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Clinical characteristics and outcome of dogs with presumed primary renal lymphoma

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Abstract

Objectives: To characterise the presentation, clinicopathologic data and outcome of 29 dogs with presumed primary renal lymphoma.
Methods: Medical records of dogs with suspected primary renal lymphoma from 11 institutions were assessed retrospectively.
Results: All dogs were substage b, and lethargy and gastrointestinal signs were common presenting complaints, as were azotaemia (n=25; 86%) and erythrocytosis (n=15; 51%) on biochemical testing. Ultrasonography typically revealed bilateral renal lesions (n=23; 79%), renomegaly (n=22; 76%) and abdominal lymphadenopathy (n=14; 48%). Chemotherapy was the only treatment in 23 dogs of which 11 responded, all considered partial response. For all dogs the median progression-free survival and median overall survival times were 10 days (range: 1 to 126) and 12 days (range: 1 to 212), respectively, and for dogs that responded to chemotherapy 41 days (range: 10 to 126) and 47 days (range: 10 to 212), respectively. **Clinical Significance**: Primary renal lymphoma in dogs appears to be associated with a poor prognosis and short-lived response to chemotherapy.

Keywords
canine, chemotherapy, extranodal, lymphoma, renal

Introduction

Malignant lymphoma comprises 90% of all canine haematopoietic cancers. Diffuse large B-cell lymphoma is most common and for which chemotherapy can result in high response rates with median survival times of 7–11 months (Davies *et al.* 2017). The high grade T-cell form may carry a worse prognosis although newer protocols rich in alkylating agents have demonstrated comparable survival times (Brown *et al.* 2018). However, extranodal lymphoma such as hepatic, hepatosplenic and gastrointestinal are less frequently described in the dog; these are mainly T-cell tumours and often associated with poor outcome (Frank *et al.* 2007, Keller *et al.* 2013).

Renal involvement in canine multi-centric lymphoma has been previously recognised, typically at an advanced tumour stage (Breshears *et al.* 2011). The prevalence of primary

renal lymphoma (PRL), in which the neoplasia originates from the kidneys, is unknown in dogs. Literature regarding presumed primary renal lymphoma is sparse and only a small number (<30 cases) have been reported. Most had bilateral involvement and five were T-cell and one was B-cell (Baskin & De Paoli 1977, Batchelor *et al.* 2006, Breshears *et al.* 2011, Cook & Lothrop 1994, Cotchin 1954, Durno *et al.* 2011, Froment & Gara-Boivin 2015, Hilscher 2004, Klein *et al.* 1988, Lane & Lobetti 2002, Lascelles *et al.* 2003, Nelson *et al.* 1983, Osborne *et al.* 1971, Snead 2005, Szatmári *et al.* 2004, Walter *et al.* 1987, Zhao *et al.* 1993). An association with erythrocytosis, as seen with other canine renal neoplasms, and CNS (central nervous system) involvement, was common. Only five cases received chemotherapy, of which one was managed for 11 months. One dog with unilateral disease underwent nephrectomy but died postoperatively of aspiration pneumonia. Full staging was not documented in any dog, but at necropsy four fit the definition of primary renal lymphoma and another had concurrent disease in the central nervous system only. Feline renal lymphoma has typically been described with involvement of other anatomical locations, although as a primary form in 24% of cats. It is mostly of B-cell origin and intermediate to high grade, and up to 40% of affected cases can have the CNS affected. Though chemoresponsive, the median survival time is less than 7 months (Mooney *et al.* 1987, Gabor *et al.* 1998, 1999, Taylor *et al.* 2009).

In humans both Non-Hodgkin lymphoma with renal involvement and primary renal lymphoma are uncommon (Stallone *et al.* 2000, Villa *et al.* 2011). Flank pain, inappetence, weight loss, haematuria, fever and acute renal failure are common as is CNS involvement at diagnosis or relapse (Okuno *et al.* 1995, Saito 1996, Stallone *et al.* 2000, Villa *et al.* 2011). Prognosis is poor, although long-term survivors have also been reported (Okuno *et al.* 1995, Saito 1996). For unilateral disease, extended disease-free intervals can occur following surgical resection, chemotherapy and consolidation radiation therapy (Okuno *et al.* 1995). CHOP protocols are recommended for bilateral disease (Navas Martinez *et al.* 2011, Villa *et al.* 2011).

The purpose of this retrospective study was to gather information from dogs with presumed primary renal lymphoma, characterising the clinical presentation, response to chemotherapy and outcome, and to examine prognostic factors for this rare condition.

Materials and Methods

Inclusion/exclusion criteria

Dogs were included if they had a cytologic or histologic diagnosis of lymphoma obtained from renal samples and tumour burden was largest in the kidney(s). The latter was determined by a board certified veterinary radiologist. Dogs diagnosed at necropsy that had not received cytotoxic treatment but fulfilled the above criteria were included. Cases with suspected or confirmed hepatic, splenic, peritoneal, abdominal or thoracic lymph node involvement were eligible for inclusion if the main clinical presentation was associated with renal infiltration (azotaemia with decreased urine concentration). Dogs with peripheral lymphadenomegaly, lymphoma in peripheral nodes (regardless of size) or a mediastinal mass at diagnosis were excluded.

Case selection and medical record review

Cases were recruited through the American College of Veterinary Internal Medicine Oncology email listserv and by contacting other institutions in the UK. Medical records were retrospectively searched at seven veterinary teaching hospitals (Royal Veterinary College; University of Liverpool; The Ohio State University; Michigan State University; University of Cambridge; Washington State University; Hokkaido University) and four private veterinary referral centres (Veterinary Oncology Consultants; Veterinary Oncology & Hematology Center; Veterinary Specialty Care) from 2003 to 2017. Follow-up information was obtained from records and telephone conversation with referring veterinarians.

