

Morphology of Retinal Axon Arbors Induced to Arborize in a Novel Target, the Medial Geniculate Nucleus. II. Comparison With Axons From the Inferior Colliculus

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ABSTRACT

Specific neonatal lesions in ferrets can induce retinal axons to project into the medial geniculate nucleus (MGN). In the accompanying paper (Pallas et al., this issue), we described the morphology of these retinal ganglion cell axons. Those results and others (Roe et al. [1993] *J. Comp. Neurol.* 334:263) suggest that these axons belong to the W class of retinal axons.

In this paper, the retino-MGN axons are compared with the normal inputs to the MGN from the brachium of the inferior colliculus (BIC). We first sought to determine further the extent to which a novel target might influence retinal axon arbor morphology. The second issue concerns retinal topography. Ferrets with retinal projections to the MGN have a two-dimensional retinotopic map in the MGN and the primary auditory cortex rather than the one-dimensional tonotopic map normally present (Roe et al. [1990] *Science* 250:818). To investigate whether there might be an anatomical substrate for a two-dimensional retinotopic map in the MGN, we compared the space-filling characteristics of the retino-MGN axons with the IC-MGN axons.

Our results show that the branched retino-MGN axons resemble normal retinal W axons much more closely than they resemble the normal inputs to MGN. In addition, most of the axon arbors from the BIC are elongated along the rostrocaudal (isofrequency) axis, whereas the branched retino-MGN axons are more spatially restricted, suggesting an anatomical substrate for a retinotopic map in the MGN of the rewired ferrets. © 1994 Wiley-Liss, Inc.

Key words: cross-modal plasticity, ferret, auditory topography, retinotopic map, auditory development

In an ongoing series of studies in our laboratory, we have examined the effect of visual inputs on the morphological and functional development of auditory brain structures in the ferret (Sur et al., 1988; Pallas et al., 1990, Pallas and Sur, 1993; Roe et al., 1990, 1991, 1993). In this series of two papers, we have used this preparation to describe the morphology of retinal axons that are induced to arborize anomalously in the medial geniculate nucleus (MGN) and to examine the role of target identity in the morphological development of afferent axon arbors.

In the first paper (Pallas et al., this issue), we examined whether the retinal axons in the MGN showed arborization patterns similar to what they exhibit in their normal targets (the lateral geniculate nucleus and the superior colliculus). We described two different populations of retinal axons projecting to the MGN; branched axons, which form terminal arbors in MGN, and unbranched axons, which pass through the MGN with only en passant boutons. We found that the branched retinal axons arborizing

in MGN resembled retinal ganglion cell axons (presumably W axons) arborizing in the ventral C-laminae of the lateral geniculate nucleus (LGN) or in the superior colliculus (SC), in that they had fine-caliber axons, small arbor volumes, and similar bouton numbers. (W cells are a heterogenous group, however, and it is possible that not all types of W cells project to the MGN.) Our reconstructed branched retino-MGN axons did not resemble X- and Y-retinal axons arborizing in the LGN, which had much larger arbors, axon diameters, and numbers of boutons. This result suggested that the branched retinal axons are not substantially altered morphologically by their presence in the novel target.

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In the present study, we continued to investigate to what degree the target structure influences the morphology of axons arborizing within it. To this end, we examined whether the branched retino-MGN axons resemble axons from the inferior colliculus that normally make up the main ascending afferent input to the MGN (Kudo and Niimi, 1980). There is support in the literature for both afferent and target control of axon arbor morphology. Our data provide support for the idea that arbor morphology is at least in part independent of target identity.

Another issue that we wished to investigate concerned the topography of afferent projections into the MGN. Previous work in our laboratory has shown that, in ferrets with retinal afferents directed to the MGN by neonatal lesions, both the MGN and the primary auditory cortex (AI) contain a two-dimensional map of the retina (Roe et al., 1990, 1991, 1993). Normally, the various way stations in the central auditory system contain a one-dimensional representation of the cochlea (Aitkin et al., 1984). This is reflected in the fact that the projections from the inferior colliculus (IC) to the MGN and from the MGN to AI are restricted only along the tonotopic dimension and are overlapped extensively along the isofrequency dimension (Andersen et al., 1980a,b; Imig and Morel, 1985). Because of the "one-dimensional" nature of the IC to MGN projection, one might expect that normal IC axons would arborize extensively along the isofrequency plane. In contrast, to subserve a visual map in the MGN, the retinal ganglion cell arbors there should be restricted in two dimensions. It was thus of interest to us to determine whether retinal axon arbors in the rewired MGN were indeed more spatially restricted than IC arbors in the normal MGN.

There is very little information available on the morphology of single IC axon arbors in the MGN (but see Winer, 1985; Wenstrup et al., 1994). Part of the intent for this study, then, was to provide information on this subject in addition to comparing the normal axon arbors with retinal arbors in the MGN. Parts of this work have been reported previously in preliminary form (Pallas et al., 1989, 1991).

MATERIALS AND METHODS

The methods used in this study were substantially similar to those described in the previous paper (Pallas et al., this issue). Thus they will be described only briefly here.

Animals

We used three normal pigmented ferrets (*Mustela putorius furo*) for this portion of the study and compared them to the six lesioned animals described in the preceding paper. The animals used for filling of inferior collicular axons were normal females obtained as adults from Marshall Farms

(North Rose, NY). The lesioned animals were offspring of dams shipped to us 2 weeks prior to parturition.

Neonatal surgery

Retinal axons can be induced to innervate the MGN by a set of specific lesions performed within 24 hours of birth in ferret kits. The procedure involves unilateral cautery of the occipital cortex, complete ablation of the superior colliculus (SC), and sectioning of the brachium of the inferior colliculus (BIC) in ferrets anesthetized by hypothermia. The cortical lesion causes partial retrograde degeneration in the lateral geniculate nucleus (LGN) and, together with the SC lesion, removes or reduces the normal targets of the retina. The BIC lesion deafferents the MGN, allowing the retina to terminate there. Following the ablations, the kits were revived and returned to the dam for rearing to adulthood. There was no neonatal surgery performed on normal animals.

Horseradish peroxidase injections

Individual inferior collicular axons in MGN were examined by the horseradish peroxidase (HRP) bulk-filling method (Mason, 1982; Sretavan and Shatz, 1984, 1986; Hahm et al., 1991). Briefly, animals were deeply anesthetized with sodium pentobarbital and perfused transcardially with 1 liter of cold (4°C), oxygenated, artificial cerebrospinal fluid. The brain was then quickly removed, and the inferior colliculi were exposed to view. Small crystals of HRP (Type VI; Sigma, St. Louis, MO) were introduced into the brachium of the inferior colliculus caudal to the MGN (Fig. 1). The brain tissue was then maintained in oxygenated, artificial cerebrospinal fluid at room temperature for 3–4 hours and subsequently fixed by immersion in 1% paraformaldehyde/2% glutaraldehyde for 2 hours, followed by 30% sucrose in phosphate buffer. The tissue was embedded in albumin-gelatin and sectioned frozen at 100 µm in the sagittal plane. Sections were treated with diaminobenzidine to visualize the HRP reaction product (Adams, 1981).

