**Mediastinal lymphoma in dogs is homogeneous compared to thymic epithelial neoplasia and is more likely to envelop the cranial vena cava in computed tomographic images.**

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Key words: computed tomography, dog, lymphoma, mediastinum, neoplasia, thymoma

Running head: CT of mediastinal neoplasia

**Abstract**

In order to identify computed tomographic (CT) signs that could be used to distinguish cranial mediastinal lymphoma and thymic epithelial neoplasia, a retrospective case-control study was done of 62 dogs that had thoracic CT and confirmed diagnosis of lymphoma (n=33) or thymic neoplasia (n=29). Thymic neoplasms included 24 thymomas and 5 thymic carcinomas. Associations between CT signs and diagnosis were tested using binary logistic regression and results expressed as odds ratio (OR) and 95% confidence interval (CI). Dogs with thymic epithelial neoplasia were significantly older than dogs with lymphoma (median age 8.6y versus 6.0y, p=0.007), but there were no significant differences in prevalence of clinical signs. Regarding subjective CT signs; diagnosis of thymic epithelial neoplasia was associated with heterogeneous attenuation in pre- (OR 23.3, CI 4.5-121.1) and post-contrast (OR 30.7, CI 3.6-265.0) images. Conversely, envelopment of the cranial vena cava by the mass was less likely with thymic epithelial neoplasia than lymphoma (OR 0.07, CI 0.007-0.66). Greater standard deviation (SD) of HU values in post-contrast images was associated with thymic epithelial neoplasia (p=0.005). Based on ROC analysis, SD >17HU of the mass in post-contrast images had a sensitivity of 72% and specificity of 79% for thymic epithelial neoplasia. There were no significant differences in morphology, prevalence of calcification, mediastinal lymphadenopathy, cranial vena cava invasion, collateral vessels or pleural fluid associated with these tumors. Thymic epithelial neoplasms tended to occur in older dogs and were heterogeneous in CT images, whereas mediastinal lymphoma was more homogeneous and more likely to envelop the cranial vena cava.

**Introduction**

Lymphoma and thymoma are, respectively, the first and second most common mediastinal neoplasia in dogs.1 Other differential diagnoses for a cranial mediastinal mass include cyst, abscess, granuloma, haematoma or other neoplasia (for example ectopic thyroid carcinoma). Thymoma is a neoplasm of thymic epithelial cells and contains epithelial cells and small lymphocytes in varying proportions.2 Mediastinal lymphoma is a neoplasia of lymphoid cells that may arise in the thymus or the mediastinal lymph nodes. Cytologic or histologic samples of both thymoma and lymphoma can contain small lymphocytes with similar morphology which, in combination with only a small number of thymic epithelial cells in a sample, could lead to misdiagnosis of a thymoma as a lymphoma, or give inconclusive results.1,2 In humans, the most widely used classification for thymic neoplasia is the World Health Organization (WHO) classification, which includes four thymoma subtypes, atypical thymoma and thymic carcinoma.3,4 Canine thymic epithelial tumors have similar pathologic features as described in humans, with classifications ranging from benign thymoma to malignant atypical thymoma and thymic carcinoma.5 Grossly, thymomas are frequently well-circumscribed with a capsule of variable thickness that can contain calcified plaques. Lobulation of thymic tumors due to intersecting connective tissue bands may be observed6,7,8,9 however, lobulation is also observed in some mediastinal lymphomas.6 Thymomas frequently contain cystic structures2,8,10 that may be observed sonographically.11 It is important clinically to distinguish thymoma and lymphoma because the optimal treatment differs, with surgical excision preferred for thymoma and chemotherapy the treatment of choice for mediastinal lymphoma.8,12

Computed tomography (CT) is frequently used to examine mediastinal masses in humans and characteristics of thymoma and mediastinal lymphoma have been described.13 CT features suggesting thymoma are a well-circumscribed mass with a rounded or oval shape and an off-midline location, whereas a poorly circumscribed, lobular or multinodular mass and pericardial involvement favors the diagnosis of lymphoma.13 In humans, CT features of thymic neoplasms have been correlated with the WHO classification; with lobulation, irregular contour, fat invasion, and heterogeneous contrast enhancement associated with the higher-grade thymic epithelial neoplasms.14,15

