The Laminar Pattern of Connections between Prefrontal and Anterior Temporal Cortices in the Rhesus Monkey is Related to Cortical Structure and Function

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The laminar pattern of axonal termination from prefrontal (caudal orbitofrontal, rostral orbitofrontal and lateral areas) to anterior temporal areas (entorhinal cortex, perirhinal cortex and area TE) and from temporal to prefrontal areas was investigated with the aid of anterograde tracers. Both regions are characterized by structural heterogeneity, and include agranular, dysgranular and granular cortical types, denoting, respectively, the absence, incipience and presence of granular layer 4. In addition, both the prefrontal and anterior temporal cortices are composed of areas that have related though specialized functions. The pattern of cortical axonal termination was associated with both the structural type of the cortex of origin and the structure of the destination cortex. Thus, efferent fibers from a single origin in either prefrontal or anterior temporal cortex terminated in different patterns depending on their target area. Conversely, axons terminated in different patterns in a single target area, prefrontal or anterior temporal, depending on their area of origin. Projections from agranular or dysgranular type cortices (e.g. medial temporal areas and caudal orbitofrontal areas) terminated mostly in the upper layers of granular cortices (e.g. area TE and lateral prefrontal areas), and projections from granular cortices terminated mostly in the deep layers of agranular or dysgranular cortices. A robust projection from dysgranular orbitofrontal areas terminated in the deep layers of the agranular entorhinal cortex. Projections from prefrontal areas to area TE terminated in the upper layers, and may facilitate focused attention on behaviorally relevant stimuli processed through reciprocal pathways between prefrontal and temporal cortices.

Introduction

Anatomical studies have demonstrated the existence of connections linking medial and inferior temporal areas with prefrontal areas, and the general topography and laminar origin of neurons giving rise to the connections between these areas are known (Barbas, 1986, 1988, 1993; Seltzer and Pandya, 1989; Morecraft *et al.*, 1992; Rodman and Consuelos, 1994; Suzuki and Amaral, 1994a; Webster *et al.*, 1994). However, a fundamental question that has not been addressed is whether different prefrontal and temporal areas have a common or distinct pattern in their anatomic communication. For example, do efferent fibers from all prefrontal areas terminate in the same laminar pattern when their target in the temporal lobe is the same? Conversely, does a single prefrontal area terminate in the same or a different pattern when it targets several temporal cortices? Since the chemical, connectional and physiological properties of neurons differ across layers within a single column of cortex (Lund, 1988; Hof *et al.*, 1999), axons terminating in different layers convey potentially distinct information.

The organization of the laminar pattern of connections between prefrontal and anterior temporal areas may be considered within at least two frameworks. One possibility is that the laminar pattern of connections between these areas is related to the functions of the connected areas. The anterior temporal region is composed of a series of areas with interrelated but specialized functions (Buckley *et al.*, 1997; Buffalo *et al.*, 1998, 1999). Occupying the medial flank of the anterior temporal lobe, the medial temporal entorhinal and perirhinal cortices are involved in long-term memory for various sensory modalities, including the visual (Jones and Mishkin, 1972; Fuster *et al.*, 1981, 1985; Voytko, 1986; Zola-Morgan *et al.*, 1989; Meunier *et al.*, 1993; Suzuki *et al.*, 1993, 1997; Murray *et al.*, 1998). The laterally adjacent inferior temporal cortex (area TE) has visual perceptual functions and its role in memory appears limited to the visual modality [for a review see (Gross, 1994)]. The prefrontal cortex is similarly a large and heterogeneous region [for reviews see (Goldman-Rakic, 1987; Barbas, 1997)]. Functional differences have been proposed for lateral and orbitofrontal areas (Rosenkilde, 1979; Barbas, 1995, 2000; Dias *et al.*, 1996). For example, orbitofrontal areas have been implicated in long-term memory and emotional functions (Jones and Mishkin, 1972; Stamm, 1973; Passingham, 1975; Mishkin and Manning, 1978; Fuster *et al.*, 1985; Suzuki *et al.*, 1993; Nakamura and Kubota, 1996; Meunier *et al.*, 1997; Van Hoesen *et al.*, 1999), while lateral prefrontal cortices are important for working memory [for reviews see (Goldman-Rakic, 1996; Owen, 1997)]. In addition, there are differences in the connections of caudal and rostral orbitofrontal areas, suggesting they may be involved in different aspects of behavior (Barbas, 1993).

Another possibility is that the laminar pattern of connections between prefrontal and anterior temporal areas is related to differences in their cortical structure, as we noted previously for prefrontal interconnections (Barbas and Rempel-Clower, 1997). Both prefrontal and anterior temporal regions are structurally heterogeneous, composed of cortices that have fewer than six layers and lack a granular layer 4 (agranular cortex) or have an incipient granular layer 4 (dysgranular cortex), or have six layers (granular cortex) (Moran *et al.*, 1987; Barbas and Pandya, 1989; Morecraft *et al.*, 1992). The medial temporal cortices are agranular or dysgranular (e.g. temporal pole, entorhinal and perirhinal cortices), whereas area TE is granular. In the prefrontal cortices, the lateral areas and the rostral orbitofrontal areas are granular, whereas the caudal orbitofrontal areas are agranular or dysgranular [for a review see (Barbas, 1997)].

In the present study, we sought to characterize the laminar pattern of termination of the connections linking caudal orbital, rostral orbital and lateral prefrontal areas with anterior temporal cortices. Our analysis in the temporal region was restricted to medial temporal and inferior temporal cortices, which are known to have robust interconnections with functionally and structurally distinct orbital as well as lateral prefrontal areas [for reviews see (Suzuki, 1996b; Barbas, 1997]. It should be noted, however, that the prefrontal cortex has additional distributed connections with other cortices, including other temporal cortices [for a review see (Cusick, 1997)]. Analyses in this study

focused exclusively on the termination of efferent fibers, which clearly shows laminar patterns. Our goal was to investigate whether there is a consistent relationship in the pattern of connections between distinct prefrontal and temporal cortices and their functional specificity or their cortical structure.

Materials and Methods

Surgical and Histological Procedures

Experiments were conducted on 11 rhesus monkeys (*Macaca mulatta*) according to the NIH guide for the Care and Use of Laboratory Animals (NIH publication 86-23, revised 1987). The animals were anesthetized with ketamine hydrochloride (10 mg/kg, i.m.) followed by sodium pentobarbital administered i.v. through a femoral catheter until a surgical level of anesthesia was achieved (cumulative dose ~30 mg/kg). Additional anesthetic was administered during surgery as needed. The monkey's head was firmly positioned in a holder that left the cranium unobstructed for surgical approach. A craniotomy was made and the dura retracted to expose the cortex. All injections were made with a microsyringe (Hamilton, 5 μl) mounted on a microdrive. Injections of HRP-WGA (Sigma, St Louis, MO) were placed in the prefrontal cortices in six animals, [^{3}H]leucine and [^{3}H]proline were injected in the prefrontal cortices of three animals and biotinylated dextran amine (BDA, mol. wt 3000, lysine fixable; Molecular Probes, Eugene, OR) was injected in the anterior temporal cortices of two animals. In each case, a neural tracer was injected 1.5 mm below the pial surface in the following quantities and concentrations: 0.05–0.1 μl of 8% HRP-WGA; 0.4–1.0 μl of [^{3}H]-leucine and [^{3}H]proline, sp. act. 40–80 Ci/mmol; and 1.5 μl of 10% BDA.

In the HRP experiments, the monkeys were anesthetized deeply 40–48 h after injection and perfused through the heart with saline followed by 2 l of fixative (1.25% glutaraldehyde, 1% paraformaldehyde in 0.1 M phosphate buffer, pH 7.4), followed by 2 l of cold (4°C) phosphate buffer (0.1 M, pH 7.4). The brains were then removed from the skull, photographed and cryoprotected in glycerol phosphate buffer (10% glycerol and 2% DMSO in 0.1 M phosphate buffer, pH 7.4) for 1 day and in 20% glycerol phosphate buffer for another 2 days. The animals injected with BDA were anesthetized 14 days after surgery and perfused with 4% paraformaldehyde in 0.1 M phosphate buffer (pH 7.4). The brains were cryoprotected in sucrose solutions of 10, 15, 18 and 25% in 0.1 M phosphate buffer. The brains with injections of HRP or BDA were then frozen in −75°C isopentane, transferred to a freezing microtome and cut in the coronal plane at 40 μm in ten series. For the HRP experiments, one series of sections was treated to visualize HRP (Mesulam *et al.*, 1980). The tissue was mounted, dried and counterstained with neutral red. For the BDA experiments, a series of sections was treated in a Vector ABC Elite solution (PK-6100) for 3 h, and 2–5 min in 3,3-diaminobenzidene tetrahydrochloride (DAB-Plus, Zymed Laboratories).

In animals injected with [^{3}H]amino acids, the survival period was 10 days. The animals were anesthetized deeply and perfused with saline followed by 10% formalin. The brains were removed, photographed, stored in 50% ethanol, embedded in paraffin and cut in the coronal plane at 10 μm thickness. The tissue was processed for autoradiography according to the procedure of Cowan *et al*. (1972). Exposure time ranged from 3–6 months. Architectonic areas and their borders were determined by staining with thionin, acetylcholinesterase (AChE) or myelin (Geneser-Jensen and Blackstad, 1971; Gallyas, 1979).

Data Analysis

Outlines of brain sections, the location of the injection site and the general regional distribution of labeled terminals and fibers ([^{3}H]amino acids, HRP or BDA) were transferred from the slides onto paper by means of a digital plotter (Hewlett Packard, 7475A) electronically coupled to the stage of the microscope and to a computer (Austin 486). In this system the analog signals are converted to digital signals via an analog-to-digital converter (Data Translation, Marlboro, MA) in the computer. Movement of the microscope stage was recorded via linear potentiometers (Vernitech, Axsys, San Diego, CA) mounted on the x and y axes of the stage of the microscope and coupled to a power supply. Every other prepared section through the cortex in one series was examined and charted.