Axon reconstruction

Well-filled and well-isolated axons were identified with axon trunks extending into the BIC for IC axons and into the optic tract (OT) for retinal axons. The completeness of filling could be assessed by noting that small branches were darkly stained and did not fade out at their terminations. Individual axons were serially reconstructed with the aid of a camera lucida. Most axons were reconstructed with a ×100 objective; the largest arbors were reconstructed with a ×63 objective. In cases in which details of the arbor were ambiguous, we used differential interference contrast microscopy to increase resolution. We analyzed both the branched retino-MGN and the inferior collicular axons quantitatively, but measurements taken from unbranched axons were limited to axon diameter. Quantitative analysis of axon diameter was done by measuring from the first branch of the arbor, taking measurements every 20 µm or until 140 µm had been covered, and then averaging the values. Arbor area was calculated from a digitizing tablet by drawing a perimeter closely around the outermost boutons, and arbor volume was estimated by multiplying arbor area by the number of 100 µm sections in which the axon was contained. Arbor symmetry was measured by calculating the ratio between the dorsoventral extent of the axon arbor and its rostrocaudal extent. Individual boutons were counted to arrive at the value for bouton number, and bouton

Abbreviations

AI	primary auditory cortex
BIC	brachium of the inferior colliculus
BIN	nucleus of the BIC
CP	cerebral peduncle
IC	inferior colliculus
LGN	lateral geniculate nucleus
MGd	dorsal nucleus of the MGN
MGm	magnocellular nucleus of the MGN
MGN	medial geniculate nucleus
OT	optic tract
SC	superior colliculus

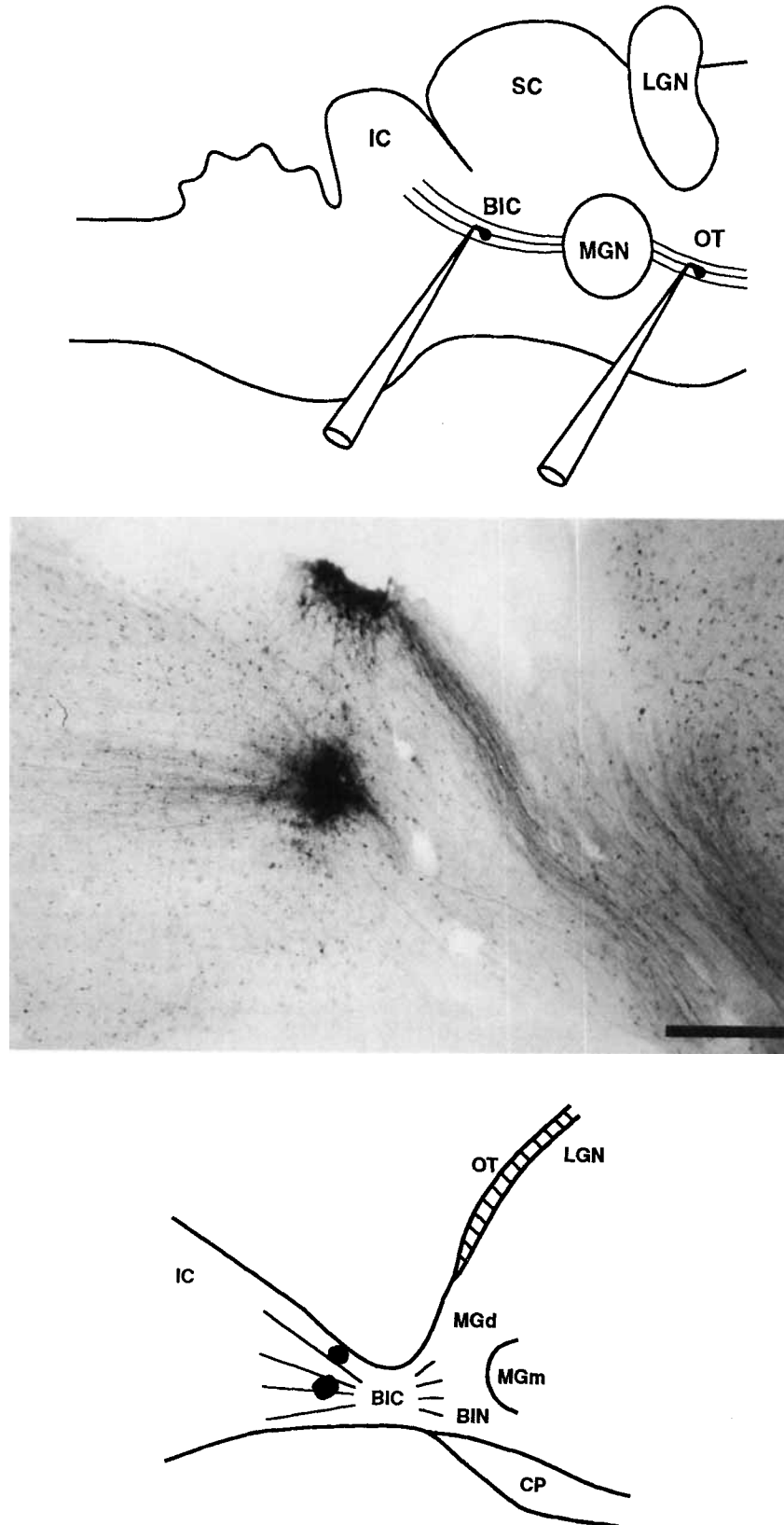


Fig. 1. Method for bulk-filling retinal and inferior collicular axons. At the top is a schematic showing where horseradish peroxidase (HRP) injections were made, within the optic tract for the retinal axons and within the BIC for the IC axons. In the center is a photomicrograph of

two injection sites in the BIC in a normal ferret. A low-power schematic showing the location of the injection sites is shown at the bottom. Scale bar = 200 μ m.

