Normal and neoplastic canine lymphocytes express luteinizing hormone receptors
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Introduction
Lymphoma is a common malignant cancer of dogs. Spayed and castrated dogs are 3-4 times more likely to develop lymphoma.\textsuperscript{1,2} Circulating luteinizing hormone (LH) concentrations are significantly and persistently elevated in spayed and castrated dogs compared to intact dogs. Luteinizing hormone receptors (LHR) are expressed in normal rat lymphatic tissue.\textsuperscript{3}

Hypothesis
That LHR were also present in normal and neoplastic canine lymph nodes. The study aim was to determine if LHR were expressed and to quantify the level of expression.

Methods
Normal (n=1) and neoplastic (n=4) lymph node tissue was removed postmortem, formalin-fixed, paraffin-embedded, and sectioned (6 μm) onto charged slides. Testicular tissue from a separate dog obtained following castration was treated in the same manner for a positive control. All slides were deparaffinized, rehydrated, subjected to heat-induced epitope retrieval (#S1700, Dako, Carpinteria, CA). Endogenous peroxidase activity was inactivated with 3% H\textsubscript{2}O\textsubscript{2} and nonspecific binding was blocked with 1% horse serum. Goat polyclonal anti-human LHR antibody (SC-26341, Santa Cruz Biotechnology, Dallas, TX,) was applied at a 1:50 dilution. Negative controls from each tissue were treated in the same way except in absence of primary antibody. Slides were reacted with biotinyalted horse anti-goat IgG (Vector Laboratories, Burlingame, CA) and incubated with preformed avidin-biotin-peroxidase complex (#PK6105, ABC kit, Vector Laboratories) followed by Nova Red Peroxidase substrate (#SK4800, Vector Laboratories). Slides were counter-stained with hematoxylin, dehydrated, and mounted. The percentage of cells positive for LHR was determined at 400X magnification by a single observer.

Results
Canine lymphocytes express LHR in 4% of cells in normal lymph nodes and 12.37% of the cells in those with lymphoma based on a single observer counting the cells (Figure). There was no positive staining evident in the negative control tissue sections.

Discussion
This is the first report of LHR expression in canine lymphatic tissue. Studies are underway to determine the immunophenotype (B- or T-) of the lymphocytes expressing the LHR. The long-range goal of this research is to provide evidence to support using a complementary treatment for canine lymphoma by down-regulating LH with a commercially-available canine gonadotropin-releasing hormone (GnRH) agonist.

Keywords Castrate, dog, lymphoma, spay

References