After creating outlines of brain sections indicating the location of anterograde label, the density and laminar distribution of anterograde label in the cortex were determined using an image analysis system (MetaMorph, Universal Imaging System Corp., West Chester, PA). This high resolution system uses a CCD camera (Dage-MTI, Michigan City, IN) mounted on the microscope to capture images directly from brain sections. Measurements from the HRP and [^{3}H]amino acid cases were made under dark-field illumination using a fiber optic illuminator (Optical Analysis Corp., Nashua, NH), and for the BDA cases under bright-field illumination at a magnification of 100×. If more than one site contained label within a single architectonic area, then each site was measured separately. An initial density measure in each section was taken in an area with no anterograde label to determine the level of background. The background density was subtracted from subsequent measures to determine the density of anterograde label. Measurements of density were taken from multiple samples in each layer in each site of anterograde label to avoid retrogradely labeled neurons. The cumulative density of label within the entire extent of each architectonic area was calculated from serial coronal sections for layers 1, 2/3, 4 and 5/6. To determine the laminar pattern of anterograde label, data were normalized so that the density in the upper layers (mean density in layers 1–3) and the lower layers (mean density in layers 4–6) was expressed as a percentage of the total in that area (total density = density in upper layers + density in lower layers). In previous studies, this quantitative method has provided the same results as qualitative analyses (Barbas and Rempel-Clower, 1997; Rempel-Clower and Barbas, 1998; Barbas *et al.*, 1999).

Images for photomicrographs were captured directly from histological brain slides using a CCD camera, imported into Adobe Photoshop for assembly and labeling, and adjusted for brightness and contrast but not retouched.

Reconstruction of Injection Sites

The cortical regions containing the injection sites were reconstructed serially by using the sulci as landmarks, as described previously (Barbas, 1988), and are shown on a diagram of the surface of the cortex. References to architectonic areas of the prefrontal cortex are according to a previous study (Barbas and Pandya, 1989). Each injection extended through the depth of the cortex to include all layers.

Results

Medial and Inferior Temporal Cortex

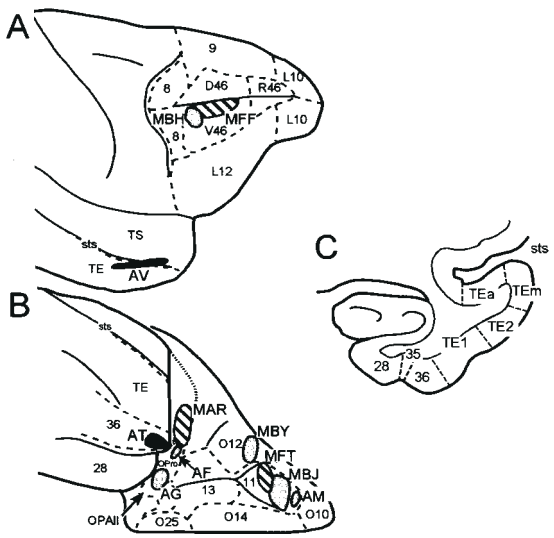
The nomenclature used for the anterior temporal cortices in this study is based primarily on the maps of Seltzer and Pandya (Seltzer and Pandya, 1978) and Suzuki and Amaral (Suzuki and Amaral, 1994a). Our analysis included all medial temporal and inferior temporal cortices. The medial temporal region is composed of a series of areas, including the ventral temporal polar cortex, the entorhinal cortex (area 28), the perirhinal cortex (areas 35 and 36) and the parahippocampal cortex (areas TH and TF). The anterior inferior temporal region includes areas TE1, TE2, TE3, TEm and TEa (Fig. 1). Borders for areas 28, 35, TH and TF are based on the maps of Suzuki and Amaral (Suzuki and Amaral, 1994a). The borders used for area 36 were consistent with the medial, lateral and posterior boundaries described by Suzuki and Amaral, but we did not include the cortex extending to the tip of the temporal pole in area 36. The ventral portion of the temporal pole is continuous with areas 28, 35, 36 and TE1. In previous studies this area has been referred to as area 38 (Brodmann, 1909), area TG (Von Bonin and Bailey, 1947), area Pro (Pandya and Sanides, 1973; Galaburda and Pandya, 1983), the agranular and dysgranular components of the temporal pole [TPa-p, TPdg (Mesulam and Mufson, 1982; Moran *et al.*, 1987)], the periallocortical and ventral proisocortical divisions of the temporal pole [TPpAll, TPproV (Gower, 1989)] and rostral area

36 [36r (Suzuki and Amaral, 1994b)]. We refer to this area simply as the temporal polar cortex.

Injection Sites

Figure 1 includes a composite diagram showing the location of injection sites of neural tracers in prefrontal and temporal cortices. Prefixes are used throughout the text and figures to indicate the more precise location in a particular prefrontal area (C, caudal; R, rostral; L, lateral; M, medial; O, orbital; V, ventral). The HRP injections were located in five orbital areas (area OPro/OPAll, case AG; area OPro, case AF; area 11, cases MBJ and AM; area O12, case MBY) and in one lateral prefrontal area (V46, case MBH). The [^{3}H]amino acid injections were located in area OPro (case MAR), area 11 (case MFT) and area V46 (case MFF). The BDA injections were located in anterior temporal cortices in area 36 (case AT) and area TE1 (case AV). The injection site in case AV extended slightly above the superior temporal sulcus into area TS1 from the map of Seltzer and Pandya (Seltzer and Pandya, 1978).

Most of the prefrontal cases described here appeared in previous studies investigating connections with the hypothalamus (Rempel-Clower and Barbas, 1998), amygdala (Barbas and De Olmos, 1990), thalamus (Barbas *et al.*, 1991; Dermon and Barbas, 1994), hippocampal formation (Barbas and Blatt, 1995) or other cortical areas (Barbas, 1993). The analyses in the previous studies investigating connections with other cortical areas were restricted to retrogradely labeled neurons. Recent studies use the same identification codes we use here, and refer to the designations used in older studies (Barbas, 1993; Dermon and Barbas, 1994; Barbas and Blatt, 1995; Barbas and Rempel-Clower, 1997; Rempel-Clower and Barbas, 1998).



D

Region	Areas	Cortical Type
Caudal Orbitofrontal	OPAll, OPro, 13, O25	Agranular, Dysgranular
Rostral Orbitofrontal	11, O12, O14,	Granular, narrow layer 4
Lateral Prefrontal	L12, L10, V46	Granular, wide layer 4
Medial Temporal	temporal pole, 28, 35, 36	Agranular, Dysgranular
Inferior Temporal	TE1, TE2, TEm, TEa	Granular

Figure 1. (*A*,*B*) Composite of injection sites shown on the lateral (*A*) and orbital (*B*) surfaces of the right cerebral hemisphere. The injection sites are superimposed on an architectonic map of the prefrontal cortex and anterior temporal cortex. Shaded sites were injected with horseradish peroxidase conjugated to wheatgerm agglutinin (HRP-WGA), black sites were injected with biotinylated dextran amine (BDA) and the striped sites were injected with [^{3}H]labeled amino acids. The temporal pole was removed from the orbital view (*B*) to show an injection in caudal orbitofrontal area proisocortex (OPro). (*C*) Coronal section showing borders for medial (areas 28, 35 and 36) and inferior temporal areas (areas TE1–2, TEm and TEa). (*D*) Prefrontal and temporal cortices consist of several regions, each including multiple areas. Cortical type or structure varies by region in a graded fashion. In *A-C*, dashed lines demarcate architectonic areas indicated by numbers. Prefixes for numbers indicate location on the orbital (O) or lateral (L) surface, or the ventral (V), dorsal (D) or rostral (R) part of area 46. OPAll, OPro, TE, TE1, TE2, TEm, TEa and TS also indicate architectonic areas; sts, superior temporal sulcus. Other letter combinations refer to cases (see Results for details).

Termination of Projections from Prefrontal Cortices in Medial and Inferior Temporal Areas

Anterograde label was evident in medial and/or inferior temporal cortex in all cases with tracer injections in caudal orbitofrontal area OPro, more rostral orbital areas 11 and O12, and lateral prefrontal area V46 (Table 1). In the three cases with injections in area OPro (cases AG, AF and MAR; Fig. 1*B*), anterograde label was seen in medial temporal areas 28 and 35, and in area 36 in two of these cases (AG and AF). In case AF, additional anterograde label was apparent in the anterior portions of the subdivisions of area TE. No anterograde label was noted in the caudally situated medial temporal areas TH or TF in any of the cases.

In the four cases with injections in more rostral orbital areas 11 or O12 (cases MBJ, MFT, AM and MBY; Fig. 1*B*), anterograde label was present in area TE1 (Table 1). Other areas containing anterograde label in at least two of these cases included other subdivisions of area TE, the ventral temporal pole, and areas 35 and 36.

Table 1

Distribution of labeled terminals in the anterior temporal cortex after injection of anterograde tracers in prefrontal areas

Label site	Injection site (case)								
	OPro/OPAll (AG)	OPro (AF)	OPro (MAR[a])	11 (MBJ)	11 (MFT[a])	11 (AM)	O12 (MBY)	V46 (MBH)	V46 (MFF[a])
Area 28	+	+	+	–	–	–	–	–	–
Area 35	+	+	+	+	+	–	+	–	–
Area 36	+	+	–	+	–	–	+	–	–
Temp. pole	+	–	–	–	+	+	+	–	–
Area TE1	+	+	–	+	+	+	+	–	–
Area TE2	–	+	–	+	–	–	+	+	–
Area TEm	–	+	–	+	+	–	+	+	+
Area TEa	–	+	–	+	+	–	+	+	+

–, no label; +, light–moderate label; ++, moderate–heavy label.

[a]Cases with injections of [^{3}H]amino acids; all other cases were injected with WGA-HRP.

