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ORIGINAL ARTICLE

Rabacfosadine for relapsed canine B-cell lymphoma: Efficacy and adverse event profiles of 2 different doses

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Rabacfosadine (RAB), a novel double prodrug of the acyclic nucleotide phosphonate PMEG, preferentially targets neoplastic lymphocytes with reduced off target toxicity. Historical studies have suggested that every 21-day dosing is effective with acceptable toxicity. The purpose of this study was to evaluate RAB's safety and efficacy at 2 different doses every 21 days in dogs with relapsed B-cell lymphoma. Dogs that had failed 1 doxorubicin-based chemotherapy protocol were eligible for inclusion in this prospective trial. Once enrolled, dogs were randomized to receive RAB at either 0.82 mg/kg or 1.0 mg/kg as a 30-minute IV infusion every 21 days for up to 5 treatments. Response assessment and adverse event (AE) evaluation were performed every 21 days via VCOG criteria. Fifty dogs were enrolled, with 16 treated at 0.82 mg/kg and 34 treated at 1.0 mg/kg. The overall response rate was 74%, with 45% of dogs experiencing a complete response (CR). The median progression free intervals (PFIs) were 108 days, 172 days and 203 days for all dogs, all responders, and all CRs, respectively. Response rates and PFIs were similar in both treatment groups. The incidence of AEs, dose delays, dose reductions and withdrawals were not statistically different between the 2 groups. The AEs observed were similar to those previously reported and included hematologic, gastrointestinal, dermatologic and pulmonary AEs. One dog had grade 5 pulmonary fibrosis; otherwise, AEs resolved with supportive treatment. Rabacfosadine is a generally well tolerated, effective chemotherapy option for dogs with relapsed B-cell lymphoma.

KEYWORDS

chemotherapy, dogs, lymphoma, relapsed, resistant

1 | INTRODUCTION

Multicentric lymphoma is one of the most common cancers in dogs.[1] The "gold standard" treatment generally consists of a multi-agent doxorubicin (DOX)-based chemotherapy regimen (eg, a CHOP-based protocol including cyclophosphamide, doxorubicin, vincristine, and prednisone), and clinical remission is a realistic goal. Approximately 85%-90% of dogs with lymphoma will respond to this treatment; however, few dogs are cured because of the nearly inevitable development of drug-resistant relapse.[1–4] When resistance to CHOP-based treatment occurs, clinicians must utilize different cytotoxic agents, preferably agents with distinctive mechanisms of action and mechanisms of resistance from drugs in the CHOP-based regimen.

Rabacfosadine (RAB, formerly known as GS-9219 and subsequently VDC-1101) is a nucleotide analog with a unique mechanism of action, making it an attractive treatment option for lymphoma, including CHOP-relapsed or refractory disease. Rabacfosadine, a prodrug of 9-(2-phosphonylmethoxyethyl) guanine (PMEG), has a short half-life in plasma and preferentially targets activated or neoplastic lymphoid cells. Once inside the cell, RAB undergoes enzymatic hydrolysis to form 9-(2-phosphonylmethoxyethyl)-N(6)-cyclopropyl-2,6-diaminopurine (cPrPMEDAP) which is deaminated to yield PMEG. PMEG is subsequently diphoshphorylated to the active metabolite, PMEGpp. PMEGpp induces cytotoxicity through inhibition of DNA polymerases α, δ and ε, ultimately inhibiting DNA synthesis and/or repair.[5]

The intracellular metabolism of RAB to PMEG and ultimately to PMEGpp permits clinical advantages over administration of PMEG itself. When compared with PMEG, it has been demonstrated that RAB more effectively loads peripheral blood mononuclear cells (PMBCs) and lymph nodes with significantly lower distribution in, and toxicity to, the kidneys and gastrointestinal tract.[5]

Previous studies evaluating RAB in both treatment naive and relapsed or treatment-refractory dogs with lymphoma have reported overall response rates of ~50%-100%.[5–7] As seen with most other cytotoxic agents, dogs with B-cell lymphoma are more likely to respond with significantly longer response durations.[6] A variety of dosing regimens have been evaluated, with 0.82-1.0 mg/kg every 21 days used most frequently and with a lower likelihood of adverse effects as compared with alternate dosing frequencies, with apparently equivalent efficacy.[6–8] While the drug has been well tolerated, dose limiting toxicities have included neutropenia, dermatopathy and gastrointestinal signs. The dermatopathy is most often characterized as a pruritic focal otitis externa or focal erythemic skin lesions on the dorsum and in the inguinal areas.[6–9] With supportive therapy and treatment interruption, the dermatopathy generally resolves. Another unique but potentially life-threatening idiosyncratic toxicity seen with RAB administration is pulmonary fibrosis, which has been recognized in a small number of treated dogs, necessitating careful monitoring of thoracic radiographs for evidence of pulmonary pathology.[6,9]

