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Phase I dose escalating study of oral cyclophosphamide in tumour-bearing cats

C.M. Chan[a,*], K.M. Rassnick[b], A.E. Frimberger[c], S.M. Nguyen[d], A.S. Moore[c]

[a] The Pet Oncologist, 1A Norman Street, Fig Tree Pocket, QLD 4069, Australia
[b] Veterinary Medical Center of Central New York, 5841 Bridge Street, East Syracuse, NY 13057, USA
[c] Veterinary Oncology Consultants, 379 Lake Innes Drive, Wauchope, NSW 2446, Australia
[d] Small Animal Specialist Hospital, 1 Richardson Pl, North Ryde, NSW 2113, Australia

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ABSTRACT

Cyclophosphamide is an alkylating agent used to treat cats with lymphoma, carcinomas and sarcomas. However, no clear consensus exists regarding the maximum tolerated dose (MTD) of oral cyclophosphamide in cats. Toxicities are rarely reported at published oral dosages of cyclophosphamide (200–300 mg/m^2). The primary aim of this prospective study was to determine the MTD of oral cyclophosphamide in tumour-bearing cats via a modified phase I trial. A secondary aim was to define any toxicity. Forty-six client-owned tumour-bearing cats were enrolled. The cyclophosphamide dosage was escalated by approximately 10% (300, 330, 360, 400, 440, 460 and 480 mg/m^2) in cohorts of at least six cats.

The MTD of oral cyclophosphamide in this study was 460 mg/m^2 with an inter-treatment interval of two to three weeks. Haematology is recommended 7 and 14 days after first cyclophosphamide treatment, and immediately before each subsequent dosage of cyclophosphamide or any potentially myelosuppressive chemotherapy agent. The dose-limiting toxicity was neutropenia with nadir at 7–21 days. This higher dosage was considered safe in combination with prednisolone and L-asparaginase. However, the higher dose of oral cyclophosphamide has not been evaluated in combination with other chemotherapy agents and thus should not be administered with these agents.

Introduction

Cyclophosphamide is an alkylating agent used to treat cats with lymphoma, carcinoma and sarcoma (Mauldin et al., 1988; Barber et al., 2000). However, no clear consensus exists regarding the maximum tolerated dose (MTD) of oral cyclophosphamide in cats. Toxicities are rarely reported at the currently recommended dosages of between 200 and 300 mg/m^2 (Cotter, 1983; Teske et al., 2002; Collette et al., 2016; Stroda et al., 2017). The principal toxicity seen in cats is myelosuppression, affecting neutrophils more than platelets (Moore et al., 2018). Gastrointestinal toxicity appears rare except at very high dosages (Fetting et al., 1982; Moore et al., 2018). Sterile haemorrhagic cystitis is a common adverse effect in humans and dogs, but it has not been convincingly reported in cats (Crow et al., 1977; Henness, 1985). In a recent study of 99 tumour-bearing cats treated with intravenous cyclophosphamide, the recommended dosage was 460 mg/m^2 every three weeks with dose limiting toxicity (DLT) of neutropenia (Moore et al., 2018). There is equal bioavailability of the active metabolite 4-hydroxycyclophosphamide seen in healthy cats administered 200 mg/m^2 via the oral, intravenous and intraperitoneal routes (Stroda et al., 2017). Therefore, the primary aim of this prospective study was to determine the MTD of oral cyclophosphamide in tumour-bearing cats via a modified phase I trial. A secondary aim was to define any toxicity.

Materials and methods

Animals

Prospective clinical trial was conducted for client-owned cats with cytologic or histologic confirmation of cancer that received oral cyclophosphamide between 2013 and 2019. Cases were recruited from four specialty veterinary practices (Queensland Veterinary Specialists, Veterinary Medical Center of Central New York, Animal Referral Hospital, and Small Animal Specialist Hospital) and via case consultations (Veterinary Oncology Consultants). Pre-treatment evaluation consisted of a physical examination, haemogram, serum biochemistry, and urinalysis. Follow-up haematology and serum biochemistry were performed in cats that presented systemically unwell including with febrile neutropenia. Cats that presented with comorbidities and / or in clinical substage b were permitted to enter this study. Staging with thoracic radiography, abdominal ultrasonogram and

* Corresponding author.
E-mail address: info@thepetoncologist.com (C.M. Chan).

