

A STUDY OF 76 CANINE HEMANGIOSARCOMAS
RECORDED AT MICHIGAN STATE UNIVERSITY
FROM 1956 TO 1965

Thesis for the Degree of M. S.
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ABSTRACT

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by Ernest E. McConnell

This thesis is a retrospective study of 76 canine hemangiosarcomas diagnosed in the Department of Pathology from 1956 to 1965. The study was divided into 3 basic parts: (1) occurrence and gross characteristics, (2) microscopic characteristics, and (3) radiographic characteristics of the tumor.

Hemangiosarcomas occurred with greater incidence in the large breeds of dogs, particularly the Boxer. There was a sex ratio of approximately 2 males to 1 female. The mean age was 9 years, a little earlier in males (8.5 years) and a little later in females (10.7 years). The most common primary sites were the subcutis and the spleen. If metastasis occurred the most common site was the lung. The tumor was difficult to diagnose without histologic examination. Hemorrhage was an almost constant finding.

The neoplastic endothelial cell was generally spindle shaped, resembling a short fibroblast. The nucleus was generally oval and of moderate size (9 x 12 microns), often containing large eosinophilic single and multiple nucleoli. There was a tendency for the cytoplasm of 1 cell to fuse with the next, presenting a syncytial type of arrangement rather than individualized cells. There were both normal and abnormal mitoses. Most of the nuclei had a moderate

amount of hyperchromatism. The cellular arrangement varied from normal appearing vascular cells to undifferentiated cellular sheets arranged in whorls or clumps with little vessel formation. The use of a reticulum stain revealed a fine reticular meshlike network, even in the most anaplastic areas of the tumor. There was a variable amount of necrosis, especially associated with hemorrhages. The amount of stroma varied from large amounts in the vascular forms to scanty in the more anaplastic cellular forms. Invasion of blood vessels occurred in 29 of 59 hemangiosarcomas. There were inflammatory cells in many of the tumors, particularly those which had an eroded surface.

Two radiographic characteristics were observed. First, the metastatic lesions in the lungs presented a "snowflake" pattern. Secondly, osteolysis occurred in those neoplasms that were in or near bone.

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INTRODUCTION

"Tumors of animals are not fundamentally different from those of man. Therefore, a much broader view on the comparative pathology should be taken; in accordance with the trends of cancer chemotherapy, tumor pathogenesis, and etiology, natural cases of cancer in animals could be utilized to considerably greater advantage for the study of fundamental problems of cancer research than has been the case thus far."

(Meier, 1963)

This thesis is a retrospective study of 76 canine hemangiosarcomas collected from 1956 to 1965. The tumors were either diagnosed in dogs submitted to the Michigan State University Veterinary Clinics or diagnosed on the basis of specimens referred to the Department of Pathology by private veterinarians.

The specific objectives of this study were to learn more about (1) epizootology, (2) histologic characteristics, (3) radiographic characteristics, and (4) diagnostic criteria.

REVIEW OF THE LITERATURE

Arey (1954) stated that both the blood cells and the blood vessels arose from mesenchyme. The earliest definitive cell with this potential was called the angioblast, while the process of vessel development was known as angiogenesis. Originally, there was a solid mass of these angioblastic cells which eventually developed lumen. In the process, the peripheral cells formed flattened endothelium. For a short time, the arteries and veins were not distinguishable. Ham (1957) pointed out that normal endothelium was a continuous lining formed by the junction of 1 endothelial cell with another. Palade (1953) demonstrated in a detailed ultrastructural study that in addition to the usual cellular elements (nucleus, mitochondria, endoplasmic reticulum, and small granular components) endothelial cells possessed 2 additional characteristic structural elements: (1) intracellular fibrils of $240 \text{ \AA}$ diameter and (2) a large number of vesicles concentrated immediately under the cell membrane facing both the capillary lumen and intercapillary spaces. These averaged $650 \text{ \AA}$ in diameter. Many of these vesicles appeared to open at the surface of the cell membrane. In addition, a thin, irregular layer of relatively dense material surrounded the outside surface of the endothelium. In places it appeared as a belt of extraordinarily fine fibrils. This layer corresponded to the basement membrane. In addition, Stout (1943) pointed out that the growth of capillaries in granulation tissue occurred by the sprouting of endothelial cells

from pre-existing capillaries, forming first a solid cord which secondarily became canalized. Referring to hemangiosarcoma, he stated that "probably all of the malignant tumors exhibit this type of growth in their infiltrative growth".

Joest (1924) cited Leisering (1861) and Siedamgrotsky (1874) as being the first 2 authors to describe tumors of vascular origin in the dog. Leinaux (1899) described 2 dogs with hemangioendotheliomas of the parotid region. However, these appeared to be benign tumors. Ragenbagen (1907) reported 4 hemangiomas of the cavernous type in dogs between 6 and 10 years of age. One of these animals had 2 hemangiomas.

Kingman and Newsom (1918) were the first American authors to publish a case report of a malignant tumor of endothelial origin in animals. They described an aged German Shepherd dog which had the habit of opening a screen door that had a stout spring on it; as the dog passed through, the door usually struck him on the side. An enlargement in the area occurred over a 4-month period, measuring 8 x 14 x 4 inches. The tumor was soft, and upon insertion of a small cannula, blood exuded. At necropsy examination, nodules of the same type as on the side of the body were observed throughout the lungs. Microscopically, the tumor had large cavernous spaces filled with blood, although in some areas there was little blood. A diagnosis of malignant endothelial blastoma was made.

Feldman (1932) classified this tumor in his textbook on veterinary oncology as a malignant hemangioendothelioma. He described the tumor as usually localized but possibly metastasizing to widely separated areas of the body. It usually infiltrated tissue, but it

was exceptional for a hemangioendothelioma to become severely destructive. Grossly, the tumor was typically dark blood red to purple and had a soft consistency. On pressure blood exuded, to be replaced when pressure was relieved. If punctured, hemorrhage was profuse. Microscopically, there was a tendency to form multiple, irregular, and immature capillaries. The capillary spaces or cavities sometimes varied in size. The cell lining tended to push into the lumen and eventually filled most of it. Frequently, solid nests or irregular sheets of cells resembled a sarcomatous structure. Some had small whorls with no lumens. Blood disintegrated due to being removed from circulation. The type of cell also varied. Many cells were oval or polyhedral, or sometimes flattened and almost spindle shaped. The cytoplasm was basophilic and the nuclei, which were fairly large and oval, contained many fine chromatin granules. In some, mitotic figures were numerous.

Mallory (1914) described the tumor as it occurred in man and referred to it as an endothelial blastoma. Shennan (1914) published a case report for the reason that although this tumor was histologically benign, it metastasized. There was no sarcomatous transformation in the tumor. Other authors (De Navasquez, 1936; Thiel, 1904; Langhans, 1879; and Geshickter and Keasbey, 1935) stated that while malignant tumors of blood vasculature were rare, they occurred and were usually benign in their appearance microscopically.

Magnusson (1934) stated,

"From foci with endothelium-like cells there are all sorts of transitions to tissue resembling fibrosarcoma with a fairly great amount of stroma. ... It is impossible to decide which cell of the tumor originates from the endothelium and which comes from the adventitia.

... The tumor consists partly of solid parts, partly of various sized blood-filled lumina lined with endothelium. The lumina are mainly bound by low cells provided with large lancet-shaped nuclei with a well developed chromatin network. The lining cells of some of the smaller lumina, particularly in the more central parts of the tumor, differ from this by exhibiting abundant polymorphism from round to cuboidal forms to lower and flatter cells."

Robinson and Castleman (1936) summarized this form of the tumor and gave it a specific title - "benign metastasizing hemangioma". They concluded that "the occurrence of metastases should be the deciding factor and not the histologic features" and that therefore "the primary tumor, in spite of the absence of histologically malignant properties, was not benign".

Freilich and Coe (1936) first reviewed the literature on "angiosarcomas" from 1918 to 1934. They were able to locate only 29 reports of this tumor in man. Lasserre et al. (1938), in their studies of cancer in dogs from 1933 to 1937, noted "the majority of blood vessel tumors are localized in the subcutis". They concluded that the average age of dogs afflicted was 4 to 5 years.

Stout (1943), in his detailed and authoritative article, pointed out that,

"In spite of the fact that blood vessels are ubiquitous, and benign vascular tumors exceedingly common, malignant tumors of blood vessels are exceedingly rare. Just how many of them have been recorded is impossible to say, because many tumors have been reported as such with insufficient or obviously erroneous data. After reading reports of 118 cases labeled with some name suggestive of a malignant vascular tumor, the writer felt compelled to reject 41, or 35% of them, either because there was an inadequate or no histologic report or because, in his opinion, the illustrations and text described a tumor of some other type. ... Usually such tumors, even if they appear successively and grow to a large size, are not

suspected of being malignant growths. ... There are two features of the growth of hemangioendothelioma which are striking and uniformly present in all true tumors of this kind, although they are sometimes masked and require differential staining clearly to demonstrate them. These are: first, the formation of atypical endothelial cells in greater numbers that are required to line the vessels with a simple endothelial membrane; and, second, the formation of vascular tubes with a delicate framework of reticulin fibers and a marked tendency for their lumens to anastomose. No tumor should be considered a hemangioendothelioma unless these criteria are both present. The variations found are due to the marked variability in the number, shape, size, and tinctorial peculiarities of the malignant endothelia. They may be irregularly rounded or polygonal, or they may become heaped up and more or less fill the lumen. Usually, they remain within the vascular tubes but occasionally they grow outside of it and form solid sheets of cells."

Stout, later in his article, described the use of the silver reticulum stain in differentiation of hemangioendothelioma from other malignant tumors. The silver reticulum stain will

"cause the tubes to stand out in sharp relief, because each one, even the most malignant tumor, has a delicate fibrous supporting framework and the silver brings out a distinctive pattern. This pattern will be revealed with silver even when, with other stains, it is entirely obscured by an overgrowth of cells. When ordinary stains make it impossible to decide whether cells are grouped inside or outside of the vessel wall, and so to determine whether one is dealing with endothelia or pericytes, silver staining of the vascular reticulin sheath shows the exact relationship of the cells beyond peradventure."

He also discredited the entity "benign metastasizing hemangioma" and "doubts its existence".

Kinkade (1949) published a comprehensive review of "angiosarcoma". It dealt mainly with the literature after Freilich and Coe's review of 1918 to 1934.

Hemangiosarcomas have been experimentally produced in laboratory animals by the use of chemical carcinogens. Rigdon (1952) applied 20-methylcholanthrene to the skin; Steiner (1942) implanted a pellet of it in the salivary gland; and White and Stewart (1942) added it to the diet. Furth and Furth (1938) injected 1:2-benzpyrene into the spleen. Andervont et al. (1942) injected O-aminoazotoluene into the axillary region.

Edwards et al. (1942) discovered a hemangiosarcoma in the liver of a C3H strain male mouse, which was transplantable to other mice. Metastasis occurred in 2 of the mice after the implantation of first generation tumor cells, but these metastases were restricted to the liver. The transplants resembled the original tumor in every respect.

A few case reports of hemangiosarcoma are in the veterinary literature (Kingman and Newsom, 1918; Lindsay and Gilmore, 1946; Pounden and Sprunger, 1947; Lieberman, 1955; Farrell and Farrell, 1960; Simpson, 1960; and Jäplid, 1961).

Weipers and Jarrett (1954) described a specific type of hemangioma which occurred in the scrotum of dogs. Although the tumor had the tendency to recur histologically there was no evidence of intrinsic malignancy.

The reported incidence of canine hemangiosarcoma varied with different studies. In all articles it was reported that the tumor occurred rarely. Mulligan (1961) reported the occurrence of hemangiosarcoma of the dog to be about 2.6% of all tumors submitted, and 6% of the malignant tumors. This compared with 8.8% of tumors submitted classified as hemangiopericytoma and 0.9% as benign hemangioma. He found a distinct sex difference in his study; 14 males to 6 females.

In metastatic hemangiosarcomas the lung was the most commonly affected organ, followed in frequency by the liver, regional lymph nodes, and heart. The most common sites of primary occurrence were the spleen and the subcutis.

Mulligan (1949) was the only author to describe breed predisposition. He pointed out that "the German Shepherd is definitely predisposed to neoplasms of endothelial cells".

In reviewing standard textbooks of pathology, both veterinary and human, the subject of hemangiosarcoma was treated as an insignificant topic in some, while others had exceedingly long discourses describing an individual tumor. Willis (1948) questioned the value of a separate classification for tumors of endothelial cells when he pointed out,

"Let us recall once again the intermutability of the various mesenchymal tissues, the close kinship of their tumors, and the fact that vascular tissue is, of all the mesenchymal tissues, the most ubiquitous and one of the most plastic. Then we will be prepared to regard true angiomas and angiosarcomas, not as fixed species, but merely as conspicuously vasoformative variants of the genus mesenchymoma."

Herbut (1955) pointed out that hemangiosarcomas in man were seen in all ages but there was a preponderance in childhood and youth. Both sexes were equally affected. Pack and Ariel (1958) noted that these tumors tended to occur in the subcutaneous tissues of the extremities. Robbins (1962) asserted that hemangiosarcomas were often observed in the skin, liver, spleen, lungs, and bones. Most authors stated that although the tumor characteristically produced endothelial-lined spaces containing blood, in many of the less differentiated tumors sheets of cells were laid down with no vascular characteristics.

Robbins described the use of a phosphotungstic acid - hematoxylin stain (PTAH) for the differential diagnosis of hemangiosarcoma from fibrosarcoma and leiomyosarcoma.

The radiographic characteristics of this tumor are unique and interesting. Golden (1959) in his textbook of radiology described the distribution of metastases of malignant tumors to the lungs. There were 3 types of metastatic neoplasm to lungs according to appearance. They were: (1) nodular, (2) lymphangitic, and (3) pneumonic. Sarcomas, including hemangiosarcomas, usually assumed the nodular form. The metastases were almost always multiple with sharp borders, uniform in density, usually round, and of varying size. They were usually more numerous in the lower portions of the chest. When numerous they were usually of uniform size.

Bradley and Coley (1960) pointed out that hemangiosarcomas involving bone were difficult to differentiate from osteosarcomas. Reactive bone was laid down adjacent to the tumor in layers and resembled osteomyelitis. Hemangiosarcomas had a tendency to have their primary occurrence in the shaft of a bone rather than at the epiphysis, and when they occurred as a primary bone tumor, they had a tendency to metastasize to other bones and to the lungs.

According to De Lorimier (1954), hemangiosarcomas of the bone were exceedingly rare - the rarest of primary bone tumors. They were characterized as bulky, cystic, or telangiectatic tumors and were usually single, although multiplicity could eventuate. The tumor grew rapidly, with progressive destruction and with early dissolution through the cortex but relatively late metastasis. He further stated that the basic radiographic manifestations were

(1) conspicuous rarefaction, (2) possibly slight expansion of the cortices, (3) probably dissolution through the cortices and extension to regional soft tissue, (4) little or no osteosclerotic reaction, and (5) moderate response to x-ray therapy with demineralization. Important in a differential diagnosis would be a giant-cell tumor or metastases of some other primary tumor such as an adenocarcinoma of the kidney. Lichtenstein (1952) suggested that hemangiosarcomas in bone were likely to be aggressive and so far advanced by the time they were recognized that a grave prognosis was indicated, despite radical surgical procedures. Ritvo (1955) pointed out that the tumor characteristically tended to extend through layers of new bone with the formation of parallel layers of bone above like an onion skin. This lesion resembled proliferative osteomyelitis. The fastest bone growth or reaction was at the edge of the tumor. Radiographically, the tumor usually appeared as a diffuse osteoporosis. As the tumor invaded soft tissues, the shadow was not as distinct as before. Ritvo furthermore stated, "it is the only malignant tumor which arises from the cortex of bone".

