

Research report

The associative striatum: cortical and thalamic projections to the dorsocentral striatum in rats

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Abstract

Cortico-striatal projections to the dorsocentral striatum (DCS) were investigated using retrograde fluorescent axonal tracing. The DCS is of interest because of its role in directed attention and recovery from multimodal hemispatial neglect following cortical lesions of medial agranular cortex (AGm), an association area that is its major source of cortical input. A key finding was that the multimodal posterior parietal cortex (PPC) also contributes substantial input to DCS. This is significant because PPC and AGm are linked by corticocortical connections and are both critical components of the circuitry involved in spatial processing and directed attention. Other cortical areas providing input to DCS include visual association areas, lateral agranular cortex and orbital cortex. These areas also have reciprocal connections with AGm and PPC. Less consistent labeling was seen in somatic sensorimotor areas FL, HL and Par 1. Thalamic afferents to DCS are prominent from the intralaminar, ventrolateral, mediodorsal, ventromedial, laterodorsal (LD) and lateral posterior (LP) nuclei. Collectively, these nuclei constitute the sources of thalamic input to cortical areas AGm and PPC. Nuclei LD and LP are only labeled with injections in dorsal DCS, the site of major input from PPC, and PPC receives its thalamic input from LD and LP. We conclude that DCS receives inputs from cortical and thalamic areas that are themselves linked by corticocortical and thalamocortical connections. These findings support the hypothesis that DCS is a key component of an associative network of cortical, striatal and thalamic regions involved in multimodal processing and directed attention.

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Abbreviations: ac, anterior commissure; AD, anterodorsal nucleus; AGI, lateral agranular cortex; AM, anteromedial thalamic nucleus; AV, anteroventral thalamic nucleus; cAGm, medial agranular cortex, caudal part; cg, cingulum bundle; Cg, cingulate cortex; CL, central lateral thalamic nucleus; CM, central medial thalamic nucleus; DCS, dorsocentral portion of the dorsal striatum; FL, forelimb area of cortex; fm, forceps minor; fr, fasciculus retroflexus; Fr1, frontal cortex, area 1; Fr2, frontal cortex, area 2; HL, hindlimb area of cortex; IAM, interanteromedial thalamic nucleus; ic, internal capsule; LD, laterodorsal thalamic nucleus; LH, lateral hypothalamic area; LO, lateral orbital cortex; LP, lateral posterior thalamic nucleus; LPM, lateral posterior thalamic nucleus, medial portion; MD, mediodorsal thalamic nucleus; MO, medial orbital cortex; mt, mammillothalamic tract; Oc2L, occipital cortex, area 2, lateral part; Oc2M, occipital cortex, area 2, medial part; Par 1, parietal cortex, area 1; Par 2, parietal cortex, area 2; PC, paracentral thalamic nucleus; PF, parafascicular thalamic nucleus; Po, posterior thalamic nuclear group; PPC, posterior parietal cortex; PRh, perirhinal cortex; PT, paratenial thalamic nucleus; PV, paraventricular thalamic nucleus; PVA, paraventricular thalamic nucleus, anterior part; rAGm, medial agranular cortex, rostral part; Re, reuniens thalamic nucleus; RSA, retrosplenial agranular cortex; RSG, retrosplenial granular cortex; Rt, reticular thalamic nucleus; sm, stria medullaris of the thalamus; Sm, submedial thalamic nucleus; VL, ventrolateral thalamic nucleus; VLO, ventrolateral orbital cortex; VM, ventromedial thalamic nucleus; VPL, ventral posterolateral thalamic nucleus; VPM, ventral posteromedial thalamic nucleus

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1. Introduction

The dorsocentral striatum (DCS) is defined as the major site of termination of corticostriatal inputs originating from the medial agranular cortex (AGm or Fr2) in rats [7,17,38,41]. In the region of the striatum between the level of the genu and that of the anterior commissure (ac), DCS constitutes a territory ~1 mm in diameter, centered in the dorsal striatum ~0.5–1.5 mm below the external capsule, approximately equidistant from the medial and lateral boundaries of the striatum [38]. Area AGm is a multimodal association premotor cortex with diverse cortical connections [39] and DCS has been implicated as a multimodal convergence region of striatum [33,38].

As noted by Parent and Hazrati [36], once it became clear that the striatum has sensorimotor, associational and limbic regions, it became important to determine the organizational subdivisions within these regions. Although association areas of the rat striatum have not been studied in the same detail as sensorimotor and limbic regions, several investigations have focused on striatal circuitry related to prefrontal cortical areas [5,6,20]. The study of association areas is of special interest because the influence of the striatum on motor function includes cognitive components [19] mediated by cortical areas like AGm. Thus, the associative striatum constitutes a substrate for the dynamic modulation of behaviors having a significant multimodal component involving association cortices.

The relationship between DCS and AGm is of particular interest because each of these regions is a major component of the neuronal circuitry mediating directed attention. Subsequent to the initial development of a rat model of hemispatial neglect [14,15], behavioral experiments identified AGm and the posterior parietal cortex (PPC) as being cortical areas critical for the normal functions of spatial orientation and directed attention [11,12,26,29,30]. In rats, lesions in either of these reciprocally interconnected [39,40] areas produce multimodal neglect, as does disconnection of their corticocortical connections without direct damage to AGm or PPC [9]. Spontaneous recovery from neglect resulting from cortical lesions occurs in some rats [51] and is correlated with re-establishment of symmetrical expression of immediate early genes and glutamate receptors in the dorsolateral striatum and DCS [48–50]. In addition, DCS is essential for spontaneous and pharmaceutical-induced recovery of function [46,47] and this points to the pivotal role of corticostriatal connections in directed attention.

Due to the critical role of DCS in the circuitry mediating directed attention and recovery from neglect, it is important to know which cortical areas are capable of influencing the activity of neurons in DCS. Thus, in the present study we sought to identify all the cortical inputs to DCS and to interpret this pattern with regard to the known corticocortical connections of these areas. Specifically, because of the close anatomical and functional relationship

between AGm and PPC, we wished to determine if cortical area PPC projects to DCS. In addition, we wanted to determine if the thalamic nuclei that project to DCS include those that project to the cortical areas that provide input to DCS. For these purposes we made injections of retrograde axonal tracers in DCS or along its margins. The results provide new information on corticostriatal and thalamostriatal topography and convergence, support the behaviorally derived hypothesis that DCS functions as a striatal association area that is critical for directed attention, and provide a foundation for experiments intended to promote recovery from neglect by therapeutic interventions.

2. Materials and methods

A total of 25 male Long-Evans Hooded rats were anesthetized with an intraperitoneal injection of a ketamine/xylazine cocktail (90 mg/kg:10 mg/kg) and placed in a stereotaxic device. Either Fast Blue (Sigma, 3% in H₂O) or Diamidino Yellow (Sigma, 3% in H₂O) was injected into the striatum. Both are frequently used retrograde axonal tracers which produce reliable labeling with little spreading of the injection site relative to other tracers like Fluorogold [10]. Injections were made via Picospritzer (General Valve) using two to three pulses of 20–30 p.s.i., 5–20-ms duration, or via a 33-g Hamilton syringe using a volume of 0.05–0.10 ml. Some animals received a second injection of the other tracer in the opposite hemisphere, but the present analysis involves only one tracer per animal. The injections in cases 93 and 94 were on the right side; all others were on the left. After a 3–5-day survival time, rats were injected i.p. with 3 ml chloropent (4.25% chloral hydrate, 0.9% pentobarbital) and perfused intracardially with 300 ml of potassium buffered saline (PBS) followed by 300 ml of 4% buffered paraformaldehyde. Brains were subsequently removed and stored in dilute fixative (0.4% paraformaldehyde) and cryoprotected by sinking in a 30% sucrose solution. Coronal sections were cut at 40 μ m on a freezing microtome and placed in dilute fixative until being mounted on slides. Three spaced series of sections were used for analysis: one for viewing on a fluorescent microscope, a second for cresyl violet staining for cytoarchitectural analysis, and a third for fluorescent photomicrography.