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retroviral status testing were recommended in all cats and performed at the clinician's discretion; however, complete staging was not a requirement to be enrolled in this study. For all cats, written or verbal client consent were obtained.

Toxicity

Haematologic and gastrointestinal toxicities were graded according to VCOG-CTCAE (Veterinary Cooperative Oncology Group-Common Terminology Criteria for Adverse Events) v1.1 criteria (Veterinary Cooperative Oncology Group, 2016). Neutropenia was defined as an absolute neutrophil count <3.0 $\times$ $10^3/\mu L$ (DeClue and Spann, 2017). Haemograms were performed before and 7 days after cyclophosphamide. Because prolonged grade 1 neutropenia was observed in one cat treated at 360 mg/m^2, the monitoring protocol was subsequently modified to allow for additional haemograms two weeks after cyclophosphamide and immediately before each subsequent dose of cyclophosphamide.

Dosage escalation

Cyclophosphamide dosage was escalated by approximately 10% (300, 330, 360, 400, 440, 460, and 480 mg/m^2) in cohorts of at least six cats. Dose limiting toxicity was defined as $\geq$ grade 3 toxicity, except for haematologic toxicity, for which DLT was defined as $\geq$ grade 4 toxicity. If <33% cats experienced a DLT, dosage escalation continued. If >33% cats experienced a DLT, dosage escalation would stop. However, if 33% cats experienced a DLT, another three cats were entered in the same cohort. Of these cats, if $\geq$ two of three cats experienced a DLT, dosage escalation would stop. If one of three cats experienced a DLT, another three cats were entered in the same cohort. If none of three cats experienced a DLT, dosage escalation continued. The MTD was defined as the highest dosage level at which no more than one of six cats developed a DLT. Intra-cat dosage escalation was not allowed. Cats were sometimes entered before full data were available for previously treated cats; therefore, some cohorts included more than the originally planned number of cats. A schematic of dosage escalation for cats in this study is provided in Fig. 1.

Cyclophosphamide oral preparation

Cyclophosphamide was available in five different strengths. Two were commercially available as 50 mg tablets (Endoxan, Baxter Healthcare) and 25 mg capsules (Cyclophosphamide Capsules, Amerigen Pharmaceuticals). For lower strength capsules, where required to ensure dosing as close as possible to the intended dosage, cyclophosphamide powder was compounded into capsules of 5, 10, and 20 mg, by a compounding pharmacy (BOVA Compounding) following the Australian Code of Good Manufacturing Practice. For each batch of raw cyclophosphamide powder, analyses for potency, sterility, and active ingredients were performed with high-performance liquid chromatography at an external laboratory. Additionally, each batch of powder came with a certificate of analysis to verify that the active ingredient was of appropriate identity and purity. Potency was considered acceptable if it ranged between 97.0% and 103%. At the compounding pharmacy, cyclophosphamide powder was digitally weighed and placed into capsules; the capsules were confirmed to contain the appropriate amount of powder by two pharmacists who used software on two separate occasions. The expiration date for each batch of capsules was set 180 days after the date of compounding. The stability of cyclophosphamide capsules was not routinely tested.

Other medications administered concurrently

Concurrent medications allowed included prednisolone or prednisone, antiemetics (maropitant, ondansetron) and appetite stimulants (mirtazapine, cyproheptadine). Prophylactic antibiotics (pradofloxacin, marbofloxacin, enrofloxacin) were administered in cats at one higher dosage cohort (460 mg/m^2). Cats were also allowed 10,000 IU/m^2 SQ L-asparaginase (Leunase, Sanofi-Aventis) concurrently with the first, but not subsequent, cyclophosphamide treatment. These medications were administered at the clinician's discretion.

