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Survival Analysis of 97 Cats with Nasal Lymphoma: A Multi-Institutional Retrospective Study (1986–2006)

S.M. Haney, L. Beaver, J. Turrel, C.A. Clifford, M.K. Klein, S. Crawford, J.M. Poulson, and C. Azuma

Background: Feline nasal lymphoma (NLSA) is a condition for which no standard of care exists.

Hypothesis: There is no difference in survival times of cats with NLSA treated with single or multimodality therapy.

Animals: Records from 97 cats diagnosed with NLSA were examined.

Methods: The purpose of this retrospective study was to compare the survival times of cats with NLSA treated with radiation therapy (RT) alone, chemotherapy alone, or RT + chemotherapy and identify potential prognostic variables that affected survival. Cats were grouped according to therapy: RT + chemotherapy (n = 60), RT alone (n = 19), or chemotherapy alone (n = 18).

Results: Survival was calculated with 2 methods. The 1st survival analysis (method A) included all cats, but counted only deaths caused by progressive NLSA. The median survival time (MST), regardless of therapy modality, was 536 days. The 2nd survival analysis (method B) also included all cats and counted all deaths, regardless of cause, as events. The overall MST calculated for all deaths was 172 days. A negative independent prognostic variable identified was anemia ($P < .001$), and positive independent prognostic variables were a complete response to therapy ($P < .001$) and total radiation dose >32 Gy ($P = .03$).

Conclusions and Clinical Importance: There were no significant differences in survival times among the 3 treatment groups but these results suggest that the addition of higher doses of RT to a cat's treatment protocol may control local disease and therefore influence survival.

Key words: Anemia; Chemotherapy; Feline; Radiation.

Lymphoma is the most common neoplasm in cats, accounting for approximately 90% of all hematopoietic malignancies.[1–5] Nasal lymphoma (NLSA), however, is a relatively rare manifestation, accounting for <1% of all feline tumors.[5–10] NLSA in the cat often represents a therapeutic quandary because currently no standard of care exists. Lymphoid neoplasms are not only responsive to chemotherapy but are exquisitely sensitive to radiation, which suggests that localized forms of lymphoma will respond well to treatment with radiation therapy (RT).[9–15] Two studies evaluating a total of 12 cats treated with RT alone found that several cats had extended survival times (4–55 months).[16,9] Evans and Hendrick[13] reported on 3 cats with NLSA treated with combinations of surgery and RT with or without chemotherapy with survival times of 6.3–67 months. A recent study by Sfiligoi et al retrospectively examined 19 cats diagnosed with stage I NLSA treated with a combination of local RT and systemic chemotherapy for 6 months. The median survival time (MST) for this group of cats was 955 days.[17] Lastly, larger studies of cats with lymphoma in various anatomic sites (including the nasal cavity) treated with chemotherapy alone reported survival times ranging from 98 to 358 days.[5,18] To the authors' knowledge, there are no published studies in the veterinary literature that systematically compare the survival benefits conferred by treatment with single or multimodality therapy in a large population of cats with NLSA. The purpose of this multi-institutional retrospective study was to evaluate and compare the survival times of cats diagnosed with NLSA treated with RT alone, chemotherapy alone, or combination therapy. Additionally, response to treatment, duration of response, and potential prognostic factors were evaluated.

Materials and Methods

Medical records of cats with cytologically or histopathologically diagnosed NLSA were reviewed. Data sheets were distributed to all participating institutions. Participating institutions or doctors and number of cases submitted were: Veterinary Oncology Specialties (n = 16), New England Veterinary Oncology Group (n = 14), Med Vet (n = 14), Tufts Cummings School of Veterinary Medicine (n = 13), Red Bank Veterinary Hospital (n = 8), Colorado State University College of Veterinary Medicine (n = 6), The Ohio State University College of Veterinary Medicine (n = 6), East Bay Veterinary Specialists (n = 4), Arizona Veterinary Specialists (n = 4), University of Florida College of Veterinary Medicine (n = 4), North Carolina State University College of Veterinary Medicine (n = 3), Purdue University School of Veterinary Medicine (n = 1), and Drs O'Neill (n = 1), Gallo (n = 1), Holmberg (n = 1), and Norris (n = 1). Inclusion criteria for the study were a cytologic or histopathologic diagnosis of lymphoma within the nasal cavity and negative staging tests for distant disease (stage I according to the World Health Organization clinical staging system[19]). Cats were still classified as having stage I disease if their primary nasal disease extended through the cribriform plate and into the calvarium, as

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this was considered local extension of their nasal disease. Cats were excluded from the study if there was insufficient follow-up information to allow for survival analysis or if they were lost-to-follow-up <1 month after completion of RT. Sufficient follow-up information was defined as at least 1 recheck examination or contact with owners or referring veterinarians regarding the status of the cat after the completion of a prescribed course of RT. Some of the cats included in the present study (45/97) were included in a previous unpublished study by Klein et al.[20] Information collected from medical records included signalment, presenting clinical signs, duration of clinical signs, routine clinical staging results (CBC/serum biochemistry, abdominal ultrasound findings, local and regional lymph node aspirates, bone marrow aspirates, cytologic or histopathologic findings, and radiologic findings), results of retroviral testing, and date of initiation of treatment, details concerning individual treatment regimens were collected including machine type and energy, minimum total tumor dose and RT fractionation scheme as well as chemotherapy protocol, response to treatment, duration of response, and survival times. In some cases, detailed morphologic descriptions of tumor samples allowed grades to be assigned according to the National Cancer Institute working formulation of feline lymphoproliferative diseases (NCI WF).[21,22] Lymphomas were classified as low, intermediate, or high grade.

