

MEDIAL PREFRONTAL CORTEX LESIONS ABOLISH CONTEXTUAL CONTROL OF COMPETING RESPONSES

J. E. HADDON AND A. S. KILLCROSS

CARDIFF UNIVERSITY

There is much debate as to the extent and nature of functional specialization within the different subregions of the prefrontal cortex. The current study was undertaken to investigate the effect of damage to medial prefrontal cortex subregions in the rat. Rats were trained on two biconditional discrimination tasks, one auditory and one visual, in two different contexts. At test, they received presentations of audiovisual compounds of these training stimuli in extinction. These compounds had dictated either the same (*congruent* trials) or different (*incongruent* trials) responses during training. In sham-operated controls, contextual cues came to control responding to conflicting information provided by incongruent stimulus compounds. Experiment 1 demonstrated that this contextual control of responding was not evident in individual rats with large amounts of damage that included the prelimbic and cingulate subregions of the prefrontal cortex. Experiment 2 further dissociated the result of Experiment 1, demonstrating that lesions specific to the anterior cingulate cortex were sufficient to produce a deficit early on during presentation of an incongruent stimulus compound but that performance was unimpaired as presentation progressed. This early deficit suggests a role for the anterior cingulate cortex in the detection of response conflict, and for the medial prefrontal cortex in the contextual control of competing responses, providing evidence for functional specialization within the rat prefrontal cortex.

Key words: prefrontal cortex, Stroop, conditional discrimination, response conflict, lever press, rat

Multiple rat and primate studies have provided evidence that subregions of the prefrontal cortex may be responsible for independent but complementary forms of cognitive processing (Goldman-Rakic, 1987; O'Reilly, Noelle, Braver, & Cohen, 2002; Owen, Evans, & Petrides, 1996; Petrides, 1994; Wise, Murray & Gerfen, 1996), drawing on data from anatomical, lesion, and electrophysiological recording studies.

Anatomically, the prefrontal cortex (PFC) frequently has been identified based on reciprocal projections to the mediodorsal nucleus of the thalamus, (Krettek & Price, 1977; Ray & Price, 1992; Rose & Woolsey, 1948). The rat frontal cortices can be divided into three principal areas: medial prefrontal cortex, orbital regions, and agranular insular cortex, with the medial prefrontal cortex seen as most analogous to primate dorsolateral prefrontal

cortex (Krettek & Price, 1977). The medial prefrontal cortex of the rat has been further subdivided into four areas: infralimbic and prelimbic cortices, and anterior cingulate and precentral areas (Krettek & Price, 1977; Öngür & Price, 2000; Ray & Price, 1992). Each subregion has a distinct relation with other cortical and subcortical areas (Condé, Maire-Lepoivre, Audinat, & Crepel, 1995; Hurley, Hernert, Moga, & Saber, 1991; Öngür & Price, 2000; Reep, Chandler, King, & Corwin, 1984; Sesack, Deutch, Roth, & Bunney, 1989), exhibited by dissociable afferent, efferent, and intrinsic pathways.

Lesion studies have further indicated functional specialization within the frontal cortices of the rat (see Kesner, 2000, for a review). Studies have shown that anterior cingulate and precentral cortex lesions produce deficits in visual-motor associative conditional discriminations (Winocur, 1991; Winocur & Eskes, 1998) but spare the acquisition of simple visual, spatial, or olfactory discriminations (Ragozzino, Wilcox, Raso, & Kesner, 1999). Similarly, combined prelimbic and infralimbic cortex lesions produce deficits on cross-modal switching (Ragozzino, Detrick & Kesner, 1999) but do not impair performance on a visual conditional associative task (Bussey, Muir, Everitt & Robbins, 1996). Evidence for a dou-

This research was funded by a UK BBSRC CASE studentship, sponsored by Merck, Sharp and Dohme UK, to J. E. Haddon, and by an Independent Investigator Award for the National Alliance for Research on Schizophrenia and Depression (Southwest Florida Investigator) to A. S. Killcross.

Address correspondence to A. S. Killcross, School of Psychology, Tower Building, Park Place, Cardiff University, Cardiff, CF10 3AT, United Kingdom (e-mail: KillcrossAS@cardiff.ac.uk).

doi: 10.1901/jeab.2005.81-04

ble dissociation between the prelimbic and infralimbic cortices in the contribution to operant performance also has been reported (Coutureau & Killcross, 2003; Killcross & Coutureau, 2003). Lesion studies in nonhuman primates similarly indicated functional specialization of prefrontal subregions. For example, Dias, Robbins, and Roberts (1997) observed dissociations between orbitofrontal and lateral prefrontal cortex lesions on a reversal and attentional-set-shifting task in marmosets.

Electrophysiological recording studies in nonhuman primates similarly have provided evidence for differentiation of function within the prefrontal cortex (Funahashi, Bruce, & Goldman-Rakic, 1989; Kojima & Goldman-Rakic, 1982, 1984; Watanabe, 1986). These single-cell recording studies demonstrate the highly specific properties of prefrontal neurons, with neurons responding exclusively to spatial, delay, and task-related information during performance of a spatial working-memory task.

In contrast, Duncan and Owen (2000) recently have suggested that evidence for functional specialization within the human prefrontal cortex is less clear. Functional imaging studies reveal activation of three common frontal regions (midsolateral, midventrolateral, and dorsal anterior cingulate cortex) in response to many different types of task demands. Systematic comparison of the frontal activations produced by five different types of task demands (response conflict [Stroop], task novelty, number of elements in working memory, working memory delay, and perceptual difficulty) demonstrated similar clusters of prefrontal activation for each of the tasks. Duncan and Owen suggested that much the same prefrontal subregions are activated by these different forms of task demand.

Moreover, Duncan and Owen (2000) argue against the evidence for functional differentiation in animals, suggesting that much of the research results are ambiguous. They argue that restricted lesions result in small deficits in tasks that tap into a broad range of task demands. A similarly ambiguous picture is seen with electrophysiological studies, with some research indicating that some prefrontal neurons can adapt their firing properties to match the current behavioral demands of the

task (Rao, Rainer, & Miller, 1997). Duncan and Owen also suggest that the differentiated architecture and connectivity of prefrontal subregions does not inevitably lead to functional differentiation, as this would result in functional modules consisting of multiple, small, widely distributed clusters.

A classic assessment of frontal function in humans is the Stroop task (MacLeod, 1991; Stroop, 1935). Patients with frontal damage have been found to be impaired at producing the task-appropriate response to stimulus compounds that provide conflicting response information. This is not due to a failure to produce appropriate stimulus-response mappings as performance on congruent trials remains unimpaired (Cohen & Servan-Schreiber, 1992; Perret, 1974; Vendrell, Jungue, Pujol, Jurado, Molet, & Grafman, 1995). In the Stroop task (MacLeod, 1991; Stroop, 1935), participants are required to read the word or name the color of stimulus compounds. These compounds are composed of either *congruent* stimulus-response mappings (in which both stimulus elements map onto the same response, that is, GREEN in green ink) or *incongruent* stimulus-response mappings (in which the stimulus elements map onto different responses, that is, GREEN in red ink). When incongruent compounds are presented (mapping onto two different responses), the task instructions (color naming or word reading) are required to disambiguate the conflicting stimulus information and enable production of the correct response. Typical findings indicate that producing the task-appropriate response to incongruent stimuli is more difficult when participants are required to name the color, as demonstrated by longer latencies to produce a response and increased errors (Stroop, 1935).

The Stroop task essentially can be viewed as an instructed conditional-discrimination task, in which specific stimuli indicate specific responses. Thus, following one task instruction (T1), a bivalent stimulus (S) evokes a particular response (R1), but following a different task instruction (T2) the same S now evokes a different response (R2). Thus correct responding is dependent on appropriate stimulus-response mappings and production of the correct, or task-appropriate, response in the presence of alternatives. Consistent with the idea of the Stroop task as an instructed

conditional discrimination, the prefrontal cortex has been implicated in conditional discrimination task performance in rats (Winocur, 1991; Winocur & Eskes, 1998), nonhuman primates (Murray, Bussey, & Wise, 2000; Petrides, 1982, 1985, 1991), and humans (Petrides, 1990, 1997; Petrides, Alivisatos, Evans, & Meyer, 1993), in particular when multiple responses are possible.

Evidence for a specific role for the anterior cingulate cortex on performance of the Stroop task has been provided by functional imaging research. Activation of the anterior cingulate cortex has been reported to be greatest during mismatched or incongruent trials (Carter, Mintun & Cohen, 1995; MacDonald, Cohen, Stenger, & Carter, 2000; MacLeod & MacDonald, 2000; Milham, Banich, & Barad, 2003), resulting in suggestions that this region of the prefrontal cortex is involved in tasks in which response conflict is high and/or task-appropriate responding must be produced in the presence of distracters. However, Duncan and Owen (2000) have pointed out a number of problems with the evidence indicating a specific role for the anterior cingulate cortex in Stroop interference. In particular, imaging studies are confounded as there are numerous reports of heightened activation in this region during congruent trials, which requires neither the detection of response conflict nor the use of task-appropriate responding, as well as of the activation of other prefrontal regions (dorsolateral and ventrolateral PFC) during performance of the same task. Patient studies also are problematic as few patients have focal damage limited to one prefrontal subregion alone.

The aim of this study was to establish the involvement of the prefrontal cortex on performance of tasks (like the Stroop) that involve the resolution of conflicting response information, and the possible specific involvement of the anterior cingulate cortex on trials that involve response conflict. This aim was achieved by investigating the effects of excitotoxic lesions of the medial prefrontal cortex (Experiment 1) and the anterior cingulate cortex (Experiment 2) in the rat on performance of a task designed to reflect some of the response-competition aspects seen in the human Stroop task.

