Journal of Veterinary Internal Medicine
Open Access

Standard Article

J Vet Intern Med 2017

Alternating Rabacfosadine/Doxorubicin: Efficacy and Tolerability in Naïve Canine Multicentric Lymphoma

D.H. Thamm, D.M. Vail, G.S. Post, T.M. Fan, B.S. Phillips, S. Axiak-Bechtel, R.S. Elmslie, M.K. Klein, and D.A. Ruslander

Background: Standard of care treatment for multicentric lymphoma in dogs remains doxorubicin (DOX)-based combination chemotherapy, but owners may hesitate to commit the time and financial resources to complete such a protocol, typically requiring 12–16 visits. Rabacfosadine (RAB), a double prodrug of the nucleotide analog 9-(2-phosphonylmethoxyethyl) guanine, has substantial single-agent activity in dogs with lymphoma, and a different mechanism of action than DOX.

Hypothesis/Objectives: Our objective was to evaluate the efficacy and adverse effect (AE) profile of alternating doses of RAB and DOX in dogs with naïve multicentric lymphoma.

Animals: Fifty-four dogs with previously untreated lymphoma.

Methods: Open-label, multicenter prospective clinical trial. Dogs received alternating RAB (1.0 mg/kg IV weeks 0, 6, 12) and DOX (30 mg/m^2 IV weeks 3, 9, 15). Dogs that achieved complete response (CR) were followed by monthly evaluations. Complete clinicopathological evaluation and assessment of remission and AEs were performed every 21 days.

Results: The overall response rate was 84% (68%; CR; 16%; partial response [PR)]. The overall median progression-free interval (PFI) was 194 days (216 for CR and 63 for PR). Most AEs were mild and self-limiting: gastrointestinal and hematologic AEs were most common. Thirteen dogs experienced dermatologic AEs, and 2 dogs developed grade 5 pulmonary fibrosis.

Conclusions and Clinical Importance: Alternating RAB/DOX generally was well tolerated and resulted in PFIs comparable to standard DOX-based multi-agent protocols, with fewer treatment visits. Most adverse events were mild or moderate and self-limiting. Further studies are warranted to explore long-term outcome and other RAB chemotherapy combinations.

Key words: Chemotherapy; Dog; Guanine; Lymphosarcoma.

From the Department of Clinical Sciences, Flint Animal Cancer Center, Colorado State University, (Thamm); Cell and Molecular Biology Graduate Program, Colorado State University, Fort Collins, (Thamm); Developmental Therapeutics Program, Comprehensive Cancer Center, University of Colorado, Aurora, CO (Thamm); School of Veterinary Medicine, University of Wisconsin-Madison, Madison, WI (Vail); The Veterinary Cancer Center, Norwalk, CT (Post); College of Veterinary Medicine, University of Illinois at Urbana-Champaign, Urbana, IL (Fan); Veterinary Specialty Hospital of San Diego, San Diego, CA (Phillips); College of Veterinary Medicine, University of Missouri, Columbia, MO (Axiak-Bechtel); Veterinary Referral Center of Colorado, Englewood, CO (Elmslie); Southern Arizona Veterinary Specialty and Emergency Center, Tucson, AZ (Klein); and the Veterinary Specialty Hospital of the Carolinas, Cary, NC (Ruslander).

This work was performed at the following sites: Colorado State University, Fort Collins, CO; University of Wisconsin-Madison, Madison, WI; The Veterinary Cancer Center, Norwalk, CT; University of Illinois at Urbana-Champaign, Urbana, IL; Veterinary Specialty Hospital of San Diego, San Diego, CA; University of Missouri, Columbia, MOi; Veterinary Referral Center of Colorado, Englewood, CO; Southern Arizona Veterinary Specialty and Emergency Center, Tucson, AZ; Veterinary Specialty Hospital of the Carolinas, Cary, NC.

The preliminary results of this study were presented at the 2016 American College of Veterinary Internal Medicine Annual Forum in Denver, CO.

Corresponding author: D.H. Thamm, Flint Animal Cancer Center, Colorado State University, 300 W Drake Road, Fort Collins, CO 80523-1620; e-mail: dthamm@colostate.edu.

Submitted January 11, 2017; Revised February 13, 2017; Accepted February 23, 2017.