In the two cases with injections in lateral area V46 (cases MBH and MFF; Fig. 1*A*), the distribution of anterograde label was restricted to areas TE2, TEm and TEa, with the heaviest concentration in area TEa (Table 1).

Table 2
Distribution of anterograde label in prefrontal areas after tracer injections in anterior temporal cortices

Label site	Injection site (case)	
	Area 36 (AT)	Area TE1/TS1 (AV)
Area OPAll	+	–
Area OPro	+	+
Area 13	+	+
Area O25	+	–
Area M25	+	+
Area O12	+	+
Area O14	–	+
Area M14	–	+
Area 11	–	+
Area L12	–	+
Area R46	–	+
Area L10	–	+

–, no label; +, light–moderate label; ++, moderate–heavy label.

Termination of Projections from Medial and Inferior Temporal Areas in Prefrontal Areas

Axonal terminations were evident in prefrontal cortices in the two cases with injections in medial temporal area 36 or inferior temporal area TE1 (Table 2). In the case with an injection in area 36 (case AT), terminal axons with BDA labeled synaptic boutons were seen primarily in basal prefrontal areas, including medial area 25 (M25) and orbital areas 13 and OPro. Fewer sites with labeled boutons were observed in areas OPAll, O25 and O12. In case AV, the injection was located in area TE1 and extended somewhat into area TS1 (Fig. 1*A*). Like in case AT, labeled boutons were observed primarily in ventral prefrontal cortices (Table 2), and were most densely distributed in orbital areas OPro, O12 and O14 and in area M25. Other prefrontal areas with labeled boutons included areas M14, 11, R46, 13, L12 and L10.

Laminar Pattern of Termination of Prefrontal Axons in Temporal Cortices

Our goal was to investigate whether the laminar patterns of axonal termination depend on the laminar organization of the cortex of origin as well as the area of termination. We first examined connections from prefrontal to temporal cortices and asked whether axons from a single prefrontal area terminated in the same or different pattern in distinct, though adjacent,

Table 3
Laminar distribution of labeled terminals in the anterior temporal cortex after injection of anterograde tracers in prefrontal areas

Label site	Injection site (case)								
	OPro/OPAll (AG)	OPro (AF)	OPro (MAR)	11 (MBJ)	11 (MFT)	11 (AM)	O12 (MBY)	V46 (MBH)	V46 (MFF)
Area 28	(1)	(2)	(3)						
Layer 1	5	14	6						
Layers 2/3	25	13	15						
L. dissecans	40	40	58						
Layers 5/6	30	33	20						
Area 35	(1)	(9)	(5)	(2)	(2)		(3)		
Layer 1	0	38	38	0	0		3		
Layers 2/3	0	16	18	25	17		20		
Layers 5/6	100	46	44	75	83		77		
Area 36	(2)	(2)		(1)			(2)		
Layer 1	0	36		0			4		
Layers 2/3	0	13		7			24		
Layers 4–6	100	51		93			72		
Temp. pole	(3)				(2)	(1)	(2)		
Layer 1	33				35	27	24		
Layers 2/3	11				23	9	22		
Layers 4–6	56				42	64	54		
Area TE1	(3)	(5)		(7)	(5)	(2)	(3)		
Layer 1	65	46		43	30	24	11		
Layers 2/3	21	16		28	18	18	36		
Layer 4	7	25		17	21	51	35		
Layers 5/6	8	13		12	31	7	18		
Area TE2		(1)		(8)			(1)	(2)	
Layer 1		58		15			28	49	
Layers 2/3		35		31			47	30	
Layer 4		0		24			20	11	
Layers 5/6		6		29			5	10	
Area TEm		(3)		(1)	(3)		(2)	(7)	(2)
Layer 1		34		51	28		18	38	52
Layers 2/3		14		28	20		27	34	8
Layer 4		25		9	18		24	11	20
Layers 5/6		27		12	34		32	17	20
Area Tea		(4)		(4)	(3)		(10)	(9)	(15)
Layer 1		45		28	49		28	38	41
Layers 2/3		15		51	20		32	21	17
Layer 4		20		10	13		21	19	21
Layers 5/6		20		11	18		19	22	21

Data are expressed in percentages of total anterograde label for each area. Numbers in parentheses are the total number of sites of anterograde label that were observed for each area.

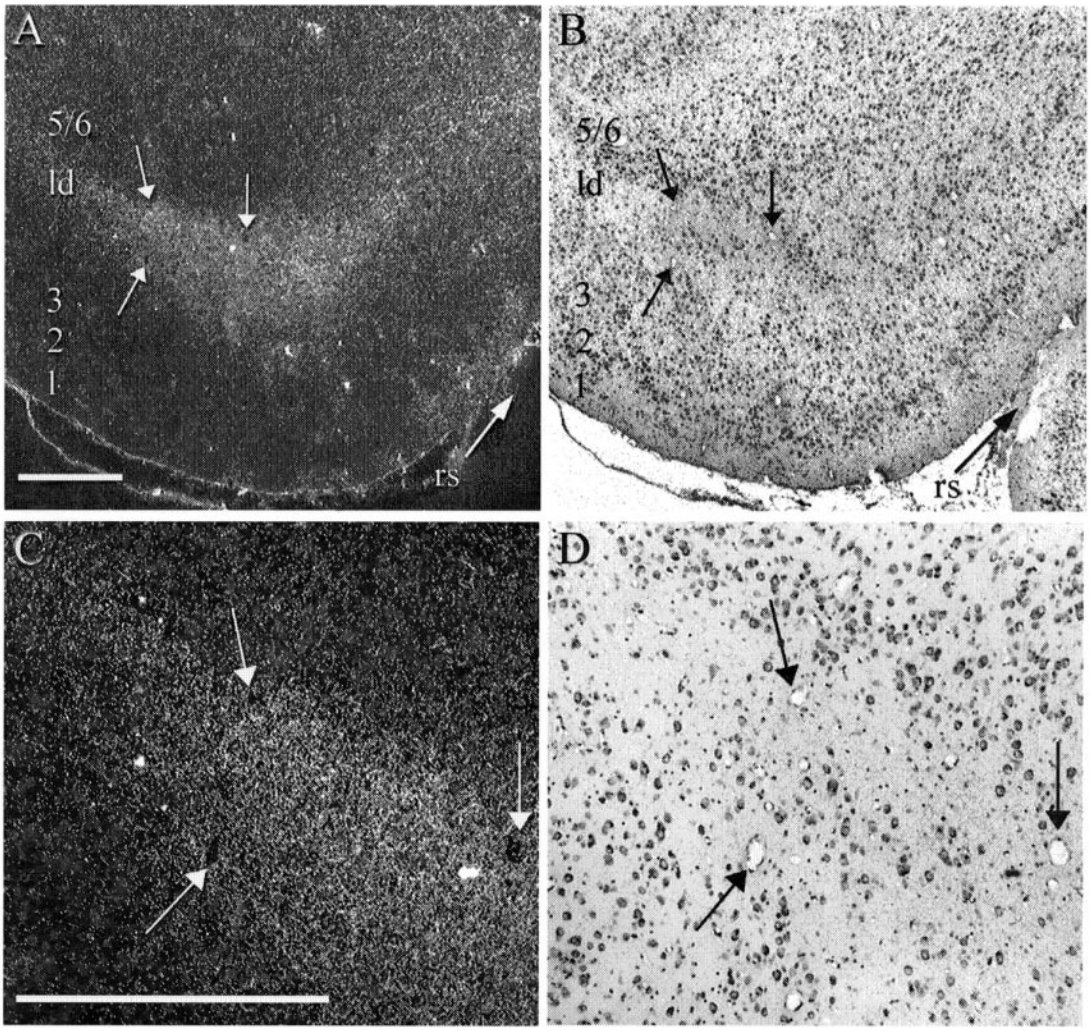


Figure 2. Projections from prefrontal area OPro terminated in area 28 (entorhinal cortex). (*A*) Darkfield image of a coronal section showing anterograde label (white grain) in the lamina dissecans of area 28 after injection of [^{3}H]amino acids in orbital area OPro (case MAR). (*B*) Brightfield image of the same Nissl-stained coronal section. (*C*) Higher magnification darkfield image of the same section showing anterograde label. (*D*) Higher magnification brightfield image of the same section showing the cell sparse lamina dissecans. Arrows indicate matching blood vessels in all images. Numbers indicate cortical layers, and are positioned at the start of each layer in these and the following images. ld, lamina dissecans; rs, rhinal sulcus. Scale bars, 500 μm. The scale bar for *A* applies to *B*; that for *C* applies to *D*.

temporal areas. We addressed this question by examining the pattern of termination in medial and inferior temporal cortices after injection of anterograde tracers in three regions of the prefrontal cortex, including caudal orbitofrontal, rostral orbitofrontal and lateral prefrontal areas (Fig. 1*A–D*). These areas represent the spectrum of cortical structural types in the prefrontal region: the caudal orbitofrontal region includes agranular and dysgranular areas, the rostral orbitofrontal region includes granular areas, and the lateral region includes granular areas that are distinguished from the orbital granular areas by a thicker layer 4 and overall more distinct laminar borders (Fig. 1*D*).

In the first group of cases with injections in dysgranular area OPro in the caudal orbitofrontal region, we noted three general laminar termination patterns in anterior temporal cortices: (i) a majority of anterograde label in the deep layers; (ii) a column of anterograde label relatively equally distributed between upper and deep layers; and (iii) a majority of anterograde label in the upper layers, particularly layer 1. Label in area 28 fell into the first category, and was most heavily, though not exclusively, distributed in the deep layers (Table 3, Fig. 2). In particular, dense anterograde label was apparent in the lamina dissecans, the cell-free region between layers 3 and 5 in area 28 (Fig. 2*A*,*C*). In contrast, in areas 35 and 36 label was distributed in the upper and deep layers. In area 35, label was evident particularly in layers 1 and 5/6 (Table 3, Fig. 3*A*,*B*). A similar pattern was observed in area 36, where anterograde label was detected in all layers, but was denser in layers 1 and 4–6 (Table 3). In a third pattern, label in inferotemporal areas TE1, TE2, TEm and TEa was noted predominantly in layer 1 and to a lesser degree in layers 2–6 (Table 3, Fig. 3*C*).