Three-dimensional reconstructions of tracer injection sites were produced by outlining areas of visible fluorescence and the boundaries of the striatum and cortex from fluorescent sections, transferring these sketches into vector drawings (Adobe Illustrator 10), then making three-dimensional models and rendering final images using an M5 imaging system (Imaging Research). Cases 93 and 94 are represented on the left for comparison purposes, even though their injections were located on the right side.

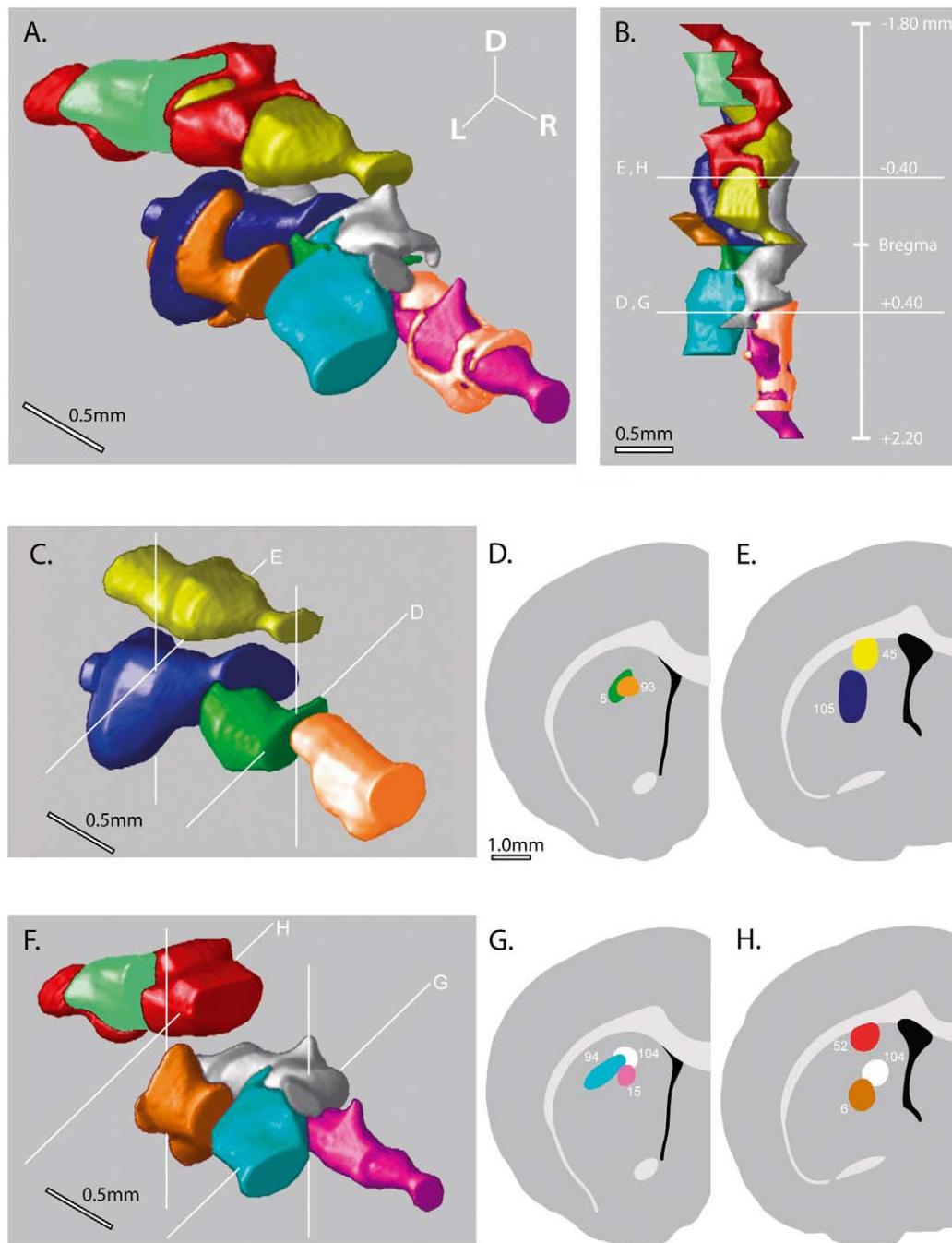


Fig. 1. Injection sites in DCS. (A) Three dimensional reconstruction of the ten principal injection sites seen from a lateral oblique perspective, to show their relative locations within the dorsal striatum. Stick diagram indicates dorsal, lateral and rostral directions and also applies to panels C and F. (B) Dorsal perspective of the same set of injections as in A, rostral toward the bottom. Vertical white line indicates rostrocaudal coordinates with respect to Bregma. Horizontal white lines indicate the plane of sections D,E and G,H. (C) Three dimensional renderings of the injection sites in the four cases discussed in detail in the Results. White lines indicate the planes represented by the corresponding two-dimensional depictions of the injection sites at two levels. (D,E) Location of injection sites for the four cases in C, plotted on two sections between the level of the genu and ac. Scale bar in D also applies to panels E, G and H. (F) Three dimensional reconstructions of the remaining six cases. White lines indicate the planes represented by the two-dimensional depictions in G and H. (G,H) Sectional views of the injections in F, at the same rostrocaudal levels as panels D and E. The injection for case 84 (light green in panel F) is not shown because it begins caudal to the level in H.

injections in dorsal DCS and these are presented separately below.

3.2. Retrograde labeling in cortex and thalamus

Here we provide detailed descriptions of the injection

sites and labeling patterns for four important cases, with the other cases discussed in reference to these.

3.2.1. Case 5: central DCS

The injection site for case 5 was centered 1 mm rostral

to the anterior commissure, at the level of the septum (Fig. 1). It was centered 0.7 mm ventral to the white matter, 1.5 mm lateral to the lateral ventricle, and 1.5 mm medial to the lateral boundary of the striatum. This represents an injection in the central portion of the AGm projection field in DCS. As discussed below, cases 93 and 105 represent rostral and caudal extensions (respectively) of the territory encompassed by case 5. The injection site was cylindrical and extended rostrally from its center to the level of the genu and caudally to the level of the ac. A slight track of fluorescence lining the pipette trajectory was seen in cortical area AGl on the centermost section, but this track does not include the white matter. There was no label in contralateral AGl at the level of the injection, indicating that the track did not influence the labeling pattern observed. In the cortex, retrogradely labeled cell bodies were present in several areas (Fig. 2, Table 1). Area AGm was labeled most intensely on sections greater than 1 mm rostral to the level of the ac commissure, in layers II/III and V (Fig. 2A–C). In all sections in which AGm was labeled, layer V was labeled most strongly. Areas VLO and LO were weakly labeled in layer V only. In areas PPC and Oc2M, label was only present in layer V and was most robust rostrally. In the thalamus, nuclei VL, VM, MD, the intralaminar nuclei (CM, PC, CL), and PF all contained labeled cells (Fig. 2D–F). Nucleus VL appeared more strongly labeled medially than laterally, but the other nuclei were labeled throughout.

The injection in case 94 overlapped that of case 5, but extended farther rostrally and lay somewhat lateral to case 5 (Fig. 1). Case 94 produced a broader distribution of cortical labeling (Table 1) that included all of AGm, somatic sensory and motor areas AGl, FL, HL, Par 1 and Par 2, and visual association areas Oc2M and Oc2L. The pattern of thalamic labeling was nearly identical to case 5 (Table 1).

3.2.2. Case 93: rostral DCS

The injection site in case 93 was centered at the level of the genu and represents a more rostral injection than case 5, but one that is also centered in DCS. The center of the injection was located 1.1 mm ventral to the white matter. The injection was oval shaped and spanned a distance of 1.1 mm in the dorsoventral plane, 0.7 mm in the mediolateral plane, and 1 mm in the rostrocaudal plane. There was a very faint fluorescent track visible along the trajectory the pipette followed through AGl. Contralateral AGl contained no labeled cells. No fluorescence was evident in the white matter. Multiple cortical areas were labeled by the striatal injection. Labeled cells were evident in layers II/III and V of rAGm (Fig. 3A,B). Label in layer V of AGm became sparser caudally and was limited to layer II/III by the level of the anterior commissure. The labeling in layer II/III of area AGm also became less intense caudal to the anterior commissure, but remained faintly visible throughout cAGm. Label was present in layer V of

area Cg at and caudal to the level of the ac. Labeled cells in area HL were limited to layer V and were present only in small numbers (Fig. 3B). Labeled cells in layer V of areas Oc2M, PPC, Par 1, Par 2, RSA and RSG were distributed evenly throughout their rostrocaudal extent (Fig. 3C,D). In the thalamus, label was observed in VL, VM, MD, the intralaminar group (CM, PC, CL), and PF with no observable topography (Fig. 3E–G).