DOI: 10.1111/jvim.14700

Abbreviations:

AE	adverse event
ALT	alanine aminotransferase
AST	aspartate aminotransferase
CHOP	cyclophosphamide doxorubicin vincristine prednisone
CR	complete response
DOX	doxorubicin
ORR	objective response rate
PFI	progression-free interval
PR	partial response
RAB	rabacfosadine
SD	stable disease
VCOG	Veterinary Cooperative Oncology Group

The current standard of care for the treatment of dogs with multicentric lymphoma is combination chemotherapy including cyclophosphamide, doxorubicin (DOX), vincristine, and prednisone (CHOP).[1] Objective response rates (ORR) of ≥85%, median response durations of 6–10 months, and median overall survival times of 8–12 months have been reported with CHOP-based protocols.[2–11] Most dogs that achieve remission after a CHOP-based chemotherapy protocol eventually relapse, with <25% of patients experiencing survival times >2 years.[1] Because treatment of lymphoma in dogs is not curative, owners may be hesitant to commit the time and financial resources necessary to complete a lengthy, multidrug protocol.[12]

Rabacfosadine (RAB), formerly referred to as VDC-1101 or GS-9219, is a double prodrug of the guanine nucleotide analog 9-(2-phosphonylmethoxyethyl) guanine (PMEG), which was designed to preferentially deliver PMEG and its active phosphorylated metabolite,

PMEG diphosphate (PMEGpp) to lymphoid cells while avoiding systemic PMEG exposure.[13] Cytotoxic activity mediated by PMEG/PMEGpp is by inhibition of nuclear DNA polymerases α, δ, and ε.[14] The clinical utility of PMEG is limited by poor cellular permeability as well as gastrointestinal and renal toxicity.[15–17] Rabacfosadine, however, is hydrolyzed intracellularly to 9-(2-phosphonylmethoxyethyl)-N^6-cyclopropyl-2,6-diaminopurine (cPrPMEDAP), deaminated to PMEG and then rapidly converted to PMEGpp.[13] Rabacfosadine selectively depletes replicating lymphoid tissues at doses that spare most other organ systems in normal laboratory dogs, and demonstrates substantial antineoplastic activity in dogs with lymphoma.[13,18,19] Additional studies recently have identified activity in dogs with cutaneous T-cell lymphoma and multiple myeloma.[20,21] Commonly encountered adverse events (AEs) include self-limiting gastrointestinal toxicity and myelosuppression; cumulative dermatologic changes and rare delayed pulmonary fibrosis also have been observed in some dogs.[19–21]

In the management of multicentric lymphoma in dogs, DOX exerts robust single-agent activity with ORRs ranging from 65 to 85% and median response durations of 100–170 days.[22–26] Common AEs include gastrointestinal signs and myelosuppression, as well as a dose-dependent, cumulative cardiomyopathy that can limit the total number of DOX doses that can be administered.

Given the substantial activity of RAB in dogs with lymphoma and the different mechanism of action, resistance, and cumulative AE profiles between RAB and DOX, the goal of our multicenter study was to prospectively evaluate the efficacy and AE profile associated with administration of alternating RAB and DOX at 3-week intervals in dogs with lymphoma.

Materials and Methods

Patient Selection

Studies were carried out with approval of each site's Institutional Animal Care and Use Committee, Clinical Review Board, or both. Signed informed consent was obtained from all owners before study enrollment. Dogs with a cytologic or histologic diagnosis of multicentric lymphoma were prospectively enrolled at 9 sites across the United States from December 2014 to August 2015. Dogs were eligible for inclusion if they had immunophenotype information available and had not received previous treatment. Exclusion criteria included West Highland White terrier breed, a Veterinary Cooperative Oncology Group (VCOG) performance status of <2,[27] absolute neutrophil count <2,000 cells/μL, hematocrit <25%, platelet count <50,000 cells/μL, serum creatinine concentration >2.5 mg/dL, or serum bilirubin concentration exceeding the normal reference range. Dogs with alanine aminotransferase (ALT) or aspartate aminotransferase (AST) activity >3× the normal reference range limit were required to have normal fasting and postprandial serum bile acid concentrations. All dogs had a CBC, serum biochemistry profile (SBC), and urinalysis performed before study entry. Thoracic radiography was strongly recommended but not required. Dogs were assigned an approximate clinical stage based on the World Health Organization clinical staging system.[28]