To test if the three patterns of termination described above were related to the cortical structure of the connected areas, we combined data from temporal areas that had anterograde label into three groups based on their general laminar architecture. For cases with injections in caudal orbitofrontal area OPro (cases AG, AF, MAR), we compared the relative proportion of anterograde label in the upper and deep layers for all sites within each of three groups of temporal areas representing different architectonic types: agranular cortex (area 28), dysgranular

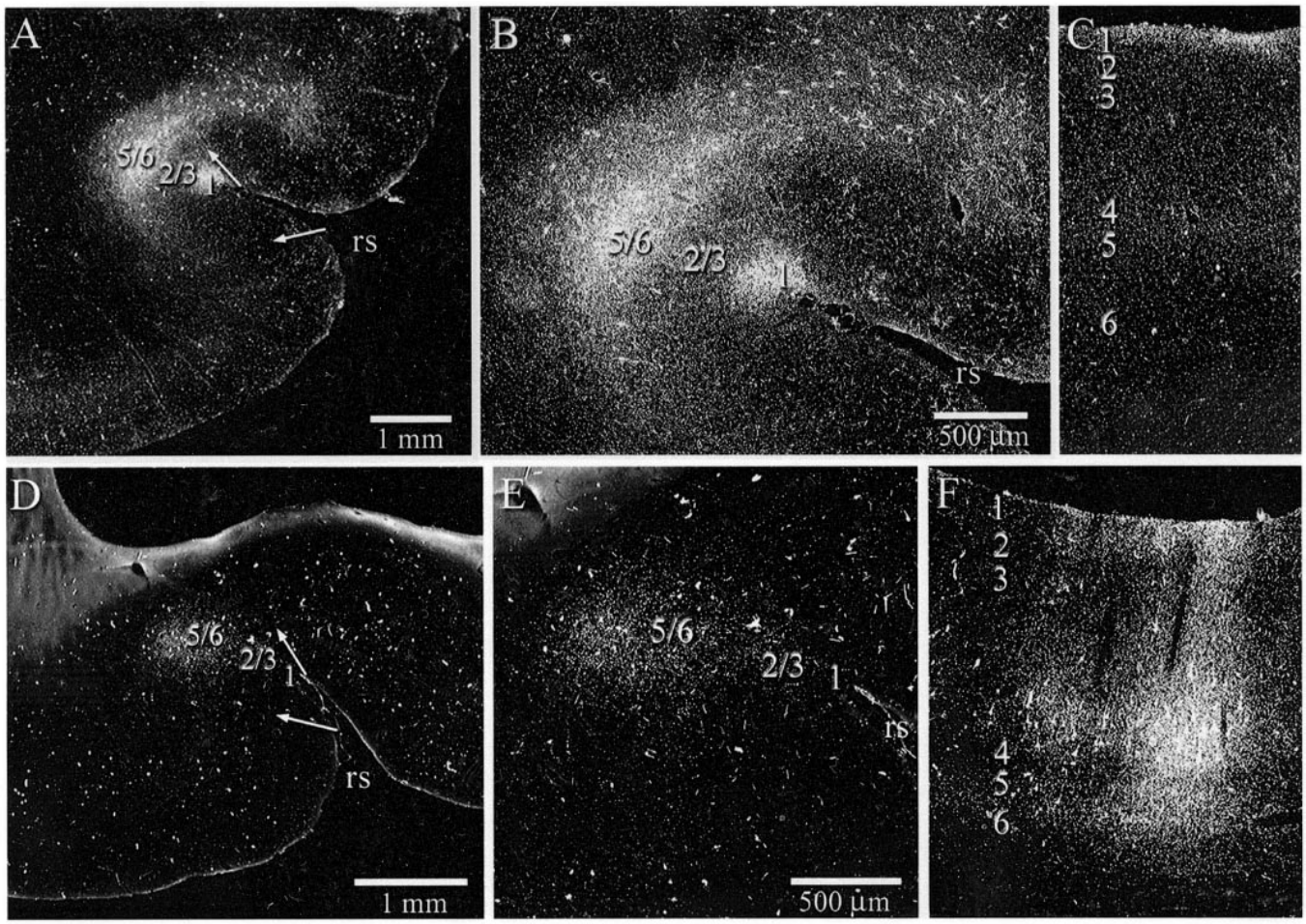


Figure 3. Comparison of projections from prefrontal areas OPro and O12 terminating in areas 35 and TEa. (*A*,*B*) Darkfield images of area 35 (between arrows in *A*) in the rhinal sulcus in case AF showing anterograde label (white grain) concentrated most heavily in layers 1 and 5/6 after injection of WGA-HRP in orbital area OPro. Retrogradely labeled neurons are visible as well. In *A*, area 36 is shown below and to the left of the lower arrow, and area 28 is visible to the right of the upper arrow. (*C*) Darkfield image of area TEa in the same case (AF) with anterograde label visible primarily in layer 1. (*D*,*E*) Darkfield images of area 35 (between arrows in *D*) in the rhinal sulcus in case MBY showing anterograde label (white grain) concentrated most heavily in layer 5/6 after injection of WGA-HRP in area O12. In *D*, area 36 is located to the left of the lower arrow and area 28 is shown to the right of the upper arrow. (*F*) Area TEa in the same case (MBY) with anterograde label distributed across all layers of cortex, and especially clustered in layers 1 and 3. Retrogradely labeled neurons are visible, particularly in the deep portion of layer 3. Numbers indicate cortical layer. The scale bar for *B* applies to *C*; that for E applies to F.

cortex (the ventral temporal pole, area 36, area 35) and granular cortex (areas TE1, TE2, TEa, TEm). We included area 35 with dysgranular cortices rather than agranular area 28 because, other than having no granular layer 4, the cortical structure of area 35 more closely resembles the dysgranular areas than area 28, which has a very distinctive appearance. An analysis of variance (ANOVA) for the percentage of anterograde label in the upper layers for the three groups was significant ($P < 0.01$); subsequent *t*-tests showed significantly less label in the upper layers in area 28 compared with areas 35 and 36 ($P < 0.01$), and in areas 35 and 36 compared with subdivisions of area TE ($P < 0.01$; Fig. 4).

The second group of prefrontal cases included injections in granular areas 11 and O12, situated in the rostral orbitofrontal region (Fig. 1*B*,*D*). Axonal termination in the medial and inferior temporal cortices fell into two general patterns. The first termination pattern was evident in medial temporal areas 35 and 36, in which anterograde label was predominantly found in the deep layers and avoided layer 1 (Table 3, Fig. 3*D*,*E*). In the ventral temporal pole label was more evenly distributed across cortical layers, although it was less densely distributed in layers 2–3 than in layers 4–6 or 1 (Table 3). The second termination pattern after injections in areas 11 and O12 was noted in areas TE1, TE2, TEa and TEm, where label was more equally distributed across the upper and deep layers (Table 3, Fig. 3*F*). Figure 5 shows the mean percentage of label in the upper and deep layers in all sites in areas 35, 36 and the temporal pole compared with sites in the subdivisions of area TE. The percentage of anterograde label in the upper layers of subdivisions of area TE was significantly higher than the percentage of label in the upper layers of areas 35, 36 and the temporal pole ($P < 0.001$).

The third group of prefrontal cases included injections in area V46 on the lateral surface of the frontal lobe (Fig. 1*A*,*D*). A particularly interesting finding pertained to the pattern of anterograde label in the subdivisions of area TE. Some inferotemporal areas span a long rostrocaudal distance, and we observed variations in the pattern of termination in cases where anterograde label spanned this distance. For example, in the two cases with an injection in area V46 (cases MFF and MBH), anterograde label was distributed across a considerable rostrocaudal distance of area TEa. In these cases in rostral area TEa label was noted in the upper and deep layers, and was most densely distributed in layers 2, 3 and 4 (Figs 6*A* and 7). In contrast, in

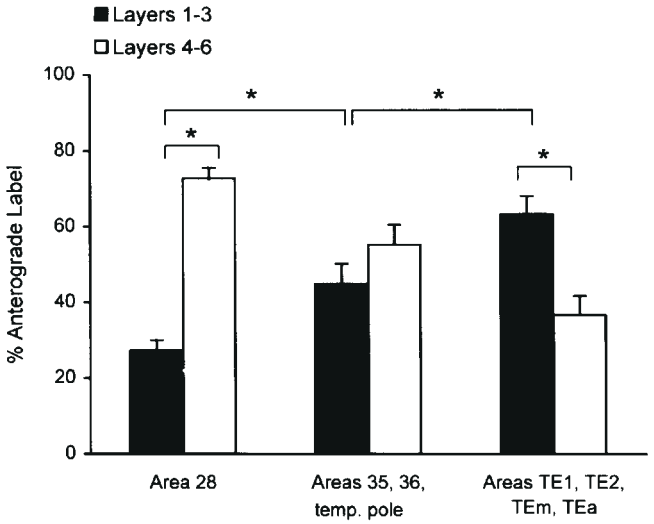


Figure 4. The laminar distribution of anterograde label after injections in caudal orbitofrontal area OPro differed in three groups of anterior temporal cortices. Anterograde label was heavier in the deep layers of area 28, equally distributed in the upper and deep layers of areas 35, 36 and the dysgranular cortex of the ventral temporal pole, and more dense in the upper layers of inferotemporal areas (TE1, TE2, TEm and TEa). Bars indicate the mean percentage of total anterograde label in the upper (1–3; black bars) and deep (4–6; white bars) layers of three cases with injections in caudal orbitofrontal area OPro (cases AG, AF and MAR). In all bar graphs (Figs 4–5, 7, 9 and 11), vertical lines on the bars indicate the standard error. Asterisks indicate statistically significant differences between the percentage of label in the upper and deep layers in a single group of areas or between the percentage of label in the upper layers of two different groups of areas.