Data recorded included signalment, presenting clinical signs and duration, physical exam findings, date of diagnosis and method, lymphoma grade and immunophenotype, diagnostic imaging and laboratory results. Clinicopathological results were classified as normal or abnormal by comparison with established reference ranges for the relevant laboratory. If reference ranges were not listed, dogs were considered to have anaemia if the haematocrit was <37% and erythrocytosis if >55%. The International Renal Interest Society (IRIS) Classification Scheme for Acute Kidney Injury (AKI) was used to categorise dogs using serum creatinine concentration (and clinical parameters, if available): Grade I (≤1.6 mg/dl, <140 µmol/l; non-azotaemic with historical, imaging or laboratory evidence of AKI), Grade II (1.7–2.5 mg/dl, 141–220 µmol/l; mild AKI), Grade III (2.6–5.0 mg/dl, 221–439 µmol/l; moderate to severe AKI), Grade IV (5.1–10.0 mg/dl, 440–880 µmol/l), and Grade V (>10.0 mg/dl, >880 µmol/l; moderate to severe AKI) (Ettinger *et al.* 2017). Urine specific gravity (USG) 1.008 to 1.012 was considered isosthenuric and hyposthenuric when >1.008. A systolic blood pressure ≥160 mmHg was considered hypertensive. Protocols, responses, cause of death, and necropsy results were recorded when available.

Dogs were classified using the World Health Organization (WHO) staging system and further characterised as substage a or b (Owen 1980). Because the kidney constitutes an extranodal form, dogs were also classified according to the Ann Arbor staging system in which stage I is defined as a single extranodal organ; II as for I but also including nodes on the same side of the diaphragm with or without spleen involvement (in the case of renal lymphoma); III including nodes on both sides of the diaphragm; and, IV defined by liver involvement or further dissemination (Carbone *et al.* 1971). Information regarding renal lesions and extent of disease was gathered from imaging reports (CT, thoracic radiography, abdominal ultrasonography or a combination thereof). Abnormalities that were not sampled were presumed to be involved in the disease process based on imaging reports and the opinion of the overseeing clinician. Bone marrow sampling was not routinely performed.

Information on chemotherapy protocols was noted. For comparative purposes, induction protocols were classified as COP (an 8-week protocol utilising cyclophosphamide, vincristine and prednisolone with or without cytarabine on the first day of treatment) or CHOP (19- or 25-week protocols consisting of cyclophosphamide, vincristine, prednisolone and doxorubicin with or without L-asparaginase at induction; epirubicin was allowed as substitution for doxorubicin) (Withrow *et al.* 2013). A protocol was required to proceed for a minimum of 28 days to be classed as either COP or CHOP; dogs that died, were euthanised,

or whose treatment was changed before this, were excluded from statistical comparison of induction protocols.

Response to treatment was determined by re-evaluation visit records and by the combination of the following when assessed a minimum of 10 days later: serum creatinine concentration, clinical signs, physical exam and renal lesions assessed by imaging. Complete response was defined as resolution of clinical signs, azotaemia, normal physical examination and/or no imaging abnormalities. Partial response was defined as a >50% but <100% improvement in one or more of the above variables. No response was defined as <50% improvement in one or more of the above variables (Taylor *et al.* 2009).

Objective response rate was defined as the sum of complete and partial response rates. Dogs that died or were euthanised within 10 days of starting chemotherapy regardless of cause were not considered as responders. Progression-free survival was defined as the time from initiating chemotherapy treatment (a cytotoxic drug, glucocorticoids or L-asparaginase) to disease relapse (after initial improvement), progression or death due to any cause. Overall survival was defined as the time from chemotherapy administration to death or euthanasia for any cause (Morrison-Collister *et al.* 2003). Dogs that remained alive and in remission at the end of the follow-up period or were lost to follow-up were censored from progression-free survival. Dogs that were alive or lost to follow-up at time of writing were censored from overall survival analysis.

Statistical analysis

The following clinical variables were evaluated: age, gender, weight, duration of clinical signs, polyuria/polydipsia, gastrointestinal signs, lethargy, inappetence, weight loss, CNS signs (seizures, cranial or spinal cord neuropathy), fever, erythrocytosis, hyperphosphatemia, hypoalbuminemia, azotaemia, IRIS AKI grade, hypertension, renomegaly, pyelectasia, loss of corticomedullary distinction, unilateral/bilateral renal lesions, Ann Arbor stage, immunophenotype, lymphoma cell size, treatment protocol, addition of cytarabine to induction protocol, addition of L-asparaginase to induction protocol and response to chemotherapy. Data were described and analysed using SPSS version 24 (IBM Corp.). Descriptive parameters were evaluated for all data variables. Categorical data were presented either as percentages or ratios. Continuous data were expressed as median and range. Following statistical description, survival analysis using Kaplan-Meier product-limit method was conducted to estimate overall progression-free interval and overall survival for the dogs treated solely with chemotherapy. Data were assessed for normality by visual plotting and the Shapiro-Wilk test. Univariable and multivariate analysis were not performed due to the small data set.

Results

Signalment and clinical signs

Twenty-nine dogs fulfilled the inclusion criteria. Seven dogs had been included in a study evaluating ultrasonographic features of canine renal lymphoma (Taylor *et al.* 2014). A summary of the clinical and clinicopathologic data is listed in Table 1. The median age was 7 years (range 3–14) and body weight 33 kg (range 9–53). Male to female ratio was 3.1:1. There were 10 Labrador Retrievers, six mixed-breed dogs, two Border Collies, and one each of Doberman, rottweiler, Newfoundland, Norwegian elkhound, Bernese mountain dog, West Highland terrier, Australian shepherd, Dalmatian, beagle, dogue de Bordeaux, and Shetland sheepdog.

Clinical presentation data were available for all dogs. Median duration of clinical signs before presentation was 18 days (range: 2–210). Lethargy (n=2-; 69%), vomiting (n=24; 48%), polyuria/polydipsia (n=12; 41%), decreased appetite (n=11; 38%), weight loss (n=6; 21%), diarrhoea (n=6; 21%), and gross haematuria (n=4; 14%) were common.

