ORIGINAL ARTICLE

Computed tomographic staging of dogs with anal sac adenocarcinoma

D. R. Sutton*,[1], T. Hernon*, M. J. Hezzell[†], L. B. Meakin*, S. M. Gould*, K. J. Bradley[†] and A. C. Major*

*Langford Vets, Langford House, Langford BS40 5DU, UK
[†]Bristol Veterinary School, University of Bristol, Langford House, Langford, BS40 5DU, UK

[1]Corresponding author email: ds8203@bristol.ac.uk

Objectives: To describe the CT appearance of anal sac adenocarcinoma lesions in a population of dogs including the relations between primary tumour, and locoregional and distant metastasis.

Materials and Methods: Retrospective review of dogs with confirmed anal sac adenocarcinoma and available CT images of the thorax, abdomen and pelvis.

Results: A population of 70 dogs were included in the study. No association was found between anal sac mass size and presence or absence of iliosacral lymph node enlargement. The prevalence of local metastatic disease characterised by iliosacral lymphadenomegaly in this study was 71%, with pulmonary metastases identified in 11% of cases. There were no cases of distant pulmonary metastasis without concurrent locoregional lymphadenomegaly.

Clinical Significance: In our population of dogs local metastatic spread of anal sac adenocarcinoma was common, with a relatively low prevalence of pulmonary metastasis. The study demonstrates the importance of thorough rectal examination and/or imaging to assess the iliosacral lymph centre in this disease irrespective of the size of the anal sac mass.

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INTRODUCTION

Canine anal sac adenocarcinoma (ASAC) is a neoplastic disease affecting the apocrine glands of the walls of the anal sacs (Withrow *et al.* 2012). It is the most common malignant neoplasia of the perianal region accounting for 17% of these and 2% of skin tumours overall (Withrow *et al.* 2012). Affected dogs tend to be geriatric with a median age of 10 to 11 years. No clear sex predilection has been established (Goldschmidt & Zoltowski 1981, Meuten *et al.* 1981, Williams *et al.* 2003, Potanas *et al.* 2015). Cocker spaniels are significantly overrepresented in British studies, followed by other spaniel breeds and German shepherd dogs (Goldschmidt & Zoltowski 1981, Williams *et al.* 2003, Polton 2006, Polton *et al.* 2006). However, in a study by Williams *et al.* (2003), based in the USA, only six cocker spaniels were included out of a total of 113 dogs.

ASAC is often locally invasive and a wide range of metastasis rates are reported (36-96%; Ross *et al.* 1991, Bennett *et al.* 2002, Emms 2002, Williams *et al.* 2003). The most common site of metastatic spread is the iliosacral lymph centre (commonly referred to as the sub-lumbar lymph nodes) which is made up of the paired medial iliac (MILN) and internal iliac lymph nodes (IILN – sometimes referred to as hypogastric lymph nodes) and the sacral lymph node. The right and left medial iliac lymph nodes (R and L MILN) are consistently positioned in relation to the caudal abdominal vasculature, between the respective deep circumflex iliac and external iliac arteries in a crease created by the psoas major muscle, the aorta and the caudal vena cava. They usually lie ventral to the bodies of the fifth and sixth lumbar vertebrae. In most cases a single left and right node is present, but occasionally two can be present on one or both sides. The right and left internal iliac lymph nodes (R and L IILN) lie caudal to the medial iliac lymph nodes situated ventral to sixth and seventh lumbar vertebrae between the right and left internal iliac and the median sacral artery. Again, these are usually single nodes, but can be multiple. Finally, the sacral lymph node or nodes, which are only present in around 50% of dogs, lie further caudally again, ventral to the sacrum and caudal vertebrae, and closely associated with the median sacral artery (Evans & Lahunta 2012).

Fifty to eighty percent of dogs are found to have disease extending beyond the primary site at the time of initial diagnosis of ASAC with the medial iliac lymph nodes being the most common site of confirmed metastatic disease (Goldschmidt & Zoltowski 1981, Ross *et al.* 1991, Bennett *et al.* 2002,

Emms 2002, Williams *et al.* 2003, Withrow *et al.* 2012). Distant/disseminated metastatic disease is also reported and tends to occur at a later stage in the disease process. Site of distant metastasis can include the abdominal organs, lungs and bones (Linden *et al.* 2018).

Screening for metastatic disease in canine patients with ASAC is typically performed using a combination of physical examination, including rectal palpation, and diagnostic imaging. The normal CT appearance of the abdominal lymph nodes (LN) has been previously described (Beukers *et al.* 2013). CT is considered superior to ultrasound for overall assessment of the iliosacral lymph centre, particularly its more caudal components, as an ultrasonographic window for the internal iliac and sacral lymph nodes is not always available (Beukers *et al.* 2013, Palladino *et al.* 2016). CT has been found to have a higher sensitivity than abdominal ultrasound in detecting abdominal lymphadenopathy during routine anal and rectal cancer screenings in human medicine (Muller *et al.* 1993, Otto *et al.* 2009, Samee & Selvasekar 2011, Linden *et al.* 2018).

