Objectives: To evaluate the response of measurable canine mast cell tumours unsuitable for other treatment modalities to a chemotherapy protocol comprising chlorambucil and prednisolone.

Methods: Dogs bearing measurable mast cell tumours, unsuitable for treatment by surgery or radiotherapy, were treated with orally administered prednisolone and chlorambucil, and their responses assessed.

Results: Twenty-one dogs were enrolled in the study; 13 had intermediate-grade mast cell tumour, six were high grade and two were diagnosed by cytology alone. Eight dogs had multiple tumours and 13 dogs had single tumours, and six dogs had lymph node metastases and no dogs had visceral metastases detected. Three dogs achieved complete remission, five achieved partial remission (overall response rate 38 per cent), nine had static disease and four dogs had progressive disease. Median progression-free interval for the eight responders was 533 days, and median survival time for all dogs in the study was 140 days. Progression-free interval and median survival time were not influenced by the age, sex, weight or neutering status of the patient, by the grade or stage of the tumour or whether the patient had single or multiple tumours. No toxicity was detected.

Clinical significance: Response and survival rates of inoperable canine MCT to chlorambucil and prednisolone are comparable to previously described protocols, with no apparent toxicity.

Introduction

Canine mast cell tumours (MCTs) are common, comprising seven to 21 per cent of all canine skin tumours (Thamm and Vail 2007). Tumours are graded on their histological characteristics, whereby tumours are assigned grades I (low) to III (high), depending on their morphology, degree of differentiation and invasiveness (Patnaik and others 1984). It is well documented that the majority of dogs affected by low-grade and mid-grade tumours can be treated effectively with surgery, and adjuvant radiotherapy can be used where complete excision has not been possible (Turrel and others 1988, Al-Sarraf and others 1996, Frimberger and others 1997, LaDue and others 1998, Dobson and others 2004, Hahn and others 2004, Poirier and others 2006). Unfortunately, a small proportion of dogs with intermediate-grade tumours and the majority of dogs with high-grade tumours can experience aggressive local tumour recurrence and/or metastatic disease and thus carry a poor prognosis (Bostock 1973, Patnaik and others 1984, Seguin and others 2001, 2006, Michels and others 2002, Weisse and others 2002, Murphy and others 2004). In these situations, chemotherapy could be indicated, and a variety of drugs and protocols have been used in an attempt to extend survival (McCaw and others 1994, 1997, Gerritsen and others 1998, Rassnick and others 1999, Thamm and others 1999, 2006, Davies and others 2004). All these studies have included dogs treated postoperatively for microscopic residual disease as well as those with gross disease, despite published evidence that suggests that two thirds to three quarters of grade II MCTs with microscopic residual disease following surgery alone do not recur (Michels and others 2002, Seguin and others 2006).

The aim of this prospective study was to evaluate a protocol combining chlorambucil and prednisolone in dogs with measurable MCTs that were unsuitable for local therapies such as surgery and radiotherapy.
Chlorambucil and prednisolone chemotherapy for dogs

measurable MCT deemed unsuitable for locoregional therapy, such as surgery and/or radiotherapy. This included dogs with extensive local disease, multiple tumours and/or metastatic disease. Dogs with local lymph node (LN) metastases suitable for locoregional therapy (surgery and/or radiotherapy) were not included in this study. Dogs with suspected residual disease following surgery were not included.

Dogs were not included if they had had any prior therapy for their MCT, except for cases with recurrent MCT that had had prior surgical treatment only.

All tumours were histologically or cytologically confirmed as MCTs by a commercial laboratory. All the histologically diagnosed tumours were graded according to the Patnaik’s grading system (Patnaik and others 1984) by a veterinary histopathologist.