Existing descriptions of CT findings in dogs with mediastinal masses suggest that lymphoma and thymoma may share similar features, including esophageal displacement, tendency to wrap around the cranial mediastinal vessels with variable degrees of vascular invasion, regional lymphadenopathy and pleural effusion.7,8,12,16 A study of 9 dogs found no associations between the CT appearance and histologic characteristics of mediastinal masses.7 In contrast, a study comparing the sonographic features of thymoma and mediastinal lymphoma in 50 dogs found that internal cystic structures and heterogeneous echogenicity were associated with thymoma.11

The aim of the present study was to evaluate a greater range of subjective and objective CT features, within a larger cohort than has been previously evaluated, to identify any significant differences in the CT appearance of canine mediastinal lymphoma and thymic epithelial neoplasms that could be used as a basis for tentative diagnosis, or to usefully guide the diagnostic approach and case management where cytological or histological results are inconclusive.

**Materials and methods**

 This was a retrospective case-control study, ethical approval was granted the Animal Welfare and Ethical Review Body, University of Bristol. Clinical records were searched across three centers (Langford Vets University of Bristol, The Royal Veterinary College University of London, Institute of Veterinary Science University of Liverpool) for dogs that had thoracic CT, including post-contrast images, and a confident diagnosis of thymoma, thymic carcinoma or lymphoma based on cytology or histology, and in some cases with additional information from immunohistochemistry, flow cytometry or PCR for antigen receptor rearrangements (PARR). Cases were excluded, and not recorded, when a confident final diagnosis was not achieved by the treating clinician. At all three centers thoracic CT is part of the normal diagnostic evaluation for mediastinal masses, all dogs were treated according to standard hospital protocols, and all hospital consent forms include permission to use patient data in retrospective studies.

Signalment, history and clinicopathological data were recorded for each dog, including patient identification number, age, breed, sex, CT study date, main clinical presenting signs, presence of hypercalcemia, final diagnosis, and method of diagnosis (cytology or histology). Search and clinical data extraction were done by ER, EM and FS at their respective institutions.

As part of the study inclusion criteria, all CT scans of the thorax required soft tissue reconstruction algorithms and included series obtained after intravenous contrast administration. Due to this being a multi-center retrospective study the protocol for CT was not standardized; however, all studies were performed using third generation, multi-slice CT units (see Appendix) with high kVp (120–130) in helical scan mode with variable pitch. CT images were reconstructed with a 512 x 512 matrix and variable field of view depending on the size of the dog. Slice thickness varied between 2 and 3mm. All dogs were sedated or anaesthetized for CT, being a retrospective study this was not standardised. Thoracic CT studies were anonymized, reviewed and objective measurements made using commercially available DICOM software (Osirix, Pixmeo, Switzerland) by three Board-certified radiologists (CWS, CL, TM) who were blinded to the final diagnosis. Decisions about CT findings were reached by consensus for every feature.

Transverse CT images were assessed with respect to the following subjective features: 1. location (mainly to left/ midline/ mainly to right); 2. lesion morphology (round/lobular/conforming); 3. circumscription (well defined/ill-defined); 4. margin (regular/irregular); 5. subjective attenuation pre- and post-contrast (homogeneous/heterogeneous); 6. presence of foci of fat attenuation within the mass (yes/no); 7. presence of calcification within or around the mass (yes/no); 8. presence of lymphadenopathy (slight/marked); 9. displacement of adjacent structures (i.e. mass effect) (yes/no); 10. cranial vena cava enveloped by the mass (yes/no); 11. degree of vena cava compression (none/partial/complete occlusion); 12. Signs of invasion of adjacent structures (vascular, pleural, pericardial, pulmonary or thoracic wall); 13. presence of intrathoracic venous collateral vessels (yes/no); and 14. presence of pleural fluid (yes/no).

