

What's up, Doc?

A case report of Osteosarcoma in the mandible of an Angora rabbit (*Oryctolagus cuniculus*).

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Introduction

Osteosarcoma is the most common bone cancer in children and dogs. It is ten times more common in dogs than in people (4). These primary malignant bone tumors make up more than 80% of bone tumors in dogs and 70% in cats but are rare in domestic rabbits. In dogs, the typical signalment is a middle-age to old, large or giant breed, with an increased prevalence in Rottweilers, Greyhounds, and Great Danes. The typical lifespan of a rabbit is anywhere from 4-10 years however a geriatric rabbit is any over the age of 4 years old (2). Though giant breeds of rabbits exist, (i.e. Flemish Giant), there does not appear to be an increased prevalence of osteosarcoma in giant rabbit breeds (4). There have been reports of osteosarcoma in hamsters, a rat, a hedgehog, a wolf, and a goat (12). The most common tumor in rabbits is the uterine adenocarcinoma followed by lymphoma (6). In the rare instances when rabbits are diagnosed with osteosarcoma, they are usually middle-age to older. The age range of cases reported is 1.5 years to ten years with the median age being seven years old (5). There does not appear to be a gender predilection. Rabbits with osteosarcoma usually present with fever, anorexia, and lameness or soft-tissue swelling around lesions. In rabbits with mandibular or facial bone osteosarcoma, the first signs owners notice are dysphagia, dropping food, and ptyalism (9). Owners may claim these signs are acute, however, it is an insidious process.

Osteosarcomas most commonly occur in the extremities, specifically the metaphyseal region of long bones, with forelimbs being approximately twice as likely to be affected than hindlimbs. This parallels the ratio of weight distribution (60% forelimb, 40% hindlimb). In dogs, the most common site for osteosarcoma is the distal radius, followed by the proximal humerus, and then the distal femur (away from the elbow and towards the knee). Mandibular osteosarcoma has been reported most frequently in laboratory rabbits, but the predilection sites are unknown.

Once the diagnosis has been confirmed, the tumor then needs to be staged. The current staging system used is the Musculoskeletal Tumor Society staging system. This system uses tumor grade (*I* being low-grade and *II* being high-grade), tumor extension (*A* being intraosseous involvement only and *B* being extra osseous extension on top of the interosseous involvement), and the presence of metastasis (11). Histologically, these tumors are classified into variants in dogs, but no classification system has been developed for rabbits. In dogs, the tumors are classified as poorly differentiated, osteoblastic, chondroblastic, fibroblastic, telangiectatic and giant cell types. Osteoblastic tumors have been reported most often in rabbit osteosarcomas, using the canine classification system (5). Fibroblastic and giant cell types have also been reported (5,6). The osteosarcomas present in the skull or mandible have all been reported as productive osteoblastic tumors.

Histologic examination of osteosarcomas usually is composed of sheets of round to polyhedral neoplastic cells resembling histiocytes with round to oval nuclei (6). Karyomegaly and anisokaryosis are usually present (8). Osteosarcoma cells are characterized by basophilic cytoplasm, hyperchromatic nuclei, and prominent nucleoli. The cytoplasm will often exhibit granules, indicating osteoid production (8). These tumors are highly cellular and pleomorphic, with high mitotic figures and numerous giant cells. A histologic feature specific to the rabbit is either the presence or absence of osteocalcin. Also known as bone gamma carboxyglutamic acid containing protein, Osteocalcin is a non-collagenous protein and specific protein maker for osteoblasts, which helps to regulate osteogenesis (12). Healthy cytoplasm of rabbit bone osteoblasts are positive for osteocalcin. In contrast, osteoblasts invaded by osteosarcoma are negative for osteocalcin (6).

Without treatment, the rabbit may die within a few weeks (8). Drugs used to treat osteosarcoma are Cisplatin and Doxorubicin. Complete surgical resection is crucial in treating osteosarcoma. If adequate surgical margins are not achieved, local recurrence rates are about 25% (11). If lesions are present in the lungs, the patient should undergo a thoracotomy. Microscopic lesions of tumor may exist in the lungs and 90% of recurring osteosarcomas affect the lungs (11).