Results

Animals

Forty-six cats were included in this study, and all cats had measurable disease when treatment was started. The diagnosis was confirmed by histopathology in 18 cats (39.1%) and by cytology in 27 cats (58.7%). One cat had both cytologic and histologic confirmation of cancer. Thirty-eight cats (82.6%) had lymphoma, four had sarcoma, two had myeloma-related disorder, one had squamous cell carcinoma, and one had mammary carcinoma. For cats diagnosed with lymphoma, the gastrointestinal tract was the most common location (n = 18), and 16 of these 18 cats (88.9%) presented with gastrointestinal signs.

The median age was 11 years (range, 1.5–14 years of age). Thirty-three were castrated males, and 13 were spayed females. There were nine breeds included, with Domestic Shorthair cats being the most common (n = 32). Other breeds represented were Domestic medium hair (n = 3), Domestic longhair (n = 3), Siamese (n = 2), Devon Rex (n = 2), Maine Coon (n = 1), Persian (n = 1), Russian blue (n = 1) and Birman (n = 1). Median bodyweight was 4.67 kg (range, 2.95–7.26 kg).

Haematologic data were available in all cats before and 7 days after cyclophosphamide except for one cat that was found dead 5 days after receiving cyclophosphamide. One cat with lymphoma treated at 360 mg/m^2 experienced grade 1 neutropenia 14 days after cyclophosphamide; therefore, the remaining cats had haematologic data collected 7 days and 14 days after cyclophosphamide. On presentation (prior to receiving cyclophosphamide), one cat had marked neutropenia (0.71 $\times$ $10^3/\mu L$) and moderate thrombocytopaenia (41 $\times$ $10^3/\mu L$), one cat had mild thrombocytopaenia (129 $\times$ $10^3/\mu L$), and one cat had mild neutropenia (2.64 $\times$ $10^3/\mu L$). The remaining cats (n = 43) had normal neutrophil (>3.0 $\times$ $10^3/\mu L$) and adequate platelet counts (150 $\times$ $10^3/\mu L$ or platelet clumps on blood smear evaluation). Median neutrophil count on presentation was 7.56 $\times$ $10^3/\mu L$ (range, 0.71–22.07 $\times$ $10^3/\mu L$). Median platelet count on presentation was 326 $\times$ $10^3/\mu L$ (range, 41–680 $\times$ $10^3/\mu L$). Eighteen cats (39.1%) had anaemia (packed red cell volume [PCV] <30) on presentation. Median PCV on presentation was 32 (range, 10–48).

Treatment

All cats with lymphoma were given prednisolone or prednisone. Median dosage of prednisolone or prednisone was 2 mg/kg PO q 24 h (range, 1–2.75 mg/kg). Twenty cats (43.5%) also received L-asparaginase concurrently with the first, but not subsequent, cyclophosphamide treatment. Due to the available tablet and / or capsule sizes, the actual dosage of cyclophosphamide administered varied between −4.0 and +6.0% (median, 0%) from the intended cyclophosphamide dosage. The total number of cyclophosphamide doses administered per cat was between 1 and 4 (median = 2). The dosing interval of cyclophosphamide was every two weeks and every three weeks in 42 and four cats, respectively. Twenty-four cats (52.2%) received commercial cyclophosphamide capsules or tablets, and 22 cats (47.8%) received a combination of commercial and compounded cyclophosphamide.

Dosage escalation and toxicity

The dosage escalation and toxicity data for the cats in this study are summarised in Table 1. Seven cats received oral cyclophosphamide at 300 mg/m^2. Three cats experienced no toxicity. One cat experienced grade 1 neutropenia 7 days after cyclophosphamide, one cat experienced grade 1 thrombocytopaenia 7 days after cyclophosphamide, and two cats experienced grade 2 inappetence within the 7 days after cyclophosphamide. There were no DLT toxicity seen in this cohort; therefore, dosage escalation continued.

Six cats received oral cyclophosphamide at 330 mg/m^2. Three cats experienced no toxicity. Two cats experienced grade 1 inappetence within 7 days after cyclophosphamide, and one cat experienced grade 1 anaemia 7 days after cyclophosphamide. There were no DLT toxicity seen in this cohort; therefore, dosage escalation continued.

