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Differences in Projection Patterns between Large and Small Corticothalamic Terminals

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ABSTRACT

We injected tracer into wide regions of visual cortex in the cat to produce retrograde and orthograde labeling in the thalamus, chiefly in the lateral geniculate nucleus and lateral posterior–pulvinar complex (LP-Pulvinar). We used the electron microscope to measure the sizes of orthogradely labeled terminals in thalamus and used these measurements to help determine whether the terminals were "RL" (large, presumed excitatory) or "RS" (small, presumed excitatory). We also distinguished *reciprocal regions*, which were zones of corticothalamic feedback defined by the presence of many retrogradely labeled cell bodies and orthogradely labeled terminals, from *nonreciprocal regions*, which were zones of feedforward corticothalamic projections defined by the presence of orthogradely labeled terminals alone. The lateral geniculate nucleus, a reciprocal region, had retrogradely labeled cell bodies as well as labeled RS terminals. Likewise, reciprocal regions in LP-Pulvinar were dominated by labeled RS terminals. In contrast, nonreciprocal regions were dominated by labeled RL terminals. Based on other evidence of corticothalamic projections that RL and RS terminals derive, respectively, from layer 5 and layer 6, we suggest the same relationship here, leading to the conclusion that the corticothalamic input from layer 6 is largely feedback, whereas that from layer 5 is largely feedforward. This finding lends credence to a recent hypothesis that layer 5 corticothalamic axons represent the afferent limb of a cortico-thalamo-cortical pathway that is critical for corticocortical communication. J. Comp. Neurol. 475:406–415, 2004.

Indexing terms: thalamus; cortex; pulvinar; first and higher-order thalamic relays

A major source of input to thalamic circuitry originates in cerebral cortex (reviewed in Sherman and Guillery, 1996, 2001). Much of this input is feedback, projecting back to thalamic targets that innervate the cortical area providing the projection. This is mostly true for the cat's lateral geniculate nucleus, although there are exceptions to a strict feedback arrangement. For instance, the geniculate A layers project to cortical areas 17 and 18 and receive a feedback projection from those areas, but they also receive a projection from cortical area 19, which thus is not strictly feedback (Garey and Powell, 1967; LeVay and Gilbert, 1976; Updyke, 1975, 1977).

However, the lateral geniculate nucleus and other comparable first-order relays, such as the ventral posterior (medial and lateral) nucleus and ventral part of the medial geniculate nucleus receive only or mainly layer 6 innervation from cortex (reviewed in Sherman and Guillery, 2001, 2002; Guillery and Sherman, 2002; see also Rouiller, et al., 1998; Darian-Smith et al., 1999). Higher-order thalamic relays, in addition to a layer 6 input, also receive a layer 5 input (Sherman and Guillery, 2001, 2002; Guillery and Sherman, 2002). An example of such a higher-order relay receiving both layer 6 and layer 5 inputs is the lateral posterior–pulvinar complex (LP-Pulvinar). Guillery and Sherman (Guillery, 1995; Sherman and Guillery, 2001, 2002; Guillery and Sherman, 2002) have suggested that the largely feedback layer 6 projections are modulatory, providing subtle regulation of thalamic relay properties, while the layer 5 projections are

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part of a feedforward cortico-thalamo-cortical information pathway. Although this hypothesis requires considerable rigorous testing to determine its validity, it does suggest that there might be extensive differences in corticothalamic projections that are feedback vs. those that are not feedback, the latter being candidates for a feedforward projection.

To explore this possibility, we used techniques of retrograde and orthograde labeling to study corticothalamic projections from wide areas of visual cortex to the LP-Pulvinar complex and the lateral geniculate nucleus in the cat. We operationally distinguished feedback projections from those that were not feedback and used the electron microscope to describe differences in the patterns of termination between these two types of projection. A preliminary report of these experiments was published in abstract form (Van Horn et al., 2002).

MATERIALS AND METHODS

Experiments were performed on two adult cats. The animals were treated strictly in accordance with NIH guidelines for the care and use of laboratory animals and the protocols used were approved by the Institutional Animal Care and Use Committee at the State University of New York at Stony Brook. Multiple injections of biotinylated dextran amine (BDA), spaced approximately 1 mm apart anterior to posterior, were made into each of cortical areas 17, 18, 19, and 21a. We used the maps of Tusa and colleagues (Palmer et al., 1978; Tusa et al., 1978, 1979; Tusa and Palmer, 1980) to judge location of each injection. Each injection consisted of 3–4 μl of 10% BDA administered by means of a 30-gauge needle.

After a 2-week survival period, the animals were deeply anesthetized with pentobarbital (100 mg/kg) and perfused transcardially with 4% paraformaldehyde, 0.05–0.1% glutaraldehyde, and 0.2% picric acid in 0.1 M phosphate buffered saline (0.9%). By using histological techniques that we have described previously (Erişir et al., 1997; Van Horn et al., 2000), we blocked areas of interest, post-fixed them overnight in the fixative perfusate, and sectioned them coronally on a Leica VT 1000 S Vibratome at 50 μm. A glucose oxidase, nickel intensified, diaminobenzidine reaction was performed to visualize the BDA injection sites and regions of orthograde and retrograde label in the thalamus. Light level digital images of the injection sites and labeled areas of thalamus were taken with a Zeiss Axiocam digital camera.

For electron microscopy, the 50-μm vibratomed sections were osmicated, dehydrated, and flat embedded in Durcupan resin. Areas of interests were blocked, and ultrathin sections were cut at 80 nm on a Reichart Ultracut E ultramicrotome. Three regions of interest were sampled. One was a region of the A layers of the lateral geniculate nucleus showing retrograde and orthograde labeling. The other two were in LP-Pulvinar and were based on the presence or absence of BDA-labeled cell bodies: *reciprocal*, being a region that includes retrogradely labeled cell bodies as well as orthogradely labeled terminal arbors; and *nonreciprocal*, being a region that is relatively free of labeled cell bodies but that contains orthogradely labeled terminal arbors. It is important to note that these definitions of reciprocal and nonreciprocal are strictly operational and depend on the negative result of failing to find cell bodies in the latter region (see also Discussion section).