Rats received training on two biconditional discrimination tasks (one auditory, one visual) in two contexts (C1 and C2). In context C1,

a press on one lever (LP1) during one auditory cue (A1) produced reinforcement; a press on the other lever (LP2) produced reinforcement during a different auditory cue (A2). Similarly, in context C2, reinforcement followed a lever press (LP1) during one visual cue (V1), whereas reinforcement followed a press on the other lever (LP2) during a different visual cue (V2). At test, rats were presented with audiovisual compounds of the training stimuli in each of the training contexts (C1 and C2), in extinction. These audiovisual compounds were either composed of stimulus elements that evoked the same lever press response during training (i.e., A1V1 and A2V2) or stimulus elements that evoked different responses during training (i.e., A1V2 and A2V1), termed congruent and incongruent stimulus compounds, respectively. As in the human Stroop task, responses during incongruent stimulus compounds were defined as correct or incorrect according to whether they were appropriate to the test context. For example, if presented with the incongruent stimulus compound, A1V2, in context C1, then the context appropriate response would be LP1 because in context C1, LP1 during auditory cue A1 had been followed by reinforcement.

Examination of performance on this task allows the detailed comparison of how cell loss within different prefrontal subregions relates to control of responding by both congruent and, especially, incongruent trial types, allowing for the examination of the relative involvement of prefrontal subregions, in particular the anterior cingulate cortex, in Stroop-like interference and/or response conflict. If the anterior cingulate cortex is responsible for performance on incongruent trials, then damage to this subregion alone should reflect the basic pattern of performance seen on incongruent trials, providing clear evidence for functional specialization within the prefrontal cortex. If there is no specialization of function, however, then the amount of overall damage to the medial prefrontal cortex should best relate to the degree of performance during test sessions.

EXPERIMENT 1

The Effect of Medial Prefrontal Lesions on Contextual Control of Biconditional Discriminations

The first experiment examined the effect of large excitotoxic lesions of the medial pre-

frontal cortex on the contextual control of conditioned responding, in particular examining the impact of differential damage to prefrontal subregions on the pattern of responding.

METHOD

Subjects

Twelve naive adult male hooded-lister rats (supplied by Harlan OLAC, UK) served in Experiment 1. The colony room housing the rats operated on a 12:12 hr light/dark cycle (lights on at 8:00 a.m.) and was maintained at a temperature of $21^{\circ}\text{C} \pm 1^{\circ}$ and humidity of $55\% \pm 5\%$. The rats were housed in pairs. Eight of the rats received pretraining excitotoxic lesions of the medial prefrontal cortex and 4 served as sham-operated controls. Following surgery, the rats were maintained at 85% of age-matched ad lib weights (range: 300 g to 350 g) and had free access to water.

Surgery and Histology

Surgery. Rats were first anaesthetized with Isoflurane, their heads shaved, and placed in a stereotaxic frame (Kopf Instruments, Tujunga, CA). An incision was made in the scalp and then a skull flap overlying the prefrontal cortex was drilled out. Injections of Ibotenic acid (63 mM, Sigma-Aldrich Co. Ltd., UK) were made with a 2 μl Hamilton syringe (Reno, NV) mounted on the stereotaxic frame. Injections of 0.5 μl at a rate of 0.1 $\mu\text{l}/\text{min}$ were made at four sites within the medial prefrontal cortex (AP: +2.7, +3.7, ML: ± 0.7 , DV: -4.5 and -4.0, respectively). Following each injection the needle was left in position for 10 min to allow absorption of the bolus and to minimize spread of the toxin into other brain regions. Sham-operated controls received an identical procedure with the exception that no toxin was infused. Following a minimum of a week's postoperative recovery, rats were gradually reduced to 85% of age-matched ad-lib weights.

Histology. Following completion of testing, rats were given a lethal overdose of sodium-pentobarbitone (Euthatal) and perfused with saline (0.9%) and formal-saline (10%, w/v). Brains were taken out and postfixed in formal saline and before cutting were transferred to a 25% sucrose solution in which they

remained for 24 hr. Slices (40 μm thick) were made using a cryostat (Leica Instruments) and were mounted onto gelatin-coated slides. These were subsequently dried, first at room temperature and then in an oven (at 40°C), before being stained with Cresyl-violet. The extent and location of cell loss were verified with a light microscope and the brain atlas of Paxinos and Watson (1998).

Apparatus

Eight operant chambers (30 cm wide by 24 cm deep by 21 cm high; supplied by Med Associates Inc., St Albans, VT), housed in sound attenuating chambers and arranged in a 2×4 array, were used. The chambers were housed in a room that remained dark during the experiment. Each chamber consisted of three aluminum walls and ceiling, with a Perspex door serving as the fourth wall. The walls and ceiling were lined with transparent Perspex behind which context cue "wallpapers" were fixed. Four of the chambers had wallpapers consisting of black and white checks, and the other four had white wallpaper with black dots. Each chamber had a floor constructed of 19 stainless steel rods (4.8 mm in diameter, spaced 1.6 cm apart). The chambers were illuminated by a 3-W houselight located at the top center of the left wall. Food pellets (45 mg; Formula A/I, P.J. Noyes, Lancaster, NH) were delivered into a recessed magazine located in the right wall of each chamber. Twenty percent (w/v) sucrose solution (made daily) was delivered via a dipper (vol = 0.1 ml) into the same magazine. Access to the magazine could be determined by means of infrared detectors mounted across the mouth of the recess. Two flat-panel retractable levers could be inserted to the left and right of the magazine. Two panel lights (diameter: 2 cm) were located in the right wall of the chamber, and a magazine light was located in the top of the magazine and illuminated blue. Auditory stimuli consisted of a 2 kHz tone and a 10 Hz train of clicks delivered from speakers located in the ceiling. Visual stimuli were flashing panel lights and steady panel lights plus the magazine light. A computer equipped with MED-PC[®] software (Med Associates Inc.) controlled the operant chambers and recorded the data.

Table 1
Experimental design used in all the experiments.

	Biconditional training	Test sessions	
		Congruent	Incongruent
C1	A1:LP1→O1, A2:LP2→O1	A1V1, A2V2	A1V2, A2V1
C2	V1:LP1→O2, V2:LP2→O2	A1V1, A2V2	A1V2, A2V1

Note. C1/C2, A1/A2, V1/V2, LP1/LP2, and O1/O2 refer to alternative experimental chambers (checked/spotted), auditory stimuli (tone/clicker), visual stimuli (steady/flashing lights), lever presses (right/left) and outcomes (pellets/sucrose), respectively.

Behavioral Procedure

Rats received two sessions per day throughout training, one in the morning in one context (checked or dotted wallpapers) and another session in the alternative context in the afternoon. In each of the different contexts, they received a different reinforcer; for example, in the checked context sucrose solution served as the reinforcer, whereas in the dotted context food pellets served as the reinforcer. This allocation remained consistent throughout, but was counterbalanced across animals.

Pretraining. On day 1, the rats received two sessions of magazine training, one in each context. Sessions lasted for 48 min and sucrose or a food pellet was presented approximately every 120 s. Following this there were 2 days (four sessions) of lever-press training. During these sessions, the rats received 24 lever presentations (12 each of the right and left levers), each lasting for 60 s. Session duration was again 48 min. On the 1st day of lever-press training, each press was reinforced; this was altered on the second day to a random-interval (RI) 15-s schedule.

Biconditional discrimination training. Table 1 shows the experimental design for all animals. Rats were trained concurrently, for a total of 14 days, on two biconditional discriminations, auditory and visual, in the two discriminable contexts (C1 and C2). Rats received two sessions a day, one of each discrimination. In context C1, rats received presentations of the auditory cues (A1 or A2) during which different lever presses (LP1 or LP2, respectively) led to reinforcement (O1). Similarly, lever presses (LP1 and LP2) led to reinforcement (O2) in the presence of visual cues (V1 and V2, respectively) in sessions in context C2. Correct responses were reinforced with pellets in one context and with sucrose in the other. Allocation of contexts, cues, levers, and re-

inforcers was counterbalanced as far as was possible. Sessions consisted of 24 trials (12 of each trial type; A1 and A2 or V1 and V2) with an intertrial interval (ITI) of 2 min. Stimulus presentations lasted 60 s. Reinforcement was unavailable during the first 10 s of this period (S1), and was available during the final 50 s (S2) on a RI 15-s reinforcement schedule. Both levers were presented at the start of each trial and were retracted during the ITI. Houselights were illuminated for the duration of the session.

Test sessions. Following acquisition of the biconditional discriminations, rats received two test sessions, one in each training context. Test sessions consisted of presentations of audiovisual compounds of the training stimuli, in extinction, that is, A1V1, A2V2, A1V2, and A2V1. There were four presentations of each trial-type and stimulus duration was again 60 s. Trial order was block randomized, with each trial-type being presented once in each block of four trials. Houselights were illuminated for the duration of each test session. There was one test session per day, with the context in which rats were first tested counterbalanced across animals.

Test stimuli. The test stimulus compounds were categorized as congruent or incongruent depending on the responses previously required during training (see Table 1). Congruent stimulus compounds were those comprised of elements that had required the *same* lever-press response during acquisition. For the example given above, both A1V1 and A2V2 compounds required the same lever-press responses in training, LP1 and LP2, respectively. Incongruent stimulus compounds were those composed of elements that required *different* lever press responses during initial training, A1V2 and A2V1. Correct responses to incongruent stimulus compounds were designated based on the stimulus ele-

ments previously trained in the test contexts. Thus, if the test session occurred in the context in which the auditory discrimination had been acquired, then the auditory stimulus element governed the designation of the correct response; similarly, if tested in the context in which the visual discrimination had been learned, then the visual stimulus elements were the relevant cues. Thus, if the incongruent stimulus compound A1V2 was presented in context C1 (in which the auditory discrimination was trained), then the correct response would be LP1 since this response previously was associated with the stimulus element A1. If the same compound was presented in context C2, in which the visual discrimination was acquired, then the correct response would be LP2 since this response previously had been associated with the stimulus element V2. Thus the test context is used to disambiguate the conflicting responses associated with the individual elements of the test compounds.

Behavioral measures. For both biconditional discrimination training sessions and test sessions, discrimination ratios (DR = number of responses on correct lever/total number of lever-press responses) were calculated for each animal.