Lasser (1955) described an interesting technique for the demonstration of the vascularity of tumors in bone. He used a radio-opaque solution which was injected into the blood vessels that supplied the bone tumor. Radiographs were then taken. He found that the various kinds of tumors in bone have a characteristically different vascular network. The object of the study was to help in the diagnosis of primary bone cancers.

In veterinary medicine, Carlson (1961), in his textbook of radiology, described a situation similar to that seen in man. This was especially true with reference to this tumor when it metastasized to the lungs.

MATERIALS AND METHODS

Preparation of Material

All clinical tissues were fixed in 10% neutral formalin. Each clinical tissue carried a number from the Veterinary Clinics and a number from the Department of Pathology. The clinical numbers were arranged in chronologic order beginning with 1 and continuing to over 100,000 at the time of completion of this study. Numbers from the Department of Pathology were listed according to alphabetical letter followed by a series of digits in chronologic order. The letter "A" covered the fiscal year 1956-1957, the letter "B" covered the fiscal year 1957-1958, etc., continuing to the last year of this study, the letter "J", covering the fiscal year 1964-1965. The letter "I" was omitted so as not to be confused with a numeral.

After the tissues were submitted, each tissue was trimmed, embedded, and sectioned at 6 microns. Each section was stained with hematoxylin and eosin (H & E) (Armed Forces Institute of Pathology, 1960, page 30). Tissues referred by private practitioners were usually fixed in 10% neutral formalin.

Selected slides in this study were stained with Gomori's iron reaction stain (Armed Forces Institute of Pathology, 1960, page 151). A second special stain used was a modification of reticulum technique as outlined by Lillie (1954).

An outline for the staining technique is as follows:

1. Deparaffinize sections through 2 changes each of xylene, absolute alcohol, 95% alcohol, and distilled water.
 2. Oxidize for 20 minutes in 0.5% periodic acid and rinse in distilled water.
 3. Dry thoroughly in dryer (heat).
 4. Lay slides face up on glass rods over large pan and deposit on each about 1.5 to 2 cc. of diamine silver hydroxide.* Let stand 3 minutes and decant.
 5. Rinse quickly in distilled water.
 6. Reduce 2 minutes in 10% neutral formalin and wash 3 minutes in running water.
 7. Tone 2 minutes in 0.2% acid gold chloride (by visual examination) and rinse in tap water.
 8. Fix 2 minutes in 5% sodium thiosulfate (not more) and wash in tap water.
 9. Counterstain for 5 minutes with nuclear fast red.**
 10. Rinse in distilled water and dehydrate through ascending concentrations of alcohols.
 11. Two changes of xylene.
 12. Mount with permount.
-

*Diamine silver hydroxide solution: Place 1 cc. 28% ammonia water in a small flask. Add 10 cc. of 10% silver nitrate, the first 7 to 8 cc. fairly rapidly, the rest cautiously, shaking between each addition to clear the brown clouds of silver oxide until a faint, slightly brown permanent opalescence remains. Add an equal volume of distilled water (solution to be made approximately 10 minutes before staining). In preparation of this stain all glassware must be acid clean.

**Nuclear fast red solution: Dissolve 0.1 Gm. of nuclear fast red in 5% solution of aluminum sulfate with aid of heat. Cool, filter, and add a grain of thymol as a preservative.

Study Procedures

The case files of the Department of Pathology from 1956 to 1965 were reviewed. All case reports of hemangiosarcoma in the dog were retrieved, along with the corresponding slides. If a case originated from the university's small animal clinic, the clinical history was obtained. These histories were reviewed and epidemiologic information from each was recorded in table form, similar to Brodey (1960). Column headings included pathology number, clinic number, breed, sex, age, primary location, size of tumor, tentative clinical diagnosis, rate of development, and metastasis.

The H & E-stained slides were examined by the author and 2 pathologists of the Department of Pathology for review and confirmation of the original diagnosis. The microscopic characteristics of each tumor were catalogued according to cell morphology, size and shape of nuclei, size and tinctorial qualities of nucleoli, and tendency for cytoplasm to fuse. The mitotic figures were classified according to their frequency and types of abnormal forms. The neoplasms were graded for hyperchromatism, invasiveness, lack of organization, and type and size of vessel formation. The section was examined for necrosis, amount and type of stroma, invasion of pre-existing blood vessels or lymphatics, and miscellaneous characteristics, such as hemorrhage and inflammation.

The reticulum-stained sections were evaluated for the amount, type, and structure of the reticulum present. The sections stained for iron were evaluated for identification of iron pigment within macrophages which were present in some of the neoplasms.

Cases from the small animal clinic with radiographs were studied with regard to the characteristics of the tumor in or near bone and metastases to the lungs.

Gross photographs and photomicrographs were taken of selected lesions.

All measurements taken through the microscope were obtained by use of an ocular micrometer. The ocular micrometer was calibrated in a routine manner by use of a stage micrometer.

TERMINOLOGY

This tumor of endothelial cell origin has been given many different names, most of which are confusing to the student of pathology. For this reason, the following discussion on terminology is presented.

The following terms have been used in describing this tumor: angiosarcoma, hemangioendothelioma, angioendothelioma, endothelioma, hemendothelioma, angioblastoma, intracranial angioblastoma, angiofibrosarcoma, endothelial sarcoma of blood vessel, fibroangioendothelioma, hematoblastoma, hemangioblastoma, hemangiosarcoma, angioblastic neoplasm, angioblastic meningioma, malignant aneurysm, capillary angiosarcoma, capillary sarcoma, hemangioendothelialblastoma, hemangioendothelialsarcoma, angioblastic sarcoma, telangioendothelioma, telangiectatic sarcoma (American Cancer Society, 1953).

Moulton (1961) defined the tumor as "a malignant tumor of endothelial cells". The terminology is just as much a problem today as in the past, since most authors still disagree on the term that should be used. Anderson (1961) used the term "malignant hemangioendothelioma" or "angiosarcoma". Herbut (1955) used both "hemangioendothelioma" and "hemangiosarcoma". Boyd (1958) used the term "hemangioendothelioma", with the adjective "benign" or "malignant", depending upon the cell type. He pointed out that the tumor would more conveniently be called "hemangioendothelial sarcoma". Pack and Ariel (1958) used the term "angiosarcoma", which is a relatively older term. Robbins recently (1962) used the term "hemangioendothelioma" to "represent an

intergrade between the well differentiated hemangiomas and the frankly anaplastic, totally cellular hemangioendothelialsarcoma". He also stated that "certain writers use the term hemangioendothelioma to refer to a malignant endothelial tumor. This usage is confusing and inappropriate, since the malignant counterpart should properly be referred to as a sarcoma". On the other hand, "hemangioendothelioma" seems to be a popular term in the veterinary literature (Moulton, 1961).

In this study the term "hemangiosarcoma" was selected for the malignant neoplasm for the following reasons:

1. The term hemangioendothelioma was most inadequate and inappropriate for the reasons stated by Robbins (1962).
2. The term hemangiosarcoma has been recommended by the National Cancer Institute (Public Health Service, U. S. Department of Health, Education and Welfare, 1964).
3. The word hemangiosarcoma connotes both malignancy and histogenesis.

Hopefully, progress has been made in limiting the terminology that is used in referring to this tumor. In a recent publication (American Cancer Society, 1965), only 4 terms were recognized for this neoplasm: hemangiosarcoma, angiosarcoma, malignant hemangioendothelioma, and hemangioendothelialsarcoma.

RESULTS

Occurrence and Gross Characteristics

TABLE 1 is a compilation of the salient features of each case as outlined in Materials and Methods.

Breed incidence. The breeds which had the highest incidence of hemangiosarcomas were Cocker Spaniel (14), Boxer (14), German Shepherd Dog (6), and Labrador Retriever (4). The tumor was diagnosed in 22 other breeds. The number of hemangiosarcomas based on the weight of the dog is shown (Figure 1). The method for categorizing the dogs by weight was that used by Tjalma (1965) in his study of bone cancer. The largest number of hemangiosarcomas occurred in the large breeds of dogs.

Sex distribution. In those animals of known sex, a total of 44 hemangiosarcomas was found in the male and 25 in the female, or a ratio of 1.76 males to 1 female. The influencing factor for the sex difference was the great preponderance of the tumor in male dogs of the large breeds (Figure 1). Excluding this group, the ratio of males to females was approximately equal.

Age distribution. The number of hemangiosarcomas according to age is presented (Figure 2). The mean age was 9.0 years. The mean age for males was 8.5 years, while the mean age for females was 10.7 years.

TABLE 1. Occurrence and gross characteristics of 76 canine hemangiosarcomas.

Path. No.	Clinic No.	Breed	Sex	Age (yrs.)
A325	---	---	---	---
A522	---	---	---	---
A1503	---	Doberman Pinscher	M	---
A1983	7163	Cocker Spaniel	M	6
B2412	---	---	M	4
B3408	32424	Cocker Spaniel	M	13
B4928	32908	Weimaraner	F	4
B6134	33282	English Setter	M	8
C376	---	Cocker Spaniel	M	8
C720	---	Cocker Spaniel	F	12
C858	33849	Boxer	M	8
C3343	---	Mixed	F	17½
C5391	---	Boston Terrier	M	8
C5497	7163	Cocker Spaniel	M	8½
C5550	30359	Boxer	M	5
C5838	36238	Pointer	F	3½
C5887	---	Labrador Retriever	F	6
D683	---	Boxer	M	9

Primary Location	Size (cm.)	Rate of Development	Metastasis
mouth	---	---	---
tongue	1.1 x .7 x .5	---	---
3rd premolar area	---	---	---
l. ear	---	---	none
free in abdomen	---	---	numerous masses in abd. cavity
mouth	5 dia.	rapid-3 wk.	mandibular l.n.
femur	from acetabulum to mid shaft	6 wk.	regional l.n.
r. prox. foreleg	---	---	lungs
l. scapular area	7 dia.	---	---
l. cheek vent. to ear	6 x 3 x 2	several mo.	none
prox. r. femur	large	---	lysis of bone
liver (caudate lobe)	6 x 9	---	none
muscle between scapulae	---	2 mo.	---
spleen	---	---	numerous nodules in lung, l.n.
sheath of penis	---	6 mo.-1 yr.	inguinal l.n. pre- viously removed
l. femur and adj. area	1 encircled femoral neck; 2 others, 2 dia.	4 wk.	---
upper jaw	4.5 length	rapid-2 mo.	lysis of bone
l. front leg	---	---	cervical l.n., lung, liver, kidney, heart

TABLE 1--continued

Path. No.	Clinic No.	Breed	Sex	Age (yrs.)
D1250	---	Beagle Hound	F	11
D1349	5820	Dachshund	F	12
D3651	38357	Cocker Spaniel	M	10
D3752	---	Cocker Spaniel	M	12
D4327	---	Boxer	M	11
D4615	38631	Boxer	M	7
D5863	37853	Setter	M	12
E1019	---	Fox Terrier	F	14
E1628	38310	Kerry Blue Terrier	M	10
E2374	---	Boxer	M	6
E3701	---	Cocker Spaniel	---	8
E3900	---	German Shorthaired Pointer	F	10
E4161	757	Cocker Spaniel	F	8
E4186	37149	German Shepherd Dog	M	7
E4227	---	Dachshund	M	9½
E4792	---	Boxer	F	11
E5197	41651	German Shepherd Dog	M	11
E5432	40841	Springer Spaniel	M	9

Primary Location	Size (cm.)	Rate of Development	Metastasis
mesentery	2 nodules-6x9 dia.	rapid	liver, spleen
spleen	5 dia.	---	liver
spleen	---	2 wk.	none
liver	7 x 12	unknown	lungs, kidney, intestines
---	---	---	small intestine, lung
spleen	---	---	mesentery, lung, liver
spleen	11 x 16	---	none
prox. 1/3 radius	---	---	---
---	---	---	numerous l.n., lung
---	---	---	numerous nodules, lungs
---	---	---	---
S.C., r. 11-13th rib	10 dia.	rapid-2 mo.	---
mammary gland	4 dia.	---	---
elbow	5 x 7	1 yr.	---
mesentery	---	---	kidney, liver, mesentery, lung
S.C., scrotum	1 dia.	rapid	none
liver, spleen	---	slow	none
lg. mass involving kidney, spleen, omentum, liver	---	---	see primary location

TABLE 1--continued

Path. No.	Clinic No.	Breed	Sex	Age (yrs.)
E5763	41222	German Shepherd Dog	M	11
E5907	---	Cocker Spaniel	M	9
F247	---	Terrier	F	13
F814	---	Terrier	F	11
F890	---	English Setter	M	5
F1494	42828	Dalmatian Coach Dog	M	12
F2644	---	Golden Retriever	M	7
F3788	43982	Boxer	M	5
F3970	---	Boxer	F	9
F5137	---	Scottish Terrier	F	13
F6173	---	German Shepherd Dog	M	8
G107	---	Labrador Retriever	---	---
G294	---	Labrador Retriever	M	13
G1013	---	Pointer	F	14
G3833	39715	Boxer	---	4
G4150	---	Poodle	F(S)	11
G4373	---	Dachshund	M	4
G4446	---	German Shepherd Dog	M	10
G4752	---	Cocker Spaniel	F	10
G4792	---	Boxer	---	11
G5327	47472	Cocker Spaniel	M	5

Primary Location	Size (cm.)	Rate of Development	Metastasis
S.C. of l. flank	3 dia.	slow-4 mo.	none
spleen	3 dia.	rapid	none
S.C., inguinal area	2.2 dia.	---	spleen, lung, heart, jejunum, kidney
base of tail	---	*	---
skin	---	---	---
S.C., r. side post. last rib	---	---	none
r. kidney	5 dia.	---	none
S.C., r. side of chest	---	---	---
r. nares	---	5 mo.	none
lung	small, numerous	---	none
pancreas, duodenum	---	---	---
post. antebrachium	4.5 dia.	5 mo.	none
liver, spleen	variable	2 mo.	mandibular, mediasti- nal l.n.
various	1 x 4 dia.	3-6 mo.	liver, spleen, l.n., heart, lung, kidney
spleen	10 dia.	rapid	none
ovary	4 x 3 x 2	---	none
spleen	18 dia.	sudden	none
S.C., cervical	1 x 3	rapid	lung, kidney, cere- brum, cerebellum
spleen	4.5 dia.	---	none
ovary	---	---	spleen, omentum
r. oral commissure	2 dia.	---	none

TABLE 1--continued

Path. No.	Clinic No.	Breed	Sex	Age (yrs.)
G6848	---	Pointer	M	9
H472	48205	Poodle	M	7
H1075	48483	Standard Poodle	F	7½
H1202	---	German Shepherd Dog	M	8
H2156	---	Boxer	M	10
H2796	---	Beagle Hound	F	---
H4957	---	Boxer	M	11
H5628	---	Boxer	M	11
H5657	---	Bassett Hound	F(S)	9
H6057	---	Cocker Spaniel	F	13
H6119 H6360	101469	Vizsla	M	5
J1766	102835	Golden Retriever	M	7½
J2611	---	Wirehair Terrier	F	6
J2652	---	Miniature Poodle	M	8
J2691	103252	Labrador Retriever	F	7
J2814	---	---	---	12
J2958	103080	Cocker Spaniel	F	13
J3526	---	---	M	10
J4075	103820	Airedale Terrier	M	10½

Primary Location	Size (cm.)	Rate of Development	Metastasis
---	---	---	lung, omentum
S.C., ant. to penis	0.2 x 0.6	slow	none
spleen	2 x 3 dia.	---	liver, lung
l. hind leg muscle	---	---	heart, lung, kidney, mesentery
S.C., prepuce	---	1 mo.	---
spleen, liver	---	2-3 wk.	spleen, liver
bladder	---	1-2 mo.	lung
r. femur	---	---	liver, kidney, lung
l. hock(soft tis.)	---	---	---
3rd phalanx	---	---	---
S.C., vent. abd.	---	slow	numerous l.n., mesen- tery, peritoneum, diaphragm, stomach, muscle of abd. wall
lung	none	---	heart, lung
spleen	---	---	---
---	---	---	lung, heart
diaphragm	7 dia.	---	pleura, ileum, peri- cardium
S.C., inguinal area	5 dia.	3 mo.	---
---	---	---	lung, spleen
intestine	---	---	---
S.C., l. metacarpus	---	---	prescapular l.n.