The OvidMEDLINE(R) database was searched with the keywords anal, canidae, canine, dog, gland, perianal and sac on 15.11.19; the following textbooks have been consulted (Withrow & MacEwen's Small Animal Clinical Oncology). No previous reports of UK-based CT descriptions of anal sac adenocarcinoma staging with case numbers exceeding 50, have been found via these searches.

The aims of this study are: (1) to report the CT findings in a large population of dogs with histologically confirmed ASAC, including an assessment of interobserver agreement for presence of iliosacral lymph node enlargement on CT studies, (2) to compare clinical variables, including CT evidence of local lymph node enlargement, between dogs with and without CT evidence of pulmonary metastasis, and (3) to compare clinical variables between dogs with and without CT evidence of local lymph node enlargement. We hypothesised that pulmonary metastasis would not be identified in dogs without concurrent iliosacral lymphadenomegaly.

MATERIALS AND METHODS

This is a retrospective, cross-sectional study for which ethical approval was granted by the Animal Welfare and Ethical Review Body of the University of Bristol. In order to identify dogs for inclusion in the study the medical database of our institution was searched for dogs that underwent anal sacculectomy between September 2012 and December 2019. The history text was searched using the key words: anal, sac and adenocarcinoma. In a separate search all cases in the database which had undergone CT during the same period were identified and those whose imaging reports included the keyword 'anal' were isolated. To be included in the final study population dogs had to have cytologically or histologically confirmed ASAC from either the primary mass or from a metastatic lymph node and have full thoracic and abdominal CT scans, with pre- and post-contrast images available for assessment. Dogs were excluded if there was any evidence of additional primary neoplasia.

All CT studies were acquired using a Siemens Somatom Emotion 16 (Siemens AG) with helical acquisition parameters as follows: 130 kV, reference 120 mA (CARE Dose 4D), pitch 0.8, rotation time 0.6 seconds, slice thickness 3 mm. Intravenous contrast medium (iopamidol; Niopam Bracco) was administered via a single-barrel power injector system (ACIST Medical Systems Inc.) at a standard dose of 600 mg/kg at a rate of either 2 or 3 mL/s, depending on the size of the patient. Pre-contrast studies were reconstructed in soft tissue and lung algorithms and post-contrast studies in soft tissue algorithms only.

Images were evaluated using dedicated, freely available, DICOM viewing software (Horos v2.0.1, The Horos Project; www.horosproject.org). All available images were evaluated independently by two board-certified radiologists and a third-year diagnostic imaging resident without knowledge of the final clinical outcome of each case. Each observer described the presence or absence of an anal sac mass in each scan. Each of the left and right medial and internal iliac, and the sacral lymph nodes were assigned as enlarged or not enlarged by each observer. If a discrepancy in findings occurred, the images were re-reviewed by the three observers in combination to achieve a consensus. Height, width and length of all visible components of each of the iliosacral lymph nodes that were considered enlarged were recorded by one observer, using multi-planar image reconstruction. All three observers assessed other thoracic and abdominal organs for variations in parenchymal attenuation, size, shape and contrast pattern, and for the presence of nodules with particular focus on the pulmonary parenchyma, liver, spleen and lymph nodes. Where visible the primary ASAC was also measured in three planes. The volume was calculated for the anal sac mass and for any enlarged lymph nodes using the equation for the volume of an ellipsoid ($1/6\ \pi$ abc, where a is the length, b is the height and c is the width).

Statistical analysis was performed using Microsoft Excel (Microsoft Corporation) and IBM SPSS Statistics, v. 27.0 (IBM Corporation). Data were assessed for normality graphically and by use of the Shapiro–Wilk test. Interobserver agreement was reported as frequencies and percentages. The primary outcome of interest was the presence or absence of pulmonary metastasis; the secondary outcome of interest was the presence or absence of iliosacral lymph node enlargement. Dogs were divided into groups, first by CT evidence of pulmonary metastasis (yes/no) and second by CT evidence of iliosacral lymph node enlargement (yes/no). Independent samples t-tests, Mann-Whitney U tests and Fisher's exact tests were used to compare clinical variables [age, bodyweight, breed (cocker spaniel (yes/no)] and CT findings (anal sac mass volume) between groups, as appropriate; for presence of pulmonary metastases (yes/no), iliosacral lymph node enlargement (yes/no) and total lymph node volume were also compared between groups. The correlation between anal sac mass volume and bodyweight was explored using Spearman's correlations. For calculation of 95% confidence intervals for percentages, the expected number of dogs affected by ASAC in the UK during the study period was estimated as follows: reported annual incidence of skin tumours (1437 per 100,000 dogs)×125 (estimated number of dogs in the UK per 10,000)×reported

percentage of skin tumours that are ASAC (2%)×number of years in the study period (7.17)=25,758 dogs (Dobson *et al.* 2002).