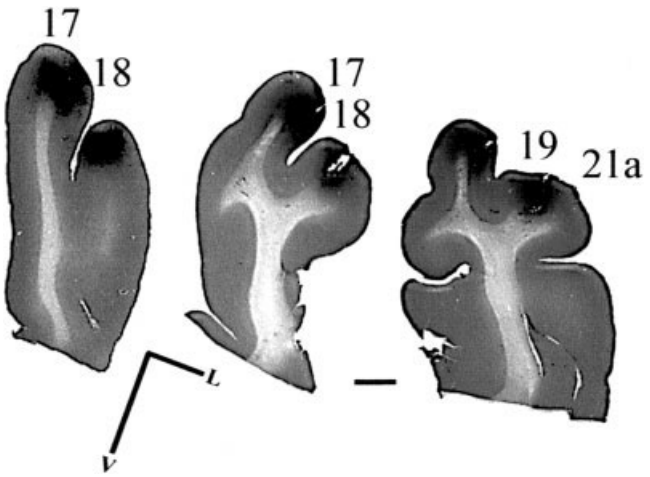

Fig. 1. Injection sites in visual cortex. Series of light-level digital images, left to right from posterior to anterior, depicting actual injection sites in cortical areas 17, 18, 19, and 21a. V, ventral; L, lateral. Scale bar = 2 mm.

Every second section was picked up on a formvar-coated nickel slot grid, counterstained with lead citrate and viewed with a JEOL 1200ExII electron microscope. Every BDA-labeled terminal in a field making a synaptic contact was photographed and the area of that terminal at the contact site was measured with BioScan/Optimas software.

Terminal identification was based on morphological criteria from Guillery's (1969a,b) classification, which includes terminal size (large or small), vesicle shape (round or pleomorphic), synaptic zone appearance (symmetrical or asymmetrical), and often mitochondria contrast (pale or with contrast). RL terminals (for *R*ound vesicles and *L*arge terminal) also had asymmetric synaptic zones, and these typically had pale mitochondria. RS terminals (for *R*ound vesicles and *S*mall terminal) also had asymmetrical synaptic zones, and these typically had dark mitochondria. Only RS and RL terminals were found to be labeled from cortex. The other major terminal type, known as F (for *F*lattened vesicles) also had symmetric synapses, but because these were not labeled from cortex, they are not considered further. Terminal size was derived in two ways (see Results for details): by measuring the area in a single section in which a synaptic contact was evident, or by estimating the volume from reconstructions using serial sections.

Light-level photomicrographs were captured with a Zeiss, AxioCam digital camera. Electron photomicrographs were printed from Kodak #4489 film and scanned in with an Epson Perfection 1260 scanner. Images were then taken into Adobe Photoshop 6.0 and CorelDRAW 9 programs and prepared for final presentation.

RESULTS

Light microscopic evidence

Injection sites. Figure 1 shows the injection sites for one of the animals. A comparison of Figure 1 with maps of

cat cortex (Palmer et al., 1978; Tusa et al., 1978, 1979; Tusa and Palmer, 1980) suggests that label was placed widely into visual cortical areas 17, 18, 19, and 21a. In any case, details of the extent of label are irrelevant for the main purpose of this study, which was to place orthograde and retrograde label into wide areas of visual cortex, including area 17, and clearly we succeeded with this.

Labeling in thalamus. Figure 2 shows the patterns of labeling seen in thalamus. This was of two general forms. One included many retrogradely labeled cell bodies embedded in what appeared to be many orthogradely labeled axonal terminal arbors, presumably reflecting labeling of corticothalamic axons (Figs. 2, 3). We refer to this as a *reciprocal* region or *reciprocal* labeling, because it suggests a region of thalamus that receives a reciprocal projection from the region of cortex to which it projects. The other included heavy labeling of axonal arbors without a dense aggregate of retrogradely labeled cell bodies, although a few scattered cell bodies may be found in or near such regions (Figs. 2, 3). We refer to this as a *nonreciprocal* region or *nonreciprocal* labeling. Note that this definition of different regions is purely operational (see also Discussion section). The retrograde labeling both in the LP-Pulvinar and lateral geniculate nucleus was limited to the cell bodies and most proximal dendrites but rarely extended to the first branch point and never into distal dendrites (see also below).

All of the labeling seen in the lateral geniculate nucleus was of a reciprocal nature (Figs. 2, 3), but the LP-Pulvinar contained regions of both reciprocal and nonreciprocal labeling (Figs. 2, 3). Each type of zone for analysis was chosen by inspection. It is interesting to note that the dense labeling of cell bodies in the lateral geniculate nucleus is similar to that seen with other retrograde labeling procedures (e.g., LeVay and Ferster, 1977; Montero and Zempel, 1985), thus suggesting that the retrograde labeling we have achieved with our technique is likely to be comparable to that in prior studies.

Note that the retrograde labeling in the lateral geniculate nucleus showed a high density of labeled neurons (Fig. 2C), as expected, because roughly three fourths of geniculate cells are relay cells (Fitzpatrick et al., 1984; Montero and Zempel, 1985). However, the retrograde label in both the lateral geniculate nucleus and LP-Pulvinar was largely limited to the cell body and most proximal dendrites (Fig. 2B,D). Our impression from light microscopy was that the nonreciprocal region of labeling contained labeled arbors with thicker axons and larger terminal boutons than did the reciprocal region. This was verified by electron microscopy (see below).

Electron microscopic evidence

As expected from light microscopic evidence, retrograde labeling was evident in the lateral geniculate nucleus and reciprocal regions of the LP-Pulvinar, but it was effectively limited to cell bodies and thick, presumably proximal, dendrites. We did not see labeling in fine processes that could be interpreted as distal dendrites. We also saw many orthogradely labeled synaptic terminals in the zones indicated. Figures 4–6 shows examples of labeled terminals found in the lateral geniculate nucleus and LP-Pulvinar.

Terminal sizes. Our general observation was that terminals labeled in reciprocal regions, including the lateral geniculate nucleus, were smaller than those in nonreciprocal regions. One measure of this was to determine the cross-sectional area of labeled terminals having a clear synaptic contact within a sampling region. To avoid sampling bias, we started at one corner of an area and chose every terminal forming a clear synaptic contact, moving row by row, until a predetermined number of terminals was sampled. From each of the two cats, we chose a sampling region within the A layers of the lateral geniculate nucleus and from both a reciprocal and nonreciprocal region of the LP-Pulvinar. We found consistency within matched pairs of these labeling regions, so that, for instance, there was no significant difference in cross-sectional area between the similar zones of each cat (i.e., lateral geniculate nucleus, reciprocal region of LP-Pulvinar, and nonreciprocal region; $P > 0.1$ on a Mann–Whitney U test). We thus pooled data of these pairs to create the three distinct sampling zones.

