

The Visual Pulvinar in Tree Shrews I. Multiple Subdivisions Revealed through Acetylcholinesterase and Cat-301 Chemoarchitecture

DAVID C. LYON, NEERAJ JAIN, AND JON H. KAAS*
Department of Psychology, Vanderbilt University, 301 Wilson Hall,
Nashville, Tennessee 37203

ABSTRACT

Tree shrews are highly visual mammals closely related to primates. They have a large visual pulvinar complex, but its organization and relation to visual cortex is only partly known. We processed brain sections through the pulvinar with seven different procedures in an effort to reveal histologically distinct compartments. The results revealed three major subdivisions. A dorsal subdivision, Pd, stains darkly for acetylcholinesterase (AChE) and occupies the dorsoposterior one-third of the pulvinar complex. A ventral subdivision, Pv, stains darkly when processed with the Cat-301 antibody and occupies the ventroanterior fifth of the pulvinar complex along the brachium of the superior colliculus. Unexpectedly, part of Pv is ventral to the brachium. A large central subdivision, Pc, stains moderately dark for AChE and cytochrome oxidase (CO), and very light for Cat-301. Pc includes about half of the pulvinar complex, with parts on both sides of the brachium of the superior colliculus. These architectonic results demonstrate that the pulvinar complex of tree shrews is larger and has more subdivisions than previously described. The complex resembles the pulvinar of primates by having a portion ventral to the brachium and by having histochemically distinct nuclei; the number of nuclei is less than in primates, however. *J. Comp. Neurol.* 467:593–606, 2003.

© 2003 Wiley-Liss, Inc.

Indexing terms: cerebral cortex; lateral posterior nucleus; primate; thalamus; Tupaia; vision

In the visual system of mammals, the pulvinar complex, sometimes identified as the lateral posterior nucleus, is the main source of subcortical input to extrastriate cortex (Jones, 1985). In monkeys, the relatively large pulvinar (Grieve et al., 2000) has been parsed into several anatomically defined subdivisions, each with a distinct chemoarchitecture (Cusick et al., 1993; Gutierrez et al., 1995, 2000; Stepniewska and Kaas, 1997; Gray et al., 1999) and a unique pattern of connections with visual cortex (Cusick et al., 1993; Gutierrez and Cusick, 1997; Beck and Kaas, 1998; Stepniewska et al., 1999, 2000; Adams et al., 2000; Shipp, 2001; Soares et al., 2001; Weller et al., 2002).

As in primates, the pulvinar complex of the tree shrew is relatively large (Diamond et al., 1970; see Results). Portions of the pulvinar receive visual inputs from the retina (Hubel, 1975; Ohno et al., 1975; Somogyi et al., 1981) or the superior colliculus (Harting et al., 1973b; Casagrande and Harting, 1975; Albano et al., 1979; Lupino et al., 1988). The pulvinar projects to striate and extrastriate visual cortex (Diamond et al., 1970; Harting

et al., 1973a; Carey et al., 1979; Lupino et al., 1988). Previously, two subdivisions of the pulvinar have been described as receiving either “diffuse” or “specific” (or topographic) projections from the superior colliculus (Lupino et al., 1988). The region receiving “diffuse” input from

Grant sponsor: National Eye Institute; Grant number: EY-02686; Grant number: 5T32 EY07135.

Dr. Lyon's current address: Picower Center for Learning and Memory, Department of Brain and Cognitive Sciences, Massachusetts Institute of Technology, 45 Carleton Street, E25-235, Cambridge, MA 02139.

Dr. Jain's current address: National Brain Research Centre, Nainwal Mode, Manesar, Haryana 122 050, India.

*Correspondence to: Jon H. Kaas, Department of Psychology, Vanderbilt University, 301 Wilson Hall, Nashville, TN 37203.
E-mail: jon.h.kass@Vanderbilt.edu

Received 20 June 2003; Revised 14 August 2003; Accepted 18 August 2003
DOI 10.1002/cne.10939

Published online the week of November 3, 2003 in Wiley InterScience (www.interscience.wiley.com).

the superior colliculus is located dorsomedially and projects to extrastriate visual cortex located ventral and posterior in the temporal lobe, whereas the remainder of the pulvinar receives "specific" projections from the superior colliculus and projects to V2 and cortex located dorsally in the temporal lobe. No further pulvinar subdivisions have been described.

Tree shrews are of particular interest to us because they are generally considered to be a close relative of primates (Kaas and Preuss, 1993; Kaas, 2002). Tree shrews belong to the mammalian order of Scandentia, which includes a small radiation of 18 species distributed in Southeast Asia. Like primates, tree shrews are highly visual mammals. Tree shrews have good visual acuity (Petry et al., 1984), possess retinas in which cones form a large percentage of photoreceptor cells (Petry and Kelly, 1991), a six-layered LGN containing both X- and Y-like ganglion cells (Conley et al., 1984; Holdefer and Norton, 1995), a large and distinctly layered superior colliculus (Casagrande et al., 1972; Albano et al., 1979), ocular dominance (Hubel, 1975; Humphrey et al., 1977), on/off cells (Kretz et al., 1986) sublayers in layer 4 of striate cortex (V1), myelin anti-blobs in superficial layers of V1 (Lyon et al., 1998), intricate patterns of intrinsic connections between layers (Fitzpatrick, 1996), long-range lateral connections in superficial layers of V1 (Rockland and Lund, 1982; Rockland et al., 1982) that interconnect neurons of similar orientation selectivity (Mitchison and Crick, 1982; Bosking et al., 1997), band-like modular compartments in V2 (Sesma et al., 1984; Lyon et al., 1998), and at least seven distinct extrastriate visual areas (Sesma et al., 1984; Luppino et al., 1988; Jain et al., 1994; Lyon et al., 1998).

Recently, a number of chemoarchitectonic and immunohistochemical techniques have been used to reveal previously undetected subdivisions of the primate pulvinar (Stepniewska and Kaas, 1997; Gutierrez et al., 2000). However, such methods have not been used to reveal subdivisions of the pulvinar complex in tree shrews. Because of their close ties to the primate lineage, tree shrews may offer clues to the evolution of the primate visual system. Therefore, we employed these new staining techniques to investigate whether tree shrew pulvinar can

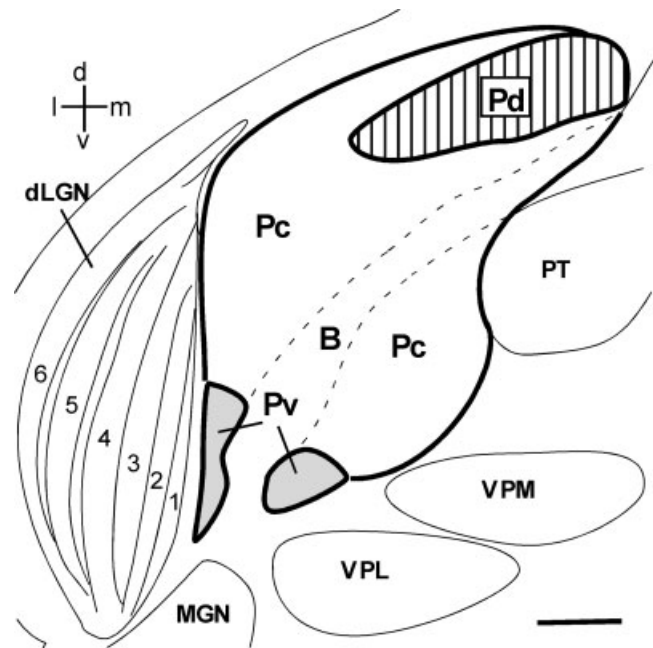


Fig. 1. Subdivisions of the pulvinar in tree shrews. The pulvinar is subdivided into dorsal (Pd; hatching), central (Pc; gray shading) nuclei based on staining patterns of Cat-301 and AChE (Fig. 2). The pulvinar complex (Pd, Pc, and Pv) is outlined in bold. The dashed line through the pulvinar represents the intervening brachium (B) of the superior colliculus. Sections were made in a coronal plane. Dorsal (d) is toward the top, and lateral (l) is to the left. For abbreviations, see list. Scale bar = 500 μ m.

also be further subdivided architectonically. We show that at least three architectonically defined subdivisions of the pulvinar nucleus exist in tree shrews. In a companion paper we describe the cortical connections of these subdivisions (Lyon et al., 2003).

MATERIALS and METHODS

Histology and data analysis

All procedures were approved by the Vanderbilt Institutional Animal Care and Use Committee, and followed NIH guidelines. Three adult tree shrews (*Tupaia belangeri*) were anesthetized with ketamine hydrochloride (30 mg/kg) and xylazine (10 mg/kg) and then given a lethal dose of sodium pentobarbital (50 mg/kg). When areflexive, they were perfused transcardially with 0.9% phosphate-buffered saline (PBS; pH 7.4) followed by a solution of 4% paraformaldehyde fixative, and subsequently 4% paraformaldehyde with 10% sucrose in PBS. The brain was removed and the cortex separated from the thalamus. Additional material came from tree shrews used in studies of thalamocortical connections (Lyon et al., 2003) in which 2% fixative was used to allow for easier flattening of the cortex.