The injection in case 15 was virtually identical to case 93, but extended slightly more rostrally and was somewhat more focal (Figs. 1C,D and 6C). Cortical labeling was most dense in areas AGm, PPC and orbital cortex. Labeling was dense in rAGm, sparse in cAGm. Thalamic labeling was similar to case 93, differing only in relative intensity of cellular labeling (Table 1).

3.2.3. Case 105: caudal DCS

The injection site for case 105 was centered at a level 0.5 mm rostral to the ac, and represents a location caudally adjacent to case 5 (Figs. 1 and 6A). The center of the injection was located 1 mm ventral to the white matter and 1.1 mm lateral to the lateral wall of the lateral ventricle. The injection measured 1.4 mm in the dorsoventral plane, 0.6 mm in the mediolateral plane, and 1.5 mm in the rostrocaudal plane (it extended from 0.5 mm caudal to the level of the genu to the level of the ac). There was no evidence of fluorescent tracer lining the path through which the pipette was inserted, nor was there any fluorescent material in the white matter. In the cortex, several areas contained labeled cells. Areas VLO, LO, and the rostral portions of AGm and AGl contained labeled cells in layers II/III and V (Fig. 4A,B). Area AGm remained labeled throughout its rostrocaudal extent and densely in cAGm, though layer V was no longer labeled caudal to the level of the ac. Labeled cells in area AGl were seen only until the level of the ac. Label was observed in area Cg at the level of the ac and continued to its caudal boundary. Areas HL, PPC, Oc2M, Par 1, Par 2, and PRh all contained labeled cells in layer V throughout their rostrocaudal extents (Fig. 4C,D). In the thalamus, nuclei AV, VL, VM, the intralaminar nuclei (CM, PC, CL), MD, PF, and rostral LP all contained labeled cells (Fig. 4E–G).

The injection in case 6 partially overlaps that of case 105 throughout most of its extent, but is centered more ventrally (Fig. 1; compare panels D and G). Cortical labeling included many of the same areas as in case 105, but AGm was labeled more intensely in rostral portions and somatic sensorimotor labeling did not include area HL. Thalamic labeling was very similar to case 105 with the addition of labeling in LD (Table 1).

Case 104 had an injection site located just medial to that of case 105 (Fig. 1). It also extended more rostrally than case 105. These two injections overlap partially in the middle of their a–p ranges. The pattern of cortical labeling for case 104 is similar to the pattern seen in case 105, with the addition of labeling in areas FL, Par 2, and Oc2L, and

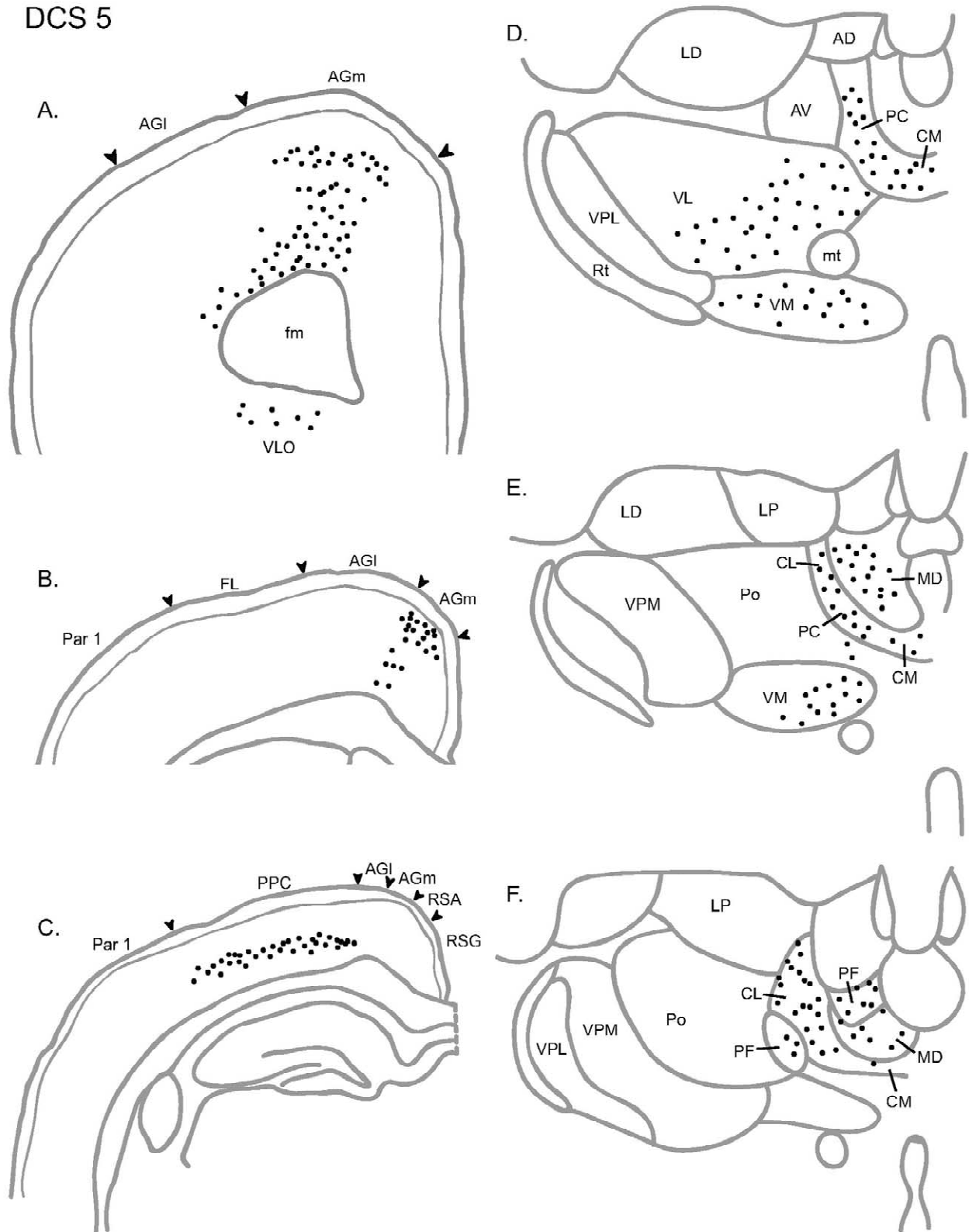


Fig. 2. Distribution of labeled cells in case 5, an injection in central DCS, depicted on selected coronal sections through the cerebral cortex (A–C) and thalamus (D–F).