RESULTS

Histological Analysis

Table 2 reports the extent of cell loss in various medial prefrontal subregions exhibited by the rats. Lesioned rats were categorized according to the extent of the damage to the



Fig. 1. The maximum and minimum extent of the damage to prelimbic and cingulate subregions of the medial prefrontal cortex. Outlines are reproduced from Paxinos and Watson (1998) and represent sections ranging from 2.2 to 4.7 anterior to bregma. The maximum amount of damage to the prefrontal cortex was observed in Rat R31 (HIGH P+C) and is depicted by the striped regions. The minimum amount of damage was observed in Rat R14 (LOW P+C) and is depicted by the grey regions.

preflimbic cortex and cingulate cortex, with rats showing greater than 70% cell loss in these regions categorized as HIGH P+C, and rats exhibiting less than 50% damage in these areas categorized as LOW P+C. Four rats were categorized as HIGH P+C, (Rats R23, R25, R29, R31) and 4 were categorized as LOW P+C (Rats R12, R14, R17, R21). Figure 1 represents the maximum and minimum extent of this damage to prelimbic and cingulate cortex subregions in rats classified as HIGH P+C and LOW P+C.

Table 2

The extent and location of cell loss within the medial prefrontal cortex for each rat in Experiment 1.

Group	Rat	Overall mPFC damage (%)	Combined prelimbic and cingulate cortex damage (%)	Prelimbic cortex damage (%)	Cingulate cortex damage (%)
HIGH P+C	23	73	96	92	100
	25	40	79	83	75
	29	56	87.50	100	75
	31	74	93.75	100	87.50
LOW P+C	12	41	49	67	31
	14	17	33	42	25
	17	17	33	42	25
	21	21	40	42	37.50

Note. Assessment of regional cell loss was based on the atlas of Paxinos and Watson (1998) and is given as a percentage. P = Prelimbic cortex and C = Cingulate cortex. HIGH P+C animals were those that exhibited greater than 70% combined damage to the cingulate cortex and prelimbic cortex, and LOW P+C animals were those that had less than 50% damage to these regions.

In the HIGH P+C group, overall medial prefrontal damage ranged from 40% (Rat R25) to 74% (Rat R31), with the combined prelimbic and cingulate damage ranging from 79% (Rat R25) to 96% cell loss (Rat R23). In the LOW P+C group, overall medial prefrontal damage was quite low ranging from 17% (Rats R14 and R17) to 41% cell loss (Rat R12). Combined damage to the prelimbic and cingulate cortex subregions ranged from 33% (Rats R14 and R17) to 49% (Rat R12).

Rat R25 (HIGH P+C) and Rat R12 (LOW P+C) had equivalent overall medial prefrontal damage (40% and 41%, respectively), but differed with respect to the amount of prelimbic (83% and 67%) and cingulate damage (79% and 49%), and thus served as an explicit comparison for the effect of damage to these prefrontal subregions.

Sham-operated controls (Rats R9, R11, R13, and R15) exhibited no cell damage within the medial prefrontal cortex.

Behavioral Analysis

Biconditional discrimination performance during training. Biconditional discrimination performance was assessed based on responses during the first 10 s of the stimulus presentation and is shown in Figure 2 (response rates are shown in Table 3). The discrimination ratio was derived from responding averaged across the two biconditional discriminations tasks and calculated based on responses during the first 10 s of the stimulus presentation because reinforcement was unavailable during this period, and therefore responding was uncontaminated by reinforcement. Acquisition of the biconditional discriminations progressed rapidly, with all animals demonstrating a discrimination ratio of more than 0.60 on the last day of training. Discrimination ratios ranged from 0.60 (Rat R23, HIGH P+C) to 0.84 (Rat R13, SHAM). Comparison of the discrimination ratios of the rats with equivalent overall medial prefrontal damage (Rat R25, HIGH P+C and Rat R12, LOW P+C) indicated roughly comparable responding, with discrimination ratios of 0.77 and 0.70, respectively, indicating high levels of performance in both animals regardless of the degree of damage to the prelimbic and cingulate cortices.

Test session performance. Test performance was averaged across both test sessions because a similar pattern of responding was seen in

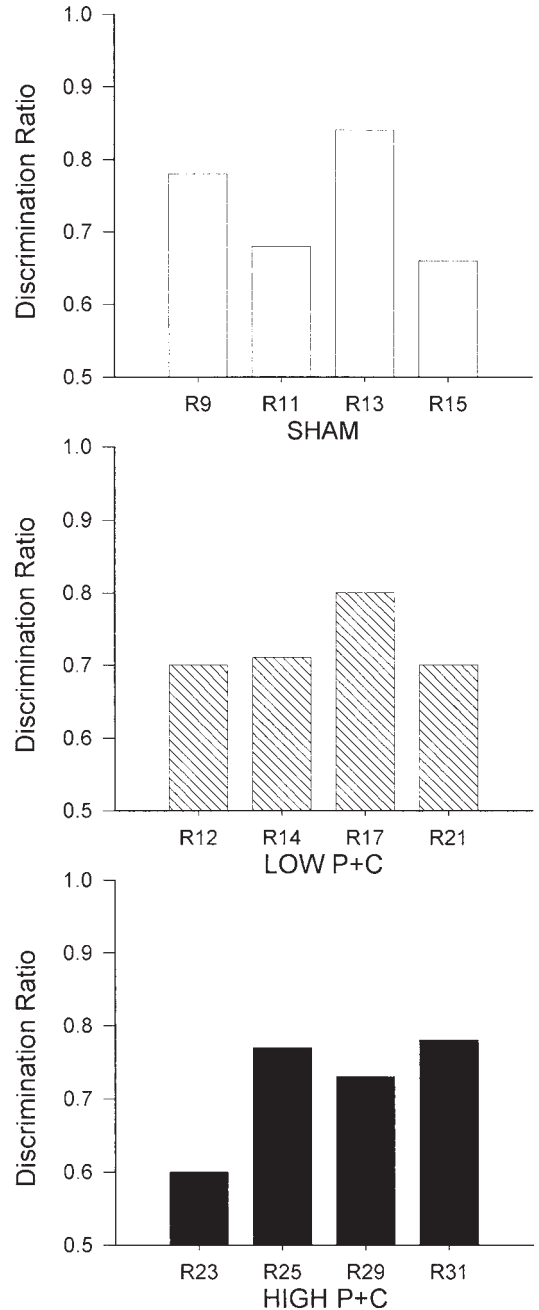


Fig. 2. Overall biconditional discrimination performance on the last day of training for each rat in each condition (SHAM, LOW P+C, HIGH P+C) of Experiment 1.

both contexts and is shown in Figure 3 (response rates are shown in Table 3). Responding across the whole stimulus period was analyzed.

Table 3

Response rates (lever presses per minute) during cue presentation for each rat during the last day of biconditional discrimination training and during test sessions in Experiment 1.

Group	Rat	Extinction test sessions					
		Biconditional training (last day)		Congruent		Incongruent	
		Correct	Incorrect	Correct	Incorrect	Correct	Incorrect
HIGH P+C	23	8.75	5.875	14.00	5.75	10.75	9.25
	25	18.375	5.375	22.50	4.75	18.75	15.75
	29	17.625	6.625	33.75	2.75	13.50	15.00
	31	21.125	6.00	26.75	5.50	19.50	31.00
LOW P+C	12	19.75	8.75	17.25	7.50	24.75	7.25
	14	23.00	9.50	31.00	5.00	28.50	16.75
	17	13.375	3.375	26.75	12.25	21.75	12.75
	21	33.375	14.875	31.75	13.50	30.25	14.75
SHAM	9	45.625	13.50	40.75	23.00	46.25	19.75
	11	21.75	10.375	24.75	13.00	26.25	12.50
	13	21.375	4.00	39.25	23.50	30.25	21.00
	15	36.25	19.00	29.75	5.25	19.75	15.50

Congruent Stimulus Compounds

In all animals, discrimination ratios to congruent stimulus compounds were greater than 0.60, ranging from 0.63 (Rat R9, SHAM) to 0.92 (Rat R29, HIGH P+C). Comparison of Rat R12 (LOW P+C) and Rat R25 (HIGH P+C) with equivalent overall damage demonstrates better responding in the HIGH P+C rat with Rat R25 producing a ratio of 0.83 to congruent compounds compared to 0.70 by Rat R12. Other HIGH P+C rats also demonstrate improved responding to congruent compounds compared to LOW P+C and SHAM rats with discrimination ratios of 0.71, 0.71, and 0.92. Thus responding to congruent stimulus compounds indicates successful performance to compound cues based on initial biconditional training in all animals.

Incongruent Stimulus Compounds

Performance to incongruent stimulus compounds indicates differences between HIGH P+C rats compared to those with LOW P+C damage and SHAMS. Most rats demonstrated performance on incongruent trials that was equivalent to, or worse than, that on congruent trials, with performance ranging from discrimination ratios of 0.39 (Rat R31, HIGH P+C) to 0.77 (Rat R12, LOW P+C). SHAM and LOW P+C rats demonstrated a similar pattern of responding to incongruent compounds, demonstrating greater responding on the correct compared to the incorrect lever. Thus, in SHAM and LOW P+C animals, contextual

cues that were incidental during training came to control responses during incongruent stimulus compounds at test. In contrast, performance of HIGH P+C rats did not demonstrate this transfer of contextual control to the novel audiovisual compounds. Discrimination ratios ranged from 0.39 (Rat R31) to 0.54 (Rat R25). Rats R25, R23, and R29 demonstrated minimal differences between responses on the correct and incorrect levers, with Rat R31 responding more on the incorrect than the correct lever, despite demonstrating accurate performance on congruent trials.

Comparison of rats with equivalent overall medial prefrontal cortex damage further highlights the difference between HIGH and LOW P+C rats. Rat R12 (LOW P+C) demonstrated good contextual control of performance to incongruent trial types (0.77), whereas this pattern of results is not evident in Rat R25 (HIGH P+C), with responding roughly equivalent on both levers (0.54).