Treatment Protocol

Rabacfosadine[a] was provided under United States Food and Drug Adminstration INAD# 012-117. Doxorubicin was purchased from commercial vendors by the individual participating sites. Dogs were given alternating doses of RAB and DOX every 3 weeks. Rabacfosadine was administered at a dosage of 1.0 mg/kg as a 30-minute IV infusion on weeks 0, 6, and 12. Doxorubicin was administered at a dosage of 30 mg/m^2 (1.0 mg/kg for dogs weighing <15 kg) as an approximately 20-minute IV infusion on weeks 3, 9, and 15. Concurrent cytotoxic chemotherapy of any kind and concurrent corticosteroids were not allowed. A CBC was performed 1 week after the first RAB and DOX treatments (weeks 1 and 4). All dogs had a physical examination with lymph node measurements, owner history, CBC, SBC, and urinalysis performed at each chemotherapy visit. Thoracic radiographs were recommended at week 15 and approximately every 2 months thereafter in responding dogs. Dose delays, reductions, or both, and supportive treatment for AEs were carried out at the discretion of the attending clinician. Dogs that experienced a complete response (CR) were followed with monthly physical examinations after week 15 until relapse, when they were considered off study. Dogs that experienced partial response (PR) or stable disease (SD) at week 15 were considered off study and censored from analysis at that time. At the time of study withdrawal, dogs were eligible for additional treatment at the discretion of the attending clinician.

Response and Toxicity Evaluation

Response to treatment was determined using the VCOG Response Evaluation Criteria for Peripheral Nodal Lymphoma in Dogs.[29] A CR was defined as the disappearance of all measurable peripheral lymph nodes (ie, returned to a size considered nonpathologic in the judgement of the evaluator). A PR was defined as at least 30% reduction in the sum of widest diameters of peripheral lymph nodes measured at first treatment. Stable disease was defined as <30% reduction or <20% increase in the sum of the widest diameters of the peripheral lymph nodes measured at first treatment. Progressive disease was defined as >20% increase in the sum of the widest diameters of measurable peripheral lymph nodes or the appearance of new lesions.

Adverse events were determined from owner medical history obtained at each visit as well as clinicopathologic evaluation, and graded prospectively based on the VCOG Common Terminology Criteria for Adverse Events v1.1.[27]

Statistical Analysis

Continuous data were expressed as median and range, and categorical data as frequencies and percentages. Objective response rate 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 were withdrawn or lost to follow-up before PD development. Continuous variables were compared between subsets of patients by a 2-tailed, unpaired *t*-test or Mann–Whitney test as appropriate. Categorical variables were compared between groups 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 log-rank analysis. Variables with values of $P \leq .05$ were considered significant. All statistical analysis was performed with a commercial software package[b] .

Results

Patient Population

Fifty-four dogs were prospectively enrolled. Three dogs were excluded from analysis for violation of exclusion criteria. Fifty were evaluable for response assessment and 51 were evaluable for PFI and AE assessment. Information regarding age, weight, approximate stage, substage, and immunophenotype is presented in Table 1. The most common breeds enrolled were mixed-breed dogs (9) and golden retrievers (7). Immunophenotyping was performed by flow cytometry in 22 dogs, immunocytochemistry in 23 dogs, immunohistochemistry in 3 dogs, and PCR for antigen receptor rearrangement in 3 dogs.

Adverse Events

Fifty-one dogs were evaluable for AE assessment. Thirty-three dogs completed the prescribed 6-dose protocol. The most common AEs are summarized in Table 2, and a complete list of AEs is provided in Table S1. Seven dogs experienced a grade 4 AE, and 2 dogs experienced a grade 5 AE. The most common AEs reported were gastrointestinal. Grade 3 gastrointestinal AEs included hyporexia (n = 4), diarrhea with or without hematochezia (n = 4), and weight loss (n = 6). The most common hematologic AE was neutropenia, with 5 dogs experiencing grade 4 AEs; 4 of these were after RAB administration.

Seventeen dogs developed an increase in the activity of at least 1 liver enzyme during the study. In 3 dogs, these increases coincided with the development of PD. The median time to increase liver enzyme activity was 42 days (range, 21–136 days). No dose modifications or drug discontinuations were required in any patients, and the small number of grade 3 or 4 increases in liver enzyme activity resolved spontaneously despite continued chemotherapy.