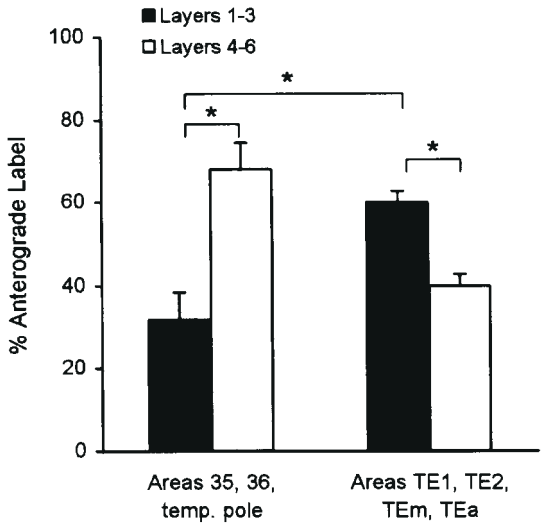


Figure 5. The laminar distribution of anterograde label after injections in rostral orbitofrontal areas 11 and O12 differed in two groups of anterior temporal cortices. Anterograde label was heavier in the deep layers of areas 35, 36 and the dysgranular cortex of the ventral temporal pole, and more dense in the upper layers of inferotemporal areas (TE1, TE2, TEm and TEa). Bars indicate the mean percentage of total anterograde label in the upper (1–3; black bars) and deep (4–6; white bars) layers of four cases with injections in orbital area 11 and O12 (cases MBJ, MFT, AM and MBY).

caudal area TEa label was noted primarily in layer 1, and to a lesser degree in layers 4–6 (Figs 6*B* and 7). The percentage of label in the upper layers differed significantly between the rostral and caudal area TEa sites ($P < 0.05$).

In a second series of analyses to investigate the relationship of cortical structure to the laminar pattern of termination we asked the converse question: When several prefrontal areas with distinct cortical structure project to a single temporal area, is their pattern of termination similar or different? We found that projections that originated in different prefrontal areas and terminated in the same temporal area frequently showed different patterns of anterograde label (Table 3). For example, after injections in dysgranular area OPro anterograde label was distributed relatively equally in the upper and deep cortical layers in area 35, and was concentrated most densely in layers 1 and 5/6 (Fig. 3*B*). In contrast, after injections in granular area O12 anterograde label was located in the deep layers of area 35 and avoided layer 1 (Fig. 3*E*). The mean percentage of label in the upper layers for all sites in area 35 was greater after injections in area OPro than after injections in areas 11 and O12 ($P < 0.05$; Table 3). Similarly, after injections in area OPro anterograde label in area TE1 was found predominantly in the upper layers, especially layer 1. In contrast, after injections in areas 11 or O12 label was more equally distributed between the upper and deep layers of area TE1 (Table 3), although the difference between these two groups did not reach statistical significance ($P > 0.05$).

Laminar Pattern of Termination of Temporal Axons in Prefrontal Cortices

The second phase of this study examined whether the laminar termination pattern of the reciprocal projections followed the same rules based on the cortical structure of the connected areas. We began by investigating whether axons from a single temporal area terminated in different patterns depending on the cortical structure of their prefrontal destination (Table 4). Evidence was obtained from two cases with injections in structurally distinct medial and inferior temporal areas. As with the prefrontal cases, these areas represent different types of cortex (Fig. 1*C*,*D*). In the first case, after injection in dysgranular area 36 (case AT), axonal terminations were distributed relatively equally across the upper and deep layers of dysgranular orbital area OPro (Fig. 8*A*). In contrast, in granular area O12, axonal terminations were essentially limited to layers 1–3 (Fig. 8*B*,*C*). To address the question of whether the pattern of termination is related to the cortical structure of the destination cortex, we divided the sites of label into three groups that included agranular, dysgranular and granular cortices, as described previously (Barbas and Rempel-Clower, 1997). In agranular areas (area OPAll) axonal terminations were distributed nearly equally in the upper and deep layers, in dysgranular areas (areas OPro, 13, O25 and M25) they were somewhat denser in the upper layers and in granular area O12 terminations were essentially limited to the upper layers (Fig. 9). An ANOVA for the three groups was significant ($P < 0.01$), and subsequent *t*-tests demonstrated a lower percentage of label in the upper layers in agranular than in dysgranular prefrontal areas ($P < 0.05$) and in dysgranular than in granular prefrontal areas ($P < 0.001$).

In a second case with an injection in granular area TE1/TS1 (case AV), the pattern of termination was also related to the structure of the destination cortex. In dysgranular prefrontal areas (areas OPro, 13 and M25), axonal terminations tended to be more densely distributed in the deep layers (Fig. 10*A*,*B*). In contrast, a higher proportion of terminations was observed in the upper layers of granular prefrontal areas (Fig. 10*C*,*D*). We divided the granular prefrontal areas with label into two groups according to our previously reported divisions (Barbas and Rempel-Clower, 1997). The first group included granular cortices on the rostral orbital surface (areas O14, M14, O12 and

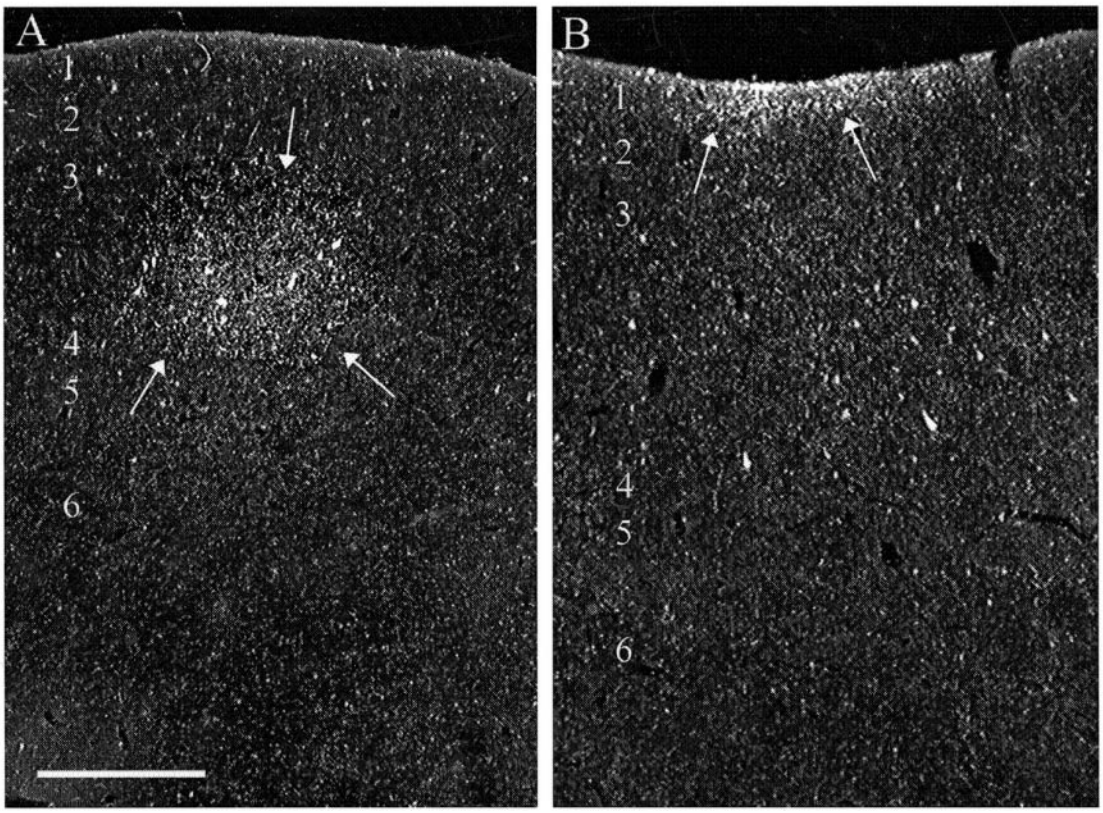


Figure 6. Projections from prefrontal area V46 terminated most densely across layers 2, 3 and 4 in the rostral part of area TEa but terminated primarily in layer 1 of the caudal part of area TEa. (*A*) Darkfield image of the rostral part of area TEa showing anterograde label (white grain) concentrated most heavily in layers 2–3 after injection of WGA-HRP in prefrontal area V46 (case MBH). (*B*) Darkfield image of the caudal part of area TEa in the same case (MBH) with anterograde label seen primarily in layer 1. Arrows indicate location of anterograde label. A few retrogradely labeled neurons are visible in both images. Numbers indicate cortical layer. Scale bar, 500 μm.

11) and the second group of granular cortices included those on the lateral surface (areas L12, L10 and V46). Compared with lateral granular areas, orbital granular areas have a narrower layer 4 and less laminar distinction (Fig. 1*D*). The highest proportion of labeled terminals in the upper layers (1–3) was seen in the lateral areas, followed by the granular orbital areas, and the least was noted in the dysgranular orbital areas (Fig. 11). An ANOVA showed a significant difference for the three groups ($P < 0.01$), and subsequent *t*-tests showed differences between the percentage of label in the upper layers in dysgranular cortices and the first group of granular cortices ($P < 0.01$), and between the two groups of granular prefrontal cortices ($P < 0.05$).