Cats that received only prednisone were considered to have received chemotherapy and were grouped accordingly. For statistical analysis, cats were grouped according to RT protocol intent (ie, palliative or definitive). Because of variability in RT protocols (fraction size, number of fractions, and total dose), cats were assigned to the palliative-intent group if their protocol met at least 2 of the following criteria: if they received RT treatments <3 times a week, if the individual fraction size was ≥4 Gy, or if the total dose was ≤32 Gy. If the protocol did not meet at least 2 of these criteria, they were assigned to the definitive-intent group.

The response to treatment (complete response [CR], partial response, stable disease, and progressive disease) was a subjective assessment by the submitting clinician based on partial or complete resolution of clinical signs such as termination of nasal discharge, reduction in external masses, and lessening of sneezing. Stable disease was defined as no change in clinical signs or physical appearance (ie, no change in external mass) and progressive disease was defined as a worsening of clinical signs. Duration of response was defined as time from initial response to treatment until return of clinical signs related to disease progression.

Outcome after treatment, which included duration of response and development of local recurrence or distant disease, was subjectively determined by the submitting clinician based on results of physical examination, return of clinical signs, and diagnostic imaging. In cats in which follow-up diagnostic imaging was not performed, progressive unilateral or bilateral mucoid nasal discharge, facial deformity, or exophthalmos typically was taken to represent a recrudescence of lymphoma whereas nonmucoid or clear chronic nasal discharge typically was classified as chronic radiation-induced rhinitis. In addition, response to antibiotic therapy often was used by clinicians to differentiate between return of disease and upper respiratory tract infections or rhinitis. Cause of death was reported by the submitting clinician or by the primary care veterinarian. Early death was defined as death <1 month after initiation of treatment, either RT or chemotherapy. Additional follow-up information was gathered from the medical records or by phone calls to referring veterinarians and owners.

Statistical Analysis

Cats with NLSA treated with RT, chemotherapy, or combination therapy that met the inclusion criteria were included in the survival analysis. Using the Kaplan-Meier product limit method, between-group differences in survival were assessed with respect to 2 different event variables. First, only deaths attributable to local disease progression were considered events, and deaths due to other causes and cats that were still alive or lost-to-follow-up were censored (method A, disease-free survival). Second, all deaths were considered events, including both lymphoma and nonlymphoma-related deaths, and only cats alive at the time of analysis or lost-to-follow-up were censored (method B, overall survival). Statistical testing was performed on both the overall survival group and the disease-free survival group, but unless specifically noted, results are reported only for the overall survival group. Survival time was defined as the time, in days, from the 1st treatment, either chemotherapy or RT, to the time of death. The date of the last contact served as the date of censor for cats that were lost-to-follow-up, and the date of statistical analysis was the date of censor for cats that were still alive. Fisher's exact test was used to identify predictors of response to treatment. Statistical significance of categorical variables such as sex, presenting clinical signs (anemia, sneezing, facial deformity, epistaxis, mucoid nasal discharge, ocular discharge, sterterous breathing, dyspnea, buphthalmos, and epiphora), serum biochemistry and CBC abnormalities (anemia, neutrophilia, and hyperglobulinemia), the inclusion of RT, chemotherapy or both in a treatment protocol, computed tomography (CT) and radiographic findings (the presence of bony lysis or invasion through the cribriform plate), and histopathology were assessed for any between-group differences with regard to survival by the Wilcoxon test. Bivariate analysis to assess for prognostic value of additional variables was performed using the Kaplan-Meier product limit method. Variables tested for influence on survival included those tested for between-group differences, as well as signalment, duration of clinical signs before presentation, retroviral status, total radiation dose, and a definitive or palliative RT protocol. The 3 treatment groups were also compared with respect to duration of response by nonparametric analysis of variance (Kruskal-Wallis test). Variables with P values $\leq .05$ were considered statistically significant. Multivariate analyses were performed using the Cox proportional hazards model for any variables that were significant on the bivariate analysis. All analyses were conducted using SAS version 9.1.[23]

Results

Patient Population

A total of 118 cats with nasal lymphoma presented to 16 referral institutions between the years of 1986 and 2006 were identified and 97 cats met the inclusion criteria. Patient characteristics were as follows: 62 cats were castrated males and 35 were spayed females with a median age of 10 years (range, 2–19 years). There were 60 domestic shorthair cats, 20 domestic longhair cats, 11 Siamese purebreds, and 6 other types of purebred cats. The median weight at presentation was 4.8 kg (range, 1.9–10 kg). Results of retroviral testing were available for 51 cats, with 41 negative for FeLV and FIV, 5 positive for FeLV, and 6 positive for FIV. Retroviral status was unknown for 46 cats.

Presenting Clinical Signs and Diagnostic Tests

Cats commonly presented with 1 or more of the following clinical signs: purulent or mucoid nasal discharge (n = 57), facial deformities (n = 22), epistaxis (n = 21), and sneezing (n = 20). Other presenting clinical signs included sterterous breathing (n = 18), anorexia (n = 14), dyspnea (n = 11), buphthalmos (n = 11), and epiphora (n = 9). The duration of clinical signs before

1st examination was known for 64 cats, and the median duration was 60 days (range, 3–1,095 days).

Twenty-three cats had records of full staging procedures (including radiographs with or without CT of the nasal cavity, chest and abdominal radiographs, serum biochemistry, CBC, and abdominal ultrasound examination) and 74 cats had partial staging procedures recorded (including combinations of the above). Imaging was performed in 64 cats and consisted of skull radiographs (n = 35), CT (n = 27), and magnetic resonance imaging (n = 2). Bony lysis was noted in 30 studies (CT, n = 17 and skull radiographs, n = 13). Seven cats had evidence of local invasion of their tumor through the cribriform plate and into the calvarium. Laboratory abnormalities at presentation included: anemia (8/70 cats; median hematocrit, 24%; range, 15–35%), neutrophilia (14/65 cats; median, 18.2 $\times$ 10^9 dL; range, 14.7–38.0 $\times$ 10^9 dL), and hyperglobulinemia (16/64 cats; median, 5.7 g/dL; range, 5.0–6.9 g/dL).