A total of 13 dogs developed dermatologic changes suspected to be related to treatment. These were characterized as grade 1 in 7 dogs, grade 2 in 5 dogs, and grade 3 in 1 dog. The reactions generally consisted of otitis or pruritic and alopecic or erythematous skin lesions, often on the dorsum. The skin lesions resolved with dose modification or delay and symptomatic treatment including topical treatments, systemic analgesics, H1 blockers, and systemic antibiotics as needed. One dog received prednisone every other day. Several dogs developed hyperpigmentation, especially in the inguinal area, unassociated with other skin changes and requiring no dose modification or specific treatment. Two dogs developed severe dyspnea 130 and 142 days after treatment initiation, which led to euthanasia owing to respiratory signs. Both of these dogs had experienced CR at the time of euthanasia, and pulmonary fibrosis was confirmed on necropsy. A third asymptomatic dog developed a diffuse interstitial lung pattern radiographically interpreted as compatible with fibrosis, which resolved with corticosteroid treatment. All 3 of these dogs had pretreatment radiographs that showed no interstitial lung disease.

Eight RAB dose reductions and 7 DOX dose reductions were utilized for management of AEs. A 20% dose reduction was most commonly employed. No delays in dosing were necessary. Five dogs were withdrawn from the trial by owners as a result of AEs or a perceived decrease in quality of life. Withdrawal occurred a median of 27 days after treatment initiation (range, 19–74 days).

Table 1. Baseline characteristics of patient population (n = 51)

Age (years)	Median (range)	7 (2–14)
Weight (kg)	Median (range)	28.7 (3.3 to 84.1)
Sex (%)	Male	27 (53)
	Female	24 (47)
Approximate stage (2 NR) (%)	3	28 (57)
	4	13 (27)
	5	8 (16)
Substage (%)	a	39 (76)
	b	12 (24)
Immunophenotype (%)	B	43 (84)
	T	8 (16)

NR, not reported.

Table 2. Frequency of common adverse events by grade after rabacfosadine/doxorubicin administration (n = 51).[a]

Event	Grade 1	Grade 2	Grade 3	Grade 4	Grade 5
Gastrointestinal					
Hyporexia	10	4	4		
Dehydration		2			
Diarrhea	17	8	3		
Hematochezia			1		
Vomiting	19	4			
Weight loss	12	16	6		
Constitutional					
Lethargy	12	2			
Hematologic					
Anemia	9				
Neutropenia	5	1	8	5	
Thrombocytopenia	3	4	1	1	
Hepatic					
Increased ALT	6	1	2	1	
Increased AST	5	2	1		
Increased GGT		1			
Increased bilirubin	1	1			
Increased alkaline phosphatase	7	1	3		
Cutaneous/Pulmonary					
Dermatopathy	7	5	1		
Edema	1				
Otitis	6	1			
Pulmonary fibrosis		1			2
Cough	1				

ALT, alanine aminotransferase; AST, aspartate aminotransferase.
[a]Complete list is provided in Table S1.

Acute AEs, occurring within 21 days after administration of the first dose of each agent, were compared between RAB and DOX in 46 dogs receiving at least 1 dose of each agent. Selected comparisons are depicted in Figure 1, and a complete list of acute AEs by agent is provided in Table S2. The incidence of acute diarrhea of any grade was significantly higher after the first RAB dose than after DOX (50% versus 24%, $P = .017$). Other observed differences were not statistically significant.

Patient Outcomes

The overall response rate (ORR) was 84% with 34 (68%) dogs experiencing a CR and 8 (16%) dogs experiencing a PR as their best response. Five dogs had SD and 3 dogs had PD as their best response. The median time to first response was 21 days (range, 21–42 days), and the median time to maximal response was 42 days (range, 21–113 days). The median PFI for all dogs was 194 days (range, 7–435+ days). The median PFI for those dogs that experienced a CR was 216 days (range, 105–435+ days), and the median PFI for those that experienced a PR was 63 days (range, 42–124 days). The median PFI for dogs that experienced SD was 78 days (range 42–93 days). Fourteen dogs were censored from PFI analysis: 7 were in CR at the time of last follow-up (median follow-up time, 220 days), and 6 were withdrawn by the owners because of AEs, decreased quality of life, or lack of response (median follow-up time, 42 days). One was withdrawn on day 26 because of worsening of pre-existing back pain from chronic intervertebral disk disease requiring corticosteroids.