DISCUSSION

In SHAM animals, the biconditional discrimination stimuli came to control correct lever-press responses during training, as evident by high levels of correct responding to both the congruent stimulus compounds during the extinction tests. Moreover, responding in SHAM animals to incongruent stimulus compounds was dictated by the context-appropriate stimulus-response map-

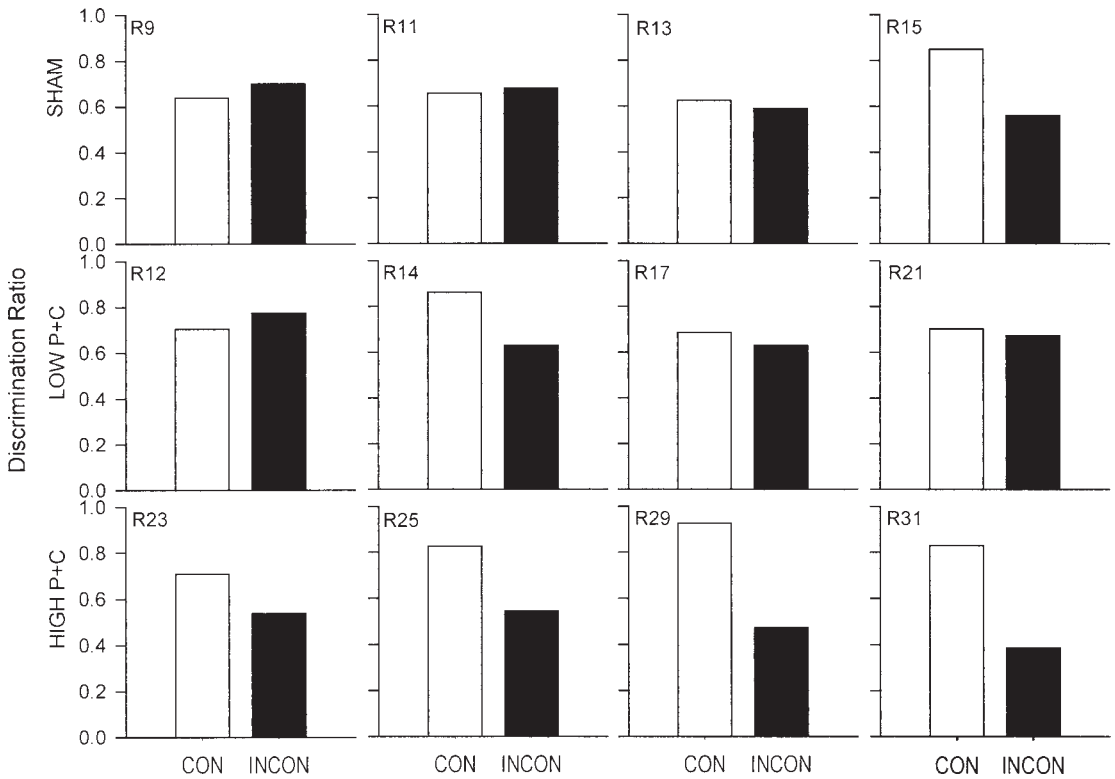


Fig. 3. Performance on congruent and incongruent test trials for SHAM, LOW P+C, and HIGH P+C rats in Experiment I. Open bars represent discrimination ratios during congruent trials, and solid bars represent discrimination ratios during incongruent trials.

tings. These results suggest that the contextual information provided during initial training came to control responding to stimulus compounds at test, especially when the individual elements of the stimulus compound mapped onto different lever press responses (i.e., incongruent compounds).

Rats with small amounts of damage (less than 50% cell loss) to prelimbic and cingulate subregions of the medial prefrontal cortex (LOW P+C) demonstrated a similar pattern of responding to that of SHAM animals. They responded normally to congruent stimulus compounds, and like SHAM animals, contextual cues came to control responding to incongruent stimulus compounds. As with SHAM animals, responding to incongruent compounds was somewhat less accurate, on average, than performance on congruent trials, indicating some impact of competing stimulus-response mappings.

Damage to large amounts of prelimbic and cingulate cortex regions (greater than 70%

cell loss, HIGH P+C) resulted in a different pattern of results from that seen in rats in both SHAM and LOW P+C groups. The training stimuli came to control appropriate responding, as demonstrated by accurate responding on congruent trials during test sessions, comparable to SHAM and LOW P+C rats. In contrast, responding on incongruent compounds did not yield the same pattern of results in HIGH P+C animals as it did in SHAMS and LOW P+C rats. HIGH P+C rats were unable to respond normally to the conflicting stimulus-response mappings activated by the incongruent stimulus compounds. Thus the contextual cues did not come to control responding to incongruent compounds in rats with large amounts of prelimbic and cingulate cortex damage. This effect of prelimbic and cingulate damage did not appear to be a result of overall damage to the medial prefrontal cortex. Comparison of individual rats with equivalent overall damage, but differing damage to prelimbic and cingu-

Table 4

The extent and location of cell loss within the anterior cingulate cortex and surrounding regions for each rat in Experiment 2a.

Group	Rat	Overall mPFC damage (%)	Cingulate cortex damage (%)	Prelimbic cortex damage (%)
HIGH C	2	47	75	0
	8	53	75	75
	14	38	80	6
LOW C	18	22	37.50	25
	27	33	40	37.50
	29	21	30	25

Note. Assessment of regional cell loss was based on the atlas of Paxinos and Watson (1998) and is given as a percentage. C = Cingulate cortex. HIGH C animals were those that exhibited greater than 70% damage to the cingulate cortex, and LOW C animals were those that had less than 40% damage to this region.

late subregions, demonstrated a similar pattern of results as that described above.

Although this experiment demonstrates that damage to the prefrontal and cingulate, rather than other subregions of the medial prefrontal cortex, disrupts performance on incongruent trials, it is not possible to differentiate the possible involvement of each of these subregions because damage to these areas tended to covary. As a consequence, a second experiment was conducted to determine the possible locus of the effect within the prefrontal and cingulate cortices.

EXPERIMENT 2A

The Effect of Anterior Cingulate Cortex Lesions on Contextual Control of Conditional Responding

Experiment 2a attempted to further dissociate the effect of prefrontal and anterior cingulate subregions on performance of incongruent trials by investigating the effect of excitotoxic lesions specific to the anterior cingulate cortex on performance of this task.

METHOD

Subjects

Nine rats were maintained and housed as described for Experiment 1. Ad-lib weights were in the range of 400 to 450 g.

Surgery and Histology

Surgical and histological procedures were as described for Experiment 1. Injections of Quinolinic acid (0.09 M, Sigma-Aldrich Co. Ltd., UK.) were made with a 2 μ l Hamilton syringe (Reno, NV) mounted on the stereotaxic frame. Quinolinic acid was chosen

because this excitotoxin was found in pilot studies to be appropriate for producing selective damage to the anterior cingulate cortex. Injections of 0.2–0.3 μ l at a rate of 0.1 μ l/min were made at six sites within the anterior cingulate cortex (AP: +2.2, +2.7, and +3.2, ML: \pm 0.5, DV: –2.4, –2.6, and –2.4, respectively). Following each injection the needle was left in position for 2 min to allow absorption of the bolus and to minimize spread of the toxin into other brain regions.

Apparatus and Behavioral Procedure

All details were as described for Experiment 1.

RESULTS

Histological Analysis

Lesioned animals were grouped into those with low and high anterior cingulate cortex damage groups (LOW C and HIGH C, respectively) according to the extent and location of cell loss within the anterior cingulate cortex (see Table 4). HIGH C rats (Rats R2, R8, and R14) exhibited greater than 70% cell loss in area Cg1, as defined by Paxinos and Watson (1998), whereas LOW C rats (Rats R18, R27, and R29) demonstrated less than 40% cell loss in the same region. Overall medial prefrontal damage in the HIGH C rats ranged from 38% (Rat R14) to 53% (Rat R8), with damage to the cingulate cortex ranging from 75% (Rats R2 and R8) to 80% (Rat R14). The overall medial prefrontal cell loss in the LOW C rats ranged from 21% (Rat R29) to 33% (Rat R27), with cingulate cortex damage ranging from 30% (Rat R29) to 40% (Rat R27). Three sham-operated animals (Rats R7, R11, and

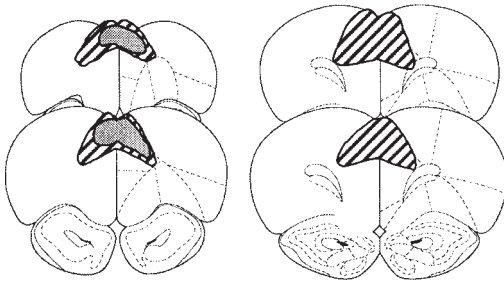


Fig. 4. The maximum and minimum extent of the damage to the anterior cingulate cortex. Outlines are reproduced from Paxinos and Watson (1998) and represent sections ranging from 2.2 to 3.7 anterior to bregma. The maximum amount of damage to the anterior cingulate cortex was observed in Rat R14 (HIGH C) and is depicted by the striped regions. The minimum amount of damage was observed in Rat R29 (LOW C) and is depicted by the grey regions.

R26) provided control comparisons. Sham surgical procedures resulted in negligible cell damage.

As in Experiment 1, 2 animals with approximately equal overall medial prefrontal cell loss (Rat R27, LOW C = 33% and Rat R14 HIGH C = 38%) were selected to serve as more direct comparisons for the loss of cingulate cortex tissue (40% and 80%, respectively). The maximum and minimum extent of the damage in rats classified as HIGH C and LOW C is shown in Figure 4.

Behavioral Analysis

Biconditional discrimination performance during training. As in Experiment 1, performance on the biconditional discriminations was assessed according to responding during the 10-s period at the beginning of the stimulus presentation when reinforcement was unavailable. The discrimination ratio was calculated based on responding averaged across the two biconditional discrimination tasks.