 Objective measurements made in transverse CT images were: 1. mass height and width in millimeters (measured at the level of greatest transverse area), and the relative height and width of the mass at the same level (mass height and width divided by the internal height and width of the thorax in the same image, result expressed as a percentage); 2. attenuation value in a polygonal region of interest (ROI) fitted manually to the margin of the mass with the ROI placed at the same position in pre- and post-contrast images and the mean Hounsfield units (HU) and standard deviation (SD) recorded. This ROI was measured on a single slice considered to be representative of the mass as a whole.

Statistical testing (IBM SPSS Statistics Version 24) was used (CL) to test the null hypothesis that there were no differences in the signalment of affected dogs, prevalence of clinical signs or CT features of lymphoma and thymic epithelial neoplasms using binary logistic regression and results expressed as odds ratio (OR) and 95% confidence interval (CI). Differences with p<0.05 were considered significant. A receiver-operating characteristic (ROC) plot was used to test the diagnostic accuracy of Hounsfield unit (HU) measurements to distinguish between lymphoma and thymic neoplasia.

**Results**

Records of 62 dogs that satisfied all the inclusion criteria were found. One dog had no pre-contrast CT images available but was retained because all features remained measurable except pre-contrast subjective attenuation and HU measurement. Post-contrast images were available for all dogs. Of 62 dogs, 33 had lymphoma and 29 had thymic epithelial neoplasia, including 24 thymoma and 5 thymic carcinoma. Diagnosis of lymphoma was based on histology in 9 dogs and cytology in 24; diagnosis of thymic epithelial neoplasia was based on histology in 24 dogs and cytology in 5.

The contribution of cases is presented in table 1. (Insert table 1)

Signalment of dogs and their clinical signs are summarized in table 2. Dogs with thymic epithelial neoplasia were significantly older than those with lymphoma (median age 8.6y versus 6.0y, p=0.007), but there were no significant differences in prevalence of clinical signs or hypercalcemia. A wide range of breeds were present in each group. The 33 dogs in the lymphoma group comprised Labrador retriever (6), cross breed (6), boxer (5), English springer spaniel (3), cocker spaniel (2), dogue de Bordeaux (2) and one each of border collie, Leonberger, American bulldog, golden retriever, Staffordshire bull terrier, Australian sheepdog, Newfoundland, St. Bernard, Cavalier King Charles spaniel. The 29 dogs in the thymic neoplasia group comprised Labrador retriever (13), cross breed (3), German shepherd (4), Jack Russell terrier (2) and one each of beagle, English springer spaniel, golden retriever, Cavalier King Charles spaniel, Siberian husky, Irish terrier, and a shih tzu.

(Insert table 2)

(Insert table 3)

Subjective CT features are summarized in table 3. On the basis of logistic regression analysis, diagnosis of thymic epithelial neoplasia was associated with heterogeneous attenuation in pre- (OR 23.3, CI 4.5-121.1) and post-contrast (OR 30.7, CI 3.6-265.0) CT images. Conversely, envelopment of the cranial vena cava by the mass was less likely with thymic epithelial neoplasia than lymphoma (OR 0.07, CI 0.007-0.66). (figure 1A). There were no other significant differences between the lymphoma and thymic epithelial neoplasia groups in morphology, prevalence of calcification, mediastinal lymphadenopathy, cranial vena cava invasion, collateral vessels, foci of fat attenuation, or pleural fluid.

(Insert table 4)

Results of quantitative CT measurements are summarized in table 4. A greater standard deviation (SD) of HU values in pre- and post-contrast CT images were associated with thymic epithelial neoplasia (SD pre-contrast p=0.005, SD post-contrast p=0.001). As standard deviation is a measure of variation of the data, a larger standard deviation over a ROI indicates heterogeneity. Based on ROC analysis, a SD >17HU in post-contrast CT images had sensitivity 72% and specificity 79% for the diagnosis of thymic epithelial neoplasia (figure 2). Absolute and relative mass size were not significantly different between the two types of neoplasia.