TABLE 1--continued

Path No.

A - July 1, 1956, to June 30, 1957	F - July 1, 1961, to June 30, 1962
B - July 1, 1957, to June 30, 1958	G - July 1, 1962, to June 30, 1963
C - July 1, 1958, to June 30, 1959	H - July 1, 1963, to June 30, 1964
D - July 1, 1959, to June 30, 1960	J - July 1, 1964, to June 30, 1965
E - July 1, 1960, to June 30, 1961	

Note: Cases A1983 and C5497 are from the same animal but were counted as 2 cases since 2½ years had elapsed between occurrences.
Cases H6119 and H6360 are from the same animal and are counted as 1 case because of the short period of time between occurrences.

Sex

M - male

F - female

F(S) - female, spayed

Primary Location

abd. abdomen
r. right
prox. proximal
l. left

vent. ventral
adj. adjacent
S.C. subcutis
post. posterior

ant. anterior
lg. large
l.n. lymph node(s)
tis. tissue

Size

dia. diameter

Rate of Development

* Present for several months, increasing rapidly last few days.

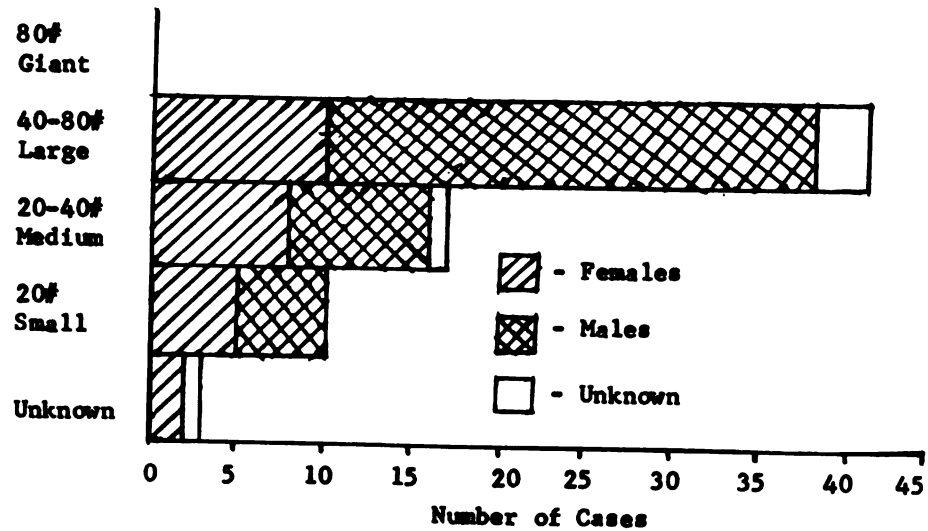


Figure 1. Incidence of hemangiosarcomas in male and female dogs of different weight classes.

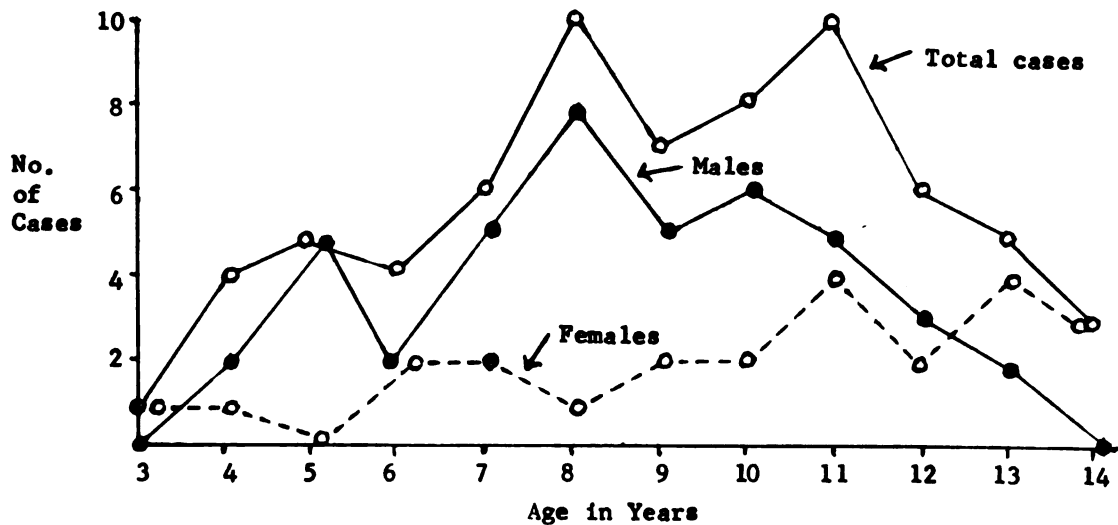


Figure 2. Age and sex distribution of hemangiosarcomas in dogs.

Primary location. The most common primary sites were the skin, including the subcutis (24), and the spleen (15), followed by bone (6), and liver (5). The primary location was also classified by the area of body involved. The most commonly affected area was the trunk (39) followed by the extremities (16) and head and neck (10).

Metastases. Fifty-four histories had information on metastases. Thirty-two (59%) had metastasized to various organs. The lungs were the most common site (19), followed in frequency by distant lymph nodes (11), liver (9), kidney (8), mesentery (7), spleen (6), heart (6), and regional lymph nodes (5). Typical numerous metastases are shown (Figures 3 and 4).

Rate of development. The rate of development was obtained from the history, given by the owner or the veterinarian. The rate was divided into 3 categories: (1) 0 to 1 month, (2) 2 to 5 months, and (3) 6 months and over. Six neoplasms were placed in the 0- to 1-month, 11 in the 2- to 5-month, and 3 in the 6 months and over category. Growth rate of 3 tumors was reported to be slow and 6 rapid.

Size and shape of tumor. Tumor size varied from a few millimeters to several centimeters. It was often round, bluish-black, and poorly encapsulated. The metastatic lesions were usually round and less hemorrhagic than the primary lesion. The typical gross appearance of a primary lesion on cut surface is shown (Figure 5). A single, circular metastatic lesion which has ruptured resulting in an adjacent clot is illustrated (Figure 6).

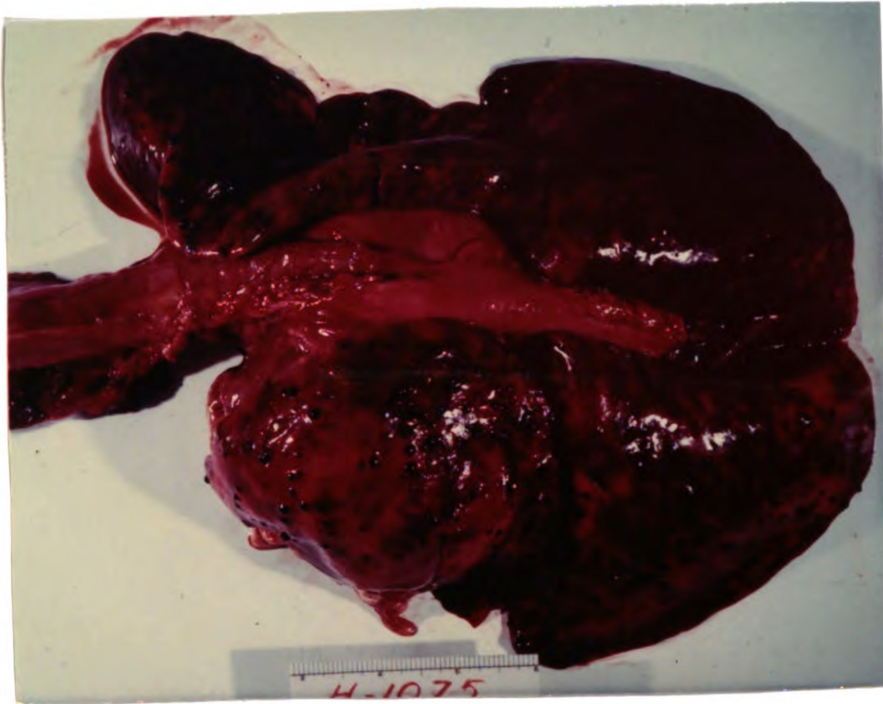


Figure 3. Metastatic hemangiosarcoma in the lungs.

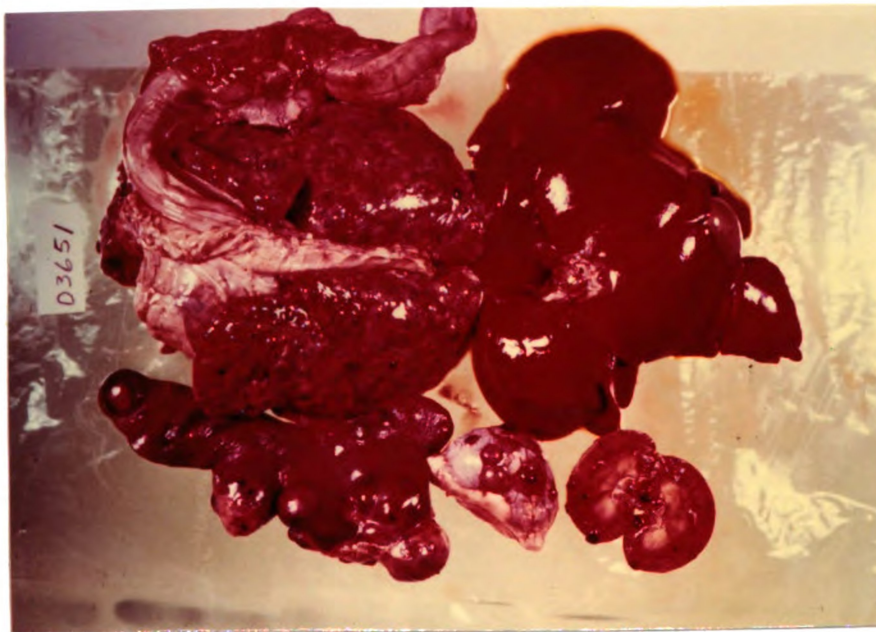


Figure 4. Metastatic hemangiosarcoma in lung, spleen, kidney, and liver.

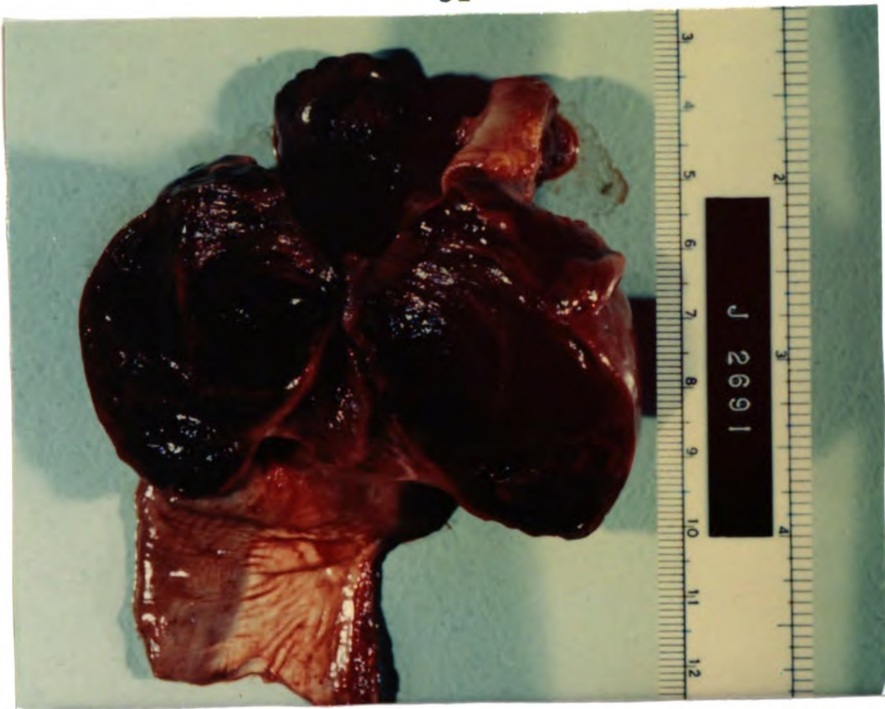


Figure 5. Cut surface of hemangiosarcoma in diaphragm (primary site).

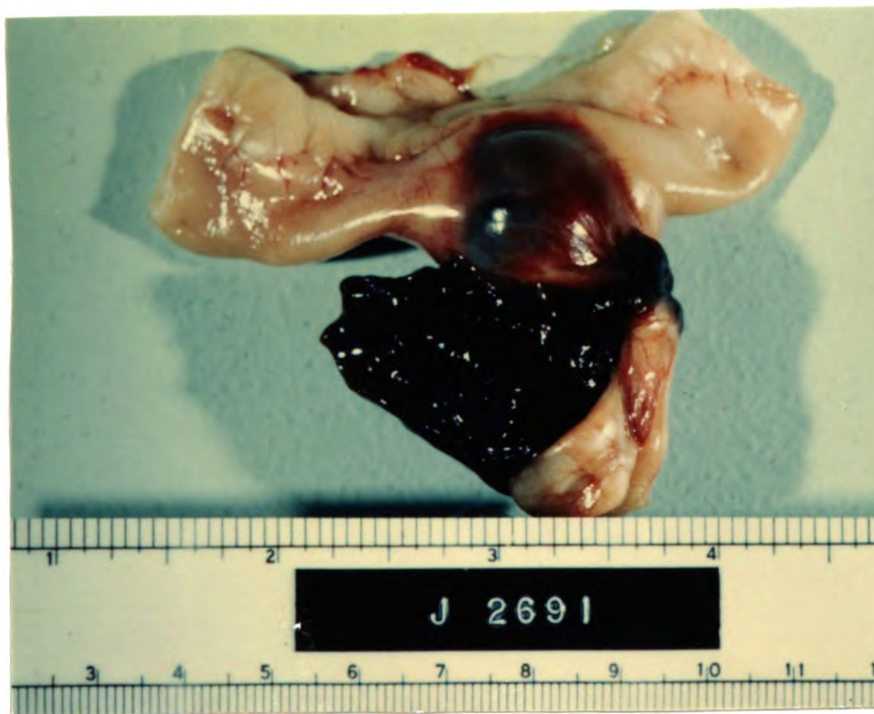


Figure 6. Metastatic hemangiosarcoma in small intestine with adjacent blood clot.

Recurrence. In 11 cases where information was available, 7 were known to recur. The diagnosis was made from necropsy specimens in 32 cases and biopsy specimens in 26 cases.

Tentative diagnosis. Although not recorded in TABLE 1, the tentative diagnosis by the clinician was noted in each case where this information was given. Of the 21 cases in which this information was found, only 6 were diagnosed as hemangiosarcoma.

Microscopic Characteristics

Tissue sections of hemangiosarcomas from 76 dogs were examined microscopically. The tumor characteristics are presented (TABLE 2).