On physical examination, renomegaly was suspected in 16 (55%) dogs and 12 (41%) had pain on abdominal palpation. Neurologic signs (n=4; 14%) were comprised of Horner's syndrome and trigeminal neuropathy (n=1; 3%), facial nerve paralysis (n=1; 3%), nystagmus (n=1; 3%), and upper motor neuron signs and hindlimb ataxia (n=1; 3%). Two dogs (7%) were pyrexic. Hypertension (range 160–230 mmHg) was present in 15 of 22 (68%) dogs.

Clinicopathologic findings

Haematological tests revealed mild anaemia (n=4, 14%; range 27–36%) and erythrocytosis (n=15, 51%; range 56–76%). Erythropoietin levels were measured in four cases and were normal in two and above reference interval in two. There were no reports of abnormal circulating lymphocytes on routine haematology or smear (when performed). No dogs had bone marrow sampling performed.

Serum biochemical profile of all dogs revealed elevated creatinine concentration in 25 (86%; range 1.61–9.86 mg/dL), hyperphosphatemia in 14 (48%; range 4.9–11.6 mg/dL), hyperkalemia in 8 (28%; range 2.8–6.8 mEq/L), hypoalbuminemia in 5 (17%; range 2.1–2.7 g/dL), and elevated alkaline phosphatase (ALP) and alanine aminotransferase activity (ALT) in 4 (14%) patients each (range 150–4549 IU/L and 84–885 IU/l, respectively). With the exception of one dog with an intrahepatic thrombus and markedly increased ALP and ALT, elevation of ALP and ALT were mild or moderate. None of the dogs was hypercalcemic. Based on IRIS AKI guidelines, four (14%) were Grade I; ten (34%) Grade II; nine (31%) Grade III; five (17%) Grade IV and one (3%) Grade V.

Urine samples, acquired prior to fluid therapy when possible, were evaluated in all dogs. Findings included normal concentrating ability (n=1; 3%), USG >1.012 and <1.025 (n=15; 52%), isosthenuria (n=11; 38%) and hyposthenuria (n=1; 3%). Urine sediment or dipstick analysis of 27 dogs revealed haematuria (n=18; 67%), proteinuria with inactive sediment (n=4; 15%), bacteriuria (n=4; 15%), pyuria and casts (n=3; 11% each), and atypical lymphocytes (n=1; 4%). Urine culture on four of 17 (24%) samples were positive for bacterial growth.

Diagnostic imaging findings

Three-view thoracic radiographs performed in 25 dogs detected no abnormalities in 19 (76%), and concurrent thoracic CT in one dog was normal. Abnormalities included sternal lymphadenopathy (n=3; 12%), mild pleural effusion (n=1; 4%), tracheobronchial lymphadenopathy (n=1; 4%) and a diffuse peribronchial and interstitial lung pattern and concurrent tracheobronchial lymphadenopathy (n=1; 4%). Bronchoalveolar lavage in the latter dog did not detect evidence of lymphoma. Cytology of an enlarged sternal lymph node in one dog confirmed lymphoma.

Abdominal ultrasonography was performed in all dogs at presentation. Bilateral renal abnormalities were described in 23 (79%) and unilateral lesions in 6 (21%). Lesions included renomegaly (n=22; 76%), pyelectasia (n=21; 72%), loss of corticomedullary distinction (n=17; 59%), subcapsular hypoechoic lesions (n=3; 10%) and diffuse hypoechogenicity (n=6; 21%) or hyperechogenicity (n=2; 7%). Mild renal abnormalities were described in two dogs with erythrocytosis and azotaemia, consisting of bilateral pyelectasia (n=2) and focal unilateral loss of corticomedullary distinction (n=1). Bilateral lymphoma was confirmed in both, one by cytology and biopsy and one at necropsy following deterioration a few days later in which a diffuse renal infiltration without renomegaly was found.

Other lesions reported on abdominal ultrasonography included lymphadenopathy (n=14) including renal (n=4) and other nodes (mesenteric or sublumbar; n=8), both renal and other abdominal nodes (n=2), hepatic abnormalities (n=6) including one additional dog with an intrahepatic portal thrombus, and splenic abnormalities (n=4). Sampling of a solitary 1.7 cm diameter nodule revealed benign hyperplasia, and nodules in the other two dogs and one dog with hepatomegaly were consistent with lymphoma. Splenic cytology for two of three dogs were confirmed as lymphoma. Other ultrasonographic lesions included focal gastrointestinal wall abnormalities (n=4), adrenal enlargement or mass (n=3), peritoneal effusion (n=4), hyperechoic mesentery (n=2), thickened gall bladder wall (n=1), thickened and hypoechoic pancreas in which lymphoma was not found (n=1), and prostatomegaly (n=1). Fine needle aspiration of non-renal lesions confirmed neoplastic infiltration in four of nine liver, three of 11 spleen, one of one peritoneal effusion and eight of 10 abdominal lymph node samples. A cytologically reactive mesenteric node in one dog was confirmed as lymphoma by histopathology.

Abdominal CT of one dog demonstrated heterogeneous contrast enhancement of the kidneys with suspected haemorrhage. A CT/excretory urogram in another dog revealed decreased unilateral contrast uptake and abnormal filling of the pelvis and ureter. No other lesions were reported.

Diagnosis and disease staging

A cytological diagnosis of lymphoma was obtained in 24 of 26 dogs that underwent fine-needle aspiration from the kidney, and in five dogs by biopsy and histopathology, includeing one diagnosed at necropsy 6 days after presentation. Immunotyping resulted in 6 of B-cell

and 10 T-cell origin, diagnosed by immunocytochemistry (n=3), immunohistochemistry (n=7), flow cytometry (n=3), and PARR (n=3) designated six as B-cell and 10 as T-cell tumours. All PARR results demonstrated clonal rearrangement. One additional case was an LGL variant. Neoplastic lymphoid morphology was described in 24/29 cases: large (n=12; T-cell n=5), intermediate-to-large (n=4; B-cell n=1), intermediate (n=3; T-cell n=2), small-to-intermediate (n=3; T-cell n=1) and small (n=2; T-cell n=1).