The only factor predictive of response was immunophenotype (ORR = 95% for B cell versus 25% for T cell, respectively. $P = .0001$). Prognostic factors identified on univariate analysis as significant predictors of PFI included substage and immunophenotype (Table 3). The effects of these factors on PFI are depicted in Figure 2.

Discussion

The chemotherapy protocol described in our study was associated with a lower ORR (84%; 68% CR) when compared with contemporary previously reported CHOP protocols (ORR 93–100%; 73–96% CR),[2–9,11] but the response rate appeared equivalent to that reported after single-agent DOX treatment (ORR, 74–87%; 52–78% CR).[22–26] Interestingly, despite the lower ORR, the 194-day median PFI reported in our study appears comparable to PFIs after CHOP-based treatment as reported by others (140–219 days),[2–8] and

Table 3. Factors identified by univariate analysis to be prognostic for PFI.

Factor		n	Median PFI (days)	HR (95% CI)	*P* value
Substage	a	39	203	2.048	.045
	b	12	138.5	(0.84–5.02)	
Immunophenotype	B	43	215	6.62	<.0001
	T	8	43	(0.95–45.99)	

PFI, progression-free interval; HR (95% CI), hazard ratio (95% confidence interval).

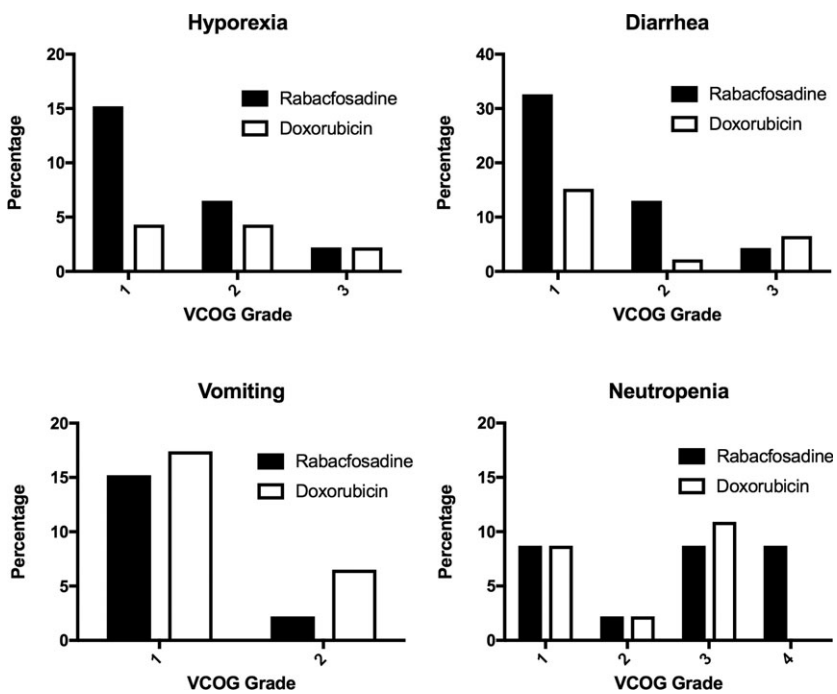

Fig 1. Adverse event frequency after the first dose of rabacfosadine or doxorubicin (n = 46).

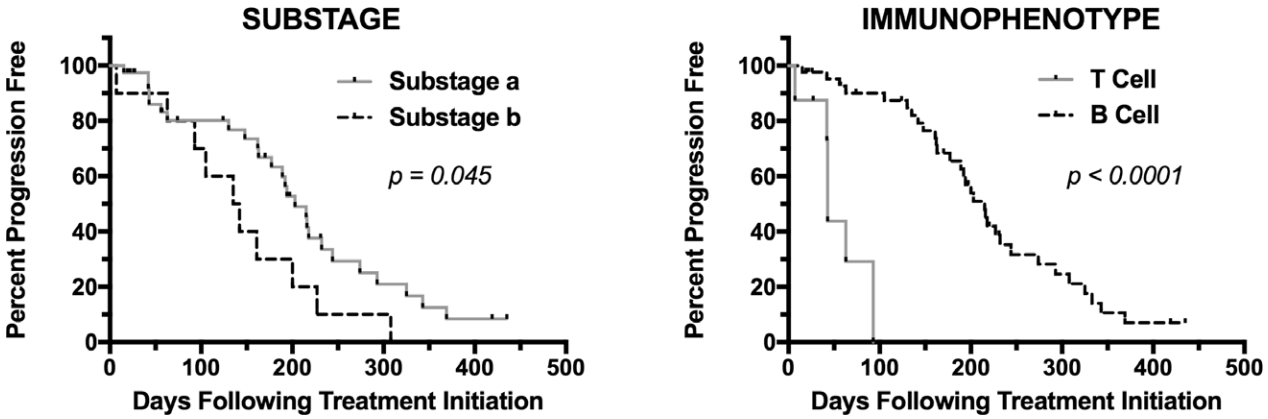

Fig 2. Kaplan–Meier curves depicting effects of substage and immunophenotype on progression-free survival. *P* values indicate univariate log-rank values.