Shape of the cell. The cells generally were spindle shaped except that in the more anaplastic tumors the cells were pleomorphic (Figures 7 and 8).

Size and shape of nuclei. The shape was elongate, oval, round, or pleomorphic. In general, the round or pleomorphic nuclei were larger, more vesicular, and more anaplastic (Figure 9). The size was variable. The elongate nuclei ranged from 3 x 12 microns to 6 x 16 microns. The oval nuclei, which were by far the most common type seen, ranged from 6 x 8 microns to 12 x 18 microns, while the round forms were 7 to 18 microns in diameter. Size measurements were usually not determined on nuclei which were pleomorphic.

Size and staining properties of nucleoli. There was a great variation in the size of the nucleoli, from those which could barely be seen to those as large as 7 to 9 microns in diameter. The larger forms were usually seen in the large oval and round nuclei (Figure 10). They were invariably round, and there was a correlation between the size and the staining characteristics. The smaller nucleoli (0.5 to

TABLE 2. Microscopic characteristics of 76 canine hemangiosarcomas.

Path. No.	Shape of Cell	Size & Shape of Nuclei (μ)	Size & Color of Nucleoli (μ)	Mitoses/hpf	Types of Mitoses	Hyperchromatism
A325	spindle	oval (9x13) to round (20 dia.)	7-9 dia. single & multiple eosinophilic	0-1	bizarre	+
A522	spindle	oval (7x10)	0.5-1 basophilic	0	---	++
A1503	pleomorphic to spindle	round (11) to oval (10x12)	4-5 multiple eosinophilic	2	bizarre	++
A1983	spindle	oval (9x12)	1-2 single eosinophilic	0-1	normal	++
B2412	elongate to spindle	oval (9x12)	0.5-1 single basophilic	0-1	blast	++
B3408	spindle	oval (7x13)	1 single not many	0	---	++
B4928	pleomorphic to spindle	elongate (6x16) to round (15)	1 few; most not seen	2-3	blast	++
B6134	spindle	oval (8x15)	most not seen; 1 a few up to 3		blast	++
C376	elongate to spindle to pleomorphic	elongate to oval (4x15) to round (12)	2-3 single eosinophilic	1	blast & bizarre	++
C720	spindle	oval (6x8)	only a few observed .5-1 single basophilic	0	---	++
C858	pleomorphic	round (8)	none observed	0	---	++

Degree of Anaplasia		Necrosis	Amount of Stroma	Invasion of Pre-existing Blood or Lymph Vessels	Miscellaneous		
Cellular	Vascular				Infl.	Hem.	Macro.
+++	++	in eroded area	+ *	---	+	+	0
++	+++	in eroded area	++	no	+	+	0
++	+++	in eroded area	+	---	+	+	0
+	++	in eroded area	++	no	+	+	0
	+++	in areas of hemor-rhage	+ *very little	---	0	+	0
	+++	in eroded area	+	---	+	+	0
+++	+++	in areas of hemor-rhage	+++	no	0	+	0
	+++	none	++ *	yes	0	0	0
+++	+++	in areas of hemor-rhage	++ *	yes	0	+	0
	+++	none	+ *	---	0	+	0
+++		none	+	no	0	+	0

TABLE 2--continued

Path. No.	Shape of Cell	Size & Shape of Nuclei (μ)	Size & Color of Nucleoli (μ)	Mitoses/hpf	Types of Mitoses	Hyperchromatism
C3343	pleomorphic	round (9to15)	3-4 single eosinophilic	1	blast & bizarre	++
C5391	spindle to pleomorphic	oval	1-3 single & multiple eosinophilic	0-1	blast	++
C5497	very pleomorphic	round (15)	2-3 single - a few multiple eosinophilic	3-4	blast	++
C5550	spindle	oval (7x11)	1-2 single & multiple	2-3	blast	++
C5838	spindle	elongate to oval (5x14)	1 single basophilic	2-3	normal	+++
C5887	pleomorphic	oval (10x14) a few tumor giant cells	2 single & multiple eosinophilic	2	normal & blast	+++
D683	spindle	oval (6x12)	none observed	0	---	++
D1250	pleomorphic	elongate to oval (4x12) to round (10)	.5-1 basophilic	3	blast	++
D1349	spindle	pleomorphic to round (9-10)	4 single eosinophilic	2-3	blast	+++
D3651	spindle	oval (9x12) to round (10)	2-3 eosinophilic	5	normal & blast	++

Degree of Anaplasia		Necrosis	Amount of Stroma	Invasion of Pre-existing Blood or Lymph Vessels	Miscellaneous		
Cellular	Vascular				Infl.	Hem.	Macro.
++	+++	in areas of hemorrhage	++ *poor stain	---	0	+	0
+++		in areas of hemorrhage	++	---	0	+	0
+++		in center of cellular areas	+ *very little in cellular areas	yes	0	+	0
	+++	in eroded areas	++ *very scanty	---	+	0	0
+++	++	none	++ *	---	+	+	+
	+++	none	+	---	+	0	0
	+++	in areas of hemorrhage	+	---	0	+	0
	+++	in areas of hemorrhage	+++ *	yes	+	+	0
+++	+++	in center of cellular area	+	---	+	0	0
	+++	in areas of hemorrhage	++	yes	0	+	0

TABLE 2--continued

Path. No.	Shape of Cell	Size & Shape of Nuclei (μ)	Size & Color of Nucleoli (μ)	Mitoses/ hpf	Types of Mi- toses	Hyper- chroma- tism
D3752	spindle	oval (6x8) to round (12-14)	2-3 eosinophilic	1	normal	++
D4327	pleomorphic to spindle	round (12)	.5-1 single basophilic	1-2	normal	---
D4615	spindle	elongate (4x12)	.5-1 single basophilic	1	normal & blast	++
D5863	elongate to spindle	elongate to oval to round	2-3 single basophilic & eosinophilic	2-3	blast & bizarre	+++
E1019	spindle	oval (7x13)	.5-1 single a few large (2-3)	1	normal & blast	++
E1628	spindle to pleomorphic	oval (7x12)	1-2 single eosinophilic	1	blast	+
E2374	elongate to spindle	oval some round	most not seen; 1 few .5-1 basophilic	1	normal	++
E3701	spindle	oval (8x17) to round	2-3 single eosinophilic	1	blast	+++
E3900	spindle	round (8-10)	1-2 single eosinophilic	2-3	normal & blast	++
E4161	spindle to pleomorphic	oval (8x10) to pleomorphic	1 single & multiple	5-10	blast & bizarre	+++

Degree of Anaplasia		Necrosis	Amount of Stroma	Invasion of Pre-existing Blood or Lymph Vessels	Miscellaneous		
Cellular	Vascular				Infl.	Hem.	Macro.
+++	+++	in areas of hemorrhage	++	yes	0	+	0
+++		in areas of hemorrhage	+ *	yes	0	+	0
	+++	none	+++ *	yes	0	0	0
	+++	in areas of hemorrhage	+	no	0	+	0
	+++	none	+++	no	0	0	0
++	+++	none	+ *	---	0	+	0
	+++	in areas of hemorrhage	++	no	+	+	+
	+++	in areas of hemorrhage	+	---	+	+	0
	+++	none	++	---	0	0	0
+++	++	in areas of hemorrhage	+	yes	+	+	0

TABLE 2--continued

Path. No.	Shape of Cell	Size & Shape of Nuclei (μ)	Size & Color of Nucleoli (μ)	Mitoses/ hpf	Types of Mi- toses	Hyper- chroma- tism
E4186	pleomorphic to spindle	1, oval (12x18) to round (11)	2 single & multiple	5	normal & bizarre	+++
E4227	pleomorphic	round to oval (7x13)	1 single - a few multiple	3-4	normal	+++
E4792	pleomorphic to spindle	oval (9x11) to round	.5-1 single - a few mul- tiple	1	normal	+++
E5197	spindle	oval (8x15) a few round (18)	most not seen - a few .5-1	1	normal & blast	+++
E5432	pleomorphic to spindle	oval (11x18) to round (16)	3-5 single & some mul- tiple eosinophilic	5-10	bizarre	+++
E5763	very pleo- morphic	oval (8x15) to round (18)	1-3 eosinophilic	2-3	normal & blast	+++
E5907	pleomorphic	oval to round to pleomorphic	2-3	3	bizarre & blast	+++
F249	pleomorphic	pleomorphic	small (1)	1	normal	+++
F814	spindle to pleomorphic	round (15- 18) to pleomorphic	2 single & multiple	1	---	++
F890	spindle to pleomorphic	oval to round	3-4 single eosinophilic	1-2	normal	++

Degree of Anaplasia		Necrosis	Amount of Stroma	Invasion of Pre-existing Blood or Lymph Vessels	Miscellaneous		
Cellular	Vascular				Infl.	Hem.	Macro.
+++	+++	none	++	no	+	+	+
++	+++	in areas of hemorrhage	++	no	+	+	+
++	+++	none	+	no	0	+	+
very little tissue		none	+	no	+	+	+
+++	++	in areas of hemorrhage	++	yes	+	+	0
+++	++	none	++	yes	0	+	0
+++	++	none	+	no	0	+	0
+++	+++	in areas of hemorrhage	+	no	+	+	+
++	+++	in areas of hemorrhage	+	yes	0	+	+
+++	++	none	+	yes	0	0	0

TABLE 2--continued

Path. No.	Shape of Cell	Size & Shape of Nuclei (μ)	Size & Color of Nucleoli (μ)	Mitoses/ hpf	Types of Mi- toses	Hyper- chroma- tism
F1494	spindle to pleomorphic	oval (9x11) to round	1 single	2-3	blast	++
F2644	pleomorphic	round (13-15)	5 single eosinophilic	3	blast	+++
F3788	spindle	oval (9x11) to round	none observed	3-4	normal & blast	++
F3970	spindle	oval (8x10) to round (12)	1 single basophilic	2	normal & blast	++
F5137	pleomorphic	irregular round to oval (15x18)	3-4 single eosinophilic	5-6	bizarre	+++
F6173	spindle	oval to elongate (3x12)	none	0	---	+++
G107	pleomorphic	round (13) to oval (9x16)	3 single eosinophilic	0	---	+++
G294	pleomorphic	round (15)	3-4 single & multiple eosinophilic	7-8	bizarre	+++
G1013	pleomorphic	oval (10x14) to round (13)	3 single eosinophilic	1-2	normal	+++
G3833	pleomorphic	oval (8x10)	1 single basophilic	0	---	++
G4150	spindle to pleomorphic	round (11) to oval (11x15)	3 single & multiple basophilic	1-2	normal & blast	+

Degree of Anaplasia		Necrosis	Amount of Stroma	Invasion of Pre-existing Blood or Lymph Vessels	Miscellaneous		
Cellular	Vascular				Infl.	Hem.	Macro.
	+++	in areas of hemorrhage	+	yes	+	+	0
	+++	none	+	yes	0	+	0
++	+++	none	+	yes	0	+	0
+++	++	none	+	no	0	0	0
+++	++	in areas of hemorrhage	+	yes	+	+	0
+++	++	in areas of hemorrhage	+	yes	0	+	+
+++		in areas of hemorrhage	+	no	+	+	+
	+++	in areas of hemorrhage	+	no	+	+	+
	+++	none	+	no	+	0	0
+++		none	+++	yes	+	+	0
	+++	in areas of hemorrhage	+	no	0	+	+

*very little observed

TABLE 2--continued

Path. No.	Shape of Cell	Size & Shape of Nuclei (μ)	Size & Color of Nucleoli (μ)	Mitoses/ hpf	Types of Mi- toses	Hyper- chroma- tism
G4373	spindle to pleomorphic	oval (7x12)	1 basophilic	1	---	++
G4446	spindle to pleomorphic	round (7) to oval (6x11)	1-2 eosinophilic	1-2	normal & blast	+++
G752	spindle	round to oval	2-3 single & multiple eosinophilic	1-2	normal & blast	++
G4792	pleomorphic to round	oval (6x15) some large bizarre	1 eosinophilic	0	---	+++
G5327	spindle	pleomorphic	3-4 single eosinophilic	4-5	bizarre	+++
G6848	spindle to pleomorphic	oval (7x11)	1-2 single & a few multiple eosinophilic	0	---	+
H472	spindle	pleomorphic (9x11)	1-2 basophilic	0	---	++
H1075	spindle to pleomorphic	pleomorphic (10x12), a few round (15)	1 single & multiple basophilic	1-2	blast	+++
H1202	spindle	oval (9x12) to round (11)	1-4 single & multiple basophilic & eosinophilic	1-2	blast	++
H2156	spindle	oval (8x13)	most not seen	2-3	normal	++

Degree of Anaplasia		Necrosis	Amount of Stroma	Invasion of Pre-existing Blood or Lymph Vessels	Miscellaneous		
Cellular	Vascular				Infl.	Hem.	Macro.
+++	++	none	+	no	0	0	0
	+++	in center of tumor	++ *	no	0	+	0
+++	+++	in areas of hemorrhage	+	no	+	+	0
++	+++	in areas of hemorrhage	+	no	+	+	0
+++	+++	in eroded areas	+	yes	+	+	0
+++		in areas of tumor	+	no	+	+	0
very little tissue		necrosis at surface	++	no	+	0	0
+++	++	in areas of hemorrhage	+	yes	0	+	0
	+++	in areas of hemorrhage	+	no	0	+	+
	+++	in eroded areas	++ *atypical	no	+	+	0

TABLE 2--continued

Path. No.	Shape of Cell	Size & Shape of Nuclei (μ)	Size & Color of Nucleoli (μ)	Mitoses/hpf	Types of Mitoses	Hyperchromatism
H2796	spindle	oval (9x15)	2 single & multiple eosinophilic	0	---	+
H4957	pleomorphic	oval (9x14) to round (12)	2-3 single & multiple eosinophilic	1	blast	+
H5628	spindle	elongate (3x12) to oval (6x18)	1 single basophilic	3-4	bizarre	+++
H5657	pleomorphic	oval (10x16)	2 single & multiple eosinophilic	3	normal & blast	++
H6057	spindle	pleomorphic to round (10)	1-2 single eosinophilic	4-5	normal & blast	+
H6119 H6360	spindle	elongate to oval (8x10)	1 basophilic	0	---	++
J1766	spindle	oval (6x14) to round (15)	1-3 basophilic & eosinophilic	5	normal & bizarre	++
J2611	spindle	round (6) to oval (6-8)	1-2 single & multiple basophilic	0	---	+++
J2652	spindle	oval (7x12)	1 basophilic	1-2	normal & bizarre	++
J2691	pleomorphic	pleomorphic	1-2 basophilic	1-2	normal blast bizarre	++

Degree of Anaplasia		Necrosis	Amount of Stroma	Invasion of Pre-existing Blood or Lymph Vessels	Miscellaneous		
Cellular	Vascular				Infl.	Hem.	Macro.
	+++	in areas of hemorrhage	+	yes	+	+	0
+++		in areas of hemorrhage	+++ *	no	0	+	+
+++	++	in center of cellular areas	++	no	+	+	+
+++		none	+	no	+	+	0
+++	++	in areas of hemorrhage	+	no	0	+	0
++	+++	in center of cellular areas	++ *	yes	+	+	+
+++	+++	in center of cellular areas	++	yes	+	+	+
	+++	none	+	---	0	0	+
	+++	in areas of tumor	++	yes	0	+	+
+++	+++	in areas of hemorrhage	+	yes	+	+	+

TABLE 2--continued

Path. No.	Shape of Cell	Size & Shape of Nuclei (μ)	Size & Color of Nucleoli (μ)	Mitoses/ hpf	Types of Mi- toses	Hyper- chroma- tism
J2814	pleomorphic	oval (10x14)	1-3 single eosinophilic	1-2	---	++
J2958	spindle	round (9-10) to oval (8x11)	1-2 multiple eosinophilic	2-3	normal	+++
J3526	spindle to pleomorphic	oval (10x14) to round (13)	1-4 multiple basophilic & eosinophilic	4-5	blast & bizarre	++
J4075	spindle	pleomorphic (10)	most not seen	0	---	+++

*Fine meshlike network of reticulin as determined with a special stain.