Five cats had a diagnosis of lymphoma based on cytology and 92 cats had a histopathologic diagnosis. A histopathologic grade was assigned for 77 cats. In 15 cats, the pathology report did not specifically state a grade, but a detailed morphologic description of the cellular and histopathologic appearance allowed a grade to be assigned. Eight lymphomas were classified as low grade, 19 as intermediate grade, and 39 as high grade. For the purposes of statistical analysis, low and intermediate grade were grouped together and high grade was considered separately to evaluate for differences in survival among the histopathologic subgroups. Further analysis considered intermediate and high-grade lymphoma grouped together and low-grade lymphoma was considered separately. Histopathologic grade did not have a significant bearing on overall survival time regardless of grouping (P = .85).

Survival Analysis

Cats were grouped according to therapy received: RT and chemotherapy (n = 60), RT alone (n = 19), and chemotherapy alone (n = 18). Seventy-nine cats had died or been euthanized at the study end point. Deaths were categorized as: due to progressive local disease (n = 36), due to both progressive local disease and distant nonnasal lymphoma (n = 5), due to distant nonnasal lymphoma (n = 5), due to other disease unrelated to lymphoma (n = 24), or due to unknown causes (n = 9). Seven cats were still alive at the study end point and 11 were lost-to-follow-up; these 18 cats were censored in survival analyses. Forty-one cats died of progressive local disease, and 56 cats were censored for this survival analyses (method A). The MST for cats dying of progressive disease was 536 days (range, 12–1,917 days; 95% CI, 268–1,431 days). The Kaplan-Meier estimate computes survival at a given time based on the number of subjects still "at risk" at that time. Thus, subjects who have died previously as well as subjects who have been censored are not in that risk set.[23] Consequently, MST could be estimated despite censoring $\geq$50% of the subjects. Method A then was reevaluated to include all cats that died of lymphoma, either nasal or distant disease, as it was most likely that distant nonnasal lymphoma represented spread of the lymphoma and not denovo cancer. When the 5 cats that died of distant lymphoma were included in the group that died of progressive NLSA, the MST was 473 days (range 12–1,917; 95% CI, 218–1,131days). In survival analysis method B, 79 cats died and 18 cats were censored. The overall MST, including all deaths, was 172 days (range, 4–3,749 days; 95% CI, 146–320 days). When the cats that received prednisone only were omitted from the survival analysis, the survival times were not significantly different (P = .14).

In the 1st survival analysis, which included only deaths caused by progressive local disease as events (method A, disease-free survival) the 3 treatment groups were individually evaluated with the following results: the MST for group 1 cats (chemotherapy and RT) was 473 days (range, 57–1,015 days; 95% CI, 192–1,015 + days). The MST for cats in group 2 (RT alone) was 1,431 days (range, 12–1,917 days; 95% CI, 268–1,917 + days). The MST for cats in group 3 (chemotherapy alone) was 320 days (range, 17–1,131days; 95% CI, 92–1,015 + days). Given the available follow-up observation period and the small number of cats in each group, we were unable to estimate exact upper confidence bounds. In this survival analysis, no significant difference was detected among the 3 treatment groups (P = .41). The overall 1- and 2-year survival rates in this group were 56.4 and 44.2%, respectively (see Fig 1).

In the 2nd survival analysis, which included all deaths as events (method B, overall survival), evaluation of the 3 treatment groups yielded the following results: The MST for group 1 cats (chemotherapy and RT) was 174 days (range, 29–3,749 days; 95% CI, 150–388 days). The MST for group 2 cats (RT alone) was 456 days (range, 12–2,169 days; 95% CI, 75–1,511 days). The MST for group 3 cats (chemotherapy alone) was 116.5 days (range, 4–1,131 days; 95% CI, 49–300 days). No significant difference was detected among survival times for these groups (P = .07). The overall 1- and 2-year survival rates were 37.7 and 25.2%, respectively (see Fig 2).

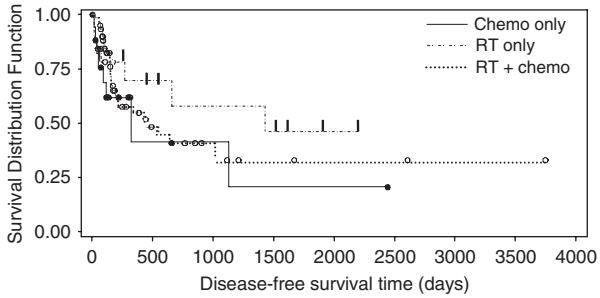

Fig 1. Disease-free survival: Kaplan-Meier survival curve illustrating differentiation of the 3 groups according to treatment modality. Radiation therapy (RT) and chemotherapy median survival time (MST) 473 days, RT alone MST 1,431 days, chemotherapy alone MST 320 days (P = .41). Only deaths caused by progressive disease were considered events in this survival analysis. Circles and hatch marks indicate cats censored for analysis.