Separate thalami were sectioned at 40 μ m in different planes, including coronal, parasagittal, and horizontal. Series of sections were processed for acetylcholinesterase (AChE; Stepniewska and Kaas, 1997), calbindin (Celio, 1990), Cat-301 (Hockfield and McKay, 1983; Hendry et al., 1984, 1988; Jain et al., 1994), cytochrome oxidase (CO;

Abbreviations

AChE	acetylcholinesterase
B	brachium of the SC
CO	cytochrome oxidase
IC	inferior colliculus
IGL	intergeniculate leaflet
Intra.	intralaminar nucleus
Lat.	lateral intermediate nucleus
LD	lateral dorsal nucleus
LGN	dorsal lateral geniculate nucleus
MGN	medial geniculate nucleus
Pc	central nucleus of the pulvinar
Pd	dorsal nucleus of the pulvinar
Po	posterior nucleus
PT	pretectum
Pv	ventral nucleus of the pulvinar
R	reticular nucleus
SC	superior colliculus
VL	ventral lateral nucleus
vLGN	ventral lateral geniculate nucleus
VP	ventroposterior nucleus
VPL	lateral ventroposterior nucleus
VPM	medial ventroposterior nucleus

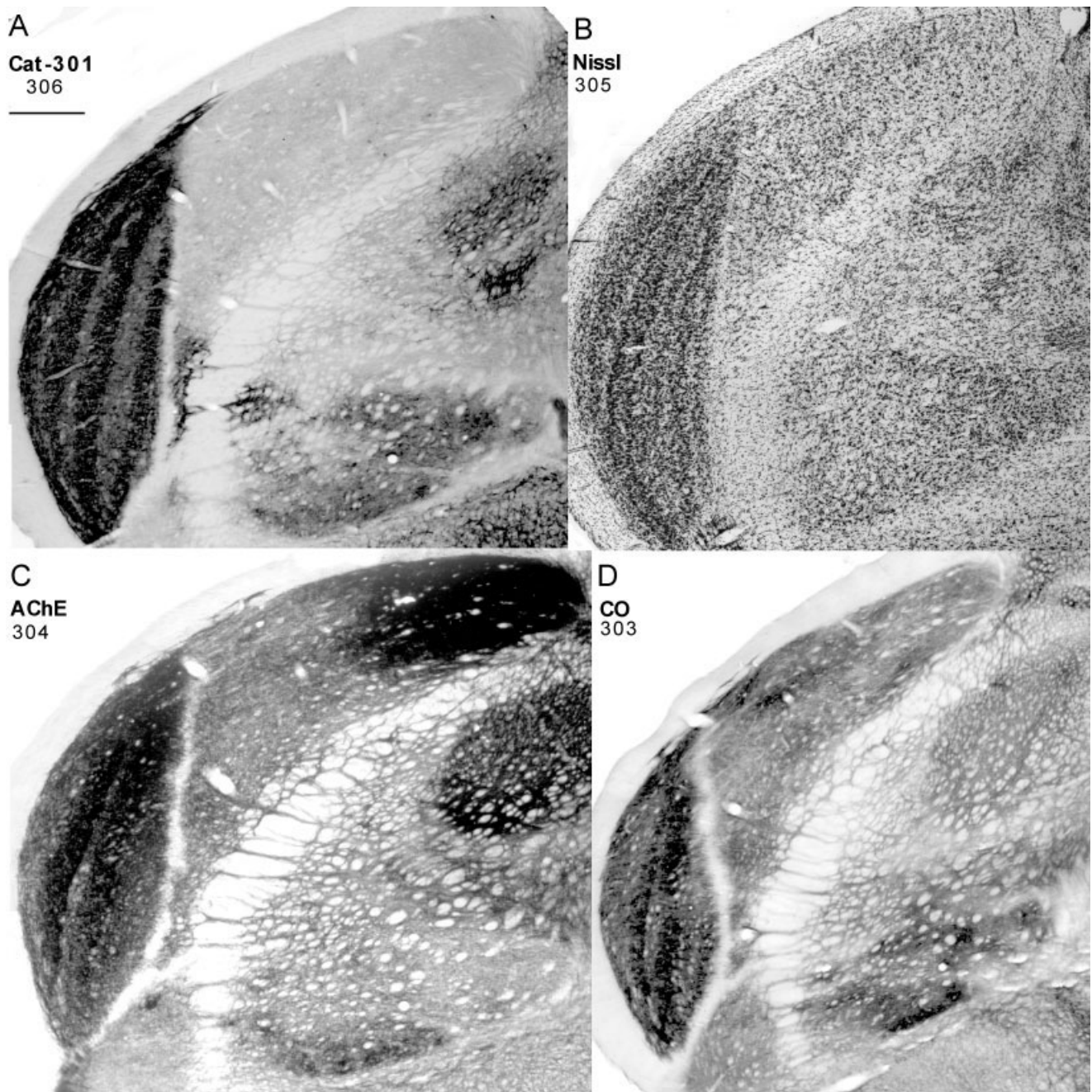


Fig. 2. Four consecutive coronal sections from case 96-3 stained for Cat-301 (A), Nissl (B), AChE (C), and cytochrome oxidase (CO; D). Nuclei are identified in Figure 1. Pv is darkly stained for Cat-301 in A, whereas Pd is darkly stained for AChE in C. Dorsal (d) is toward the top, and lateral (l) is to the left. Scale bar = 500 μ m.

Wong Riley, 1979), myelin (Gallyas, 1979), Nissl (thionin), and parvalbumin (Cusick et al., 1993). All seven stains were not performed for each case, but at least three of the procedures were performed for any given case.

Volumetric measures were made for the case shown in Figures 1, 2 and 3 by measuring the surface area of pulvinar subdivisions in every sixth section. These values were multiplied by the thickness of six sections (240 μ m), and

the total number of values involving the pulvinar were summed. We calculated the surface area of every sixth section because one of the AChE and Cat-301 defined subdivisions was included only once in every series of six sections. Thus, one section was used to represent average surface areas of all six sections.

The images in Figures 2, 3, 4, and 5 were captured by using a Spot 2 camera mounted on a Nikon E800 micro-

A

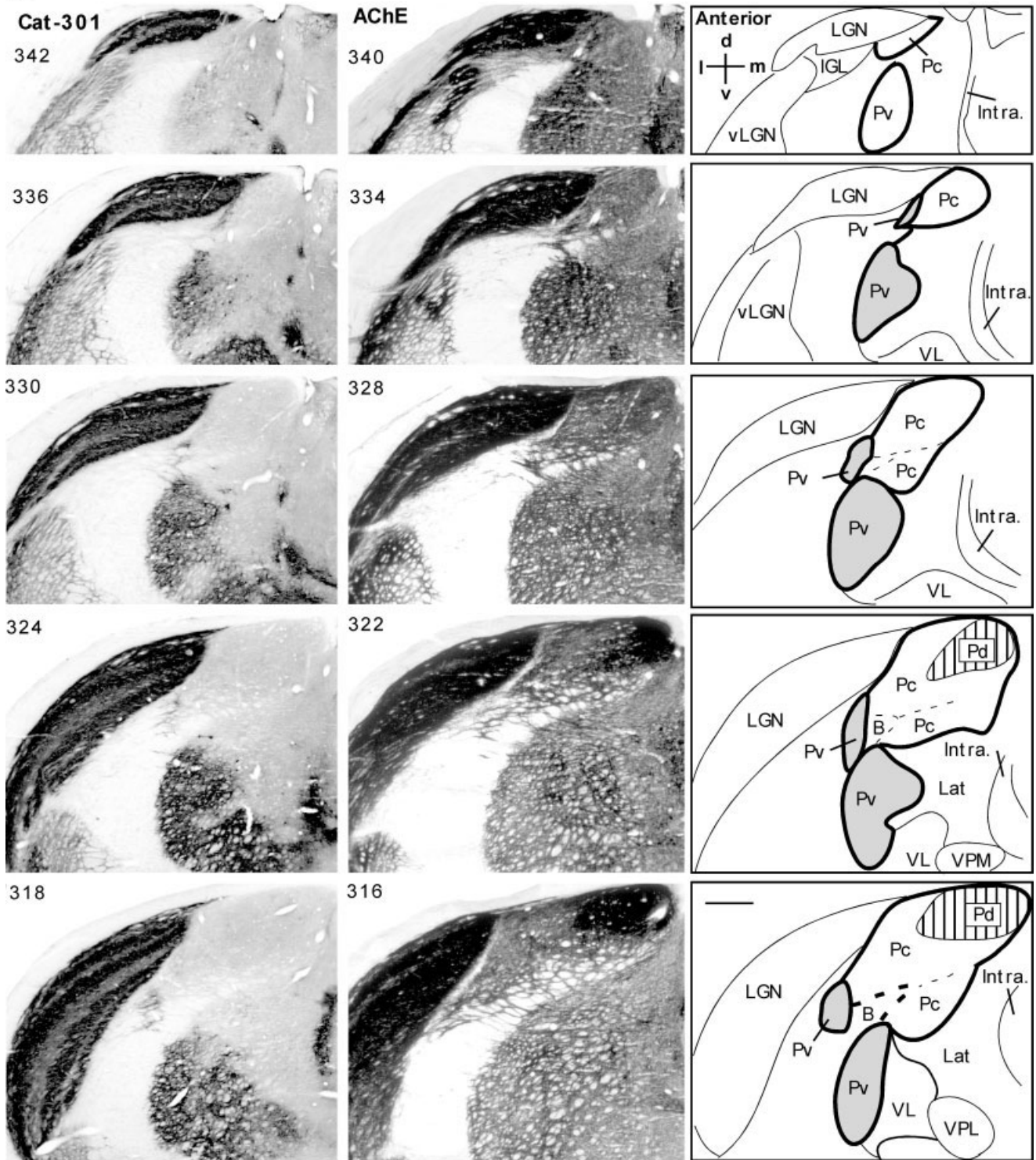


Fig. 3. Two series of coronal sections stained for Cat-301 (column 1) or AChE (column 2). Sections are from case 96-3 also shown in Figure 2 (see sections 306 and 304). Each row shows a series of nearly adjacent sections whose thalamic nuclei are identified in column 3. In all, there are 10 matched sets of sections that progress from anterior (top row in **A**) to posterior (bottom row in **B**). The 40- μ m sections are numbered consecutively from anterior to posterior. In column 3, the

entire pulvinar complex is outlined in bold. Hatching marks the AChE dark Pd subdivision. Gray shading marks the Cat-301 dark Pv subdivision. The Pc subdivision is left unshaded. The dashed line indicates the brachium (B) of the superior colliculus. See Results for more details. Dorsal (d) is toward the top, and lateral (l) is to the left. For abbreviations, see list. Scale bars = 500 μ m.