Based on clinical exam findings and staging tests, all dogs were considered WHO stage Vb (Withrow *et al.* 2013). The Ann Arbor system classified six dogs as stage I, six stage II, three stage III and fourteen stage IV.

Treatment and outcome

Twenty-four dogs were treated; 23 received chemotherapy alone and one dog with a unilateral renal mass and mesenteric lymph node involvement underwent nephrectomy followed by a COP protocol. It was euthanised due to unknown causes at 2046 days, and was excluded from survival analyses because of its multimodal therapy. The five untreated dogs were euthanised at diagnosis (n=2) or 6, 8, and 15 days later.

Of the 23 dogs treated only with chemotherapy, the intention-to-treat protocol was COP (n=12, including induction with cytarabine, n=7; or L-asparaginase, n= 4) and CHOP (n=8, including L-asparaginase at induction, n=4). The intended protocol was not clear for two dogs that received L-asparaginase and prednisone, and one dog vincristine and prednisolone. Twelve dogs (52%) were excluded in response evaluation that were euthanised due to progressive renal disease (n=7, considered non-responders) or died within 1 week of treatment (n=5) due to respiratory arrest, sepsis, biliary obstruction secondary to cholelithiasis, persistent seizures and unknown causes, respectively.

Eleven dogs (48%) responded to chemotherapy, all with a partial response, treated with CHOP (n=5), COP (n=2) or a protocol that could not be defined because duration of therapy was <28 days (n=4). The median progression-free survival was 10 days (1–126) for all dogs treated with chemotherapy and 41 days (10–126) for responders. The median PFS for dogs treated with COP was 8 days (3–126) and 41 days (3–96) for CHOP.

At follow up dogs were euthanased for suspected unrelated causes (n=2, sepsis and thromboembolism, respectively) and progressive disease (n=13). Nine of these died or were euthanased because of renal disease, and four because of CNS signs (presumed lymphoma-related). Various rescue treatments were used in four dogs but responses were short-lived with a median progression-free survival of 12 days (range 1–16).

The median overall survival for dogs treated with chemotherapy was 12 days (range 1–212) with 30% alive one month after treatment initiation and only one alive at 6 months. The median overall survival for dogs that responded to chemotherapy was 47 days (range 10–212). Outcome data are listed in Table 2, and the corresponding survival curve is show in Fig. 1.

Four dogs underwent necropsy of which the diagnosis was achieved at that time in one. Findings included cream-coloured masses that replaced or compressed renal parenchyma and focal haemorrhages (Fig. 2). Microscopically there were neoplastic cells, suppurative inflammation and necrosis, with compression and occasional fibrosis of tubules (Fig. 3). Of the chemotherapy-treated dogs, one had no extra-renal lesions at necropsy three days after receiving chemotherapy, and the other had renal lymphadenopathy and hepatic nodules found by ultrasound but additional lesions (adrenal, mediastinal and mesenteric node) at necropsy 4 days after starting chemotherapy. Of the two untreated dogs, no extra-renal lesions were found on ultrasound or necropsy in one, and ultrasound of the other revealed only renal lesions and a hypoechoic pancreas but involvement of pituitary, adrenal, spleen and liver at necropsy a few days later. None of these dogs had liver or spleen sampled. Thoracic imaging findings were reported as unremarkable in all four, including the dog diagnosed with a mediastinal mass at necropsy that underwent CT. All four exhibited neurological signs before death but CNS involvement was confirmed in only one dog.

Discussion

Our data demonstrate the poor prognosis of dogs with presumed primary renal lymphoma, even with initiation of a multiagent chemotherapy protocol. Despite many (48%) responding to treatment, the median progression-free survival for these was 41 days and none had a complete response. The median overall survival, 47 days, was short.

The reason for these grave outcomes are likely multifactorial. Dogs with an extranodal abdominal form typically present with non-specific signs and diagnosis is more likely at an advanced stage (Couto *et al.* 1989, Dank *et al.* 2011). All dogs were systemically ill at diagnosis and in poor condition, and substage is a known prognostic factor for canine lymphoma (Keller *et al.* 1993). Similarly, the response duration for responders was short and the overall response rate to chemotherapy was substantially low compared to canine peripheral nodal lymphoma (Simon *et al.* 2008). Moreover, there were no complete responses. Finally, T-cell phenotype appeared to predominate, which may have contributed to the poor chemotherapy response observed; intrinsic drug resistance is common with this form and is associated with increased ABCG2 expression (Zandvliet *et al.* 2015).

The study was limited by variable re-evaluation intervals and methods. In addition, we chose to use 10 days as a minimum duration of response rather than 42 days as for peripheral nodal lymphoma (Vail *et al.* 2010) because many dogs in our study were found to respond to chemotherapy despite short survival times. In an effort to avoid understating treatment response in this aggressive form, a shorter duration was evaluated instead, because some dogs had reasonable outcomes.

Most dogs in this study were azotaemic at presentation. However, the AKI grading system, as determined by a single measurement, reflects a moment in the disease course. In addition, the mechanism of renal failure in human primary renal lymphoma is thought to be neoplastic infiltration and compression of the tubular lumen resulting in intrarenal obstruction, leading to tubular atrophy and necrosis (Glicklich *et al.* 1986). The canine necropsies demonstrated these lesions microscopically. As such, the degree of azotaemia

may not be truly reflective of the damage done to the kidneys. Finally, interpretation bias in the face of severe azotaemia may result in reluctance to treat.