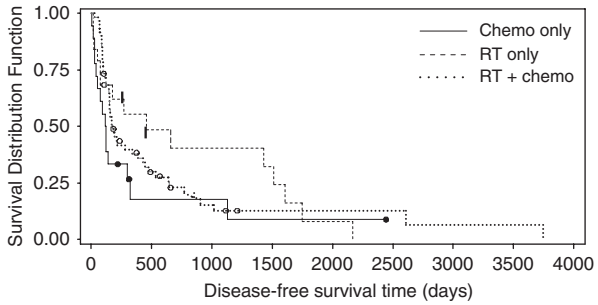

Fig 2. Overall survival: Kaplan-Meier survival curves differentiating the 3 groups according to treatment modality. Radiation therapy (RT) and chemotherapy median survival time (MST) 174 days, RT alone MST 456 days, chemotherapy alone MST 116.5 days ($P = .07$). All deaths were considered events in this analysis. Circles and hatch marks indicate cats censored for analysis.

The status of the cats for the 3 treatment groups was as follows: In the chemotherapy and RT group (n = 60), 21 cats (35%) died from progressive local disease, 5 cats (8.3%) died of both progressive local disease and distant lymphoma (ultrasound-guided fine needle aspirates indicated involvement of the liver, kidney, mesenteric lymph nodes, and spleen), 4 cats (6.6%) died of distant lymphoma alone (gastrointestinal lymphoma; n = 1, renal lymphoma; n = 1, and multiple abdominal organ involvement; n = 2), 12 cats (20%) died of causes other than lymphoma, 7 cats (11.6%) died of unknown causes, 5 cats (8.3%) were lost-to-follow-up, and 6 cats (10%) were still alive at the time of data analysis. In the RT only group (n = 19), 7 cats (36.8%) died of progressive local disease, 8 cats (42.1%) died of causes other than lymphoma, and 4 cats (21%) were lost-to-follow-up. In the chemotherapy only group (n = 18), 8 cats (44.4%) died of progressive local disease, 1 cat (5%) died of renal lymphoma, 4 cats (22.2%) died of causes other than lymphoma, 2 cats (11.1%) died of unknown causes, 2 cats (11.1%) were lost-to-follow-up, and 1 cat (5%) was still alive. A total of 36 cats (37.1%) died of progressive local disease alone, 5 cats (5.1%) died of progressive local disease plus distant lymphoma, and 5 cats (5.1%) developed distant lymphoma with no local progression of their nasal disease. Of the cats that developed systemic disease, 2 (20%) were positive for FeLV and all cats had received systemic chemotherapy. Seven cats in the current study died an early death. The causes for early death included progressive local disease (n = 4), development of distant nonnasal lymphoma (n = 1), progressive local disease and distant lymphoma (n = 1), and other non-lymphoma-related causes (n = 1). Of these cats, 3/7 (43%) were anemic at presentation.

Prognostic Factors

Regarding presenting clinical signs, anemia ($P < .001$) and anorexia ($P = .05$) at presentation were significantly associated with decreased survival times on bivariate analysis. The MSTs for cats with (n = 8) and without (n = 70) anemia at presentation were 81and 268 days, respectively. The MSTs for cats with (n = 14) and without (n = 64) anorexia at presentation were 135 and 320 days, respectively. Intracranial extension of disease did not significantly influence survival ($P = .26$) nor did any other presenting clinical sign or diagnostic testing abnormality.

Because of the differences in protocols among submitting institutions, the chemotherapeutic regimens varied widely. The most commonly used chemotherapeutics were combinations of L-asparaginase[a] (range, 200–400 IU/kg), vincristine[b] (range, 0.025–0.75 mg/m^2), cyclophosphamide[c] (range, 50–300 mg/m^2), prednisone (range, 0.5–2 mg/kg), doxorubicin[d] (range, 20–25 mg/m^2), mitoxantrone[e] (range, 3–5 mg/m^2), and lomustine[f] (range, 30–60 mg/m^2). Cats typically were treated on a weekly basis with a multidrug protocol.

The use of any chemotherapy in a cat's protocol, regardless of RT, also was evaluated, and no statistically significant survival benefit was found ($P = .47$). Cats that received any chemotherapy in their treatment regimen survived a median of 157 days (range, 4–3,749 days; 95% CI, 136–282 days) and cats that did not receive chemotherapy survived a median of 456 days (range, 12–2,169 days; 95% CI, 75–1,511 days). Because of the variability in chemotherapy regimens among submitting institutions, individual chemotherapy drugs and specific protocols could not be evaluated. Three cats received single agent prednisone with or without RT. Two cats received prednisone alone with no other therapy and were included in the chemotherapy only group. These cats survived 49 and 120 days and died of unknown causes. The 2nd cat received prednisone and RT, survived 67 days and died due to renal lymphoma. All cats in the RT and chemotherapy group either received chemotherapy and RT concurrently (n = 51), or chemotherapy after completion of RT (n = 9).

Regarding cats that received any RT in their treatment protocols, regardless of initial grouping by treatment modality, the overall median number of fractions administered was 5 (range, 2–19), the overall median fraction size was 4 Gy (range, 3–10 Gy), and the overall median total dose was 32 Gy (range, 10–57 Gy). Forty-two cats were treated using a 4- or 6-MV linear accelerator, 23 cats were treated using orthovoltage units, and 12 cats were treated using Cobalt-60 units. In the 2 groups of cats that received RT as part of their protocol, the median dose of radiation for group 1 (chemotherapy and RT group) was 32 Gy (range, 10–57 Gy) and the median dose of radiation for group 2 (RT only) was 30 Gy (range, 15–48 Gy). The addition of any RT to a cat's protocol was evaluated for an influence on survival, and was found to significantly improve overall survival in the bivariate analysis ($P = .02$). Cats receiving any RT in their treatment protocols survived a median of 192 days (range, 12–3,749 days; 95% CI, 152–429 days), compared with a median of 116 days (range, 4–2,445 days; 95% CI, 49–300 days) for cats not treated with RT. The total dose of radiation administered to each cat was evaluated and was found to be predictive of the duration of response to treatment ($P = .05$). More specifically, total doses of $\geq$32 Gy were found to be associated with longer MSTs ($P = .01$). Cats that received $\geq$32 Gy, survived a median of 388 days (range, 6–3,749 days; 95% CI, 172–1,015 days) compared with cats that received $\leq$32 Gy,