+ - scanty
 ++ - moderate
 +++ - marked
 Infl. - inflammation
 Hem. - hemorrhage
 Macro. - pigment-laden macrophages
 dia. - diameter

Degree of Anaplasia		Necrosis	Amount of Stroma	Invasion of Pre-existing Blood or Lymph Vessels	Miscellaneous		
Cellular	Vascular				Infl.	Hem.	Macro.
+++	+++	in areas of tumor	++	no	0	+	0
++	+++	in areas of hemor- rhage	+	yes	0	0	0
+++		in center of cellu- lar areas	++	yes	+	+	+
	+++	none	++	yes	0	+	+

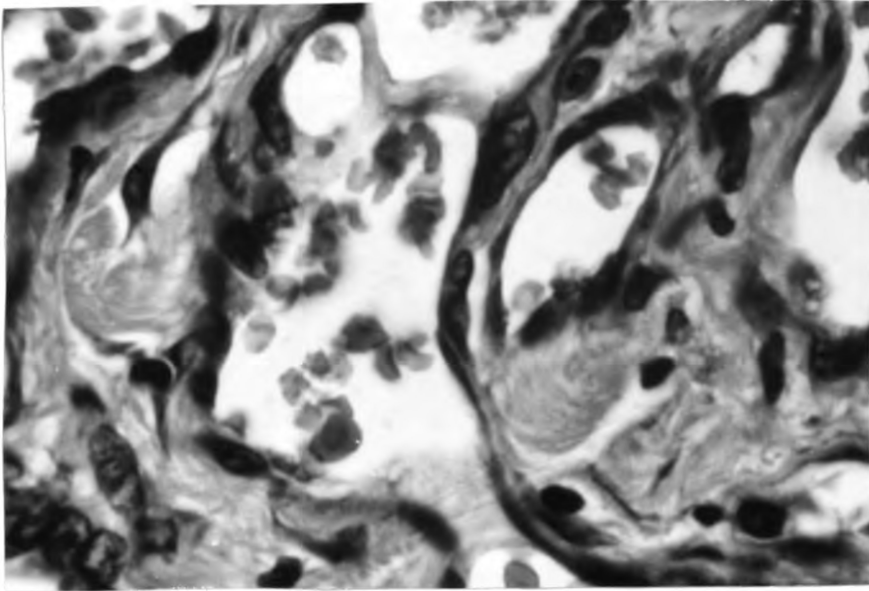


Figure 7. Numerous spindle-shaped cells lining vascular spaces of a hemangiosarcoma. Hematoxylin and eosin, x 750.

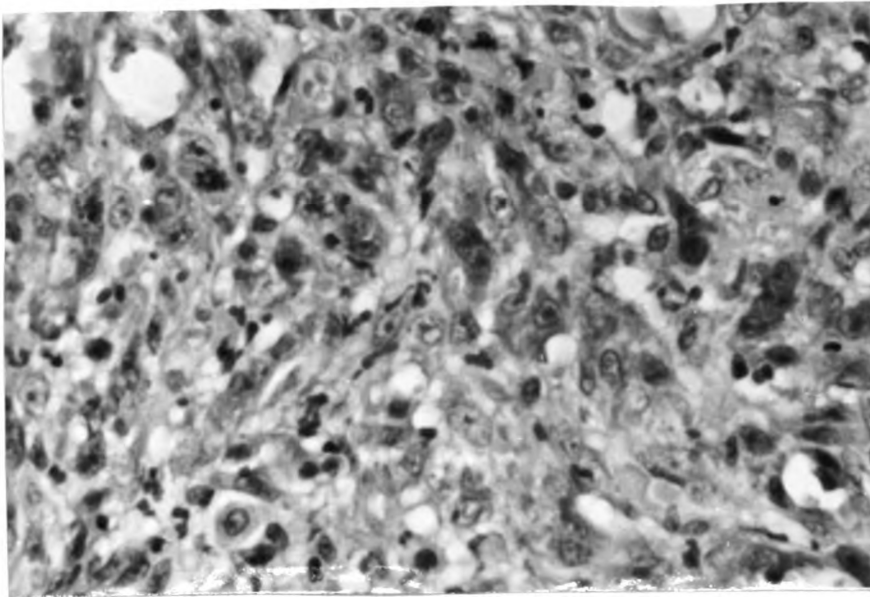


Figure 8. Cellular type of hemangiosarcoma with little vessel formation. Hematoxylin and eosin, x 468.

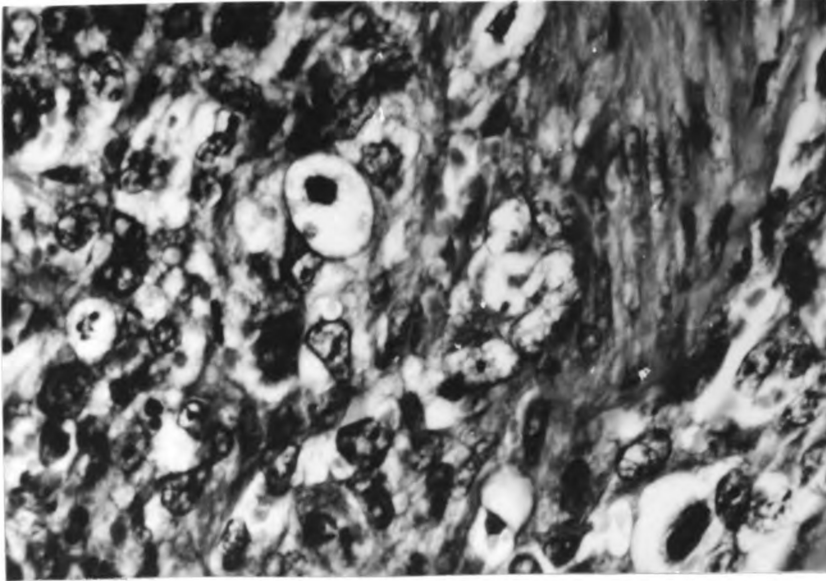


Figure 9. Cellular type of hemangiosarcoma showing vesicular nucleus and large nucleolus (arrow). Hematoxylin and eosin. x 750.

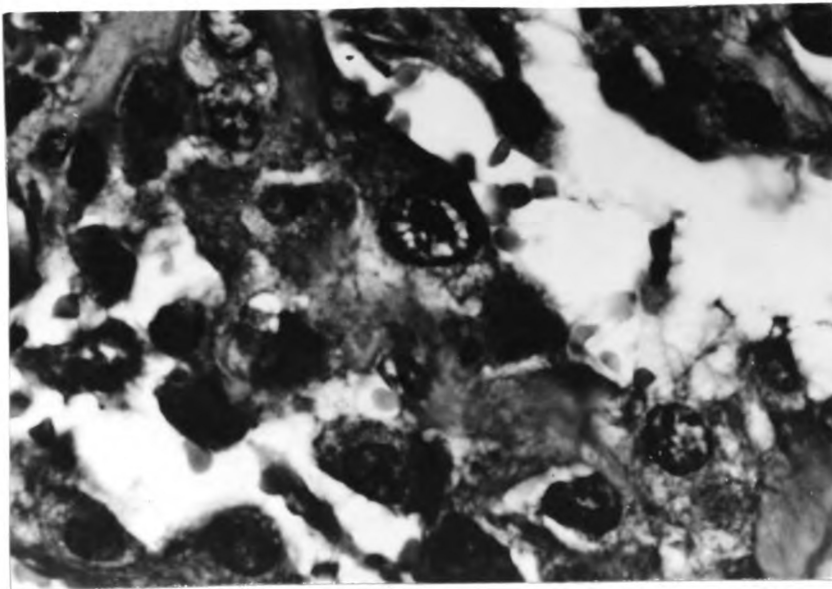


Figure 10. Large nucleolus in malignant cell of a hemangiosarcoma (arrow). Hematoxylin and eosin. x 750.

1.5 microns diameter) were characteristically basophilic, while the larger nucleoli (2 to 5 microns diameter) were usually eosinophilic. If nuclei were large and pleomorphic, they almost always contained multiple nucleoli.

Tendency of the cytoplasm to fuse. In every instance the cytoplasm appeared continuous with that of an adjacent cell, so that no distinct cell membrane was observed. This was especially noticeable in the more anaplastic forms.

Number of mitoses per high power field (hpf). An average of 5 to 6 hpf was examined to determine the number of mitoses. This figure varied from 0 to 10/hpf. There were usually 2 to 3 mitoses per hpf.

Type of mitoses. This characteristic differed from tumor to tumor. Most of the mitoses were normal, but abnormal forms were present in some. One type that was numerous was the so-called "blast" form. This was a form in which the chromatin strands appeared to be exploding from the center toward the periphery in an unorganized pattern. Other abnormal types included the so-called "X form", "Y form", and shrunken constricted form. These latter 3 forms were all classified as bizarre (Figures 11 and 12).

Hyperchromatism. There was some correlation between the degree of hyperchromatism and the degree of anaplasia. In those tumors which characteristically had large vascular spaces filled with blood, there was only slight to moderate hyperchromatism. There was a greater degree of hyperchromatism in the more anaplastic hemangiosarcomas.

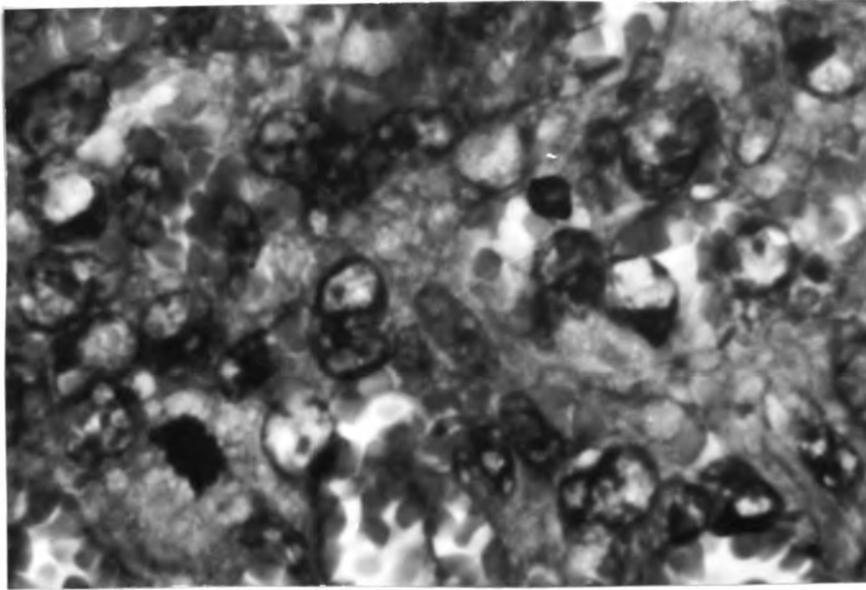


Figure 11. Cellular type of hemangiosarcoma containing large abnormal mitotic figure (arrow). Hematoxylin and eosin. x 750.

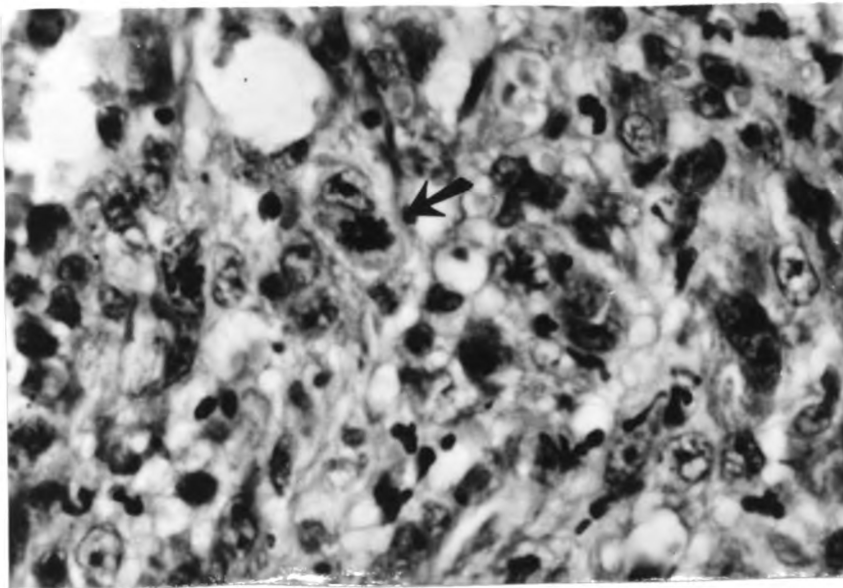


Figure 12. Cellular type of hemangiosarcoma containing mitotic figure (arrow). Hematoxylin and eosin. x 750.

Degree of anaplasia. The term anaplasia in this paper refers to the degree of differentiation. The well differentiated tumors were classified as vascular and less differentiated as cellular. It should be noted that even in those that were classified as cellular, a certain amount of vascularity was seen; otherwise, the diagnosis of hemangiosarcoma would not have been made. The term "cellular" meant, in most cases, whorl or clump formation of tumor cells or lack of organization (Figures 8, 11, and 12), as compared to those which had well defined vascular channels (Figures 13, 14, 15, and 16). The classification of vascular was used, not only in those areas which contained blood, but also in those areas which were forming nonfunctional neocapillaries.

Necrosis. Little necrosis was observed in most of the neoplastic tissues; however, liquefaction necrosis was constant in old areas of hemorrhage. In a few of the more anaplastic forms there was caseation necrosis deep in the cell structure of the tumor.

Amount of stroma. A variable amount of stroma was observed. There was little stroma in the more anaplastic forms, while more stroma was present in those hemangiosarcomas that were well differentiated (Figure 15). The stroma in these tumors consisted of wide bands of collagenous fibers. Few normal blood vessels were found in the tumors. Fine meshlike networks were seen in most of the reticulum-stained sections. This was present even in the most anaplastic tumors (Figures 17, 18, and 19).

Invasion of pre-existing blood vessels and lymphatics. Among the 59 hemangiosarcomas that had pre-existing blood vessels and/or lymphatics,

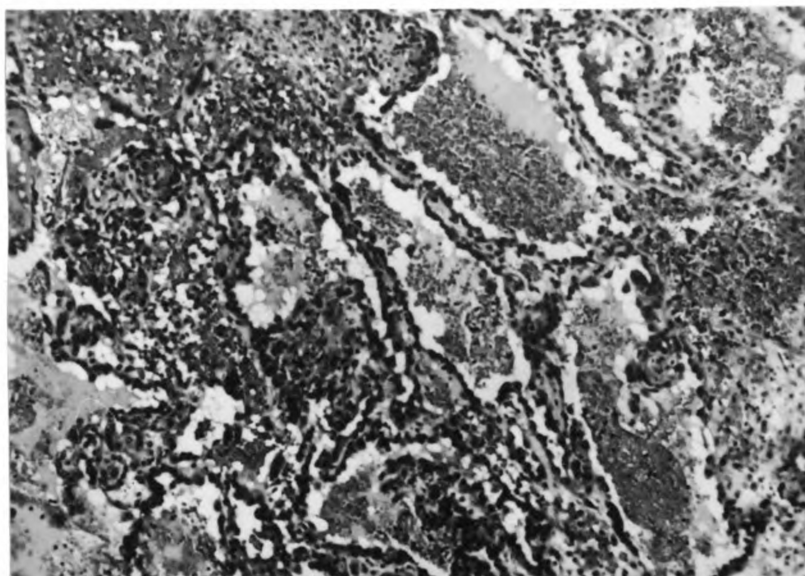


Figure 13. Vascular type of hemangiosarcoma.
Hematoxylin and eosin. x 75.