In this study, clinical signs, physical exam, imaging and biochemical findings were used to determine response, as standardised response evaluation criteria are not available for canine extra-nodal lymphoma. While data reassessing renal values were not available for all, 5/11 dogs had improved or resolved azotaemia. Similarly, improvement in serum creatinine values is associated with longer survival in human primary renal lymphoma (Glicklich *et al.* 1986). In addition, one dog demonstrated improved clinical signs while ultrasound abnormalities remained unchanged. Sonographic lesions in human primary renal lymphoma may persist despite resolution of symptoms and histopathologic absence of disease (Choi *et al.* 1997).

The longest survivor was a dog with unilateral disease and mesenteric lymph node involvement that lived over five years following nephrectomy and a COAP protocol. The prognosis for human primary renal lymphoma is guarded with conventional chemotherapy alone. However, in unilateral cases with no extra-renal involvement, nephrectomy followed by chemotherapy is recommended, with long-term survivors reported (Okuno *et al.* 1995). This suggests that dogs with unilateral disease and no extra-renal involvement may benefit from nephrectomy.

Thirteen dogs exhibited neurologic signs either at diagnosis or disease progression. Renal involvement of lymphoma is an independent risk factor for CNS relapse in people, possibly due to chemokines, adhesion molecules, and matrix metalloproteinases aiding invasion into the nervous system (Villa *et al.* 2011). Necropsy of only one of four dogs revealed infiltration in the CNS. Five presented with concurrent neurologic signs and erythrocytosis (range 56–73%); erythropoietin levels were normal when measured (n=2). While it is possible that dehydration caused a relative polycythemia, total protein was elevated in only one dog; after fluid therapy, the total protein normalized but erythrocytosis persisted. Blood hyperviscosity can lead to complications such as neurologic signs, cardiac or renal function compromise, weakness, or thromboembolic disease (Ettinger *et al.* 2017). Erythrocytosis, common in this study and comprising seven T-cell, one B-cell, and one LGL type, has been associated with various canine renal neoplasms (Bennett 2004, Cook & Lothrop 1994) in addition to renal lymphoma. Finally, hypertensive encephalopathy can initiate CNS signs through rapid rise in pressure altering cerebrovascular flow (Becker *et al.* 2007, Ettinger *et al.* 2017). Although blood pressure measurements can be falsely elevated in the veterinary hospitals, 9/11 dogs with neurological signs were diagnosed as hypertensive.

Ultrasonography demonstrated renal abnormalities in all dogs. In two, subtle findings were nonspecific and not considered clinically meaningful at the time. Further investigations revealed azotaemia and erythrocytosis in both and lymphoma was diagnosed either at necropsy or when marked imaging abnormalities later prompted sampling. Preservation of renal shape and size is documented in human primary renal lymphoma, indicating an infiltrative rather than expansile growth pattern in some cases (Hartman *et al.* 1988). This suggests that if erythrocytosis or azotaemia is diagnosed but imaging lesions are nonspecific, an infiltrative process such as lymphoma should be a differential diagnosis and renal sampling may be indicated.

There are several limitations of the data presented in the current study. First, the low number of cases – because of the rarity of this disease – limits statistical power to detect prognostic markers. Because dogs with predominant disease in non-renal locations were excluded, those with initial renal localisation and subsequent rapid dissemination may have been omitted (Krol *et al.* 2003). Conversely, those with disease originating elsewhere and then disseminating to the kidneys may have been erroneously included; in this study only six dogs had disease limited to the kidney according to available staging data, though one did not have thoracic imaging and not all had both spleen and liver sampled. However, those with Stage I disease did not have improved or worse outcomes compared to dogs of higher Ann Arbor stage.

Clear guidelines to classify primary renal lymphom *versus* secondary renal lymphoma are lacking in both human and veterinary medicine, but recommendations for staging in humans include advanced thoracoabdominal imaging and bone marrow biopsy (Cheson *et al.* 2014). Primary renal lymphoma cases with disseminated disease have been reported using a broader definition of primary extra-nodal lymphoma that consideres extent of disease (Geetha *et al.* 2014, Jipp *et al.* 2016), although this risks erroneously including cases that have originated elsewhere and subsequently disseminate to the kidney. Evaluation of the impact of alternative definitions for human extra-nodal lymphoma revealed similar patient outcomes regardless of categorisation and, as a result it is advocated to include those with disseminated disease at presentation provided the extranodal component is clinically predominant (Krol *et al.* 2003). This criterion reduces selection bias for lower-stage disease. In the veterinary medicine, classification of primary forms of lymphoma have been less restrictive, allowing inclusion of cases with concurrent abdominal lymph node, splenic, or hepatic involvement (Dank *et al.* 2011, Desmas *et al.* 2017), and cats with other organ involvement have been included in large studies describing feline renal lymphoma (Mooney *et al.* 1987, Taylor *et al.* 2009). As such, the use of more liberal criteria for certain cases of primary extra-nodal lymphoma (Krol *et al.* 2003) seems reasonable because the outcome does not appear affected.

As seen with two dogs in this study, cases may go undiagnosed if subtle renal lesions do not prompt sampling. In addition, systematic sampling of bone marrow, both kidneys or non-renal lesions was not always performed, and so bilateral disease or other organ involvement may have been underrepresented, and four dogs lacked thoracic imaging. This may have resulted in Ann Arbor stage migration. Histology and cytology slides did not undergo second review and immunophenotyping and cell morphology data were not available for all. Three dogs were diagnosed as B- or T-cell by PARR; detection of clonal antigen receptor rearrangement can also occur with infectious diseases (Burnett *et al.* 2003) though these are rare where the dogs resided (UK and Japan). As with other veterinary studies, survival times may be affected by decisions of owners to euthanise rather than reflecting the true nature of the disease. Finally, cases were managed by different clinicians with varied chemotherapeutic protocols and heterogeneous assessment of response, precluding evaluation of the efficacy of particular drugs/protocols.

Presumed primary renal lymphoma in dogs is associated with high morbidity, a short-lived response to multi-agent chemotherapy and poor prognosis. Further studies are needed to

identify effective therapy and prognostic factors and to assess the role of surgery and adjuvant chemotherapy in the treatment of unilateral Stage I disease.

