Evaluation of the interaction of gonadotrophin releasing hormone (GnRH) and kisspeptin (Kp) as peptides that modulate the higher order control mechanisms in reproductive physiology has been a recent development. A novel investigation into the anatomical location and possible interaction of GnRH and Kp neurons in the canine brain was undertaken with immunohistochemical (IHC) evaluation of domestic canids. The aim of this study was to characterize the location of GnRH and Kp neurons within the canine hypothalamus and to assess their interactions.

Four canines were humanely euthanized, with the pituitary and hypothalamus preserved for analysis. Antibody specificity was imperative for the IHC, in order to discriminate Kp from other RF-amide peptides. Therefore, the antibody selection was: 1°: rabbit anti-Kp and mouse anti-GnRH; 2°: Alexa fluor (594) goat anti-rabbit IgG and Alexa fluor (488) goat anti-mouse IgG. The mounted sections were evaluated with an epifluorescence microscope with wavelength filters for analysis of each fiber type (480nm - GnRH, green fluorescence; 560nm - Kp, red fluorescence).

Gonadotropin releasing hormone fibers were concentrated in the medial basal hypothalamus (MBH) with dense fibers and terminals throughout the median eminence (ME) extending into the arcuate nucleus (ARC). The Kp fibers had poorly defined boundaries and were scattered throughout the MBH and the lateral hypothalamic area, with clear demarcation within the lateral ARC. In contrast to the high frequency of GnRH fibers, Kp fibers were scarce within the ME. With regard to the morphological interactions between GnRH and Kp, the dual immunofluorescence illustrated very closely apposed Kp structures to GnRH neurons in the ARC of the caudal MBH. Characterization of the distribution of the GnRH and Kp fibers in the canine enables inferences regarding their neuro-endocrinological activity.

Keywords: Kisspeptin, gonadotrophin releasing hormone, immunohistochemistry, hypothalamus, canine

(Editors note: The photographs in this paper appear in color in the online edition of Clinical Theriogenology.)