The objective of this study was to evaluate the safety and efficacy of RAB at 2different doses in dogs with B-cell lymphoma that had relapsed following one initial DOX-based treatment regimen. To achieve this goal, a prospective multi-institutional trial was initiated with the inclusion of 9 tertiary referral oncology specialty sites across North America. The deliverable findings would serve as future guiding principles for the administration of RAB in the setting of relapsed B-cell lymphoma.

2 | MATERIALS AND METHODS

2.1 | Inclusion/exclusion criteria

This study was conducted across 9 study sites including Hope Veterinary Specialty Specialists, Colorado State University, University of Wisconsin-Madison, Red Bank Veterinary Hospital, Tufts University, University of Illinois, Veterinary Specialty Hospital of San Diego, VCA Animal Diagnostic Clinic and University of Georgia. Client-owned dogs of at least 1 year of age with a cytologic or histologic diagnosis of B-cell multicentric lymphoma were eligible for inclusion in this prospective randomized, double-arm, open-label clinical trial. Confirmation of B-cell immunophenotype using immunohistochemistry, immunocytochemistry, flow cytometry or polymerase chain reaction (PCR) for antigen receptor rearrangement (PARR) was required. In an attempt to evaluate RAB as a first line rescue agent and without the confounding challenges that arise with varying degrees of pretreatment between patients, eligible dogs had received at least one, but no more than one DOX-based treatment regimen, consisting of administration of DOX alone or in combination with one or more cytotoxic agents including vincristine, vinblastine, cyclophosphamide, and asparaginase with or without corticosteroids. A lapse of at least 1 week between treatment with the DOX-based chemotherapy regimen and entry into the trial was required. Prior treatment with short-acting corticosteroids and/or homeopathic or alternative therapies was permissible if discontinued ≥72 hours and ≥24 hours prior to study initiation, respectively. Prior radiation therapy was also permissible as long as there was a ≥6 week lapse between radiation therapy and entry into the trial.

Adequate bone marrow and organ function, defined as absolute neutrophil count ≥2000 cells/μL, hematocrit ≥ 25%, platelet count ≥ 75 000 cells/μL, creatinine ≤ 2.5 mg/dL, total bilirubin ≤ the upper limit of normal (ULN), ALT ≤ 3 times ULN or if > 3 times ULN, serum bile acids ≤ ULN, were required. A modified ECOG performance score ≤ 1 was required for inclusion.[8]

West Highland white terrier dogs, dogs previously treated with bleomycin and/or dogs with pulmonary pathology possibly predisposing to fibrosis were excluded from the study. The study protocol was approved by each institution's Animal Care and Use Committee and/or Clinical Review Board.

2.2 | Trial design

Rabacfosadine was provided by VetDC, Inc. (Fort Collins, Colorado). Signed informed consent was obtained from all owners prior to study entry. Within 7 days of trial entry, screening tests including physical examination, complete blood count (CBC), serum biochemical profile, urinalysis and thoracic radiographs were performed to ensure dogs met all inclusion criteria. Once enrolled, each dog was randomized to receive RAB at 0.82 mg/kg (Treatment Group A) or 1.0 mg/kg (Treatment Group B). The protocol was subsequently modified to include more dogs in Treatment Group B, as 1.0 mg/kg was the intended label dose. Rabacfosadine was reconstituted and diluted with Sodium Chloride for Injection, USP to achieve a total infusion volume of 2 mL/kg and was administered intravenously (IV) over 30 minutes. Treatments were repeated every 21 days for up to 5 total treatments, per the intended label dose. Dogs were evaluated prior to each treatment as outlined in Table 1.