(insert figure 1)

(insert figure 2)

**Discussion**

The thymic epithelial masses were subjectively and objectively more heterogeneous in pre and post contrast images when compared to cranial mediastinal lymphomas. This finding likely reflects the presence of cystic fluid-containing cavities, which have been observed previously in thymomas,11 and possibly an adipose component, although this was only observed in CT images of 4 dogs in the present study: all were thymic epithelial neoplasia cases. The invasion or infiltration of fat by the tumour is included as a sign of malignancy in humans and is one of the criteria used to indicate more advanced disease in the human staging systems for thymoma.17 However, without dissection it is unclear if the fat observed within thymomas in the present study is mediastinal fat that has been enveloped or invaded by the tumor.17

A limitation of the present study is the use of CT images obtained by several different scanners, although, this would not be expected to complicate interpretation of subjective CT features, the measured attenuation (HU) for a given material can vary substantially between CT scanners18,19 because of differences in kVp, slice thickness, pitch, and reconstruction algorithm. This effect will have introduced additional variability in our data that will tend to diminish the observed differences in attenuation between cranial mediastinal lymphomas and thymic epithelial neoplasms; however, it was not sufficient to obscure the significantly greater heterogeneity of thymic epithelial neoplasms, which was evident based on both subjective and objective assessments, and all CT machines were regularly calibrated. Also, due to the multicenter retrospective nature of the study, the cytologic or histologic diagnosis would have been made by various pathologists, although this is not considered a major limitation. Similarly, the sedation or anesthesia protocol differed between dogs, but is considered unlikely to have had a significant effect on any of the evaluated CT features.

Among thymic epithelial neoplasms in dogs, thymoma is the most common and thymic carcinoma relatively less frequently observed. In the present series, thymic epithelial neoplasms were classified as either thymoma or thymic carcinoma; however, insufficient numbers were included to allow any meaningful assessment of differences in CT features that might reflect malignancy. The authors’ opinion is that, in addition to possible invasion of mediastinal fat noted above; poorly defined borders, lymphadenopathy, and the presence of pleural nodules may all be more likely to be present in dogs with higher-grade thymic neoplasia. Canine mediastinal lymphoma is most commonly associated with a T-cell phenotype, however, the retrospective nature of this study meant that lymphoma subtypes were not available for all cases. Therefore, for the evaluation of CT features, lymphoma cases were considered as one group.

Envelopment of the cranial vena cava was a CT finding significantly associated with lymphoma in the present series. This appearance could reflect tumor infiltration of multiple mediastinal nodes that become confluent around the mediastinal vessels. Intraluminal tissue compatible with vascular invasion was observed in only one dog with thymic epithelial neoplasia in the present study. CT assessment of cranial vena cava invasion by mediastinal neoplasms has been considered inaccurate by previous authors7,16 with both false positives7 and false negatives 7,16 reported. In contrast, invasion of the caudal vena cava by adrenal neoplasms was accurately detected by CT in a recent study.20 Identifying vascular invasion is most difficult when there is wall invasion but not yet a tumour thrombus, or when the vessel is markedly compressed by the mass and the lumen is difficult to visualise.

It was anticipated that mediastinal lymphadenopathy would be more frequently observed in dogs with lymphoma; however, no significant difference was found. In dogs with thymic epithelial neoplasia it is uncertain if lymphadenopathy represented a reactive change or local metastasis, as the nodes were not independently sampled. Only thoracic CT images were reviewed for the present study, hence the possibility of generalized lymphadenopathy was not assessed; this finding would have supported a diagnosis of lymphoma, but would also have potentially biased the observers.

In humans, an off-midline location of a mediastinal mass has been considered a sign of thymoma.13 In the present study, left of midline was the most common location for both mediastinal lymphomas and thymic epithelial neoplasms. This likely reflects the fact that the ventral part of the cranial mediastinum differs from human anatomy and lies to the left of midline in most dogs due to the position of the cranioventral part of the right cranial lung lobe extending across the midline,21 although a few examples were observed of mediastinal masses entirely to the right of midline. The large size of the majority of mediastinal masses will tend to hide any initial signs of lateralization of the mass.