B

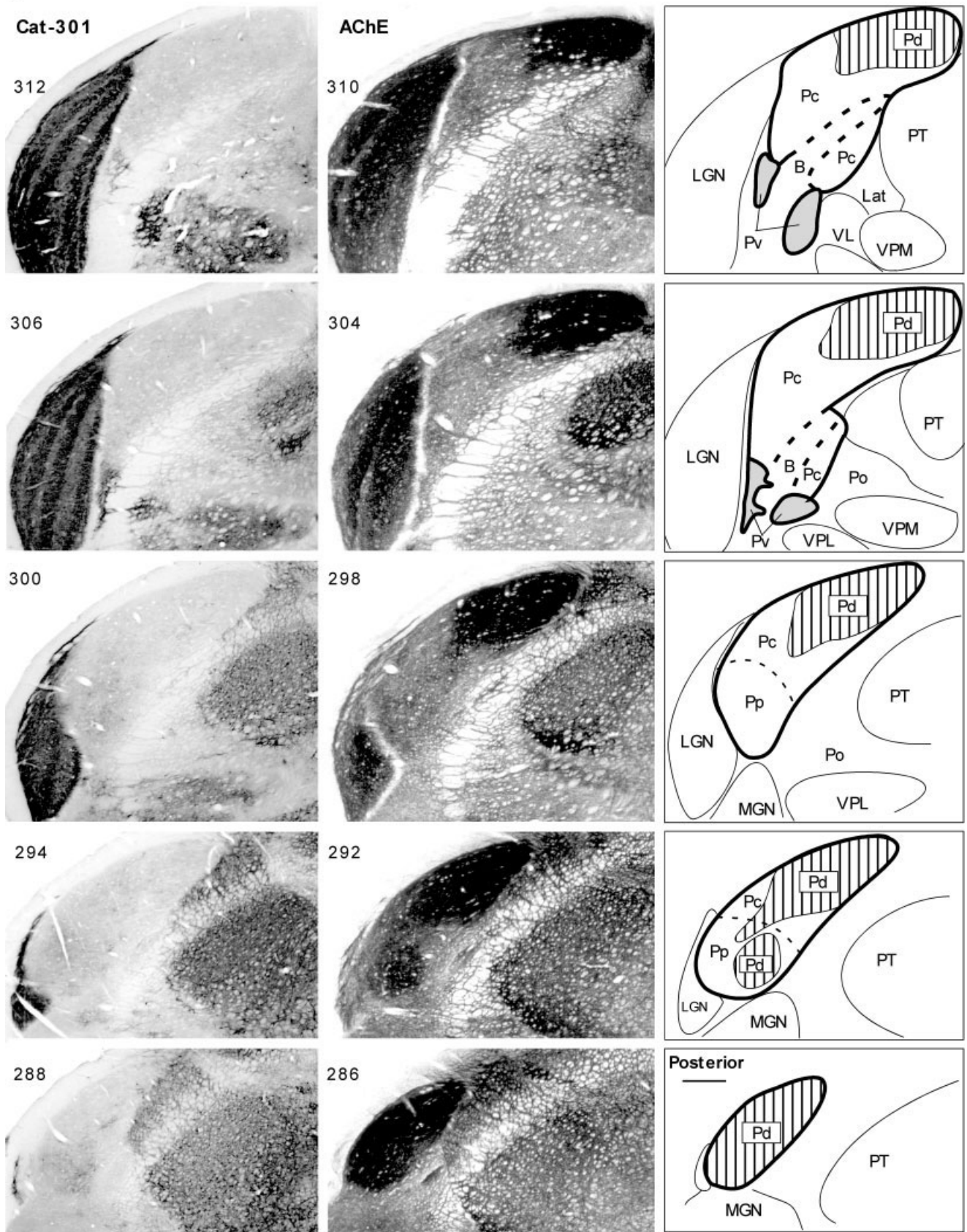


Figure 3 (Continued)

scope, acquired through Adobe Photoshop 6.0 software, and adjusted for brightness and contrast. None of the images were altered in any other way.

RESULTS

Subdivisions of the pulvinar complex of tree shrews were most clearly revealed in brain sections processed for the Cat-301 antigen or AChE. Each procedure revealed distinct regions in the tree shrew pulvinar. From these results, the pulvinar was subdivided into three separate regions: the ventral pulvinar (Pv), which stained dark for the Cat-301 antibody (Figs. 1, 2A), the dorsal pulvinar (Pd), which stained dark for AChE (Figs. 1, 2C), and the central pulvinar (Pc), which stained light for both AChE and the Cat-301 antigen (Figs. 1, 2A,C). fourth subdivision, the posterior pulvinar (Pp; Fig. 3B, rows 3 and 4) was distinguished from Pc by connection patterns (Lyon et al., 2003). In addition, the staining patterns revealed that the pulvinar is much larger than previously described, as it clearly includes regions on the ventromedial side of the brachium of the superior colliculus (Fig. 1). Previously this brachium was used to delineate the medial border of the visual pulvinar.

The architectonic evidence for subdivisions of the tree shrew pulvinar is presented in two sections. The first includes a description of variously stained brain sections cut in the coronal plane, and the second compares the evidence from sections cut in the parasagittal plane. Horizontal sections were included in the initial analysis, but they are not described here because sections cut in this plane added little to the identification of the pulvinar subdivisions.

Coronal plane

The commonly portrayed view of the tree shrew thalamus is in the coronal plane of section (e.g., Tigges and Shantha, 1969; Marrocco et al., 1970). For our purposes, coronal sections were useful in revealing three architectonically defined subdivisions of the tree shrew pulvinar.

Overview. Consecutive thalamic sections from the same case were each processed in one of four ways to show different aspects of thalamic architecture in tree shrews. Traditionally, subdivisions of the visual thalamus have been defined in Nissl-stained sections, as seen in Figure 2B. Along the lateral extent of the Nissl section, the dorsal lateral geniculate nucleus (LGN) is darkly stained and layers are discernable. The LGN is comprised of six layers separated by interlaminar regions (Glickstein, 1967; Harting et al., 1973a), both of which are also easily recognized in sections processed with the Cat-301 antibody (Fig. 2A; Jain et al., 1994) or for CO (Fig. 2D; Wong-Riley and Norton, 1988). As previously reported (Brauer et al., 1981), AChE is more densely expressed in layers 4 and 5, whereas a low level of AChE is apparent in layer 3. Just medial to the LGN, a large teardrop-shaped structure comprises the classically defined pulvinar (Fig. 1). The pulvinar stains uniformly and moderately dark in Nissl (Fig. 2B), CO (Fig. 2D), calbindin, myelin, and parvalbumin (not shown) preparations, but it also has a ventral dark region in Cat-301 sections (Fig. 2A) and a dorsal dark region in AChE sections (Fig. 2C). Thus, the Cat-301 and AChE stains reveal three subdivisions within the classically defined pulvinar. In keeping with each subdivision's location, we term the dorsally located dark AChE region,

the dorsal pulvinar (Pd; hatching; Fig. 1), the ventrally located dark Cat-301 region, the ventral pulvinar (Pv; gray shading; Fig. 1), and the central region that stains light for both AChE and Cat-301, the central pulvinar (Pc; Fig. 1).

An unexpected observation was that part of the dense Cat-301 region in the ventral pulvinar (Fig. 2A; also see Fig. 1) extended ventrally across the brachium of the superior colliculus (dashed lines in Fig. 1), occupying a region outside of the classically defined pulvinar. We conclude that this staining pattern identified the remainder of Pv and that the pulvinar is larger than previously described. Based on the uniformly dark staining in Nissl and CO preparations, as well as the uniformly light staining in Cat-301 and AChE preparations (Fig. 2), the Pc division also crosses the brachium (Fig. 1). The cortical projection patterns shown in the companion paper further support the view of the visual pulvinar extending ventral to the brachium (Lyon et al., 2003).

A more complete series of coronal sections stained for Cat-301 and AChE from the case in Figure 2 is shown in Figure 3. This case (96-3) was especially valuable in that nearly all of the consecutive sections were undamaged during cutting and processing. Nevertheless, the Cat-301 and AChE staining patterns revealed the Pv and Pd subdivisions in all other cases processed (Figs. 4, 5). As the sections progress from anterior (row 1; Fig. 3A) to posterior (row 5; Fig. 3B), the pulvinar increases in size and then becomes slightly reduced (see the third column in Fig. 3 for outlines of the entire extent of the pulvinar drawn in bold).

Cat-301 antibody. At the anterior pole of the pulvinar (row 1; Fig. 3A), a smaller part of the pulvinar is dorsal to the brachium of the superior colliculus and a larger portion is ventral. Neither region stained darkly in either Cat-301 (section 342) or AChE (section 340) sections. The subsequent Cat-301 section (336; row 2; Fig. 3A) is 240 μm posterior (six sections at 40 μm each). At this level the darkly stained Cat-301 cells that identify Pv start to appear. Pv is split in two at this level so that a small sliver of Cat-301-stained cells is visible at the ventral pole of the classically defined tree shrew pulvinar, whereas the larger remainder of Pv is located ventral to the brachium. In previous reports, the portion of Pv ventral to the brachium was identified as the lateral intermediate nucleus (Carey et al., 1979; Luppino et al., 1988). However, the histochemical characteristic of this region identifies it as part of Pv. In the next three more posterior sections (rows 3–5; Fig. 3A), the portion of Pv ventral to the brachium increases somewhat in size before becoming much more reduced in section 312 (row 1; Fig. 3B). At 1,680 μm posterior to the anterior end of the pulvinar, only a small remnant of this part of Pv remains (section 300; row 3; Fig. 3B). In all sections, the portion of Pv dorsal and lateral to the brachium is comparatively smaller than its ventral counterpart (see columns 1 and 3 of rows 2–5 in Fig. 3A and rows 1 and 2 in Fig. 3B).