Treatment response was based on measurements of peripheral target lesions using the Veterinary Cooperative Oncology Group (VCOG) Response Evaluation Criteria for Peripheral Nodal Lymphoma.[10] Dogs experiencing CR received a total of 5 RAB treatments; thereafter, monthly rechecks were performed until PD was noted. Dogs experiencing PR or SD after 5 treatment cycles were considered off-study upon completion of the fifth treatment cycle and censored from outcome analysis at that point. Dogs experiencing PD were removed from the study and were eligible for other treatment as deemed appropriate by the investigator.

2.3 | Adverse event assessment

Hematological adverse events (AEs) were evaluated 7 days after the first treatment. Thereafter, clinical, hematological, and biochemical AEs were assessed every 21 days based on patient history provided by the owner, physical examination and blood work (Table 1).

TABLE 1 Study schedule

Day	RAB treatment	PE	LN evaluation	CBC	Serum chemistry	UA	Thoracic radiographs
Pre-enrollment (day −7 to −1)				X	X	X	X
Day 0	X	X	X	X[a]	X[a]	X[a]	
Day 7		X		X			
Day 21	X	X	X	X	X	X	
Day 28[b]		X		X			
Day 42	X	X	X	X	X	X	
Day 63	X	X	X	X	X		
Day 84	X	X	X	X	X	X	X
Monthly rechecks		X	X				Every other month

Abbreviations: CBC, complete blood count; LN, lymph node; PE, physical examination; RAB, rabacfosadine; UA, urinalysis.

[a] If CBC, serum chemistry, and urinalysis were performed and evaluated within 7 days of day 0, these were not repeated on day 0.

[b] This visit was only required in dogs experiencing a dose-limiting toxicity following the first treatment.

Adverse events were graded according to the Veterinary Cooperative Oncology Group Common Terminology Criteria for Adverse Events (VCOG-CTCAE) v1.1.[11] Dose-limiting toxicities (DLT) were defined as any grade 3 or 4 non-hematologic toxicity, any uncomplicated (eg, no fever, bleeding, etc.) grade 4 hematologic toxicity, or any complicated grade 3 or 4 hematologic toxicity. In addition, dermatological lesions deemed less than grade 3 according to VCOG-CTCAE v1.1[11] criteria but considered clinically substantial and/or extensive enough to warrant protocol alteration were considered DLTs. Exceptions that were not considered DLT included AEs not related to RAB; hyporexia, vomiting, or diarrhea remediable within 24 hours by supportive medical therapy; elevations in liver enzymes or total bilirubin which resolved without medical intervention.

Dose reductions and/or delays of up to 2 weeks were permissible to manage AE. If a DLT was observed, the dose was reduced by up to 20% for future RAB administrations.

2.4 | Statistical analysis

Continuous data were expressed as median and range, and categorical data as frequencies and percentages. The objective response rate (ORR) and progression-free interval (PFI) were the primary efficacy endpoints. The ORR was defined as the percentage of evaluable patients experiencing CR or PR as their best response. The PFI was calculated from the date of treatment initiation to the date of PD. Dogs were censored if they had not developed PD at the time of data analysis, or if they were withdrawn or lost to follow up before PD development. Continuous variables were compared between groups of patients using a 2-tailed, unpaired *t* test or Mann-Whitney test depending on data normality, which was assessed using a D'Agostino Pearson omnibus test. Categorical variables were compared between cohorts using a 2-tailed Fisher's exact test. The Kaplan-Meier method was used to estimate and display the distribution of PFI. Differences between potential prognostic subsets were compared using logrank analysis. Variables with values of $P \leq .05$ were considered significant. All statistical analysis was performed with a commercial software package (Prism v. 6.0b, GraphPad Software, La Jolla, California).

3 | RESULTS

Fifty dogs were included; 16 were in Treatment Group A and 34 were in Treatment Group B. Patient demographics are outlined in Table **S1**, Supporting Information.

The ORR for all treated dogs was 74%. The ORR for Group A was 69%, with 8 dogs (50%) experiencing CR, 3 dogs (19%) experiencing PR, 2 dogs (13%) experiencing SD and 3 dogs (19%) experiencing PD. The ORR for Group B was 77%, with 13 dogs (42%) experiencing CR, 11 dogs (35%) experiencing PR, 5 dogs (16%) experiencing SD and 2 dogs (6%) experiencing PD. In Group B, 1 dog was euthanized and 2 were withdrawn from the study because of owner reported AEs prior to the first response evaluation. Table 2 summarizes the median PFI, median response duration and median complete response duration for all dogs, with comparisons between Treatment Groups A and B. Twenty-one dogs were censored from survival analysis. One dog was still on study in CR 546 days following treatment initiation. Fourteen dogs were removed due to AEs and/or declining quality of life, 2 were removed at the owner's request, 3 were lost to follow up prior to progression and one was removed as a result of a splenic infarct resulting in euthanasia 8 days after the first RAB treatment. The median follow-up time for all censored patients was 61 days (range: 7 to >546 days). Three of the censored dogs (14%) were in Treatment Group A, and 18 (86%) were in Treatment Group B.