Conflict of interest

The authors do not have any conflicts of interest to declare.

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References

Baskin, G. B. & De Paoli, A. (1977) Primary renal neoplasms of the dog. *Vet Pathol* **14**, 591-605

Batchelor, D. J., Bright, S. R., Ibarrola, P., Tzannes, S. & Blackwood, L. (2006) Long-term survival after combination chemotherapy for bilateral renal malignant lymphoma in a dog. *N Z Vet J* **54**, 147-150

Becker, A. M., Bowers, D. C., Margraf, L. R., Emmons, J. & Baum, M. (2007) Primary renal lymphoma presenting with hypertension. *Pediatr Blood Cancer* **48**, 711-713

Bennett, F. (2004) Unilateral renal cell carcinoma in a Labrador retriever. *Can Vet J* **45**, 860-862

Breshears, M. A., Meinkoth, J. H., Stern, A. W., Buoncompagni, S. & Thomason, J. D. (2011) Pathology in practice. Renal lymphoma. *J Am Vet Med Assoc* **238**, 167-169

Brown, P. M., Tzannes, S., Nguyen, S., White, J. & Langova, V. (2018) LOPP chemotherapy as a first-line treatment for dogs with T-cell lymphoma. *Vet Comp Oncol* **16**, 108-113

Burnett, R. C., Vernau, W., Modiano, J. F., Olver, C. S., Moore, P. F. & Avery, A. C. (2003) Diagnosis of canine lymphoid neoplasia using clonal rearrangements of antigen receptor genes. *Vet Pathol* **40**, 32-41

Carbone, P. P., Kaplan, H. S., Musshoff, K., Smithers, D. W. & Tubiana, M. (1971) Report of the Committee on Hodgkin's Disease Staging Classification. *Cancer Res* **31**, 1860-1861

Cheson, B. D., Fisher, R. I., Barrington, S. F., Cavalli, F., Schwartz, L. H., Zucca, E. & Lister, T. A. (2014) Recommendations for initial evaluation, staging, and response assessment of Hodgkin and non-Hodgkin lymphoma: the Lugano classification. *J Clin Oncol* **32**, 3059-3068

Choi, J. H., Choi, G. B., Shim, K. N., Sung, S. H., Han, W. S. & Baek, S. Y. (1997) Bilateral primary renal non-Hodgkin's lymphoma presenting with acute renal failure: successful treatment with systemic chemotherapy. *Acta Haematol* **97**, 231-235

Cook, S. M. & Lothrop, C. D., Jr. (1994) Serum erythropoietin concentrations measured by radioimmunoassay in normal, polycythemic, and anemic dogs and cats. *J Vet Intern Med* **8**, 18-25

Cotchin, E. (1954) Further observations on neoplasms in dogs, with particular reference to site of origin and malignancy. Part II: Male genital, skeletal, lymphatic and other systems. *Br Vet J* **110**, 274–286

Couto, C. G., Rutgers, H. C., Sherding, R. G. & Rojko, J. (1989) Gastrointestinal lymphoma in 20 dogs. A retrospective study. *J Vet Intern Med* **3**, 73-78

Dank, G., Rassnick, K. M., Kristal, O., Rodriguez, C. O., Clifford, C. A., Ward, R., Mallett, C. L., Gieger, T. & Segev, G. (2011) Clinical characteristics, treatment, and outcome of dogs with presumed primary hepatic lymphoma: 18 cases (1992-2008). *J Am Vet Med Assoc* **239**, 966-971

Davies, O., Szladovits, B., Polton, G., Garden, O. A., Leo, C. & Lara-Garcia, A. (2018) Prognostic significance of clinical presentation, induction and rescue treatment in 42 cases of canine centroblastic diffuse large B-cell multicentric lymphoma in the United Kingdom. *Vet Comp Oncol* **16**, 276-287

Desmas, I., Burton, J. H., Post, G., Kristal, O., Gauthier, M., Borrego, J. F., Di Bella, A. & Lara-Garcia, A. (2017) Clinical presentation, treatment and outcome in 31 dogs with presumed primary colorectal lymphoma (2001-2013). *Vet Comp Oncol* **15**, 504-517

Durno, A. S., Webb, J. A., Gauthier, M. J. & Bienzle, D. (2011) Polycythemia and inappropriate erythropoietin concentrations in two dogs with renal T-cell lymphoma. *J Am Anim Hosp Assoc* **47**, 122-128

Ettinger, S. J., Feldman, E. C. & Côté, E. (2017) Textbook of veterinary internal medicine: diseases of the dog and the cat, Eighth edn. Elsevier, St. Louis, Missouri.

Frank, J. D., Reimer, S. B., Kass, P. H. & Kiupel, M. (2007) Clinical outcomes of 30 cases (1997-2004) of canine gastrointestinal lymphoma. *J Am Anim Hosp Assoc* **43**, 313-321

Froment, R. & Gara-Boivin, C. (2015) Bilateral renal T-cell lymphoma with hepatic infiltration and secondary polycythemia in a dog: Utility of cytology slides. *Can Vet J* **56**, 1287-1291

Gabor, L. J., Canfield, P. J. & Malik, R. (1999) Immunophenotypic and histological characterisation of 109 cases of feline lymphosarcoma. *Aust Vet J* **77**, 436-441

Gabor, L. J., Malik, R. & Canfield, P. J. (1998) Clinical and anatomical features of lymphosarcoma in 118 cats. *Aust Vet J* **76**, 725-732

Geetha, N., Shahid, A., Rajan, V. & Jacob, P. M. (2014) Primary renal lymphoma—a case report. *Ecancermedicalscience* **8**, 466

Glicklich, D., Sung, M. W. & Frey, M. (1986) Renal failure due to lymphomatous infiltration of the kidneys. Report of three new cases and review of the literature. *Cancer* **58**, 748-753