superior to outcomes reported after single-agent DOX (median response durations, 80.5–169 days).[22–26] Many earlier studies failed to report PFI, a measure that incorporates both responding and nonresponding dogs, and rather reported response duration, which excludes dogs experiencing SD or PD as their best responses, making comparisons with these studies challenging. When compared statistically with the raw data from a recently published retrospective study evaluating CHOP-based treatment,[8] there appeared to be no difference in PFI between the entire populations or specific subsets thereof (data not shown), providing further support for the contention that outcomes are relatively similar. A large randomized trial would be necessary to demonstrate this unequivocally.

The median time to first response was 21 days (after the first RAB treatment), which was the first time point for response assessment. Previously published data suggest that clinical responses actually may occur more rapidly. A previous study evaluating RAB in dogs with lymphoma reported a median time to response of 7 days.[19]

In our study, substage and immunophenotype were strong predictors for PFI, similar to previous reports after CHOP-based treatment,[30–34] as was response to treatment. Thus, despite the novel agent being used with DOX in our study, the predictors of response were very similar to those reported after CHOP-based treatment. Clinical stage did not have prognostic relevance, but complete staging was not required for study entry and thus the staging information provided was approximate. Although dogs with substage b lymphoma were included in our study, severely debilitated dogs (VCOG performance status >1) were not eligible for inclusion for ethical reasons.

Gastrointestinal toxicities (self-limiting hyporexia, diarrhea) were the most common acute AEs noted in our study and, with the exception of diarrhea, were reported with relatively equal frequency after the initial DOX and RAB treatments. This is reflected by a relatively equal likelihood of dosage reductions between the 2 drugs. Transient grade 4 neutropenia was noted in 4 dogs after RAB and in 1 after DOX, but only 1 dog developed signs consistent with sepsis or fever requiring hospitalization for treatment after RAB administration. Increases in liver enzyme activity were noted in 17 dogs, but none required dose modification. Many of these increases in liver enzyme activity resolved or improved spontaneously without medical intervention. Sporadic increases in liver enzyme activity have been noted in previous studies with RAB.[19,21] More frequent cumulative AEs included weight loss and dermatopathy, as well as 2 confirmed cases of delayed pulmonary fibrosis. One hypothesis with our study design was that the decreased frequency of admission and decreased total cumulative dose of RAB might result in decreased occurrences of dermatopathy and pulmonary fibrosis, but the frequency of occurrence of both of these AEs was relatively equivalent to what has been observed with single-agent RAB.[19–21] This observation suggests that either frequency of administration or total dose are not relevant predictors of these AEs or, alternatively, that DOX administration could potentiate them to some degree. Future studies should investigate the ability of concurrent corticosteroids to mitigate these cumulative AEs, and careful monitoring of thoracic radiographs and assessment for respiratory signs are warranted.

Five dogs were withdrawn from study by the owners for AEs or a perceived diminished quality of life. Two of these occurred after development of grade 4 neutropenia (1 post-RAB #1, 1 post-DOX #2), 2 were for sustained hyporexia, lethargy, and weight loss after RAB, and 1 was for acute diarrhea, hyporexia, and lethargy after the first DOX treatment. Many of these AEs possibly could have been addressed by dose reductions, prophylactic medication administration, or both rather than study withdrawal. Prophylactic therapies (e.g., antiemetics, antidiarrheals) generally were not used after the first treatment with either agent.