Other thalamic nuclei also stained darkly for the Cat-301 antibody. Lateral to the pulvinar, the LGN, as noted above, was very darkly stained. More medially, dark patches of Cat-301 staining were found in the intralaminar nuclei (Fig. 3A), a structure that resembles the pulvinar by having connections with visual cortex (Jones, 1985; Lyon et al., 2003). Ventral to Pv, the ventral lateral nucleus (VL; Tigges and Shantha, 1969) also stained

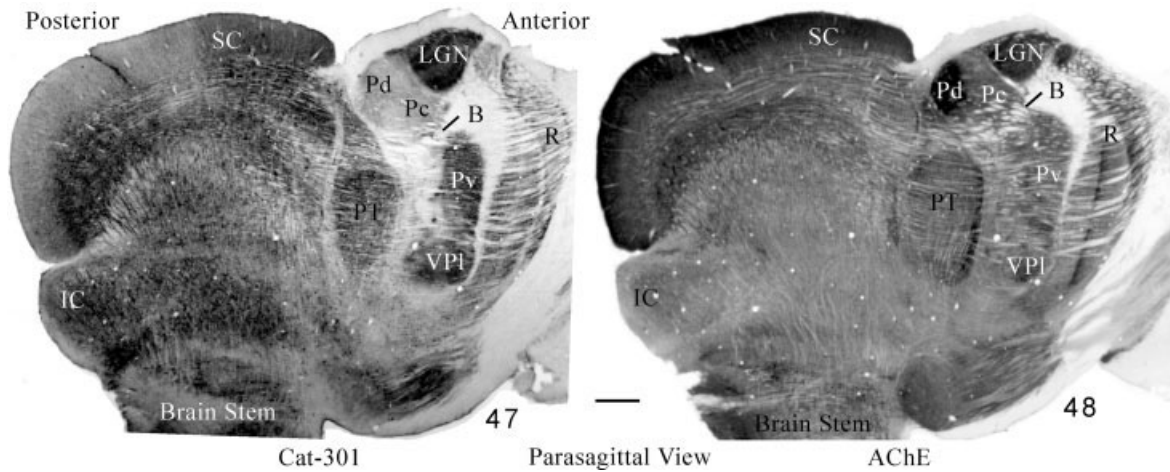


Fig. 4. Two adjacent parasagittal sections from case 99-52 stained for Cat-301 (left) or AChE (right). The images include the reticular nucleus (R) at the extreme anterior end, and the superior (SC) and inferior (IC) colliculi at the posterior end as a reference to view the

series of closely cropped parasagittal sections in Figure 5. For abbreviations see list. Dorsal is toward the top, and anterior is to the right. Scale bar = 500 μ m.

darkly for Cat-301. In some sections, a bridge (section 330) or complete filling-in of darkly stained Cat-301 tissue gave the appearance that VL is fused with Pv (sections 324 and 318; Fig. 3A). However, this bridge of Cat-301 staining between Pv and VL is probably the tree shrew equivalent of the anterior pulvinar because this region has connections with both visual and somatosensory cortex (Lyon et al., 2003). In other sections VL at its anterior and posterior ends is clearly distinguished from Pv (sections 336 in Fig. 3A; section 312 in Fig. 3B). The two somatosensory relay subnuclei, VPL and VPM, both stained darkly for Cat-301 (sections 312, 306; also see Diamond et al., 1993).

Acetylcholinesterase. The AChE sections clearly revealed another subdivision of the pulvinar. A much darker AChE region was located dorsomedially in the pulvinar (column 2; illustrated by hatching in column 3; Figs. 3A,B), whereas the remainder of the pulvinar stained moderately for AChE. This darker dorsal region identifies the dorsal subdivision (Pd) of the pulvinar. This subdivision extends from near the anterior pole (section 322) to the posterior pole (section 286; approximately 1,440 μ m from anterior to posterior). Furthermore, Pd increases in relative size posteriorly, so that the dark AChE region eventually comprises all of the pulvinar (section 286, row 5; Fig. 3B). The AChE dark subdivision (Pd) never overlaps with the Cat-301 dark subdivision (Pv).

Dark AChE staining did not occur in any part of the pulvinar ventral to the brachium of the superior colliculus. The pretectal region stained darkly for AChE, but the pretectum is distinct from the pulvinar. As for the Cat-301 stain the LGN stained darkly for AChE, but the layers were less distinct. Portions of the intralaminar nucleus also stained darkly for AChE.

Cytochrome oxidase. We also examined series of thalamic sections stained for CO (not shown; see next section for parasagittal results for CO-stained sections; Fig. 5A,B). The entire pulvinar stained moderately dark to dark for CO, and subdivisions were not obvious. Most surrounding nuclei stained moderately dark to dark for CO as well. However, CO stains are useful for visualizing

lamina of the LGN (Fig. 2D; Wong-Riley and Norton, 1988). The uniform staining of CO in the pulvinar included the part ventral to the brachium of the superior colliculus (Fig. 1D), thus providing further evidence that a significant portion of the central subdivision of the pulvinar (Pc) is located ventral to the brachium.

Calbindin and parvalbumin. The distributions of cells and neuropil labeled by antibodies for calbindin or parvalbumin in the posterior visual thalamus of tree shrews have been briefly described by Diamond et al. (1993). In agreement with this previous description, the pulvinar complex was lightly to moderately stained with both protocols, but subdivisions were not obvious. The LGN and ventroposterior nucleus stained darkly for calbindin. Layers 3 and 6 of the LGN stained moderately dark for calbindin.

Parasagittal plane

The parasagittal plane of tree shrew thalamus has been used in previous anatomical studies (Oliver and Hall, 1978), but this plane has been portrayed less often than the coronal plane. The low-magnification images of mid-brain sections in Figure 4 provide a frame of reference for interpreting the higher magnification images of thalamic nuclei in parasagittal sections shown in Figure 5. The dorsal thalamus lies just anterior to the large superior colliculus and pretectum (PT). The ventral subdivision of the pulvinar complex lies anterior to PT. The dorsal (Pd) and central (Pc) subdivisions of the pulvinar lie just dorsal and slightly anterior to PT, and posterior and ventral to the LGN.

Subdivisions of the pulvinar were easily distinguished in both Cat-301 and AChE in parasagittal brain sections (Fig. 4). In sections about midway through the lateromedial dimension of the pulvinar (see row 1 in Fig. 5B), the dark AChE region, Pd, is dorsal and posterior to the rest of the pulvinar. At this location, Pd is rather small. The dark Cat-301 subdivision, Pv, is much larger at this level than Pd. Pv is primarily anterior and ventral to the brachium of the superior colliculus. However, a very tiny portion of Pv