Finally, we addressed the converse question, i.e. whether the laminar pattern of termination in prefrontal areas was related to the cortical structure of the temporal area of origin. After anterograde tracer injections in temporal area 36 (case AT) and area TE1/TS1 (case AV), labeled fibers were observed in some of the same prefrontal areas, but in different laminar patterns. For example, axons from dysgranular area 36 terminated equally in the upper and deep layers of orbitofrontal area OPro (Fig. 8*A*; $P > 0.05$); however, axons from area TE1/TS1 terminated more densely in the deep layers of area OPro (Fig. 10*A*,*B*; $P < 0.01$). The opposite pattern was observed in area O12. After injection in area 36, axons terminated predominantly in the upper layers of area O12 (Fig. 8*B*,*C*; $P < 0.001$), whereas after injection in area TE1/TS1 axons terminated more evenly across the upper and deep layers of area O12 (Fig. 10*D*; there was no difference between the percentage of label in upper and deep layers,

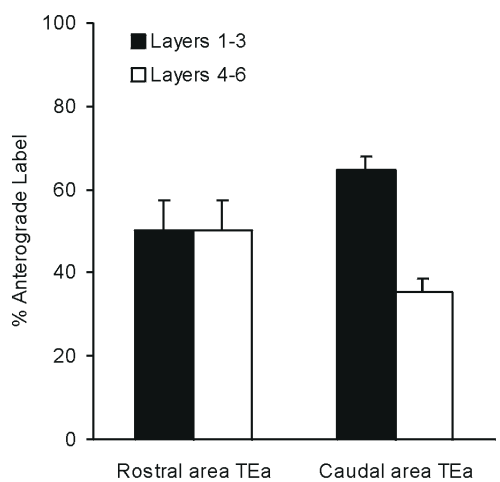


Figure 7. The laminar distribution of anterograde label after injections in prefrontal area V46 differed in the rostral and caudal parts of area TEa. Anterograde label was equally distributed in the upper and deep layers of rostral area TEa, whereas label was more dense in the upper layers of the caudal part of area TEa. Bars indicate the mean percentage of total anterograde label in the upper (1–3; black bars) and deep (4–6; white bars) layers of two cases with injections in prefrontal area V46 (cases MBH, MFF).

$P > 0.05$). The two other prefrontal areas that showed axonal terminations in both cases, areas 13 and M25, also revealed different termination patterns (Table 4). Efferent fibers from

Table 4
Laminar distribution of anterograde label in the prefrontal cortex after injection of tracers in anterior temporal areas

Injection site	Label site											
	OPAll[a]	OProb	13[b]	O25[b]	M25[b]	O12	O14	M14	11	L12	V46	L10
Area 36	(4)	(9)	(8)	(3)	(18)	(3)	(0)	(0)	(0)	(0)	(0)	(0)
Layer 1	24	19	29	36	34	46						
Layers 2/3	22	28	26	35	33	48						
Layer 4	–	–	–	–	–	2						
Layers 5/6	54	53	45	29	33	4						
Area TE1/TS1	(0)	(10)	(1)	(0)	(7)	(11)	(5)	(3)	(2)	(1)	(2)	(1)
Layer 1		13	16		34	25	42	43	53	17	46	43
Layers 2/3		19	10		21	33	24	16	45	51	37	39
Layer 4		–	–		–	13	13	7	1	6	5	7
Layers 5/6		68	74		45	29	21	34	1	26	12	11

Data are expressed in percentages of total anterograde label for each area. Numbers in parentheses are the total number of sites of anterograde label that were observed for each area.

[a]Areas in which there was no layer 4.

[b]Areas in which layer 4 was not clearly delineated, and thus measurements were taken across the region of layers 4–6 and listed in the row marked layers 5/6.

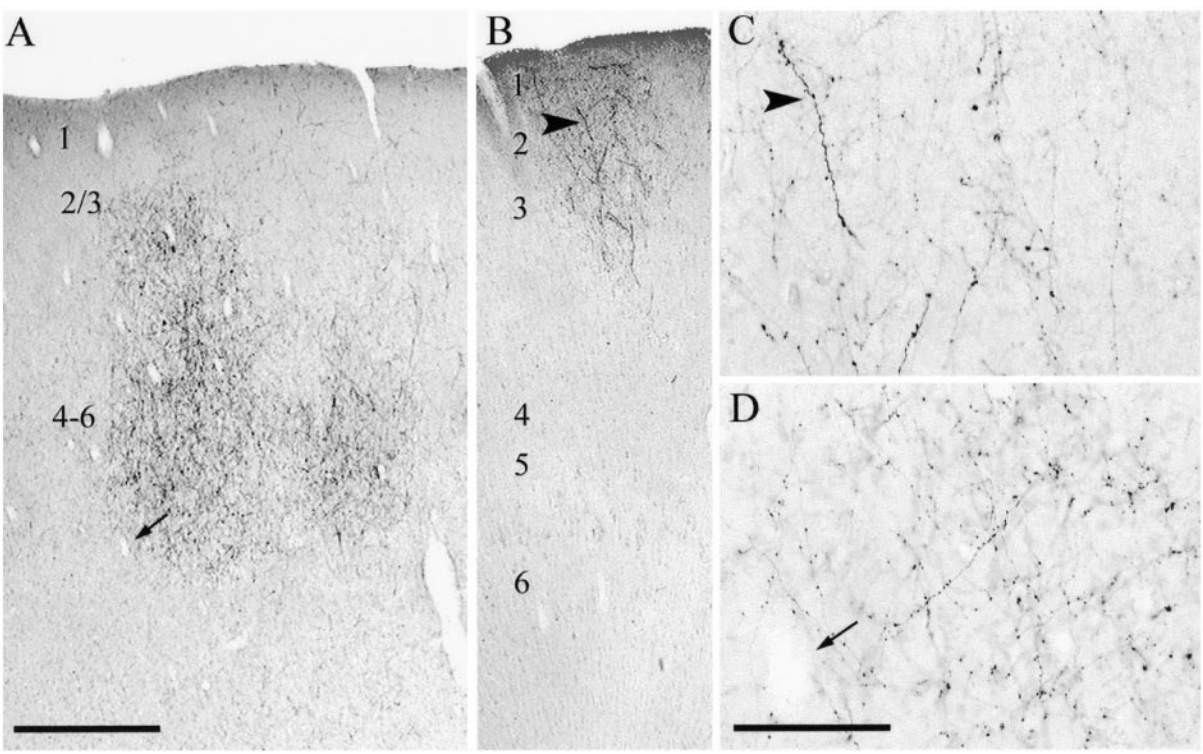


Figure 8. Projections from temporal area 36 terminated across all layers of prefrontal area OPro and terminated mostly in the upper layers of prefrontal area O12. (*A*) Brightfield image of prefrontal area OPro showing axonal terminations (black fibers) distributed across cortical layers 2–6 after injection of BDA in temporal area 36 (case AT). (*B*) Brightfield image of prefrontal area O12 in the same case (AT) with labeled terminals visible primarily in layers 1–3. (*C*) Higher magnification showing labeled synaptic boutons on fibers in *B*. (*D*) Higher magnification showing labeled synaptic boutons on fibers in *A*. Arrows indicate the same blood vessel in *A* and *D;* arrowheads indicate the same labeled fiber in *B* and *C*. Numbers indicate cortical layer. Scale bar in *A* is 500 μm and applies to *B*. Scale bar in *D* is 100 μm and applies to *C*.

dysgranular area 36 terminated in all layers of dysgranular area 13, with slightly more fibers targeting the upper layers than the deep, whereas fibers from granular area TE1/TS1 terminated predominantly in the deep layers of area 13. In dysgranular area M25, the same proportion of labeled fibers from area 36 and area TE1/TS1 terminated in layer 1. However, the pattern of termination in layers 2–6 of area M25 differed for these two injection sites. In the case with an injection in area 36, the same proportion of labeled fibers was observed in layers 2/3 as in layers 4–6, but after the injection in area TE1/TS1, more axonal terminations were observed in layers 4–6 than in layers 2/3.

Discussion

The Laminar Pattern of Termination is Related to Cortical Structure

The present findings demonstrated that the laminar pattern of termination of connections between prefrontal and temporal

areas is related to the cortical structure of both the origin and the target areas. As a consequence, projections arising from the same origin terminated in different laminar patterns in distinct target areas. Conversely, the laminar termination pattern in a single area differed for projections arising from different origins. A simplified summary of the patterns of connections is illustrated in Figure 12. Our findings are generally consistent with previous reports of laminar termination patterns for connections between these areas (Goldman-Rakic *et al.*, 1984; Seltzer and Pandya, 1989; Webster *et al.*, 1994).

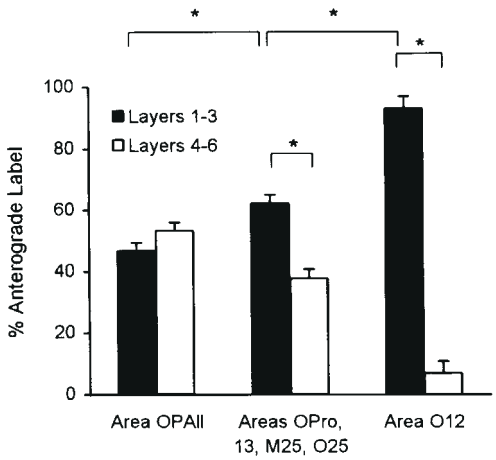


Figure 9. The laminar distribution of axonal terminations from temporal area 36 differed in three groups of prefrontal cortices. Labeled terminals were equally distributed in the upper and deep layers of agranular area OPAll, more densely distributed in the upper layers of dysgranular areas OPro, 13, M25 and O25, and nearly restricted to the upper layers of granular area O12. Bars indicate the mean percentage of total anterograde label in the upper (1–3; black bars) and deep (4–6; white bars) layers in case AT.