Nine cats received oral cyclophosphamide at 360 mg/m^2. In the first six cats receiving treatment at this dosage, two cats experienced no toxicity, one cat experienced grade 1 neutropenia 14 days after cyclophosphamide, one cat experienced grade 2 thrombocytopaenia 7 days after cyclophosphamide, one cat experienced grade 3 anorexia and grade 1 neutropenia 7 days

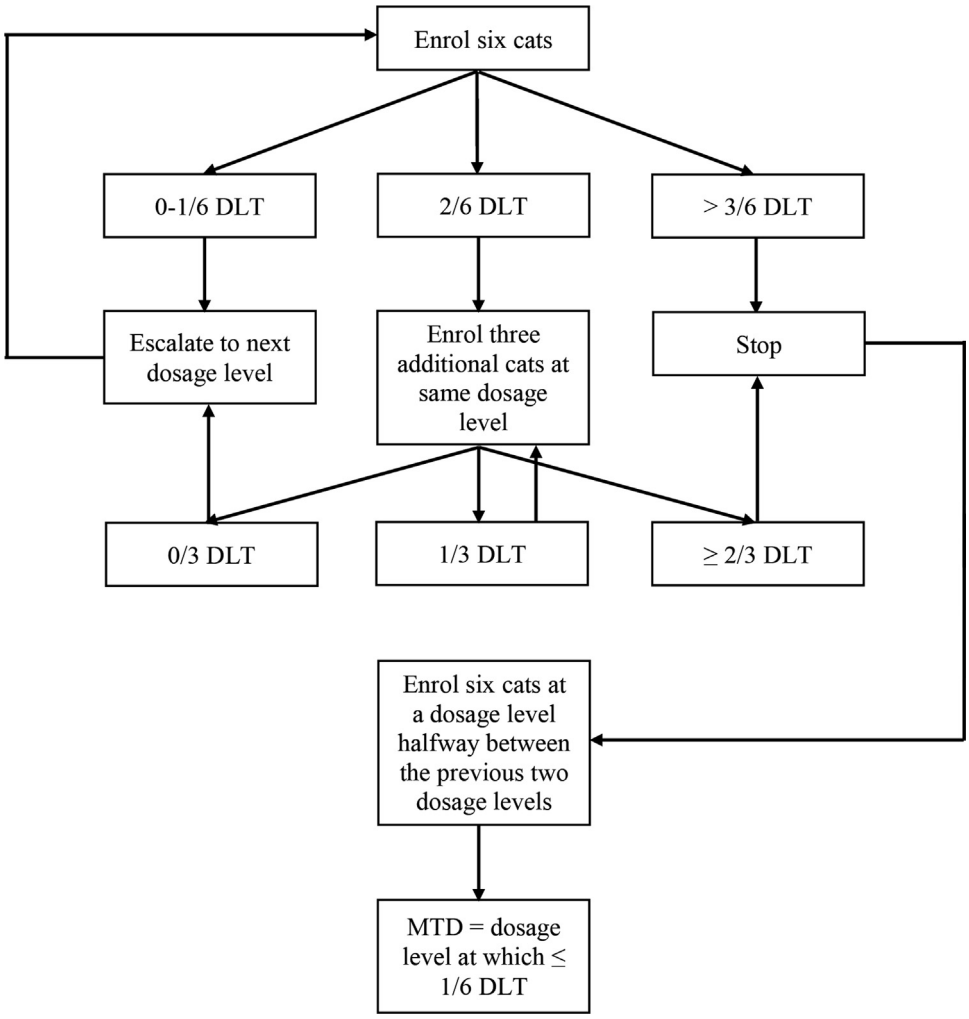

Fig. 1. Modified 6 + 3 phase I dosage escalation study design schematic. DLT, dose limiting toxicity.

Table 1
Highest toxicity grade and dose limiting toxicity (DLT) following different dosages of cyclophosphamide in tumour-bearing cats.