As previously reported,1 we found that the median age of dogs with thymic epithelial neoplasia was greater than those with lymphoma.Labrador retrievers were the most frequently affected breed in both the lymphoma and thymoma groups. Their prevalence in the thymic tumor group corresponds with previous reports that thymoma occurs more frequently in Labrador retrievers,5,6,12 although the low numbers of dogs in the present study do not provide strong support for a breed predisposition, and Labrador retrievers are a common breed in the UK.

The presenting clinical signs observed in the present study were mainly non-specific and correspond with those listed in previous reports.1,6,12 No significant differences in prevalence of clinical signs between the groups was observed. Three dogs in each group had regurgitation, likely due to compression of the esophagus or, possibly, as a paraneoplastic syndrome (myasthenia gravis) in the dogs with thymoma. Myasthenia gravis occurs due to the production of autoantibodies to nicotinic acetylcholine receptors at the neuromuscular junction, with the assumption that autoreactive T-lymphocytes produced by thymic neoplasms stimulate antibody production against the neuromuscular antigens.22,23 Myasthenia gravis causing megaesophagus has been reported in up to 40% of dogs with thymoma.1,8 All dogs in the present study were sedated or anaesthetized for CT, hence assessment of megaesophagus was not possible.

In the present study, diagnosis was based on cytology in a greater proportion of the dogs with lymphoma than with thymic epithelial neoplasms. This likely reflects use of cytology as the first choice for tissue sampling when lymphoma is suspected because in cases where cytology enables confident diagnosis of lymphoma, no further sampling may be necessary. If necessary, additional supporting evidence for diagnosis of lymphoma may be obtained from the cytology sample, including flow cytometry or PCR for antigen receptor rearrangements. In dogs with thymoma, where the tumor is composed of variable proportions of neoplastic thymic epithelial cells and lymphocytes, cytology less often enables thymoma to be distinguished from a well-differentiated lymphoma.2 Inconclusive cytology results are an indication for needle biopsy or surgical biopsy, hence more dogs with thymic epithelial neoplasia had histology. While histology might be considered preferable as an inclusion criterion in the present study, cytology and histology of mediastinal masses in dogs have relatively good agreement12,24 and obtaining histological samples may not be as strongly indicated when a confident cytological diagnosis has been obtained. As a result, reliance on cytology is not considered an important limitation of the present study.

**Conclusions**

Thymic epithelial neoplasms tended to occur in older dogs and were more heterogeneous in CT images than cranial mediastinal lymphoma, probably because of fluid-containing cavities and an adipose component. Cranial mediastinal lymphomas were more homogeneous and were more likely to envelop the cranial vena cava. These findings may be useful for guiding case management, particularly when cytologic or histologic results are inconclusive.

**List of Author Contributions**

Category 1

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Category 2

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Category 3

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**Conflicts of interest**

The authors’ have declared no conflict of interest.

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Table 1. Contribution of cases from each institution.

 Lymphoma Thymoma Thymic

 Carcinoma

The Royal Veterinary College, Univeristy of London 11 9 4

Institute of Veterinary Science, University of Liverpool 10 12 1

Langford Vets, University of Bristol 12 3 0

Table 2. Signalment of dogs and their clinical signs

 Lymphoma Thymic epithelial

 neoplasia p-value

 (n=33) (n=29)

Median age (y) (range) 6.0 (1 - 12 years) 8.6 (6 – 12 years) p=0.007

Males : females 21 (64%) : 12 (36%) 14 (48%) : 15 (52%) NS

Clinical signs

 Lethargy 13 (39%) 11 (38%) NS

 Dyspnea or tachypnea 12 (36%) 7 (24%) NS

 Cough 10 (30%) 4 (14%) NS

 Inappetence or anorexia 8 (24%) 5 (17%) NS

 Polydipsia/polyuria 7 (21%) 9 (31%) NS

 Weight loss 5 (15%) 4 (14%) NS

 Vomiting 5 (15%) 4 (14%) NS

 Regurgitation 3 (9%) 3 (10%) NS

 Diarrhea 3 (9%) 1 (3%) NS

 Caval syndrome 2 (6%) 2 (7%) NS

 Seizures 1 (3%) 0 NS

 Weakness 0 3 (10%) NS

Hypercalcemia 4 (12%) 6 (21%) NS

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NS, Not significant

Table 3. Prevalence of subjective CT signs in dogs with lymphoma and thymic neoplasia