which had a MST of 170 days (range, 12–2,610 days; 95% CI, 97–268 days). For statistical analysis, cats receiving RT were assigned to either a palliative intent (n = 41) or definitive intent (n = 38) group. The MST for the definitive intent group was 388 days (range, 18–3,748 days; 95% CI, 152–906 days) and the MST for the palliative intent group was 171 days (range, 12–2,610 days; 95% CI, 123–268 days). There were no differences in survival between the definitive and palliative intent groups ($P = .07$). Additionally, there were no differences in survival among the cats treated with different machine energies (orthovoltage versus Cobalt-60 versus 4- or 6-MV linear accelerators) ($P = .54$). Lastly, none of the factors that were found to have prognostic value were significantly changed when the 5 cats that died of distant lymphoma alone were reevaluated as part of the group of cats that died of progressive NLSA.

Response and Outcomes

Fifty-one cats achieved a CR and 18 cats achieved a partial response for an overall response rate of 70%. The chemotherapy and RT group had a response rate of 82%, the RT only group had a response rate of 93% and the chemotherapy only group had a response rate of 67%. Response to therapy was significantly associated with survival time ($P < .001$). Cats that achieved a CR (MST: 536 days; range, 17–3,749 days; 95% CI, 300–906 days) survived significantly longer compared with cats that achieved PR (MST: 120 days; range, 73–386 days; 95% CI, 97–157 days) ($P < .001$). Four cats achieved SD and survived a median of 90 days (range, 27–1,131 days; 95% CI, 27–1,131 days). Ten cats developed PD and survived a median of 53 days (range, 4–155 days; 95% CI, 29–85 days).

Duration of response was evaluated for all cats and for each individual treatment group. The duration of response was known for 83/97 cats. The overall median duration of response for all groups was 120 days. The median duration of response for cats receiving chemotherapy and RT was 120 days (range, 0–1,812 days; 95% CI, 84–180 days), 264 days for cats receiving RT only (range, 0–1,775 days; 95% CI, 74–471 days), and 73 days for cats receiving chemotherapy only (range, 0–2,445 days; 95% CI, 19–219 days); these differences were not statistically significant ($P = .09$). Only 2 cats had a documented follow-up CT scan performed, both of which had no evidence of disease. The variables that were significant on the bivariate analyses for survival are summarized in Table 1.

After multivariate analyses, when accounting for all other prognostic variables and evaluating all deaths as events, anemia and a CR to therapy remained statistically significant, and anorexia was no longer significant ($P = .79$) (see Fig 3).

Additionally, in the population of cats that died of known PD (local disease), the addition of doses >32 Gy was significant ($P = .03$) (see Fig 4). The results of multivariate analyses are summarized in Table 2.

Table 1. Prognostic variables using bivariate analyses.

Variable Influencing Survival	Number of Cats (%)	Median Survival Time (Days)	*P* Value
Anemia	8 (8.3)	81	< .001
Anorexia	14 (14.4)	135	.05
Addition of any RT to protocol	79 (81.4)	192	.02
RT dose >32 Gy	32 (32.9)	388	.01
CR achieved from therapy	52 (53.6)	536	< .001

RT, radiation therapy; CR, complete response.

Discussion

In this study, there were no differences in survival times among the 3 treatment groups in cats with stage I disease, using either survival method. The overall MSTs were 536 and 172 days, using survival analyses A and B, respectively. When the 5 cats dying of distant lymphoma were not censored from method A survival analysis, the MST was 473 days. The median age of cats in this study was 10 years, which is consistent with the median age reported by other studies evaluating cats with NLSA (range 8.4–13 years).[5,9,10,17] There were nearly twice as many males as females, although this difference was not related to survival.

Two different survival analyses were performed in the current study; the 1st analysis (method A, disease free survival) considered only deaths caused by progressive disease as events. This analysis evaluated fewer cases overall but included only identifiable cases of death caused by progressive local disease. Although this analysis is a less conservative method of evaluating survival, it may be more accurate in reflecting true survival times. The 2nd survival analysis (method B, overall survival) was performed as a comparison with the 1st because it is considered a more conservative approach to evaluating survival data in client-owned cats for which follow-up information may be lacking and a precise cause of death cannot be determined.

Regarding doses of radiation received: the RT alone group received a median total dose of 30 Gy, whereas the chemotherapy and RT group received a median total dose of 32 Gy. The MST of the 2 groups was not signifi-

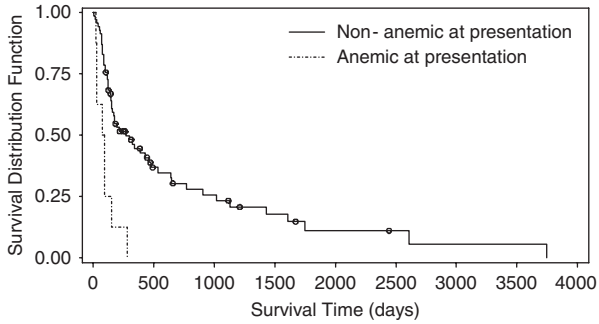

Fig 3. Kaplan-Meier survival curves depicting the differences in cats with and without anemia at presentation. Anemic cats median survival time (MST) 81 days, nonanemic cats MST 269 days ($P <$ .001). Circles indicate cats censored for analysis.