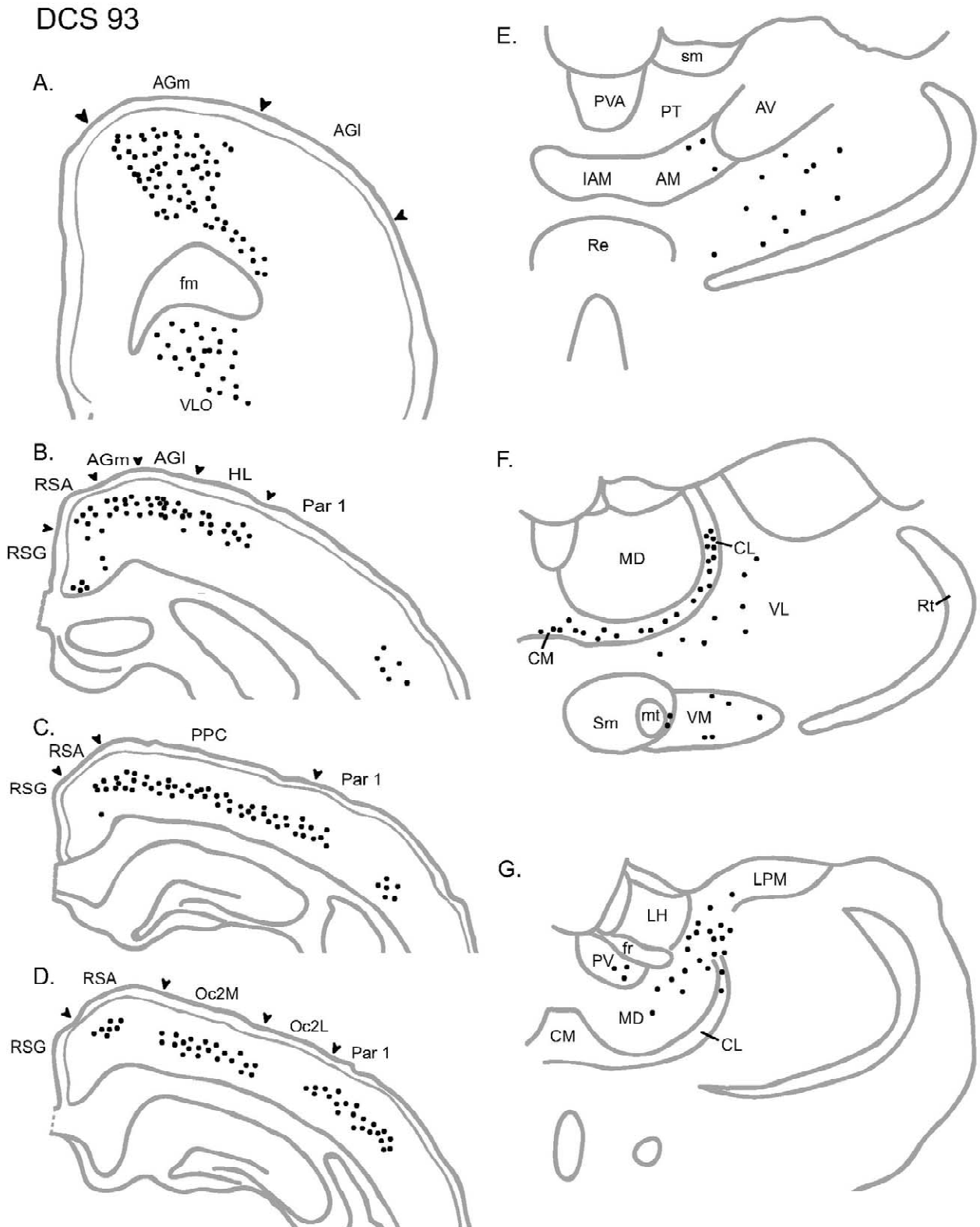


Fig. 3. Distribution of labeled cells in case 93, an injection in rostral DCS, depicted on selected coronal sections through the cerebral cortex (A–D) and thalamus (E–G).

DCS 105

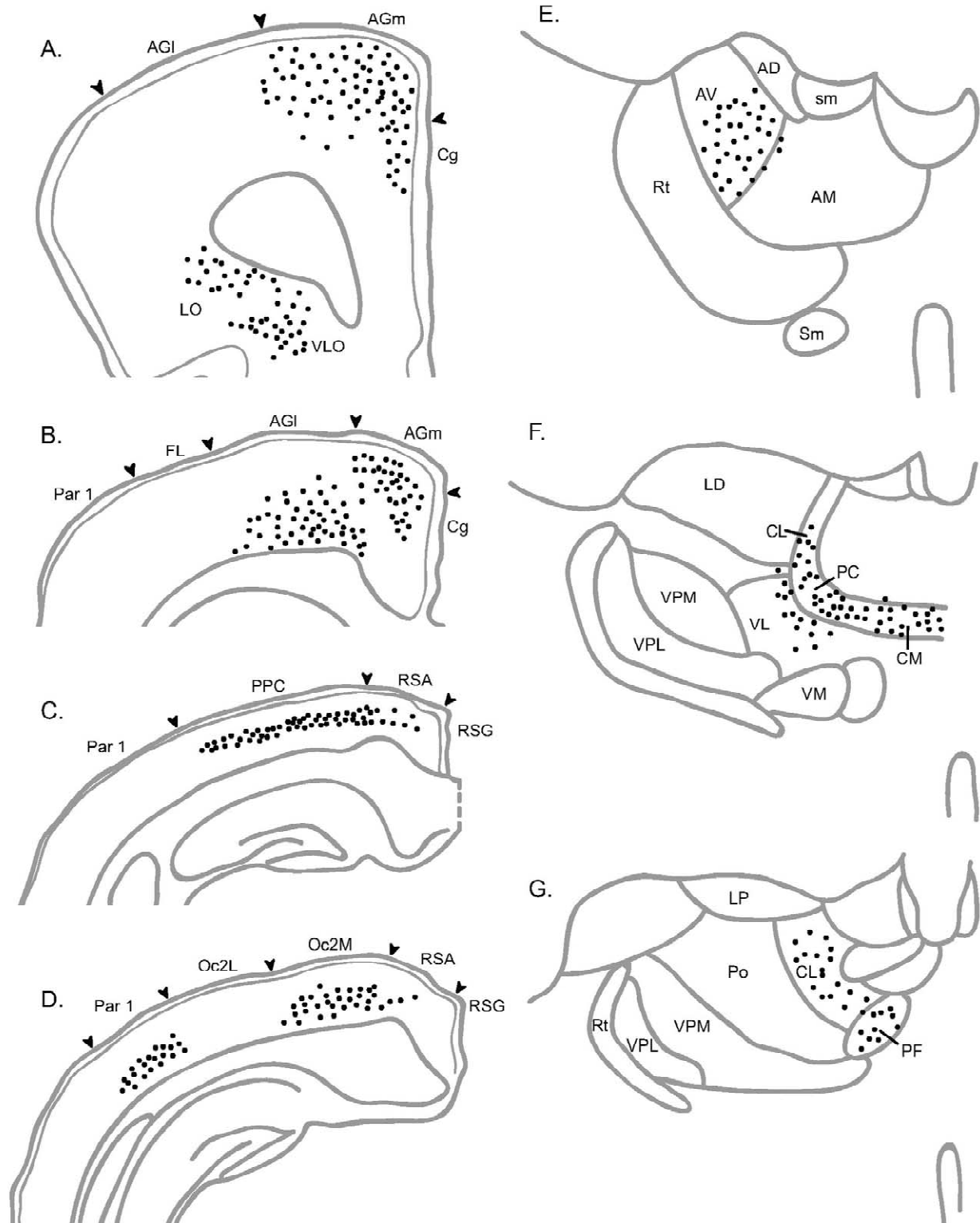


Fig. 4. Distribution of labeled cells in case 105, an injection in caudal DCS, depicted on selected coronal sections through the cerebral cortex (A–D) and thalamus (E–G).

a lack of labeled cells in RSA. Thalamic labeling differed from case 105 in the addition of LD, VPL, and VPM (Table 1).

3.2.4. Case 45: dorsal DCS

The injection site in case 45 was centered at the level of the anterior commissure and extended caudally to the level

of the fimbria (Fig. 1). The injection was spherical in shape and measured 0.5 mm in diameter. The dorsal border of the injection lay against the ventral border of the white matter. Therefore, this case represents a dorsal injection with respect to the boundaries of DCS. A moderate fluorescent track outlining the path of the pipette could be seen in all layers of area FL immediately above the center of the injection. Contralateral FL was not labeled, indicating that the track did not cause a change in the labeling pattern of the striatal injection. Because of the location of the injection site, a slight amount of fluorescence was visible in the white matter. In the cortex label could be seen in areas AGm, AGl, PPC, Oc2M, Oc2L, Par 1, Par 2, MO, VLO, LO, Cg, FL, HL, and PRh. Throughout its extent area AGm was labeled most densely in layers II/III, with infrequent labeling in layer V. Fluorescence was strongest within 1 mm of the ac (Fig. 5A,B). Labeled cells in layer V of area AGl could be seen from the level of the anterior commissure through the caudal border of AGl. Area PPC contained numerous labeled cells in layer V throughout its extent (Fig. 5C). Label in layer V of areas Oc2M and Oc2L began at their rostral pole and continued caudally through the remaining sections (Fig. 5D). Area Par 1 contained lightly labeled cells in layer V only (Fig. 5C,D). Perirhinal cortex contained labeled cells in layer V only. In the thalamus, nuclei AV, AD, VL, LD, medial LP, the intralaminar nuclei (CM, PC, CL), and PF contained labeled cells throughout their extent. Thalamic nucleus MD was labeled more strongly rostrally (Fig. 5E–G).