Two limitations of this study were incomplete patient staging and the fact that study participation ended at the time of first relapse, resulting in inconsistent use of rescue treatment and very limited follow-up information for overall survival reporting. Inconsistent rescue treatment and the option of euthanasia make overall

survival time a difficult endpoint to evaluate in veterinary oncology when compared to studies performed in human patients. However, given that alternating RAB/DOX resulted in fewer drugs and doses utilized in the first-line setting, this approach theoretically preserves a larger pool of potentially effective agents for use in the rescue setting, which could improve overall survival. Future studies should explore the efficacy of RAB/DOX retreatment, single-agent RAB retreatment, or CHOP re-induction at relapse. A final limitation is that histopathology was not required for study entry. Although relatively uncommon, some cases of low-grade or indolent lymphoma could have been enrolled, and such dogs might have experienced prolonged PFIs as part of their natural history rather than as a result of treatment.

In conclusion, the alternating RAB/DOX regimen reported here was generally well tolerated and resulted in PFIs comparable to those observed after standard CHOP-based protocols, with substantially fewer treatment visits (6 versus 12–16). Most adverse events were mild or moderate and self-limiting, and similar in spectrum and frequency to those observed after treatment with single-agent RAB. Additional studies are warranted to explore long-term outcome in dogs treated with this combination, as well as with other RAB chemotherapy combinations.

Footnotes

[a] TANOVEA-CA1, VetDC, Fort Collins, CO
[b] Prism v. 6.0b, GraphPad Software, La Jolla, CA

Acknowledgments

Grant support: This study was supported by a grant from VetDC, Inc. DMV, GSP, BSP, MKK, and DR are members of VetDC's Clinical Advisory Board.

Conflict of Interest Declaration: DHT is a consultant for and shareholder in VetDC.

Off-label Antimicrobial Declaration: Authors declare no off-label use of antimicrobials.

References

1. Vail DM, Pinkerton ME, Young KM. Hematopoietic tumors. In: Withrow SJ, Vail DM, Page RL, eds. Withrow & MacEwen's Small Animal Clinical Oncology, 5th ed. St. Louis, MO: Elsevier; 2013:608–678.

2. Elliott JW, Cripps P, Marrington AM, et al. Epirubicin as part of a multi-agent chemotherapy protocol for canine lymphoma. Vet Comp Oncol 2013;11:185–198.

3. Sorenmo K, Overley B, Krick E, et al. Outcome and toxicity associated with a dose-intensified, maintenance-free CHOP-based chemotherapy protocol in canine lymphoma: 130 cases. Vet Comp Oncol 2010;8:196–208.

4. Greenberg CB, Boria PA, Borgatti-Jeffreys A, et al. Phase II clinical trial of combination chemotherapy with dexamethasone for lymphoma in dogs. J Am Anim Hosp Assoc 2007;43:27–32.

5. Hosoya K, Kisseberth WC, Lord LK, et al. Comparison of COAP and UW-19 protocols for dogs with multicentric lymphoma. J Vet Intern Med 2007;21:1355–1363.

6. Zandvliet M, Rutteman GR, Teske E. Prednisolone inclusion in a first-line multidrug cytostatic protocol for the treatment of canine lymphoma does not affect therapy results. Vet J 2013;197:656–661.

7. Burton JH, Garrett-Mayer E, Thamm DH. Evaluation of a 15-week CHOP protocol for the treatment of canine multicentric lymphoma. Vet Comp Oncol 2013;11:306–315.

8. Curran K, Thamm DH. Retrospective analysis for treatment of naive canine multicentric lymphoma with a 15-week, maintenance-free CHOP protocol. Vet Comp Oncol 2016;14(Suppl 1):147–155.

9. Garrett LD, Thamm DH, Chun R, et al. Evaluation of a 6-month chemotherapy protocol with no maintenance therapy for dogs with lymphoma. J Vet Intern Med 2002;16:704–709.

10. Keller ET, MacEwen EG, Rosenthal RC, et al. Evaluation of prognostic factors and sequential combination chemotherapy with doxorubicin for canine lymphoma. J Vet Intern Med 1993;7:289–295.

11. MacDonald VS, Thamm DH, Kurzman ID, et al. Does L-asparaginase influence efficacy or toxicity when added to a standard CHOP protocol for dogs with lymphoma? J Vet Intern Med 2005;19:732–736.

12. Mellanby RJ, Herrtage ME, Dobson JM. Owners' assessments of their dog's quality of life during palliative chemotherapy for lymphoma. J Small Anim Pract 2003;44:100–103.

13. 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;14:2824–2832.

14. Kramata P, Downey KM, Paborsky LR. Incorporation and excision of 9-(2-phosphonylmethoxyethyl)guanine (PMEG) by DNA polymerase delta and epsilon in vitro. J Biol Chem 1998;273:21966–21971.