Dosage (mg/m^2)	Cats (*n*)	Grade (n)						DLT *n* (%)
		0	1	2	3	4	5	
300	7	3	2	2	0	0	0	0
330	6	3	3	0	0	0	0	0
360	9	4	1	2	1	0	1	2/9 (22)
400	9	2	4	1	2	0	0	2/9 (22)
440	6	2	2	1	1	0	0	1/6 (17)
460	6	2	3	1	0	0	0	0
480	3	0	1	0	0	2	0	2/3 (67)

after cyclophosphamide, and one cat was found dead 5 days after receiving oral cyclophosphamide. This cat had localised small intestinal and cranial mediastinal lymphoma. A post-mortem was not permitted; therefore, the cause of death for the latter cat was unknown. Two of six cats (33%) experienced a DLT at this cohort; therefore, a further three cats were entered in the same cohort. Two cats experienced no toxicity and one cat experienced grade 2 diarrhea within 7 days after cyclophosphamide. Overall, two of nine cats (22%) experienced a DLT in this cohort; therefore, dosage escalation continued.

Nine cats received oral cyclophosphamide at 400 mg/m^2. In the first six cats receiving treatment at this dosage, two cats experienced grade 1 neutropenia 7 days after cyclophosphamide; one cat experienced grade 1 vomiting and thrombocytopaenia 7 days after cyclophosphamide; one cat experienced grade 2 neutropenia and grade 1 thrombocytopaenia 7 days after cyclophosphamide; one cat experienced grade 3 febrile neutropenia, thrombocytopaenia and anorexia 7 days after cyclophosphamide that required prompt management in hospital with intravenous fluid therapy, broad-spectrum antibiotics and supportive care; and one cat experienced grade 3 inappetence and grade 2 thrombocytopaenia 7 days after cyclophosphamide. Thirty-three percent of cats experienced a DLT in this cohort; therefore, another three cats were entered in the same cohort. Two cats experienced no toxicity, and one cat experienced grade 1 inappetence within 7 days after cyclophosphamide. Overall, two of nine cats (22%) experienced a DLT in this cohort; therefore, dosage escalation continued.

Six cats received oral cyclophosphamide at 440 mg/m^2. Two cats experienced no toxicity. One cat experienced grade 1 weight loss, lethargy and inappetence within 7 days after cyclophosphamide; one cat experienced grade 1 neutropenia and thrombocytopaenia 7 days after cyclophosphamide; one cat experienced grade 2 gastrointestinal toxicity (inappetence and dehydration) and neutropenia, and grade 1 thrombocytopaenia 7 days after cyclophosphamide; and one cat experienced grade 3 lethargy and anorexia within 7 days of cyclophosphamide. Overall one of six cats (16.7%) experienced a DLT in this cohort; therefore, dosage escalation continued.

Three cats received oral cyclophosphamide at 480 mg/m^2. One cat experienced grade 1 inappetence and weight loss within 7 days of cyclophosphamide. Two cats experienced grade 4 febrile neutropenia and sepsis at 5 days and 14 days, respectively, after receiving cyclophosphamide. Both cats recovered with prompt management in hospital with IV fluid therapy, broad-spectrum antibiotics and supportive care. Overall, two of three cats (66.7%) experienced a DLT in this cohort; therefore, dosage escalation was ceased.

An additional cohort was entered at a dosage of 460 mg/m^2 as halfway between the dosage levels of 440 mg/m^2 and 480 mg/m^2. All cats in this cohort received prophylactic pradofloxacin, marbofloxacin or enrofloxacin at 5 mg/kg orally once daily for 14 days after cyclophosphamide. Two cats experienced no toxicity. One cat experienced grade 1 neutropenia and inappetence 7 days after cyclophosphamide, one cat experienced grade 1 neutropenia 21 days after cyclophosphamide, one cat experienced grade 1 weight loss, diarrhea and vomiting 7 days after cyclophosphamide and one cat experienced grade 2 neutropenia 14 days after cyclophosphamide. There were no DLT seen in the six cats in this cohort; therefore, this dosage was determined to be the MTD.

A summary of the neutrophil data for cats in this study is found in Table 2 and Fig. 2. There was a trend to increased frequency and grade of neutropenia with increased dosage of cyclophosphamide; however, statistical evaluation was not performed. Neutropenia was recorded in 14 cats (30.4%). Four cats (8.7%) experienced delayed neutropenia. Three cats experienced delayed neutropenia 14 days after cyclophosphamide, and one cat experienced delayed neutropenia 21 days after cyclophosphamide. All cases of neutropenia normalised by day 21, except for the one cat that experienced delayed neutropenia 21 days after cyclophosphamide, which normalised by day 28. At the MTD of 460 mg/m^2, 50% of cats experienced neutropenia which were mild (grade 1 or 2). No cats were humanely euthanased or died due to neutropenia in this study.