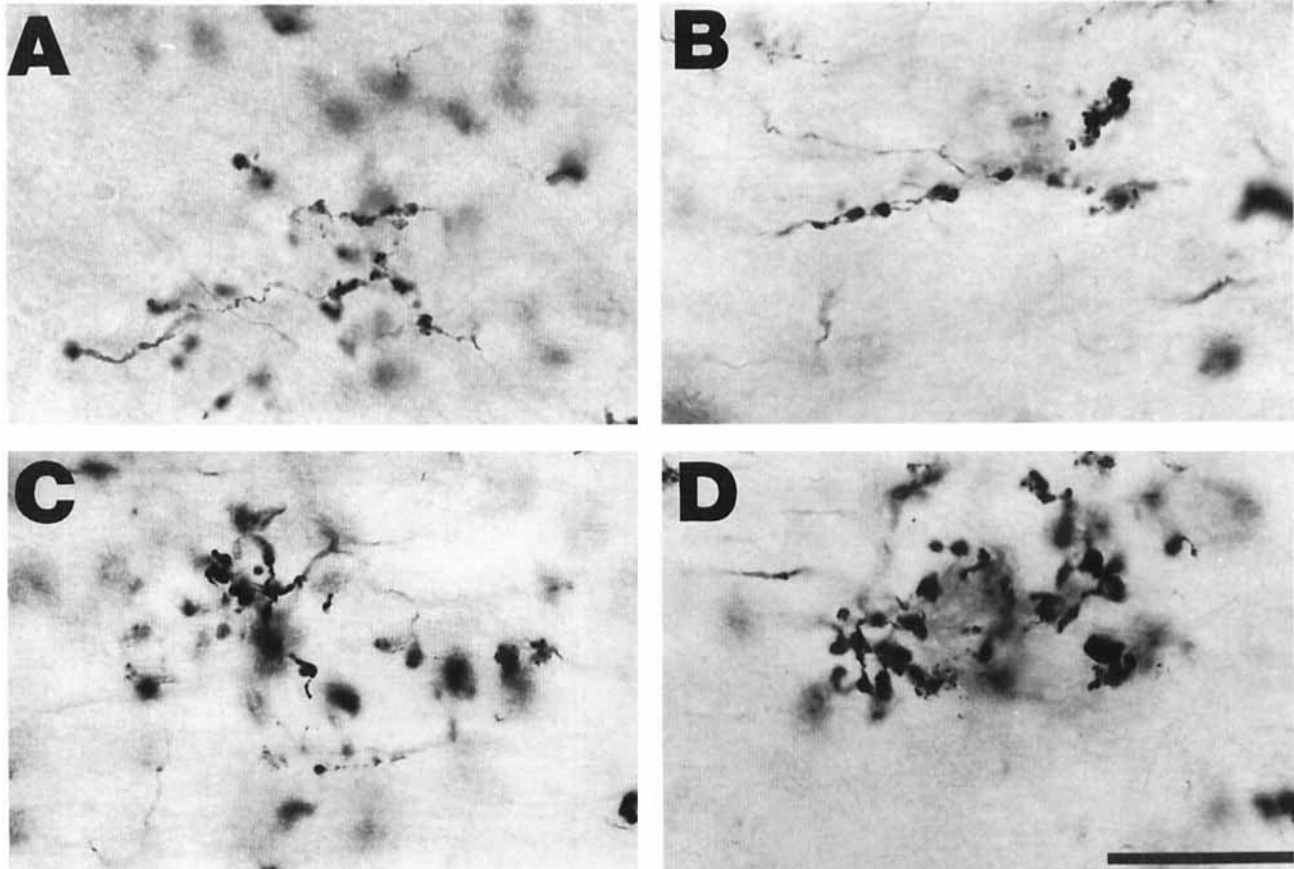


Fig. 2. **A–D:** Photomicrographs of inferior collicular axons arborizing in the MGN. Dorsal is up and anterior is to the right in each panel. These axons are in the same approximate location as the axons in Figure 4. The arbors often had elaborate bouton structures, arranged

mainly in strings of small boutons (such as in A,B) or in clusters with large boutons (C,D). These are 100- μ m-thick sagittal sections. Scale bar = 50 μ m.

density was derived simply by dividing number of boutons into arbor volume. Population means were compared with *t* tests, with *n* equal to the number of axons in each group. Because we have not rounded off the averages, extra decimal places in the values should not be taken to reflect actual resolution.

RESULTS

We have reconstructed a total of 12 IC-MGN axons. These were compared both qualitatively and quantitatively to the sample of 26 branched retino-MGN axons from the preceding paper (Pallas et al., this issue). We also compared the IC-MGN axons qualitatively to the unbranched retinal axons seen in the MGN (Pallas et al., this issue), but quantitative comparisons with the unbranched axons were limited to measurements of axon diameter.

Qualitative observations

Axons from the inferior colliculus enter the MGN primarily via the brachium of the inferior colliculus caudal to the MGN. The individual boutons of IC axon arbors (Fig. 2) are similar to those of retinal ganglion cell axon arbors in the MGN in a number of respects. These boutons can be quite large and complex and are often arranged in strings of small

boutons (Fig. 2A,B) or clusters with very large boutons (Fig. 2C,D).

Axons terminating in the MGN did not appear to have collateral branches to other destinations. All reconstructions were done in the sagittal plane in sections roughly parallel to the isofrequency laminae (Morest, 1965a; Pallas et al., 1990). We have included camera lucida reconstructions of all the IC-MGN axons in this study to illustrate the variability they exhibit, and because such axon arbors have not been previously described in the literature (but see Wenstrup et al., 1994). The groupings of the axons in the figures are somewhat arbitrary; they are loosely arranged by their similarity.

Most of the axons (seven of the 12) had their arbors elongated along the rostrocaudal dimension as seen in sagittal section (Figs. 3, 4). The others were fairly uniform in their two-dimensional shape. Most had long axon trunks branching far into the nucleus anteriorly. They varied quite a bit in their complexity.

Figure 3 shows two axons with very simple arbors and long primary branches bearing only en passant boutons. These collaterals extended far from the more highly branched part of the arbor. Differential interference-contrast microscopy confirmed that they were part of the

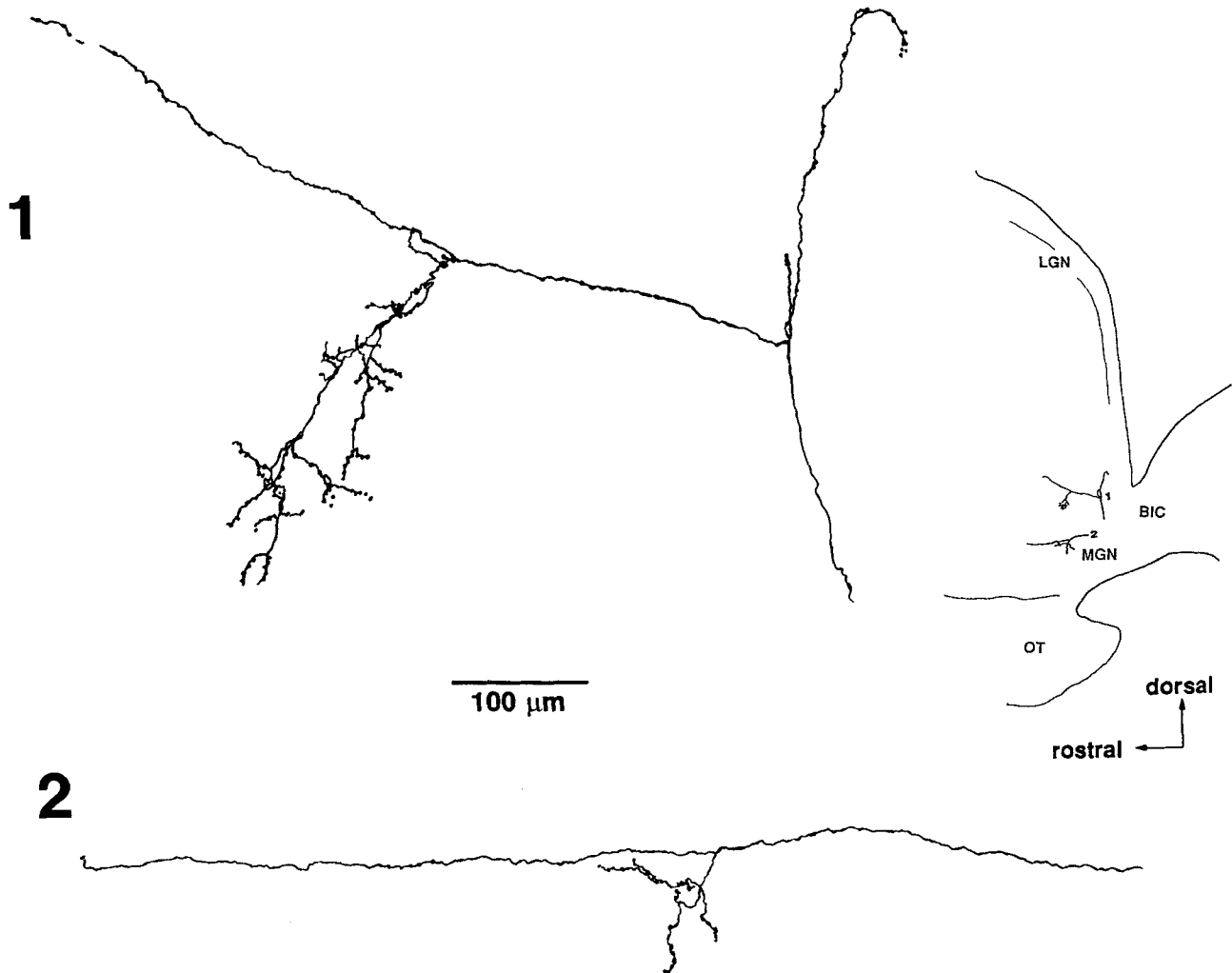


Fig. 3. **1,2:** Camera lucida reconstructions of two IC-MGN axons. The **inset** shows the approximate location and size of the arbors within the MGN. Here, the brachium of the inferior colliculus (BIC) is to the right. The axons are elongated in the rostrocaudal dimension in

contrast to the retino-MGN arbors shown in the accompanying paper (Pallas et al., this issue). These two arbors were the simplest with respect to number of branches. All reconstructions were made from 100- μ m-thick serial sagittal sections.

same arbor. Such collaterals were not seen on the branched retinal ganglion cell axons in MGN.