All AEs are summarized in Table 3. Figure 1 shows the highest AE reported per patient. Fifteen dogs required dose reductions (3 in Treatment Group A and 12 in Treatment Group B), 5 dogs required

TABLE 2 Overall response rate (ORR), median progression free interval (PFI), median response duration (RD) and median complete response (CR) duration (in days) for all dogs, with comparisons between Treatment Groups A and B

	Overall	0.82 mg/kg	1.0 mg/kg	*P*
ORR	74%	69%	77%	.43
Median PFI	108 d (range: 7-332 d)	72	108	.32
Median RD	172 d (range: 42-332 d)	172	148	.79
Median CR duration	215 d (range: 72-332 d)	203	264	.17

TABLE 3 All adverse events (AEs) by grade per treatment group

	1	2	3	4	5		1	2	3	4	5
0.82 mg/kg						1.0 mg/kg					
Gastrointestinal											
Anorexia	2	2	1				5	5	8		
Dehydration	1						1	1	1		
Diarrhea	6	3	1				12	9	6		
Hematochezia							1				
Nausea/vomiting	6	3					8	9			
Weight loss	4	3					8	7	3		
Constitutional											
Lethargy		1	2				4	7	2		
Haematologic											
Anemia	2	1					1	1			
Neutropenia	1	1					5	4			
Thrombocytopenia		1					3	1			
HEPATIC											
AST	1						1				
GGT	1										
Bilirubin							1				
SAP	1						1		1		
Urinary											
BUN	2						2	1			
Creatinine	3						3	1			
Polyuria/polydipsia							2				
Proteinuria	6						5				
Pyuria	2										
Casts	1										
Stranguria		1									
Other biochemical											
Amylase	1										
CPK	2	1					1				
Hypercalcemia	1										
Hypercholesterolemia	2						1	1			
Hyperglobulinemia	2										
Hyperkalemia	1						1				
Hypermagnesemia	1										
Hypernatremia	1						1				
Hypertriglyceridemia							2				
Hypocalcemia							1				
Hypochloremia							1				
Hypoglobulinemia	1						4				
Hypoglycemia							1				
Pancreatitis		1									
Cutaneous/pulmonary											
Dermatopathy	6	1					3	5	2		
Edema								1			
Otitis							2	3	3		
Pulmonary fibrosis					1			3			
Dyspnea		1						1			
Cough	2	2					2				
Pneumonia			1								
Other											

(*Continues*)

TABLE 3 (Continued)

	1	2	3	4	5	1	2	3	4	5
Behavior change						1				
Pruritus	2									
Pain						1	1			
Fever		1								
Conjunctivitis	1									

Abbreviations: AST, aspartate aminotransferase; BUN, blood urea nitrogen; CPK, creatinine phosphokinase; GGT, gamma-glutamyl transferase; SAP, serum alanine transferase.

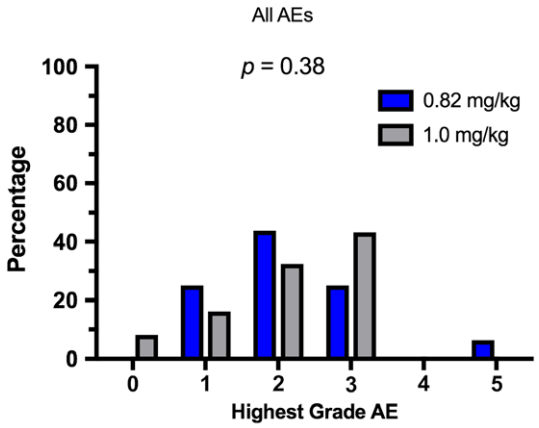

FIGURE 1 Highest grade adverse event (AE) reported per dog for each group. The blue boxes represent dogs in Group A (0.82 mg/kg). The gray boxes represent dogs in Group B (1.0 mg/kg). There was no statistically significant difference in reported AE between groups (P = .38)

dose delays (1 in Treatment Group A and 4 in Treatment Group B), and 14 dogs were withdrawn from the study (3 in Treatment Group A and 11 in Treatment Group B). For each of these variables, there was no statistically significant difference between Treatment Groups (P = .51, P = 1.0 and P = .25, respectively). However, dogs experiencing dose delay or dose reduction had a significantly longer PFI than dogs not experiencing dose delay or dose reduction (203 days vs 63 days, P = .023).