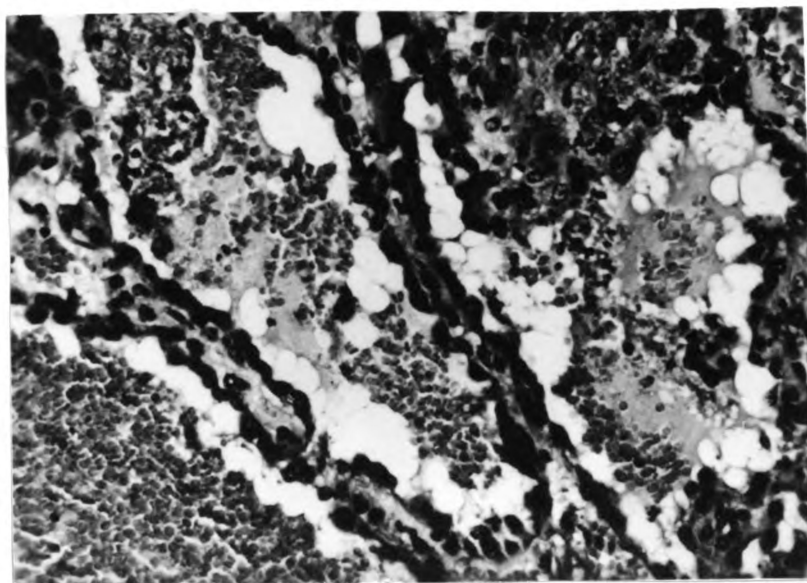


Figure 14. Higher magnification of Figure 13.
Hematoxylin and eosin. x 187.

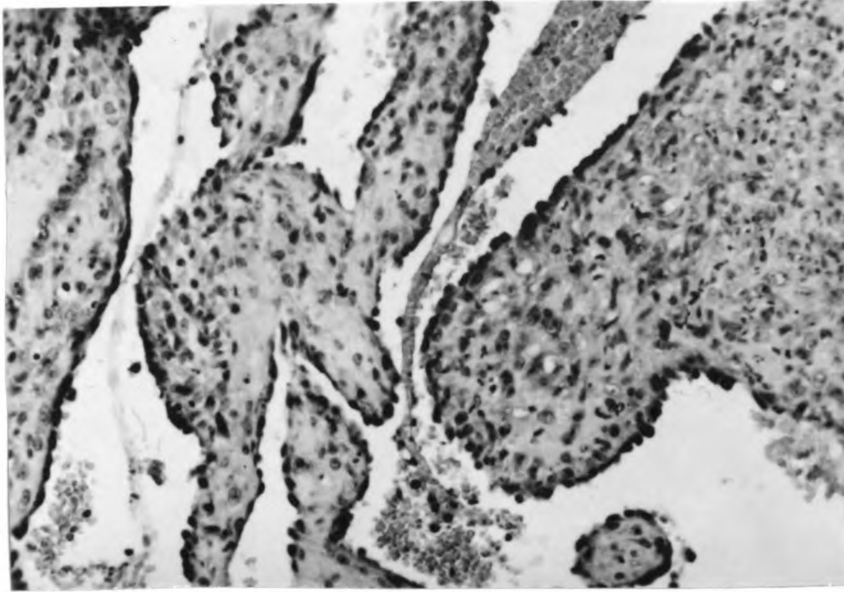


Figure 15. Vascular type of hemangiosarcoma with vein-like channels and abundant stroma. Hematoxylin and eosin, x 187.5.

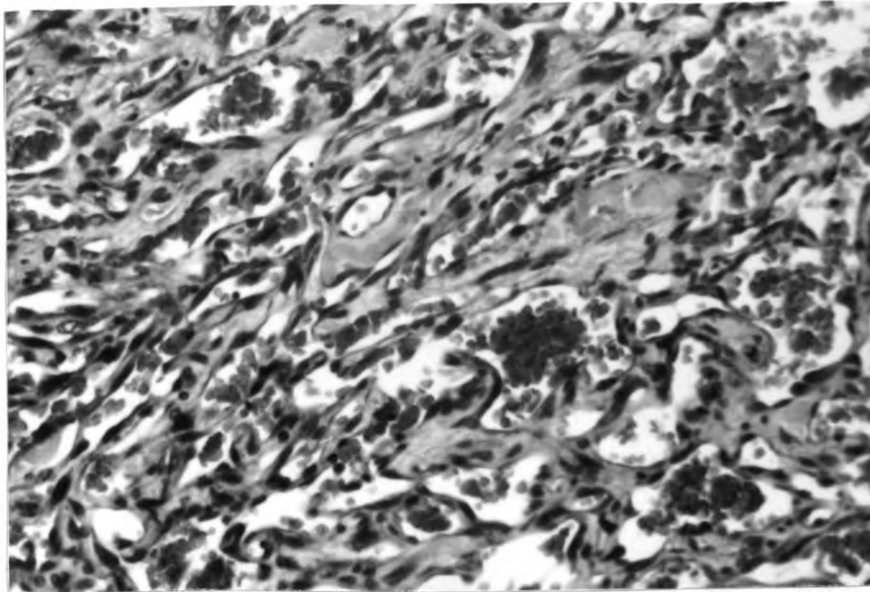


Figure 16. Vascular type of hemangiosarcoma with capillary-like channels. Hematoxylin and eosin, x 187.5.

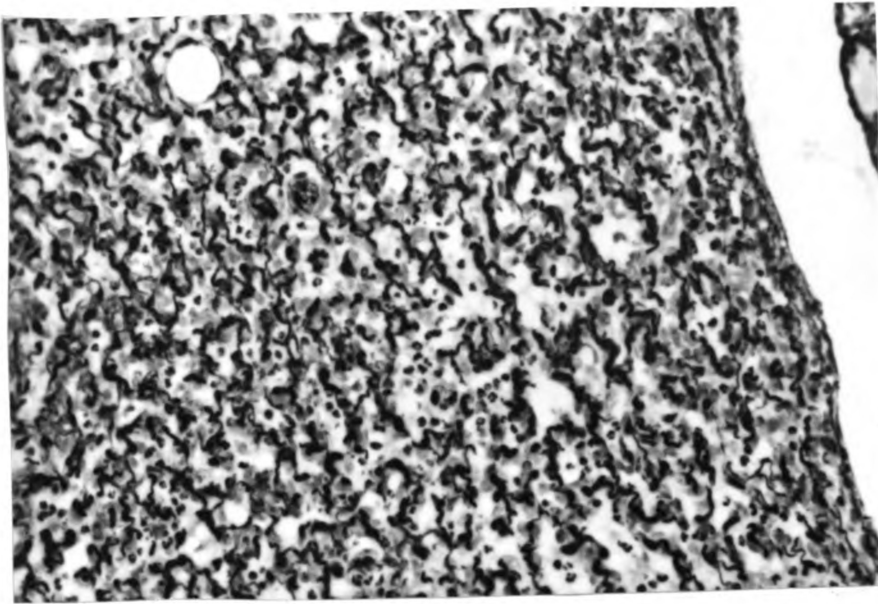


Figure 17. Cellular type of hemangiosarcoma with reticular fibers. Reticulum stain. $\times 187.5$

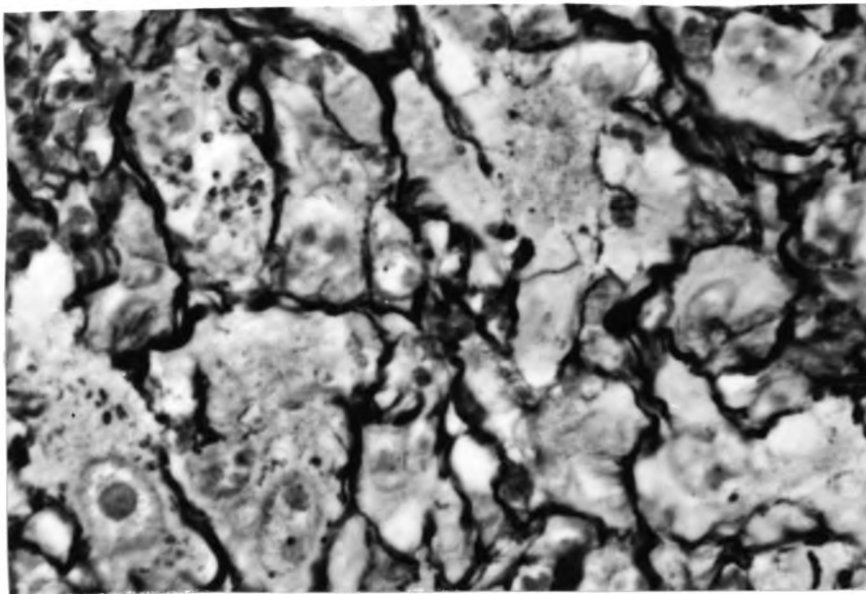


Figure 18. Cellular type of hemangiosarcoma with fine reticular fibers. Note large nucleolus (arrow). Reticulum stain. $\times 750$.

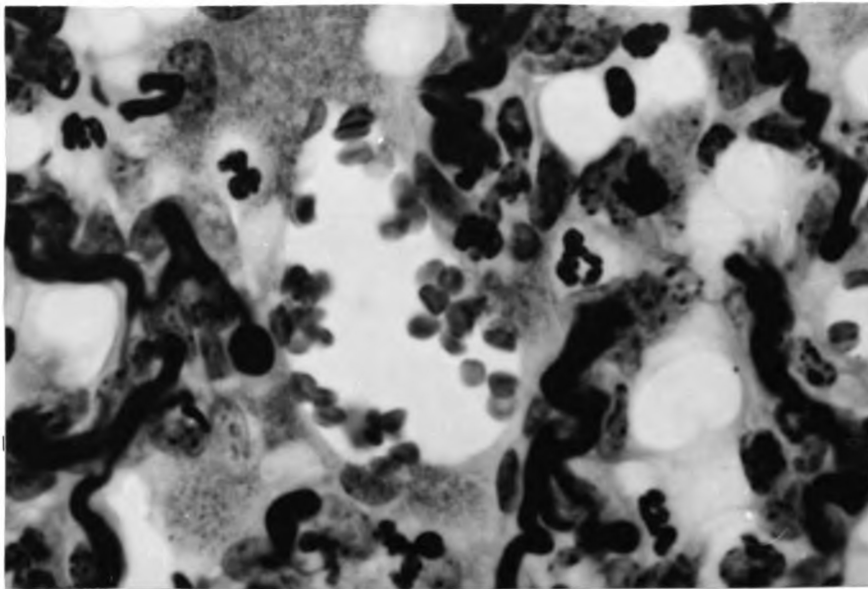


Figure 19. Vascular type of hemangiosarcoma with thick bands of reticular fibers. Reticulum stain. x 750.

malignant cells had invaded these structures in 29. In several instances tumor emboli were found within veins (Figure 20) or arteries (Figure 21). This was particularly true in tissues which represented a metastasis from a primary tumor.

Miscellaneous. Characteristics included under this heading were inflammation, hemorrhage, and pigment-laden macrophages. There was hemorrhage in a number of the tissues which varied from small to large amounts; in fact, many of the tissues superficially resembled large blood clots. Infiltration with inflammatory cells was seen in numerous sections. The most common inflammatory cell was the neutrophil, followed by lymphocytes and plasma cells. In those tumors which had an eroded surface, there was more likely to be inflammation of the purulent type than in those tumors which were deeper. It was not unusual to find many macrophages containing an iron-bearing pigment resembling hemosiderin in hemangiosarcomas which contained a great deal of hemorrhage.

Radiographic Characteristics

While only 12 dogs in this study had radiographs taken during the course of examination, some definite characteristics were observed. Metastasis to the lung produced small miliary-type lesions throughout. All lobes of the lung were equally affected, and the spread seemed to be hematogenous in origin. This gave the lung a so-called "snowflake" appearance (Figure 22). Metastases were usually more radio-opaque than the original tissue.

Osteolysis was observed (Figure 23) in those dogs in which the hemangiosarcoma occurred in or near bone. This varied from a so-called

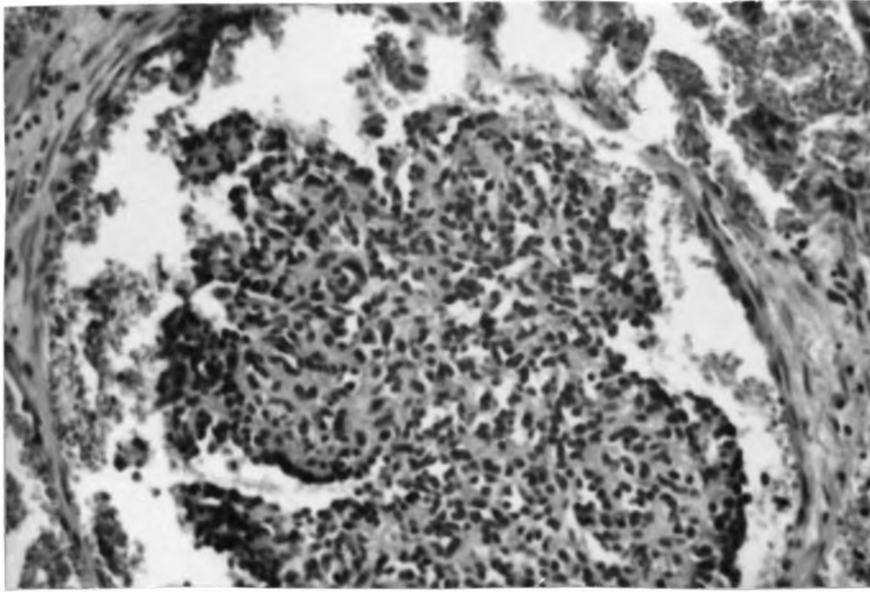


Figure 20. Hemangiosarcoma embolus in vein.
Hematoxylin and eosin. x 187.5.

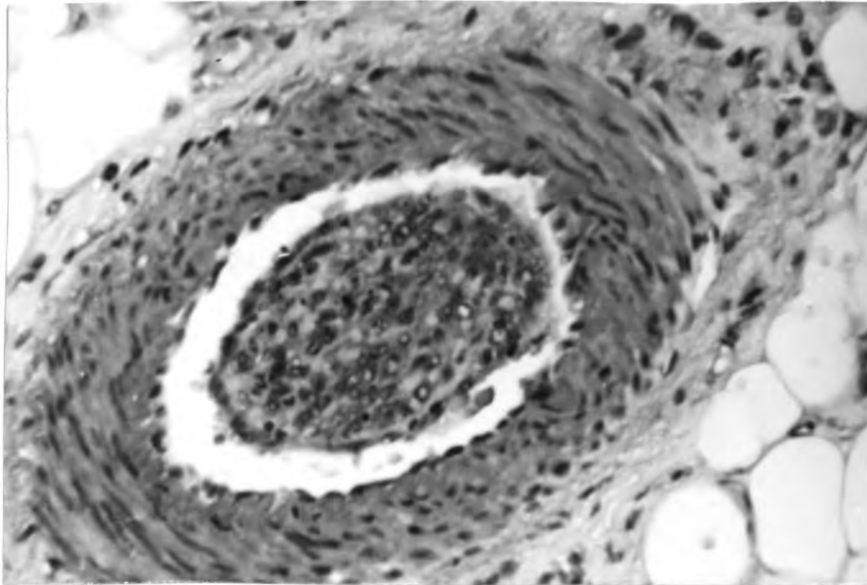


Figure 21. Hemangiosarcoma embolus in artery.
Hematoxylin and eosin. x 187.5.