Acquisition of the biconditional discrimination tasks progressed rapidly, with all animals demonstrating a discrimination ratio greater than 0.60 by the last day of training (see Figure 5; Table 5 presents response rates). Performance, as assessed by discrimination ratios, ranged from 0.62 (Rat R18, LOW C) to 0.86 (Rat R26, SHAM), indicating that the training stimuli controlled appropriate responding in all animals. Comparison of animals with equivalent overall damage (Rat R27, LOW C and Rat R14, HIGH C) indicated

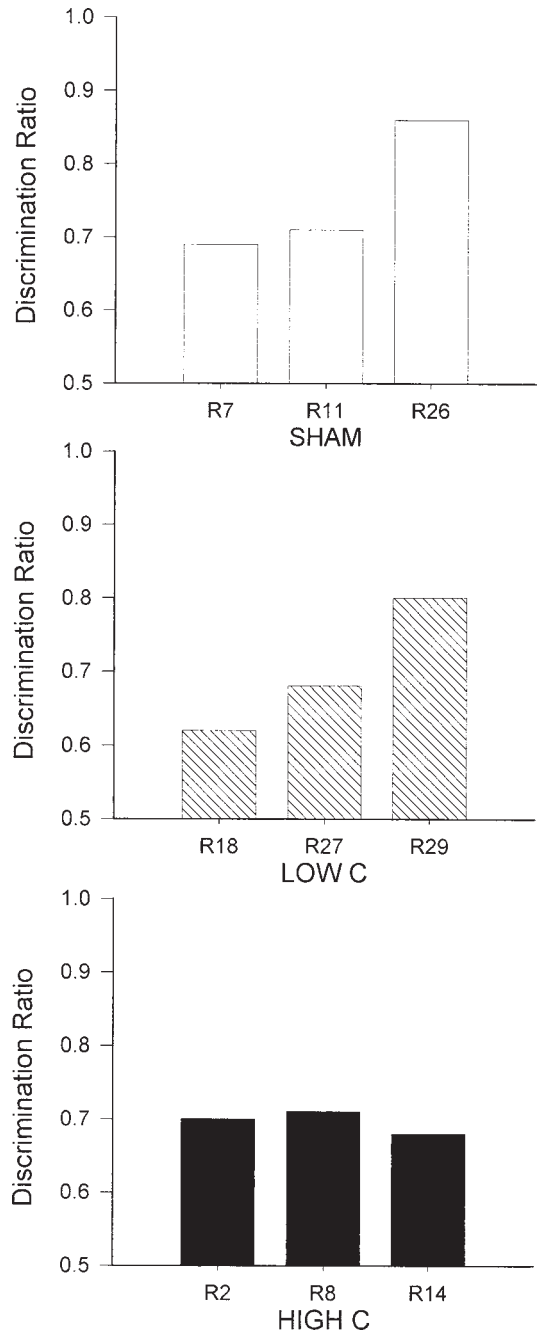


Fig. 5. Overall biconditional discrimination performance on the last day of training for each rat in each condition (SHAM, LOW C, HIGH C) of Experiment 2a.

the same level of performance as assessed by discrimination ratios (0.68).

Test session performance. Figure 6 presents the discrimination ratio of each rat to the test

Table 5

Response rates (lever presses per minute) during cue presentation for each rat during biconditional discrimination training and test sessions in Experiment 2a.

Group	Rat	Extinction test sessions					
		Biconditional training (last day)		Congruent		Incongruent	
		Correct	Incorrect	Correct	Incorrect	Correct	Incorrect
HIGH C	2	30.75	13.375	33.125	10.125	17.625	22.50
	8	17.125	6.875	15.50	3.75	8.875	10.50
	14	29.75	14.25	30.375	12.125	14.625	13.25
LOW C	17	25.50	15.375	19.125	7.50	16.375	10.25
	27	19.875	9.50	31.875	4.625	26.50	14.00
	29	49.25	12.375	45.375	11.75	40.25	18.625
SHAM	7	24.25	10.875	22.625	14.625	21.375	13.375
	11	38.25	16.00	29.875	7.875	28.25	13.75
	26	11.75	1.875	15.00	2.375	12.625	6.50

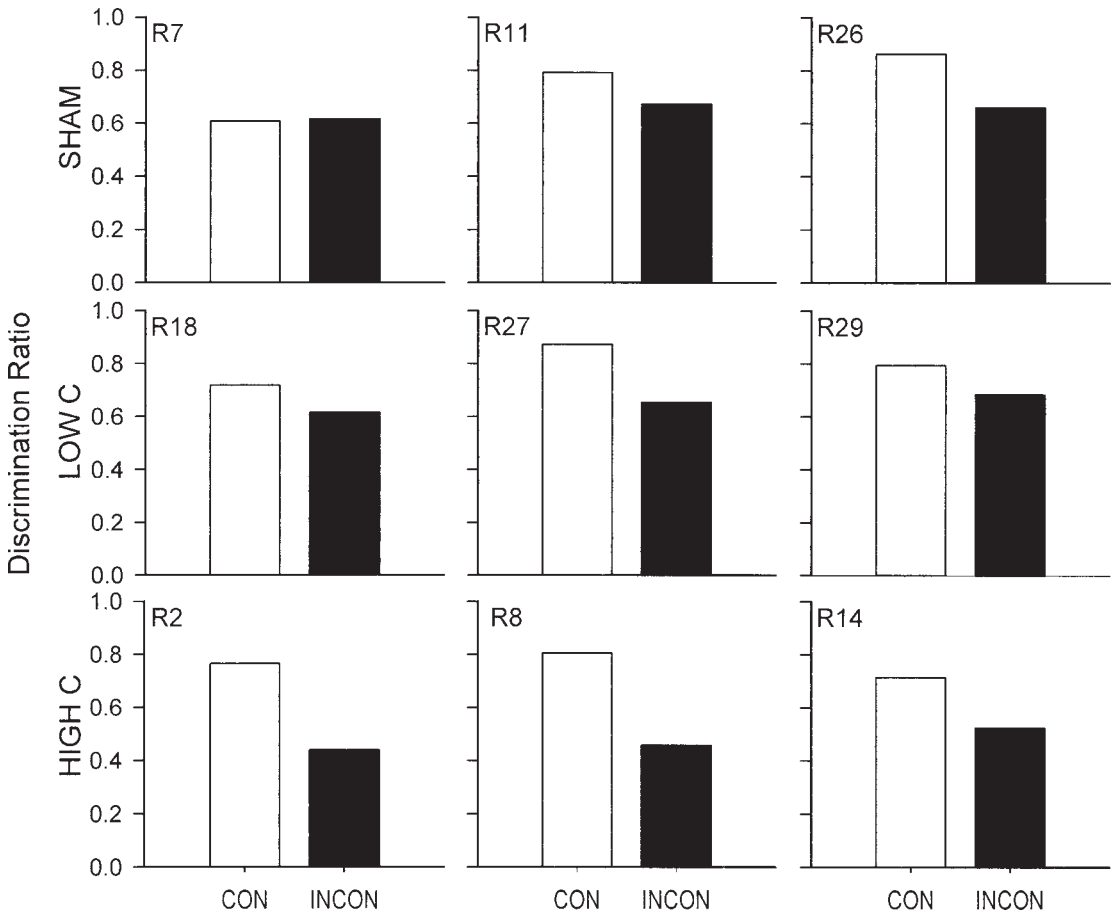


Fig. 6. Performance on congruent and incongruent test trials for SHAM, LOW C, and HIGH C rats in Experiment 2a. Open bars represent discrimination ratios during congruent trials, and solid bars represent discrimination ratios during incongruent trials.

stimuli. Test performance was averaged across both test sessions, and responding over the whole stimulus period was analyzed (see Table 5 for response rates during congruent and incongruent stimulus presentation).

Congruent Stimulus Compounds

All animals responded accurately to congruent stimulus compounds exhibiting discrimination ratios greater than 0.60. Performance as assessed by discrimination ratios ranged from 0.61 (Rat R7, SHAM) to 0.87 (Rat R27, LOW C), and was similar across all animals. Comparison of Rat R27 (LOW C) and Rat R14 (HIGH C) demonstrated good performance on congruent trials by both rats (0.87 and 0.71, respectively).

Incongruent Stimulus Compounds

Performance on incongruent trials demonstrated a similar pattern of responding in SHAM and LOW C rats, with greater responding on the correct lever than on the incorrect lever on incongruent trials. Discrimination ratios were greater than 0.60 for all the rats, ranging from 0.62 (Rat R7, SHAM) to 0.68 (Rat R29, LOW C). Thus, as in Experiment 1, the contextual cues came to disambiguate conflicting responses, guiding responding according to the stimulus-response mapping previously trained in the test context.

In contrast, rats with greater than 70% damage to the cingulate cortex (HIGH C) failed to demonstrate accurate responding to incongruent stimulus compounds, resulting in discrimination ratios of around 0.5 or less. For 2 of the 3 rats (Rats R2 and R8), responding was greater on the incorrect lever than the correct lever, resulting in discrimination ratios that were less than 0.5. Performance of Rat R14 was not much better, with a discrimination ratio of 0.52. Comparison of rats with equivalent overall damage further established this finding. Rat R14 (HIGH C) failed to perform correctly to incongruent trials (0.52) compared to a discrimination ratio of 0.65 demonstrated by Rat R27 (LOW C), despite both animals having comparable overall damage to the medial prefrontal cortex.

DISCUSSION

SHAM and LOW C rats (with less than 40% cingulate cortex cell loss) responded accurately

ly to both types of test trial (congruent and incongruent), replicating the effect seen in Experiment 1. These results demonstrate the successful acquisition of stimulus-response mappings and that contextual information came to control responding when rats were presented with novel audiovisual compounds that provided conflicting response information.

Animals with greater than 70% cell loss in the cingulate cortex (HIGH C) showed accurate responding to congruent trials, comparable to SHAM and LOW C rats. In contrast, these animals were unable to perform as well to incongruent stimulus compounds, responding equally on both the correct and incorrect levers. This deficit was not a result of a failure of the training stimuli to control responding, since these animals were able to perform correctly to the congruent stimulus compounds and to the training stimuli. As in Experiment 1, the effect does not appear to be related to the degree of overall damage to the medial prefrontal cortex.

