AJNR

Histochemical Characterization and Functional Significance of the Hyperintense Signal on MR Images of the Posterior Pituitary

John Kucharczyk, Walter Kucharczyk, Isabelle Berry, Jack de Groot, William Kelly, David Norman and T. H. Newton

AJNR Am J Neuroradiol 1988, 9 (6) 1079-1083
http://www.ajnr.org/content/9/6/1079

Histochemical Characterization and Functional Significance of the Hyperintense Signal on MR Images of the Posterior Pituitary

John Kucharczyk[1,2]
Walter Kucharczyk[3]
Isabelle Berry[1,4]
Jack de Groot[5]
William Kelly[6]
David Norman[1]
T. H. Newton[1]

MR imaging of the pituitary fossa characteristically shows a well-circumscribed area of high signal intensity in the posterior lobe on T1-weighted images. We used a combination of high-field MR, electron microscopy, and histologic techniques in experimental animals to determine whether the hyperintensity of the posterior lobe might be functionally related to hormone neurosecretory processes, and to attempt to establish its chemical nature. Histologic sections of a dog's pituitary gland processed with lipid-specific markers showed intense staining in the posterior lobe but not in the anterior lobe, thus documenting the location of fat in the posterior pituitary. Administration of vasoactive drugs known to influence vasopressin secretion to anesthetized cats produced changes in the volume of high-intensity signal in the posterior pituitary. Subsequent electron microscopy showed a significant increase in posterior lobe glial cell lipid droplets and neurosecretory granules in dehydration-stimulated cats.

The data suggest that the pituitary hyperintensity represents intracellular lipid signal in the glial cell pituicytes of the posterior lobe or neurosecretory granules containing vasopressin. The volume of the signal may, in turn, reflect the functional state of hormonal release from the neurohypophysis.

While CT and MR imaging can both be used to evaluate patients with suspected pituitary disease, high-field high-resolution MR imaging is increasingly the method of choice [1–3]. The inherently superior contrast provided by MR is especially advantageous in the region of the sella turcica, where numerous biochemically heterogeneous soft-tissue structures and fluid compartments are located in close anatomical association.

Several MR studies of the sella have shown that a well-defined area of high signal intensity on T1-weighted images, located just anterior to the dorsum, corresponds to a variable portion of the posterior lobe [4, 5]. Furthermore, the size, shape, and exact position of this bright signal are influenced by intrasellar and suprasellar lesions [1–5]. In particular, the absence of high signal intensity in the posterior lobe of patients with central diabetes insipidus suggests a possible functional relationship to hypothalamic–pituitary hormone secretory processes [1–3].

The peptide hormones vasopressin and oxytocin are synthesized in the supraoptic and paraventricular nuclei of the hypothalamus and are transported by axoplasmic flow into the posterior lobe for subsequent release into the circulation. Histologically, the posterior pituitary is composed of unmyelinated nerve fibers, axon terminals containing the neurosecretory products, and a variable number of astrocytic glial cells or "pituicytes" [6–10]. The pituicytes are found in close contact with axon terminals containing vasopressin and, in both humans [11] and various experimental animals [12–20], show signs of raised metabolic activity and increased intracellular lipid content under conditions that promote increased hormone release. In the present study, we investigated the possibility that the high-intensity signal in T1-weighted images of the posterior lobe may be due to intracellular lipid in the pituicytes.

This article appears in the November/December 1988 issue of *AJNR* and the January 1989 issue of *AJR*.

Received December 16, 1987; accepted after revision July 18, 1988.

Presented at the annual meeting of the American Society of Neuroradiology, New York, May 1987.

[1] Department of Radiology, L371, University of California, San Francisco, 505 Parnassus, San Francisco, CA 94143. Address reprint requests to J. Kucharczyk.

[2] Department of Physiology, University of Ottawa, Ottawa, Ontario, Canada K1H 8M5.

[3] Department of Radiology, The Toronto Hospital, Toronto, Ontario, Canada M5G 2C4.

[4] Service de Neuroradiologie, Hopital Purpan, Tourlouse, France.

[5] Department of Anatomy, University of California, San Francisco, CA 94043.

[6] Department of Radiology, David Grant USAF Hospital, Travis Air Force Base, CA 94535.