Case 52 had an injection site that overlapped that of case 45 from the level of the ac to the level of the fimbria, but extended more caudally (Fig. 1). Cortical labeling differed from case 45 by being sparse in rAGm, absent in MO, and present in areas RSA and RSG. In the thalamus the distribution was identical to case 45, with the addition of robust labeling in nuclei VPM and VPL (Table 1).

Case 84 had an injection site located lateral to both cases 45 and 52, and mostly caudal to case 45 (Fig. 1). Cortical labeling was similar to case 45 with the addition of labeled cells in RSA and RSG, and an absence of label in HL. The thalamic pattern was the same as case 45 (Table 1, Fig. 6D).

Case 76 (Fig. 6B) involved a large injection that was dorsally located, but did not overlap with the more caudal dorsal injections represented by cases 45, 52 and 84. It did overlap significantly with several rostral injections including cases 5, 15, 93, 94 and 104. Cortical labeling was extensive, but excluded Par 1 and Par 2 (Table 1). The thalamic labeling pattern was similar to the other dorsal cases presented above (Table 1).

3.3. Topography

From the cases presented above it was possible to discern some topographic patterns of connectivity. With

regard to the cortex it is notable that all injections produced labeled cells in areas AGm, PPC, orbital cortex, and Oc2M. No other cortical areas were labeled with this degree of consistency. The pattern of labeling in AGm was correlated with the location of the injection in DCS. Most of our injections had a longer rostrocaudal than dorsoventral extent and produced at least some labeling across the full rostrocaudal extent of areas AGm and PPC. However, injections centered rostrally in DCS (cases 5, 15, 93, 94) tended to produce their most robust labeling in rAGm, whereas injections located in caudal DCS (104, 105) exhibited sparser labeling in rAGm. One important exception is case 6, which had an injection located more ventrally in caudal DCS than cases 104 and 105, and exhibited only slight labeling in cAGm. Area PPC and visual association cortical areas Oc2M and Oc2L were labeled most densely in cases with injections on the dorsal border of DCS (cases 45, 52, 76, 84) and those placed caudally in DCS (104, 105). A rostrocaudal pattern of topography was also noticeable to a lesser degree in area AGl.

In the thalamus, the nuclei with the most evident topographical relationship with striatum were LD and LP. Labeled cells were found in LD and LP only in cases with injections in dorsal portions of DCS.

4. Discussion

Two important findings of the present study are the identification of cortical areas that project to DCS, and the discovery of a high degree of overlap in the projections from AGm and PPC. Also of significance is the finding that the thalamic nuclei projecting to DCS are also those that project to the cortical areas that provide input to DCS. Finally, there is evidence of some topography within DCS.

4.1. Corticostriatal connections

Many cortical areas have some projections to DCS, but the most frequent contributors are areas AGm, AGl, PPC, VLO, LO, and Oc2M. The projections from AGm and other frontal cortical areas to the striatum are arranged along rostrocaudal and mediolateral gradients [4,7,17,38], and the rostrocaudal topography that we observed in the present study with regard to the projections from AGm is consistent with these previous reports. There is also evidence of dorsoventral topography with respect to the labeling pattern in AGm. The injection in case 6 was the most ventral in our sample, and produced very little labeling in cAGm. This is consistent with the fact that cAGm projects to a more dorsal region within caudal DCS [38].

Evidence for spatial overlap in the terminal fields of

DCS 45

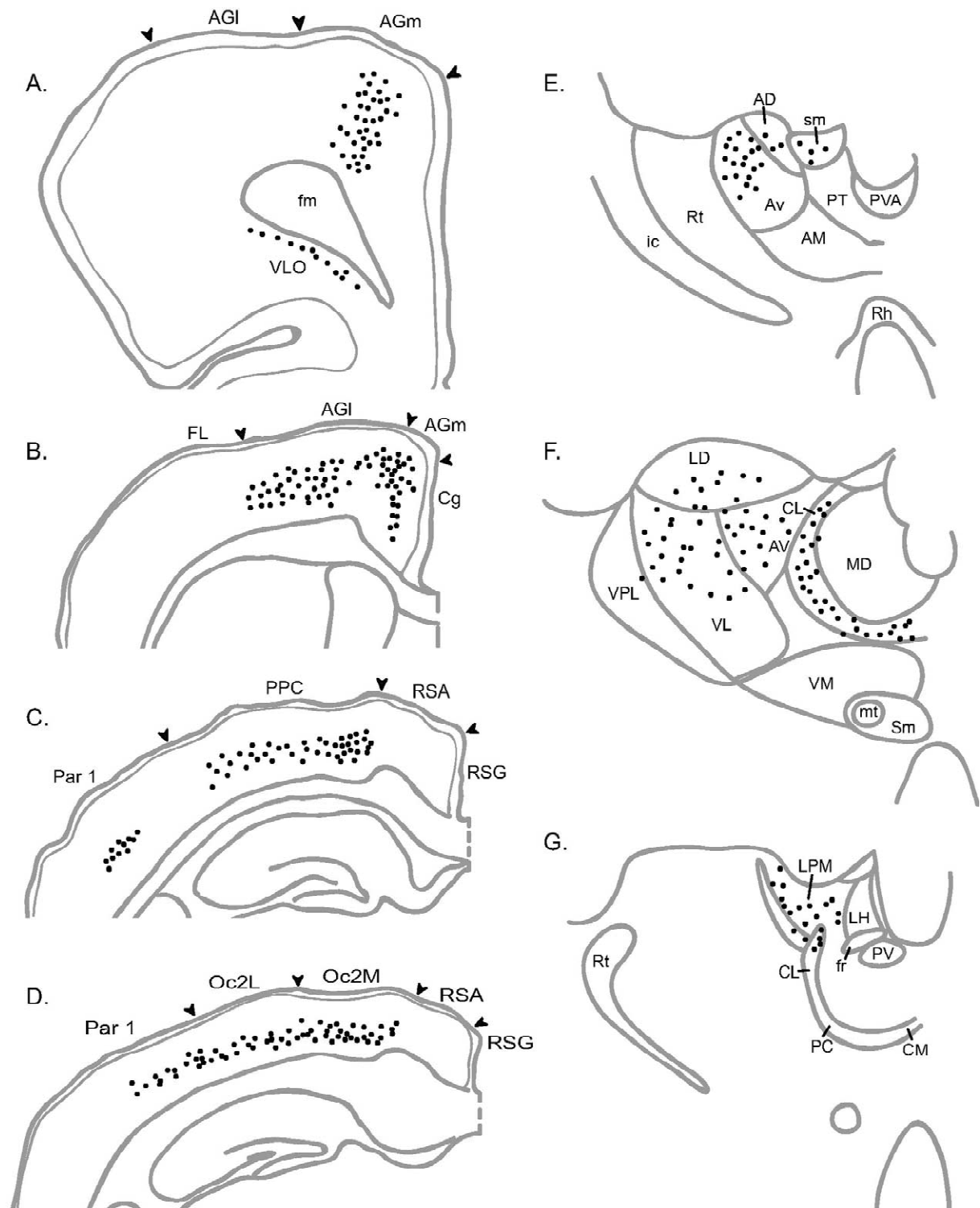


Fig. 5. Distribution of labeled cells in case 45, depicted on selected coronal sections through the cerebral cortex (A–D) and thalamus (E–G).

axons projecting from AGm and PPC to the striatum was noted previously [38,40]. In the present cases, all injections produced some degree of labeling in both PPC and

AGm, confirming the previous anterograde findings based on single injections in different brains. This finding is in itself suggestive of a potential relationship between in-