RESULTS

The database search revealed 153 dogs with both a CT scan and presumed ASAC all of which were assessed for eligibility. Eighty-three dogs were excluded due to a lack of cytological or histological diagnosis, an incomplete CT scan and/or evidence of other primary neoplasia. Seventy dogs met the inclusion criteria; three dogs had an initial staging CT followed by a post-operative CT, giving a total of 73 CT scans. Of the 70 dogs included in the study, ASAC was confirmed via cytology in 50 (71.4%), histopathology in 55 (78.6%) and both cytology and histopathology in 35 (50%). Cytological assessment confirmed the presence of ASAC metastasis in liver and/or spleen in three dogs (4.3%). Data from the three repeat studies was used for calculation of interobserver agreement but was not included in other image analysis. The study population consisted of 22 different breeds, with cocker spaniels being the most frequently represented (n=17), followed by cross breed (n=12, with two of these being cocker spaniel crosses), Labrador retriever (n=9), German shepherd dog (n=4), springer spaniel (n=3), golden retriever (n=3), Border collie (n=3), cavalier King Charles spaniel (n=3); additional breeds were represented by two or fewer individuals each. The mean [±standard deviation (SD)] age was 9.0±2.0 years. Forty-four dogs were male (two entire, 42 neutered) and 26 were female (eight entire, 18 neutered). The median (minimum, maximum) bodyweight was 23.0 kg (5.0-50.0).

Anal sac adenocarcinomas were identified on the CT scan and measurements acquired in 64 of the 70 dogs (Fig 1). In the remaining six dogs the anal sac mass was either not identifiable or had already been removed before CT. The mass was left sided in 48% [31/64; 95% confidence interval (CI)=35.8-60.2%] and right sided in 52% (33/64; 95% CI=39.8-64.2). Median (minimum, maximum) ASAC volume was 5.826 cm^3 (0.088-154.46). Median (minimum, maximum) largest ASAC dimension was 2.6 cm (0.8-7.8). There was no significant correlation between the volume of the anal sac mass and bodyweight (P=0.063).

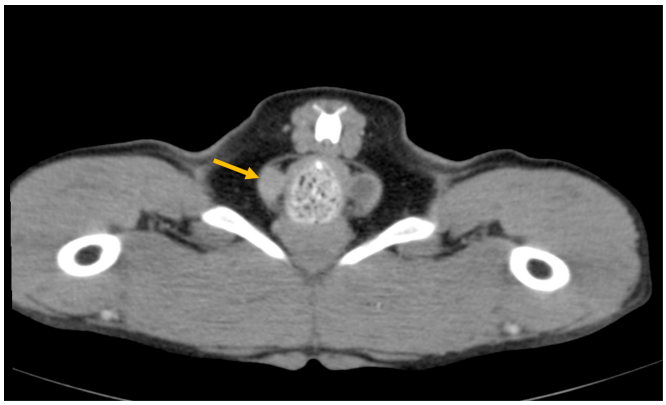

FIG 1. Post-contrast transverse CT image of the anal sacs in a dog with a right sided anal sac adenocarcinoma mass lesion. The lesion is homogeneously contrast enhancing (see yellow arrow)

For each CT, three observers independently assessed the L MILN and R MILN, L IILN and R IILN and the sacral lymph nodes and designated each node as enlarged or not enlarged. When the status of each lymph node was assessed separately (R MILN, L MILN, R IILN, L IILN or sacral LN) all three observers agreed in 78-90% (95% CI=68.5-96.9%) of CT scans (Table 1). Three observers agreed on the combination of lymph nodes judged to be enlarged in 56% (95% CI=44.6-67.4%) of CT scans, suggesting that agreement between observers for the same combination of lymph nodes being enlarged was low (Table 2). When all iliosacral lymph nodes were considered together as a single centre, all three observers agreed in 93.2% (95% CI=87.4-99.0%) of cases when determining whether any nodes within the group were enlarged (Table 2).