15. Rose WC, Crosswell AR, Bronson JJ, et al. In vivo antitumor activity of 9-[(2-phosphonylmethoxy)ethyl]-guanine and related phosphonate nucleotide analogues. J Natl Cancer Inst 1990;82:510–512.

16. Naesens L, Hatse S, Segers C, et al. 9-(2-phosphonylmethoxyethyl)-N6-cyclopropyl-2,6-diaminopurine: A novel prodrug of 9-(2-phosphonylmethoxyethyl)guanine with improved antitumor efficacy and selectivity in choriocarcinoma-bearing rats. Oncol Res 1999;11:195–203.

17. Valerianova M, Votruba I, Holy A, et al. Antitumour activity of N6-substituted PMEDAP derivatives against T-cell lymphoma. Anticancer Res 2001;21:2057–2064.

18. Lawrence J, Vanderhoek M, Barbee D, et al. Use of 3′-deoxy-3′-[18F]fluorothymidine PET/CT for evaluating response to cytotoxic chemotherapy in dogs with non-Hodgkin's lymphoma. Vet Radiol Ultrasound 2009;50:660–668.

19. 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;15:3503–3510.

20. Morges MA, Burton JH, Saba CF, et al. Phase II evaluation of VDC-1101 in canine cutaneous T-cell lymphoma. J Vet Intern Med 2014;28:1569–1574.

21. 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.

22. Higginbotham ML, McCaw DL, Roush JK, et al. Intermittent single-agent doxorubicin for the treatment of canine B-cell lymphoma. J Am Anim Hosp Assoc 2013;49:357–362.

23. Lori JC, Stein TJ, Thamm DH. Doxorubicin and cyclophosphamide for the treatment of canine lymphoma: A randomized, placebo-controlled study. Vet Comp Oncol 2010;8:188–195.

24. Mutsaers AJ, Glickman NW, DeNicola DB, et al. Evaluation of treatment with doxorubicin and piroxicam or doxorubicin alone for multicentric lymphoma in dogs. J Am Vet Med Assoc 2002;220:1813–1817.

25. Valerius KD, Ogilvie GK, Mallinckrodt CH, et al. Doxorubicin alone or in combination with asparaginase, followed by cyclophosphamide, vincristine, and prednisone for treatment of multicentric lymphoma in dogs: 121 cases (1987–1995). J Am Vet Med Assoc 1997;210:512–516.

26. Postorino NC, Susaneck SJ, Withrow SJ, et al. Single agent therapy with Adriamycin for canine lymphosarcoma. J Am Vet Med Assoc 1989;25:221–225.

27. 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 2016;14:417–446.

28. Jaffe ES, Harris NL, Diebold J, et al. World Health Organization classification of neoplastic diseases of the hematopoietic and lymphoid tissues. A progress report. Am J Clin Pathol 1999;111:S8–S12.

29. Vail DM, Michels GM, Khanna C, et al. Response evaluation criteria for peripheral nodal lymphoma in dogs (v1.0)–a Veterinary Cooperative Oncology Group (VCOG) consensus document. Vet Comp Oncol 2010;8:28–37.

30. Greenlee PG, Filippa DA, Quimby FW, et al. Lymphomas in dogs. A morphologic, immunologic, and clinical study. Cancer 1990;66:480–490.

31. Marconato L, Stefanello D, Valenti P, et al. Predictors of long-term survival in dogs with high-grade multicentric lymphoma. J Am Vet Med Assoc 2011;238:480–485.

32. Ruslander DA, Gebhard DH, Tompkins MB, et al. Immunophenotypic characterization of canine lymphoproliferative disorders. In Vivo 1997;11:169–172.

33. Teske E. Prognostic factors for malignant lymphoma in the dog: An update. Vet Q 1994;16(Suppl 1):29S–31S.

34. Beaver LM, Strottner G, Klein MK. Response rate after administration of a single dose of doxorubicin in dogs with B-cell or T-cell lymphoma: 41 cases (2006–2008). J Am Vet Med Assoc 2010;237:1052–1055.

Supporting Information

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

Table S1. Frequency of all adverse events by grade following rabacfosadine/doxorubicin administration (n = 51).

Table S2. Frequency of acute adverse effects by grade following the first dose of rabacfosadine or doxorubicin (n = 46).