A

Parasagittal from lateral to medial
CO Cat-301

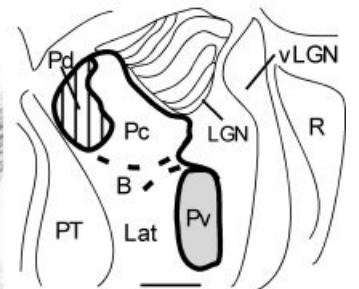
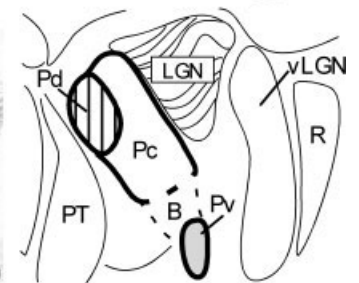
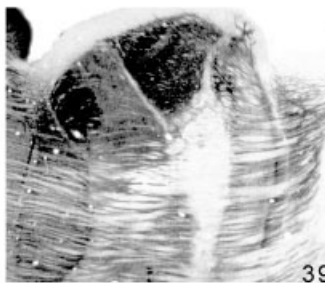
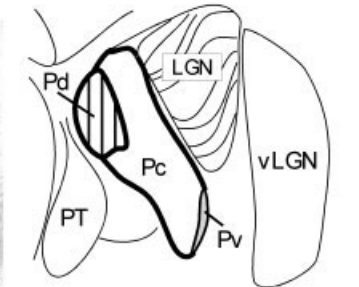
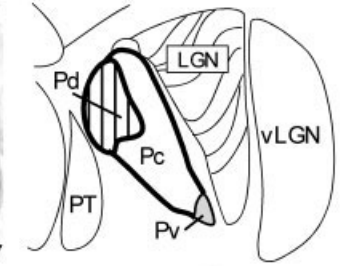
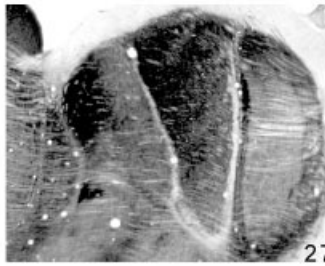
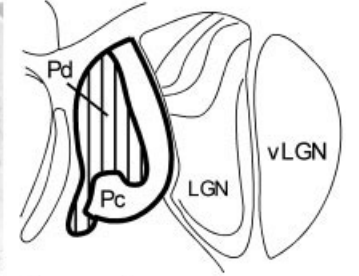
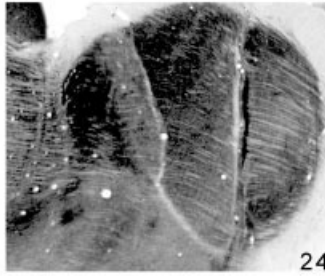
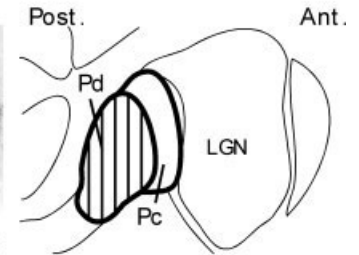
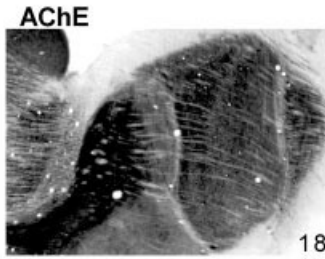
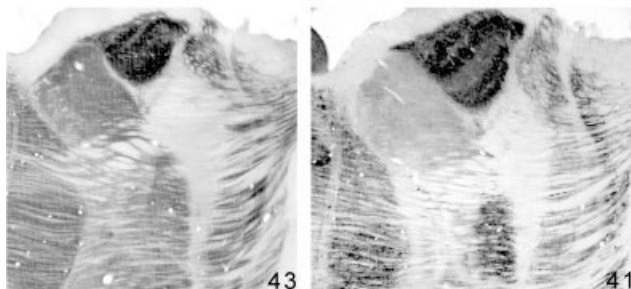
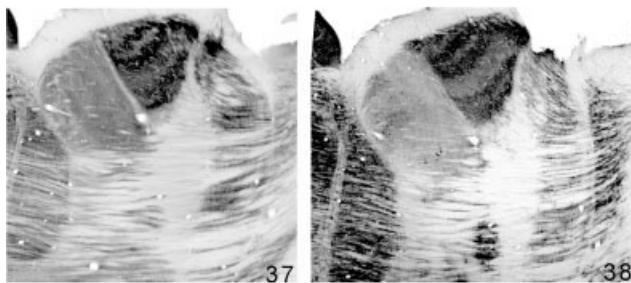
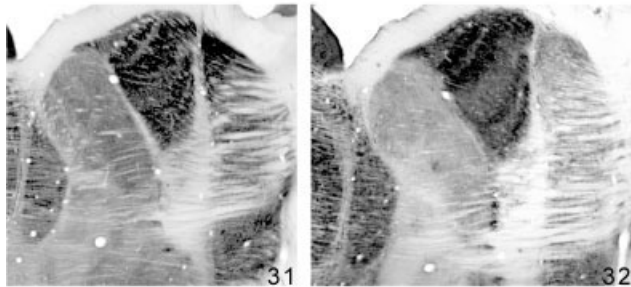
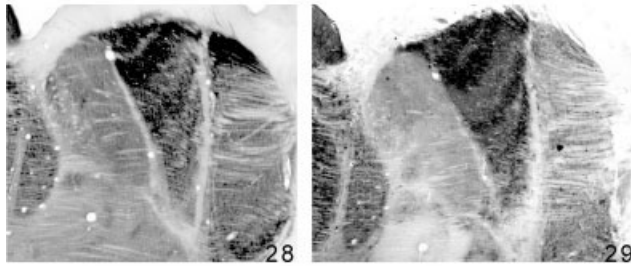
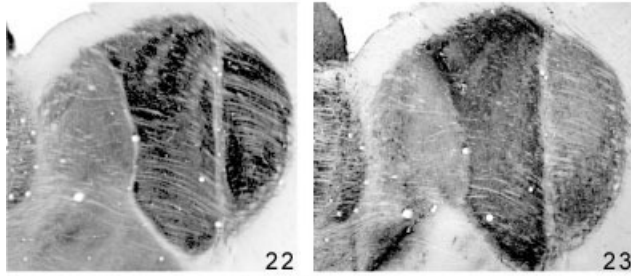
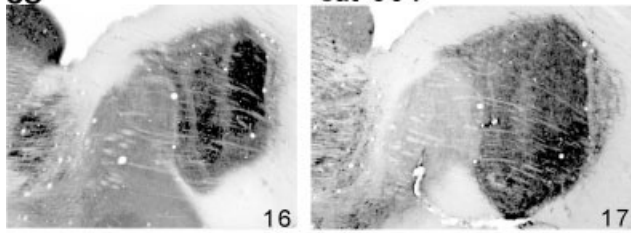
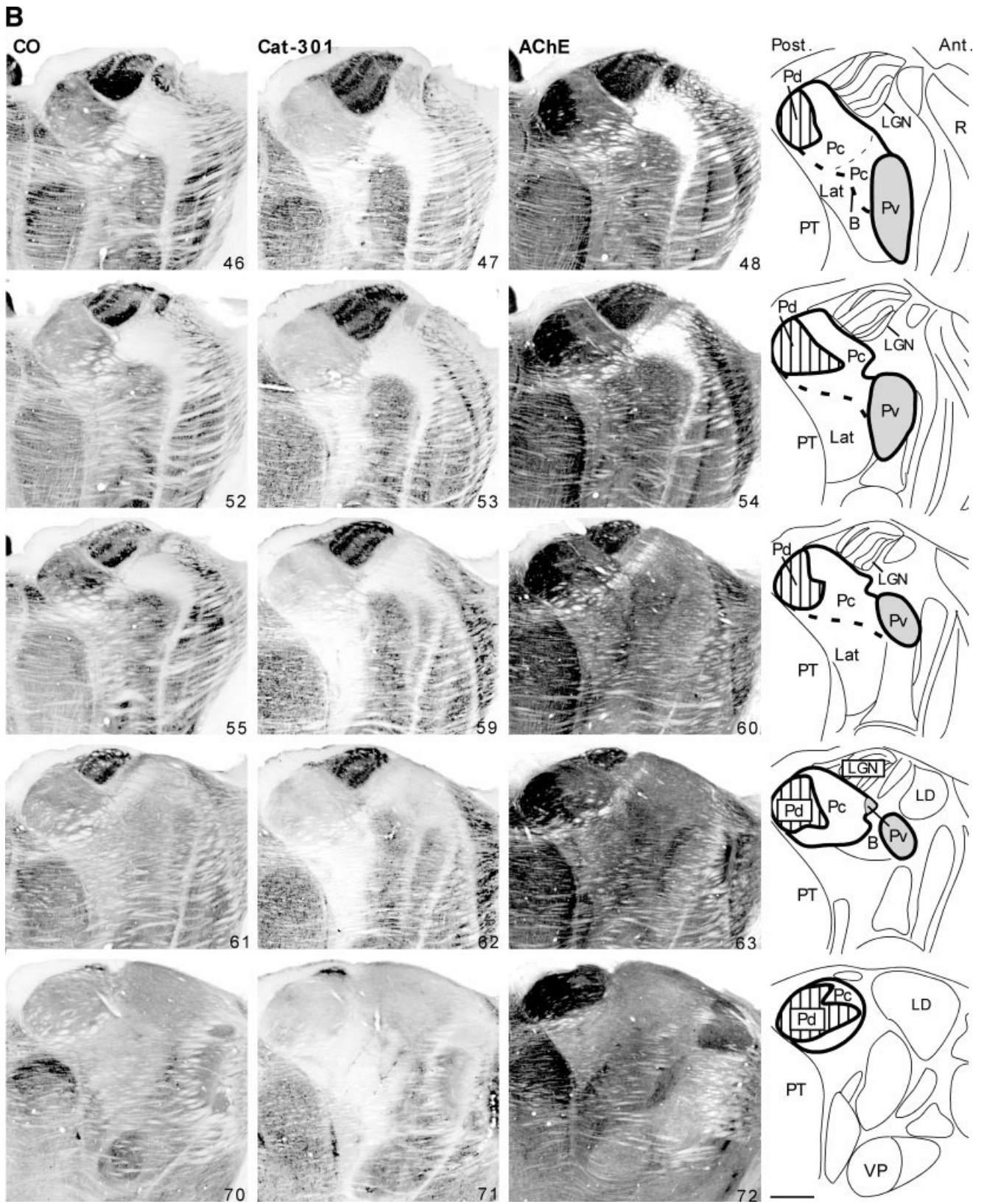


Fig. 5. Three series of parasagittal sections stained for CO (column 1), Cat-301 (column 2), or AChE (column 3). Sections are from the same case (99-52) shown in Figure 4 (see sections 47 and 48). Each row contains a progression of adjacent sections whose thalamic nuclei are identified in column 4. In all there are 11 rows of 40- μ m sections that are consecutively numbered from lateral to medial. In column 4,

the entire pulvinar complex is outlined in bold. Hatching marks the AChE dark Pd subdivision. Gray shading marks the Cat-301 dark Pv subdivision. The Pc subdivision is left unshaded. The dashed line outlines the brachium (B) of the superior colliculus. See Results for more details. Dorsal (d) is toward the top, and anterior is to the right. For abbreviations, see list. Scale bars = 500 μ m.



is dorsal to the brachium, but ventral to the main body of the pulvinar, Pc.

Other nuclei are visible in Cat-301 and AChE preparations as well. Among these, the reticular nucleus is located at the most anterior end of the section with the tectum at the posterior end.

Cat-301 antibody. The darkly stained portion of the pulvinar with the Cat-301 antibody is first noticeable near the lateral edge of the pulvinar (Fig. 5A, section 29). This marker for the Pv subdivision is found both anterior and ventral to the AChE-defined, Pd subdivision (row 3). At this lateral level only the upper portion of the pulvinar (dorsal to the brachium of the superior colliculus) is present. Dark Cat-301 antibody staining in Pv is present on this upper portion of the pulvinar for the next three sections in the series (32, 38, and 41), spanning a total of 480 μm , and it reappears briefly 740 μm medial in section 62. As in the coronal sections, this upper portion of Pv is much smaller and spans less of the mediolateral distance than the larger ventral portion of Pv.

The larger, ventral portion of Pv is first present 360 μm medial (section 38; row 5; column 2; Fig. 5A) to the initial appearance of the dorsal Pv portion. The ventral Pv subdivision increases in size medially for another 600 μm (until section 53; row 2; column 2; Fig. 5B). The ventral part of the Pv subdivision then reduces in size and, about 400 μm medially, is no longer present. In total, the ventral part of the Pv subdivision spans at least 1,000 μm .

Acetylcholinesterase. The hatching in column 4 of Figure 5A and B marks the dorsal subdivision of the pulvinar, Pd, as defined by dark AChE staining (column 3). This dark AChE subdivision extends from the most lateral to the most medial sections (18 and 72, respectively), spanning at least 2,160 μm . In the two most lateral sections (18 and 24), the AChE-dark region is posterior and occupies most of the pulvinar. In the middle sections (33–48), the AChE-dark region occupies only a relatively small portion of the dorsoposterior pulvinar (see illustrations in column 4; rows 4–6 in Fig. 5A; and row 1 in Fig. 5B). Dark AChE staining is noticeably absent in the portion of the pulvinar located ventral to the brachium of the superior colliculus. In the medialmost section shown (72; row 5 in Fig. 5B) the dark AChE staining has extended to the anterior portion of the pulvinar to occupy the entire nucleus.

Cytochrome oxidase. CO preparations are commonly used to identify thalamic nuclei, and they did distinguish such structures as the LGN, but they were not very useful in subdividing the pulvinar (Fig. 5A,B; column 1). Nevertheless, in several CO sections (28–43; rows 3–6; Fig. 5A) a distinct lighter stained region was identified. This region coincides with the AChE-dark (column 3) subdivision, Pd (see illustrations in column 4). For the most part, however, the CO staining was moderate and uniform. This uniform staining pattern was useful in revealing that the central and ventral nuclei of the pulvinar cross the brachium (as noted for coronal CO sections). Sections 37–55 (column 1; Fig. 5A,B) clearly show that CO staining is similar on either side of the brachium. In sections 37 and 43 (rows 5 and 6; Fig. 5A) the CO region ventral to the brachium coincides with the dark Cat-301 (column 2) subdivision, Pv (see illustrations in column 4).

Size of pulvinar and each subdivision

Volumetric measures were calculated for the pulvinar and each of its three architectonic subdivisions, Pd, Pc, and Pv. For comparison, the volume of the dorsal LGN was calculated as well. In total, the tree shrew pulvinar occupies about 4.4 mm^3 of the thalamus. The LGN, on the other hand, fills about 2.7 mm^3 , around 61% of the pulvinar volume. Even though these measurements do not account for cell density within either nucleus, this comparatively large size of the pulvinar illustrates its importance in the tree shrew visual system.