Figure 7 shows the results of this for cross-sectional area. Labeled terminals in the lateral geniculate nucleus were all small as were most of those in the reciprocal region of LP-Pulvinar, but nonreciprocal regions of LP-Pulvinar had many larger terminals. Statistically, the labeled terminals in nonreciprocal regions of LP-Pulvinar ($1.281\ \mu m^2 \pm 0.083$; n = 107) were larger than those either in reciprocal regions ($0.534\ \mu m^2 \pm 0.082$; n = 69) or in the lateral geniculate nucleus ($0.175\ \mu m^2 \pm 0.012$; n = 48), and those in reciprocal regions of LP-Pulvinar were larger than those in the lateral geniculate nucleus ($P < 0.001$ in all comparisons on a Mann–Whitney U test). The histograms of Figure 7 suggest a bimodal distribution of terminal sizes (separated by the vertical dashed line), with those in the lateral geniculate nucleus being exclusively of the smaller mode (Fig. 7A), those in the reciprocal region of LP-Pulvinar being mostly of the smaller mode with a minority of larger terminals (Fig. 7B), and those in the nonreciprocal region being mostly of the larger mode with a minority of smaller terminals (Fig. 7C).

To confirm the terminal size differences between reciprocal and nonreciprocal regions of LP-Pulvinar were not due to a sectioning artifact (e.g., sampling eccentric sections through many large terminals, making them appear small), we serially reconstructed a subset of 15 randomly selected terminals from each area. All of the terminals in the reciprocal region were completely reconstructed, but we stopped the reconstruction of 11 terminals in the nonreciprocal region as soon as it became clear that each was larger than any in the reciprocal region ($>0.65\ \mu m^3$). Figure 8 shows the results. Again, terminals in the nonreciprocal region were significantly larger ($P < 0.001$ on a Mann–Whitney U test).

Synaptic relationships and terminal types. Labeled terminals were typically found on profiles deemed to be dendrites, but we never found a labeled terminal on a labeled postsynaptic profile, even in the lateral geniculate nucleus or reciprocal regions of the LP-Pulvinar. There are two likely reasons for this. First, some of the postsynaptic targets are likely to be interneurons and would not be retrogradely labeled in any case. Second, relay cells had retrograde labeling limited to their cell bodies and very proximal dendritic stumps (Fig. 2B,D), and this finding was also evident in electron photomicrographs from the reciprocal regions (not illustrated). Thus, virtually all synaptic inputs to these cells, labeled or unlabeled, would terminate on dendritic processes without label.