On the basis of consensus there was enlargement of at least one lymph node in the iliosacral lymph centre in 50 dogs (71.4%; 95% CI=61.1-81.8%) (Fig 2). Median (minimum-maximum) largest lymph node volume was 16.035 cm^3 (0.17-254.78) Enlargement of the L MILN, R MILN, L IILN, R IILN and sacral lymph nodes was found in 37.1% (95% CI=26.0-48.2%), 25.7% (95% CI=15.7-35.7%), 35.7% (95% CI=24.7-46.7%), 37.1% (95% CI=26.0-48.2%) and 51.4% (95% CI=40.0-62.9%), respectively. Enlargement of the left and/or right MILN was present in 62.9% (95% CI=51.8-74.0%) of dogs and enlargement of the L IILN and/or R IILN was present in 72.9% (95% CI=62.7-83.1%) of dogs. Enlargement of one, two, three, four and five of the iliosacral lymph nodes was found in 30% (95% CI=19.5-40.5%), 18% (95% CI=9.2-26.8%), 28% (95% CI=17.7-38.3%), 14% (95% CI=6.1-22.0%) and 10% (95% CI=3.1-16.9%) of the dogs with lymph node enlargement, respectively. Median (minimum-maximum) of the largest lymph node height was 2.4 cm (0.5-7.7). Median (minimum-maximum) lymph node height for the enlarged L MILN, R MILN, L IILN, R IILN and sacral LNs was 2.1 cm (0.8-5.2), 1.8 cm (1.0-6.9), 1.4 cm (0.4-3.7), 1.9 cm (0.6-7.7) and 1.6 cm (0.4-6.5), respectively. Of the dogs with lymph node enlargement 76% (95% CI=66.2-85.8%) had at least one lymph node with a maximum diameter over 2 cm and 48% (95% CI=36.6-59.4%) had a node with maximum diameter over 4.5 cm (Table 3). No significant differences were detected in any of the clinical or CT

Table 1. Frequency and percentages of observer agreements for enlargement (yes/no) in each individual lymph node

	L MILN frequency [percentage (95% CI for percentage)]	R MILN frequency [percentage (95% CI for percentage)]	L IILN frequency [percentage (95% CI for percentage)]	R IILN frequency [percentage (95% CI for percentage)]	Sacral frequency [percentage (95% CI for percentage)]
Two agree	14 [19% (10.0-28.0%)]	8 [11% (3.8-18.2%)]	8 [11% (3.8-18.2%)]	7 [10% (3.1-16.8%)]	16 [22% (12.5-31.5%)]
Three agree	59 [81% (72.0-90.0%)]	65 [89% (81.8-96.2%)]	65 [89% (81.8-96.2%)]	66 [90% (83.1-96.9%)]	57 [78% (68.5-87.5%)]

L MILN Left medial iliac lymph node, R MILN Right medial iliac lymph node, L IILN Left internal iliac lymph node, R IILN Right internal iliac lymph node, Sacral Sacral lymph node, CI Confidence interval

Table 2. Observer agreements for iliosacral lymph centre enlargement (yes/no) as a combination of individual lymph nodes *versus* as one group

Number of observers in agreement	Frequency [percentage (95% confidence interval)] for the number of observers in agreement for the exact combination of lymph nodes enlarged on each CT	Frequency [percentage (95% confidence interval)] for the number of observers in agreement when assessing if there is ANY iliosacral lymph centre enlargement (yes/no)
Zero agree	11 [15.1% (6.9-23.3%)]	0 [0% (0-0%)]
Two agree	21 [28.8% (18.4-39.2%)]	5 [6.8% (1.0-12.6%)]
Three agree	41 [56.2% (44.8-67.6%)]	68 [93.2% (87.4-99.0%)]

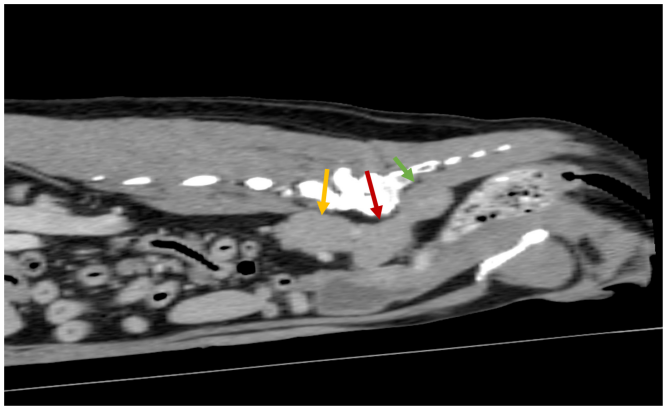

FIG 2. Post-contrast sagittally reconstructed CT image of the right side of the iliosacral lymph centre in the same dog as Fig 1 with moderate enlargement of the medial iliac (yellow arrow), internal iliac (red arrow) and sacral (green arrow) lymph nodes with mild effacement between individual nodes. Note the dramatic enlargement in lymph node size compared to relatively small anal sac mass size in this dog as shown in Fig 1

Table 3. Number of dogs in each stage of disease according to Polton & Brearley (2007) Clinical Stage Scheme Recommendations for Canine Anal Sac Gland Carcinoma

Tumour stage according to Polton & Brearley (2007)	Definition according to Polton & Brearley (2007)			Number of dogs in this study (percentage (95% confidence interval for percentage)
	T	N	M	
1	<2.5 cm	None	None	13 [18.6% (9.7-27.5%)]
2	>2.5 cm	None	None	7 [9.6% (2.9-16.4%)]
3a	Any size	<4.5 cm	None	22 [31.4% (20.8-42.0%)]
3b	Any size	>4.5 cm	None	20 [28.6% (18.3-39.0%)]
4	Any size	Any size	Present	8 [11.4% (4.1-18.7%)]

T Primary tumour status, N Regional lymph node status, M Distant metastasis status

variables assessed between dogs with and without evidence of iliosacral lymph centre enlargement (Table 4).