Two pseudo 3-dimensional reconstructions of the pulvinar are illustrated in Figure 6. The frontal (Fig. 6B,C) and lateral (Fig. 6D,E) views illustrate the height and depth of the pulvinar, as well as the relative sizes and locations of each of the three architectonically defined subdivisions. Although the dimensions of the pulvinar and its subdivisions in Figure 6 were calculated from coronal series of sections (Fig. 3; see Fig. 6A), they are consistent with the results obtained from the parasagittal series (Fig. 5).

Pc. The central subdivision, Pc (orange), which stains lightly for Cat-301 and moderately dark for both AChE and CO, makes up nearly half of the pulvinar (49%; 2.1 mm^3). As illustrated in the frontal view (Fig. 6B,C), the Pc subdivision is relatively small at the anterior end (front) and gradually enlarges in a posterior direction. The lateral view (Fig. 6D,E) shows that at the posterior pole (back) of the pulvinar Pc is again relatively small.

Pd. The dorsal subdivision, Pd (gold), is defined by dark AChE staining. Pd comprises a third of the volume (33%; 1.4 mm^3) of the entire visual pulvinar, making it the second largest subdivision. The frontal view shows that Pd is partially encapsulated within the Pc subdivision, both lateral and ventral to Pd. Furthermore, as portrayed in the lateral view (Fig. 6D,E), the Pd subdivision starts slightly posterior to the beginning of the pulvinar (dominated by Pc) where it is relatively small. More posteriorly, Pd increases in size to occupy from half to nearly the entire pulvinar.

Pv. The ventral subdivision, Pv (blue), defined by dark Cat-301 staining, is located at the ventralmost portion of the pulvinar. The Pv subdivision is the smallest of the three, occupying just less than 20% (0.8 mm^3) of the pulvinar. The frontal view (Fig. 6B,C) shows that the portion of Pv ventral and medial to the brachium of the superior colliculus (this portion shown as transparent in Fig. 6B) is situated more anteriorly than the smaller dorsolateral portion of Pv that is part of the classically defined portion of the visual pulvinar. The lateral view (Fig. 6D,E) shows that Pv is found ventrally along the anterior two-thirds of the pulvinar but is absent in the posterior third.

DISCUSSION

The present results lead to a new interpretation of the pulvinar in tree shrews. Although the pulvinar appeared rather homogenous in previous descriptions based on Nissl-stained sections, stains for AChE and the Cat-301 revealed three architectonically distinct subdivisions of the tree shrew pulvinar (summarized in Fig. 6). Contrary to earlier descriptions, the results also indicate that two subdivisions of the visual pulvinar complex, Pc and Pv, extend ventromedially to cross the

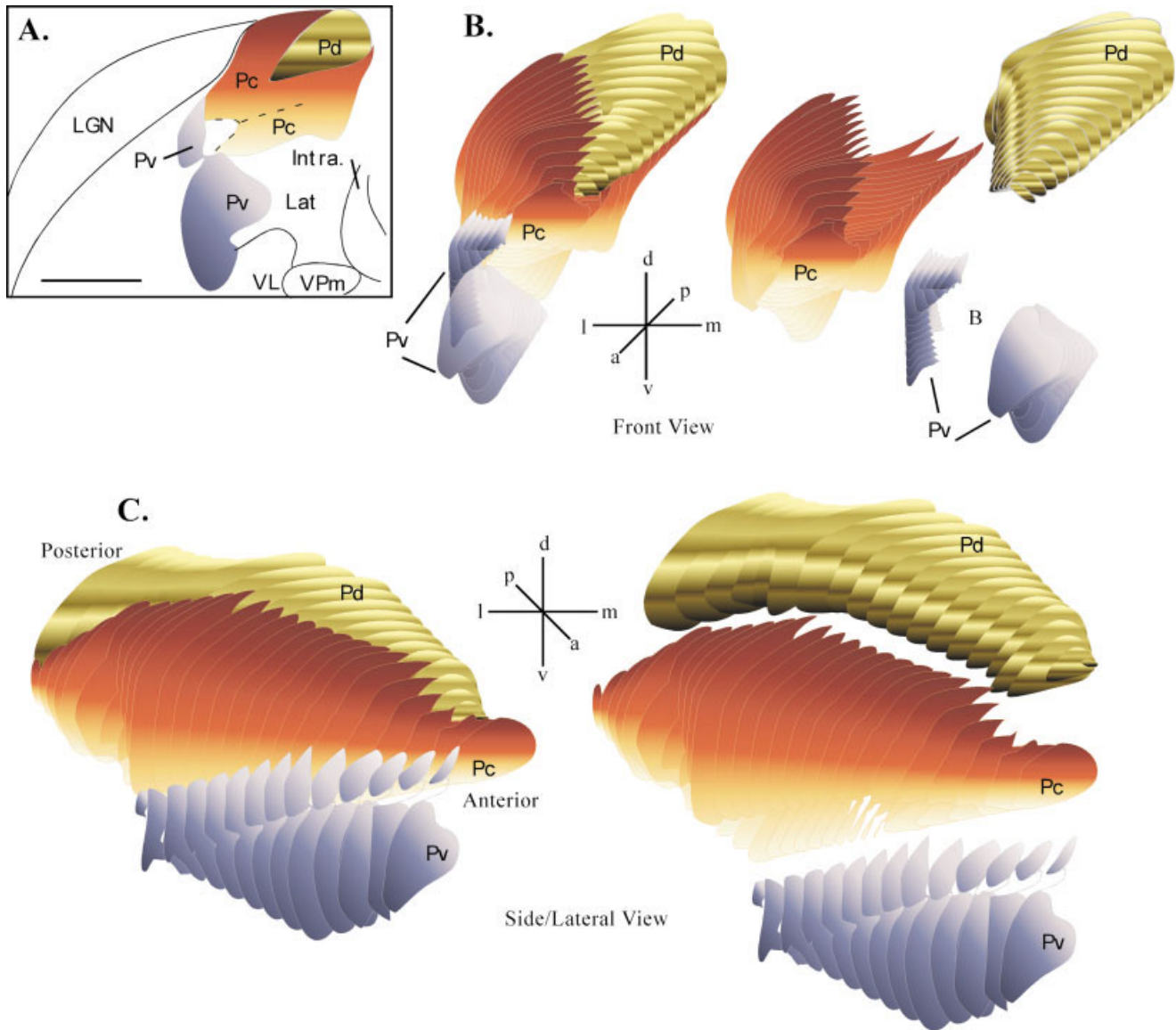


Fig. 6. Pseudo-3-dimensional reconstructions of the pulvinar and its architectonic subdivisions shown in frontal (B,C) and lateral views (D,E). Gold represents the AChE dark Pd subdivision, which comprises 33% of the total pulvinar. Orange represents the CO and AChE moderate Pc subdivision, which comprises 49% of the pulvinar. Blue represents the Cat-301 dark Pv subdivision, which comprises 18% of the pulvinar. See Results for more details. To create these figures coronal sections of the pulvinar (A) were aligned within a rectangular

cube 2,160 μm deep using Canvas 7 software (Deneba, Miami, FL). The 2,160 μm -length was used because the illustrations (column 3) of the final 9 of the 10 sections shown in Figure 3 were used in the reconstruction. Each section in the reconstruction represents six 40- μm sections for a total thickness of 240 μm (see Materials and Methods section on volume calculations). Thus, nine sections in the reconstruction would span 2,160 μm ($9 \times 6 \times 40$). For abbreviations, see list. Scale bar = 500 μm .

brachium of the superior colliculus. Thus, the pulvinar is more subdivided and larger than previously thought. The dorsal subdivision, Pd, stains darkly for AChE, is found dorsomedial and caudal, and occupies 33% of the pulvinar nucleus. The ventral subdivision, Pv, stains darkly for the Cat-301 antibody and is located ventral and anterior, and on either side of the brachium of the superior colliculus. This subdivision is the smallest of the three, occupying just less than 20% of the entire pulvinar nucleus. The remainder of the pulvinar makes up nearly 50% of the nucleus. It stains moderately dark

for AChE and CO and very light for Cat-301. This region, termed the central subdivision, Pc, is located centrally throughout the pulvinar, and is also found on either side of the brachium of the superior colliculus. A fourth subdivision was distinguished in the posterior-most portion of Pc (see illustrations in rows 3 and 4 of Fig. 3B) by its unique connection pattern with extrastriate cortex (Lyon et al., 2003). This region comprises only a small portion of the pulvinar, 6%, by occupying about 12% of the Pc subdivision. Because this subdivision was not identified architectonically, it is discussed in the

companion paper exploring the thalamocortical connections in tree shrews.

Here we relate previous descriptions of the tree shrew pulvinar to our newly discovered architectonic subdivisions and compare these subdivisions with those described for primates and cats. In addition we discuss the possible significance of the localized expression of the Cat-301 antigen and AChE in the pulvinar.

Previous descriptions of the tree shrew pulvinar

The conspicuous size and cytoarchitectonic distinctiveness of the pulvinar in tree shrews was noted long ago by Le Gros Clark (e.g., Le Gros Clark, 1929), who argued that similarities in the visual thalamus and cortex of tree shrews and other primates were compelling enough to place tree shrews in the primate order. The tree shrew is no longer considered a primate but is now classified as a close relative, due to several anatomical similarities. One similarity is the rather large and conspicuous size of each species visual pulvinar complex. However, in Nissl preparations, the pulvinar of tree shrews is more uniform in appearance than in primates, leading early investigators to consider it as single structure (Snyder and Diamond, 1968; Tigges and Shantha, 1969; Abplanalp, 1970; Diamond et al., 1970; Marocco et al., 1970; Harting et al., 1973a). Consistent with this view, it was noted that projections of the superior colliculus appeared to fill the entire pulvinar (Harting et al., 1973b). In addition, the boundaries of the pulvinar seemed obvious, as the border with the LGN was distinct, the pulvinar was the most dorsal nucleus in the caudal thalamus, and the complete ventromedial border was thought to correspond to a band of fibers, the brachium of the superior colliculus (e.g., Snyder and Diamond, 1968).