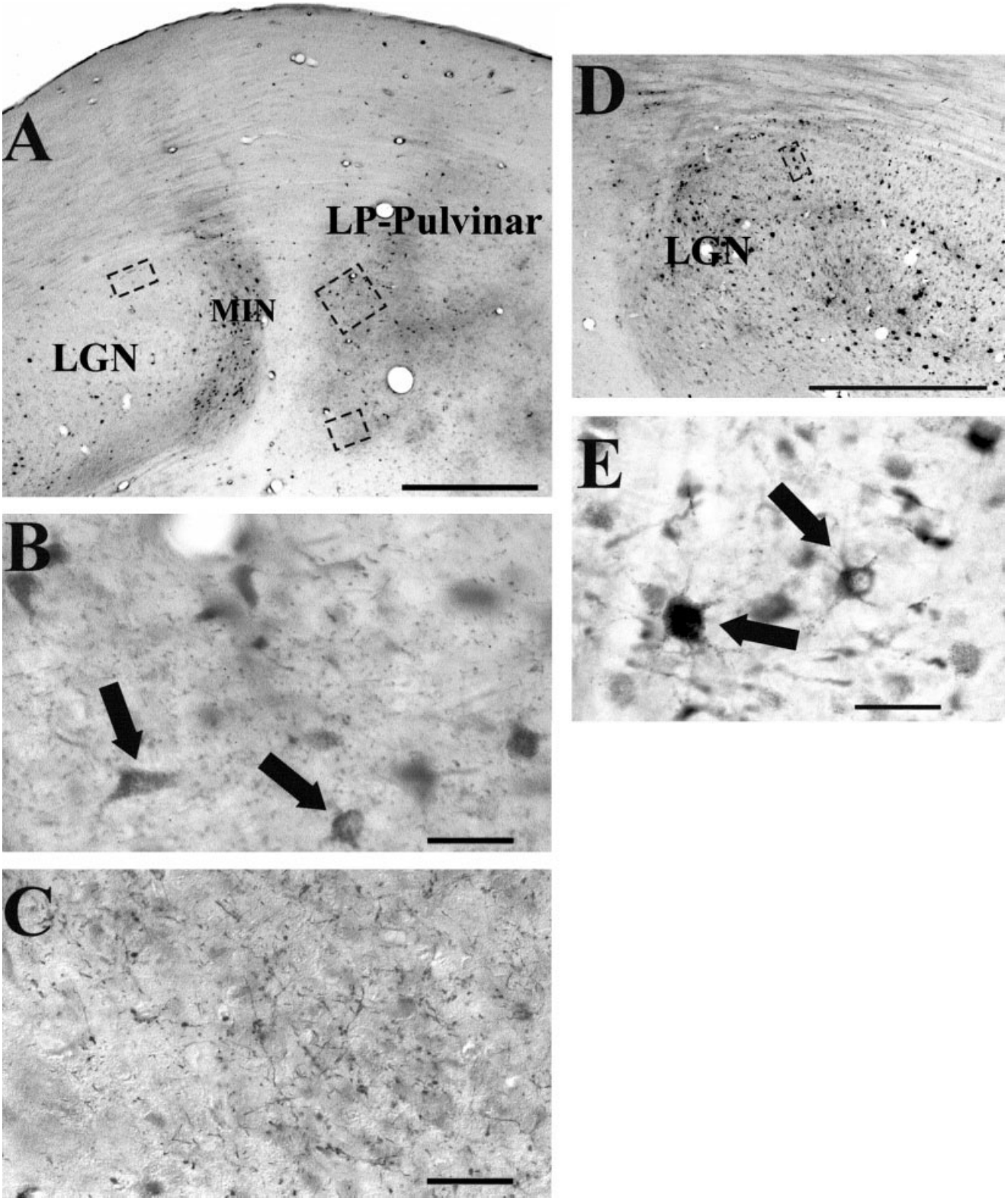

Fig. 2. Areas of interest sampled viewed with the light microscope. **A:** Low-power image showing transport of biotinylated dextran amine label. Three distinct sampled areas are shown (boxes): one in the lateral geniculate nucleus (LGN; but with label mainly in the medial interlaminar nucleus [MIN], which is part of the lateral geniculate nucleus) and two in the lateral posterior–pulvinar complex (LP-Pulvinar) complex. Of the latter, the upper one has retrogradely labeled cells and is thus a *reciprocal region*, and the lower has orthogradely labeled terminals only and is thus a *nonreciprocal region* (see text for details). **B:** Higher magnification of reciprocal region in LP-Pulvinar showing retrogradely labeled cell bodies (arrows). **C:** Higher magnification of nonreciprocal region in LP-Pulvinar showing only orthogradely labeled terminals and no labeled cell bodies. **D:** Low-power image showing labeling in A layers of lateral geniculate nucleus. **E:** Higher magnification of labeling from boxed region within D showing retrogradely labeled cells (arrows). Scale bars = 1 mm in A,D; 50 μm in B,C,E

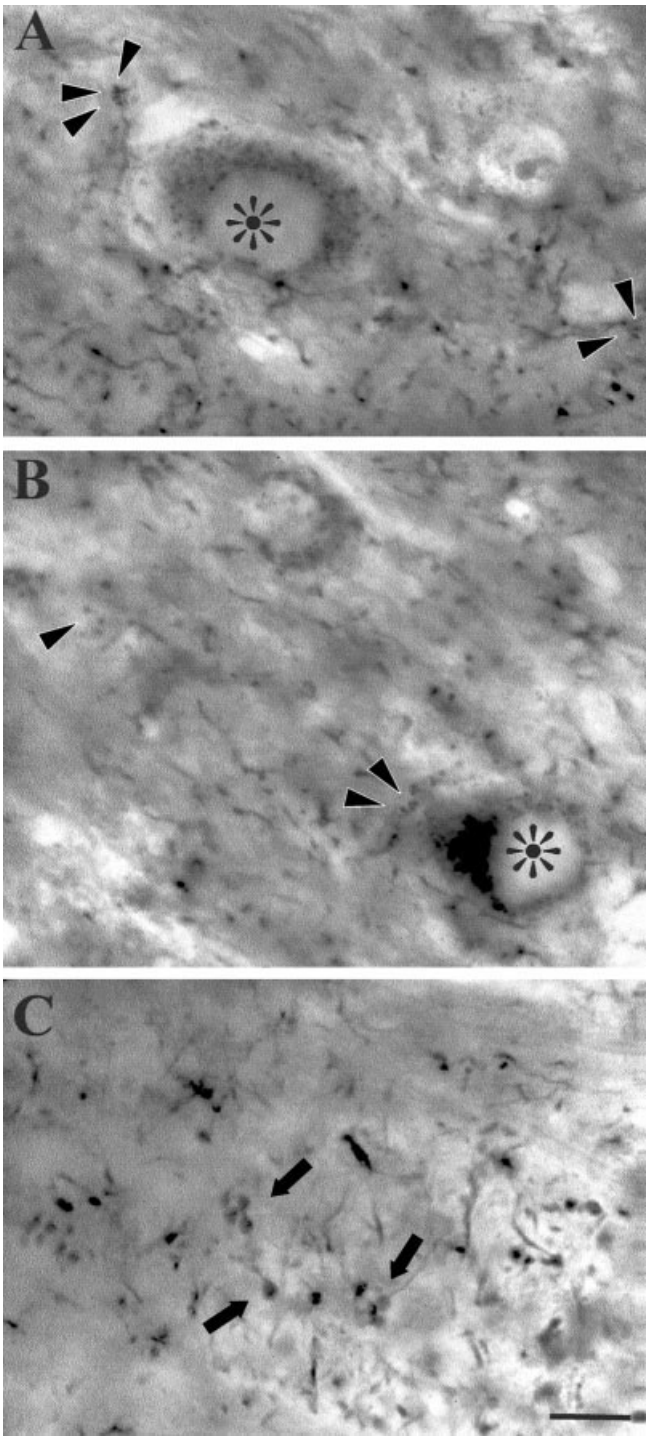

Fig. 3. Light microscopic views of biotinylated dextran amine label in sampled areas. **A:** View in lateral geniculate nucleus showing retrogradely labeled cell body (asterisk) and small orthogradely BDA-labeled terminals (arrowheads). **B:** View in *reciprocal region* of lateral posterior–pulvinar complex (LP-Pulvinar); conventions as in A. **C:** View in *nonreciprocal region* of LP-Pulvinar showing large orthogradely labeled terminals (arrows). Scale bar = 10 μm in C (applies to A–C).

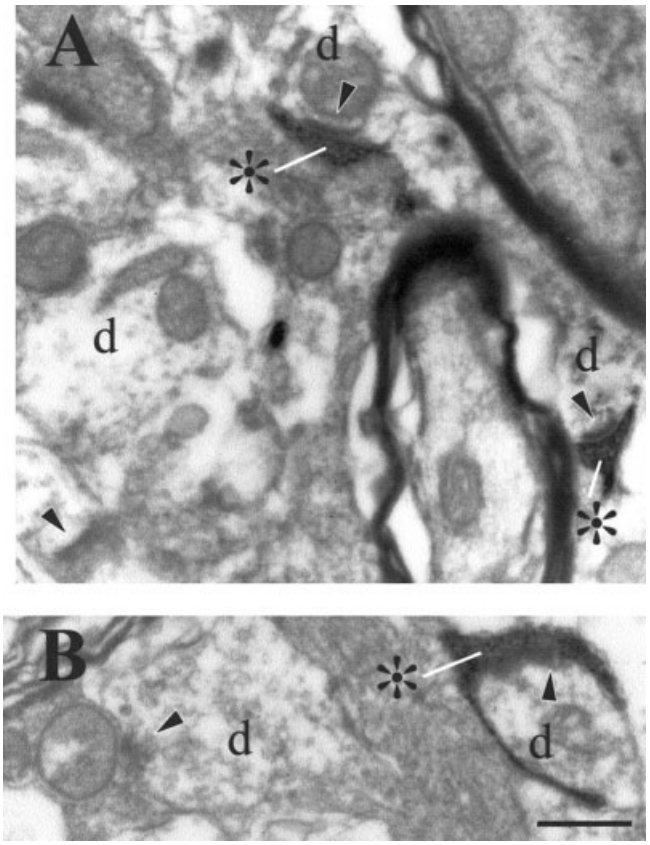

Fig. 4. **A,B:** Electron photomicrographs in lateral geniculate nucleus showing small orthogradely labeled terminals forming synapses onto small caliber dendrites. Asterisks indicate labeled terminals; arrowheads indicate synapses, and d indicates dendrite. Scale bar = 1 μm in B (applies to A,B).