History & Presentation

Mischa, an approximately 10-year-old male intact Angora rabbit, presented to MSU-CVM Community Veterinary Services on April 15, 2019 for excessive salivation and alopecia. Three weeks prior, the owner noted that Mischa was struggling to keep food in his mouth and started salivating to the point that it would drench his face, neck, and chest. Alopecia was noticed in these areas.. The alopecia appeared to be associated with excessive grooming. Mischa was reported to be lethargic and uninterested in his normal activities. He was taken to the North Madison Animal Hospital in Jackson, TN, where he was prescribed enrofloxacin to treat a potential abscess in his mouth. The previous year Mischa had an infection in his mouth that cultured *Pasteurella multocida*. The infection had been treated with enrofloxacin and resolved uneventfully. It was also recommended 3 weeks ago to switch Mischa to a critical care diet, which the owner did. Mischa did not improve with the enrofloxacin, so he was referred to MSU. Mischa lived as a free roaming pet in the living room. He urinated and defecated on pee pads that the owner once soiled. He remained inside only with one guinea pig. He normally ate Oxbow pellets but since his clinical signs started, his diet was switched to Oxbow Critical Care Diet. Mischa was bought from Tractor Supply 10 years ago as a kit.

Upon presentation to MSU- CVM, Mischa was bright and alert. He had a decreased body condition score of 3/9, with 4-5/9 being ideal, and weighed 1.59 kg. He had hyperemic and moist mucous membranes, with a capillary refill time of less than 2 seconds. He was tachycardic with a heart rate of 176 beats per minute, with normal being 120-136 bpm, and his respiratory rate was normal at 32 breaths per minute (2). Mischa's heart and lungs auscultated within normal limits with no murmurs, arrhythmias, wheezes, or crackles noted. Mischa exhibited ptyalism and had saliva covering his face and chest. There was a palpable square mass 2cm x 2 cm just dorsal to the lower incisors. It extended past the teeth, firm on palpation, and was white in color. The mass was adhered to the lower gum line. Mischa had severe gingivitis and marked alopecia in his dewlap region, as well as moist, hyperemic skin. Exophthalmos was noted of his right eye but was absent in the left. His right orbital bone was bulging but vision did not appear to be impaired.

Pathophysiology

Osteosarcomas develop from mesenchymal stem cells with the capacity to make osteoid. They can develop from bone, fibrous tissue or cartilage (4). The most common recurrent abnormality in canine osteosarcoma is loss or mutation of TP53 tumor suppressor gene. There is also evidence to suggest that the elimination of p53 function is required for the development of osteosarcoma in dogs, however this has not been studied (7.). Approximately 60% of canine osteosarcomas over-express the p53 protein, correlating to the presence of missense point mutations within the DNA (4).

Radiographs usually show bone lysis, soft tissue swelling, enlarged lymph nodes, enlarged liver, and micrometastases to the lungs (5). Though it may not be radiographically evident (less than 15% of the time (4)), pulmonary metastasis is present 90% of the time at

diagnosis (5). To confirm diagnosis, biopsy is always required (11). However, plain radiographs of the involved bone showing a sclerotic or lytic lesion are characteristic for osteosarcomas. The cortex of the bone is eroded through by the tumor, elevating the periosteum. This results in soft tissue swelling. Computed tomography (CT) or magnetic resonance imaging (MRI) are usually required to determine tumor size and involvement.

Another common abnormality found in patients with osteosarcoma is an increased serum alkaline phosphatase (ALP) with the heightened range of ALP being 77- 164 U/l (5). The normal reference range of ALP in rabbits is 4-16 U/l (6). ALP is an enzyme found in liver, intestine, bone, kidney, placenta and many other tissues. The higher the level of ALP, the worse the prognosis. An elevated ALP is usually seen in patients with osteosarcoma, however, this can be difficult to detect in young patients as their normal ALP levels are naturally higher due to growth and development (11).

It is uncommon for osteosarcoma to cross or spread through joint space and into the connecting bone. According to the hematogenous route supported by rabbit necropsies, which have shown metastatic emboli in blood vessels, the normal first place of metastasis is the lungs, followed by proximal bone, and finally lymph nodes and other soft tissues (8).

Diagnostic Approach

After completing a thorough physical exam, it was determined that the gingival mass was likely to be the cause of Mischa's illness. A plan was developed to locate the origin of the mass and the cause of exophthalmos. Mischa was sedated using meloxicam 5mg/ml 1mg/kg (0.32ml) IM and butorphanol 10mg/ml 0.5mg/kg IM (0.08ml). He was maintained on 1% isoflurane while thoracic and skull radiographs were performed. Five skull images were evaluated from 4 different radiographic views. Irregular new bone formation was seen around the tooth roots of

the left and right maxillary and mandibular premolars and molars. Permeative lysis and expansion of the left zygomatic arch and the left facial tuber of the maxilla were present. There was soft tissue swelling along the ventral aspect of the mandible with undulating cutaneous margins. There was protruding soft tissue opaque material lateral to the left and right orbits. Possible causes for these radiographic changes were chronic tooth root abscessation with granuloma formation, dystrophic mineralization, osteomyelitis or neoplasia. The soft tissue swelling may have been due to cellulitis, edema, or less likely, neoplastic infiltration.