***AJNR* 9:1079–1083, November/December 1988**
0195–6108/88/0906–1079

Materials and Methods

To establish the location of the high signal intensity seen on T1-weighted images of the sella turcica, the pituitary gland and sella turcica of a dog were examined under light microscopy. After the animal was sacrificed by barbiturate overdose, the sphenoid bone with its intrasellar contents intact was excised and immediately imaged with T1-weighted sagittal and T1-weighted axial sections, 600/20 (TR/TE). The pituitary gland was then removed, fixed, and stained with Oil Red O, a lipid-specific stain, and examined at ×100 magnification. The staining characteristics of the anterior and posterior lobes of the pituitary were compared.

MR studies were then carried out on five 16–24-hr food-deprived adult cats and a single dog tranquillized with acepromazine (2–4 mg/kg, IM), and subsequently anesthetized with sodium pentobarbital (30–35 mg/kg, IV) or halothane. Animals were ventilated through an endotracheal tube in order to maintain normal pO_2 (100–150 torr) and pCO_2 (27–35 torr) levels. A femoral artery and vein were catheterized for blood pressure monitoring and drug administration. The rectal temperature was maintained at 37± 0.5°C with an electronically controlled body heating pad.

Images were acquired on a GE CSI-II imager/spectrometer operating at 2T and an 8.3-cm inner-diameter homebuilt "birdcage" head coil tuned to the proton resonant frequency. T1-weighted (450/20) and T2-weighted (3000/40,80) 2-mm-thick (2DFT) or 1.25-mm-thick (3DFT) contiguous images were obtained in the sagittal and coronal planes in a 3 × 8-cm field of view, and 128 × 256-pixel acquisition matrix. The scan range was adjusted to ensure that the median eminence was completely within the margins of a single section location. All images were retrospectively reviewed by three or more observers with attention focused on the architectural features and intrinsic signal characteristics of the hypothalamic–pituitary axis.

Subsequently, T1- and T2-weighted midsagittal images were obtained before and after administration of epinephrine (1–10 μg/kg, bolus IV, three injections in three cats), isoproterenol (10–50 μg/kg, bolus IV, four injections in three cats) and sodium pentobarbital (10–30 mg/kg, IV, four injections in four cats). Images were produced at 5–10 min intervals for 30–60 min after administration of each drug in order to determine whether the intensity or volume of the pituitary "bright spot" had been altered. Arterial blood pressure was recorded continuously during this period. At the conclusion of each MR study, the animal was sacrificed by IV barbiturate overdose. The pituitary fossa of one intact cat was reimaged 72 hr postmortem.

Ultrastructural features of the pituitary were examined in 24–48-hr water-deprived cats ($n = 2$) and rats ($n = 4$), and in normally hydrated controls (one cat and two rats). Animals were killed by halothane or isoflurane overdose and prepared for electron microscopy in accordance with methods described previously [12, 21]. Briefly, animals were perfused via IV with 0.9% NaCl followed by 4% glutaraldehyde in 0.1 M sodium cacodylate buffer (pH 7.4) containing 0.5% dimethyl sulfoxide. The pituitary gland was removed, separated into anterior and posterior lobes under ×40 magnification, and the two lobes were stored overnight in fixative. Blocks of the neurohypophysis were then removed with a hypodermic needle, treated with Os 04, and embedded. Uranyl acetate en bloc staining was carried out after osmication.

Sections 1–2 μm thick were cut through the blocks to establish the presence of neural lobe tissue. Thin sections were cut at various levels and stained with lead citrate. Prints of electron micrographs of individual pituicytes were analyzed at a final magnification of ×7500–26,000.