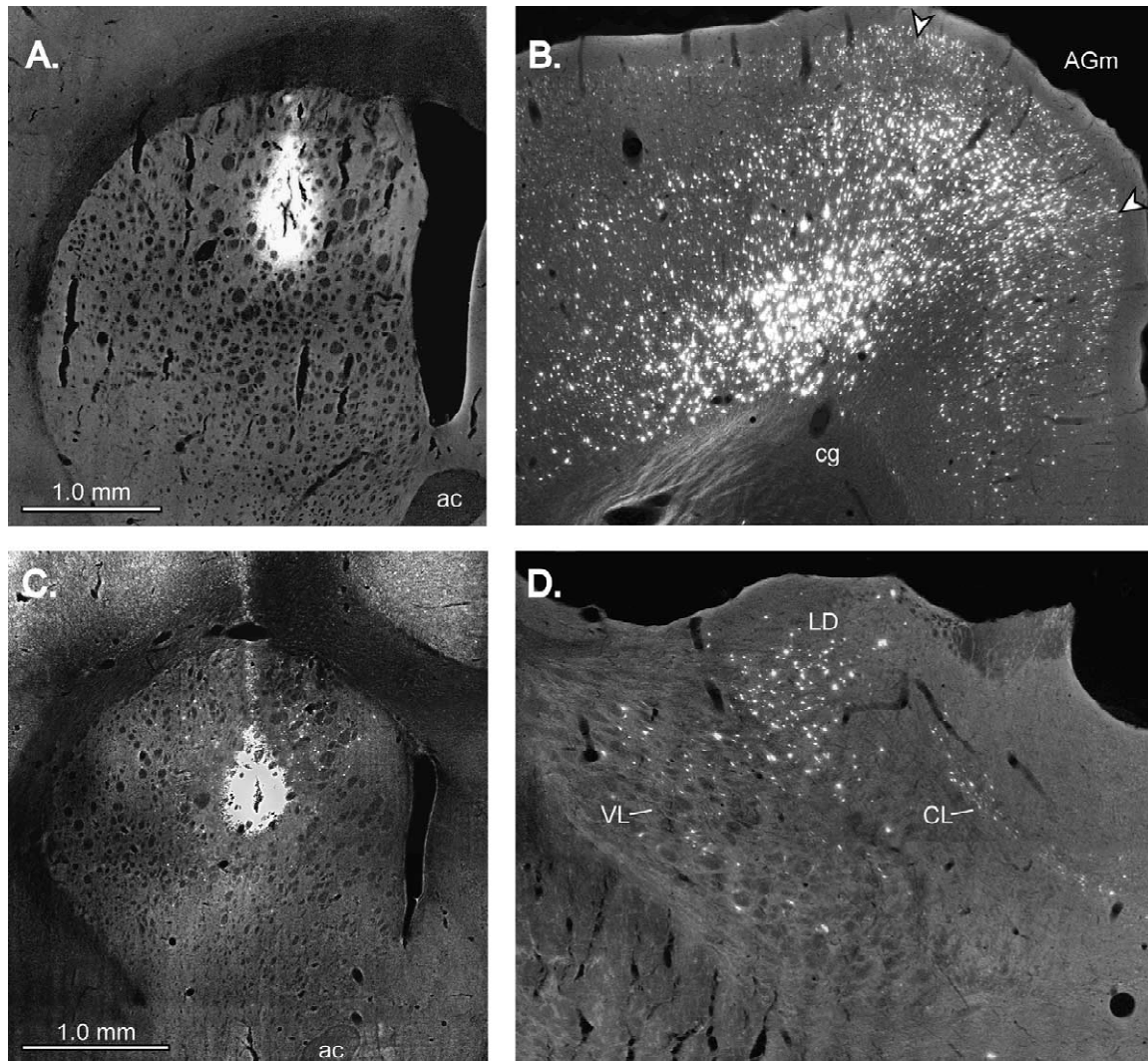


Fig. 6. Fluorescent photomicrographs of representative sections from select cases. (A) The Diamidino Yellow (DY) injection site in case 105 at the level of the septum. (B) Retrograde labeled cells in cortical area AGm, case 76. (C) The Fast Blue (FB) injection site in case 15 at the level of the genu. (D) Retrograde labeled cells in the thalamus, case 45.

formation coming from AGm and PPC into DCS, and relates to functional studies demonstrating that AGm and PPC are parts of an interconnected network mediating spatial processing and directed attention [11]. This network also includes orbital cortex and Oc2M, the final two regions labeled in every brain.

The present retrograde findings demonstrating convergence in the region of DCS of inputs from AGm, PPC and orbital cortex at the light microscopic level are suggestive of a possible interaction among these inputs. In primates, Yeterian and Van Hoesen [54] first demonstrated convergent striatal projections from distant cortical areas and related this to the corticocortical connectivity between these areas. A later study by Selemon and Goldman-Rakic [43] showed that most convergence of this kind actually consists of zones of interdigitating projections rather than

true overlap, and that convergence occurs even among projections from cortical areas that are not themselves interconnected. In rodent somatic sensory striatum there is significant true overlap in addition to interdigitation in the case of projections from cortical barrels representing the same whisker row [3]. It is possible that AGm, PPC, and VLO each project to a discrete territory within DCS, with little or no overlap. Alternatively, there may be convergence onto single striatal neuron dendritic trees [53]. These issues are discussed in more detail in a companion study [42], in which we found that dense patches of segregated labeling from AGm and PPC are interspersed with less dense patches of overlapping label in DCS.

The convergence in DCS of input from AGm and PPC is also interesting because of behavioral and pharmacological data implicating DCS as a critical site necessary for

normal directed attention and spatial processing, and recovery from deficits induced by cortical lesions [46,47]. The cannula placements in these studies tended to involve the dorsal portion of DCS between the levels of the septum and ac, exactly that territory implicated by the present findings as the AGm–PPC overlap zone.

In the present study visual association areas Oc2m and Oc2L were consistently labeled most densely with dorsal and caudal injections, and this is consistent with the topography defined by others [32,33]. We found no labeling in area Oc1, which projects to a circumscribed region of dorsomedial striatum bordering the lateral ventricle [32], outside the territory we injected. These authors noted that areas Oc2M and Oc2L project not only to this dorsomedial sector but also to the more deeply located territory encompassed by our injections, which is consistent with our findings.

Several other cortical areas labeled by injections in DCS have their densest corticostriatal terminations outside of DCS. Physiologically identified motor areas for the forelimb and hindlimb project in a banded pattern to the dorsolateral striatum, but the less dense medial portions of their projection fields appear to be located on the lateral margin of DCS [17]. The physiologically identified vibrissal-eyefield area located in AGm projects in a topographic manner to DCS and a dorsolateral shell [17]. Somatic sensory areas also project most densely to the dorsolateral striatum in banded patterns [8,23], but in each case the medial-most band is located within DCS between the levels of the genu and ac [8]. The focal size of these medial bands together with individual variation may explain the variability we observed in labeling of somatic sensory areas. Orbital and cingulate cortical areas project to territories along the ventral and medial borders of DCS and appear to have their densest terminations outside of DCS [4,7].

Some discrepancies observed in the present study may relate to the focal, distributed nature of corticostriatal axon arborizations [2,13,27,28,31] and their potential for variability across individuals. Cases 15 and 93 appeared to occupy virtually overlapping regions, but there was more widespread cortical labeling in case 93. Likewise, the dorsal cases 45, 84, and 76 exhibited labeled cells in orbital area MO; case 52 is also a dorsal injection, but did not produce labeling in MO.

The laminar distribution of labeled cortical neurons varied across cortical areas and within AGm. Labeled neurons were present in layers II/III and V in AGm and AGl, but only in layer V of the other cortical areas. Within AGm it was common to find only layers II/III labeled caudal to the level of the ac, as described for cases 93, 105 and 45. Wilson [52] also reported labeling in layers II/III and V within ipsilateral AGm, and labeling restricted to layer V in somatic sensory cortex following injection of a retrograde tracer into the striatum.

Due to the long rostrocaudal extent of our injections, our ability to make statements about highly discrete topog-

raphy of cortical projections to DCS is limited. The observed rostrocaudal expansion of our injections could be due to diffusion along corticostriatal axons, which are known to traverse long distances in the striatum, synapsing on striatal neurons at varying rostrocaudal levels [25]. Selemon and Goldman-Rakic [43] demonstrated that some cortical areas project axons that traverse the entire rostrocaudal extent of the primate caudate nucleus. Diffusion of this type presents a unique problem for discrete retrograde analyses in DCS and should be addressed in future studies.