Three images depicting three different radiographic views of the thorax were evaluated. A mild, diffuse unstructured interstitial pulmonary pattern was seen on all views. On the lateral projections, there were ill-defined, ovoid soft tissue opaque nodules, the largest measuring approximately 2 mm x 1 mm. Possible causes for the unstructured interstitial pulmonary pattern were atelectasis, infectious/inflammatory etiologies, and neoplasia. The ill-defined nodules within the lung parenchyma may be due to end-on vessels, metastatic neoplasia, or granulomas.

After his radiographs were taken, Mischa was transported to the Small Animal Dental Procedures room for a thorough oral exam under anesthesia. In addition to the mass noted rostral to the left mandibular incisor, a second larger mass was noted rostral to the mandibular cheek teeth approximately 8mm x 8mm. Three punch biopsies of the gingival masses were removed, placed in formalin, and submitted to the lab for histopathology. At this point, our differential diagnoses were neoplasia, granulomatous disease, or abscessation. A preliminary evaluation of cytology indicated osteosarcoma; however, a definitive diagnosis could not be made without the biopsy results.

Due to Mischa's grave prognosis and age, palliative care or euthanasia were our best options. Surgery to remove the tumors could have been performed, however the risks were

significant. Because of the evidence of metastasis and multiple tumors, Mischa's prognosis was grave. Chemotherapy and radiation protocols have been done infrequently in rabbits but were not an option at this time. We recommended that Mischa be continued on pain medication (Meloxicam 1mg/kg dose- administer 0.5mL orally every 12 hours) and that his owner continue to monitor his quality of life. Euthanasia needed to be strongly considered within the next few days due to the extent of the disease. It was unfortunate that only palliative care could be recommended, however, due to the advanced nature of the disease and type of cancer, it was the best option available.

Treatment & Management

The treatment of choice for osteosarcoma is surgery. However, due to the likelihood that pulmonary metastasis will be present, anesthesia is a high risk. Surgical complications are most consistently due to respiratory disease and cardiac arrest during anesthesia (9). In this case, the preferred treatment would have been a radical hemimandibulectomy. However, rabbits have elodont teeth which make malocclusion a probable post-operative complication. Assisted feeding and frequent dental care would be required for the remainder of the rabbit's life (9). Prognosis is good in surgical cases without metastasis and patients have a relatively long survival time once recovered from surgery.

Meloxicam is the most studied NSAID used in rabbits (3). It is thought to have less side effects than other NSAIDs because of its greater COX-2 selective activity (2). Based on recent studies, rabbits appear to metabolize meloxicam faster than other mammals. The appropriate dose to achieve optimal analgesic effects is 1mg/kg (2). In dogs, chemotherapy protocols with either cisplatin or carboplatin and doxorubicin have shown improved survival time after amputation (4),

Radiation therapy has been tried in the treatment of some rabbit tumors. Rabbit thymomas appear to respond well to radiation therapy (1). Though this therapy improved long-term survival, it came with potentially fatal side effects. Rabbits experienced radiation-induced pneumonitis, myocardial failure, and liver failure (1). In another study, radiation was used to treat adenocarcinoma in rabbits. The radiation therapy was started 4 weeks after surgical removal of the tumor (10). All rabbits undergoing this treatment developed binocular cataracts and coat changes. The coat changes resolved after treatment ceased and the cataracts remained static. These patients remained healthy and cancer-free on CT at the one, six, and twelve-month follow up examinations (10). Another treatment option that has been studied experimentally is doxorubicin chemotherapy to treat renal carcinoma in rabbits (13). Local administration of doxorubicin was performed at 5mg/kg of doxorubicin IV via renal artery perfusion. In this case, the doxorubicin significantly reduced tumor volume (13).

Case Outcome

A few days after Mischa's discharge, the official biopsy results were confirmed. Expanding the lamina propria was a poorly demarcated, unencapsulated, densely cellular infiltrative neoplasm composed of mesenchymal cells entrapped in an amorphous eosinophilic matrix (osteoid) and woven bone. Cells had indistinct borders, scant eosinophilic cytoplasm, a round to elongate nucleus, coarsely stippled chromatin and multiple small nucleoli. There were few karyomegalic and multinucleated cells with marked anisocytosis and anisokaryosis. There was a large number of mitotic figures. The histopathology report confirmed the presumptive diagnosis of osteosarcoma. A follow up phone call to North Madison Animal Hospital revealed that Mischa was humanely euthanized about a week after diagnosis due to deteriorating quality of life.

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