Results

MR imaging of the dog's excised sella turcica and pituitary gland revealed the typical hyperintense region in the posterior

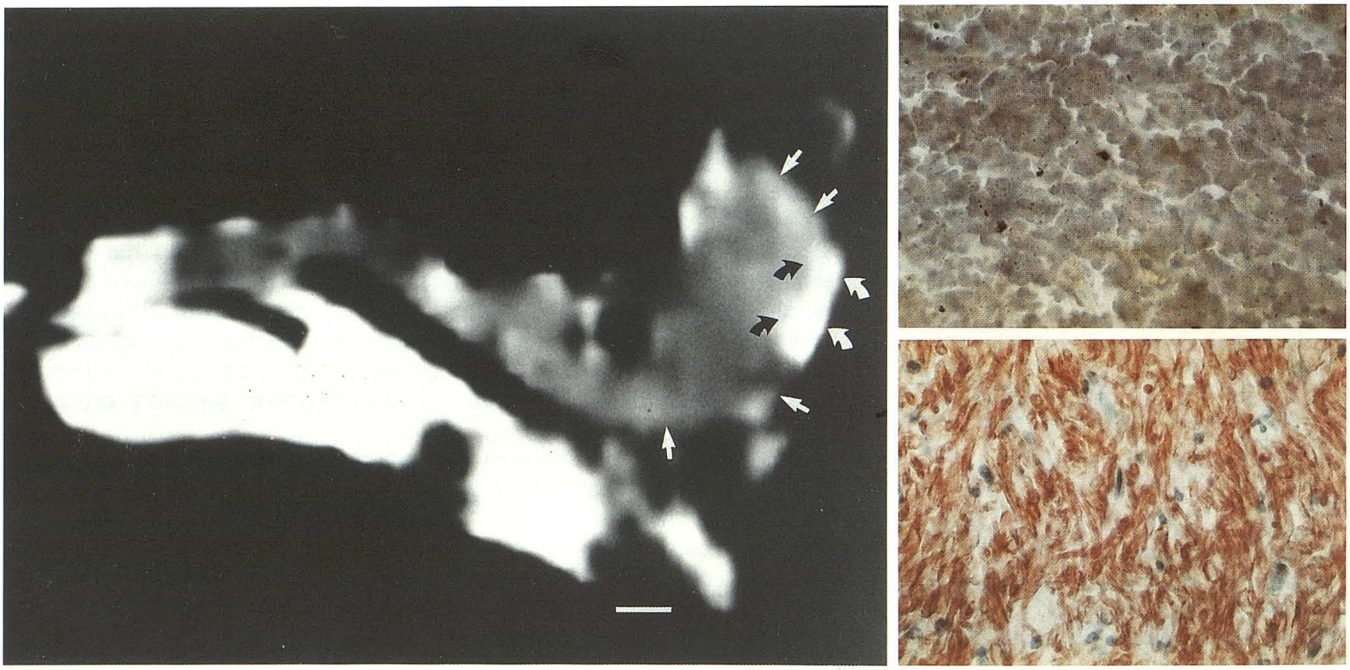


Fig. 1.—*A*, T1-weighted sagittal MR image of excised sphenoid bone of dog. Large area of high signal intensity represents marrow within sphenoid bone. Low-signal cortical bone of sella floor is identified by *straight arrows*. Intermediate-intensity tissue superior to floor is the anterior pituitary. High-intensity posterior pituitary is identified by *curved arrows*. Horizontal bar = 1 mm.

***B*, Oil Red O stain of anterior (*top*) and posterior (*bottom*) lobes at ×100 magnification shows typical architectures. Note relative paucity of fat in anterior lobe compared with abundant red-staining lipid material in posterior pituitary.**

sella (Fig. 1A). Histologic sections with Oil Red O staining revealed abundant fat in the posterior lobe (Fig. 1B), which we believe is the source of the bright signal on T1-weighted images. An earlier report [22] had assigned this hyperintensity to the "sellar fat pad," a point with which we disagree.

MR studies of the cats' pituitary fossa also showed areas of hyperintense signal on T1-weighted images in the posterior–inferior part of the sella (Fig. 2A) similar to MR studies of human pituitary [2, 3, 5]. As in the human, this high signal intensity was usually biconvex or semilunar in shape and of variable size between individual animals.

On T2-weighted images, the posterior lobe hyperintensity differed from adjacent areas of high signal. Although the intensity diminished with progressively increased T2 weighting, the degree of signal loss was less than that of marrow in the dorsum and clivus. Thus, the MR signal characteristics of the posterior pituitary lobe of the cat and dog appear to be very similar to those reported in human studies [5].