Hartman, D. S., Davidson, A. J., Davis, C. J., Jr. & Goldman, S. M. (1988) Infiltrative renal lesions: CT-sonographic-pathologic correlation. *AJR Am J Roentgenol* **150**, 1061-1064

Hilscher, K., and N. Eberle (2004) Polyzythämie bei einem Hund mit renalem T-Zell-Lymphom. *Prakt Tierarzt* **6.7**, 470-475

Jipp, J., Sadowski, D., Kay, P. & Schwartz, B. (2016) Primary Renal Lymphoma Identified in a Robot-Assisted Laparoscopic Nephroureterectomy Specimen. *J Endourol Case Rep* **2**, 96-98

Keller, E. T., MacEwen, E. G., Rosenthal, R. C., Helfand, S. C. & Fox, L. E. (1993) Evaluation of prognostic factors and sequential combination chemotherapy with doxorubicin for canine lymphoma. *J Vet Intern Med* **7**, 289-295

Keller, S. M., Vernau, W., Hodges, J., Kass, P. H., Vilches-Moure, J. G., McElliot, V. & Moore, P. F. (2013) Hepatosplenic and hepatocytotropic T-cell lymphoma: two distinct types of T-cell lymphoma in dogs. *Vet Pathol* **50**, 281-290

Klein, M. K., Cockerell, G. L. & Harris, C. K. (1988) Canine primary renal neoplasms: a retrospective review of 54 cases. *J Am Anim Hosp Assoc* **24**, 443–452

Krol, A. D., le Cessie, S., Snijder, S., Kluin-Nelemans, J. C., Kluin, P. M. & Noordijk, E. M. (2003) Primary extranodal non-Hodgkin's lymphoma (NHL): the impact of alternative definitions tested in the Comprehensive Cancer Centre West population-based NHL registry. *Ann Oncol* **14**, 131-139

Lane, E. P. & Lobetti, R. G. (2002) Renal T-cell lymphoma with cerebral metastasis in a dog with chronic canine ehrlichiosis. *J S Afr Vet Assoc* **73**, 83-85

Lascelles, B. D., Monnet, E., Liptak, J. M., Johnson, J. & Dernell, W. S. (2003) Surgical treatment of right-sided renal lymphoma with invasion of the caudal vena cava. *J Small Anim Pract* **44**, 135-138

Mooney, S. C., Hayes, A. A., Matus, R. E. & MacEwen, E. G. (1987) Renal lymphoma in cats: 28 cases (1977-1984). *J Am Vet Med Assoc* **191**, 1473-1477

Morrison-Collister, K. E., Rassnick, K. M., Northrup, N. C., Kristal, O., Chretin, J. D., Williams, L. E., Cotter, S. M. & Moore, A. S. (2003) A combination chemotherapy protocol with MOPP and CCNU consolidation (Tufts VELCAP-SC) for the treatment of canine lymphoma. *Vet Comp Oncol* **1**, 180-190

Navas Martinez, M. C., Molina Escudero, R., Soto Delgado, M., Jimenez Romero, M. E. & Jimenez Jimenez, J. (2011) Bilateral primary renal lymphoma: case report and bibliographic review. *Arch Esp Urol* **64**, 904-907

Nelson, R. W., Hager, D. & Zanjani, E. D. (1983) Renal lymphosarcoma with inappropriate erythropoietin production in a dog. *J Am Vet Med Assoc* **182**, 1396-1397

Okuno, S. H., Hoyer, J. D., Ristow, K. & Witzig, T. E. (1995) Primary renal non-Hodgkin's lymphoma. An unusual extranodal site. *Cancer* **75**, 2258-2261

Osborne, C. A., Johnson, K. H., Kurtz, H. J. & Hanlon, G. F. (1971) Renal lymphoma in the dog and cat. *J Am Vet Med Assoc* **158**, 2058-2070

Owen, L. (1980) TNM Classification of tumors in Domestic Animals. 1980. *World Health Organization, Geneva* **34**

Saito, S. (1996) Primary renal lymphoma. Case report and review of the literature. *Urol Int* **56**, 192-195

Simon, D., Moreno, S. N., Hirschberger, J., Moritz, A., Kohn, B., Neumann, S., Jurina, K., Scharvogel, S., Schwedes, C., Reinacher, M., Beyerbach, M. & Nolte, I. (2008) Efficacy of a continuous, multiagent chemotherapeutic protocol versus a short-term single-agent protocol in dogs with lymphoma. *J Am Vet Med Assoc* **232**, 879-885

Snead, E. C. (2005) A case of bilateral renal lymphosarcoma with secondary polycythaemia and paraneoplastic syndromes of hypoglycaemia and uveitis in an English Springer Spaniel. *Vet Comp Oncol* **3**, 139-144

Stallone, G., Infante, B., Manno, C., Campobasso, N., Pannarale, G. & Schena, F. P. (2000) Primary renal lymphoma does exist: case report and review of the literature. *J Nephrol* **13**, 367-372

Szatmári, V., Favier, R. P. & Teske, E. (2004) Absolute erythrocytosis due to malignant lymphoma of the kidneys in a dog. European Veterinary Conference - Voorjaarsdagen. Amsterdam, Holland. pp 206-207

Taylor, A. J., Lara-Garcia, A. & Benigni, L. (2014) Ultrasonographic characteristics of canine renal lymphoma. *Vet Radiol Ultrasound* **55**, 441-446

Taylor, S. S., Goodfellow, M. R., Browne, W. J., Walding, B., Murphy, S., Tzannes, S., Gerou-Ferriani, M., Schwartz, A. & Dobson, J. M. (2009) Feline extranodal lymphoma: response to chemotherapy and survival in 110 cats. *J Small Anim Pract* **50**, 584-592

Vail, D. M., Michels, G. M., Khanna, C., Selting, K. A. & London, C. A. (2010) Response evaluation criteria for peripheral nodal lymphoma in dogs (v1.0)--a Veterinary Cooperative Oncology Group (VCOG) consensus document. *Vet Comp Oncol* **8**, 28-37