In Treatment Group A, there were 6DLTs including 1 grade 3 hyporexia, 1 grade 3 diarrhea, 2 grade 3 lethargy, 1 grade 3 pneumonia and 1 grade 5 pulmonary fibrosis. The dogs with grade 3 pneumonia and grade 5 pulmonary fibrosis were withdrawn from the study as a result of these AEs. The dog with grade 5 pulmonary fibrosis was first documented to have pulmonary changes characterized as grade 2 pulmonary fibrosis at the time of its fifth RAB treatment. Because of this finding, the fifth treatment was not given, and the dog was removed from the study. Three months later the dog was euthanized as a result of progressive pulmonary signs, characterized as grade 5 pulmonary fibrosis. However, on necropsy examination, only mild pulmonary fibrosis was observed in addition to evidence of pulmonary infiltration with lymphoma.

Although one of the dogs in Treatment Group A did not experience a DLT as defined by the study protocol, it developed suspected acute kidney injury defined by a grade 1 creatinine elevation after the first treatment; as a result, this dog was removed from the study based on the attending clinician's judgement.

In Treatment Group B, there were 29 DLTs including 8 grade 3 hyporexia, 1 grade 3 dehydration, 6 grade 3 diarrhea, 3 grade 3 weight loss, 2 grade 3 lethargy, 1 grade 3 SAP elevation, 1 grade 2 pulmonary fibrosis, 4 grade 2 dermatologic AEs and 3 grade 3 dermatologic AEs. Ten dogs were withdrawn from the study as a result of these DLTs. The number of dogs experiencing DLTs was not significantly different between Treatment Groups A and B (P = .23).

Hematologic toxicity was mild in both groups. In Treatment Group A, hematologic AEs included 2 grade 1 anemia, 1 grade 2 anemia, 1 grade 1 neutropenia, 1 grade 2 neutropenia and 1 grade 2 thrombocytopenia. In Treatment Group B, hematologic AEs included 1 grade 1 anemia, 1 grade 2 anemia, 5 grade 1 neutropenia, 4 grade 2 neutropenia, 3 grade 1 thrombocytopenia and 1 grade 2 thrombocytopenia.

When looking at the unique dermatologic toxicity which includes dermatitis, hyperpigmentation, alopecia, pruritus, and otitis, there were 8 grade 1 and 1 grade 2 AEs reported in Treatment Group A. None of these dogs were withdrawn from the study as a result of these AEs, but 2 were removed from the study because of PD at the time their dermatopathies were noted. In Treatment Group B, dermatologic AEs included 5 grade 1, 8 grade 2 and 5 grade 3. Six dogs were withdrawn from the study as a result of these AEs. Four of these dogs were in CR and 2were in PR.

In addition to the aforementioned dog with grade 5 pulmonary fibrosis in Treatment Group A, there were 3 dogs with grade 2 possible pulmonary fibrosis in Treatment Group B. All the 3 were suspected based on lung appearance on thoracic radiographs, but none were confirmed with lung histopathology. Two were removed from the study as a result of this AE. One remained on study; with discontinuation of RAB and the addition of prednisone and a bronchodilator, subjective improvement was observed on thoracic radiographs performed 1 month later.

4 | DISCUSSION

The results of this study provide evidence that RAB is an effective treatment for dogs with B-cell lymphoma that have relapsed following an initial DOX-based chemotherapy regimen. The overall response rate was 74% with a median PFI in responders of ~6 months. There were no significant differences between the 2dosages with respect to response rate, PFI, response duration, or AEs; however, it should be noted that enrolment in Treatment Group A was discontinued early to allow treatment of more dogs at the intended label dose of 1.0 mg/kg, resulting in limited power to detect