The most significant revision of the theory that the pulvinar of tree shrews is a single structure came from a more detailed study of the distributions of inputs from the superior colliculus (Luppino et al., 1988). These investigators distinguished a dorsomedial portion of the pulvinar with less dense inputs from the superior colliculus that were diffusely distributed without a topographic pattern. The location of this subdivision closely corresponds with Pd of the present report. Most or all of the rest of the pulvinar received a denser, patchy input from the superior colliculus that varied in location according to the location of the injection site in the retinotopic map in the superior colliculus. Injections in the medial superior colliculus, representing the upper visual quadrant, labeled projections to more rostral portions of the pulvinar than injections in the lateral superior colliculus representing the lower quadrant. In addition, the projections of the pulvinar to visual cortex were no longer seen as forming a single topographic pattern. Instead, Luppino and colleagues (1988) reported that injections in visual cortex of the lower temporal lobe labeled neurons in the dorsomedial portion of the pulvinar that has diffuse inputs from the superior colliculus, whereas injections in area 18 (V2) and the adjoining temporal dorsal area (TD) labeled locations in the larger part of the pulvinar (Lyon et al., 2003 for further discussion) that has retinotopic inputs from the superior colliculus. In agreement with the retinotopy of the ventrolateral pulvinar as suggested by superior colliculus inputs, injections in parts of V2 or TD representing the lower visual quadrants labeled more caudal zones in

the pulvinar than injections in the upper quadrant representations of V2 and TD.

In the present proposal, a fiber band separates the bulk of the pulvinar complex from more medial portions (Fig. 1). In contrast, previous depictions had the fiber band separating the pulvinar from a more ventromedial thalamic region identified as the lateral intermediate nucleus (e.g., Diamond et al., 1979). There is previously reported evidence that the pulvinar extends medially across this fiber band. First, there was no notable difference in the appearance of the Nissl-stained tissue across this band. In addition, tissue on both sides of the band receives inputs from the superior colliculus (Luppino et al., 1988) and projects to visual cortex (Carey et al., 1979; Luppino et al., 1988). More specifically, a portion of the lateral intermediate region appears to have the "specific" type of superior colliculus inputs, as well as cortical connections with V2 and TD. We elaborate on this pattern of cortical connections in our companion paper (Lyon et al., 2003). Here we note that the evidence from these previously reported connections is consistent with our proposal that the Pd nucleus does not cross the fiber band, whereas the Pc nucleus does (Fig. 1), as Pc would be the nucleus with "specific" superior colliculus inputs, and projections to V2 and TD.

In summary, a dorsal division (Pd) of the pulvinar in tree shrews that is darkly stained for AChE appears to be the same division that receives a diffuse pattern of direct retinal input, as well as a diffuse pattern of superior collicular inputs. This division projects to visual areas of the ventral temporal lobe. In contrast, the rest of the pulvinar is devoid of retinal inputs, and much of the region receives a topographic pattern of superior collicular inputs and projects topographically to V2 and TD. The connection patterns indicate that the lower visual quadrant is represented caudal to the upper visual quadrant in this portion of the pulvinar. Earlier studies did not distinguish between the central pulvinar (Pc) and the ventral pulvinar (Pv), and they did not recognize that both of these subdivisions extend ventromedially across the fiber band containing corticotectal axons.

Comparison of pulvinar nuclei in tree shrews and other mammals

Le Gros Clark (1930) was one of the first to recognize that the pulvinar of the thalamus is not a structure unique to primates but is part of the visual thalamus of all mammals, although the region was usually termed the lateral posterior nucleus. A problem for Le Gros Clark was that the pulvinar was not yet very well defined in primates, and the lateral posterior nucleus in other mammals was also poorly delimited. The situation has improved as recent studies have denoted histochemical subdivisions and connections of the pulvinar complex in primates (e.g., Cusick et al., 1993; Gutierrez and Cusick, 1997; Beck and Kaas, 1998; Gray et al., 1999; Stepniowska et al., 1999, 2000; Adams et al., 2000; Gutierrez et al., 2000; Soares et al., 2001; Weller et al., 2002) and cats (Patel and Bickford, 1997; Patel et al., 1999; Fitzgibbon et al., 1999), but there is still little understanding of the organization of the pulvinar (lateral posterior nucleus) of other mammals, such as rodents (however, see Robson and Hall, 1977). In addition, as the numbers of proposed subdivisions of the pulvinar increase in different taxa, the problem of relating these subdivisions to each other becomes greater. Nevertheless,

an attempt to do so is found in our companion paper (Lyon et al., 2003) where we consider connection patterns in addition to architecture.

AChE and Cat-301 preparations reveal subdivisions of the pulvinar in other mammals studied. In cats, the visual pulvinar has been divided into a "pulvinar" and two divisions of the lateral posterior nucleus. The "pulvinar" division and the medial division of the lateral posterior nucleus stain darkly for AChE (Graybiel and Berson, 1980; Patel and Bickford, 1997; Patel et al., 1999), as does Pd in tree shrews. In monkeys, a dorsal division of the lateral pulvinar (PLd) stains darkly for AChE (Gray et al., 1999). In addition, a subdivision of the inferior pulvinar of monkeys, the medial subdivision (PI_m), stains darkly with the Cat-301 antibody (Stepniewska and Kaas, 1997; Gray et al., 1999), much like Pv in tree shrews.

The significance of Cat-301 and AChE in tree shrew pulvinar

In the present report, we distinguish a ventral subdivision of the pulvinar (Pv) of tree shrews, by its high level of the antigen for Cat-301, and a dorsal subdivision (Pd), by its high level of AChE. In the visual system of cats and monkeys, Cat-301 selectively labels large, rapidly conducting neurons in the LGN and to a lesser extent in their target neurons in cortex (Hendry et al., 1984; De Yoe et al., 1990; Hockfield and Sur, 1990). A similar labeling of the large cell pathways occurs in the LGN of tree shrews (Jain et al., 1994). Thus, Cat-301 appears not only to be a useful marker of subdivisions of the brain, but also one that may identify neuronal populations with large rapidly conducting axons. However, this possibility needs to be evaluated for Pv of tree shrews, in which neurons do not appear to be notably different from those in the rest of the pulvinar in Nissl preparations.

AChE inactivates the neurotransmitter acetylcholine (ACh) and thus can be considered to vary with and be a marker for ACh. Various structures in the brain express high levels of AChE, sometimes over a period of development and sometimes throughout adult life. Major roles for ACh in the central nervous system are to modulate neural circuits and to promote synaptic plasticity (Dani, 2002). AChE is highly expressed in sensory nuclei and sensory areas in cortex, especially during stages of development when synaptic remodeling is occurring (e.g., Robertson et al., 1991). However, during adulthood high levels are maintained in some areas of the brain, such as the primary auditory cortex (e.g., Hackett et al., 2001), suggesting a maintained role for plasticity in such regions. The high levels of AChE in the Pd subdivision of the pulvinar in adult tree shrews suggest that the intrinsic circuits of this nucleus remain plastic throughout life.

ACKNOWLEDGMENTS

We thank Iwona Stepniewska for helpful comments on this article and Susan Hockfield for generously providing the Cat-301 antibody. We also thank Laura Trice and Mary Varghese for their assistance with histological procedures.

LITERATURE CITED

Abplanalp P. 1970. Some subcortical connections of the visual system in tree shrews and squirrels. *Brain Behav Evol* 3:155–168.

- Adams MM, Hof PR, Gattass R, Webster MJ, Ungerleider LG. 2000. Visual cortical projections and chemoarchitecture of macaque monkey pulvinar. *J Comp Neurol* 419:377–393.
- Albano JE, Norton TT, Hall WC. 1979. Laminar origin of projections from the superficial layers of the superior colliculus in the tree shrew, *Tupaia glis*. *Brain Res* 173:1–11.
- Beck PD, Kaas JH. 1998. Thalamic connections of the dorsomedial visual area in primates. *J Comp Neurol* 396:381–398.
- Bosking WH, Zhang Y, Schofield B, Fitzpatrick D. 1997. Orientation selectivity and the arrangement of horizontal connections in tree shrew striate cortex. *J Neurosci* 17:2112–2127.
- Brauer K, Werner L, Winkelmann E, Hans-Joachim L. 1981. The dorsal lateral geniculate nucleus of *Tupaia glis*: a Golgi, Nissl and acetylcholinesterase study. *J Hirnforsch* 22:59–74.
- Carey RG, Fitzpatrick D, Diamond IT. 1979. Layer 1 of Striate cortex of *Tupaia glis* and *Galago senegalensis*: projections from thalamus and claustrum revealed by retrograde transport of horseradish peroxidase. *J Comp Neurol* 186:393–438.
- Casagrande VA, Harting JK. 1975. Transneuronal transport of tritiated fucose and proline in the visual pathways of tree shrew *Tupaia glis*. *Brain Res* 96:367–372.
- Casagrande VA, Harting JK, Hall WC, Diamond IT, Martin GF. 1972. Superior colliculus of the tree shrew (*Tupaia glis*): evidence for a structural and functional subdivision into superficial and deep layers. *Science* 177:444–447.
- Celio MR. 1990. Calbindin D-28k and parvalbumin in the rat nervous system. *Neuroscience* 35:375–475.
- Conley M, Fitzpatrick D, Diamond IT. 1984. The laminar organization of the lateral geniculate body and the striate cortex in the tree shrew (*Tupaia glis*). *J Neurosci* 4:171–197.
- Cusick CG, Scriptor JL, Darensbourg JG, Weber JT. 1993. Chemoarchitectonic subdivisions of the pulvinar in monkeys and their connective relations with the middle temporal and rostral dorsolateral visual areas, MT and DLr. *J Comp Neurol* 336:1–30.
- Dani JA. 2001. Overview of nicotinic receptors and their roles in the central nervous system. *Biol Psychiatry* 49:166–174.
- De Yoe EA, Hockfield S, Garren H, Van Essen DC. 1990. Antibody labeling of functional subdivisions in visual cortex: Cat-301 immunoreactivity in striate and extrastriate cortex of the macaque monkey. *Vis Neurosci* 5:67–81.
- Diamond IT, Snyder M, Killackey H, Jane J, Hall WC. 1970. Thalamocortical projections in the tree shrew (*Tupaia glis*). *J Comp Neurol* 139:273–306.
- Diamond IT, Fitzpatrick D, Schmechel D. 1993. Calcium binding proteins distinguish large and small cells of the ventral posterior and lateral geniculate nuclei of the prosimian galago and the tree shrew (*Tupaia belangeri*). *Proc Natl Acad Sci USA* 90:1425–1429.
- Fitzgibbon T, Bittar RG, Dreher B. 1999. Projections from striate and extrastriate visual cortices of the cat to the reticular thalamic nucleus. *J Comp Neurol* 410:467–488.
- Fitzpatrick D. 1996. The functional organization of local circuits in visual cortex: insights from the study of tree shrew striate cortex. *Cereb Cortex*. 6:329–341.
- Gallyas F. 1979. Silver staining for myelin by means of physical development. *Neurosci Res* 1:203–209.
- Glickstein M. 1967. Laminar structure of the dorsal lateral geniculate nucleus in the tree shrew (*Tupaia glis*). *J Comp Neurol* 131:93–102.
- Gray D, Gutierrez C, Cusick CG. 1999. Neurochemical organization of inferior pulvinar complex in squirrel monkeys and macaques revealed by acetylcholinesterase histochemistry, calbindin and Cat-301 immunostaining, and wisteria floribunda agglutinin binding. *J Comp Neurol* 409:452–468.
- Graybiel AM, Berson DM. 1980. Histochemical identification and afferent connections of subdivisions in the lateralis posterior-pulvinar complex and related thalamic nuclei in the cat. *Neuroscience* 5:1175–238.
- Grieve KL, Acuna C, Cudiero J. 2000. The primate pulvinar nuclei: vision and action. *Trends Neurosci* 23:35–39.
- Gutierrez C, Cusick CG. 1997. Area V1 in macaque monkeys projects to multiple histochemically defined subdivisions of the inferior pulvinar complex. *Brain Res* 765:349–356.
- Gutierrez C, Yaun A, Cusick CG. 1995. Neurochemical subdivisions of the inferior pulvinar in macaque monkeys. *J Comp Neurol* 363:545–562.
- Gutierrez C, Cola MG, Seltzer B, Cusick CG. 2000. Neurochemical and connective organization of the dorsal pulvinar complex in monkeys. *J Comp Neurol* 419:61–86.