EXPERIMENT 2B

The Effect of Anterior Cingulate Cortex Lesions on Contextual Control of Conditional Responding Early in Incongruent Stimulus Compound Presentation

The aim of Experiment 2b was to investigate the impact of cell damage restricted *entirely* to the anterior cingulate cortex on performance during incongruent stimulus compounds, in particular, investigating the impact of these lesions at different periods of time during the incongruent stimulus presentation (S1/early and S2/late). This investigation was conducted to establish whether the impairment observed during Experiment 2a was a consequence of a complete failure of the context to control responding in the lesioned animals or a result of some other, related process, for example, the detection of response conflict.

METHOD

Subjects, Surgery, and Histology

Three rats were studied, and their housing, surgery, and histology were as described for Experiment 2a.

Apparatus and Behavioral Procedure

All details were as described for Experiment 1.

Table 6

The extent of cell loss within the anterior cingulate cortex for each of the rats in Experiment 2b.

Group	Rat	Cingulate cortex damage (%)	Overall mPFC damage (%)
C ONLY	10	50	17
	12	53	18
	19	65	22

Note. Assessment of regional cell loss was based on the atlas of Paxinos and Watson (1998) and is given as a percentage. C = Cingulate cortex. C ONLY animals were those that exhibited cell loss restricted to the anterior cingulate cortex.

RESULTS

Histological Analysis

The 3 rats demonstrated lesions restricted entirely to the anterior cingulate cortex (Rats R10, R12, and R19—C ONLY group) and exhibited 50% or greater damage to the anterior cingulate cortex, ranging from 50% (Rat R10) to 65% (Rat R19), with no cell loss observed within other medial prefrontal cortex subregions (see Table 6).

Behavioral Analysis

Biconditional discrimination performance during training. As in Experiment 1, performance on the biconditional discriminations was assessed according to responding during the 10-s period at the beginning of the stimulus presentation when reinforcement was unavailable. Acquisition of the biconditional discrimination tasks progressed rapidly, with all animals demonstrating a discrimination ratio greater than 0.60 by the last day of training (see Figure 7). Discrimination ratio performance ranged from 0.65 (Rat R12) to 0.76 (Rat R10), indicating that the biconditional stimuli had come to control appropriate lever-press responding in all 3 lesioned animals.

Test session performance. Figure 8 presents the performance of the 3 C ONLY rats to the test stimuli. Test performance was averaged across both test sessions. In contrast to Experiments 1 and 2a, responding across the entire stimulus period was not analyzed; instead, responses were separated into the first 10 s (S1) and the last 50 s (S2) of a trial, and responses per minute were calculated for each period. These analyses were conducted to establish whether the pattern of the impairment observed in Experiment 2a was similar during the two recording periods. For com-

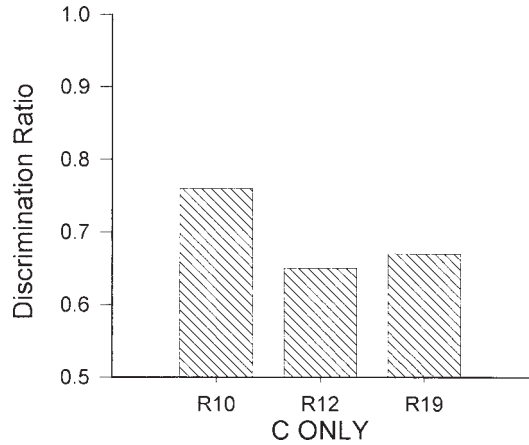


Fig. 7. Overall biconditional discrimination performance on the last day of training for each rat in the C ONLY group of Experiment 2b.

parison purposes, data from SHAM and HIGH P+C rats from Experiment 1 are included (response rates for all animals are shown in Table 7).

Congruent Stimulus Compounds

All 3 C ONLY rats responded accurately to congruent stimulus compounds during both S1 and S2 recording periods, exhibiting discrimination ratios greater than or equal to 0.65. Performance remained accurate throughout the duration of congruent stimulus compound presentation in all 3 rats. During the first 10 s of congruent stimulus compound presentations (S1 recording period), discrimination ratio performance ranged from 0.65 (Rat R10) to 1.0 (Rat R19), whereas later on (S2 recording period) performance ranged from 0.65 (Rat R10) to 0.81 (Rat R19). A similar pattern also was observed in SHAM and HIGH P+C rats from Experiment 1. Discrimination ratio performance during congruent trials ranged from 0.72 (Rat R13, SHAM) to 1.0 (Rats R23, R31, HIGH P+C, and Rat R15, SHAM) early on in stimulus presentation, and ranged from 0.62 (Rat R13, SHAM) to 0.92 (Rat R29, HIGH P+C) later on in stimulus presentation.

Incongruent Stimulus Compounds

Performance on incongruent trials demonstrated different patterns of performance during S1 and S2 recording periods in all 3 C ONLY rats. Performance during the S1

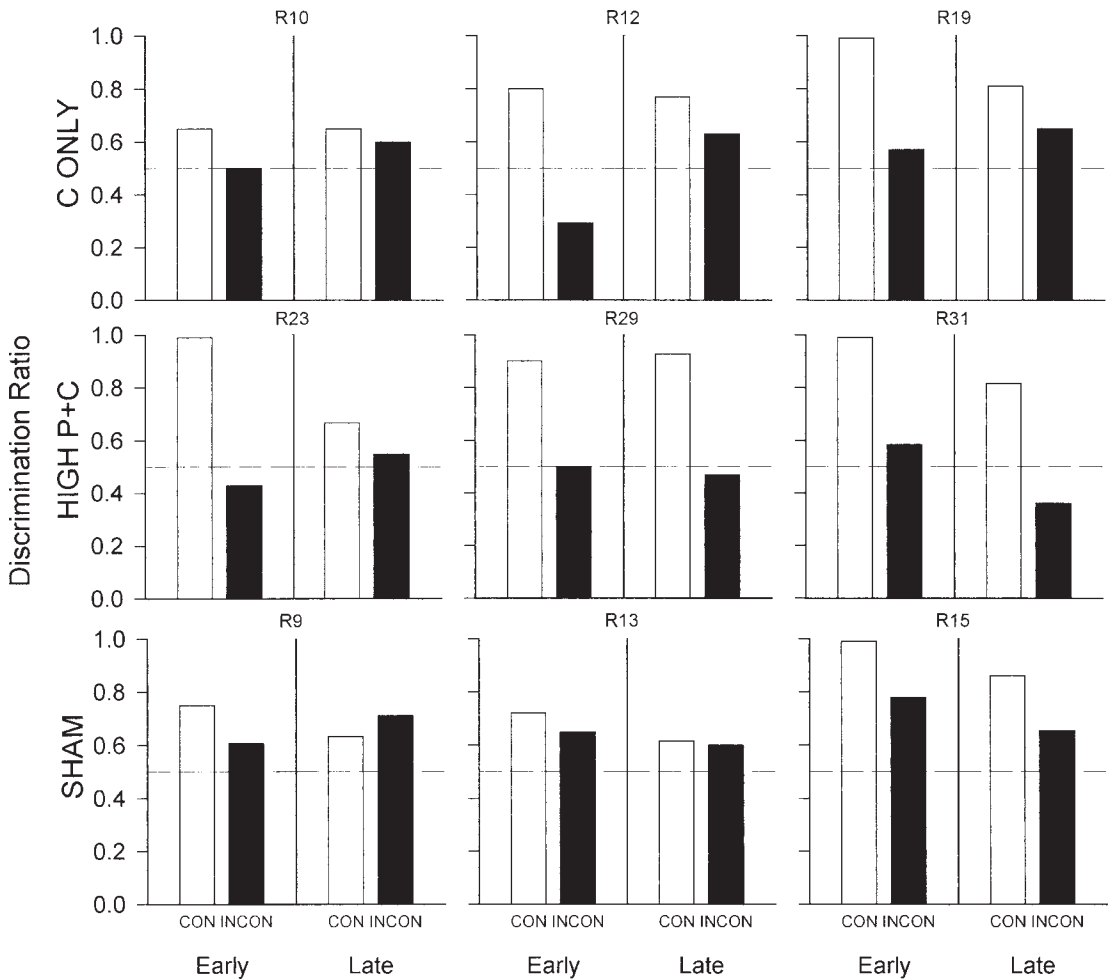


Fig. 8. Performance during the first 10 s (S1/early) and last 50 s (S2/late) of congruent and incongruent test trials for each of the C ONLY rats of Experiment 2b. Discrimination ratios also are shown for SHAM and HIGH P+C rats from Experiment 1. Dashed lines indicate the level of discrimination performance expected by chance when equal amounts of correct and incorrect responding are observed.

recording period (first 10 s of a trial) demonstrated a similar pattern of responding to that observed in HIGH C animals in Experiment 2a. All 3 rats demonstrated reduced context-appropriate responding to the incongruent stimulus compounds, ranging from 0.29 (Rat R12) to 0.56 (Rat R19) indicating that the contextual cues had not come to disambiguate the conflicting response information provided by incongruent stimulus compounds. In contrast, however, performance during the S2 recording period (the last 50 s of a trial) demonstrated increased responding on the correct lever compared to responding on the incorrect lever. Discrimination ratio perfor-

mance ranged from 0.60 (Rat R10) to 0.65 (Rat R19), suggesting that the contextual cues were able to guide responding according to the stimulus-response mapping previously trained in the test context. In summary, responding during the first 10 s of stimulus presentation was not above the level of responding expected by chance (discrimination ratio of 0.5), whereas in the last 50 s of incongruent stimulus presentation, discrimination ratio performance increased relative to chance, and consequently contextual control of responding was observed.

No differences in responding during S1 and S2 recording periods were observed in Sham

Table 7

Response rates (lever presses per minute) during cue presentation during biconditional discrimination training and test sessions for C ONLY rats of Experiment 2b and for HIGH P+C and SHAM rats from Experiment 1.