The axons shown in Figure 4 were somewhat more highly branched, with the exception of the first. The trunk of axon 1 could not be followed as far as the axon trunks of the other axons. It branched close to the BIC and thus became obscured by other axons. Axon 2 had long, unbranched collaterals, with en passant boutons, as can be seen on the axons depicted in Figure 3. Axons 3 and 5 had similar but shorter collaterals of this type. The axonal arbors were elongated rostrocaudally.

Figure 5 illustrates reconstructions of five more IC-MGN axons. They have been grouped together in the figure, because they had more symmetrical, clustered arbors than the axons depicted in Figures 3 and 4. They were also more highly branched in general, and the individual boutons were sometimes quite large (axon 3) and numerous.

An interesting feature of many of the IC-MGN axons (axon 1 in Fig. 3, axons 1–3 in Fig. 4, and axons 3–5 in Fig. 5) is that they tended to have two or three very long primary

branches. This was not as common in the branched retino-MGN axons or in the presumed W axons in the LGN and SC if they are compared on the same scale as the IC-MGN axons. The branches often came together again where the main part of the arbor was elaborated, so that the branches did not serve to spread the arbor over a larger area.

We noted differences between the population of branched retino-MGN arbors and the IC-MGN arbors, mainly in their complexity and extent. The retino-MGN arbors were generally much simpler (less branched) and smaller in overall extent. They did not show the rostrocaudal lengthening seen in most of the IC-MGN arbors. Our previous paper (Pallas et al., this issue) gives a detailed account of the retino-MGN axons.

Quantitative observations

We made a number of quantitative measurements on our data to demonstrate more clearly the differences and similarities between the two populations of axons. We compared

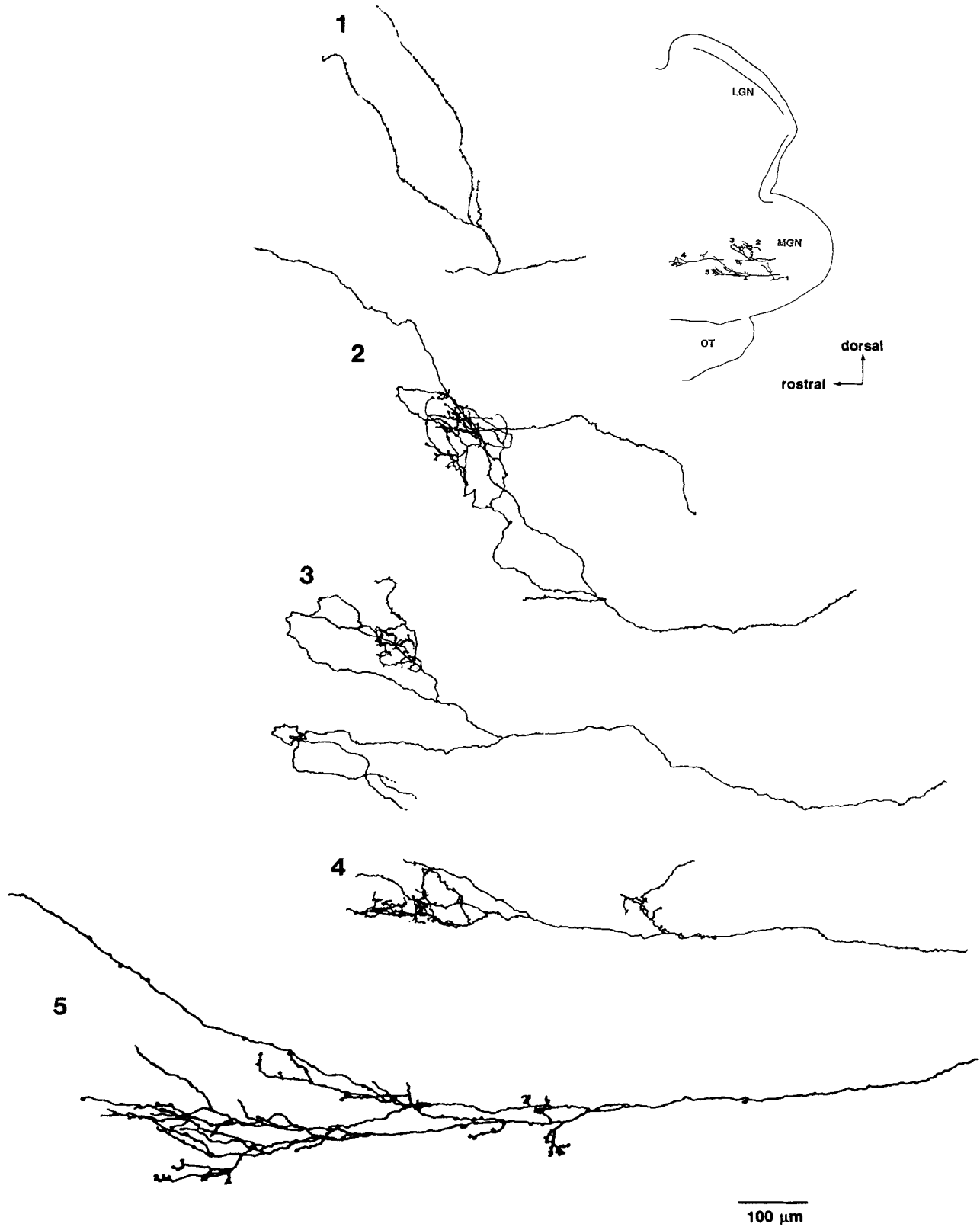


Fig. 4. 1-5: Other IC-MGN axons showing their rostrocaudally elongated axons. In this figure, as in Figure 3, there are long collaterals extending away from the main portion of the arbor that have only en passant boutons. The arbors are quite large (note the scale bar),

especially in comparison with the retino-MGN arbors (Pallas et al., this issue). The **inset** shows the approximate location and size of the arbors with respect to the MGN.

