

## PATHO-MORPHOLOGICAL DIAGNOSIS OF ORAL AMELANOTIC MELANOMA IN DOG

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Melanoma is one of the important neoplasms of canines. The present study described a histoarchitecture of amelanotic melanoma in a Doberman dog. The tumor exhibited solid, partially pigmented masses with irregular margins. Cytological analysis revealed round to oval cells with moderate anisocytosis and anisokaryosis, while histopathology showed neoplastic masses with epithelioid and spindlioid patterns. Occasional invasion of neoplastic cells into blood vessels and multifocal hyperplasia of overlying mucosa was noted. Based on these lesions the case was confirmed as amelanotic melanoma.

**Keywords:** Metastasize, Neoplasm, Pleomorphism.

Melanoma is one of the important neoplasms commonly affecting animals and humans. In dogs, melanomas account for 11.5% (Mikiewicz *et al.*, 2019) to 17.1% (Sarowitz *et al.*, 2017) of all tumours and are among the most prevalent malignant oral tumours. Malignant melanomas in dogs can arise in a wide range of anatomical locations, including the lips, mouth, skin, eyes, and digits. However, the majority of reports indicate that the skin and oral cavity are the most frequently affected areas (Lyu *et al.*, 2015). Histologically, amelanotic melanoma shows moderately pleomorphic spindled and epithelioid cells arranged into closely apposed nests within a fine fibrovascular stroma that contained occasional melanomacrophages. Spindled and epithelioid cells show moderate amounts of eosinophilic cytoplasm with distinct cell borders and occasional fine black to brown granules (Gualtieri *et al.*, 2022). The cellular features of malignant neoplasms with poor differentiation, such as carcinomas, soft tissue sarcomas, and round cell neoplasms like lymphomas, may show resemblance to neoplastic melanocytes, consequently, it makes difficult to diagnose melanoma. Immunohistochemistry targeting specific

markers like Melan-A, HMB-45 etc is being frequently used for the confirmation of malignant melanomas (Mohanapriya *et al.*, 2019). However, in field conditions at Indian context lacks the facilities for Immunohistochemistry and diagnosis of tumors relies on histo-morphological findings of the tumours. Therefore, in the present report, a case of oral amelanotic melanoma in canine is put on records.

### Materials and Methods

A six years old neutered Labrador male dog was presented to Teaching Veterinary Clinical Complex, Post Graduate Institute of Veterinary and Animal Science, Akola, Maharashtra with a history of gingival growth on right quadrant of maxilla. During the course of two and half months, the mass that extended from the right upper gingiva continued to expand. Physical examination revealed a oral mass (approx. 3 × 2 × 1.5 cm) on the right upper gingiva. Except the oral mass, no other abnormalities were noted on the physical examination. A complete blood count revealed leukocytosis with neutrophilia and mild shift to left. Further to diagnose the condition excisional biopsy was conducted and small piece of tumorous mass was

collected. Initially excised mass was clean with sterile normal saline and mobbed and impression smears were prepared and stained using Leishman stain. For histopathologic examination, the biopsy specimen was fixed in 10% neutral buffered formalin (NBF). NBF fixed tissue was processed for histopathology following standardised process, and embedded in paraffin wax. Five to six micron sections were cut using rotary microtome (Leica) and stained with routine hematoxylin and eosin (H&E) for microscopic examination.

### Results and Discussion

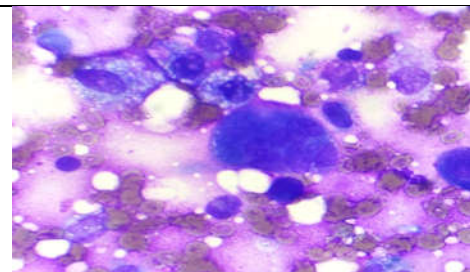
The two main types of melanocytic tumors are melanocytoma and malignant melanoma. These tumors are derived from melanocytes that originate as melanoblasts from neural crest ectoderm. Melanocytomas are the benign neoplasm of melanocytes and are most often heavily pigmented. As the name suggests, malignant melanoma is the malignant tumor of melanocytic origin and is often used synonymously with melanoma. Melanocytic tumors can be found in multiple locations; however, the integument (cutaneous) and oral cavity are common sites. In dogs, malignant melanomas are the most