Group		Biconditional training (last day)		Extinction test sessions							
				Early (S1)				Late (S2)			
				Congruent		Incongruent		Congruent		Incongruent	
Rat	Correct	Incorrect	Correct	Incorrect	Correct	Incorrect	Correct	Incorrect	Correct	Incorrect	
C ONLY	10	38.25	12.125	19.50	10.50	12.75	12.75	31.80	17.25	23.00	14.55
	12	22.00	11.625	18.40	4.50	3.75	9.00	30.00	9.15	23.50	13.95
	19	5.625	2.75	8.25	0.00	3.00	2.25	16.95	3.90	14.10	7.65
HIGH P+C	23	8.75	5.875	15.00	0.00	4.50	6.00	13.80	6.90	12.00	9.90
	29	17.625	6.625	13.50	1.50	1.50	1.50	37.80	3.00	15.90	18.00
	31	21.125	6.00	15.00	0.00	21.00	15.00	29.10	6.60	19.20	34.20
SHAM	9	45.625	13.50	18.00	6.00	25.50	16.50	45.30	26.40	50.40	20.40
	13	21.375	4.00	27.00	10.50	16.50	9.00	41.70	26.10	35.00	23.40
	15	36.25	19.00	31.50	0.00	9.00	6.00	29.40	6.30	28.00	17.40

Note. S1 = recording period during the first 10 s of stimulus presentation, and S2 = recording period during the last 50 s of stimulus presentation.

or mPFC lesioned animals in Experiment 1. Both SHAM and HIGH P+C animals demonstrated performance that remained consistent throughout the duration of congruent and incongruent stimulus compound presentation (see Figure 8). For SHAM animals, context-appropriate responding during incongruent stimulus compounds was observed throughout the duration of the stimulus presentation. During the first 10 s of stimulus presentation, discrimination ratio performance ranged from 0.60 (Rat R15) to 0.64 (Rat R13); similarly, in the last 50 s of stimulus presentation, discrimination ratio performance ranged from 0.60 (Rat R13) to 0.72 (Rat R9). Animals with mPFC lesions also demonstrated a consistent pattern of responding throughout the stimulus presentation. Context-appropriate responding was not observed at any point during the stimulus presentation as reflected by discrimination ratios around 0.5. Early on in the stimulus presentation discrimination ratio performance ranged from 0.42 (Rat R23) to 0.58 (Rat R31), and later on in stimulus presentation performance was not improved, as reflected by discrimination ratio performance that ranged from 0.36 (Rat R31) to 0.54 (Rat R23).

DISCUSSION

The performance of rats with damage restricted entirely to the anterior cingulate

cortex in Experiment 2b replicated the findings of Experiment 2a, which demonstrated normal biconditional discrimination performance in animals with anterior cingulate damage, indicated by high discrimination ratios on the last day of training and during congruent stimulus compounds at test. Moreover, a similar pattern of performance was observed during congruent stimulus compounds throughout the duration of the trial. In accord with the results from Experiment 2a, animals with damage restricted to the anterior cingulate cortex demonstrated impaired performance during incongruent stimulus compounds, suggesting a reduced ability of contextual cues to disambiguate conflicting response information provided by the compound. Further investigation of this deficit indicated that the impairment was restricted to the early stages (first 10 s) of stimulus presentation and that later on during the stimulus, context-appropriate responding to incongruent stimulus compounds was observed. This selective early impairment was not observed in animals with sham or large medial prefrontal cortex lesions.

GENERAL DISCUSSION

Rats were initially trained on two biconditional discriminations, auditory and visual, each of which was acquired in a distinct

context. At test, rats were presented with congruent and incongruent audiovisual compounds of the training stimuli in extinction. Congruent compounds were composed of stimulus elements that mapped onto the same response during training, whereas incongruent compounds were composed of stimulus elements that mapped onto different responses. The results of Experiments 1, 2a, and 2b indicate that contextual cues came to control responding when rats were presented with incongruent stimulus compounds, such that responding was in accord with the context-appropriate stimulus-response mapping. This contextual control of responding to novel compounds occurred despite the context being incidental to the solution of the initial biconditional discrimination tasks during training.

Experiment 1 demonstrated that excitotoxic lesions to the medial prefrontal cortex impaired this ability of contextual cues to control responding to incongruent stimulus compounds. This effect appeared to be dependent upon damage to the prelimbic and cingulate subregions of the prefrontal cortex since extensive damage to these regions increased incorrect responding on incongruent trials, whereas performance after limited damage to these regions did not differ from that of controls. Comparison of rats with the same overall damage to the medial prefrontal cortex but differing damage to the prelimbic and cingulate subregions indicated differences in responding to incongruent compounds consistent with the suggestion that it was the extent of cell loss within these subregions, rather than overall medial prefrontal damage, that was responsible for the impairment in responding.

The results of Experiment 2a suggest that damage focused on the cingulate cortex but also including other medial prefrontal cortical regions was sufficient to produce a disruption in correct responding during incongruent trials similar to that seen with larger medial prefrontal cortex lesions in Experiment 1. As with Experiment 1, this effect was not due to the degree of overall medial prefrontal cell loss. Experiment 2b investigated the responding to incongruent stimulus compounds in animals with damage restricted entirely to the anterior cingulate cortex, demonstrating that the deficit was restricted to the first 10 s of

stimulus presentation, with context-appropriate stimulus-response mapping emerging during the remaining 50 s of incongruent trials. Similar patterns of responding to incongruent stimulus compounds were seen during both recording periods in animals with both sham and large medial prefrontal lesions. The deficit observed with restricted damage to the anterior cingulate cortex was not a consequence of the limited nature of the medial prefrontal cortex damage, as Rats R14 and R17 (LOW P+C, Experiment 1) both exhibited restricted damage to the prefrontal cortex (17% in both cases) and demonstrated a similar pattern of responding to incongruent cues both early and late in stimulus presentation (discrimination ratios: Rat R14: early = 0.72, late = 0.70; Rat R17: early = 0.63, late = 0.63). In Experiments 1, 2a, and 2b, the disruption of performance on incongruent trials was not a result of a failure of the training stimuli to control appropriate responding since performance on congruent trials remained intact in all rats, irrespective of the extent of cell loss. In summary, damage restricted to prelimbic and cingulate regions of the prefrontal cortex resulted in a global deficit in the resolution of response conflict provided by incongruent stimulus compounds, and damage focused solely on the anterior cingulate region produced a more selective and transient deficit, suggesting that damage to this region resulted in impairments that were restricted to the detection of response conflict.

The effect of lesions to the medial prefrontal cortex on this task is consistent with previous research indicating a role for the prefrontal cortex in conditional discrimination tasks in which response conflict is present. This effect of prefrontal damage has been reported in rats (Joel, Weiner & Feldon, 1997; Winocur, 1991; Winocur & Eskes, 1998), non-human primates (Murray et al., 2000; Petrides, 1982, 1985, 1991), and humans (Petrides, 1990, 1997; Petrides et al., 1993). Furthermore, the results are consistent with performance of patients with prefrontal damage on the Stroop task. As in the current task, Stroop stimuli are able to control the production of the correct response, such that frontal patients can respond accurately to congruent stimulus compounds and individual presentations of colors or color words. However, task-appropriate control of responses to incongruent

compounds is not apparent in these patients. Thus rats with medial prefrontal cortex lesions and humans with prefrontal cortex damage perform comparably on tasks in which contextual or task-appropriate information comes to control responding to conflicting or ambiguous response information. Although the effects of prefrontal damage on the task presented here could be interpreted as reflecting task difficulty, a more specific hypothesis indicates that the impairment is a result of the increased response conflict inherent in incongruent stimulus presentations. This is manifested behaviorally as increased task difficulty, just as responding to incongruent color-word Stroop stimuli is perceived as more difficult by human participants. That is to say, the specific aspect of the task that appears more difficult is that which derives from increased response conflict, and it is this increased conflict that leads to deficits in lesioned animals.

The finding that damage to the anterior cingulate cortex produces a deficit in responding on incongruent trials is also consistent with previous research. Results from imaging studies in humans indicate activation of the anterior cingulate cortex during incongruent trials in which conflicting information is present (Carter et al., 1995; MacDonald et al., 2000; MacLeod & MacDonald, 2000; Milham, Banich, & Barad, 2003). The interpretation of these findings in humans, however, has been marred by activation of this region during performance on congruent trials and the concurrent activation of other prefrontal regions during incongruent trials, namely the dorsolateral and ventrolateral area (Duncan & Owen, 2000). The results presented above demonstrate that cell loss specific to the anterior cingulate cortex produces a deficit in performance to incongruent stimulus compounds. Experiment 2b extended the results of Experiment 2a by demonstrating that the impairment following anterior cingulate damage was restricted to early periods of incongruent stimulus presentation. Furthermore, this early/late distinction was not seen with damage to other prefrontal subregions or in sham-lesioned animals.

Evidence from human studies has implicated the anterior cingulate cortex in response-related processes, in particular in the detection of competition at the level of responses

(MacDonald et al., 2000; Milham, Banich, & Barad, 2003; Milham, Banich, Claus, & Cohen, 2003), and this also may be the case following ACC lesions in rats. The results from Experiment 2b lend support to this proposal since the deficit in the ability of the contextual cues to control responding was observed only early during the stimulus presentation, when one might suppose that the detection of competition between responses occurs. Later on during the stimulus presentation, ACC lesioned animals performed comparably to controls, whereas the deficit in mPFC lesioned animals was observed across the entire duration of the stimulus presentation. This dissociation of performance in mPFC and ACC lesioned animals suggests that ACC damage may contribute to the deficit in contextual control of responding seen with mPFC lesioned animals, but that it does not appear sufficient to produce the complete abolition of context-appropriate responding observed in Experiment 1.

The findings from the present study provide clear evidence for the involvement of the medial prefrontal cortex in performance on trials in which contextual cues have been shown to disambiguate conflicting response information, and thus control responding, and suggests a specific involvement for the anterior cingulate cortex in the detection or resolution of response conflict. This finding is consistent with research that claims a differentiation of function within the rat prefrontal cortex subregions, and is in marked contrast with the suggestion by Duncan and Owen (2000) that there is no specialization of function within the human prefrontal cortex.