Lymphadenomegaly was noted affecting the aortic chain lymph nodes in 12 dogs (17.1%), colic lymph nodes in four dogs (5.7%), sternal lymph nodes in four dogs (5.7%), hepatic lymph nodes in two dogs (2.9%) and cranial mesenteric lymph nodes in one dog (1.4%). Liver nodules were present in 15 dogs (21.4%) and splenic nodules in 12 (17.1%).

Pulmonary nodules were identified on CT in eight dogs (11.4%). No significant differences were detected in any of the clinical or CT variables compared between dogs with and without evidence of pulmonary nodules (Table 5). A sample size calculation suggested that if 4% of dogs with pulmonary metastasis are expected to have no LN enlargement, 207 cases would be required in total to achieve 80% power (69 without LN enlargement and 138 with LN enlargement, α=0.05). If 0% of dogs with pulmonary metastasis are expected to have no LN enlargement, 102 cases would be required to achieve 80% power (34 without LN enlargement and 68 with LN enlargement, α=0.05).

DISCUSSION

This study's primary aim was to report the CT findings from a larger population of UK dogs undergoing staging for anal sac adenocarcinoma than has previously been published and to assess the prevalence of pulmonary metastasis with and without local lymph node enlargement on CT. Based on the literature search this is the first time these figures have been reported from CT findings in this number of dogs undergoing staging for ASAC in the UK.

The range of previously reported rates of metastasis in dogs with ASAC is very wide (36-96%) (Ross *et al.* 1991, Bennett *et al.* 2002, Emms 2002, Williams *et al.* 2003). In this study the prevalence of regional LN enlargement was high at 71% (95% CI=60.6-81.4%) compared with the percentage of dogs with pulmonary nodules which was only 11% (95% CI=3.8-18.2%). This is consistent with previously reported rates of local metastasis at the time of initial diagnosis in this disease of between 50% and 80% (Goldschmidt & Zoltowski 1981, Ross *et al.* 1991, Bennett *et al.* 2002, Emms 2002, Williams *et al.* 2003, Withrow *et al.* 2012). Lymph node enlargement was assumed to be consistent with metastatic disease in our study based on the findings of a previous study by Barnes & Demetriou (2017) which found that all dogs with lymph node enlargement on ultrasound had cytological or histopathological evidence of metastasis. According to the literature search, this is the first time the percentage of regional LN enlargement has been reported in more than 50 dogs undergoing CT staging for ASAC in the UK. Hepatic and splenic nodules were identified in 21.4% (95% CI=12.0-30.8%) and 17.1% (95% CI=8.5-25.7%) respectively; however, given the high incidence of nodular hyperplasia and other nodular lesions found in these organs in older dogs it is difficult to draw any conclusions from this in the absence of cytology or histopathology (Bergman 1985, Cuccovillo & Lamb 2002), which we only had available in a minority of cases. Our findings both highlight and confirm the high prevalence of local metastasis in this disease and the relative rarity of metastasis to the lungs which is considered a negative prognostic indicator (Williams *et al.* 2003, Polton & Brearley 2007).

Percentage agreement between observers was low when considering whether each observer deemed the same combination of nodes within the group to be enlarged. There are a number of reasons why this could be the case. There can be difficulty determining which node is which, and even the margination of one node in relation to another, particularly when marked enlargement leads to anatomical distortion (Fig 3), and as previously described

Table 4. Comparisons of clinical variables and CT measurements between dogs with evidence of iliosacral lymph node enlargement on CT and those with no evidence of iliosacral lymph node enlargement

Variable	No evidence of iliosacral lymph node enlargement (n=21)		Evidence of iliosacral lymph node enlargement (n=52)		P value
	Mean±standard deviation/ median (minimum-maximum)	Frequency [percentage (95% confidence interval for percentage)]	Mean±standard deviation/median (minimum-maximum)	Frequency [percentage (95% confidence interval for percentage)]	
Age (years)	8.28±1.68		9.23±2.10		0.220
Bodyweight (kg)	23.0 (5.0-50.0)		22.0 (8.0-47.0)		0.888
Breed [cocker spaniel (yes/no)]		4/17 [19.1% (10.1-28.1%)]		15/37 [28.9% (18.5-39.2%)]	0.557
Anal sac mass volume (cm^3)	3686.14 (599.00-41,385.25)		6327 (87.96-154,459.54)		0.210

Presence of lymph node enlargement was determined based on consensus of three independent observers