Although the labeling can occasionally mask the vesicle shape and mitochondria contrast, the size differences alone of the terminals labeled from cortex make the distinction between RS and RL fairly straightforward. Based on the criteria described in the Materials and Methods section, in our material, all terminals having a cross-sectional area less than 0.7 μm^2 were judged to be RS, and all larger ones were judged to be RL. This division is indicated in Figure 7 by a dashed, vertical line, and this finding shows that the division does more or less divide what appears to be a bimodal population of terminal sizes. Thus, 56 of the 69 labeled terminals in the reciprocal area are RS (81.2%) and 82 of 107 (76.6%) in the nonreciprocal area are RL.

Of the 48 labeled RS terminals in the lateral geniculate nucleus, we confidently determined the postsynaptic target of 43. As we and others have reported previously (Jones and Powell, 1969; Guillery, 1969b; Vidnyanszky and Hamori, 1994; Erişir et al., 1997), every one of these 43 corticothalamic terminals contacted a relatively thin and, thus, presumably distal, dendrite, and none was found within a glomerulus (e.g., Fig. 4). We saw the same general relationships for reciprocal regions of LP-Pulvinar. Of the 69 labeled terminals sampled, the 56 that were RS were presynaptic to dendrites only. Of the 13 that

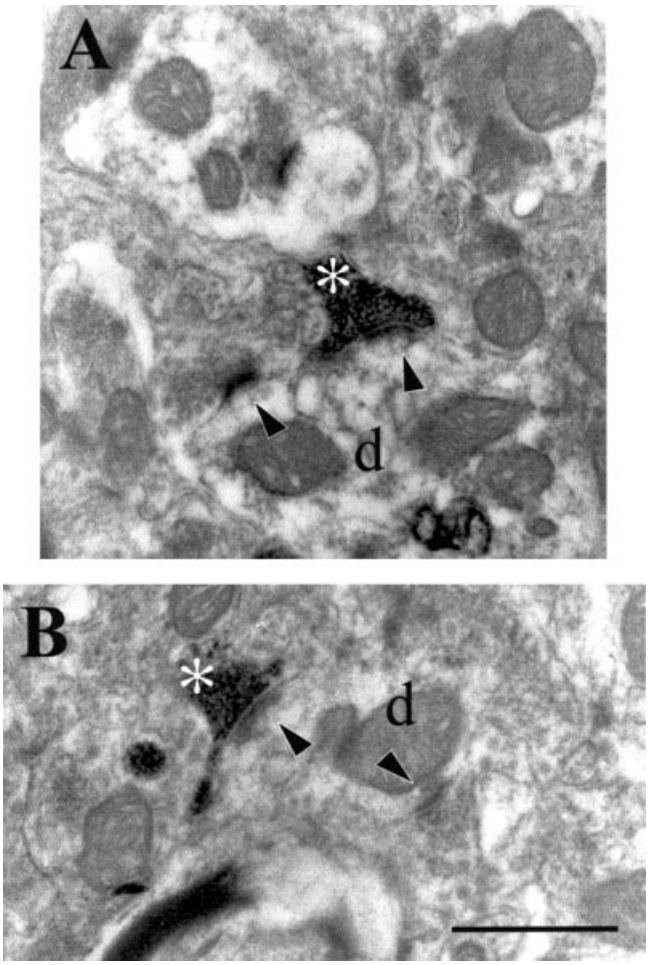

Fig. 5. **A,B:** Electron photomicrographs in *reciprocal region* of lateral posterior–pulvinar complex, showing small orthogradely labeled terminals forming synapses onto small caliber dendrites. Conventions as in Figure 4. Scale bar = 1 μm in B (applies to A,B).

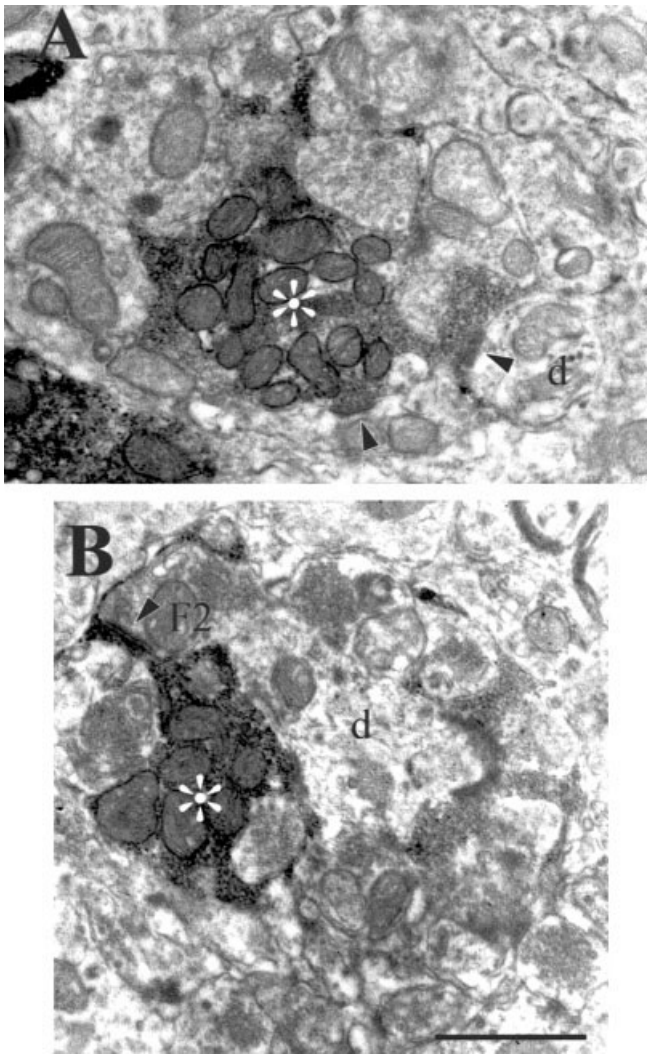

Fig. 6. Electron photomicrographs in *nonreciprocal region* of lateral posterior–pulvinar complex, showing large orthogradely labeled terminals in glomerular-like regions. **A:** Large labeled terminal (asterisk) forming a synapse onto dendrite (d); synapses indicated by arrowheads. **B:** Large labeled terminal (asterisk) forming a synapse (arrowhead) onto vesicle filled profile (F2). Scale bar = 1 μm in B (applies to A,B).