Similarly, the placement of a tracer in the rat striatum raises the possibility of uptake not only by the axons of interest but also those fibers that are only passing through the striatum to form terminals elsewhere in the CNS. This is known to occur with retrograde fluorescent tracers [10]. However, the coordinated pattern of thalamic and cortical labeling we observed suggests uptake by axon terminals, because it seems unlikely that interconnected cortical and thalamic regions would be consistently labeled via uptake by fibers of passage. Rather, it would appear more parsimonious to suggest that the axons from such linked cortical and thalamic regions terminate in the same striatal territory. Ultimate resolution of this question will depend on double anterograde tracing.

4.2. Thalamostriatal projections

The pattern of thalamic projections to DCS bears a striking resemblance to the thalamic projections to those cortical areas that provide input to DCS. The rostral portion of area AGm receives thalamic input from lateral MD, VL, VM and the intralaminar nuclei, whereas inputs to caudal AGm emphasize VL, LD, LP and Po [22,38,41]. In the present study we found that with the exception of Po and the addition of the parafascicular nucleus, these same thalamic nuclei project to DCS. It is known that single neurons of the central lateral nucleus project to both the striatum and cortex [16], and if this is also an attribute of the projections of the other thalamic nuclei projecting to DCS it could underlie the registration observed. Anterograde tracing has demonstrated that the intralaminar nuclei (CM, PC, CL) have dense projections to the dorsal striatum [5,6]. Although their terminal fields are generally located medially (CM), centrally (PC) and laterally (CL), they overlap to some extent in the dorsocentral striatum, especially rostral to the level of the ac. This may explain why all three nuclei were labeled in all of our cases.

We found evidence of topography in the thalamic projections to DCS from LD and LP, and the observed pattern is consistent with the known organization of thalamocortical projections involving these nuclei and the topography of corticostriatal projections from their cortical targets. Injections in dorsal DCS consistently labeled thalamic nuclei LD and LP. Together with the fact that LD and LP project to cAGm [38], this topography suggests

that dorsal DCS is preferentially related to cAGm. Consistent with this claim, corticostriatal projections from cAGm project most densely to dorsal DCS [38]. Thalamic nuclei LD and LP also project to cortical areas PPC and Oc2M [40], which are densely connected with cAGm rather than rAGm [39,40]. Finally, cortical input to dorsal DCS is densest from areas PPC and Oc2M (present study and unpublished anterograde observations). Thus, dorsal DCS appears to represent a subregion of DCS that is distinguished by its network of thalamic and cortical connections. As an aside, the projections from LP to DCS and AGm [38,45] originate predominately from its medial portion, whereas the projection from LP to area PPC originates from the entire LP [40].

Our data are in agreement with the topography of labeling produced following injections of retrograde tracers (Fluorogold and the cholera toxin β -subunit) into the dorsomedial caudate-putamen, with the addition of AV and AD labeling [18]. The description of the rostrocaudal and mediolateral topography of labeled cells in the lateral and ventral thalamic nuclei following injections in the Erro et al. paper [18] is consistent with the pattern of labeling produced in our cases. They reported labeled cells in the ipsilateral intralaminar nuclei and medial thalamic nuclei in all of their cases [18]. Additionally, Erro et al. note that they found labeled cells in VPM and VPL following injections in the caudal striatum, a feature that is not widely accepted. Our injections also produced robust labeling in VPM and VPL in some, but not all, caudal (postcommissural) injections.

5. Conclusions

The present results together with previous findings demonstrate that DCS receives input from cortical and thalamic regions that are themselves interconnected. Furthermore, DCS and connected cortical areas are involved in directed attention and its dysfunctional counterpart, multimodal hemispatial neglect. We suggest that DCS is a key element of a cortico-striatal-thalamic network specialized for multimodal associations and spatial functions including directed attention. This network includes cortical areas AGm and PPC, the striatal area DCS, and the thalamic nuclei VL, MD, LD, LP, and the intralaminar group. This network is similar to the large scale networks proposed to be involved in spatial functions in primates [21,34,44]. Similar interconnected cortico-striatal-thalamic networks have been identified for sensory, motor and limbic systems [1,20,36].

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References

- [1] G.E. Alexander, M.D. Crutcher, M.R. DeLong, Basal ganglia-thalamocortical circuits: parallel substrates for motor, oculomotor, 'prefrontal' and 'limbic' functions, in: H.B.M. Uylings, C.G. Van Eden, J.P.C. De Bruin, M.A. Corner, M.G.P. Feenstra (Eds.), *The Prefrontal Cortex: Its Structure, Function, and Pathology*, Progress in Brain Research, Vol. 85, Elsevier, Amsterdam, 1990, pp. 119–146.
- [2] K.D. Alloway, J.J. Mutic, J.E. Hoover, Divergent corticostriatal projections from a single cortical column in the somatosensory cortex of rats, *Brain Res.* 785 (1998) 341–346.
- [3] K.D. Alloway, J. Crist, J.J. Mutic, S.A. Roy, Corticostriatal projections from rat barrel cortex have an anisotropic organization that correlates with vibrissal whisking behavior, *J. Neurosci.* 19 (1999) 10908–10922.
- [4] R.M. Beckstead, An autoradiographic examination of corticocortical and subcortical projections of the mediodorsal-projection (prefrontal) cortex in the rat, *J. Comp. Neurol.* 184 (1979) 43–62.
- [5] H.W. Berendse, H.J. Groenewegen, Organization of the thalamostriatal projections in the rat, with special emphasis on the ventral striatum, *J. Comp. Neurol.* 299 (1990) 187–228.
- [6] H.W. Berendse, H.J. Groenewegen, Restricted cortical termination fields of the midline and intralaminar thalamic nuclei in the rat, *Neuroscience* 42 (1991) 73–102.
- [7] H.W. Berendse, Y. Galis-DeGraaf, H.J. Groenewegen, Topographical organization and relationship with ventral striatal compartments of prefrontal corticostriatal projections in the rat, *J. Comp. Neurol.* 316 (1992) 314–347.
- [8] L.L. Brown, D.M. Smith, L.M. Goldbloom, Organizing principles of cortical integration in the rat neostriatum: corticostriate map of the body surface is an ordered lattice of curved laminae and radial points, *J. Comp. Neurol.* 392 (1998) 468–488.
- [9] K.J. Burcham, J.V. Corwin, M.L. Stoll, R.L. Reep, Disconnection of medial agranular and posterior parietal cortex produces multimodal neglect in rats, *Behav. Brain Res.* 86 (1997) 41–47.
- [10] F. Condé, Further studies on the use of the fluorescent tracers fast blue and diamidino yellow: effective uptake area and cellular storage sites, *J. Neurosci. Methods* 21 (1987) 31–43.
- [11] J.V. Corwin, R.L. Reep, Rodent posterior parietal cortex as a component of a cortical network mediating directed spatial attention, *Psychobiology* 26 (1998) 87–102.
- [12] J.V. Corwin, S. Kanter, R.T. Watson, K.M. Heilman, E. Valenstein, A. Hashimoto, Apomorphine has a therapeutic effect on neglect produced by unilateral dorsomedial prefrontal cortex lesions in rats, *Exp. Neurol.* 94 (1986) 683–698.
- [13] R.L. Cowan, C.J. Wilson, Spontaneous firing patterns and axonal projections of single corticostriatal neurons in the rat medial agranular cortex, *J. Neurophysiol.* 71 (1994) 17–32.
- [14] D.P. Crowne, M.N. Pathria, Some attentional effects of unilateral frontal lesions in the rat, *Behav. Brain Res.* 6 (1982) 25–39.
- [15] D.P. Crowne, C.M. Richardson, G. Ward, Brief deprivation of vision after unilateral lesions of the frontal eye field prevents contralateral inattention, *Science* 220 (1983) 527–530.
- [16] M. Deschences, J. Bourassa, A. Parent, Striatal and cortical projections of single neurons from the central lateral thalamic nucleus in the rat, *Neuroscience* 72 (1996) 679–687.
- [17] A. Ebrahimi, R. Pochet, M. Roger, Topographical organization of