differences between groups. Furthermore, because the 1.0 mg/kg labelled dose is more dose intense and appears reasonably well tolerated, especially with the utilization of supportive medications and possibly drug holidays to ameliorate AEs, the authors support treatment at this dose. Across both dose groups, adverse events were similar to those previously reported, including dermatopathy and pulmonary fibrosis.[6–9] Aside from 1 dog that developed signs consistent with grade 5 pulmonary fibrosis, other AEs were mostly self-limiting and resolved with supportive care and/or dosage modification. With that being said, a total of 14 dogs were withdrawn from the study, perhaps prematurely, owing to AEs or a perceived diminished quality of life. However, many of these AEs possibly could have been addressed by dose reductions and/or prophylactic medication administration. Prophylactic therapies (eg, antiemetics, antidiarrheals) generally were not used after the first treatment, and concurrent corticosteroids were not permitted. Furthermore, RAB is a novel investigational drug, so it is understandable that owners and clinicians might be less tolerant of even mild AEs until a greater expertise with the drug is established.

A multitude of single cytotoxic agents and combinations of agents have been investigated as rescue treatments for relapsed and refractory canine lymphoma, with response rates ranging from 0% to 87%[12–24]; however, attempts to make direct comparisons of RAB to these historical studies are of minimal value given the non-concurrent nature of serial investigations. With that being said, RAB's unique mechanism of action, substantial response rate and duration of response lends credence to its utility as a rescue agent. None of the cytotoxic agents in the commonly used first line multi-agent CHOP-based regimen are nucleotide analogs, and therefore their mechanisms of action and potential mechanisms of resistance are different from those of RAB.[25] Furthermore, the practiced dosing regimens for cytosine arabinoside and gemcitabine, the other non-guanine nucleoside analogs used in veterinary oncology have demonstrated minimal to no activity against relapsed and treatment-naive lymphoma respectively,[23,24] suggesting that RAB may be a more effective rescue option.

Although there was no statistical significant difference in response rate or PFI between dogs receiving RAB at 0.82 mg/kg and those receiving 1.0 mg/kg, there were numeric improvements in response rate (69% vs 77%), overall PFI (72 vs 108 days) and CR duration (203 vs 264 days) in dogs receiving 1.0 mg/kg, suggesting the possibility of improved outcome in those dogs. Given the premature termination of enrolment of the 0.82 mg/kg dose group, it is possible that there was insufficient power for detection of significant outcome differences. While there was a significant difference in PFI between dogs experiencing dose delay or dose reduction as compared with those that did not, this finding must be interpreted in light of the fact that such dose alterations were more common in dogs remaining on the study to receive more than one RAB dose as compared with those progressing or withdrawing after the first RAB dose.

The unique dermatologic toxicity reported previously with RAB[6–9] was noted in both treatment groups in this study. Although in most cases, this AE was mild and self-limiting, it did result in patient withdrawal from the study in 6 dogs. Interestingly, all 6 dogs had responded to RAB at the time of withdrawal, including 4 CR and 2 PR, and it was difficult to ascertain the rationale for premature withdrawal in retrospect. It is not clear why this dermatopathy occurs; however, it has been proposed that it is secondary to drug distribution to the skin.[6] This hypothesis prompted a phase II study evaluating the effect of RAB in the treatment of canine cutaneous T cell lymphoma (CTCL), which reported an ORR of 45%, including 1 CR and 4 PR.[8] Interestingly, a previous study reported that dermatologic toxicity occurred in 37% of dogs treated with RAB and was seen most commonly in the dogs receiving daily or weekly treatments. No skin-related AEs were seen in the every 21-day cohort of dogs.[6] With that being said, all dogs in the CTCL study were treated at a 21-day interval, yet 25% developed drug-related dermatopathies, with 1 dog exiting the study as a result of the AE.[8] The awareness that dermatopathies are possible even with less frequent dosing (ie, every 21 days) is important and should prompt clinicians to closely monitor dogs' skin and ears regardless of RAB dose intensity. With that being said, users are encouraged to manage low-grade dermatopathies with drug holidays and supportive medications, rather than complete discontinuation of RAB.

