no thrombocytes but does contain brown bilirubin crystals and macrophages that show erythrophagocytosis and accumulation of iron pigment. A few inflammatory cells, like neutrophils and lymphocytes, may also be present.

**Fat/lipoma**

Preparations that before fixing/staining contain a good amount of glassy, fatty material but after staining contain only a vague background with some empty round spots of different sizes probably contained only fat that has been dissolved by the alcohol. Sometimes there are few free pyknotic nuclei of the fat cells and/or intact three-dimensional clusters of empty fat cells. The presence of pure fat indicates an unsuccessful aspiration (subcutaneous fat) or an aspiration from a lipoma. The cells of a lipoma look the same as normal fat cells.

**Cysts**

Smears that contain large, blue, epithelial cells without nuclei are usually contaminated with flakes of keratin from the skin. If the smear contains almost exclusively large numbers of keratin flakes or amorphous blue or black material, then probably a dermoid (epidermoid) cyst has been aspirated. Such cysts also often contain cholesterol crystals. These can cause secondary inflammation. If in addition to keratin flakes there are also immature nucleated epithelial cells, these should be examined carefully for malignancy criteria.

Preparations can also contain a homogenous pink layer of protein in which there are sporadic epithelial cells and/or mesenchymal cells or intermixed blood cells. This can indicate aspiration from 1) edematous tissue, 2) a very poorly exfoliating tumour, or 3) a serous cyst (seroma, hygroma). Aspiration from a cyst is usually recognized during aspiration. By microscopic...
examination one also usually finds a few mononuclear cells (macrophages, cyst wall cells) and cholesterol crystals.

Salivary cysts are equally easy to recognize. The characteristic findings, apart from the location of the cyst and the mucoid consistency of the contents, are the presence of clumps and strings of amorphous blue material (mucus) and many foamy macrophages, which develop very strong phagocytic activity and can contain erythrocytes, bilirubin crystals, and hemosiderin, because there is always some blood in salivary cysts. Because of the high viscosity of the saliva the erythrocytes are often stretched out in rows. Preparations from salivary cysts also often contain a few groups of cells from the salivary gland. These are easy to recognize. The clumps of cells, which have a clearly acinar structure and a great amount of cytoplasm, are often enclosed in dark blue strings of mucus.

Smears from abscesses have large numbers of neutrophils. A single lymphocyte, plasma cell, or macrophage may also be found. Dependent on the cause of the abscess the neutrophils may show many degenerative signs. One should always carefully look for intracellular bacteria.

Inflammation

General inflammatory processes have been described in the introduction chapter. Some special types of inflammatory processes can be recognized on cytological evaluation:

- **Plasmacytic pododermatitis**
  Plasmacytic pododermatitis starts as a soft swelling of one or multiple footpads. Aspirates demonstrate large numbers of plasma cells. Some lymphocytes and neutrophils may also be present. Most plasma cells are well differentiated.

- **Nodular panniculitis**
  In nodular panniculitis sterile subcutaneous inflammation of fat tissue results in the formation of one or more subcutaneous nodules, which may ulcerate. A pyogranulomatous inflammation with non-degenerated neutrophils, foamy macrophages and multinucleated giant cells is characteristic for the disease. A fatty background and lipocytes are present. No microorganisms can be found.

- **Eosinophilic inflammations**
  Biopsy samples contain large numbers of eosinophilic granulocytes. This type of inflammation occurs in the eosinophilic granuloma in the cat, the lick granuloma in the dog, and also in parasitic infections and allergic reactions. Eosinophilic granulocytic infiltrates are also a characteristic finding in many mast cell tumours.

Mesenchymal cells in FNABs

Mesenchymal cells occur in all organs, as connective and supporting tissue and as a component of the blood vessels. They are also the elementary building blocks of many different tissues (connective tissue, cartilage, bone, muscle, vascular wall, adipose tissue). Mesenchymal cells are also almost always involved in repair of damaged organs, in which they can partly replace the original tissues (mesenchymal hyperplasia, scar tissue). During inflammatory processes mesenchymal cells are stimulated to cell division and proliferation to aid in tissue repair. FNABs from inflammatory processes thus also usually contain fibroblasts. These can be so strongly stimulated that they give the impression of being malignant. Hence the cytoplasm can be strongly basophilic staining and there can be remarkable large or multiple nucleoli. Giant cells with more than one nucleus, sometimes varying in size, can also be present. The presence or absence of inflammatory cells is thus of great importance in the interpretation of mesenchymal proliferations. Sporadic, well-differentiated mesenchymal cells such as fibrocytes are usually from the normal connective tissues that are also aspirated in the FNAB. If fibrocytes are found in large numbers, they can be an indication of a fibroma. In this case, there are few inflammatory cells. Malignant mesenchymal tumours can have histological characteristics of the tissue of origin (e.g. fibrosarcomas, osteosarcomas, liposarcomas, chondrosarcomas, hemangiosarcomas, leiomyosarcomas). It is not always possible to differentiate the different types of mesenchymal tumours cytologically and it can even be difficult to recognize that a malignant tumour is of mesenchymal origin. It is essential that the diagnosis of malignant tumour is only made if sufficient criteria of malignancy are confirmed and that, if the cells are of uncertain mesenchymal origin, that the proliferation of the "tumour" cannot be attributed to inflammation.
A few special types of mesenchymal tumours will be discussed:

Biopsies of osteosarcomas are usually reasonable cellular and often show cellular necrosis. Mild to moderate amounts of an eosinophilic, osteoid-like, extracellular substance surrounding the osteoblasts can be seen. Depending on the histological subtype few to moderate amounts of fibroblasts can be found as well. Less frequent osteoclasts are present. The osteoblasts have a pale blue to blue cytoplasm with eccentric nuclei, often with a Golgi like elucidation in the cytoplasm. Slight to moderate eosinophilic granulation of the cytoplasm is a distinct characteristic of malignant osteoblasts. The tumorous osteoblasts frequently have poor to moderately distinct cell borders. Distinct nucleoli, often more than 2 per nucleus, are present. Chromatin pattern is reticular to clumped. Additional malignancy criteria can be found, like angular nucleoli, anisonucleoliosis, macronucleolisation, nuclear molding and aberrant mitoses. Additional staining with alkaline-phosphatase may differentiate between osteosarcoma and other types of sarcomas.

Hemangiopericytomas are vascular neoplasms thought to be derived from pericytes. They are classified within the group of peripheral nerve sheath tumours. Cytologically, they have very distinct characteristics. The cellularity can vary from moderate to abundant. The cells are individual, spindle shape formed and have whirling-like protrusions of the cytoplasm. The nucleus is round and the cells can be binuclear or even multinucleated, forming so-called insect-head or crown like cells, respectively.

Injection-site sarcomas in the cat are located in the hypodermis and have great cellular pleomorphism and high mitotic rates. Many of these sarcomas are associated with inflammation. The inflammatory reaction is characterized by frequent aggregates of lymphocytes and smaller numbers of plasma cells. Large round macrophages with blue-gray cytoplasm, presumably associated with phagocytised adjuvant material, are commonly found within, around, or adjacent to these sarcomas. Tumours are often contiguous with granulation tissue that surrounds areas of necrosis at the vaccine site. Multinucleated giant cells are a common finding in feline vaccine-associated fibrosarcoma.

Adenomas and adenocarcinomas

Various glandular structures in and under the skin can give rise to benign tumours (adenomas) or malignant tumours (adenocarcinomas). The endocrine or exocrine gland of origin of many adenomas and adenocarcinomas is difficult to determine but a few tumours carry characteristic features, which reveal their origin.

A general diagnostic feature of a glandular epithelial tumour is of course the occurrence of tumour cells in clusters, as is true of all epithelial tumours. More specifically in cell clusters of glandular origin, an acinar structure can still be recognized. In very malignant carcinomas, however, this is lost.

Adenomas scarcely differ cytologically from normal gland tissue. The round to cuboid cells show little variation in cell size, nuclear size, and N/C ratio. The macroscopic appearance of tumour formation in combination with the cytological finding of clusters of uniform epithelial cells which may have a slightly high cell density and slightly chaotic arrangement is consistent with the diagnosis of adenoma.

Adenocarcinomas can still show some acinar formation here and there but also have a number of criteria of malignancy. In general at least four such criteria should be found for the diagnosis of "malignant" to be made. In well-differentiated adenocarcinomas, that can sometimes be difficult.

Perianal gland adenoma/carcinoma are tumours of the perianal glands that occur usually in male dogs and less frequent in bitches. They are located in the immediate surroundings of the anus, occasionally on the tail, on the prepuce, in the flank, and on the back. They are usually benign but can become malignant. The tumour is sometimes called a hepatoid tumour because the tumour cells resemble hepatic parenchymal cells. The cells lie mainly in three-dimensional clusters, but on the edge of the clusters the cells can be examined more closely. They are large cells, often egg-shaped, with an eccentric nucleus that contains one or two obvious nucleoli. The cytoplasm is usually somewhat granular/foamy. Often remnants of vascular structures can be found in the smear. Perianal tumours often ulcerate and can then be infiltrated with inflammatory cells. Sometimes there are malignancy criteria.

Anal sac adenocarcinoma is a malignant tumour that, like the thyroid carcinoma in the dog,
does not always show obvious characteristics of being malignant. Very often the biopsy reveals large numbers of monomorphous cells, frequently without any apparent cytoplasm. On careful examination, however, acinic structures can be recognized. Also some anisokaryosis, as one of the few malignancy criteria, is usually present. Morphologically the tumour cells resemble the thyroid adenocarcinomas in the dog.

**Thyroid tumours** are composed of epithelial cells that are very rich in cytoplasm. The cells exfoliate easily and also rupture quickly. The preparations are often very bloody. If, however, little blood is aspirated, the obvious acinar clusters as well as primarily loose nuclei will be found in a homogenous protein-rich background. The nuclei are about the size of lymphocytes and must not be confused with them. A FNAB from a thyroid contains in any case no lymphoglandular bodies. Here and there is usually an intact cluster of cells with an acinar structure. The nuclei show usually only sporadic malignancy characteristics such as mild anisokaryosis and multiple nucleoli. In dogs these tumours are nevertheless always considered to be malignant. In cats they are usually benign hyperplasia (adenoma).