An important finding of this study is that a given prefrontal area does not have a common mode of anatomic communication with temporal areas. Rather, axons from one prefrontal area terminated in different layers when targeting structurally distinct temporal cortices. For example, projections from dysgranular orbitofrontal area OPro terminated predominantly in the deep layers of agranular temporal area 28, in all layers of dysgranular area 36 and mostly in the upper layers of granular area TE (Fig. 12*A*). Moreover, projections from one prefrontal area can terminate in different laminar patterns in a single large temporal area with variations in its cortical structure, as we saw along the rostro-caudal extent of area TEa. The laminar definition and prominence of granular layer 4 gradually increases from the rostral to caudal parts of the temporal lobe, and this is apparent within the borders of area TEa. Efferent fibers from V4 terminated equally in the upper and deep layers of rostral TEa, whereas they terminated predominantly in the upper layers of caudal TEa. Similarly, for the reciprocal projections the pattern of axonal termination differed when a single temporal area targeted distinct prefrontal cortices. Thus, axons from rostral area TE terminated preferentially in the deep layers of dysgranular caudal orbital areas OPro and 13, in all layers of granular

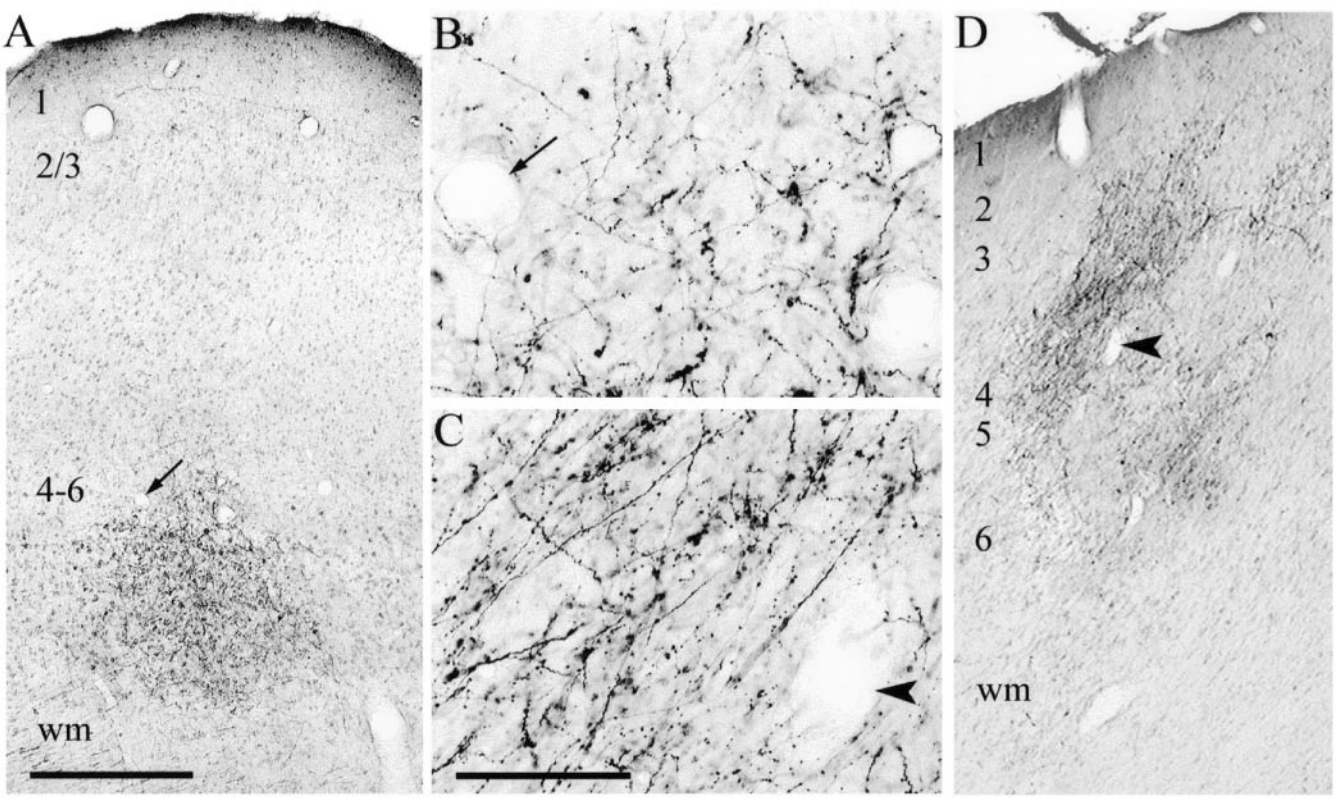


Figure 10. Projections from temporal area TE1/TS1 terminated primarily in the deep layers of prefrontal area OPro but terminated across all layers of prefrontal area O12. (*A*) Brightfield image showing axonal terminations concentrated most heavily in layers 4–6 of prefrontal area OPro after injection of BDA in temporal area TE1/TS1 (case AV). (*B*) Higher magnification showing labeled synaptic boutons on fibers in *A*. (*C*) Brightfield image of prefrontal area O12 in the same case showing labeled terminals and boutons in layer 3. (*D*) Lower magnification of the same section through prefrontal area O12 showing axonal terminations distributed across layers 2–6. Arrows in *A* and *B* and arrowheads in *C* and *D* indicate the same blood vessels for reference. Numbers indicate cortical layer. Scale bar in *A* is 500 μm and applies to *D*. Scale bar in *C* is 100 μm and applies to *B*.

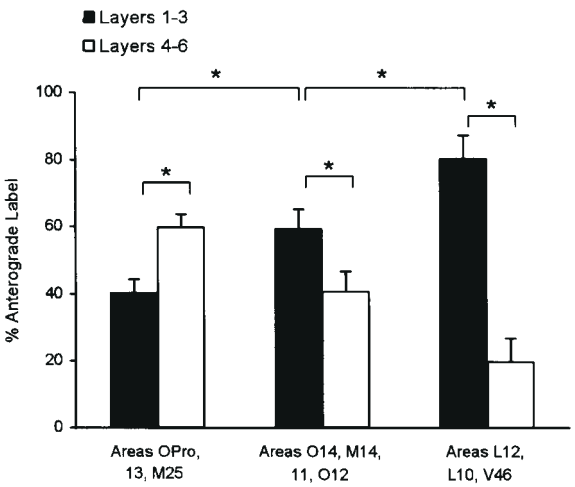


Figure 11. The laminar distribution of axonal terminations from temporal area TE1/TS1 differed in three groups of prefrontal cortices. Axonal terminations were more dense in the deep layers of dysgranular areas OPro, 13 and M25, whereas terminations were more dense in the upper layers of orbital and medial prefrontal granular areas (O14, M14, 11 and O12). In a second group of granular prefrontal cortices (areas L12, L10 and V46), which have a wider layer 4 and more laminar distinction, axonal terminations were predominantly found in the upper layers. Bars indicate the mean percentage of total label in the upper (1–3; black bars) and deep (4–6; white bars) layers in case AV.

rostral orbital area O12, and in the upper layers of granular areas V46 and L10 on the lateral surface, which have a distinct lamination (Fig. 12*B*).

The patterns of connections observed here can be summarized by a few rules based on the cortical structure of the interconnected areas. Thus, when areas with fewer than six layers and less laminar definition (i.e. agranular or dysgranular areas) projected to areas with six layers and more laminar definition, axons terminated predominantly in the upper layers. In contrast, axons terminated more densely in the deep layers of areas that have less laminar definition than the area giving rise to the projection. These termination patterns were more extreme for areas that differ substantially in cortical structure. Axonal termination in an area that was structurally similar to the area that issued the projection was relatively equally distributed in the upper and deep layers. The present findings are consistent with a previous study on the relationship of cortical structure to the pattern of interconnections between prefrontal areas (Barbas and Rempel-Clower, 1997). Further, our findings indicate that the rules based on cortical structure apply for connections linking distinct prefrontal cortices with the anterior temporal region.

There is evidence to suggest that similar rules guide the laminar pattern of connections between other association cortices. For example, projections from area 7a terminate mostly in layer 4 of rostral and intermediate parts of temporal area TPO, but span all layers in caudal area TPO (Cusick *et al.*, 1995), a pattern consistent with gradients in laminar definition in these areas [for a review see (Pandya *et al.*, 1988)]. Likewise, in the somatosensory system, efferent axons from the dysgranular insula terminate mostly in layer 1 of granular area S2 (Friedman *et al.*, 1986). In contrast, axons from the retroinsular area terminate predominantly in layers 4 and 6 of area S2 (Friedman *et al.*, 1986). Similarly, in the visual system Saleem and Tanaka (Saleem and Tanaka, 1996) showed projections from anterior dorsal area TE (TEad) that terminate most densely in layer 4 of dysgranular area 36, but in more lateral sectors of granular area TE (e.g. area TEm), the terminations increasingly target the upper layers.

The Laminar Pattern of Termination is Related to Function

In sensory cortices, the laminar pattern of termination has been associated with the functional nature of the connection in sensory processing. The projections from specific sensory thalamic relay nuclei terminating in and around layer 4 are considered forward [for a review see (Jones, 1985)]. Near the primary sensory areas, corticocortical forward projections also appear to terminate in and around layer 4, transmitting input from areas processing elementary and highly specific sensory information to areas that process integrated signals and a more global view of the sensory environment (Rockland and Pandya, 1979; Friedman *et al.*, 1986; Felleman and Van Essen, 1991). Corticocortical feedback projections between sensory areas proceed in the opposite direction and terminate most densely in layer 1, with sparser projections to the other layers (Rockland and Pandya, 1979; Friedman *et al.*, 1986; Felleman and Van Essen, 1991; Rockland and Van Hoesen, 1994). Forward and feedback type connections appear to represent the extreme patterns of corticocortical connections and apply best to early sensory areas. Axonal terminations that do not fit into the above patterns and are more or less distributed in a columnar pattern have been called 'lateral' connections (Felleman and Van Essen, 1991).

The availability of more sensitive anatomical tracing and quantitative analytic techniques in recent years has revealed a much wider spectrum of patterns of laminar termination than previously described. For example, in the visual cortices projections considered feedback terminate in the deep layers as well as the upper layers of neighboring areas, but in more distant areas they tend to terminate primarily in layer 1 [for a review see (Salin and Bullier, 1995)]. In fact, it has been proposed that the differences in connections are quantitative rather than qualitative (Einstein, 1996; Barbas and Rempel-Clower, 1997). Thus, it is the relative proportion of axonal terminals in the upper and deep layers that differs between feedback and feedforward projections. The present evidence is consistent with the latter view and further suggests that the pattern of corticocortical connections changes gradually in concert with gradual changes in cortical structure seen in all cortical systems [for a review see (Pandya *et al.*, 1988)].