Pulmonary fibrosis has also been reported previously in dogs receiving RAB.[6,9] In the previously published lymphoma study,[6] the dogs with pulmonary fibrosis had completed RAB treatment and were subsequently treated with other cytotoxic drugs. Only one of these dogs underwent a necropsy examination where pulmonary fibrosis was confirmed. Of the remaining dogs that underwent necropsy examination, pulmonary changes, characterized by hyperplasia, were seen in only one. While the authors were unable to explain the relationship between RAB administration and the pulmonary pathology, it was concluded that these findings warranted close observation of thoracic imaging in dogs receiving this treatment.[6] Pulmonary fibrosis was also noted in a subsequent study of RAB in dogs with multiple myeloma.[9]

In the current study, 1 dog was documented to have grade 5 pulmonary fibrosis, which was first noted 84 days after treatment initiation. However, on necropsy examination, the fibrosis was reported as mild, and lymphoma was found in the lungs, making attribution of the observed dyspnoea challenging. Thoracic radiographic findings are variable in dogs with pulmonary lymphoma,[26] and in some cases, such radiographic abnormalities may easily be mistaken for pulmonary fibrosis. Questions as to the most appropriate use of thoracic radiograph monitoring, as well as the potential benefit of concomitant low-dose corticosteroids are beyond the scope of this study but warrant further investigation. Although pulmonary fibrosis can be life-threatening, this unique toxicity appears to be relatively infrequent.

Another notable AE was proteinuria, which was seen in 22% of the study population. However, it is unknown if this was related to RAB administration versus the underlying lymphoma or an unrelated cause (eg, lower urinary tract infection). All incidences of proteinuria were mild, and while we do not strongly suspect they were related to RAB administration, further investigation and careful monitoring of RAB-treated dogs are warranted.

One limitation of the study design was that owners were not asked to keep daily dairies at home to prospectively record any potential AEs on a daily basis. As a result, subtle or mild constitutional and gastrointestinal AEs (eg, lethargy, hyporexia, vomiting, diarrhea, etc.) may have been under-reported.

In conclusion, RAB at a dosage of 1.0 mg/kg every 21 days is a generally well tolerated and viable option for dogs with relapsed B-cell lymphoma. A dose reduction from 1.0 mg/kg to 0.82 mg/kg, if needed to address AEs, does not appear to adversely affect RAB's efficacy. Careful monitoring for the unique dermatologic and pulmonary toxicities is warranted in dogs receiving this treatment.