 Lymphoma Thymic epithelial OR (95% CI)

 neoplasia

 (n=33) (n=29)

Location of mass:

 Centered on midline 18 11 NS

 Mainly to left 13 13 NS

 Mainly to right 2 5 NS

Morphology:

 Rounded 12 8 NS

 Lobular 14 5 NS

 Mass conforms to thoracic wall 7 16 NS

Circumscription:

 Well circumscribed 29 27 NS

 Poorly defined 4 2 NS

Margination:

 Regular border 31 23 NS

 Irregular border 2 6 NS

Subjective attenuation pre-contrast:

 Homogeneous 20 4

 Heterogeneous 12 25 23.3 (4.5-121.1)

Subjective attenuation post-contrast:

 Homogeneous 15 1

 Heterogeneous 18 28 30.7 (3.6-265.0)

Fat within mass 0 4 NS

Calcification within mass 2 3 NS

Thoracic lymphadenopathy 28 20 NS

Displacement of adjacent structures 24 23 NS

Cranial vena cava enveloped by mass 9 1 0.07 (0.007-0.66)

Cranial vena cava compressed by mass 26 21 NS

Invasion of adjacent structures:

 Cranial vena cava 0 1 NS

 Lung 2 0 NS

 Pleura 0 4 NS

Intra-thoracic venous collaterals 14 17 NS

Pleural fluid 19 15 NS

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OR, odds ratio for diagnosis of thymic neoplasia are specified for results where p<0.05

NS, Not significant

Table 4. Results of objective CT measurements in dogs with lymphoma and thymic epithelial neoplasia

 Lymphoma Thymic neoplasia p-value

 (n=33) (n=29)

Mass dimensions relative to thorax

 Median height (%) (range) 78% (47-100%) 88% (24-100%) NS

 Median width (%) (range) 73% (45-100%) 87% (26-100%) NS

Attenuation of mass

 Pre-contrast (mean HU) (SD) 37 (12.8) 42 (15.1) 0.005

 Post-contrast (mean HU) (SD) 57 (14.9) 64 (20.5) 0.001

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NS, Not significant

**Legend**

Figure 1: A, mediastinal lymphoma: post contrast transverse thoracic CT image demonstrating a homogeneous mass conforming around the vena cava (VC), the brachycephalic trunk (B) and left subclavian artery (S). B, thymic epithelial neoplasia (thymoma): post contrast transverse thoracic CT image demonstrating fluid attenuating pockets within the mass (#), and moderate volume pleural fluid (\*). C, Thymoma: post-contrast transverse thoracic CT image with white arrowheads indicating small streaks of fat attenuation within the mass. All images are reconstructed in a soft tissue algorithm with slice thickness 3 mm, matrix 512 x 512, variable field of view depending on patient size, window width 400, window level 50.

Figure 2. Receiver-operating characteristic (ROC) curve plot of the standard deviation of masses in post-contrast CT images as a diagnostic test for thymoma. Area under curve is 0.80 (p<0.001).

**Appendix:**

CT imaging equipment.

Institution CT scanner model

Institute of Veterinary Siemens Somatom Volume Zoom, Siemens Medical GmBH,

Science, University of Erlangen, Germany

Liverpool GE Medical Systems LightSpeed Plus, GE Medical Systems, Milwaukee, WI

Langford Vets, University Siemens Somatom Emotion 16, Siemens Medical GmBH,

of Bristol Erlangen, Germany

Royal Veterinary College, Philips MX-8000 IDT 16, Philips Medical Systems, 5680 DA

University of London Best, The Netherlands

 GE Medical Systems LightSpeed Pro 16, GE Medical Systems, Milwaukee, WI