Table 2
Grades of neutropenia events for cats treated at different cyclophosphamide dosages.

Dosage (mg/m^2)	Cats (*n*)	Grade (*n*)					
		0	1	2	3	4	5
300	7	6	1	0	0	0	0
330	6	6	0	0	0	0	0
360	9	7	2	0	0	0	0
400	9	5	2	1	1	0	0
440	6	4	1	1	0	0	0
460	6	3	2	1	0	0	0
480	3	1	0	0	0	2	0

Table 3
Grades of gastrointestinal events for cats treated at different cyclophosphamide dosages.

Dosage (mg/m^2)	Cats (*n*)	Grade (*n*)					
		0	1	2	3	4	5
300	7	5	0	2	0	0	0
330	6	4	2	0	0	0	0
360	9	6	0	1	1	0	0
400	9	5	2	0	2	0	0
440	6	3	1	1	1	0	0
460	6	4	2	0	0	0	0
480	3	2	1	0	0	0	0

Gastrointestinal signs were reported in 17 cats (37.0%) with anorexia and inappetence reported as the most common gastrointestinal sign, occurring in 11 of these 17 cats. At the MTD of 460 mg/m^2, 33% of cats experienced mild gastrointestinal events (grade 1). A summary of the gastrointestinal events data for cats in this study is found in Table 3.

Platelet data were available for all cats in this study. Thrombocytopaenia was recorded in eight cats (17.4%), and all occurrences of thrombocytopaenia occurred 7 days after cyclophosphamide and had resolved 14 days after cyclophosphamide. Most events (87.5%) of thrombocytopaenia were mild (grade 1 or 2). There was no clinical evidence of bleeding observed in this study.

There was no clinical evidence of sterile haemorrhagic cystitis, hepatopathy, or development or worsening of renal azotaemia in this study. However, one of the five cats with renal insufficiency, IRIS (International Renal Interest Society) stage 4 CKD treated at 400 mg/m^2 experienced grade 3 febrile neutropenia, thrombocytopaenia and gastrointestinal toxicity (anorexia), requiring prompt management in hospital with intravenous fluid therapy, broad-spectrum antibiotics and supportive care.

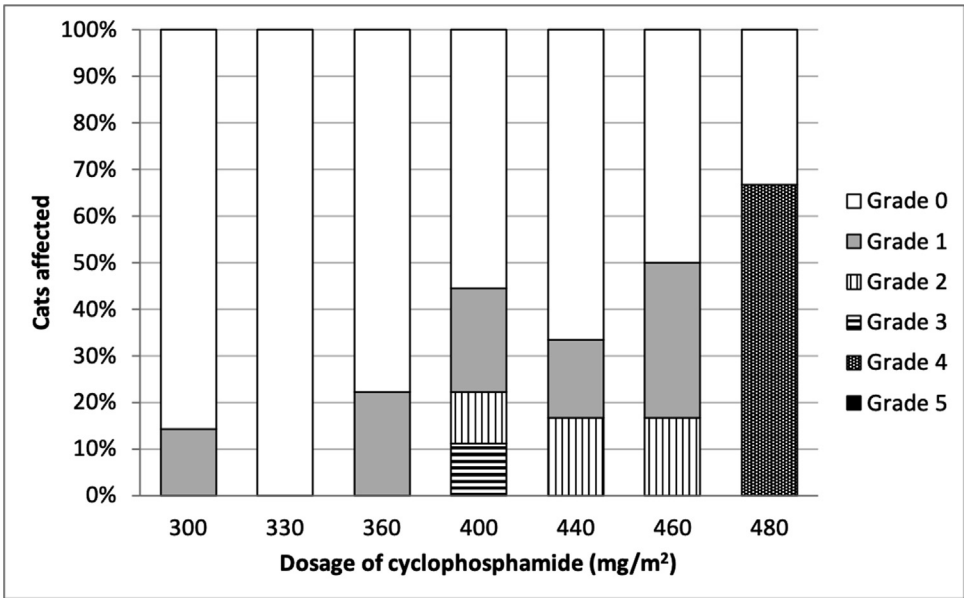

Fig. 2. Grades of neutropenia recorded for cats treated at different cyclophosphamide dosages.