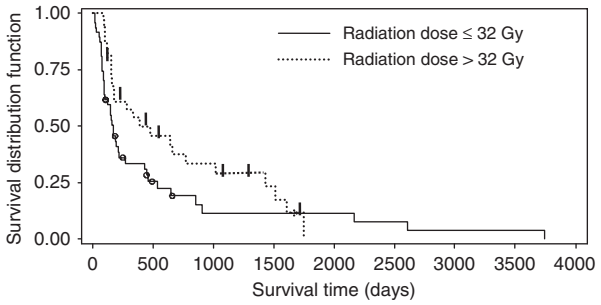

Fig 4. Kaplan-Meier survival curves comparing disease-free survival between the groups of cats that received >32 and ≤32 Gy. Circles and hatch marks indicate cats censored for analysis.

cantly different from each other in either survival analysis. A more aggressive approach utilizing combination therapy may have been taken in animals that were assumed to have worse disease (eg, facial deformity, bony lysis seen on radiologic studies, brain involvement). This also may have introduced a potential source of bias into the study and may have affected survival times.

Prognostic factors identified in the bivariate analyses were anemia, anorexia, and the addition of any RT to a cat's protocol, although the power to detect differences among groups was low because of the small numbers of patients in the chemotherapy and RT only groups. A CR to therapy and doses >32 Gy were also found to have a significant positive influence on survival in the bivariate analysis (MST 388 days for >32 Gy versus 170 days for ≤32 Gy). Also, the total dose of radiation received was a positive predictor of duration of response to therapy.

Overall, when all prognostic variables were accounted for by multivariate analyses, anemia and a CR to therapy remained statistically significant. Radiation dose was significant in the analysis that evaluated cats that died of known PD. No other variables evaluated had any bearing on prognosis, including histologic grade, bony lysis, or the addition of any chemotherapy to a cat's protocol. Invasion through the cribriform plate was not shown to be prognostic in this study, which is in contrast to the findings of Sfiligoi et al, who examined 19 cats and found that cats with tumor invasion through the cribriform plate had a MST of 76 days.[17] In the present study, 27 cats were diagnosed using CT scan, and the number of cats with invasion through the cribriform plate may be underestimated. Additionally, the number of cats with tumor-associated bony lysis of the nasal cavity also may be low, because 35 cats were diagnosed with skull radiographs but only 13 cats were shown to have bony lysis; this method of evaluation has a low sensitivity for detecting destruction of the turbinates and other surrounding bone. Thirty-three cats had no records of diagnostic imaging having been performed, which may lead to a further underestimation of cribriform plate invasion and bony lysis. It is also possible that evaluating certain parameters such as cellular immunophenotype, mitotic index, nuclear size, nucleolar size and number, and percent necrosis of the lymphoma cells may help to define other histologic subclassifications that may prove to have an association with prognosis.[21,24] Also, standardization in chemotherapy protocols may aid in establishing a correlation with prognosis, but the role of chemotherapy in the treatment of NLSA remains undefined.

Table 2. Independent variables from the multivariate analyses, corresponding hazard ratios with 95% confidence intervals plus associated *P* values.

Independent Variable	Hazard Ratio (95% CI)	*P* Value
Anemia	6.698 (2.25–19.93)	< .001
CR achieved from therapy	0.04 (0.01–0.12)	< .001
Addition of any RT to protocol	0.65 (0.37–1.15	.13
RT dose >32 Gy (including all deaths)	0.56 (0.29–1.09)	.08
RT dose >32 Gy (including only PD deaths)	0.37 (0.15–0.93)	.03
Anorexia	1.178 (0.376–3.691)	.79

RT, radiation therapy; CR, complete response.

RT has a clearly defined role in the management of NLSA. Nearly half of the cats in this study treated with RT alone died of causes unrelated to lymphoma. This observation underscores the importance of improved local control leading to increased survival times. The results of this study also suggest that higher total doses (≥32 Gy) of RT may confer an additional survival benefit to cats. However, it remains unclear which fractionation protocol is ideal for the treatment of NLSA. In the present study, there was no significant difference in survival times among cats that were treated with different RT protocols (ie, definitive or palliative intent), although small numbers of cats were treated with each protocol. Both the duration of response to radiation and overall survival times may be affected by the fractionation scheme used. Fractional dose typically is adjusted when considering the impact of long-term adverse effects (ie, larger doses per fraction may increase the risk for long-term adverse effects). The current study suggests that administering a higher total dose of radiation may be more appropriate for achieving long-term control. Therefore, using a definitive-intent protocol with smaller fraction sizes may improve survival times while minimizing potential late complications. A comparison of hypofractionated protocols (5–8 Gy) administered 1–2 times a week with more standard fraction sizes (2.5–3 Gy) given daily has not been performed. Traditionally, hypofractionated protocols have been used in a palliative setting when the overall prognosis is assumed to be poor (ie due to size or extent of local disease or high probability of metastasis). Because this disease process and its associated prognostic variables remain largely undefined, assumptions about overall prognosis should not be made when choosing a radiation or chemotherapy protocol based on assessment of local disease. A limitation of the current study is the lack of reported acute and chronic adverse effects, which also may influence a clinician's decision to use a conventional fractionation scheme or a more hypofractionated protocol.

Lymphocytes and lymphoblasts are exquisitely sensitive to ionizing radiation and undergo apoptosis relatively soon after receiving a small dose of radiation.