REFERENCES

- Bussey, T. J., Muir, J. L., Everitt, B. J., & Robbins, T. W. (1996). Dissociable effects of anterior and posterior cingulate cortex lesions on the acquisition of a conditional visual discrimination: Facilitation of early learning vs. impairment of late learning. *Behavioral Brain Research*, *82*, 45–56.
- Carter, C. S., Mintun, M., & Cohen, J. D. (1995). Interference and facilitation effects during selective attention: An H₂¹⁵O PET study of Stroop task performance. *Neuroimage*, *2*, 264–272.
- Cohen, J. D., & Servan-Schreiber, D. (1992). Context, cortex, and dopamine: A connectionist approach to behavior and biology in schizophrenia. *Psychological Review*, *99*, 45–77.

- Condé, F., Maire-Lepoivre, E., Audinat, E., & Crepel, F. (1995). Afferent connections of the medial frontal cortex of the rat. II. Cortical and subcortical afferents. *Journal of Comparative Neurology*, *352*, 567–593.
- Coutureau, E., & Killcross, S. (2003). Inactivation of the infralimbic prefrontal cortex reinstates goal-directed responding in overtrained rats. *Behavioural Brain Research*, *146*, 167–174.
- Dias, R., Robbins, T. W., & Roberts, A. C. (1997). Dissociable forms of inhibitory control within prefrontal cortex with an analog of the Wisconsin card sort test: Restriction to novel situations and independence from “on-line” processing. *Journal of Neuroscience*, *17*, 9285–9297.
- Duncan, J., & Owen, A. M. (2000). Common regions of the human frontal lobe recruited by diverse cognitive demands. *Trends in Neuroscience*, *23*, 475–483.
- Funahashi, S., Bruce, C. J., & Goldman-Rakic, P. S. (1989). Mnemonic coding of visual space in the monkey's dorsolateral prefrontal cortex. *Journal of Neurophysiology*, *61*, 331–349.
- Goldman-Rakic, P. S. (1987). Circuitry of primate prefrontal cortex and regulation of behavior by representational memory. In F. Plum (Ed.), *Handbook of physiology: The nervous system*. (pp. 373–417). Bethesda, MD: American Physiological Society.
- Hurley, K. M., Hernert, H., Moga, M. M., & Saber, C. B. (1991). Efferent projections of the infralimbic cortex of the rat. *Journal of Comparative Neurology*, *308*, 249–276.
- Joel, D., Weiner, I., & Feldon, J. (1997). Electrolytic lesions of the medial prefrontal cortex in rats disrupt performance on an analog of the Wisconsin Card Sorting Test, but do not disrupt latent inhibition: Implications for animal models of schizophrenia. *Behavioural Brain Research*, *85*, 187–201.
- Kesner, R. P. (2000). Subregional analysis of mnemonic functions of the prefrontal cortex in the rat. *Psychobiology*, *28*, 219–228.
- Killcross, A. S., & Coutureau, E. (2003). Coordination of actions and habits in the medial prefrontal cortex of rats. *Cerebral Cortex*, *13*, 400–408.
- Kojima, S., & Goldman-Rakic, P. S. (1982). Delay-related activity of prefrontal neurons in rhesus monkeys performing delayed response. *Brain Research*, *248*, 43–49.
- Kojima, S., & Goldman-Rakic, P. S. (1984). Functional analysis of spatially discriminative neurons in prefrontal cortex of rhesus monkey. *Brain Research*, *291*, 229–240.
- Krettek, J. E., & Price, J. L. (1977). The cortical projections of the mediodorsal nucleus and adjacent thalamic nuclei in the rat and cat. *Journal of Comparative Neurology*, *172*, 157–192.
- MacDonald, A. W., Cohen, J. D., Stenger, V. A., & Carter, C. S. (2000, June 9). Dissociating the role of dorsolateral prefrontal cortex and anterior cingulate cortex in cognitive control. *Science*, *288*, 1835–1838.
- MacLeod, C. M. (1991). Half a century of research on the Stroop effect: An integrative review. *Psychological Bulletin*, *109*, 163–203.
- MacLeod, C. M., & MacDonald, P. A. (2000). Interdimensional interference in the Stroop effect: Uncovering the cognitive and neural anatomy of attention. *Trends in Cognitive Science*, *4*, 383–391.
- Milham, M. P., Banich, M. T., & Barad, V. (2003). Competition for priority in processing increases prefrontal cortex's involvement in top-down control: An event-related fMRI study of the Stroop task. *Cognitive Brain Research*, *17*, 212–222.
- Milham, M. P., Banich, M. T., Claus, E. D., & Cohen, N. J. (2003). Practice-related effects demonstrate complementary roles of the anterior cingulate and prefrontal cortices in attentional control. *Neuroimage*, *18*, 483–493.
- Murray, E. A., Bussey, T. J., & Wise, S. P. (2000). Role of the prefrontal cortex in a network for arbitrary visuomotor mapping. *Experimental Brain Research*, *133*, 1–31.
- Öngür, D., & Price, J. L. (2000). The organization of networks within the orbital and medial prefrontal cortex of rats, monkeys and humans. *Cerebral Cortex*, *10*, 206–219.
- O'Reilly, R. C., Noelle, D. C., Braver, T. S., & Cohen, J. D. (2002). Prefrontal cortex and dynamic categorization tasks: Representational organization and neuromodulatory control. *Cerebral Cortex*, *12*, 246–257.
- Owen, A. M., Evans, A. C., & Petrides, M. (1996). Evidence for a two-stage model of spatial working memory processing within the lateral frontal cortex: A positron emission tomography study. *Cerebral Cortex*, *6*, 31–38.
- Paxinos, G., & Watson, C. (1998). *The rat brain in stereotaxic coordinates* (4th ed.). San Diego, CA: Academic Press.
- Perret, E. (1974). The left frontal lobe of man and the suppression of habitual responses in verbal categorical behaviour. *Neuropsychologia*, *12*, 323–330.
- Petrides, M. (1982). Motor conditional associative-learning after selective prefrontal lesions in the monkey. *Behavioral Brain Research*, *5*, 407–413.
- Petrides, M. (1985). Deficits in non-spatial conditional associative learning after periarculate lesions in the monkey. *Behavioral Brain Research*, *16*, 95–101.
- Petrides, M. (1990). Non-spatial conditional learning impaired in patients with unilateral frontal but not unilateral temporal lobe excisions. *Neuropsychologia*, *28*, 137–149.
- Petrides, M. (1991). Functional specialization within the dorsolateral frontal cortex for serial order memory. *Proceedings of the Royal Society of London*, *246*, 299–306.
- Petrides, M. (1994). Frontal lobes and working memory: Evidence from investigations of the effects of cortical excisions in nonhuman primates. In F. Boller & J. Grafman (Eds.), *Handbook of neuropsychology* (Vol. 9, pp. 59–82). Amsterdam: Elsevier.
- Petrides, M. (1997). Visuo-motor conditional associative learning after frontal and temporal lesions in the human brain. *Neuropsychologia*, *35*, 989–997.
- Petrides, M., Alivisatos, B., Evans, A. C., & Meyer, E. (1993). Dissociation of human mid-dorsolateral from posterior dorsolateral frontal cortex in memory processing. *Proceedings of the National Academy of Sciences of the United States of America*, *90*, 873–877.
- Ragozzino, R. E., Detrick, S., & Kesner, R. P. (1999). Involvement of the prelimbic-infralimbic areas of the rodent prefrontal cortex in behavioral flexibility for place and response learning. *Journal of Neuroscience*, *19*, 4585–4594.
- Ragozzino, R. E., Wilcox, C., Raso, M., & Kesner, R. P. (1999). Involvement of rodent prefrontal cortex subregions in strategy switching. *Behavioral Neuroscience*, *113*, 32–41.

- Rao, S. C., Rainer, G., & Miller, E. K. (1997, May 2). Integration of what and where in the primate prefrontal cortex. *Science*, 276, 821–824.
- Ray, J. P., & Price, J. L. (1992). The organization of the thalamocortical connections of the mediodorsal thalamic nucleus in the rat, related to the ventral forebrain-prefrontal cortex topography. *Journal of Comparative Neurology*, 323, 167–197.
- Reep, R. L., Chandler, H. C., King, V., & Corwin, J. V. (1984). Neural connections of orbital cortex in rats: Topography of cortical and thalamic connections. *Experimental Brain Research*, 100, 67–84.
- Rose, J. E., & Woolsey, C. N. (1948). The orbitofrontal cortex and its connections with the mediodorsal nucleus in rabbit, sheep and cat. *Research Publications – Association for Research in Nervous and Mental Disease*, 27, 210–232.
- Sesack, S. R., Deutch, A. Y., Roth, R. H., & Bunney, B. S. (1989). Topographical organization of the efferent projections of the medial prefrontal cortex in the rat: An anterograde tract-tracing study with *Phaseolus vulgaris* leucoagglutinin. *Journal of Comparative Neurology*, 290, 213–242.
- Stroop, J. R. (1935). Studies of interference in serial verbal reactions. *Journal of Experimental Psychology*, 18, 643–662.
- Vendrell, P., Jungue, C., Pujol, J., Jurado, M. A., Molet, J., & Grafman, J. (1995). The role of prefrontal regions in the Stroop task. *Neuropsychologia*, 33, 341–352.
- Watanabe, M. (1986). Prefrontal unit activity during delayed conditional go/no-go discrimination in the monkey. II. Relation to go and no-go responses. *Brain Research*, 382, 15–27.
- Winocur, G. (1991). Conditional learning in aged rats: Evidence of hippocampal and prefrontal cortex impairment. *Neurobiology of Aging*, 13, 131–135.
- Winocur, G., & Eskes, G. (1998). Prefrontal cortex and caudate nucleus in conditional associative learning: Dissociated effects of selective brain lesions in rats. *Behavioral Neuroscience*, 112, 89–101.
- Wise, S. P., Murray, E. A., & Gerfen, C. R. (1996). The frontal cortex-basal ganglia system in primates. *Critical Reviews in Neurobiology*, 10, 317–335.

Received August 20, 2004

Final acceptance January 12, 2005