were RL, 10 contacted dendrites only, and the other 3 contacted both a dendrite and a vesicle filled profile that, by analogy with the lateral geniculate nucleus, was probably a dendritic terminal from an interneuron (Guillery, 1969b; Ralston, 1971; Famiglietti and Peters, 1972; Hamos et al., 1985). For the subset of 15 terminals completely reconstructed in the reciprocal regions (e.g., Fig. 8), all make a single contact onto a dendrite. However, the second largest terminal not only contacts a dendrite but also makes a second contact onto a profile that we could not identify, as was the case in the lateral geniculate nucleus.

In contrast, the nonreciprocal regions of LP-Pulvinar were quite different. Of the 25 identified as RS, we could be confident of the postsynaptic target for only 11, and each of these targets was a dendrite. Of the other 82 terminals identified as RL, we could be confident of the postsynaptic target for 66. Of these, 51 make contacts onto dendrites only and 15 contact both a dendrite and vesicle-filled profile (e.g., Fig. 6). Because we stopped reconstruction of terminals in the nonreciprocal regions once their area became larger than 0.65 μm^2, only the four smallest were completely reconstructed (e.g., Fig. 8). Of these four, the smaller two contacted only dendrites, whereas the larger two contact both dendrites and vesicle filled profiles.

DISCUSSION

By determining the sizes of synaptic terminals orthogradely labeled from visual cortex in thalamus, we found that large terminals, identified as RL, tended to be labeled in areas of LP-Pulvinar distinct from retrogradely labeled thalamic relay cells, whereas small terminals, identified as RS, tended to be labeled in areas of LP-Pulvinar containing retrogradely labeled cells. In comparison, the lateral geniculate nucleus, which had many retrogradely la-

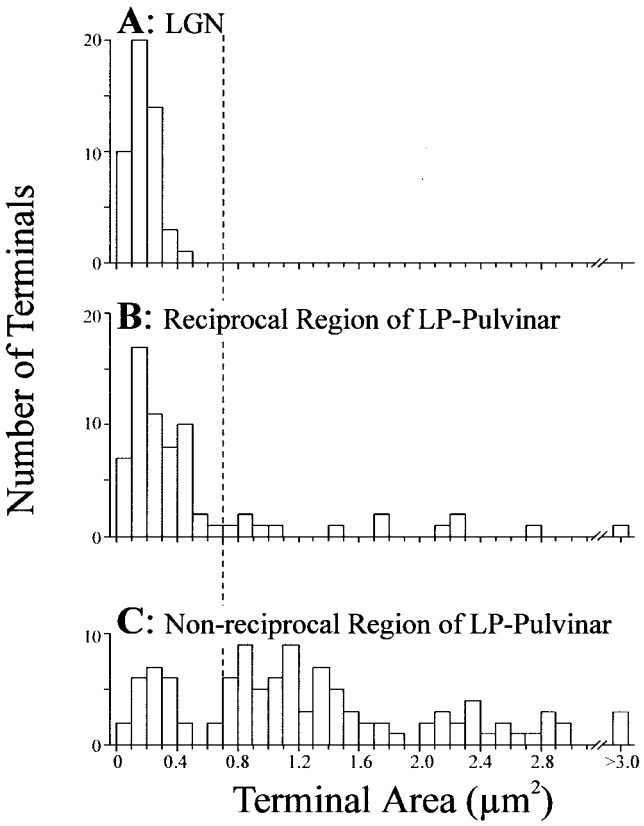

Fig. 7. Distributions of cross-sectional areas of orthogradely labeled terminals. **A:** Distribution in lateral geniculate nucleus (LGN). **B:** Distribution in *reciprocal region* of lateral posterior–pulvinar complex (LP-Pulvinar). **C:** Distribution in *nonreciprocal region* of LP-Pulvinar.

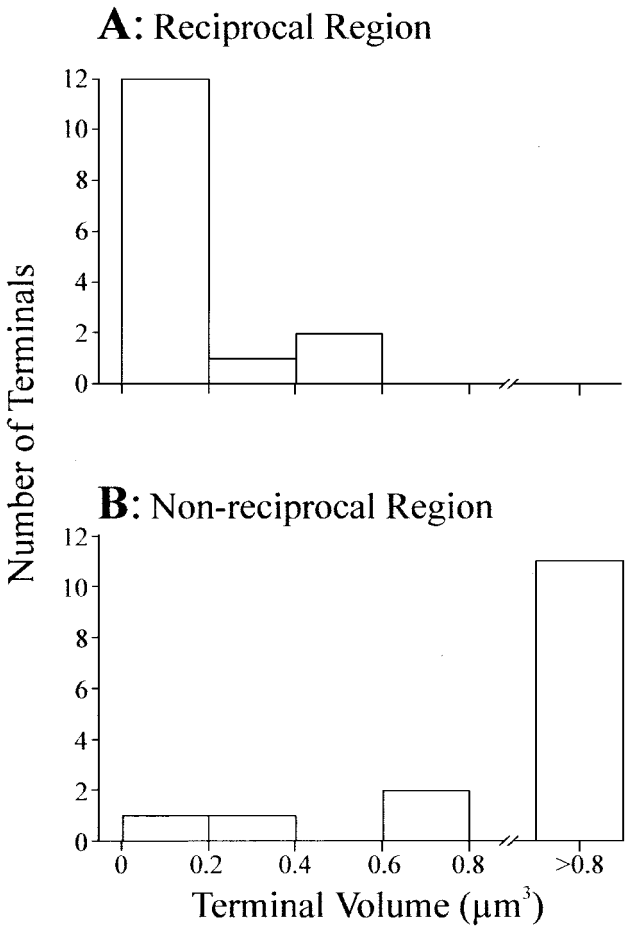

Fig. 8. Distributions of volumes of orthogradely labeled terminals. **A:** Distribution of 15 terminals in *reciprocal* region of lateral posterior–pulvinar complex (LP-Pulvinar). **B:** Distribution of 15 terminals in *nonreciprocal region* of LP-Pulvinar.

beled relay cells had only RS terminals labeled from cortex.