All dogs included in the study underwent clinical assessment, including measurement of gross tumour mass (based on measuring tumour length and width), palpation and fine needle aspiration (FNA) of local draining LNs, thoracic radiography (right and left lateral views) and ultrasound examination of abdominal organs and regional LNs in order to stage the tumour (Owen 1980). Any lesions deemed to be suspicious for MCT recurrence or metastasis were investigated using FNA and cytological examination or tissue biopsy with histopathological assessment. Multiple cutaneous tumours were not considered to be distant metastatic disease for the purposes of staging as it is recognised that some dogs are prone to development of more than one MCT, and the presence of multiple tumours has been shown not to influence prognosis (Kiupel and others 2005, Mullins and others 2006, Murphy and others 2006). Where multiple tumours occurred, all those that were excised or biopsied were histologically graded, and for statistical analyses, patients were categorised as for the highest grade tumour. Location of multiple tumours was categorised for where the majority of tumours occurred on that patient.

Chemotherapy protocol
The chemotherapy protocol comprised prednisolone (Prednicare; Animalcare) at a starting dose of 40 mg/m² orally once daily for 14 days, reducing to 20 mg/m² daily every other day, combined with 5 mg/m² chloram-

bacil (Leukeran; GlaxoSmithKline) orally every other day. In cases where a sustained complete remission (CR) occurred, the protocol was discontinued after six months, and in all other cases, the protocol was administered continuously.

All patients were assessed at the QVSH after the first two weeks of treatment. Revisits were then scheduled on a monthly basis for as long as the patient survived, with additional appointments if patients deteriorated. Where it was not possible to re-evaluate patients at the QVSH, follow-up information after the first two weeks was obtained from the referring veterinary surgeon. At each visit, tumour response was assessed by gross inspection and physical measurement with radiological, ultrasound and cytological examinations at the discretion of oncologists at the QVSH.

Evaluation of response and toxicity
Response to treatment was defined as CR: no clinical evidence of local or distant MCT, partial remission (PR): greater than 50 per cent reduction in size of tumour (by length or width measurement), static disease (SD): no change in MCT size or progression of metastatic disease or progressive disease (PD): increase in size of MCT either locally, progression of metastatic disease or deterioration in associated clinical signs [World Health Organization (WHO) 1979]. Assessment of response in patients with multiple tumours was made by summation of the longest diameter measurement for each tumour. PR in these patients was defined in a reduction in greater than 50 per cent of the sum of the diameters.

A haematology profile was performed for each patient every four weeks to assess for myelosuppression; this and any other toxicities encountered were graded according to the Veterinary Comparative Oncology Group common terminology criteria for adverse events (VCOG-CTCAE 2004).

Survival and progression-free intervals
For each patient, it was recorded if the dog died of MCT or another cause. The survival time was calculated from the start of treatment to the date of death of the patient because of MCT or other causes. Patients that were still alive at the end of the study, died because of other causes or any that were lost to follow-up were censored at that point.

The progression-free interval (PFI) was calculated from the date of start of treatment to the date where the disease progressed, either as increase in tumour size or when metastatic spread was documented. All patients that had stable disease at the end of the study were censored at this point.

Where patients that had achieved a CR and subsequently discontinued the protocol at six months, but then restarted chemotherapy at a later date because of recurrence, the PFI was extended to include the entire treatment period as long as a response was achieved.

Statistical analysis
Computer software package SPSS 15 was used for statistical analysis. The Kaplan-Meier product limit method was used to calculate survival and PFI and to compare the effect of patient variables on these. Cox proportional hazards regression model was used to determine independent prognostic factors for remission and survival duration. Dogs were censored if they were lost to follow-up, alive at the time of data analysis or if they died of causes unrelated to their MCT. Where the cause of death was not clearly defined, it was assumed that the patient died of MCT-related causes.

RESULTS
Twenty-one dogs were entered into the study. Breeds represented included nine Labrador retrievers, two each of Staffordshire bull terrier and crossbred dogs and one each of greyhound, English springer spaniel, shar pei, West Highland white terrier, German shepherd dog, Jack Russell terrier, miniature daschund and boxer. The mean age was eight years (median 10 years, range four to 13-0 years), and mean bodyweight was 25-5 kg (median 32-0 kg, range 8-0 to 38-8 kg). There were 12 males, of which seven were neutered.