Villa, D., Connors, J. M., Sehn, L. H., Gascoyne, R. D. & Savage, K. J. (2011) Diffuse large B-cell lymphoma with involvement of the kidney: outcome and risk of central nervous system relapse. *Haematologica* **96**, 1002-1007

Walter, P. A., Feeney, D. A., Johnston, G. R. & O'Leary, T. P. (1987) Ultrasonographic evaluation of renal parenchymal diseases in dogs: 32 cases (1981-1986). *J Am Vet Med Assoc* **191**, 999-1007

Withrow, S. J., Vail, D. M. & Page, R. L. (2013) Withrow & MacEwen's Small Animal Clinical Oncology, 5th edn. Elsevier, St. Louis, Missouri. pp xvi, 750 pages

Zandvliet, M., Teske, E., Schrickx, J. A. & Mol, J. A. (2015) A longitudinal study of ABC transporter expression in canine multicentric lymphoma. *Vet J* **205**, 263-271

Zhao, D., Yamaguchi, R., Tateyama, S., Yamazaki, Y. & Ogawa, H. (1993) Bilateral renal lymphosarcoma in a dog. *J Vet Med Sci* **55**, 657-659

Table 1. Clinical and clincopathologic data for dogs with presumed primary renal lymphoma

Characteristics	Number affected (range)
Clinical characteristics	
Age (year)	7 (3–14)
Sex	
Neutered male	14
Intact male	8
Neutered female	5
Intact female	2
Weight (kg)	33 (9–53)
Clinicopathologic characteristics	
Hypertension	
Yes	15
No	7
Erythrocytosis	
Yes	15
No	14
Elevated creatinine	
Yes	25
No	4
Urine concentration	
Moderate to maximal (≥ 1.025)	1
Minimal (> 1.012 but < 1.025)	15
Isosthenuric	11
Hyposthenuric	1
Ultrasonographic renal lesions	
Bilateral	23
Unilateral	6
Immunophenotype	
B-cell	6
T-cell	10
Ann Arbor stage	
I	6
II	6
III	7
IV	10

Table 2. Outcome of 23 dogs with presumed primary renal lymphoma treated with chemotherapy alone

Variable (n)	Median PFS (range)	Median OS (range)
All	10 days (1–126)	12 days (1–212)
CHOP (8)	41 days (3–96)	37 days (3–104)
L-asparaginase/prednisone (2)		(3–29) days
Vincristine/prednisolone (1)		3 days
Responded to treatment (11)	41 days (10–126)	47 days (10–212)

Fig 1. Kaplan Meier survival curves for dogs with presumed primary renal lymphoma treated with chemotherapy. The dotted line represents dogs that responded to chemotherapy, and the black line represents dogs that did not respond.

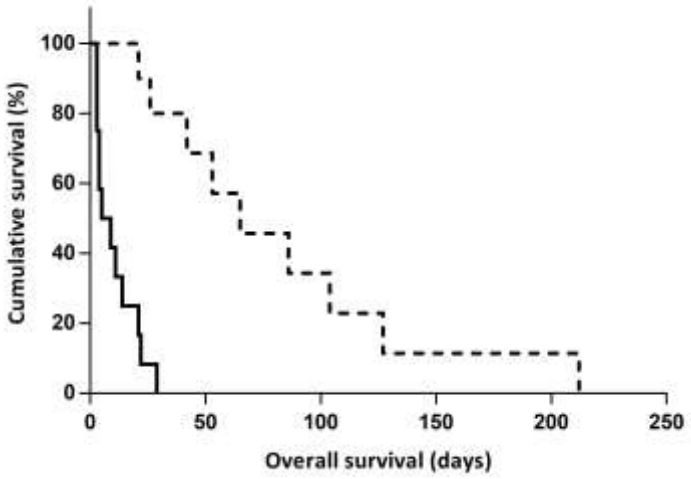

Figure 2. Haemorrhage and clotted blood extending multifocally from the capsule to the pelvis in a dog with presumed primary renal lymphoma.

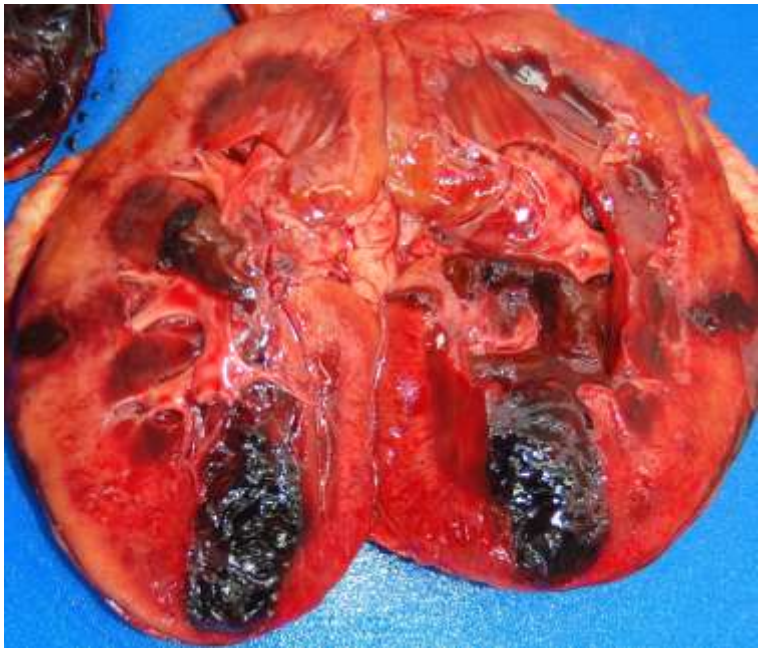

Figure 3. Marked expansion of the cortical interstitium by sheets of neoplastic lymphocytes, compressing and effacing tubules and surrounding glomeruli in a dog with presumed primary renal lymphoma (hematoxylin and eosin stain, bar=50 µm).