Human B-cells are more sensitive to radiation than are T-cells, although this association has not been proven in cats. The majority (17 of 18) of feline NLSA were of B-cell type in 1 study (12 diffuse large B-cell lymphoma, 4 lymphoblastic B-cell lymphoma, and 1 follicular B-cell lymphoma), although other studies have reported higher proportions of T-cell along with B-cell NLSA.[25–28] Although the immunophenotype has not been shown to be a prognostic factor, determining the immunophenotype of a cat's NLSA may provide some information as to an individual cat's radiosensitivity and thus be predictive of duration of response to therapy, because T-cells are more radioresistant in people.[29] Only 3 cats in the present study had records of immunohistochemical staining for B cells (CD79a) or T cells (CD3) performed, and all 3 were confirmed to be of B-cell origin. One cat was still alive at the study end point with no evidence of local or distant recurrence, the 2nd cat was euthanized due to local recurrence, and the 3rd cat died from renal failure unrelated to lymphoma.

There were a small number of cats in the current study in which treatment failed <1 month after the initiation of therapy. These cats died primarily of progressive local disease and nearly half were anemic at presentation. Information regarding the type of radiation planning (manual plan versus 3-dimensional computerized treatment plan) was not available for the cats that were treated with radiation. As a result, cats lacking CT, and therefore a 3-dimensional treatment plan, may not have had an adequate planning target volume, potentially resulting in an omission of regions of their tumor. This may have resulted in early progression of some cats' local disease resulting in early death. This dichotomy in the population of cats was similar to the findings of Sfiligoi et al,[17] in which a small subpopulation of cats died early in the course of disease (MST $<$ 6 months) whereas the remainder of the cats derived long-term benefit from combination therapy. Anemia was an independent prognostic variable in the study. A cat's hematologic status often can be influenced by the chronicity of disease along with other factors, which include inflammation, infection, or neoplasia. Hematologic stress can be induced by the release of cytokines in response to cellular injury. NLSA may be a more indolent form of lymphoma,[10] and therefore, anemia may be predictive of the subclinical duration of the disease before diagnosis. A CR to therapy was also found to be an independent prognostic variable, corroborating other studies' findings that evaluated cats with various anatomic forms of lymphoma, and indicating that lymphoma responds in a heterogenous manner as a whole.[2,5,15,27]

It is unclear why some but not all NLSA in this study progressed to distant locations. A total of 10 (10.3%) cats in the current study died due to either local plus distant lymphoma or distant lymphoma alone. In Sfiligoi's study, which identified cats with NLSA from the years 1987–2004, 7/19 (36.8%) cats developed lymphoma at local or distant sites and none were positive for FeLV.[17] In the current study, 5 cats were known to be FeLV positive. Two of these 5 cats developed distant lymphoma (40%). Although FeLV status was not found to correlate with survival, retroviral status was known for only slightly over half of the population of cats in this study and the actual prevalence of FeLV may have been underestimated. Also, in the present study, there were so few cases infected with FeLV that there was insufficient power to detect differences in survival between infected and uninfected cats. The incidence of FeLV in the United States has been estimated to have declined by 50% in the past 20 years, from an incidence of 1–3% of the total cat population.[23,30,31] Four of 5 cats with FeLV in the present study were treated between the years of 1991 and 1993, and may have been more representative of the typical population of cats during that time period. The 5th cat was treated in 2006. Also, regarding the occurrence of distant disease, staging varied among institutions. Abdominal ultrasound studies and regional lymph node aspirates were not performed on every cat, and it is possible that stage I disease was overestimated based on the absence of suspicious clinical signs or on a normal abdominal and lymph node palpation. More thorough and standardized staging may aid in identifying those cats that truly have stage I lymphoma and therefore a better prognosis.[2]

This study must be interpreted with caution, because many confounding variables were involved. Because of the multi-institutional, retrospective nature of the study, it is difficult to draw definitive conclusions as to the benefit conferred by the inclusion of chemotherapy or RT, because the protocols for both varied leading to a heterogeneous treatment population. Bias was inherent, because some owners may have chosen to pursue one therapy over another for a variety of reasons, including the cost and duration of therapy, or preconceived notions of quality of life for their cats. Clinician bias in therapy selection based on anecdotal results and personal experience represents another potential source of error into the study. In addition, the small numbers of cats in the subgroups limited the power of the study.

In conclusion, both RT and chemotherapy have benefits in treating NLSA. Results of this study indicated that there are no significant differences in survival times of cats in the three treatment groups, leading again to the question of therapy selection. It is clear, however, that addition of RT is pivotal in controlling local disease, and higher doses of radiation may lead to longer survival times. Elucidation of potential risk factors for the development of distant disease may help guide treatment protocols for cats at higher risk of developing metastases and also for cats with local disease alone. The role of chemotherapy in NLSA still is undefined but it should be considered, because multimodality therapy typically is the optimal choice for treating local disease plus disseminated spread or microscopic disease. A randomized prospective study is needed in order to establish a standard of care for NLSA in the cat.

Footnotes

[a] Elspar, Merck and Co Inc, Whitehouse Station, NJ
[b] Oncovin, Merck and Co Inc

[c] Cytoxan, Bristol Myers Squid Co, Princeton, NJ
[d] Adriamycin, Pharmacia and Upjohn, Kalamazoo, MI
[e] Novantrone, Soreno Inc, Rockland, MA
[f] CeeNU, Bristol Lab Oncology Products, Princeton, NJ

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References

1. Dorn CR, Taylor DON, Hibbard HH. Epizootiologic characteristics of canine and feline leukemia and lymphoma. Am J Vet Res 1967;28:993–1001.

2. Mooney SC, Hayes AA, MacEwen EG, et al. Treatment and prognostic factors in lymphoma in cats: 103 cases (1977–1981). J Am Vet Med Assoc 1989;194:696–699.

