Variation in Canine Neurohypophyseal Histology Associated with Sectioning Plane

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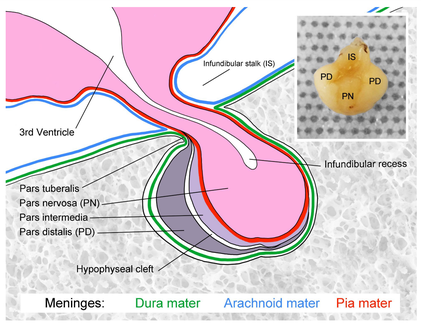


Pituitary gland diagram (lateral view) and dog specimen (inset, caudal view)

Background – Brief Review of Pituitary Gland Anatomy

- The pituitary gland is a complex organ, developing from the craniopharyngeal oral ectoderm (adenohypophysis, anterior pituitary) as well as from downgrowth of the neural ectoderm (neurohypophysis, posterior pituitary).
- Connected to the hypothalamus via the infundibular (pituitary) stalk, the pituitary gland lies ventral to the hypothalamus, on the floor of the cranial cavity in the hypophyseal fossa of the sella turcica of sphenoid bone, and is covered by dura mater (diaphragma sellae). The arachnoid membrane reflects at the level of the pituitary stalk (arachnoid sleeve of the pituitary stalk) and the pia mater extends along the hypothalamus and pituitary stalk to cover only the neurohypophysis.
- The adenohypophysis includes the pars distalis, pars tuberalis, and pars intermedia, which contain hormonally active cells sensitive to hypothalamic releasing hormones (corticotropin-, gonadotropin-, thyrotropin-, growth hormone-releasing hormones, dopamine, and somatostatin) delivered via the hypophyseal portal capillary system from the median eminence of the hypothalamus. Rathke's pouch forms the hypophyseal cleft between the pars intermedia and pars distalis.
- The neurohypophysis (an appendage of the brain) includes the pars nervosa and infundibular stalk through which axons run from hormone-producing neurons (oxytocin and vasopressin) in the hypothalamus. The neurohypophysis and median eminence form part of the floor of the third ventricle, and an extension of the third ventricle, the infundibular recess, extends into the infundibular stalk.

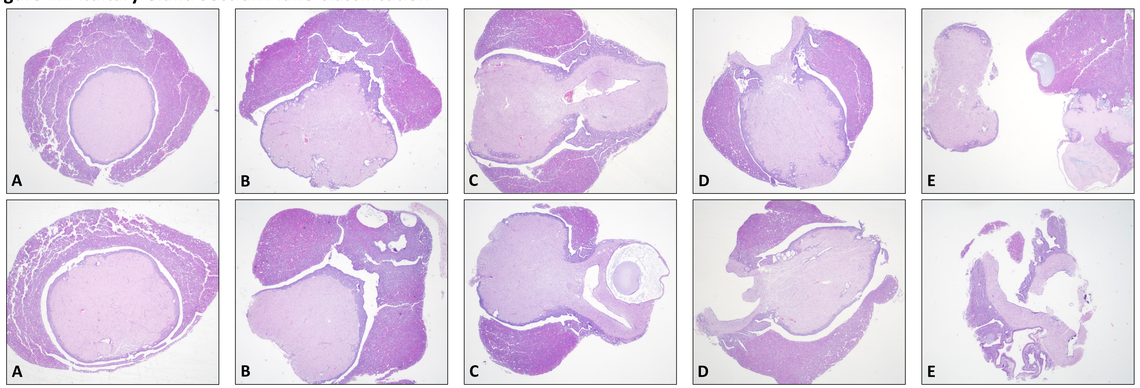
Introduction

As a master endocrine organ and relay between nervous and hormonal systems, the pituitary gland plays key roles in health and disease. Pituitary gland assessments by organ weights and microscopic examination are included in many nonclinical toxicology studies and in regulatory guidance, particularly for endocrine and carcinogenicity screenings. No defined sectioning algorithm exists for canine pituitary glands, but a recent diagnostic publication employed a mid-sagittal approach.[4] Given the organ's structural complexity, small size, and protected anatomic location, consistent retrieval and sectioning pose challenges. Histological capture of substructures and/or appearance of cell populations within substructures may also vary based on sectioning planes. Due to suspected section orientation-related observations, including an appearance of increased cellularity and/or vacuolation in the neurohypophysis, a review of canine pituitary gland slides was performed to document sectioning planes and associated histologic appearance.