- Hackett TA, Preuss TM, Kaas JH. 2001. Architectonic identification of the core region in auditory cortex of macaques, chimpanzees, and humans. *J Comp Neurol* 441:197–222.
- Harting JK, Diamond IT, Hall WC. 1973a. Anterograde degeneration study of the cortical projections of the lateral geniculate and pulvinar nuclei in the tree shrew (*Tupaia glis*). *J Comp Neurol* 150:393–440.
- Harting JK, Hall WC, Diamond IT, Martin GF. 1973b. Anterograde degeneration study of the superior colliculus in *Tupaia glis*: evidence for a subdivision between superficial and deep layers. *J Comp Neurol* 148:361–86.
- Hendry SHC, Hockfield S, Jones EG, McKay RDG. 1984. Monoclonal antibody that identifies subsets of neurones in the central visual system of monkey and cat. *Nature* 307:267–269.
- Hendry SHC, Jones EG, Hockfield S, McKay RDG. 1988. Neuronal populations stained with the monoclonal antibody Cat-301 in the mammalian cerebral cortex and thalamus. *J Neurosci* 8:518–542.
- Hockfield S, McKay RDG. 1983. A surface antigen expressed by a subset of neurons in the vertebrate central nervous system. *Proc Natl Acad Sci USA* 80:6526–6530.
- Hockfield S, Sur M. 1990. Monoclonal antibody Cat-301 identifies Y-cells in the dorsal lateral geniculate nucleus of the cat. *J Comp Neurol* 300:320–330.
- Holdefer RN, Norton TT. 1995. Laminar organization of receptive field properties in the dorsal lateral geniculate nucleus of the tree shrew. *J Comp Neurol* 358:401–413.
- Hubel DH. 1975. An autoradiographic study of the retino-cortical projections in the tree shrew (*Tupaia glis*). *Brain Res* 96:41–50.
- Humphrey AL, Albano JE, Norton TT. 1977. Organization of ocular dominance in tree shrew striate cortex. *Brain Res* 134:225–236.
- Jones EG. 1985. The thalamus. New York: Plenum Press.
- Jain N, Preuss TM, Kaas JH. 1994. Subdivisions of the visual system labeled with the Cat-301 antibody in tree shrews. *Visual Neurosci* 11:731–741.
- Kaas JH. 2002. Convergences in the modular and areal organization of the forebrain of mammals, implications for the reconstruction of forebrain evolution. *Brain Behav Evol* 59:262–272.
- Kaas JH, Preuss TM. 1993. Archontan affinities as reflected in the visual system. In: Szalay F, Novacek and M McKenna M, editors. *Mammal phylogeny: placentals*. New York: Springer-Verlag. p 115–128.
- Kretz R, Rager G, Norton TT. 1986. Laminar organization of ON and OFF regions and ocular dominance in the striate cortex of the tree shrew (*Tupaia belangeri*). *J Comp Neurol* 251:135–145.
- Le Gros Clark WE. 1929. The thalamus of *Tupaia*. *J Anat* 63:117–206.
- Le Gros Clark WE. 1930. The structure and connections of the thalamus. *Brain* 55:406–470.
- Luppino G, Matelli M, Carey RG, Fitzpatrick D, Diamond IT. 1988. New view of the organization of the pulvinar nucleus in *Tupaia* as revealed by tectopulvinar and pulvinar-cortical projections. *J Comp Neurol* 273:67–86.
- Lyon DC, Jain N, Kaas, JH. 1998. Cortical connections of striate and extrastriate visual areas in tree shrews. *J Comp Neurol* 401:109–128.
- Lyon DC, Jain N, Kaas, JH. 2003. The visual pulvinar in tree shrews II. Projections of four nuclei to areas of visual cortex. *J Comp Neurol* 467:000–000.
- Marrocco RT, DeValois RL, Boles JJ. 1970. A stereotaxic atlas of the brain of the tree shrew (*Tupaia glis*). *J Hirnforschung* 12:307–312.
- Mitchison G, Crick F. 1982. Long axons within the striate cortex: their distribution, orientation, and patterns of connection. *Proc Natl Acad Sci USA* 79:3661–3665.
- Ohno T, Migseld U, Kitai ST, Wagner A. 1975. Organization of the visual afferents into the LGd and the pulvinar of the tree shrew *Tupaia glis*. *Brain Res* 90:153–158.
- Oliver DL, Hall WC. 1978. The medial geniculate body of the tree shrew, *Tupaia glis* I. Cytoarchitecture and midbrain connections. *J Comp Neurol* 182:423–458.
- Patel NC, Bickford ME. 1997. Synaptic targets of cholinergic terminals in the pulvinar nucleus of the cat. *J Comp Neurol* 387:266–278.
- Patel NC, Carden WB, Bickford ME. 1999. Synaptic targets of cholinergic terminals in the cat lateral posterior nucleus. *J Comp Neurol* 410:31–41.
- Petry HM, Kelly JP. 1991. Psychophysical measurements of spectral sensitivity and color vision in red-light-reared tree shrews (*Tupaia belangeri*). *Vision Res* 31:101749–1757.
- Petry HM, Fox R, Casagrande VA. 1984. Spatial contrast sensitivity of the tree shrew. *Vision Res* 24:1037–1042.
- Robson JA, Hall WC. 1977. Organization of the pulvinar in the grey squirrel (*Sciurus carolinensis*) I. Cytoarchitecture and connections. *J Comp Neurol* 173:355–388.
- Robertson RT, Mostamand F., Kageyama GH, Gallardo KA, Yu J. 1991. Primary auditory cortex in the rat: transient expression of acetylcholinesterase activity in developing geniculocortical projections. *Dev Brain Res* 58:81–95.
- Rockland KS, Lund JS. 1982. Widespread periodic intrinsic connections in the tree shrew visual cortex. *Science* 215:1532–1534.
- Rockland KS, Lund JS, Humphrey AL. 1982. Anatomical binding of intrinsic connections in striate cortex of tree shrews (*Tupaia glis*). *J Comp Neurol* 209:41–58.
- Sesma MA, Casagrande VA, Kaas JH. 1984. Cortical connections of area 17 in tree shrews. *J Comp Neurol* 230:337–351.
- Shipp S. 2001. Corticopulvinar connections of areas V5, V4, and V3 in the macaque monkey: a dual model of retinal and cortical topographies. *J Comp Neurol* 439:469–490.
- Snyder M, Diamond IT. 1968. The organization and function of the visual cortex in the tree shrew. *Brain Behav Evol* 1:244–288.
- Soares JGM, Gattass R, Souza APB, Rosa MGP, Fiorani M, Brandao BL. 2001. Connectional and neurochemical subdivisions of the pulvinar in *Cebus* monkeys. *Vis Neurosci* 18:25–41.
- Somogyi G, Hajdu F, Hassler R, Wagner A. 1981. An experimental electron microscopical study of a direct retino-pulvinar pathway in tree shrew. *Exp Brain Res* 43:447–450.
- Stepniewska I, Kaas JH. 1997. Architectonic subdivisions of the inferior pulvinar in New World and old World monkeys. *Vis Neurosci* 14:1043–1060.
- Stepniewska I, Qi H-X, Kaas JH. 1999. Do superior colliculus projection zones in the inferior pulvinar project to MT in primates. *Eur J Neurosci* 11:469–480.
- Stepniewska I, Qi H-X, Kaas JH. 2000. Projections of the superior colliculus to subdivisions of the inferior pulvinar in New World and Old World monkeys. *Vis Neurosci* 17:529–549.
- Tigges J, Shantha TR. 1969. A stereotaxic brain atlas of the tree shrew (*Tupaia glis*). Baltimore: Williams & Wilkins.
- Weller RE, Steele GE, Kaas JH. 2002. Pulvinar and other subcortical projections of dorsolateral visual cortex in monkeys. *J Comp Neurol* 450:215–240.
- Wong-Riley M. 1979. Changes in the visual system of monocularly sutured or enucleated cats demonstrable with cytochrome oxidase histochemistry. *Brain Res* 171:11–28.
- Wong-Riley MT, Norton TT. 1988. Histochemical localization of cytochrome oxidase activity in the visual system of the tree shrew: normal patterns and the effect of retinal impulse blockage. *J Comp Neurol* 272:562–578.