common oral tumor and have variable breed and gender predilection. These tumors are often heavily pigmented and grossly appear black; however, in some cases, these tumors are variably pigmented or completely unpigmented (amelanotic), and appear grossly white which makes it difficult to diagnose. The present study described a histo-architecture of a case of canine oral amelanotic melanoma in a Labrador dog. On gross examination the tumor was located at right quadrant of maxilla having smooth surfaces and firm consistency. The mass was partially pigmented with irregular margination, non-pendulated and non-ulcerated in nature (Fig. 1). Leishman stained impression smear revealed round to oval cells with moderate anisocytosis and anisokaryosis, multinucleated cells, multiple and variably sized and shaped nucleoli, variable chromatin patterns, and variable nuclear-to-cytoplasmic ratios with cytoplasm that may contain few to many, small to large, clear vacuoles (Fig. 2). Neoplastic cells exhibited very little melanin pigment, and it may be difficult to ascertain cell origin; therefore, amelanocytic neoplasm was considered to classify cytologically.

Histopathological examination revealed



**Figure 1: An raised solid non-ulcerated slightly blackish mass extended from right upper gingival**



**Figure 2: Cytological impression smear showing neoplastic cells with pleomorphism and finely vacuolated cytoplasm and multinucleated cell (Oil immersion, Leishman's Staining)**

neoplastic mass composed of round to polygonal cells arranged in sheets and cell nests supported by fine fibrovascular stroma and expanded submucosa. These cells revealed moderate pleomorphism with slight eosinophilic vacuolated cytoplasm. The cells contain variable sized nucleus having two to

three prominent nucleolus and finely stippled chromatin. Numerous atypical cells with moderate anisocytosis and anisokaryosis and mitotic figures were also noted (Fig. 3). Neoplastic cells exhibited epitheloid and spindleoid pattern with presence of numerous multinucleated giant cells (Fig.4). There was

an occasional invasion of neoplastic cells into blood vessels. There was multifocal hyperplasia of overlying mucosa (acanthosis)

along with intraepithelial black brown pigments (moderate) in stratum spinosum. Different cellular morphologies have been

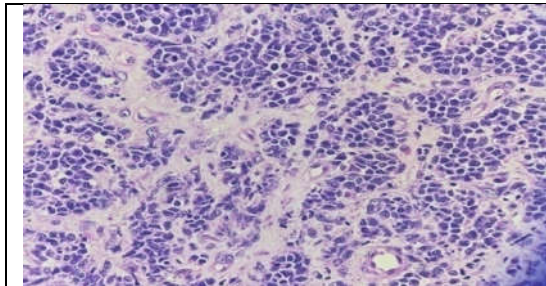


Figure 3: Photomicrograph of the tumor mass showing generalised epithelioid cells with few spindle cells arranged in small packets that are surrounded by fibrous stroma. (H&E, 200x)

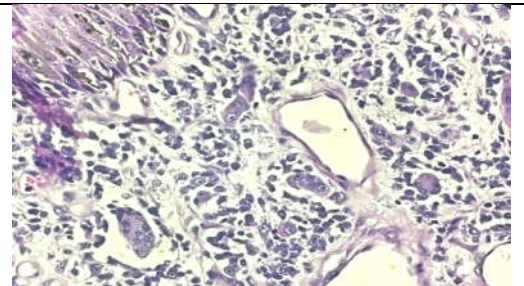


Figure 4: Photomicrograph of the tumor mass showing multinucleated giant cells. (H&E, 400x)

observed in our case, the most common being epithelioid, spindloid, and mixed epithelioid and spindloid. In our case, the tumor mass was present on the right upper gingiva of the maxilla in a mixed breed dog. Although histopathologically present case diagnosed as amelanotic melanoma, additional detection of specific immune- histochemical markers further confirms the tissue origin of the tumor as also mentioned by Przewdziecki *et al.*, 2015.

### Conclusion

In conclusion, the study elucidates the diverse histological features of canine oral amelanotic melanoma, showing its complexity and diagnostic challenges. Additionally, the identification of specific immunohistochemical markers is crucial for confirming the tissue origin, enhancing diagnostic precision. These insights contribute to a better understanding of canine melanocytic tumors, facilitating improved clinical management strategies for affected animals.

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