In the sequential MR studies of drug effects, the observers agreed that an IV dose of epinephrine, which raised mean arterial blood pressure (MAP) by 40–60 mm Hg, produced a slight decrease in the volume of the pituitary "bright spot" (Fig. 2B). On the other hand, administration of the beta-adrenergic agonist isoproterenol, in doses that reduced MAP by 40–80 mm Hg, increased the volume of the bright signal (Fig. 2C). Qualitatively similar changes were observed in each of the other animals tested with epinephrine and isoproterenol. Changes in MAP of as little as 10 mm Hg are known to influence vasopressin biosynthesis and release by baroreceptor mechanisms [23]. A small segment of the high-intensity signal was still present on a T1-weighted image taken 72 hr postmortem (Fig. 2D). The possible significance of this latter observation is not clear at this time.

In the next series of experiments, we attempted to elucidate the specific location and possible significance of the hyperintense signal by examining the ultrastructural characteristics of the posterior lobe. In electron micrographs of the cats' neurohypophysis, pituicytes were identified by the presence of nuclei, ribosomes, Golgi complexes, and liposomes in their cytoplasm. Neurosecretory axon terminals with their characteristic vesicles were also seen adjacent to or directly opposed to the pituicytes. The major difference observed between the normally hydrated (Fig. 3A) and dehydrated (Fig. 3B) neurohypophysis was a large increase in the number and

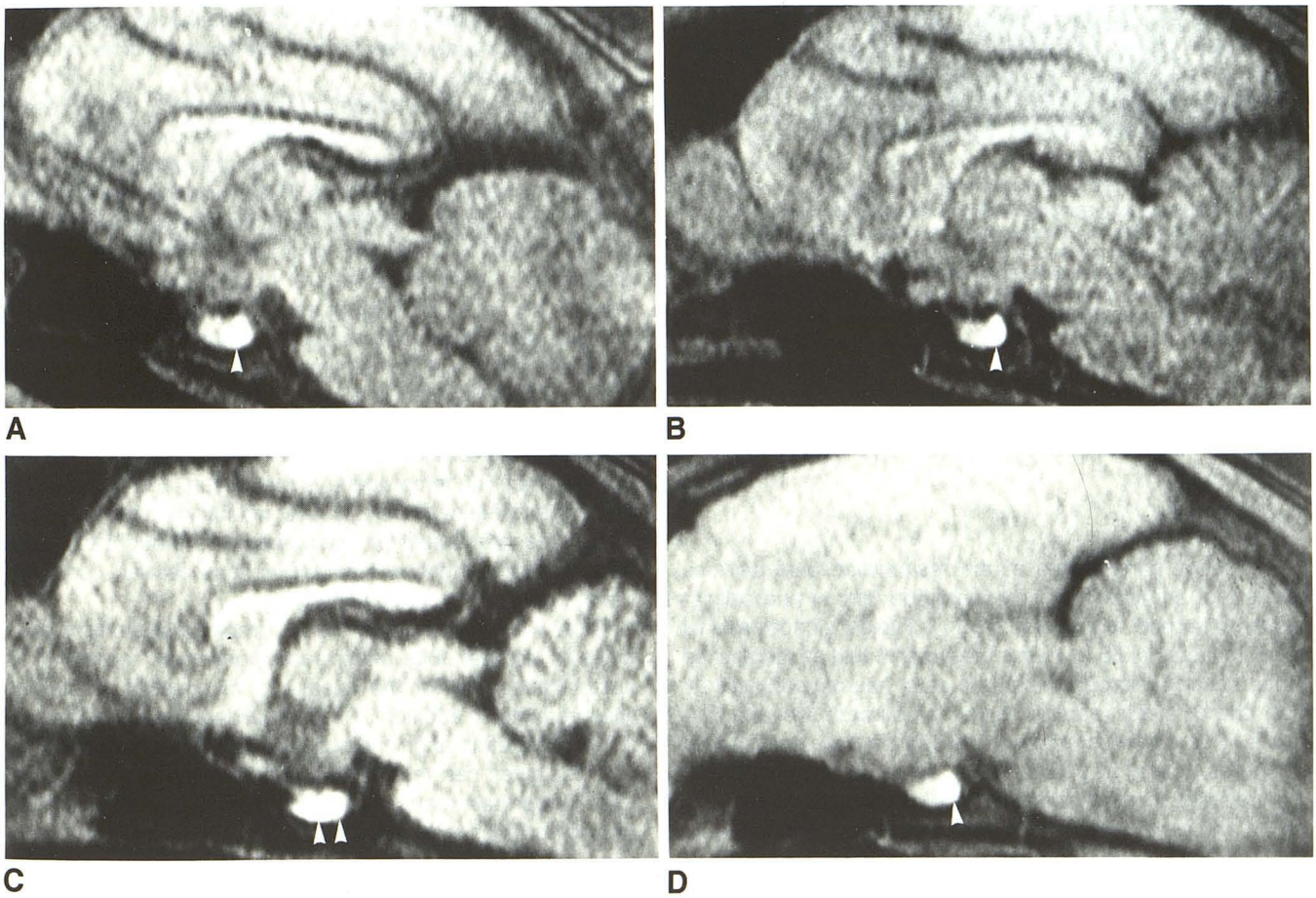
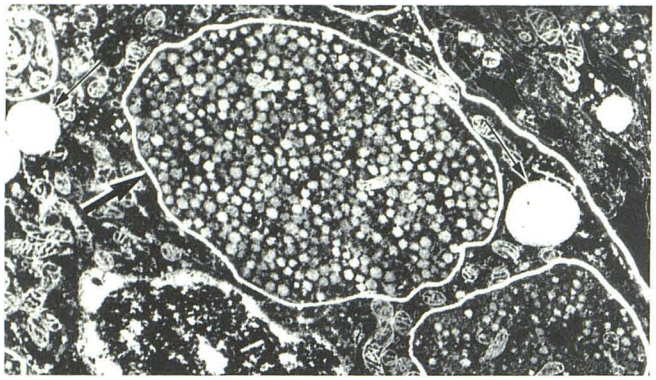
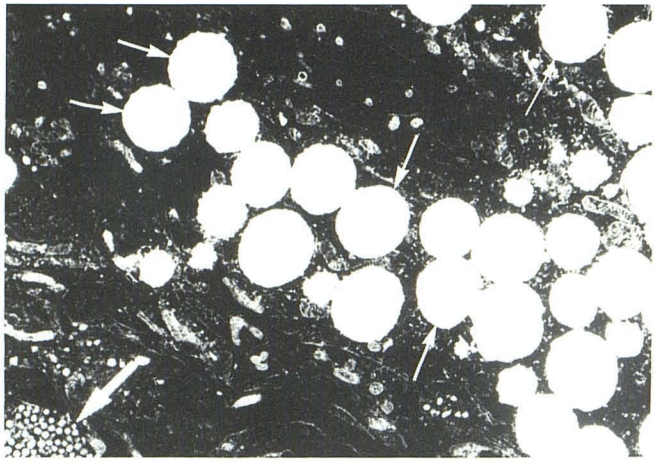