Discussion

The purpose of this study was to determine the MTD of oral cyclophosphamide in tumour-bearing cats and to define any toxicity. Based on the findings reported here, the MTD and recommended oral cyclophosphamide dosage as a single agent in tumour-bearing cats is 460 mg/m^2 with an inter-treatment interval of two to three weeks. The cyclophosphamide dosage recommended in this study is approximately twice the recommended canine dosage of 200–250 mg/m^2 (Warry et al., 2011; Goodman et al., 2016). However, it is the same as the recommended intravenous cyclophosphamide dosage in tumour-bearing cats (Moore et al., 2018), supporting previous studies that showed no pharmacokinetic difference between these two routes of administration (Warry et al., 2011; Stroda et al., 2017).

Myelosuppression, specifically neutropenia, was the acute DLT; it occurred later than 7 days in 8.7% cats in this study and in one cat was prolonged to 21 days; therefore, a dosage interval of every two to three weeks is suggested, with re-evaluation of haematology before administering the next myelosuppressive agent. These findings are similar to a recent study of 99 tumour-bearing cats treated with intravenous cyclophosphamide, where the recommended inter-treatment interval was three weeks (Moore et al., 2018). The acute DLT reported in that study was neutropenia, which was also delayed and / or prolonged in some cats. Evaluation of haematology is recommended before each dosage of cyclophosphamide, 7 and 14 days after cyclophosphamide treatment, and immediately before each subsequent dosage of any potentially myelosuppressive chemotherapy agent to confirm complete haematologic recovery.

Gastrointestinal toxicity is not a common adverse effect of cyclophosphamide administration in people or dogs. In our study, gastrointestinal toxicity, which was predominately anorexia or inappetence, occurred in 37.0% of cats; however, it was generally mild, responded to anti-nausea medications and did not require hospitalisation. This was higher than reported in tumour-bearing cats treated with intravenous cyclophosphamide, where gastrointestinal toxicity occurred in approximately 20% of cats (Moore et al., 2018). However, the incidence of grade 3 gastrointestinal toxicity was low and similar in cats treated with both intravenous and oral cyclophosphamide at 6% (Moore et al., 2018) and 8.7% of cats, respectively.

It was challenging to separate gastrointestinal signs arising from chemotherapy from those caused by gastrointestinal lymphoma. Eighteen of 46 cats (39.1%) treated in this study had macroscopic gastrointestinal lymphoma, and 16 of these 18 cats (88.9%) presented with gastrointestinal signs. Due to the large percentage of cats that presented with gastrointestinal lymphoma, the prevalence of gastrointestinal toxicities may have been higher than would be observed in cats without gastrointestinal cancer, although only one of the four cats that experienced grade 3 gastrointestinal toxicity had gastrointestinal lymphoma.

No clinical evidence for sterile haemorrhagic cystitis was observed in this study, which is consistent with the literature where sterile haemorrhagic cystitis has not been convincingly reported in cats treated with cyclophosphamide (Crow et al., 1977; Henness, 1985; Moore et al., 2018) or ifosfamide (Rassnick et al., 2006a, b).

In humans, the parent drug of cyclophosphamide is renally cleared, and clearance of cyclophosphamide is decreased in patients with reduced renal function, thereby resulting in increased systemic drug exposure. Therefore, renal insufficiency is sometimes considered a criterion for cyclophosphamide dosage reduction (Haubitz et al., 2002). In our study, renal insufficiency was identified in five cats, although there was no clinical evidence for development or worsening of renal azotaemia in this study. One cat with IRIS stage 4 CKD experienced grade 3 bone marrow and gastrointestinal toxicity. To the best of our knowledge, there have been no pharmacokinetic or pharmacodynamic studies of cyclophosphamide performed in cats with renal insufficiency. Inadequate renal function may exacerbate cyclophosphamide myelosuppression; therefore, careful monitoring of cats with renal insufficiency is recommended.

