Hope for Hemangiosarcoma -Biology Based Treatment Options
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Hemangiosarcoma (HSA) is a tumor with which most veterinary practitioners are well familiar. For most dogs, the diagnosis of HSA is a harbinger of bad things to come; most veterinarians consider this tumor to be the "kiss of death". Improvements in diagnosis and therapy of HSA have been slowly emerging, and new therapeutic opportunities are on the horizon. All HSA are highly malignant neoplasms. Most primary HSA lesions arise in the spleen, liver, right atrium or subcutaneous tissues. Approximately 25% of cases have concurrent right atrial and splenic involvement at the time of diagnosis. Regardless of the site of origin, local infiltration and systemic metastases are the common growth patterns. Some investigators have suggested that the skin and subcutaneous sites are less aggressive but metastatic behavior from these locations has been observed. True intradermal HSA can arise in non-pigmented, non-haired skin of dogs and cats as a result of chronic ultraviolet light exposure. These lesions may behave in a more benign manner, as compared to lesions that arise at deep tissue locations that are non-actinic in etiopathogenesis.

Hematogenous metastatic spread occurs as an early event in the natural history of HSA. Metastatic sites are widespread, with the lung and liver being the most frequently affected organs. Many other tissues are sites of HSA metastases including kidney, adrenal glands, lymph nodes, muscle, brain, mesentery, and skin. Regardless of location, morbidity and mortality is often due to acute internal hemorrhage secondary to tumor rupture. Dogs require stabilization and acute emergency management at the outset when the disease is diagnosed. Diagnostic evaluation consists of abdominal ultrasound or radiographs, 3 view thoracic radiographs for metastatic disease, potentially echocardiography to evaluate for an atrial mass, and a CBC, serum chemistry profile, and coagulation studies. A study published in 2004 from Ohio State suggested that contrast-enhanced CT scanning of the spleen may be valuable in differentiating benign from malignant lesions. Because up to 50% of dogs presenting for splenic lesions ultimately are found to have benign disease rather than hemangiosarcoma, it is important to establish the diagnosis through biopsy. We perform fine needle aspiration cytology of splenic and liver lesions, after warning clients that these assays may necessitate moving the schedule of surgery up if acute bleeding is induced. The intent of ultrasound-guided biopsy is to try to avoid highly cystic areas in favor of more solid areas for aspiration. Hemangiosarcoma cells are often anaplastic on cytology and resemble high-grade spindle cell tumors, with occasional epithelioid differentiation as well. Immunocytochemically, these lesions should be positive for vascular endothelial markers such as Factor VIII-related antigen and CD31. Experimentally, HSA cells are also positive for CD117 (c-Kit), a the hematogenous stem cell factor receptor, CD105 (endoglin surface marker) and alpha(v)beta(3) integrin, which is a marker
of neoangiogenic blood vessels.

HSA should be considered a systemic disease requiring a multimodality approach to therapy. After definitive treatment of the primary tumor, rigorous long term follow-up consisting of hematologic, radiographic, and ultrasonographic studies. Repeat radiographs and ultrasound exams are conducted every 6 to 8 weeks. Once diagnosed, dogs should be treated with adjuvant chemotherapy, optimally combined with some form of inhibitor of angiogenesis.

Surgical Considerations  -Ruptured splenic tumors should be considered a surgical emergency. Animals presenting with an abdominal mass, collapse, pallor and hemoperitoneum should be treated for shock, given a transfusion of packed cells or whole blood and taken to surgery when stable. Although, some clinicians prefer to stabilize overnight, I find that a significant proportion of these patients are in DIC and are at risk for fulminant, lethal hemorrhage. Removal of the primary tumor is the best treatment to prevent or address DIC and many animals rapidly stabilize after surgery. Treatment for splenic hemangiosarcoma is splenectomy followed by adjuvant chemotherapy. Splenectomy will relieve abdominal distension caused by the tumor and halt bleeding. It is very important to realize that one cannot distinguish between benign and malignant tumors of the spleen by gross examination. Similarly, the surgeon should avoid assuming that any lesions seen in the liver at the time of surgery are definitive metastases. Nodular hyperplasia may appear similar to metastatic disease; therefore, a liver biopsy is always indicated. The important message here is: never euthanize a patient suspected of having primary splenic hemangiosarcoma without histopathologic confirmation, even if it appears that metastatic disease is present in the liver. Many clinicians have been grieved to find at necropsy that the primary splenic tumor was a benign hematoma and the nodules in the liver that were thought to be metastatic foci were nodular hyperplasia.