Table 5. Comparisons of clinical variables and CT changes between dogs with evidence of pulmonary metastasis on CT and those with no evidence of pulmonary metastasis

Variable	No evidence of pulmonary metastasis (n=64)		Evidence of pulmonary metastasis (n=8)		P value
	Mean±standard deviation/ median (minimum-maximum)	Frequency [percentage (95% confidence interval for percentage)]	Mean±standard deviation/ median (minimum-maximum)	Frequency [percentage (95% confidence interval for percentage)]	
Age (years)	8.96±1.88		9.11±3.00		0.875
Bodyweight (kg)	23.0 (5.0-50.0)		18.0 (13.0-36.0)		0.451
Breed [cocker spaniel (yes/no)]		15/49 [23.4% (13.7-33.1%)]		4/4 [50% (38.6-61.5%)]	0.195
Anal sac mass volume (cm^3)	6267.48 (188.50-154,459.54)		4343.78 (87.96-85,953.98)		0.477
Iliosacral lymph node enlargement (yes/no)		43/21 [67.2% (56.5-78.0%)]		8/0 [100% (100-100%)]	0.095
Total lymph node volume (cm^3)	3675.66 (0.00-603,444.97)		12,361.65 (4460.01-244,227.41)		0.057

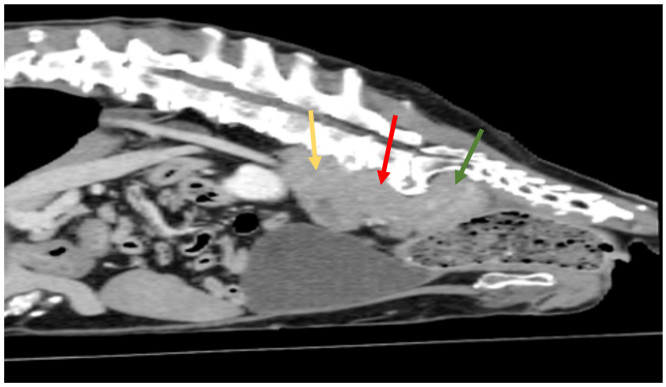

FIG 3. Post-contrast sagittally reconstructed CT image of the iliosacral lymph centre of a dog with marked enlargement of the medial iliac, internal iliac and sacral lymph nodes. Note the loss of the normal anatomical positioning and the effacement of each lymph node border

there can be significant individual anatomic variation in terms of presence/absence and number of nodes between dogs. In a 2013 study by Beukers et al the internal iliac and sacral lymph nodes could not be reliably differentiated from each other even when normal in size. Agreement between observers was far higher when assessing the medial iliac, internal iliac and sacral lymph nodes as one group, *i.e.* determining if any one or more enlarged node was present within the group.

Once consensus between observers was reached the internal iliac lymph node was noted to be the most commonly enlarged [72.9% (95% CI=62.7-83.1%)] of the iliosacral lymph centre followed by the medial iliac [62.9% (95% CI=51.8-74.0%)] and finally the sacral [51.4% (95% CI=40.0-62.9%)]. This is in agreement with a study first published by Linden *et al.* (2018) which studied sentinel lymph node mapping in a small population of dogs with ASAC using lymphoscintigraphy. However, given the aforementioned difficulty in reliably differentiating individual nodes within the iliosacral group the reliability of this is questionable. A clinically more important distinction may be the ability to differentiate enlarged lymph nodes that are intrapelvic, as these are likely to present more of a surgical challenge to remove with an increased risk of complications such as haemorrhage. Of the more distant lymph nodes the aortic chain lymph node was most commonly enlarged [17.1% (95% CI=8.5-25.7%)] followed equally by the colic and sternal [5.7% (95% CI=0.4-11.0%)].

Of the dogs with regional lymph node enlargement, 70% (95% CI=57.6-82.4) had enlargement of at least two lymph nodes in the iliosacral lymph centre and 10% (95% CI=1.9-18.2%) had enlargement of all five iliosacral lymph nodes. Involvement of a greater number of nodes is likely to increase the chance that a more invasive surgical approach would be required making this an additional consideration in treatment planning. Forty percent of the dogs with lymph node enlargement had at least one lymph node with a maximum diameter over 4.5 cm which, according to Polton & Brearley (2007), puts them into clinical stage 3b of the disease (Table 3). Seventy-six percent of the dogs with lymph

node enlargement had at least one lymph node with maximal diameter over 2 cm, which is considered an adequate size for consideration of surgical extirpation at our institution, though surgical decision making is also influenced by other factors such as the presence or absence of clinical signs associated with the lymphadenomegaly. Given the high percentage of dogs affected by regional lymphadenomegaly in our study population and the improvement in survival time afforded by amelioration of clinical signs which can be associated with extirpation of metastatic lymph nodes (Polton & Brearley 2007) we believe our findings highlight the importance of due consideration of the extent of regional metastatic disease in all dogs presenting with this disease, in order to guide treatment and advise owners on prognosis.