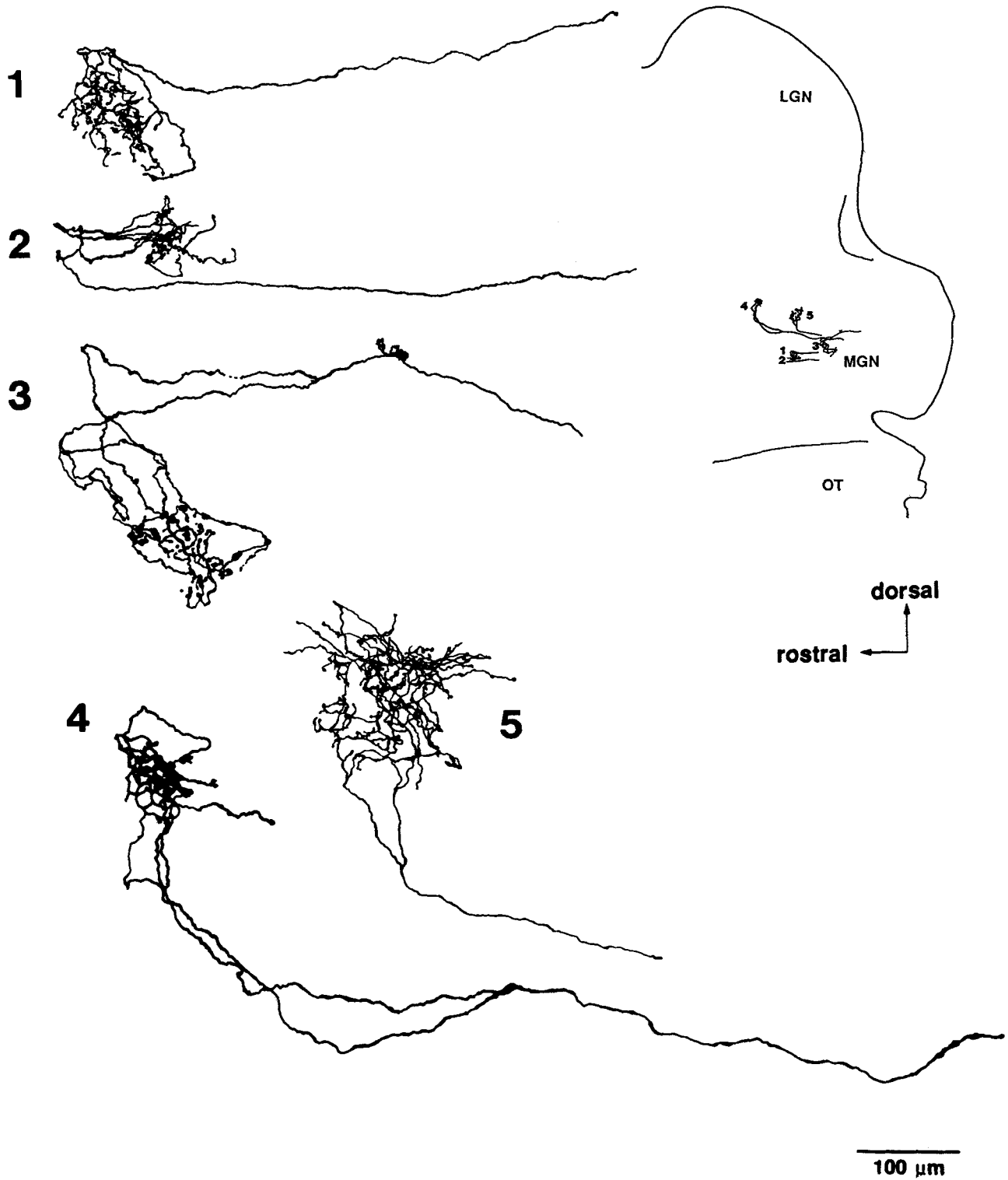


Fig. 5. 1-5: Several more examples of IC-MGN axon arbors. These are somewhat more highly branched than the ones in Figures 3 and 4 and have larger, more complex boutons.

the IC-MGN axon sample to the branched retino-MGN axons (with the exception of measurements of axon diameter, which also included the unbranched retino-MGN

axons). These measurements included axon diameter, arbor volume, arbor symmetry, bouton number, and bouton density.

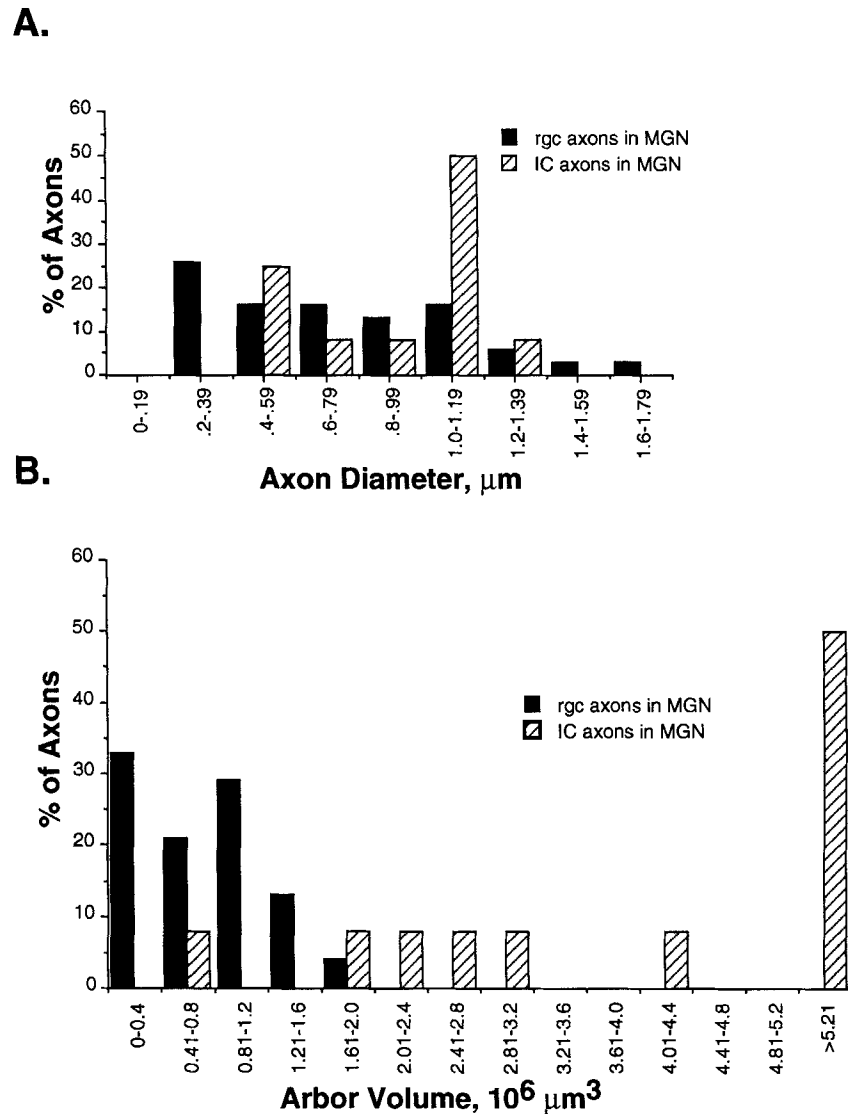


Fig. 6. Quantitative comparison of axon diameter (A) and arbor volume (B) in retino-MGN and IC-MGN axons.

Axon diameter. The values given here for axon diameter result from averaging measurements starting from the distal end of the axon trunk (see Materials and Methods). The trunks of the IC axons were quite fine, even for those axons having very large arbors. The mean axon diameter for the IC-MGN axons was $0.90 \mu\text{m}$ (SE = 0.09, $n = 12$; Fig. 6A). This is not significantly different ($P > 0.21$) from the population of retino-MGN axons (Pallas et al., this issue), which had an average diameter of $0.74 \mu\text{m}$ (SE = 0.07, $n = 31$).