- the projections from physiologically identified areas of the motor cortex to the striatum in the rat, *Neurosci. Res.* 14 (1992) 39–60.
- [18] M.E. Erro, J.L. Lanciego, J.M. Giménez-Amaya, Re-examination of the thalamostriatal projections in the rat with retrograde tracers, *Neurosci. Res.* 42 (2002) 45–55.
- [19] A.M. Graybiel, T. Aosaki, A.W. Flaherty, M. Kimura, The basal ganglia and adaptive motor control, *Science* 265 (1994) 1826–1831.
- [20] H.J. Groenewegen, H.W. Berendse, J.G. Wolters, A.H.M. Lohman, The anatomical relationship of prefrontal cortex with the striatopallidal system, the thalamus and the amygdala: evidence for a parallel organization, in: H.B.M. Uylings, C.G. Van Eden, J.P.C. De Bruin, M.A. Corner, M.G.P. Feenstra (Eds.), *The Prefrontal Cortex: Its Structure, Function, and Pathology*, Progress in Brain Research, Vol. 85, Elsevier, Amsterdam, 1990, pp. 95–118.
- [21] K.M. Heilman, R.T. Watson, E. Valenstein, Neglect and related disorders, in: K.M. Heilman, E. Valenstein (Eds.), *Clinical Neuropsychology*, 3rd Edition, Oxford University Press, New York, 1993, pp. 279–336.
- [22] R.R. Hicks, M.F. Huerta, Differential thalamic connectivity of rostral and caudal parts of area Fr2 in rats, *Brain Res.* 568 (1991) 325–329.
- [23] Z.S. Hoffer, K.D. Alloway, Organization of corticostriatal projections from the vibrissal representations in the primary motor and somatosensory cortical areas of rodents, *J. Comp. Neurol.* 439 (2001) 87–103.
- [24] E.G. Jones, *The Thalamus*, Plenum Press, New York, 1985.
- [25] E.G. Jones, J.D. Coulter, H. Burton, R. Porter, Size, laminar and columnar distribution of afferent cells in the sensory-motor cortex of monkeys, *J. Comp. Neurol.* 175 (1977) 391–438.
- [26] R.P. Kesner, G. Farnsworth, B.V. Dimattia, Double dissociation of egocentric and allocentric space following prefrontal and parietal cortex lesions in the rat, *Behav. Neurosci.* 103 (1989) 956–961.
- [27] A.E. Kincaid, C.J. Wilson, Corticostriatal innervation of the patch and matrix in the rat neostriatum, *J. Comp. Neurol.* 374 (1996) 578–592.
- [28] A.E. Kincaid, T. Zheng, C.J. Wilson, Connectivity and convergence of single corticostriatal axons, *J. Neurosci.* 18 (1998) 4722–4731.
- [29] V. King, J.V. Corwin, Spatial deficits and hemispheric asymmetries in the rat following unilateral and bilateral lesions of posterior parietal or medial agranular cortex, *Behav. Brain Res.* 143 (1992) 237–242.
- [30] V. King, J.V. Corwin, Comparisons of hemiattention produced by unilateral lesions of the posterior parietal or the medial agranular prefrontal cortex in the rat, *Behav. Brain Res.* 54 (1993) 117–131.
- [31] M. Levesque, A. Charara, S. Gagnon, A. Parent, M. Deschenes, Corticostriatal projections from layer V cells in rat are collaterals of long-range corticofugal axons, *Brain Res.* 709 (1996) 311–315.
- [32] M.O. Lopez-Figueroa, J.A. Ramirez-Gonzalez, I. Divac, Projections from the visual areas to the neostriatum in rats: a re-examination, *Acta Neurobiol. Exp.* 55 (1995) 165–175.
- [33] A.J. McGeorge, R.L.M. Faull, The organization of the projection from the cerebral cortex to the striatum in the rat, *Neuroscience* 29 (1989) 503–537.
- [34] M.-M. Mesulam, Large-scale neurocognitive networks and distributed processing for attention, language, and memory, *Ann. Neurol.* 28 (1990) 597–613.
- [35] E.J. Neafsey, E.L. Bold, G. Haas, K.M. Hurley-Guis, G. Quirk, C.F. Sievert, R.R. Terberry, The organization of the rat motor cortex: a microstimulation mapping study, *Brain Res. Rev.* 11 (1986) 77–96.
- [36] A. Parent, L.-N. Hazrati, Functional anatomy of the basal ganglia. I. The cortico-basal ganglia-thalamo-cortical loop, *Brain Res. Rev.* 20 (1995) 91–127.
- [37] G. Paxinos, C. Watson, *The Rat Brain in Stereotaxic Coordinates*, 2nd Edition, Academic Press, Sydney, 1986.
- [38] R.L. Reep, J.V. Corwin, Topographic organization of the striatal and thalamic connections of rat medial agranular cortex, *Brain Res.* 84 (1999) 43–52.
- [39] R.L. Reep, G.S. Goodwin, J.V. Corwin, Topographic organization in the corticocortical connections of medial agranular cortex in rats, *J. Comp. Neurol.* 294 (1990) 262–280.
- [40] R.L. Reep, H.C. Chandler, V. King, J.V. Corwin, Rat posterior parietal cortex: topography of corticocortical and thalamic connections, *Exp. Brain Res.* 100 (1994) 67–84.
- [41] R.L. Reep, J.V. Corwin, A. Hashimoto, R.T. Watson, Afferent connections of medial precentral cortex in the rat, *Neurosci. Lett.* 44 (1984) 247–252.
- [42] R.L. Reep et al. (2003) (submitted for publication).
- [43] L.D. Selemon, P.S. Goldman-Rakic, Longitudinal topography and interdigitation of corticostriatal projections in the rhesus monkey, *J. Neurosci.* 5 (1985) 776–794.
- [44] L.D. Selemon, P.S. Goldman-Rakic, Common cortical and subcortical targets of the dorsolateral prefrontal and posterior parietal cortices in the rhesus monkey: evidence for a distributed neural network subserving spatially guided behavior, *J. Neurosci.* 8 (1988) 4049–4068.
- [45] K. Sukekawa, Reciprocal connections between medial prefrontal cortex and lateral posterior nucleus in rats, *Brain Behav. Evol.* 32 (1988) 246–251.
- [46] T.M. Van Vleet, J.V. Corwin, K.J. Burcham, Infusion of apomorphine into the dorsal central striatum produces acute recovery from AGm-induced neglect in rats, *Soc. Neurosci. Abstr.* 25 (1999) 1896.
- [47] T.M. Van Vleet, K.J. Burcham, J.V. Corwin, R.L. Reep, Unilateral destruction of the medial agranular cortical projection zone in the dorsocentral striatum produces severe neglect in rats, *Psychobiology* 28 (2000) 57–66.
- [48] J.M. Vargo, J.F. Marshall, Time-dependent changes in dopamine agonist-induced striatal fos immunoreactivity are related to sensory neglect and its recovery after unilateral prefrontal cortex injury, *Synapse* 20 (1995) 305–315.
- [49] J.M. Vargo, J.F. Marshall, Frontal cortex ablation reversibly decreases striatal zif/268 and junB expression: temporal correspondence with sensory neglect and its spontaneous recovery, *Synapse* 22 (1996) 291–303.
- [50] J.M. Vargo, J.F. Marshall, Unilateral frontal cortex ablation producing neglect causes time-dependent changes in striatal glutamate receptors, *Behav. Brain Res.* 77 (1996) 189–199.
- [51] J.M. Vargo, J.V. Corwin, V. King, R.L. Reep, Hemispheric asymmetry in neglect produced by unilateral lesions of dorsomedial prefrontal cortex in rats, *Exp. Neurol.* 102 (1988) 199–209.
- [52] C.J. Wilson, Morphology and synaptic connections of crossed corticostriatal neurons in the rat, *J. Comp. Neurol.* 263 (1987) 567–580.
- [53] C.J. Wilson, Basal ganglia, in: G.M. Shepherd (Ed.), *The Synaptic Organization of the Brain*, 4th Edition, Oxford University Press, New York, 1998, pp. 329–376.
- [54] E.H. Yeterian, G.W. Van Hoesen, Cortico-striate projections in rhesus-monkey—organization of certain cortico-caudate connections, *Brain Res.* 139 (1978) 43–63.
- [55] K. Zilles, A. Wree, Cortex: a real and laminar structure, in: G. Paxinos (Ed.), *The Rat Nervous System: Forebrain and Midbrain*, Vol. 1, Academic Press, Sydney, 1985, pp. 375–415.