Figure 22. Lateral radiograph of chest to show "snowflake" appearance of metastatic hemangiosarcoma in lungs.

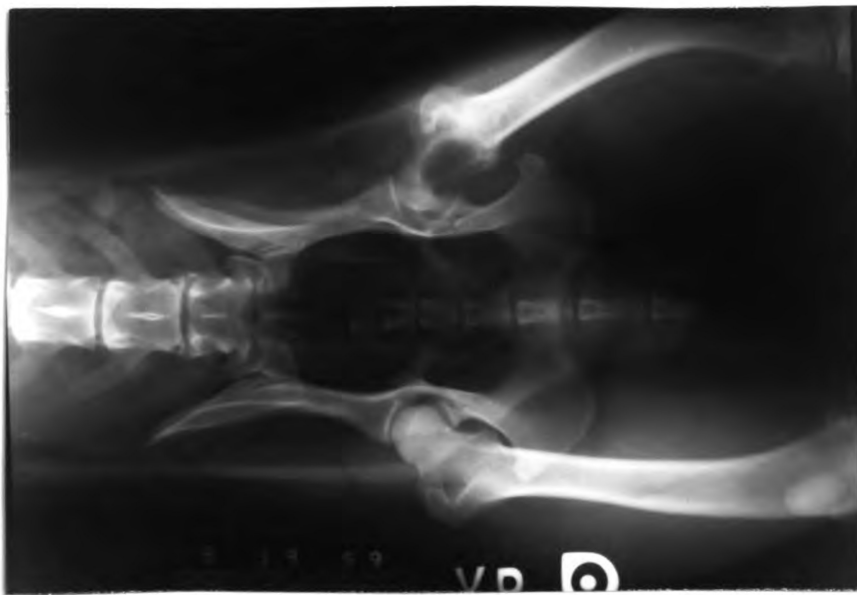


Figure 23. Ventro-dorsal radiograph of pelvic area. Note "punched out" appearance of left femoral head caused by hemangiosarcoma.

"punched out" appearance to a more diffuse reaction with bone proliferation at the edges of the lesion. This diffuse reaction resembled osteomyelitis radiographically. If the tumor occurred in the soft tissue near bone, it presented a homogeneous, somewhat radiolucent appearance, similar to other neoplasms of soft tissue; it was characteristically circumscribed and blended with the adjacent tissue.

DISCUSSION

Hemangiosarcomas are comparatively rare neoplasms. While 76 neoplasms over a 9-year period seemed like a great many, it is a relatively small number compared with the total number of tumors. During this time approximately 16,000 tumors were reviewed by the Department of Pathology. While no exact count was taken, it is reasonable to say that of these at least 14,000 were in the dog.

Occurrence and Gross Characteristics

Feldman (1932, p. 60) stated, "statistical data pertaining to the occurrence of tumors of lower animals are meager and fragmentary". While a great deal of improvement has taken place since that time, very little information of this type was found with regard to hemangiosarcoma in the dog. For this reason, the following discussion is undertaken.

Breed incidence. The number of hemangiosarcomas by breeds to the number of expected cases is compared (TABLE 3), just as Tjalma (1965) did in his study of canine bone cancer. Mulligan (1959) reported a predisposition for hemangiosarcoma in the German Shepherd Dog. In this study, the highest rate of incidence occurred in the Boxer, which had approximately 9 times the expected rate. Also of interest was the large number which occurred in the Cocker Spaniel.

The distribution of the tumor among the various weight classes of the canine population and the expected incidence in each of these

TABLE 3. Comparison of observed and expected cases of hemangiosarcoma among specific breeds and weight classes.

Breed	Range of Occurrence* (%)	% Total Composite Population Samples**	Hemangiosarcoma Cases		Excess or Deficiency of Observed to Expected Cases
			Observed	Expected	
Boxer	2.12 - 3.43	2.40	14	1.61	Excess
Cocker Spaniel	7.21 - 13.79	9.43	14	6.32	Excess
G. Shep.	0.99 - 7.15	4.21	6	2.82	Excess
Lab. Ret.	0.26 - 2.62	1.12	4	0.75	Excess
Collie	2.76 - 9.63	5.60	0	3.75	Deficiency
Beagle	6.80 - 13.72	9.89	2	6.63	Deficiency
Dachshund	1.76 - 9.13	4.10	3	2.74	Excess
F. Terr.	0.57 - 3.55	2.05	1	1.37	Deficiency
Other	---	61.20	23	41.00	Deficiency
	TOTAL	100.00	67	66.99	

Weight Class

Giant 80#	0.00 - 1.61	0.40	0	0.28	Deficiency
Large 40-80#	18.85 - 38.58	28.03	41	19.90	Excess
Medium 20-40#	23.78 - 37.72	30.77	17	21.84	Deficiency
Small 20#	11.30 - 30.40	16.98	10	12.05	Deficiency
Unknown	15.66 - 31.17	23.82	3	16.92	Deficiency
	TOTAL	100.00	71	70.99	

*Among field population samples.

**Base population of 123,123 dogs.

populations is also compared (TABLE 3). As can readily be seen, a definite excess over expected cases was found in the large breeds, while no definite statement can be made about other populations. This information compared favorably with Tjalma's work with canine bone cancer, in which he also found an increased incidence in the large breeds. This poses the question as to whether other types of cancer in the dog occur at a higher frequency in large breeds.

Sex distribution. In Tjalma's (1965) study of the normal canine population, which was based on a population of 99,088, he found the sex distribution to be 52% males and 48% females. In a 30-month study of dogs admitted to the small animal clinic at Michigan State University from July 1960 to January 1963, he found the distribution of cancer to be 226 males and 210 females, with 1 unknown. This is a ratio of 52% in the male and 48% in the female, which corresponded to the universal population. Therefore, a ratio of 1.76 males to 1 female, as presented by this study, would tend to support the findings of Mulligan (1961) and Meier (1963), in which each described a higher incidence of hemangiosarcomas in the male.

It should be pointed out that the influencing factor on the sex difference was the great preponderance of hemangiosarcomas in males of the large (40 to 80 pounds) breeds (Figure 1). Excluding this group, the ratio of males to females was approximately equal.

Age distribution. According to Mulligan (1959), the beginning of "cancer age" was 6 years, with most cases of cancer in the dog occurring between 7 and 14 years. Cotchin (1959) substantiated this figure by pointing out that 56.2% of the tumor-bearing dogs in his study

were between the ages of 6 and 10 years. Meier (1963) found hemangiosarcomas usually occurred in dogs past 6 years of age.

In this study, the mean age for the tumor was 9 years, a little earlier for males and a little later for females, and therefore would fall into the category of "cancer age" as described above.

Primary location. The most common primary sites for hemangiosarcoma were the subcutis and the spleen. These locations were substantiated both in man and in the dog (Lombard, 1935; Cotchin, 1954; Herbut, 1955; Boyd, 1958; Moulton, 1961; Mulligan, 1961; and Robbins, 1962).

Cotchin (1959), in his study of 4,187 tumors of dogs, found the skin to be the most commonly affected area (37.5%).

Pack and Ariel (1958) observed that the tumor most often occurred in the extremities in man. In this study in the dog, the tumor was most commonly found in the trunk.

Metastasis. Mulligan (1961) noted metastasis in 8 of 20 hemangiosarcomas in dogs, with 7 of these 8 to the lungs. This finding was substantiated by our study, where the most common metastatic site was the lung. Other organs affected by metastasis would suggest that the usual route of metastasis by this tumor is hematogenous. Metastasis by the lymphatic route was apparently minimal, since only 5 of 32 tumors had metastasized to the regional lymph nodes. In many instances, as shown (Figures 3 and 4), metastasis apparently occurred in a relatively short period of time, because they all were of uniform size. This was especially true in the lungs, which appeared to be completely infiltrated with the same-sized nodules.

Rate of development. While it appeared from the data that the tumor occurred suddenly, it should be remembered that this conclusion was usually based on the owners' observations. Since cancer is such a subtle disease, many are not recognized by the owner until they are in the late stages. Because of this no accurate statement can be made as to the rate of development.

Recurrence. No definite statement can be made as to the frequency of recurrence, since no follow-up data were available on most of the biopsy specimens submitted to the Department of Pathology for diagnosis. Information concerning this point was available in only 11 cases. Of these, 7 hemangiosarcomas were found to recur.

Tentative diagnosis. As pointed out in the results, a tentative diagnosis of hemangiosarcoma was made by the clinician in only 6 of 21 tumors. In surveying other diagnoses made by the clinicians, nothing specific was found as being characteristic; rather, a wide variety of diagnoses was made. The difficulty in clinical diagnosis is due to wide variability of this tumor both grossly and microscopically. No negative information was obtained in this matter, but it can be postulated that there were probably many tumors clinically diagnosed as hemangiosarcoma which later proved to be some other lesion on histopathologic examination. Just as many of these tumors were not as vascular as one would expect them to be, there were also highly vascular tumors which were not hemangiosarcomas. In bone, the tumor was often clinically diagnosed as an osteolytic osteosarcoma by the clinician.

If the tumor had metastasized to the lung, and if radiographs were taken of this area, a correct diagnosis was often made by the radiologist.

Size of tumor. This study supported the conclusions of Bell (1956) and Boyd (1958), who pointed out that these tumors are often round, variable in size, and hemorrhagic in appearance. The metastatic lesions were often less hemorrhagic than the primary lesion. Frequently, blood clots were associated with the tumors where they evidently had ruptured (Figure 6). This complication often was given as the cause of death because, while the neoplastic process had not advanced to the point of involvement throughout the body, one of the tumors had ruptured and resulted in exsanguination. This was a common finding substantiated by Moulton (1961), who pointed out that "hemorrhage and necrosis are almost constant features", and by Mulligan (1949), who stated that "rupture of metastases has resulted in sudden fatal hemorrhage".

Microscopic Characteristics

As Watson and McCarthy (1940) pointed out,

"An angioma is a true neoplastic process involving vascular or lymphatic tissue. In this respect, an angioma differs distinctly from simple self-limiting vessel hypertrophy such as occurs in granulation tissue, and it bears no relation to the ordinary dilatation of previously formed vessels such as occurs in varices."

This seemed to be the key point in the microscopic characteristics of this tumor. Some areas of the tumors had benign or even normal appearing endothelial structure, but the characteristics of other areas established the true malignancy of this tumor (TABLE 2). It

is hoped that the following discussion will point out the characteristics which will help establish the differences between malignant and benign endothelial tumors.

Shape of the cell. The general outline of the cell, as described in Results, was spindle shaped. Pack and Ariel (1962) pointed out that, in the more anaplastic tumors the endothelial cells may grow in whorls or undifferentiated sheets. This feature was also found in the study, in that the shape of the cell was less elongated in the more anaplastic forms of the tumor. This coincided with the over-all failure to form vascular channels in these same anaplastic tumors.

Size and shape of nuclei. The general shape of the nucleus was oval. The more anaplastic the hemangiosarcoma, the greater the tendency for the nucleus to round up or to attain pleomorphic characteristics. This characteristic was similar to that of other tumors of mesenchymal origin, in that the more anaplastic they are, the less they resemble the tissue or cell of origin. Also, the more nearly round nuclei had a tendency to be vesicular.

Size and staining properties of nucleoli. There was no reference to this point in the literature, but in our study the larger the nucleoli, the greater the tendency for them to be eosinophilic. It may be because of the optics involved that small nucleoli (0.5 to 1.5 microns) were darker and appeared basophilic. Therefore, the significance of this finding might be debatable. Large and multiple nucleoli were considered as factors in establishing a diagnosis of malignancy.

Number of mitoses. While this figure was variable, usually 2 to 3 mitoses were found per high power field. A greater number was usually observed in the more anaplastic tumors. This was considered as moderate in number compared to other forms of cancer. It must be remembered that this subjective observation would vary from investigator to investigator. However, this tended to support the early work by Feldman (1932) and the more recent work by Mulligan (1961) and Moulton (1961), who noted a moderate number of mitoses in hemangiosarcomas.

Types of mitoses. The appearance of bizarre mitotic figures may depend upon the angle the cell was cut. This is most likely true during prophase. However, when a tripolar or "blast" mitotic figure was observed it was described as abnormal. Mulligan (1961) was the only author to state that abnormal mitotic figures occurred in hemangiosarcomas in the dog. Robbins (1962) found most of the mitotic figures to be of normal type.

Hyperchromatism. There was no definite relationship between hyperchromatism and the degree of malignancy. In discussing hyperchromatism, various factors must be considered, such as the thickness of the section, the amount of hematoxylin, and even the type of filter in the illuminating system. The nucleus of the normal endothelial cell is dark in appearance due to the small size of the nucleus and condensation of the chromatin. In fast-growing granulation tissue, the nucleus stains somewhat lighter. A greater degree of hyperchromatism was usually seen in the more cellular areas of the hemangiosarcomas.

However, there seemed to be a point of diminishing returns. In those tumors which were highly anaplastic, with vesicular type nuclei, the reverse was found to be the rule.

Degree of anaplasia. The highly vascular hemangiosarcomas were considered well differentiated and thus less anaplastic, whereas the highly cellular tumors (less vascular) were thought to be more anaplastic. The hemangiosarcomas varied considerably in these respects. The vascular structure in many of the tumors appeared quite benign, even though they had metastasized. No effort was made to categorize the tumors as to whether they were forming veins or capillaries, since there could be areas of capillary formation and other areas in the same section where the formation tended to be more vein-like. It was considered that if a tumor were cellular in appearance this represented a more anaplastic form of the tumor.

An interesting experiment that could be undertaken with this tumor would be the use of Gomori's technique for demonstrating alkaline phosphatase (Pierce, 1960). The rationale for such was best explained by Urbach and Graham (1962), who stated that alkaline phosphatase activity appeared to be most intense only in the endarterial or capillary endothelium, while the wall of the venules or larger arterioles had little or no enzymatic activity. An experiment such as this might reveal the actual point of origin of the malignant cells. Kapf (1957) and Ellis et al. (1958) described this technique and used it as an indication of the proliferative activity of the blood vessels in a particular area. In our study this technique was not possible, since it required either frozen sections or tissues fixed in 2 to 3

changes of absolute alcohol before the staining technique could be applied.

Necrosis. A majority of the tumors investigated was necrotic to some degree, usually in areas of old hemorrhage. This was probably the result of rupture of 1 of the delicate endothelial structures. In a few, however, a caseous type of necrosis was found deep in the anaplastic areas. The question arose as to the reason for necrosis in a tumor which essentially was a mass of blood vessels, where it would seem that the cells were undoubtedly well oxygenated. An explanation for this was found in the work of Urbach and Graham (1962), who stated,

"The presence of large numbers of capillaries in tumor tissue does not necessarily imply an abundant supply of nutrient materials as indicated by tissue tension measurements, which suggests that the rich vascular bed is functionally inefficient."

Proof of this was obtained by Urbach and Noell (1958), in a study in which the oxygen tension was measured in normal tissue and tumor tissue by use of a polarographic current. There was a local state of hypoxia in tumor tissues, and it was suggested that this was the primary reason for necrosis deep in tumor tissue. Again, an alkaline phosphatase stain would be valuable because the presence of this enzyme is a direct function of the metabolic activity of the endothelial cells.