Laminar structure in many cases appears to be correlated with function, and this is particularly apparent in the visual cortices. Primary visual area V1 has a distinct laminar appearance, with a remarkably prominent layer 4, and in general, progressively rostral visual cortices have less distinct laminar borders [for a review see (Pandya *et al.*, 1988)]. Functionally, the more rostral areas, such as areas TE and 36, process more global features of visual input and visual memory (Tanaka, 1992; Gross, 1994; Eacott and Heywood, 1995; Nakamura and Kubota, 1996; Suzuki, 1996a; Gibson and Maunsell, 1997). Thus, the cortical rules based on structure coincide with differences in function (Felleman and Van Essen, 1991).

The laminar pattern of terminations is likely to have functional consequences, since axons terminating in the upper cortical layers influence a different population of neurons and processes than axons terminating in the deep layers. Laminar differences in morphology, receptors and neurochemical properties have been well documented in many cortical areas (De Lima *et al.*, 1990; Goldman-Rakic *et al.*, 1990; Hof *et al.*, 1995;

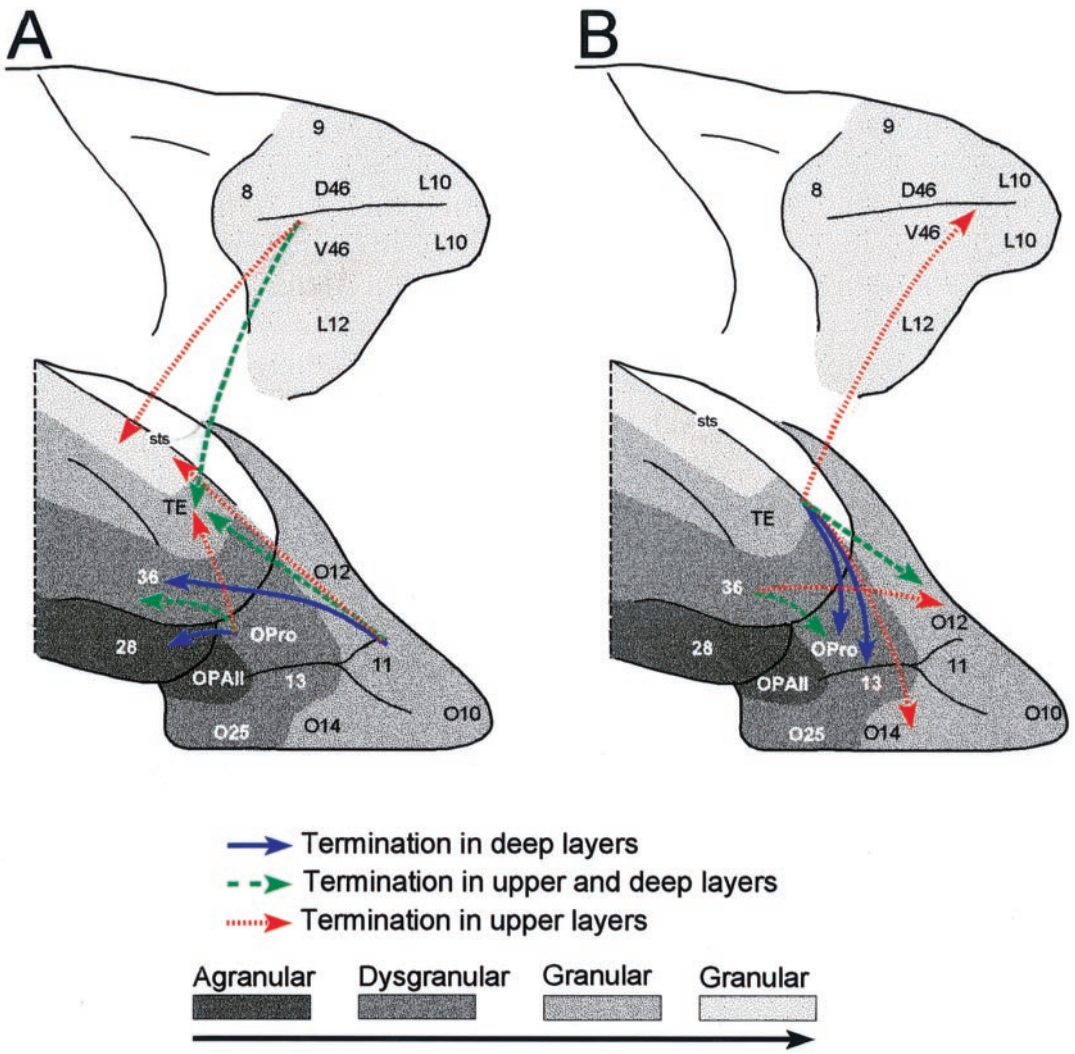


Figure 12. Simplified summary of the laminar pattern of termination of efferent fibers (*A*) from prefrontal to temporal areas and (*B*) from temporal to prefrontal areas, drawn on lateral (top) and ventral (bottom) views of the rhesus monkey brain. Shades of gray indicate cortical type with laminar definition increasing in the direction of the arrow. Two shades of light gray are used to indicate granular type cortex: the darker shade is used for areas with less laminar definition and a narrower layer 4. Termination predominantly into the deep layers (at least 60% of label in layers 4–6 in most sites) is illustrated by solid blue arrows. Termination predominantly into the upper layers (at least 60% of label in layers 1–3 in most sites) is illustrated by dotted red arrows. Termination distributed relatively equally between the upper and deep layers is illustrated with dashed green arrows.

Gabbott and Bacon, 1996). Thus far, the functional significance of different laminar patterns of connections in high-order association areas is, at best, indirect. However, there is strong evidence that the lateral prefrontal cortices affect task related activity of neurons in inferior temporal areas thought to be important for mechanisms of attention and memory for visual tasks (Fuster *et al.*, 1985; Desimone, 1996; Petrides, 1996; Rainer *et al.*, 1998; Miller, 1999; Tomita *et al.*, 1999). Neurophysiological evidence for such mechanisms was obtained for prefrontal area 46 in macaque monkeys engaged in a variety of delayed response tasks, and included enhanced cell firing when a stimulus matched the sample, delay activity that was selective for particular stimuli, and selective firing to relevant locations or to an anticipated target (Desimone, 1996; Miller *et al.*, 1996; Rainer *et al.*, 1998, 1999). Thus, the projections from area 46 to area TE may support active selection and comparison of stimuli held in short-term memory.

Further evidence for the importance of prefrontal influence on area TE for memory was provided in a recent study, where posterior commissurectomy in macaque monkeys prevented the inferior temporal cortex of one hemisphere from receiving bottom-up input from the opposite visual field (Tomita *et al.*, 1999). Recordings from single neurons in inferior temporal cortex showed stimulus selective activity that apparently came from feedback pathways from prefrontal cortex (Tomita *et al.*, 1999). In our own material, we observed that projections from lateral prefrontal area 46 terminated primarily in the upper layers of area TE, resembling a feedback pathway in early sensory cortices. In this context it is interesting that prefrontal area 8 also issues predominantly feedback-like projections to dorsolateral visual areas, such as areas MT and MST (Cusick *et al.*, 1995). Feedback pathways have been proposed to strengthen and focus the activity of neurons selective to particular stimuli or aspects of stimuli, and thus may be essential for attention and

identification of familiar stimuli (Sillito *et al.*, 1994; Ullman, 1995; Desimone, 1996; Payne *et al.*, 1996; Lamme *et al.*, 1998).

Projections from other prefrontal areas, including the orbitofrontal, also targeted predominantly the upper layers of area TE. It is reasonable to assume that orbitofrontal neurons convey somewhat different information than lateral prefrontal neurons to the upper layers of inferior temporal areas. Caudal orbitofrontal areas receive gustatory, olfactory, somatosensory and visual inputs as well as input from the amygdala (Barbas and De Olmos, 1990; Morecraft *et al.*, 1992; Barbas, 1993; Rolls and Baylis, 1994; Carmichael and Price, 1995), and have been implicated in reward-related behavior [for reviews see (Rolls, 1996; Watanabe, 1998)]. In fact, lesions of the orbitofrontal cortex in monkeys show impairments for tasks that require object-reward associations (Meunier *et al.*, 1997), and orbitofrontal neurons respond to stimuli that predict reward and appear to process the motivational value of rewarding outcomes (Tremblay and Schultz, 1999).

The orbitofrontal area OPro issued another prominent projection to the medial temporal entorhinal cortex, which is the gateway to the hippocampus [for a review see (Rosene and Van Hoesen, 1987)] associated with long-term memory (Leonard *et al.*, 1995; Nakamura and Kubota, 1995; Suzuki *et al.*, 1997). In contrast to the predominance of prefrontal projections to the upper layers of area TE, projections from area OPro terminated primarily in the deep layers of entorhinal cortex, targeting particularly the cell-free zone between layers 3 and 5. It is interesting to note that this termination pattern resembles a forward pathway in sensory areas, although its significance here is not clear. Studies of the physiologic interaction of the caudal orbitofrontal area with the entorhinal cortex are necessary to address the significance of the anatomic interaction of these areas, in general, and its specific laminar termination, in particular.

Finally, many of the connections between prefrontal and anterior temporal areas terminated in both the upper and deep layers, a pattern observed between areas with similar cortical structure (Fig. 12). Much remains to be known about this prevalent type of connection. An important issue that will need to be addressed before the functional significance of this or any other pattern of termination can be understood is that of timing. Specifically, are pathways to particular layers activated selectively in behavioral situations? Does input from a single area reach the upper and deep layers at the same time, or is the activity of neurons in one layer affected before the activity of neurons in other layers? On the basis of the pattern of connections, the present study suggests that the prefrontal cortex does not carry a monolithic dialogue, but several types of dialogues with anterior temporal cortices, whose significance must wait future physiological studies.

Notes

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