Arbor volume. Our qualitative observation that IC axons had much larger arbors in MGN than did retinal arbors was supported by quantitative comparison (Fig. 6B). Our method for measuring arbor volume from two-dimensional sections (see Materials and Methods) will lead to overestimates of volume and underestimates of bouton density, but, because we use the data for comparative purposes with other arbors measured in the same way, it is unlikely to introduce

substantial biases in our interpretation. There were 26 retinal axons that had arbors in MGN. They had a mean arbor volume of $0.72 \times 10^6 \mu\text{m}^3$ (SE = 0.09; Pallas et al., this issue). The 12 IC-MGN arbors had a much larger mean arbor volume of $7.34 \times 10^6 \mu\text{m}^3$ (SE = 2.28). Statistical comparison shows that the populations are significantly different ($P < 0.0001$).

Arbor symmetry. As was mentioned in the section concerning our qualitative observations, we noted that many of the IC-MGN arbors were elongated rostrocaudally. To quantify this impression, we measured the extent of the arbor along both the rostrocaudal and the dorsoventral axes and calculated the ratio of dorsoventral extent to rostrocaudal extent (DV/RC extent; Fig. 7). For the IC-MGN axons, we found a mean DV/RC ratio of 0.77, with a range of 0.14 to 1.59 (SE = 0.127, $n = 12$) indicating that, on average, the axons were in fact elongated rostrocaudally. The retino-

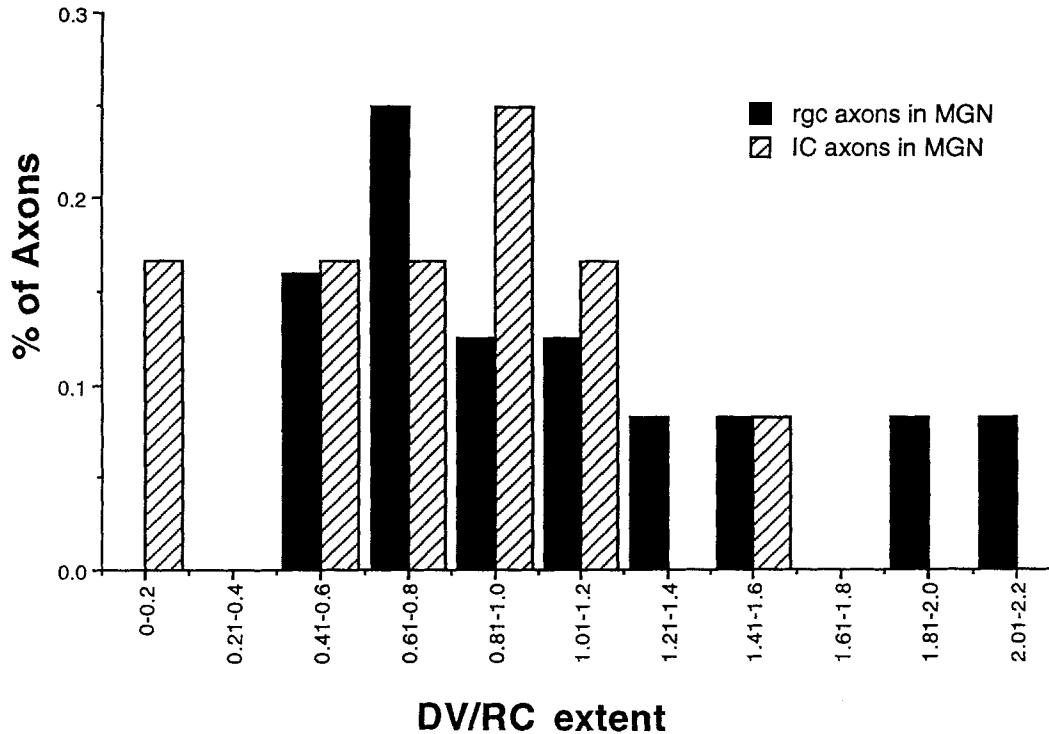


Fig. 7. Arbor symmetry. We calculated the dorsoventral (DV) and rostrocaudal (RC) extents of each axon arbor and calculated the ratio of DV extent to RC extent. Numbers less than one indicate rostrocaudal elongation. We find a significant tendency for elongation of IC arbors in MGN, but, on average, the retino-MGN arbors are more symmetric.

MGN arbors, on the other hand, were more symmetric, with an average DV/RC ratio of 1.09 (range 0.49 to 2.18, SE = 0.106, $n = 24$). These values for the two populations of axons are significantly different (t test, $P < 0.05$).

Bouton number. Given the large difference in arbor volume, it was not surprising to find that IC-MGN axons had significantly more boutons (mean = 187, SE = 26.7, $n = 12$) than retino-MGN axons (mean = 89, SE = 17.9, $n = 24$, $P < 0.004$). The comparison is presented in Figure 8A. A plot of bouton number vs. arbor volume shows very little overlap between the two populations (Fig. 8B). Although there is less variation in arbor volume of retino-MGN axon arbors compared to IC-MGN axon arbors, the retino-MGN axons show even more variation in the number of boutons than do the IC axons. There was no correlation between the number of boutons and the arbor volume for either population.

Bouton density. Interestingly, though the IC axon arbors were larger, were more elongated, and had higher numbers of boutons, inspection of the bouton density data (Fig. 9) shows that the small arbor volumes of the retinal axons result in higher bouton densities compared to the IC axons ($P < 0.0007$). The mean bouton density for the IC axons was 0.46 boutons per $10^6 \mu\text{m}^3$ (SE = 0.10, $n = 24$). For the retinal axons, the mean bouton density was 1.63 boutons per $10^6 \mu\text{m}^3$ (SE = 0.22, $n = 24$). Thus, retinal axons in MGN have a greater number of boutons per unit volume in their novel target than do the normal input fibers from the IC.