Splenectomy is rapidly accomplished using the LDS stapler, which ligates, divides and staples with one fire of the instrument. The instrument can fire 10-2 times without the need for a new cartridge. A splenectomy can be accomplished in about 20 minutes using this device. Although the staples are moderately expensive (approximately $100 per cartridge-client cost), the additional cost is often countered by significantly decreased time under anesthesia. The operative time saved may be important in critical patients.

Radiation Therapy Considerations  -The role of radiation therapy for curative treatment of hemangiosarcoma in dogs is limited. Although irradiation may be beneficial for achieving local tumor control, it is difficult to justify putting a patient through extensive treatment when metastatic disease is imminent. However, there may be a role for radiation in the palliation of bulky, non-resectable subcutaneous
hemangiosarcoma lesions. The goal of palliative radiation therapy is to relieve pain or improve function and or quality of life in patients with systemic disease, and to cause minimal treatment related effects. An overview of the palliative treatment of solid soft tissue malignancies (Blake, et al, VCS Proceedings 1998), included a description of the treatment of 5 cases of Stage III non-resectable hemangiosarcoma. All of these dogs had reported improvements in quality of life assessment, and a complete response was seen in one dog, partial remission in one dog, stable disease in 2 dogs. One dog was non-evaluable for response. Duration of responses in these cases ranged from 12-42 weeks. The radiation was administered in 6-8 gy fractions and patients received 2-4 fractions. The major reason cited for improvement in these patients was cessation of bleeding and bruising at the tumor site.

Chemotherapy and Medical Management Considerations -The initial treatment for dogs with HSA is most often surgical excision of the primary lesion. Many dogs will be presented in hypovolemic shock because of acute hemorrhage and will require an immediate whole blood transfusion. This will provide needed RBCs, platelets, and coagulation factors. Most emergency/critical care specialists have an opinion of the best treatment protocol for managing DIC; however, there is no general consensus among these specialists. At Michigan State, we tend to avoid heparin in these cases except as a last resort. The protocol in use at the University of Wisconsin, Madison includes heparinization. Dogs in DIC have heparin added to the bag of transfused blood (75 IU/kg patient body wt), which is incubated at room temperature preferably for 30 minutes prior to transfusion. This serves to activate antithrombin III, a protein that inactivates thrombin and factors XII, XI, X, and IX. Continued heparinization consists of minidoses of heparin (75 IU/kg) injected subcutaneously every 8 to 12 hours. Probably the most important goal of treating the DIC (in addition to removing the HSA) is providing a source of antithrombin III to associate with the heparin. This is best accomplished by transfusion. The low dose of heparin is unlikely to promote any added risk of surgical bleeding should emergency surgery be contemplated. Because of cardiac hypoxia, some dogs will have premature ventricular contractions, increasing their surgical risk. Arrhythmias should be managed appropriately, and surgical resection of the splenic tumor will often result in transient, self-limiting arrhythmias.

Most dogs with completely resected HSA do not experience long-term survival due to development of metastatic disease that is not amenable to surgical removal. Median survival times for dogs following splenectomy have ranged from 56 to 70 days. However, small numbers of dogs have survived more than one year following splenectomy alone for HSA. Dogs treated for right atrial HSA by surgical excision had a reported survival of 90 to 150 days. Only small numbers of patients have tumors suitable for this type of surgery, and the post-operative morbidity and prolonged recovery period after thoracotomy in the face of inevitable
and widespread metastasis makes this therapeutic recommendation questionable for most dogs. We are currently following a cohort of dogs at MSU that had right atrial appendage amputation followed by chemotherapy. The study is ongoing, but we currently have 2 dogs alive 6 months after surgery.