In our study population the median diameter of the largest primary tumour dimension was 2.6 cm with a median tumour volume of less than 6 cm^3. With respect to prognosis, this exceeds the tumour size cut off for stage 2 disease of 2.5 cm according to Polton & Brearley (2007). Additionally, according to Schlag *et al.* (2020), tumour size over 2.5 cm has been found to be significantly associated with metastatic disease at presentation. The median lymph node height was 2.4 cm which is below the cut off for stage 4 disease (>4.5 cm) (Polton & Brearley 2007). In concurrence with the secondary aim of our study we noted that the median volume of the largest lymph node in the iliosacral group, at 16.04 cm^3, was relatively large in comparison to the median primary tumour volume (Meuten *et al.* 1981). Additionally, there were cases which demonstrated marked enlargement of one or more of the iliosacral lymph node group, with only relatively small primary anal sac masses (Figs 2 and 3), a phenomenon not previously reported on the basis of our literature search, but one that we feel is anecdotally well recognised. No significant correlation was found between the volume of the anal sac mass and the presence or absence of iliosacral lymph node enlargement. This highlights the importance of a thorough rectal examination and/or diagnostic imaging for staging of these dogs particularly given that the extent of local metastatic disease can be quite marked even on initial presentation.

We hypothesised that pulmonary metastatic disease would not be identified without iliosacral lymph node enlargement on CT. Indeed, none of the dogs in this study that had normal iliosacral lymph nodes had evidence of pulmonary nodules, while all of the eight dogs with pulmonary nodules had lymph node enlargement within the iliosacral lymph centre. Although there was no difference in the proportions of dogs in each of these groups, this is likely to reflect the small number of dogs with pulmonary nodules in the present study. Our sample size calculations suggest that at least 207 cases would be required in a future prospective study to fully investigate this hypothesis.

If the iliosacral lymph nodes act as a sentinel for distant metastasis this could help guide diagnostic staging options. While we believe CT represents the diagnostic gold standard for assessment of iliosacral lymph node enlargement in these cases it would be extremely valuable to compare the usefulness of ultrasound of the illiosacral lymph centre in combination with rectal examination with CT, particularly in identification of dogs which could benefit from surgery in a first opinion setting, and those for which referral may represent a more appropriate option. Further work on this comparison is ongoing at our institute. Based on our available data there is some indication that while imaging of the thorax is an important part of disease staging in dogs with enlarged iliosacral lymph nodes, it might not be necessary in cases where the local lymph nodes are normal. This could have several potential benefits; it would negate the need for CT availability and the chemical restraint required for immobilisation during scanning and also likely reduce the financial cost of staging.

The present study has a number of limitations. First, the retrospective nature of this study may have led to inconsistencies and lack of availability of some data. This also meant that there was not always cytological or histological confirmation of metastatic disease in the local lymph nodes and only rarely cytological or histological confirmation of metastatic disease at distant sites, thus some of the above findings are based on assumptions due to imaging findings alone. However, as previously discussed lymph node enlargement has been found to be highly associated with metastatic disease in dogs with anal sac adenocarcinoma (Barnes & Demetriou 2017). Second, the study data was all obtained from a single referral centre and therefore may not be representative of the general population of dogs with ASAC. The fact that all dogs underwent CT examination will also have added bias because the patients selected are those that are amenable for surgical intervention and without financial constraint. Finally, calculating anal sac mass and lymph node volumes with an ellipsoid mathematic model using linear measurements can overestimate the actual volume of the lesion (Leffler *et al.* 2018).

In conclusion this is the largest UK-based study reporting the CT findings in dogs undergoing staging for anal sac adenocarcinoma. This study emphasises the need for a thorough rectal examination and/or diagnostic imaging due to the common occurrence of extensive local metastatic disease in this condition regardless of tumour size. It also highlights the relatively low incidence of distant metastatic disease to the lungs. This study data will provide the basis for future prospective research at our institution to devise recommendations for appropriate clinical management of these cases in both a first opinion and referral setting.

Conflict of interest

No conflicts of interest have been declared.