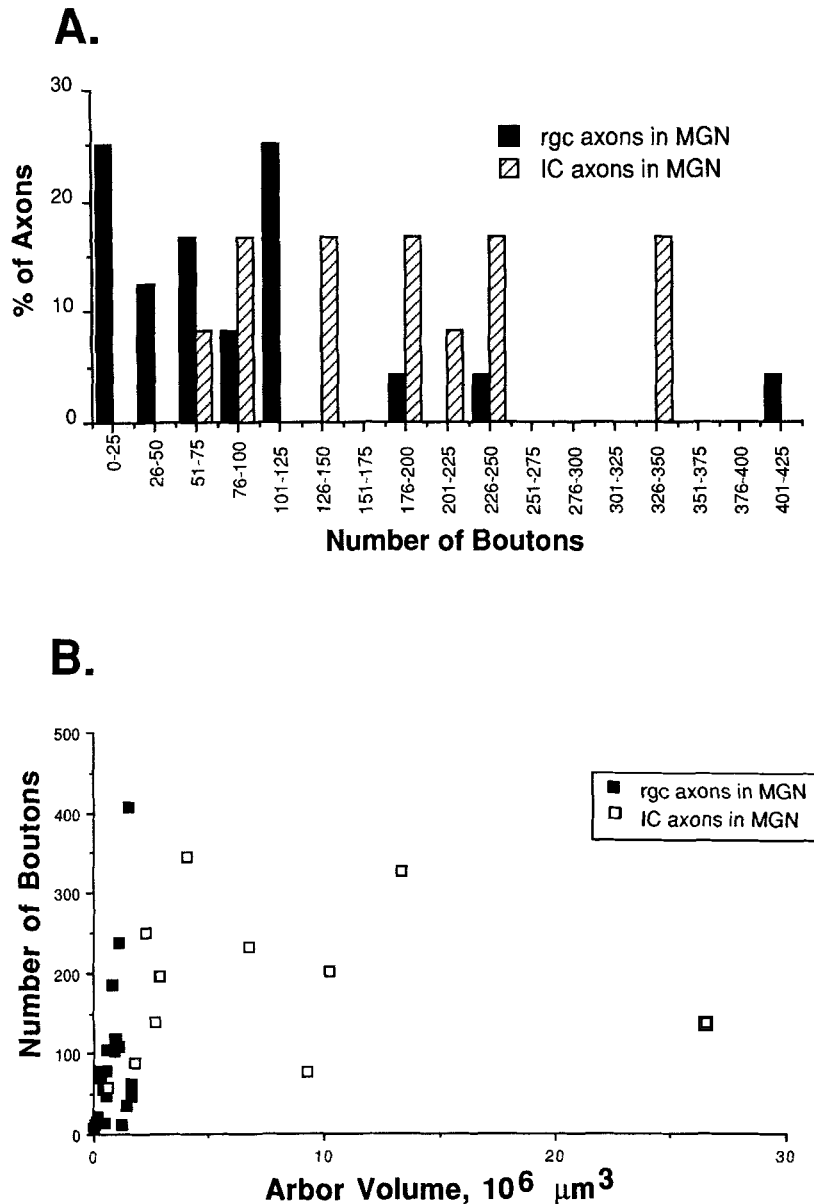
DISCUSSION

The results of this study indicate that, at least in the ferret MGN, arbor morphology is somewhat independent of target identity. Although individual boutons and clusters of boutons, as well as axon diameter, appear similar between retinal axons in the rewired MGN and IC axons in the normal MGN, the number and arrangement of boutons and the size and complexity of the axon arbors show striking differences. In particular, our results demonstrate that branched retinal arbors are much more spatially restricted in the MGN than are IC arbors, especially along the rostrocaudal axis, with smaller arbor volumes and fewer boutons but higher bouton density.

Technical considerations

As is discussed in the accompanying paper (Pallas et al., this issue), filling axons by bulk uptake of HRP rather than by intracellular injection causes more difficulty in reconstructing individual axons in their entirety. We have taken special care to ensure that each axonal process is traced to its termination and to use at least ten points to align adjacent serial sections. Differential interference-contrast optics also aided significantly in following the extremely fine branches characteristic of these axons.

We injected the BIC rather than the IC in order to use a shorter survival time in vitro and thus ensure that the axons we filled were healthy. Therefore, we must consider the possibility that we filled axons other than IC axons.



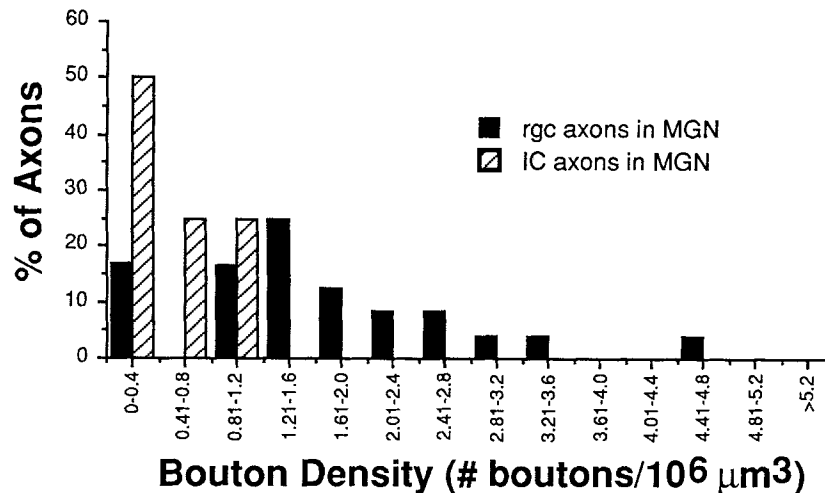


Fig. 9. Comparison of bouton density for the two axon populations. The small arbors of the retino-MGN axons result in high bouton densities, even compared to the much larger IC-MGN axons.

It should be noted that the actual identity of the normal population of MGN afferents is not critical to a comparison of the morphology of the retinal axon arbors induced to project to MGN with the morphology of normal input populations in the MGN. Other investigators who wish to use our data as an example of tectothalamic axon morphology should keep in mind, however, that the identity of the axons is not determined with certainty. This would require a morphological study of physiologically identified fibers.

Thalamic location of filled axon arbors

The details of the ferret auditory system appear to be quite similar to those of the cat (Kelly et al., 1986, 1989; King and Hutchings, 1987; Phillips et al., 1988; Brunso-Bechtold et al., 1990). Although to date there has been no study, of which we are aware, specifically describing the tectothalamic projections in the ferret or the fine structure of the MGN, we believe that it is reasonable for the purposes of this paper to assume that these structures are also similar to those in the cat.

The central nucleus of the IC in the cat projects bilaterally to the ventral, medial, and deep dorsal divisions of the MGN and to the lateral division of the posterior group of the thalamus (Morest, 1965a,b; Andersen et al., 1980a). The organization is similar in other mammals (Tarlov and Moore, 1966; Carey and Webster, 1971; Wenstrup et al., 1994) and is likely to be similar in the ferret. Thus, in order to determine in which part of the MGN our axons terminate, we have compared our unstained sections, under darkfield illumination, to structures in the cat brain (Berman and Jones, 1982) as well as to Nissl-stained sections of the ferret brain. (We did not counterstain the tissue, because we needed to have repeated access to the axons, and counterstaining makes it difficult to follow the fine-caliber axons or to find them repeatedly.) We restricted our reconstructions to the ipsilateral MGN, where we were more likely to encounter well-filled fibers. Although we lack the connective and physiological data that would prove the point, it appears that the axons we reconstructed reside mainly in the ventral, and some in the medial, subdivision of the MGN. Importantly, the location of the IC-MGN axon

arbors we describe was similar to the location of retino-MGN arbors as described in the accompanying paper.

Comparison with other studies

To our knowledge, no previous study has described the morphology of individually reconstructed afferent axons in the MGN of any mammal. Other investigators have used both the Golgi method and bulk filling with HRP to examine the morphology of populations of afferents to the MGN. Morest (1964, 1965a), using the Golgi technique in the cat, reported that the ascending input fibers from the IC are arranged in laminar fashion, as are their target cells. Fibers had axon trunks of 1–2.5 μm diameter, and their extensive terminations formed knob-like structures around the somata. The axons we reconstructed were of finer diameter than those seen by Morest, but this is perhaps not surprising, considering the fact that we are using a different technique in a different animal. Majorossy and Réthelyi (1968) and Majorossy and Kiss (1976) found grape-like terminal expansions on IC arbors in the cat MGN. Wenstrup et al. (1994), working in an insectivorous mammal, the mustache bat, found mainly complex terminal arborizations with many club-like endings in the ventral MGN. These authors also noted that the axons were organized in laminar fashion.

One difficulty in making comparisons with our data is that most of these authors have examined the afferent fibers only in the coronal plane, perpendicular to the isofrequency laminae, whereas we have sectioned in the sagittal plane in order to include more of an isofrequency lamina and, hence, an individual axon in one section. The laminar arrangement of the axons is less clear in our material due to our sectioning parallel to the laminae. However, several common features can be found in our reconstructed axons. Our normal IC-MGN axons were elongated within the isofrequency (anteroposterior) plane. In addition, they showed many large, club-like endings as well as grape-like clusters of terminals and occasional strings of en passant boutons. These similarities suggest a common origin and indicate that there is considerable

conservation of terminal morphology between ferrets and other mammals.