Methods

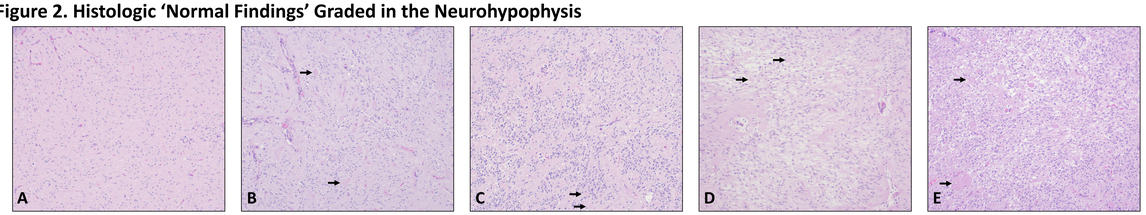
Pituitary gland slides were reviewed from two canine studies conducted according to good laboratory (GLP) practices without systemic test article-related effects. Dogs were ≥5 months old. Pituitary sections were classified by approximate section plane orientation (Figure 1), cysts were recorded, and findings (vacuolation, increased cellularity, and eosinophilic globules) within the neurohypophysis were graded on a scale of minimal (1) to severe (5). Historical control data for pituitary gland findings (Nov 2022-Dec 2024) were also compiled.

Figure 1. Pituitary Gland Section Plane Classification



Representative images (20x) of histologic section plane classification: (A) straight/distal transverse, (B) angled/proximal transverse, (C) angled/parasagittal longitudinal, (D) straight/sagittal longitudinal, and (E) fragmented/other.

Figure 2. Histologic 'Normal Findings' Graded in the Neurohypophysis



Example images (100x) of graded histologic findings: (A) no findings, (B) minimal increased cellularity, vacuolation, and eosinophilic globules (arrows indicate examples of globules), (C) mild increased cellularity, minimal vacuolation and globules, (D) mild vacuolation, increased cellularity, and minimal globules, and (E) mild vacuolation, increased cellularity, and globules.

Results

Pituitary sections of 79 dogs were reviewed, with findings summarized in Table 1.

Table 1. Summary of Pituitary Gland Section Plane Classification and Findings in the Neurohypophysis

Approximate Section Plane Orientation	Straight/ Distal Transverse	Angled/ Proximal Transverse	Angled/ Parasagittal Longitudinal	Straight/ Sagittal Longitudinal	Fragmented/ Other
Number (percent of total pituitary sections)	14 (18%)	31 (39%)	17 (22%)	7 (9%)	10 (13%)
Neurohypophysis absent in section (percent within section plane type)	1 (7%)	3 (10%)	0 (0%)	0 (0%)	1 (10%)
Cyst present (percent within section plane type)	8 (62%)	19 (68%)	10 (59%)	2 (29%)	6 (67%)
Findings Graded Within Neurohypophysis[a]					
Vacuolation	4 (31%)	20 (71%)	15 (88%)	7 (100%)	2 (22%)
Average histologic grade	0.5	1.0	1.5	1.6	0.3
Increased cellularity	4 (31%)	24 (86%)	16 (94%)	7 (100%)	9 (100%)
Average histologic grade	0.5	1.4	1.9	1.9	1.2
Eosinophilic globules (consistent with neurosecretory/Herring bodies)	2 (15%)	15 (54%)	13 (76%)	3 (43%)	2 (22%)
Average histologic grade	0.2	0.6	0.9	0.4	0.2

a = percentages calculated from pituitaries with neurohypophysis present in section

Historical control data encompassed 12 GLP studies with 87 animals (44 males, 43 females). The only observations were cysts in 18 (21%) animals (7, 16% of males; 11, 26% of females). Cyst sublocation (pars distalis or pars intermedia) was not consistently recorded.

Conclusions

- There is variability of canine pituitary gland sectioning and accompanying variation in histologic appearance.
- An appearance of vacuolation (minimal-mild) and increased cellularity (minimal-moderate) within the neurohypophysis occurred at increasing incidence and grade with section proximity to the third ventricle (infundibular) recess, from 31% in straight/distal transverse to 100% in straight/sagittal longitudinal sections. This is consistent with *normal anatomy* and mimics the "pituitary bright signal" seen on magnetic resonance imaging as a hyperintensity around the infundibular recess.
- Eosinophilic globules consistent with Herring bodies (neurosecretory bodies, reported to contain antidiuretic and oxytocin hormones) were most common in angled/parasagittal longitudinal sections, consistent with *normal anatomy* due to greater visualized area of the pituitary stalk and proximity to the hypothalamus.
- Cysts occurred in ~2/3 of most section planes but were less frequent in straight/sagittal longitudinal sections. The lower rate of cyst identification in straight/sagittal longitudinal samples was intriguing, but inconclusive due to low numbers of this section orientation. There was no correlation between cysts and appearance of cellularity and/or vacuolation.
- Despite a higher rate of 'normal findings', reviewers preferred the relative uniformity of straight/sagittal longitudinal sections and found that inclusion of the infundibular recess was a helpful reference point in contextualizing variations in histoarchitecture related to normal anatomy.

Future Directions

- We are continuing to explore standardized sectioning approaches for pituitary glands of dogs and other species in nonclinical toxicology studies.
- Standardization of pituitary gland sectioning is expected to improve interindividual consistency and avoid potential confusion with 'non-lesions'.
- Consideration of sectioning approach is especially warranted when pituitary gland is a likely target tissue.

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Acknowledgements

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