Fig. 2.—Midsagittal MR image of cat brain. T1-weighted spin-echo images (450/15/4) 3-mm slice thickness, 8 × 8-cm field of view, 128 × 256 matrix. *A*, Preinjection control image; *B*, 1 min after IV injection of 1 μg/kg epinephrine; *C*, 5 min after IV injection of 5 μg/kg isoproterenol; *D*, same cat brain reimaged 72 hr postmortem. Volume of hyperintense signal in posterior pituitary (*arrowheads* in all images) appears to be slightly decreased after epinephrine injection and increased after isoproterenol administration. Some hyperintensity persisted for 3 days postmortem.



A



B

Fig. 3.—Electron micrographs of single pituicytes from posterior pituitary of normally hydrated (*A*) and 48-hr water-deprived (*B*) cats. Shown is part of the pituicyte cytoplasm with lipid droplets (*smaller arrows*) and neurosecretory axon terminals (boundaries drawn) containing vasopressin and oxytocin (*larger arrows*). Note that the number of lipid droplets is much increased in the dehydrated animal. Magnification: ×26,400 (*A*) and ×28,700 (*B*).

size of lipid droplets interspersed throughout the pituicyte cytoplasm in the dehydrated animals.

Discussion

The neural parenchyma of the mammalian pars nervosa consists of unmyelinated neurosecretory axons and cellular elements termed "pituicytes" [7, 11]. Typical slender axonal fibers, measuring 0.5 to 1 μm in diameter, could be recognized in our electron micrographs of the cats' neurohypophysis by their characteristic parallel rows of neurotubules and filaments. The neurosecretory axons contain the usual axonal organelles as well as 100–200 nm dense core vesicles [24]. The vesicles in effect package the neurohypophysial hormones and carrier proteins that are produced in the perikarya of the supraoptic and paraventricular nuclei of the hypothalamus, and are then transported axonally to the posterior lobe for storage in the axon terminals or release into the perivascular space [8].

Studies in a number of mammalian species, including the human [11], have shown that pituicytes make up 25–30% of the volume of the neural lobe [25] and are closely associated with the neurosecretory process. In electron micrographs in our study, these astrocytic glial cells were frequently seen to have neurosecretory axonal processes indented into their cytoplasm, suggesting the possibility of neuro–glial communication in the process of hormone release [12, 13]. An even more prominent ultrastructural feature was the large increase in lipid droplets in the posterior lobe pituicytes of cats that had been water-deprived for 48-hr prior to electron microscopy. Generalized pituicyte hypertrophy [15], as well as an increase in pituicyte electron-dense cytoplasmic bodies [16] and intracellular lipid droplets [12–15, 17, 26] have previously been noted in animals under conditions in which neurosecretion of vasopressin is stimulated, such as several days of water deprivation or salt loading.

The results of our histologic study with lipid-specific marker confirm that the posterior lobe, unlike the anterior pituitary, has a high lipid content. It thus seems probable that the hyperintense signal observed on T1-weighted images of the posterior sella represents lipids contained in the pituicyte glial cells of the posterior lobe. The finding that the volume of the hyperintense area on MR could be increased by pharmacological manipulations known to increase plasma vasopressin levels further supports the view that the lipid content of the pituicytes is closely related to the state of hormone release from the posterior lobe.

The functional importance of the intracellular lipids present in the posterior pituitary is unknown. There has been speculation that phagocytosis may be a physiological role for pituicytes. After surgical transection of the hypothalamic–pituitary stalk in rats, the axons in the neural lobe degenerate and are phagocytosed by these astroglial cells [25]. On the basis of the results of their ultrastructural study of the human neural lobe, Takei et al. [11] proposed that pituicytes provide a cytoplasmic machinery to catabolize "unused or excess" neurosecretory material. In the human, at least, the large increase in pituitary fat droplets during periods of intense neurosecretory activity is paralleled in the neuronal perikarya by a great increase in the number of lysosomes. Additionally, an increase in the number of hematogenous monocytes and perivascular histiocytes is seen in the posterior pituitary at that time [11]. The increased number of macrophages is thought to reflect an elevated metabolism involving various cellular, especially membrane-bound, constituents [11].

Thus, while the origin of the fat droplet inclusions in pituicytes is not yet fully understood, they may represent an accumulation site for the release or disposal of membrane lipid products. In addition to lysosomal degradation of material in neurosecretory axon endings [20], there is indirect evidence that pituicytes can take part in lipid storage and catabolism [15]. Lipoprotein material resulting from excretory and autophagic activities in axonal endings may therefore be temporarily stored in the form of lipid droplets prior to reutilization in the formation of a new membrane. Although we are not aware of any direct proof for such a transfer of lipid material in the posterior pituitary, membrane phospholipid breakdown,

storage, and reutilization have been demonstrated in several other secretory cells [27]. Such an explanation would account for the increased liposome content of pituicytes observed in electron micrographs of the actively secreting neurohypophysis, and may explain the high signal intensity of the posterior lobe in T1-weighted MR images.