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REFERENCES

1. Vail DM, Pinkerton ME, Young KM. Hematopoietic tumors: section A: canine lymphoma and lymphoid leukemias. In: Withrow SJ, Vail DM, Page RL, eds. *Small Animal Clinical Oncology*. 5th ed. St. Louis, MO: Elsevier Saunders; 2013:608-638.
2. Chun R, Garrett LD, Vail DM. Evaluation of a high-dose chemotherapy protocol with no maintenance therapy for dogs with lymphoma. *J Vet Intern Med*. 2000;14(2):120-124.
3. Garrett LD, Thamm DH, Chun R, Dudley R, Vail DM. Evaluation of a 6-month chemotherapy protocol with no maintenance therapy for dogs with lymphoma. *J Vet Intern Med*. 2002;16(6):704-709.
4. Moore AS, Cotter SM, Rand WM, et al. Evaluation of a discontinuous treatment protocol (VELCAP-S) for canine lymphoma. *J Vet Intern Med*. 2001;15(4):348-354.
5. Reiser H, Wang J, Chong L, et al. GS-9219—a novel acyclic nucleotide analogue with potent antineoplastic activity in dogs with spontaneous non-Hodgkin's lymphoma. *Clin Cancer Res*. 2008;14(9):2824-2832.
6. Vail DM, Thamm DH, Reiser H, et al. Assessment of GS-9219 in a pet dog model of non-Hodgkin's lymphoma. *Clin Cancer Res*. 2009;15(10):3503-3510.
7. Thamm D, Clifford C, Vickery K, Phillips B, Rebhun R, Saba C, et al. Rabacfosadine for canine lymphoma: efficacy and adverse event profiles of two different doses. Paper presented at: Proceedings from Thirty-fifth Annual Meeting of Veterinary Cancer Society. Tyson's Corner, VA, USA. Oct, 2015.
8. Morges MA, Burton JH, Saba CF, Vail DM, Burgess KE, Thamm DH. Phase II evaluation of VDC-1101 in canine cutaneous T-cell lymphoma. *J Vet Intern Med*. 2014;28(5):1569-1574.
9. Thamm DH, Vail DM, Kurzman ID, et al. GS-9219/VDC-1101—a prodrug of the acyclic nucleotide PMEG has antitumor activity in spontaneous canine multiple myeloma. *BMC Vet Res*. 2014;10(30):1-7.
10. Vail DM, Michels GM, Khanna C, Selting KA, London CA. Response evaluation criteria for peripheral nodal lymphoma in dogs (v1.0)—a Veterinary Cooperative Oncology Group (VCOG) consensus document. *Vet Comp Oncol*. 2010;8(1):28-37.
11. Veterinary Cooperative Oncology Group. Veterinary cooperative oncology group—common terminology criteria for adverse events (VCOG-CTCAE) following chemotherapy or biological antineoplastic therapy in dogs and cats v1.1. *Vet Comp Oncol*. 2011;14(4):417-446.
12. Saba CF, Thamm DH, Vail DM. Combination chemotherapy with L-asparaginase, lomustine, and prednisone for relapsed or refractory canine lymphoma. *J Vet Intern Med*. 2007;21(1):127-132.
13. Saba CF, Hafeman SD, Vail DM, Thamm DH. Combination chemotherapy with continuous L-asparaginase, lomustine, and prednisone for relapsed canine lymphoma. *J Vet Intern Med*. 2009;23(5):1058-1063.
14. Flory AB, Rassnick KM, Al-Sarraf R, et al. Combination of CCNU and DTIC chemotherapy for treatment of resistant lymphoma in dogs. *J Vet Intern Med*. 2008;22(1):164-171.
15. Griessmayr PC, Payne SE, Winter JE, Barber LG, Shofer FS. Dacarbazine as single-agent therapy for relapsed lymphoma in dogs. *J Vet Intern Med*. 2009;23(6):1227-1231.
16. Back AR, Schleis SE, Smrkovski OA, Lee J, Smith AN, Phillips JC. Mechlorethamine, vincristine, melphalan, and prednisone (MOMP) for the treatment of relapsed lymphoma in dogs. *Vet Comp Oncol*. 2015;13(4):398-408.
17. Lenz JA, Robat CS, Stein TJ. Vinblastine as a second rescue for the treatment of canine multicentric lymphoma in 39 cases (2005–2014). *J Small Anim Pract*. 2016;57(8):429-434.
18. Gillem J, Giuffrida M, Krick E. Efficacy and toxicity of carboplatin and cytarabine chemotherapy for dogs with relapsed or refractory lymphoma (2000–2013). *Vet Comp Oncol*. 2017;15(2):400-410.
19. Fahey CE, Milner RJ, Barabas K, et al. Evaluation of the University of Florida lomustine, vincristine, procarbazine, and prednisone chemotherapy protocol for the treatment of relapsed lymphoma in dogs: 33 cases (2003–2009). *J Am Vet Med Assoc*. 2011;239(2):209-215.
20. Bannink EO, Sauerbrey ML, Mullins MN, Hauptman JG, Obradovich JE. Actinomycin D as rescue therapy in dogs with relapsed or resistant lymphoma: 49 cases (1999–2006). *J Am Vet Med Assoc*. 2008;233(3):446-451.
21. Van Vechten M, Helfand SC, Jeglum KA. Treatment of relapsed canine lymphoma with doxorubicin and dacarbazine. *J Vet Intern Med*. 1990;4(4):187-191.
22. Moore AS, London CA, Wood CA, et al. Lomustine (CCNU) for the treatment of resistant lymphoma in dogs. *J Vet Intern Med*. 1999;13(5):395-398.
23. Ruslander D, Moore AS, Gliatto JM, L'Heureux D, Cotter SM. Cytosine arabinoside as a single agent for the induction of remission in canine lymphoma. *J Vet Intern Med*. 1994;8(4):299-301.
24. Turner AI, Hahn KA, Rusk A, et al. Single agent gemcitabine chemotherapy in dogs with spontaneously occurring lymphoma. *J Vet Intern Med*. 2006;20(6):1384-1388.
25. Gustafson D, Page RL. Cancer Chemotherapy. In: Withrow S, Vail D, Page R, eds. *Small Animal Clinical Oncology*. 5th ed. St. Louis, MO: Elsevier Saunders; 2013.
26. Geyer NE, Reichle JK, Valdes-Martinez A, et al. Radiographic appearance of confirmed pulmonary lymphoma in cats and dogs. *Vet Radiol Ultrasound*. 2010;51(4):389-390.

SUPPORTING INFORMATION

Additional Supporting Information may be found online in the supporting information tab for this article.

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