Control of axon arbor morphology

We found that the morphology of branched retino-MGN arbors in rewired animals was quite distinct from that of IC-MGN arbors in normal animals. This result suggests a considerable degree of control by afferents in specifying arbor morphology, at least in the MGN.

There have been several other studies in support of afferent-directed control of synaptic morphology. Scalia (1987) redirected optic axons toward the olfactory region in adult frogs and found that the optic axons formed terminal plexuses in the olfactory cortex as well as several visual structures. The ultrastructural appearance of these terminals was the same regardless of the identity of the target structure. Similarly, Cantore and Scalia (1987) studied optic axons that entered the lateral thalamic neuropil in response to ablation of their normal target in the optic tectum. The terminals formed by these redirected retinal axons had the same ultrastructural morphology as in their normal target. On the light microscopic level, Graziadei and his colleagues (Graziadei et al., 1979; Graziadei and Monti Graziadei, 1985) found that olfactory axons whose normal target had been ablated early in development terminate in abnormal locations in the forebrain, where they nevertheless form the glomerular structures typical of their normal termination site.

In contrast, the synapses made onto granule cells in the cerebellum are similar regardless of the source of the input (Palay and Chan-Palay, 1974). In cats, individual Y axons from the retina appear to form different types of terminals in the superior colliculus (Behan, 1981) than they do in the LGN (Wilson et al., 1984). Campbell and Frost (1987, 1988), working with the hamster, found differences between the ultrastructure of normal retinal axon terminals in the lateral geniculate nucleus and superior colliculus and terminals of retinal axons induced to innervate either the medial geniculate nucleus or the ventrobasal nucleus of the thalamus.

These results are difficult to integrate into a unified framework. One reason is that it is not clear which aspects of a synapse or terminal arbor are the appropriate ones for comparison, and different investigators often use different measurements. Furthermore, it cannot always be assumed that either target cells or afferent axons are homogenous in any one area of the brain. For example, the retinal inputs to X and Y cells in the LGN differ in synaptic morphology and location (Wilson et al., 1984), but the identity of the retinal afferents differs in the two cases as well, with X axons innervating X cells in the LGN and Y axons innervating Y cells (see Sherman and Spear, 1982, for review). Thus, what appears to be evidence for target control may also be interpreted as evidence for afferent control.

In this study, we have very clearly changed the targets of the retinal ganglion cells, and thus we can study terminal arbor morphology of the same axons in very different targets. Although the studies of Campbell and Frost (1987, 1988) in the hamster are similar, these authors did not identify the source of their retinal ganglion cell axons by class, and there is now evidence for several types of retinal axons in the hamster (Erzurumlu et al., 1988). In the ferret, retinal ganglion cells and their axons can be clearly distinguished both physiologically and morphologically as X, Y, or W (Roe et al., 1993). Although the arbor morpholo-

gies (arbor volumes and bouton numbers) of both the branched and unbranched retinal axons in the MGN are different from those of the normal inputs from the IC, the branched retino-MGN axons (but not the unbranched axons) resemble normal retinal-W axons closely (Pallas et al., this issue; see also Roe et al. 1993). This suggests that at least certain features of axons are relatively unaffected by the identity of their target and point to a role for intrinsic determinants in regulating these features.

Topography

Roe et al. (1991, 1993) have recently shown that the MGN in rewired ferrets contains at least a rough two-dimensional map of the retina. The present study was undertaken in part to determine whether the visual map in MGN had its anatomical basis in focal projections of retinal ganglion cell axons to the MGN.

The unbranched retino-MGN axons travel across wide areas of the MGN and would not be expected to support a topographic map of visual space. However, our measurements of axon arbor volume and symmetry have shown that branched retinal arbors in the MGN are much more restricted than are the BIC inputs to MGN (Fig. 10). The normal inputs, which we assume are from the IC, are often elongated rostrocaudally. The isorepresentational (isofrequency) plane in normal MGN runs roughly rostrocaudally and dorsoventrally (Andersen et al., 1980b; Middlebrooks and Zook, 1983; Imig and Morel, 1985; Pallas et al., 1990). We would like to suggest that this difference in arbor morphology provides a substrate for a two-dimensional map of visual space in MGN of the rewired ferrets.

We have shown that topography along one dimension of the retino-MGN projection is not maintained in the projection from MGN to primary auditory cortex (AI), even though there is a two-dimensional retinotopic map in AI (Pallas et al., 1990; Roe et al., 1990). The thalamocortical projection is overlapped along the isofrequency dimension in both normal and rewired ferrets (Pallas et al., 1990), perhaps because it is formed before our rewiring lesions (Jackson et al., 1989; McConnell et al., 1989; for reviews, see Pallas, 1990; Sur et al., 1990). As a result, we have postulated that the elevational dimension of the map must be resynthesized in AI. The presence of retinotopically correlated activity in the MGN is likely to be the source of information for this resynthesis.

We noted with interest that the IC arbors are not usually as extensive dorsoventrally as they are rostrocaudally. Andersen et al. (1980a) found that injections of the IC with anterograde tracers produced curved sheets of label in MGN which extended both dorsoventrally and rostrocaudally. Adjacent sheets represented different frequencies, with high frequencies represented medially and low frequencies laterally. The fact that the individual IC arbors are not as extensive as the population may indicate another level of organization within the isofrequency plane. This may correspond to binaural organization (Middlebrooks and Zook, 1983). Alternatively, or in addition, Langner and Schreiner (1989, 1990) have shown physiologically that there is organization according to best modulation frequency (AM rate) orthogonal to the tonotopic dimension in the IC of the cat. The dorsoventrally restricted arbors of the individual IC axons may be the anatomical manifestation of this organization.

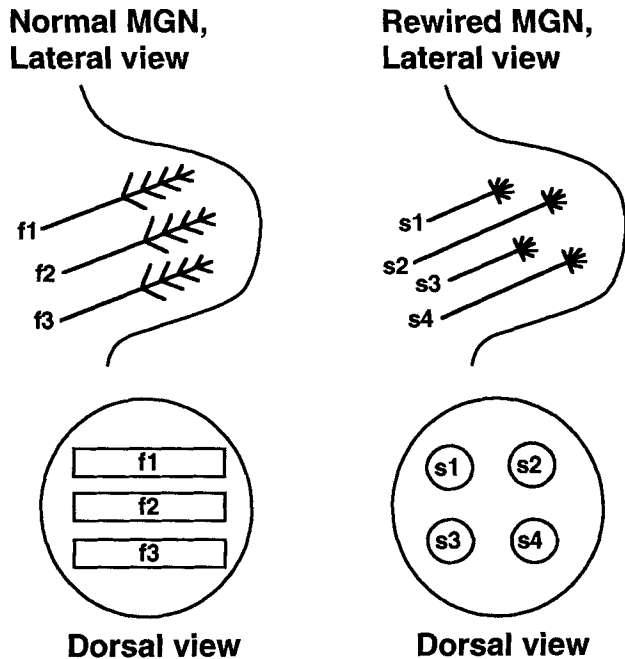


Fig. 10. Schematic showing the hypothesized topography of the two axon populations. Most of the IC-MGN axons are elongated in the isofrequency plane (f1–3 represent different isofrequency laminae) as shown in this diagram of a sagittal section through the MGN. In contrast, the retino-MGN axons are smaller and more symmetric. The more focal retinal axon arbors may provide the basis for a retinotopic map in the MGN of rewired ferrets, with each axon representing a different spatial (s) location. The IC-MGN axons would actually enter the nucleus from the right (from the BIC) rather than from the left, as they are drawn for comparative purposes with the retino-MGN axons, which enter as shown from the optic tract.

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