REFERENCES

1. Kucharczyk W, Davis DO, Kelly WM, et al. Pituitary adenomas: high resolution MR imaging at 1.5 T. *Radiology* **1986**;161:761–765
2. Fujisawa I, Nishimura K, Asoto R, et al. Posterior lobe of the pituitary in diabetes insipidus: MR findings. *J Comput Assist Tomogr* **1987**;11:221–225
3. Fujisawa I, Asato R, Nishimura K, et al. Anterior and posterior lobes of the pituitary gland: assessment by 1.5 T MR imaging. *J Comput Assist Tomogr* **1987**;11:214–220
4. Kelly WM, Kucharczyk W, Kucharczyk J, et al. Posterior pituitary ectopia: an MR feature of pituitary dwarfism. *AJNR* **1988**;9:453–460
5. Columbo N, Berry I, Kucharczyk J, et al. Posterior pituitary gland: appearance on magnetic resonance images in normal and pathologic states. *Radiology* **1987**;165:481–485
6. Decherney A, Naftolin F. Hypothalamic and pituitary development in the fetus. *Clin Obstet Gynecol* **1980**;23:749
7. Bucy PC. The pars nervosa of the bovine hypophysis. *J Comp Neurol* **1930**;50:505–519
8. Dreifuss JJ. A review on neurosecretory granules, their contents and mechanisms of release. *Ann NY Acad Sci* **1975**;258:184–201
9. Okado N, Yokata N. An electron microscopic study of the structural development of the neural lobe in the human fetus. *Am J Anat* **1980**;159:261
10. Holmes RL, Ball JN. *The pituitary gland, a comparative account*. London: Cambridge University Press, **1974**:63–94
11. Takei Y, Seyama S, Peal GS, et al. Ultrastructural study of the human neurohypophysis. II: Cellular elements of the neural parenchyma, the pituicytes. *Cell Tiss Res* **1980**;205:273–287
12. Tweedle CD, Hatton GI. Evidence for dynamic interactions between pituicytes and neurosecretory axons in the rat. *Neuroscience* **1980**;5:661–667
13. Tweedle CD. Ultrastructural manifestations of increased hormone release in the neurohypophysis. In: Cross BA, Leng G, eds. *The neurohypophysis: structure, function and control. Progress in brain research*, vol. 60. Amsterdam: Elsevier, **1983**
14. Tweedle CD, Hatton GI. Ultrastructural changes in rat hypothalamic neurosecretory cells and their associated glia during minimal dehydration and rehydration. *Cell Tiss Res* **1977**;181:59–92
15. Krsulovic J, Bruckner G. Morphological characteristics of pituicytes in different functional stages. *Z Zellforsch* **1969**;99:210–220
16. Davis E, Morris JF. Lyposomes and Herring bodies in the neural lobe of saline and pitressin-treated rats. *J Anat* **1973**;114:291–292
17. Levesque F, Small M. The relationship of the pituicyte to the posterior lobe hormones. *Endocrinology* **1959**;65:909–915
18. Tweedle CD, Hatton GI. Magnocellular neuropeptidergic terminals in neurohypophysis: rapid glial release of enclosed axons during parturition. *Brain Res Bull* **1982**;8:205–209
19. Boer GJ, van Rheenen-Verberg CMF. Acid phosphate in rat neurohypophyseal dispersions and its fractions enriched for neurosecretosomes and pituicytes after water deprivation and lactation. *Brain Res* **1976**;114:279–292
20. Kodama Y, Fujita H. Some findings on the fine structure of the neurohypophysis in dehydrated and pitressin treated mice. *Arch Histol Jap* **1975**;38:121–131
21. Karnovsky MJ. The ultrastructural basis of capillary permeability studied with peroxide as a tracer. *J Cell Biol* **1967**;35:213
22. Mark L, Pech MP, Daniels D, et al. Pituitary fossa: a correlative anatomic and MR study. *Radiology* **1984**;153:453–457
23. Dunn FL, Brennan TJ, Nelson AE, et al. The role of blood osmolality and volume in regulating vasopressin secretion in the rat. *J Clin Invest* **1973**;52:3212–3219
24. Seyama S, Pearl GS, Takei Y. Ultrastructural study of the human neurohypophysis. I: Neurosecretory axons and their dilatations in the pars nervosa. *Cell Tiss Res* **1980**;205:253–271
25. Nordmann JJ. Ultrastructural morphometry of the rat neurohypophysis. *J Anat* **1977**;123:213–218
26. Dellman HD, Stoeckel ME, Porte A, et al. Ultrastructure of neurohypophysial glial cells following stalk transection in the rat. *Experientia* **1974**;30:1220–1222
27. Hokin LE, Huebner D. Radioautographic localization of the increased synthesis of the phosphatidylinositol in response to pancreozymin of acetylcholine in guinea pig pancreas slices. *J Cell Biol* **1967**;33:521–530