Figure 9 summarizes the main conclusions of this study. The lateral geniculate nucleus (Fig. 9A) receives cortical input producing only smaller (RS) terminals from layer 6, and this is a reciprocal zone, because the cortical input overlaps cells projecting to the same cortical area. This finding suggests a thalamo-cortico-thalamic relationship that is mainly feedback. Corticothalamic projections to the LP-Pulvinar relate to two thalamic zones: mainly reciprocal and mainly nonreciprocal (Fig. 9B). For the former, the relationships are basically like those of the lateral geniculate nucleus. For the latter, the projection from a given cortical area ends in larger (RL) terminals in regions devoid of relay cells projecting to that cortical area, and this finding suggests a thalamo-cortico-thalamic relationship that is mainly feedforward.

Feedback vs. feedforward relationships

Qualification for interpretation. There are at least two qualifications that need to be discussed with the conclusion schematically shown in Figure 9. First, the identity of the nonreciprocal region depends on the failure of finding a dense aggregate of retrogradely labeled cell bodies. This is a negative result, meaning that we cannot entirely rule out the possibility that these thalamic zones indeed have cells projecting into cortically labeled areas but have failed to retrogradely transport the label. This is always a possibility, however remote, with any retrograde labeling study. Even if it were true that such a false-negative occurred, this would still represent some form of difference between reciprocal and nonreciprocal regions. However, the observation of dense aggregates of labeled cells in the lateral geniculate nucleus and other regions of the LP-Pulvinar make the possibility of such a massive false-negative unlikely.

Second, the identity of the reciprocal region is based on large injections covering multiple cortical areas. It is plausible, thus, that some limited departures from true reciprocity are masked by the extent of labeled cortex. For instance, the lateral geniculate nucleus is operationally defined here as a reciprocal region, but as noted above, the input to the A layers from area 19 is not truly reciprocal, and a similar possibility exists for reciprocal regions in LP-Pulvinar. Thus, we emphasize the point that we have operationally defined these zones with qualifications like this in mind. In any case, the reciprocal regions so defined in LP-Pulvinar are different from nonreciprocal regions with respect to the nature of corticothalamic terminals found there.

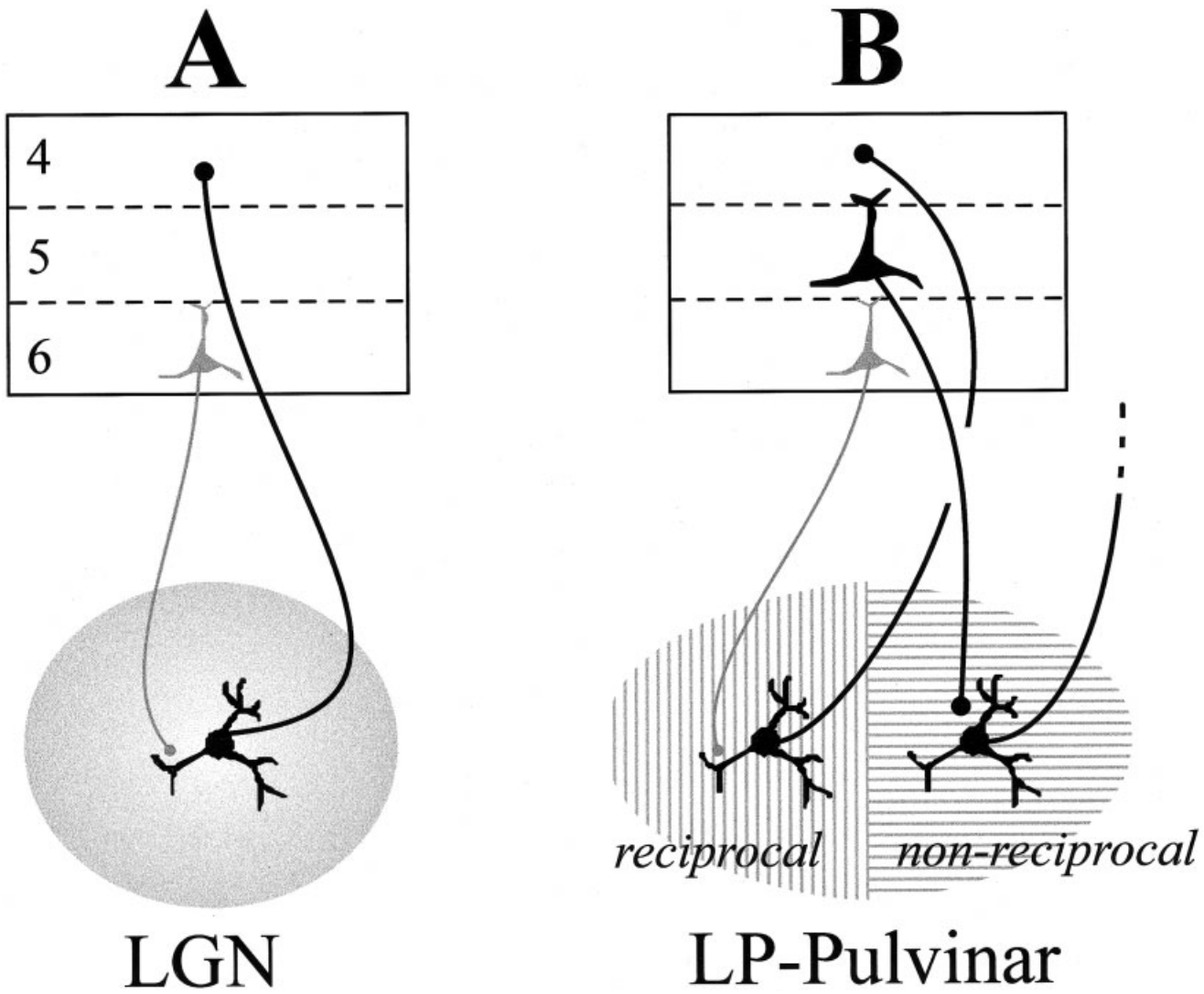

Fig. 9. Schematic summary figure of thalamocortical circuitry for the lateral geniculate nucleus (LGN) and lateral posterior–pulvinar complex (LP-Pulvinar). **A:** Circuitry for lateral geniculate nucleus, which is feedback and involves only layer 6 of cortex. **B:** Circuitry for LP-Pulvinar shown from the perspective of one cortical region. This region has a reciprocal relationship with one region of LP-Pulvinar (left) involving layer 6 and a nonreciprocal relationship with another region (right) involving layer 5.