Amount of stroma. There was variation in the amount of stroma. A large amount of stroma was observed in highly vascular hemangiosarcomas, while the more cellular tumors had less stroma. In some of the more anaplastic forms, the stroma was particularly scanty. The

reticulum stain revealed the interesting fact that the endothelial cells, even in the most anaplastic forms of the tumor, tended to lay down a fine delicate reticular meshwork. This special stain was applied to a vascular fibrosarcoma and leiomyosarcoma. In these tumors, the reticulin was associated only with endothelial cells and not with the tumor cells. For this reason, and also for the reason that the hemangiosarcomas appeared to have a characteristic meshlike network, it is suggested that this stain is of value in the differentiation of hemangiosarcomas from vascular tumors of other types. Stout (1943) also emphasized the value of this stain in diagnosing tumors of vascular origin.

Invasion of pre-existing blood vessels and lymphatics. In several instances tumor emboli were found in veins and arteries, while few were seen in lymphatics, suggesting that the former played a greater role in the metastasis from the primary site to secondary areas of the body. This point was suggested in an earlier section of this discussion. Occasionally, these tumor emboli retained their vascular form (Figure 20). It was not surprising that these tumor emboli were found in the blood vessels, since they originate from the vascular endothelium.

Miscellaneous. As pointed out in the previous areas of this discussion, hemorrhage seemed to be an almost uniform finding. Mulligan (1961) emphasized the fact that these tumor cells are delicate and have a tendency to break, resulting in small to vast areas of hemorrhage. The large number of macrophages laden with a hemosiderin-type pigment would support the observation that hemorrhage had been present

for long periods of time. The significance of inflammation in these tumors is questionable. Although many of the tumors had large numbers of inflammatory cells, this could be explained by the tissue breakdown resulting in a leukotaxic effect. Another explanation would be that in many of these tumors which had eroded surfaces, bacterial infection could easily have produced the influx of inflammatory cells.

Radiographic Characteristics

The fact that only 12 dogs with hemangiosarcoma had radiographs limited the conclusions that could be made. Two significant observations were made. First was the characteristic small miliary-type ("snowflake") distribution of metastases to the lung (Figure 22). The distribution of these can be explained by the hematogenous spread of the metastases. According to Mostosky (1965), "this characteristic distribution of metastatic lesions is pathognomonic of the hemangiosarcoma". This finding could be of great value to the small animal clinician who uses radiographs as part of his diagnostic procedure.

Second, osteolysis occurred adjacent to the tumor (Figure 23). This supports the findings of numerous authors (De Lorimier et al., 1954; Ritvo, 1955, and Bradley and Coley, 1960), who described hemangiosarcomas in bone as resembling osteomyelitis or osteosarcomas in appearance. Again, Mostosky pointed out that the radiographic appearance of tumors in bone was fairly characteristic and, along with the diagnoses mentioned above, hemangiosarcoma must be considered when viewing a radiograph which has the appearance shown (Figure 23).

SUMMARY

This thesis is a retrospective study of 76 canine hemangiosarcomas diagnosed in the Department of Pathology from 1956 to 1965. The study was divided into 3 basic parts: (1) occurrence and gross characteristics, (2) microscopic characteristics, and (3) radiographic characteristics of the tumor.

Hemangiosarcomas occurred with greater incidence in the large breeds of dogs, particularly the Boxer. There was a sex ratio of approximately 2 males to 1 female. The mean age was 9 years, a little earlier in males (8.5 years) and a little later in females (10.7 years). The most common primary sites were the subcutis and the spleen. If metastasis occurred the most common site was the lung. The tumor was difficult to diagnose without histologic examination. Hemorrhage was an almost constant finding.

The neoplastic endothelial cell was generally spindle shaped, resembling a short fibroblast. The nucleus was generally oval and of moderate size (9 x 12 microns), often containing large eosinophilic single and multiple nucleoli. There was a tendency for the cytoplasm of 1 cell to fuse with the next, presenting a syncytial type of arrangement rather than individualized cells. There were both normal and abnormal mitoses. Most of the nuclei had a moderate amount of hyperchromatism. The cellular arrangement varied from normal appearing vascular cells to undifferentiated cellular sheets arranged in whorls or clumps with little vessel formation. The use

of a reticulum stain revealed a fine reticular meshlike network, even in the most anaplastic areas of the tumor. There was a variable amount of necrosis, especially associated with hemorrhages. The amount of stroma varied from large amounts in the vascular forms to scanty in the more anaplastic cellular forms. Invasion of blood vessels occurred in 29 of 59 hemangiosarcomas. There were inflammatory cells in many of the tumors, particularly those which had an eroded surface.

Two radiographic characteristics were observed. First, the metastatic lesions in the lungs presented a "snowflake" pattern. Secondly, osteolysis occurred in those neoplasms that were in or near bone.

LIST OF REFERENCES

- American Cancer Society. Manual of Tumor Nomenclature and Coding, corrected ed. 1953. Subcommittee of the Statistics Committee, Am. Cancer Soc., New York: 53-54.
- American Cancer Society. Systematized Nomenclature of Pathology, 1st ed. 1965. Coll. of Amer. Path., Chicago, Ill.: 121.
- Anderson, W. A. D. Pathology, 4th ed. 1961. C. V. Mosby Co., St. Louis, Mo.: 576.
- Andervont, H. B., Grady, H. G., and Edwards, J. E. 1942. Induction of hepatic lesions, hepatomas, pulmonary tumors, and hemangio-endotheliomas in mice with O-amino-azotoluene. J. Nat. Cancer Inst., 3: 131-153.
- Arey, L. B. 1954. Developmental Anatomy, 6th ed. W. B. Saunders Co., Philadelphia, Pa.: 340-349.
- Armed Forces Institute of Pathology. Manual of Histologic and Special Staining Technics, 2nd ed. 1960. McGraw-Hill Book Co., Inc., New York: 30, 151.
- Bell, E. T. A Textbook of Pathology, 8th ed. 1956. Lea and Febiger, Philadelphia, Pa.: 363.
- Boyd, W. A Textbook of Pathology, 6th ed. 1958. Lea and Febiger, Philadelphia, Pa.: 259-261.
- Bradley, L., and Coley, M. D. Neoplasms of Bone, 2nd ed. 1960. Paul B. Hoeber, Inc., Medical Div. Harper and Brothers, New York: 341-363.
- Brodey, R. S. 1960. A clinical and pathologic study of 130 neoplasms of the mouth and pharynx in the dog. Am. J. Vet. Res., 21: 787-812.
- Carlson, W. D. Veterinary Radiology. 1961. Lea and Febiger, Philadelphia, Pa.: 234-239.
- Cotchin, E. 1954. Further observations on neoplasms in dogs, with particular reference to site of origin and malignancy. I. Cutaneous, female genital and alimentary systems. Brit. Vet. J., 110: 218-230.

- Cotchin, E. 1954. Further observations on neoplasms in dogs, with particular reference to site of origin and malignancy. II. Male genital, skeletal, lymphatic and other systems. *Brit. Vet. J.*, 110: 274-286.
- Cotchin, E. 1959. Some tumors of dogs and cats of comparative veterinary and human interest. *Vet. Rec.*, 71: 1040-1050.
- De Lorimier, A. A., Moehring, H. G., and Hannan, J. R. 1954. Developmental and systemic conditions and local lesions in the extremities, in *Clinical Roentgenology*, Vol. I. Charles C. Thomas, Springfield, Ill.: 335-337.
- De Navasquez, S. 1936. Angioblastoma of the spleen with metastasis in the liver. *J. Path. Bact.*, 42: 651-656.
- Ellis, R. A., Montagna, W., and Fanger, H. 1958. Histology and cytochemistry of human skin. XIV. The blood supply of the cutaneous glands. *J. Invest. Dermat.*, 30: 137-146.
- Edwards, J. E., Andervont, H. B., and Dalton, A. J. 1942. A transplantable malignant hemangio-endothelioma of the liver in the mouse. *J. Nat. Cancer Inst.*, 2: 479-490.
- Farrell, J. M., Jr., and Farrell, D. 1960. Hemangiosarcoma - A case history. *Southwest Vet.*, 14: 61.
- Feldman, W. H. 1932. *Neoplasms of Domesticated Animals*. W. B. Saunders Co., Philadelphia, Pa.: 60, 178-188.
- Freilich, E. B., and Coe, G. C. 1936. Angiosarcoma: Case report and review of the literature. *Am. J. Cancer*, 26: 269-275.
- Furth, J., and Furth, O. B. 1938. Monocytic leukemia and other neoplastic diseases occurring in mice following intrasplenic injection of 1:2-benzpyrene. *A. J. Cancer*, 34: 169-183.
- Geshickter, C. F., and Keasbey, L. A. 1935. Tumors of blood vessels. *Am. J. Cancer*, 23: 568-591.
- Golden, R. 1959. *Diagnostic Radiology*, Vol. 1. Williams and Wilkins Co., Baltimore, Md.: 190L-190N.
- Ham, A. W. 1957. The circulatory system, in *Histology*, 3rd ed. J. P. Lippincott Co., Philadelphia, Pa.: 480-507.
- Herbut, P. A. 1955. *Pathology*. Lea and Febiger, Philadelphia, Pa.: 256-258.
- Jäplid, Burtil. 1961. Haemangioendotheliomas in poultry. *J. Comp. Path.*, 71: 370-376.

- Joest, E. 1924. *Spezielle Pathologische Anatomie Der Haustiere*, Vol. III. Richard Schoetz, Berlin: 498-509.
- Kapf, A. W. 1957. Alkaline phosphatase in skin. *Arch. Dermatol.*, 75: 1-29.
- Kingman, H. E., and Newsom, I. E. 1918. An interesting tumor. Case report. *J. Am. Vet. Med. Assoc.*, 5: 703-705.
- Kinkade, J. M. 1949. Angiosarcoma: a review of the literature. *Ann. Otol. Rhin. & Laryng.*, 58: 159-167.
- Langhans, T. 1879. *Arch. Path. Anat.*, 75: 273. Cited by De Navasquez.
- Lasser, E. C., and Von Schowingen, R. S. 1955. Arteriography in bone tumors. *N. Y. State J. Med.*, 55: 3425-3430.
- Lasserre, R., Lombard, Ch., and Labatut, R. 1938. Recherches sur le cancer des animaux domestiques. *Rev. Med. Vet.*, 15, 8: 425-451.
- Leisering. 1861. Gefabgeschwulst b. e. hunde. *Ber. Vet.-Wes. Sachsen* 6. Cited by Joest.
- Lichtenstein, L. 1952. *Bone Tumors*. C. V. Mosby Co., St. Louis, Mo.: 125-128.
- Lieberman, L. L. 1955. Malignant hemangio-endothelioma of the canine heart. *J. Am. Vet. Med. Assoc.*, 126: 296.
- Lienaux, E. 1899. Deux cas d'endotheliomes de la region parotidienne chez le chien. *Ann. de Med. Vet.*, 48: 502. Cited by Feldman.
- Lillie, R. D. 1954. *Histopathologic Technic and Practical Histochemistry*. Blackiston Co., New York.
- Lindsay, S., and Gilmore, J. W. 1946. Primary splenic endothelial sarcoma in a dog. *J. Am. Vet. Med. Assoc.*, 109: 194-197.
- Lombard, C. 1964. Vegetating sublingual haemangio-endothelioma in cats. *Bull. Acad. Vet., Fr.*, 37: 163-167.
- Magnusson, R. 1934. Sarcoma of the small intestine in connection with a case of hemangiosarcoma of the jejunum. *Acta. Chir. Scandinav.*, 73: 576-590.
- Mallory, F. B. 1914. *The Principles of Pathologic Histology*. W. B. Saunders Co., Philadelphia, Pa.: 309-326.
- Meier, H. 1963. Epizootiology of cancer in animals. *Ann. N. Y. Acad. Sci.*, 108: 617-1325.

- Mostosky, U. V. 1965. Small Animal Clinic, College of Veterinary Medicine, Michigan State University, East Lansing, Mich. (personal communication).
- Moulton, J. E. 1961. Tumors of Domestic Animals. Univ. of Calif. Press, Berkeley and Los Angeles, Calif.: 77-79.
- Mulligan, R. M. 1949. Neoplasms of the Dog. The Williams and Wilkins Co., Baltimore, Md.: 73-76.
- Mulligan, R. M. 1961. Mesenchymal and neurilemmal tumors in the dog. Arch. Path., 71: 512-531.
- Pack, G. T., and Ariel, I. M. 1958. Tumors of the Soft Somatic Tissues. Paul B. Hoeber, Inc., Harper & Brothers, New York: 442-450.
- Palade, G. E. 1953. Fine structure of blood capillaries. J. Appl. Physics, 24: 1424.
- Pearse, H. G. E. 1960. Histochemistry - Theoretical and Applied, 2nd ed. J. & A. Churchill, Ltd., London: 868-869.
- Pounden, W. D., and Sprunger, E. 1947. Malignant hemangioendothelioma of a canine spleen. N. Am. Vet., 28: 461.
- Public Health Service, U. S. Department of Health, Education and Welfare. 1964. A Standard of Veterinary Diseases and Operations, 1st ed. Bethesda, Md.: 110.
- Regenbagen. 1907. Über das vorkommen der hamangiome. Monatschr. f. prakt. Thierh., 18: 293-298. Cited by Feldman.
- Rigdon, R. H. 1952. Tumors produced by methylcholanthrene in the duck. Arch. Path., 54: 368-377.
- Ritvo, M. 1955. Bone and Joint X-ray Diagnosis. Lea and Febiger, Philadelphia, Pa.: 471-472.
- Robbins, S. L. 1962. Textbook of Pathology, 2nd ed. W. B. Saunders Co., Philadelphia, Pa.: 471-472.
- Robinson, J. M., and Castleman, B. 1936. Benign metastasizing hemangioma. Ann. Surg., 104: 453.
- Shennan, T. 1914. Histologically normal malignant angioma, with numerous metastases. J. Path. Bact., 19: 139-154.
- Siedamgrotzky. 1874. Angiom e. hundes. Ber. Vet.-Wes. Sachsen 19. Cited by Joest.
- Simpson, L. 1960. Hemangioendotheliomatosis in a dog. Golden Jubilee Commemorative Vol., Coll. of Vet. Med., Univ. Philipp., Diliman, Rizal, Philippines: 82-93.

- Steiner, P. E. 1942. Comparative pathology of induced tumors of the salivary glands. Arch. Path., 34: 613-624.
- Stout, A. P. 1943. Hemangio-endothelioma: a tumor of blood vessels featuring vascular endothelial cells. Ann. Surg., 118: 445.
- Thiele. 1904. Arch. f. path. Anat., 178: 296. Cited by De Navasquez.
- Tjalma, R. A. 1965. National Cancer Institute, National Institutes of Health, U. S. Public Health Service, Bethesda, Md. (personal communication).
- Urbach, F., and Graham, J. F. 1962. Anatomy of human skin tumour capillaries. Nature, Lond., 194: 652-654.
- Urbach, F., and Noell, W. K. 1958. Effects of oxygen breathing on tumor oxygen measured polarographically. J. Appl. Physiol., 13: 61-65.
- Watson, W. L., and McCarthy, W. D. 1940. Blood and lymph vessel tumors; report of 1,056 cases. Surg. Gynec. & Obstet., 71: 569-588.
- Weipers, W. L., and Jarrett, W. F. H. 1954. Haemangioma of the scrotum of dogs. Vet. Rec., 66: 106-107.
- White, J., and Stewart, H. L. 1942. Intestinal adenocarcinoma and intra-abdominal hemangio-endothelioma in mice ingesting methylcholanthrene. J. Nat. Cancer Inst., 3: 331-347.
- Willis, R. A. 1948. Pathology of Tumors. Butterworth and Co., Ltd., London, C. V. Mosby Co., St. Louis, Mo.: 713.

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