References

Barnes, D. C. & Demetriou, J. L. (2017) Surgical management of primary, metastatic and recurrent anal sac adenocarcinoma in the dog: 52 cases. *Journal of Small Animal Practice* **58**, 263-268

Bennett, P. F., DeNicola, D. B., Bonney, P., *et al*. (2002) Canine anal sac adenocarcinomas: clinical presentation and response to therapy. *Journal of Veterinary Internal Medicine* **16**, 100-104

Bergman, J. R. (1985) Nodular hyperplasia in the liver of the dog: an association with changes in the Ito cell population. *Veterinary Pathology* **22**, 427-438

Beukers, M., Grosso, F. V. & Voorhout, G. (2013) Computed tomographic characteristics of presumed normal canine abdominal lymph nodes. *Veterinary Radiology & Ultrasound* **54**, 610-617

Cuccovillo, A. & Lamb, C. R. (2002) Cellular features of sonographic target lesions of the liver and spleen in 21 dogs and a cat. *Veterinary Radiology & Ultrasound* **43**, 275-278

Dobson, J. M., Samual, S., Milstein, H., *et al*. (2002) Canine neoplasia in the UK: estimates of incidence rates from a population of insured dogs. *Journal of Small Animal Practice* **43**, 240-246

Emms, S. G. (2002) Anal sac tumours of the dog and their response to cytoreductive surgery and chemotherapy. *Australian Veterinary Journal* **83**, 340-343

Evans, H. E. & Lahunta, A. (2012) Regional anatomy of the lymphatic system. In: Miller's Anatomy of the Dog. 4th edn. W. B. Saunders, Philadelphia. pp 551-552

Goldschmidt, M. H. & Zoltowski, C. (1981) Anal sac gland adenocarcinoma in the dog: 14 cases. *Journal of Small Animal Practice* **22**, 119-128

Leffler, A. J., Hostnik, E. T., Warry, E. E., *et al*. (2018) Canine urinary bladder transitional cell carcinoma tumor volume is dependent on imaging modality and measurement technique. *Veterinary Radiology & Ultrasound* **59**, 767-776

Linden, D. S., Cole, R., Tillson, D. M., *et al*. (2018) Sentinel lymph node mapping of the canine anal sac using lymphoscintigraphy: a pilot study. *Veterinary Radiology & Ultrasound* **60**, 346-350

Meuten, D. J., Cooper, B. J., Capen, C. C., *et al*. (1981) Hypercalcemia associated with an adenocarcinoma derived from the apocrine glands of the anal sac. *Veterinary Pathology* **18**, 454-471

Muller, M. F., Krestin, G. P. & Willi, U. V. (1993) Abdominal tumors in children. A comparison between magnetic resonance tomography (MRT) and ultrasonography. *RöFo: Fortschritte auf dem Gebiete der Röntgenstrahlen und der neuen bildgebenden Verfahren* **158**, 9-14

Otto, S. D., Lee, L., Buhr, H. J., *et al*. (2009) Staging anal cancer: prospective comparison of transanal endoscopic ultrasound and magnetic resonance imaging. *Journal of Gastrointestinal Surgery* **13**, 1292-1298

Palladino, S., Keyerleber, M. A., King, R. G., *et al*. (2016) Utility of computed tomography *versus* abdominal ultrasound examination to identify Iliosacral lymphadenomegaly in dogs with apocrine gland adenocarcinoma of the anal sac. *Journal of Veterinary Internal Medicine* **6**, 1858-1863

Polton, G. A. (2006) Examining the risk of anal sac gland carcinoma in cocker spaniels. *Journal of Small Animal Practice* **47**, 557

Polton, G. A., Mowat, V., Lee, H. C., *et al*. (2006) Breed, gender and neutering status of British dogs with anal sac gland carcinoma. *Veterinary and Comparative Oncology* **4**, 125-131

Polton, G. A. & Brearley, M. J. (2007) Clinical stage, therapy, and prognosis in canine anal sac carcinoma. *Journal of Veterinary Internal Medicine* **21**, 274-280

Potanas, C. P., Padgett, S. & Gamblin, R. M. (2015) Surgical excision of anal sac apocrine gland adenocarcinomas with and without adjunctive chemotherapy in dogs: 42 cases (2005-2011). *Journal of the American Veterinary Medical Association* **246**, 877-884

Ross, J. T., Scavelli, T. D., Matthiesen, D. T., *et al*. (1991) Adenocarcinoma of the apocrine glands of the anal sac in dogs – a review of 32 cases. *Journal of the American Animal Hospital Association* **27**, 349-355

Samee, A. & Selvasekar, C. R. (2011) Current trends in staging rectal cancer. *World Journal of Gastroenterology* **17**, 828-834

Schlag, A. N., Johnson, T., Vinayak, A., *et al*. (2020) Comparison of methods to determine primary tumour size in canine apocrine gland anal sac adenocarcinoma. *Journal of Small Animal Practice* **61**, 185-189

Williams, L. E., Gliatto, J. M., Dodge, R. K., *et al*. (2003) Carcinoma of the apocrine glands of the anal sac in dogs: 113 cases (1985–1995). *Journal of the American Veterinary Medical Association* **223**, 825-831

Withrow, S. J., Vail, D. M. & Page, R. L. (2012) Perianal umours. In: Withrow & MacEwen's Small Animal Clinical Oncology. 5th edn. Eds S. J. Withrow, D. M. Vail and R. L. Page. Saunders Elsevier, St. Louis. pp 423-430