At 480 mg/m^2, two of three cats (66.7%) experienced a DLT (specifically grade 4 febrile neutropenia and sepsis requiring hospitalisation); therefore, dosage escalation was ceased, and no further cats were recruited at this cohort. Due to concerns for potential increased risk of febrile neutropenia and sepsis, all six cats treated at 460 mg/m^2 received prophylactic antibiotics. At this dosage, there was no DLT; therefore, the use of prophylactic antibiotics may have reduced the risk of sepsis in this cohort. However, to the best of our knowledge, there have been no studies in cats to assess whether the use of prophylactic antibiotics is beneficial. Further studies treating cats at this recommended higher dosage without prophylactic antibiotics would be of interest.

A limitation of the present study was the administration of compounded cyclophosphamide capsules in approximately half cats, which all received a combination of commercial and compounded cyclophosphamide. Compounding was required to ensure dosing as close as possible to the intended dosage (range, −4.0 to +6.0%; median, 0%). All capsules were manufactured in accordance with the Australian Code of Good Manufacturing Practice by a nationally registered compounding pharmacy, which used internal quality controls for the cyclophosphamide potency of the capsules and all capsules were used within the 6-month expiration period. However, no stability validation studies were performed, and no independent quality controls were used to assure the cyclophosphamide potency of the capsules. There can be variability in potency for reformulated cyclophosphamide capsules (Burton et al., 2017; Robat and Budde, 2017), and it might have increased or decreased the toxicity in this study, although compounded capsules are commonly used in clinical practice.

L-asparaginase was administered concurrently with the first dosage of cyclophosphamide in 43.5% of cats. No interactions between cyclophosphamide and L-asparaginase have been reported in any species. Due to the small sample size in each group in our study, statistical evaluation was not performed. However, based on the recent intravenous cyclophosphamide study which revealed no impact of concurrent L-asparaginase administration on the neutrophil count, (Moore et al., 2018) we surmise that concurrent L-asparaginase administration would not impact the findings in this study.

Another limitation of our study was the inclusion of cats with pre-existing neutropenia and / or thrombocytopaenia (n = 3) or other comorbidities, which may have overestimated evaluation of DLT. In clinical practice, some cats would present with comorbidities; therefore, this was not an exclusion factor in our study.

An additional limitation of our study was the inclusion of prophylactic anti-emetics and appetite stimulants, which were permitted at the clinician's discretion. This would likely reduce the occurrences and grade of gastrointestinal toxicity seen in our study, which may have underestimated evaluation of DLT. The dosages and number of cats that received prophylactic anti-emetics and appetite stimulants were not recorded in this study.

There are published COP [cyclophosphamide, vincristine and prednisolone] based protocols that administer cyclophosphamide concurrently with other chemotherapy agents such as vincristine on the same day (Cotter, 1983; Hadden et al., 2008). This was not evaluated in this study; therefore, we do not recommend the use of this higher dosage of oral cyclophosphamide concurrently with other chemotherapy agents. Cyclophosphamide at a dosage of 460

mg/m^2 should only be administered as a single agent, or with L-asparaginase and / or prednisolone.

This study was not designed to assess the clinical efficacy (i.e. tumour response rate) for cats receiving different dosages of cyclophosphamide, nor was it designed to assess cumulative toxicity. However, further evaluation of tumour response and cumulative toxicity data in a larger population of cats to this higher dosage of cyclophosphamide is warranted.

Conclusions

The MTD of oral cyclophosphamide in tumour-bearing cats in our study was 460 mg/m^2 with an inter-treatment interval of two to three weeks. The DLT was neutropenia, which can occur 7–21 days after treatment. Evaluation of haematology is recommended before each dosage of cyclophosphamide, 7 and 14 days after cyclophosphamide treatment, and immediately before the next administration of any potentially myelosuppressive agent. This higher dosage was considered safe in combination with prednisolone and L-asparaginase, but it has not been evaluated and thus should not be used in combination with other chemotherapy agents. Further evaluation of tumour response and cumulative toxicity data in a larger population of cats treated with this higher dosage of cyclophosphamide will be of interest.

Conflict of interest

None.

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