3. Couto CG. Oncology. In: Sherding RG., ed. The Cat: Diseases and Clinical Management, 1st ed. New York: Churchill Livingstone; 1989:589–647.

4. Carpenter JL, Andrews LK, Holzworth J, et al. Tumors and tumor-like lesions. In: Holzworth J., ed. Diseases of the Cat, 1st ed. Philadelphia, PA: WB Saunders; 1987:406–596.

5. Teske E, van Straten G, van Noort R, Rutteman G. Chemotherapy with cyclophosphamide, vincristine, and prednisone (COP) in cats with malignant lymphoma: New results with an old protocol. J Vet Intern Med 2002;16:179–186.

6. Madewell BR, Priester WA, Gillette EL, Snyder SP. Neoplasms of the nasal passage and paranasal sinuses in domesticated animals as reported by 13 veterinary colleges. Am J Vet Res 1976;37:851–856.

7. Legendre AM, Krahwinkel DJ, Spaulding KA. Feline nasal and paranasal sinus tumors. J Am Anim Hosp Assoc 1981;17: 1038–1039.

8. Cotter SM. Treatment of lymphoma and leukemia with cyclophosphamide, vincristine and prednisone. II. Treatment of cats. J Am Anim Hosp Assoc 1983;19:166–172.

9. Straw RC, Withrow SJ, Gillette EL, McChesney AE. Use of radiotherapy for the treatment of intranasal tumors in cats: Six cases (1980–1985). J Am Vet Med Assoc 1986;189:927–929.

10. Elmslie RE, Ogilvie GK, Gillette EL, McChesney S. Radiotherapy with and without chemotherapy for localized lymphoma in 10 cats. Vet Radiol 1991;32:277–280.

11. Thrall DE, Harvey CE. Radiotherapy of malignant nasal tumors in 21 dogs. J Am Vet Med Assoc 1983;183:663–666.

12. Thrall DE, Dewhirst MD. Use of radiation and/or hyperthermia for treatment of mast cell tumors and lymphosarcoma in dogs. Vet Clin North Am (Small Anim Pract) 1985;15:835–843.

13. Evans SM, Hendrick M. Radiotherapy of feline nasal tumors. Vet Radiol 1988;30(3):128–132.

14. Turrel JM. Radiation and hyperthermia. In: Holzworth J, ed. Diseases of Cats, 1st ed. Philadelphia, PA: WB Saunders; 1987:606–618.

15. Gabor LJ, Canfield PJ, Malik R. Immunophenotypic and histologic characterization of 109 cases of feline lymphosarcoma. Aust Vet J 1999;7:436–441.

16. North S, Meleo M, Mooney S, Mauldin GN. Radiation therapy in the treatment of nasal lymphoma in cats. Proceedings of the 14th Annual Conference of the Veterinary Cancer Society, 1994.

17. Sfiligoi G, Theon AP, Kent MS. Response of nineteen cats with nasal lymphoma to radiation therapy and chemotherapy. Vet Radiol Ultrasound 2007;48(4):388–393.

18. Henderson SM, Bradley K, Day MJ, et al. Investigation of nasal disease in the cat— A retrospective study of 77 cases. J Feline Med Surg 2004;6:245–257.

19. Owen LN. World Health Organization TNM Classification of Tumors in Domestic Animals, 1st ed. Geneva: WHO 1980.

20. Klein MK, Powers BE, Calley C, et al Veterinary Cooperative Oncology Group. Feline Nasal Lymphoma: A Retrospective Analysis. Proceedings of the 15th Annual Conference of the Veterinary Cancer Society, 1995.

21. Non-Hodgkin's Lymphoma Pathologic Classification Project. National Cancer Institute sponsored study of classifications of non-Hodgkin's lymphomas. Cancer 1982;49:2112–2135.

22. Valli VE, Jacobs RM, Norris A, et al. The histologic classification of 602 cases of feline lymphoproliferative disease using the National Cancer Institute working formulation. J Vet Diagn Invest 12:295–306.

23. Allison PD. Survival Analysis Using SAS: A Practical Guide. Cary, NC: SAS Institute Inc; 1995.

24. Essex M. Feline leukemia and sarcoma viruses. In: Klein G., ed. Viral Oncology. New York: Raven Press; 1980; 224–243.

25. Day MJ, Henderson SM, Belshaw Z, Bacon N. Immunhistochemical investigation of 18 cases of feline nasal lymphoma. J Comp Pathol 2004;130:152–161.

26. Vail D, Moore A, Ogilvie G, Volk L. Feline lymphoma (145 cases): Proliferation indices, cluster of differentiation 3 imunoreactivity, and their association with prognosis in 90 cats. J Vet Intern Med 1998;12:349–354.

27. Mukartirwa S, van der Linde-Sipman JS, Gruys E. Feline nasal and paranasal sinus tumours: Clinicopathological study, histomorphological description and diagnostic immunohistochemistry of 123 cases. J Feline Med Surg 2001;3:235–245.

28. Milner R, Peyton J, Cooke K, et al. Response rates and survival times for cats with lymphoma treated with the University of Wisconsin-Madison chemotherapy protocol: 38 cases (1996–2003). J Am Vet Med Assoc 2005;227(7):1118–1122.

29. Hall EJ, Giaccia AJ. Radiobiology for the Radiologist, 6th ed. Philadelphia, PA: WB Saunders; 2006:337.

30. Cotter SM. Feline viral neoplasia. In: Greene CE, ed. Infectious Diseases of the Dog and Cat, 2nd ed. Philadelphia, PA: WB Saunders; 1998; 83–71.

31. Hardy Jr WD, Hess PW, MacEwan EG, et al. Biology of feline leukemia virus in the natural environment. Cancer Res 1976;36:582.