The use of chemotherapy following excision of HSA appears to provide the longest survival times. Doxorubicin is the most effective single agent in the treatment of HSA. A study from Penn evaluated 20 dogs treated with a dose intensified doxorubicin protocol for canine HSA. These dogs received 30 mg/M2 doxorubicin at 2 week intervals for 5 treatments. Results were median survival times of 257, 210, and 107 days respectively for Stage I, II, and II disease. This is not appreciably different from other reported studies. The most efficacious protocols include doxorubicin and cyclophosphamide with or without vincristine (VAC protocol). The median survival time of 18 dogs treated with VAC, in combination with surgery, was 190 days with 30% surviving at least one year. The VAC protocol has been modified to include several variations at individual cancer centers. The VAC protocol currently in use at MSU is as follows:

Day 1: Doxorubicin 30 mg/m² IV Cyclophosphamide 100-150 mg/m² IV or 50 mg/m² PO on days 3, 4, 5, and 6 of week 1 only

Day 8, 15: Vincristine 0.75 mg/m² IV (At MSU we routinely lower the dose to 0.5 mg/m² IV on Days 8 and 15 to reduce the incidence of myelosuppression and GI signs).

The cycle is repeated on day 22. Four to six cycles are administered. Myelosuppression is appreciable and for many dogs, and it may be necessary to use the lower dose of cyclophosphamide. If the neutrophil count is less than 1500/ml on day 8, the treatment is postponed for one week. Trimethoprim-sulfa can also administered orally from day 1 to 14 to minimize systemic bacterial infections. Approximately 25% of VAC-treated dogs show GI toxicity and some will require hospital attention. A protocol involving doxorubicin and dacarbazine, with or without vincristine, may also be useful for treatment of HSA in dogs. We are currently evaluating DAV as an open pilot study of dogs with both measurable non-resectable HSA and as an adjuvant treatment. We are accruing a cohort of approximately 25 dogs to assess response, toxicity, and survival duration. We have seen responses to this protocol in the face of bulky non-resectable subcutaneous, hepatic, and pulmonary metastatic disease, which allowed us to move the protocol forward into an adjuvant disease setting. Data are maturing, but the protocol shows promise as an adjunctive and salvage protocol. The DAV protocol for soft tissue sarcomas is administered at 21-day intervals for 4-6 total cycles. The protocol is as
follows:

Day 1: Doxorubicin 30 mg/m^2 IV
Dacarbazine 800 mg/m^2 IV as an 8-hour infusion or 200 mg/m^2 IV as a 15 minute infusion on days 1-4 of week one only
Day 8,15: Vincristine 0.5 mg/m^2 IV

This is an aggressive and myelosuppressive protocol that requires careful monitoring to manage leukopenia and the potential for sepsis. For dogs less that approximately 0.75 m^2, it may be necessary to perform dose reduction on the first cycle of therapy, and attempt escalation to full dose therapy on successive cycles if the protocol is tolerated by the patient. Gastrointestinal toxicity in the form of vomiting while receiving the infusion necessitates the addition of metaclopramide injection at the end of the infusion period. We generally prescribe metaclopramide orally for 3 days on a PRN basis after the 8-hour DTIC infusion.

We have developed a milder protocol for patients that are physiologically compromised. We have observed partial and complete responses with this protocol, although numbers of patients treated are limited at this time. This protocol is modified from a human soft tissue sarcoma protocol used at the Mayo clinic. What we call the "soft tissue sarcoma" protocol alternates a milder 3-week cycle (VAC with actinomycin substituted for doxorubicin) with a more aggressive chemotherapy protocol cycle (essentially DAV). This soft tissue sarcoma protocol is administered as follows:

Day 1 Actinomycin 0.6 mg/m^2IV
Days 3,4,5,6 Cyclophosphamide 50 mg/m^2 PO
Day 8 Vincristine 0.5 mg/m^2 IV
Day 15 Vincristine 0.5 mg/m^2 IV
Day 22 DTIC 800 mg/m^2 IV infusion over 8 hours
Day 23 Doxorubicin 25 mg/m^2 IV
Day 31 Vincristine 0.5 mg/m^2 IV
Day 38 Vincristine 0.5 mg/m^2 IV
Rest for 1 week if blood count indicates. Repeat cycle at least one more time if evidence of beneficial response.