Differences between feedback and feedforward projections. While our data support a mainly feedback role for the corticothalamic projections terminating with RS synapses vs. a mainly feedforward one for those terminating with RL synapses, the distinction was not perfect, because some of the synaptic terminals in the reciprocal region, suggesting feedback, appeared to be RL, and some of those in the nonreciprocal region, suggesting feedforward, appeared to be RS. There are three very different explanations, among others, that bear elaboration. First, there may be significant exceptions to the "rule" that corticothalamic axons ending in RS terminals are feedback and those ending in RL terminals are feedforward, even if these exceptions are small in number (see Rouiller et al., 1998; Darian-Smith et al., 1999). Second, perhaps there are no significant exceptions to this general rule but instead for axons with mainly RS terminals, a small percentage are RL, and vice versa, although the former seems not to be the case for corticothalamic inputs to the lateral geniculate nucleus (Fig. 7A). Third, and most plausible to us as a contributor, is that these exceptions are largely an artifact of the very large injections made in cortex. That is, a reciprocal region of LP-Pulvinar may receive a true feedback projection from the area to which its cells project in one part of the cortical area labeled, and it may also receive a true feedforward projection from another, separate region of labeled cortex to which its cells do not project. A similar argument could account for RS terminals found in the nonreciprocal region. To distinguish between these alternatives requires reinvestigating the problem with numerous, smaller injections of label into cortex. Nonetheless, even with this proviso, we believe our data make a strong case that, at least to a first approximation, corticothalamic axons terminating in RS termi-

nals are involved in feedback connections, while those terminating in RL terminals are feedforward.

Implications for thalamocortical processing of information

The conclusion here that corticothalamic RS terminals are largely involved in feedback projections while RL terminals are not is interesting in the context of a hypothesis recently proposed by Guillery and Sherman (Guillery, 1995; Sherman and Guillery, 1998, 2001; Guillery et al., 2001). Part of this is the understanding that corticothalamic axons originate from both layers 5 and 6, and there are likely to be important differences in the function of these pathways related to layer of origin. For instance, all thalamic relays seem to have a projection from layer 6 (e.g., the lateral geniculate nucleus), but some in addition have a projection from layer 5 (e.g., the LP-Pulvinar). The available evidence for the lateral geniculate nucleus and LP-Pulvinar (reviewed in Guillery, 1995; Sherman and Guillery, 1998, 2001; Guillery et al., 2001) is that the RS terminals derive from layer 6 axons, whereas the RL terminals derive from layer 5 axons. Although we cannot be absolutely certain of these relationships between terminal type and cortical layer of origin for the specific corticothalamic pathways studied here, this seems a reasonable assumption.

The tentative conclusion here that the layer 6 corticothalamic projection is largely related to reciprocal regions, suggesting a feedback projection, that can be considered in the context of its purported role as modulator (reviewed in Sherman and Guillery, 1998, 2001, 2002). The idea is that this projection is a modulator provides several relatively subtle controls over thalamic relay functions, and a number have been suggested, such as control of relay cell response modes between burst and tonic (Sherman, 1996, 2001), general effects on firing level (Kalil and Chase, 1970; Baker and Malpeli, 1977; Schmielau and Singer, 1977; Geisert et al., 1981; McClurkin and Marrocco, 1984; McClurkin et al., 1994), effects on receptive field surrounds (reviewed in Sillito and Jones, 2002), etc. Our data suggest that this role from a cortical zone, or perhaps even a cortical column, is largely limited to control over the very relay cells that innervate that zone or column. However, given the proviso noted above that some of the layer 6 projections might extend beyond relay cells that provide innervation to its region or column, this modulatory function may in some cases extend beyond simple feedback. As noted above, there is some evidence for this in the lateral geniculate nucleus of the cat in that the A layers, which innervate only cortical areas 17 and 18 (reviewed in Sherman, 1985), in addition to receiving layer 6 feedback from those areas also receive limited layer 6 input from beyond those areas (Updyke, 1975).

On the other hand, our observation that at least most of the layer 5 projection to thalamus is nonreciprocal, and thus feedforward, is consistent with the idea that this pathway serves to transmit information in the role of a driver (Sherman and Guillery, 1998). That is, it performs much like retinal input to the lateral geniculate nucleus to provide the information to be relayed, as distinct from the layer 6 input to geniculate relay cells, which is modulatory. Also, because the projection involving these putative layer 5 afferents is nonreciprocal, information it carries from one cortical area must be relayed to another cortical area. We regard this, then, as an additional piece of evidence supporting the role of the layer 5 corticothalamic projection as part of a cortico-thalamo-cortical route for processing information, the thalamic link being a higher-order relay.

Note that this scheme challenges the prevailing dogma that processing of information within cortex is based solely or nearly so on direct connections between cortical areas (e.g., Kaas, 1978, 1987; Van Essen and Maunsell, 1983; Van Essen, 1985; Zeki and Shipp, 1988; Van Essen et al., 1990, 1992; Felleman and Van Essen, 1991; Preuss et al., 1993; Van Essen and Gallant, 1994; DeYoe et al., 1994). For visual cortex, for instance, the prevailing view of the functional organization of the many discrete cortical areas is based almost entirely on the implicit assumption of information flow by means of direct corticocortical pathways that establish hierarchical relationships among areas. The main challenge we suggest here is that a consideration of cortico-thalamo-cortical pathways for information flow could radically alter these hierarchical relationships. Another consequence of the alternative view proposed is that all information targeted for a cortical area, whether originating in the periphery (e.g., the retina) or another cortical area (e.g., layer 5) benefits from a thalamic relay. That is, just as retinal input is relayed through the lateral geniculate nucleus rather than directly innervating visual cortex, most or all information passed between cortical areas is relayed through the thalamus.

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