**Immunotherapy and Novel Therapy Approaches**

Currently, surgery followed by adjuvant chemotherapy, usually doxorubicin combined with cyclophosphamide, is the standard of care for this tumor. Despite these treatments, dogs still die early from metastases. Use of immunomodulatory
drugs, such as interferon-a 2a (Roferon®) combined with the chemotherapeutic regimen postoperatively has been attempted in an investigational pilot study. In human medicine, interferon-a is used to treat tumors of endothelial origin (e.g., hemangioma, Kaposi sarcoma) where it has been shown to exert an anti proliferative effect. Following splenectomy for hemangiosarcoma, clients administer interferon-a subcutaneously to their dogs at home and its relatively low price is attractive (Roferon-A®, Roche's interferon alpha-2a recombinant). At the University of Wisconsin Madison, the protocol starts with a low daily dose (e.g., 0.25 x 10^6 IU/m^2 per day) concurrently with administration of standard chemotherapy. The dose is gradually escalated to 1-3 x 10^6 IU/m^2 daily. However, at the high-end target dose, the cost for a large dog may be more than $1000/month. Too few dogs have undergone this regimen to draw conclusions regarding the efficacy of interferon-a in canine hemangiosarcoma. Anecdotally, some treated dogs are alive over one year.

There is much enthusiasm for antiangiogenic therapies that are looming on the horizon. Canine hemangiosarcoma is an ideal tumor for these agents because it arises from a transformed endothelial cell. Clinical effects of antiangiogenic substances have been evaluated with agents such as endostatin or the canine endostatin gene in a study of dogs with cancer. Other strategies rely on blocking adhesion molecules and growth factor receptors of endothelial cells forming the growing vasculature of tumors. These same surface molecules have been found on canine hemangiosarcoma cells. The integrin avb3 is an adhesion molecule preferentially expressed on endothelial cells forming new blood vessels such as those found in the tumor environment. Antibody blockade of avb3 results in endothelial cell apoptosis prompting clinical testing of this approach in human trials. Alternatively, avb3-targeted delivery of chemotherapeutic agents using the ligand for avb3, a nine amino acid sequence that binds avb3, to target the cytotoxic agent doxorubicin, has been described. Targeting of chemotherapeutic agents or other substances that may inhibit endothelial cell growth (e.g., vascular endothelial growth factor receptor antibody) represent promising strategies for canine hemangiosarcoma. Although a trial with the nonspecific macrophage activator L-MTP-PE extended the survival of dogs with splenic hemangiosarcoma (in combination with surgery and doxorubicin/cyclophosphamide), this agent will not be available for veterinary clinical practice. The study, however, helped to prove a role for immunotherapy in treating canine hemangiosarcoma.

Current interest in novel therapy for canine HSA revolves around the combination of tumor hypoxia and vascular endothelial growth factor over-expression. Vascular endothelial growth factor (VEGF) has been shown to be increased in the serum and abdominal effusions of dogs with HSA, and has been demonstrated to be produced by the malignant endothelial cells and tumor inflammatory infiltrates.
Unfortunately, VEGF level in hemorrhagic abdominal effusion cannot differentiate between benign and malignant causes of hemoperitoneum. In human patients with heritable endothelial tumors, a mutation in the von Hippel-Lindau gene has been identified. The VHL gene functions to decrease the activity of a hypoxia-inducible factor called HIFα. Under hypoxic conditions, HIFα expression is triggered and results in transcription of a variety of genes involved in response to hypoxic stress, including apoptosis resistance pathways and VEGF. Under normal conditions, HIFα is expressed but rapidly degraded as a consequence of interaction with the VHL protein. Defects in VHL allow persistence of HIFα, with subsequent proliferation of apoptotic resistance vascular endothelial cells in human beings, although this pathway has not been definitively explored in canine HSA. It may be that this pathway will prove important in both the pathogenesis of canine HSA, and may also provide a biologically rational therapeutic window for future targeted therapies. Because of improved understanding in the underlying pathogenesis, improved therapies can be developed for this deadly and all-too-common canine
cancer.