Rhesus Monkeys with Orbital Prefrontal Cortex Lesions Can Learn to Inhibit Prepotent Responses in the Reversed Reward Contingency Task

Monkeys with lesions of the orbital prefrontal cortex (PFo) are impaired on behavioral tasks that require the ability to respond flexibly to changes in reward contingency (e.g., object reversal learning and extinction). These and related findings in rodents and humans have led to the suggestion that PFo is critical for the inhibitory control needed to overcome prepotent responses. To test this idea, we trained rhesus monkeys with PFo lesions and unoperated controls on acquisition of the reversed reward contingency task. In this task, selecting the smaller of 2 food quantities (1 half peanut [1P]) leads to receipt of the larger quantity (4 half peanuts [4P]) and vice versa. Choice of a larger quantity of food is a reliable prepotent response, and, accordingly, all monkeys initially selected 4P rather than one. With experience, however, all monkeys learned to select 1P in order to receive 4. Surprisingly, monkeys with PFo lesions learned as quickly as unoperated controls. Thus, PFo lesions do not yield a deficit in all tests that require the inhibition of a prepotent response.

Keywords: affective inhibition, response control, reward magnitude

Introduction
Efficient problem solving requires flexibility in selecting responses, including the ability to discard or inhibit previously successful responses in favor of those that produce better outcomes. Laboratory tests of such flexibility often involve suppressing a previously correct response when reward contingencies change, and the full expression of this capacity depends on having an intact orbital prefrontal cortex (PFo). For example, monkeys with lesions that include PFo are impaired on object discrimination reversal learning (ODRL) (Butter 1969; Iversen and Mishkin 1970; Jones and Mishkin 1972; Dias and others 1996) and show resistance to extinction (Butter and others 1963, 1969; Butter 1969). In both tasks, when a previously rewarded response no longer produces reward, monkeys with PFo lesions continue to choose according to the previous reward contingency. This behavior has been regarded as a failure of behavioral inhibition, that is, a failure to suppress strong, habitual responses.

Behavioral inhibition is not a unitary construct; different parts of prefrontal cortex (PF) are held to exert different kinds and levels of inhibitory control. For example, in marmosets, lateral PF subserves the inhibitory control of “attentional” selection, which involves choices based on which stimulus features are currently relevant to that choice, whereas PFo subserves the inhibitory control of “affective” selection, which involves choice according to stimulus–reward associations (Dias and others 1996). Consistent with this idea, rhesus monkeys with PFo lesions do not appropriately alter their responses to objects after changes in either the value or contingency of the associated food rewards (Izquierdo and others 2004), and similar findings have been reported in humans (Bechara and others 1994; Fellows and Farah 2003) and rats (Gallagher and others 1999; Chudasama and Robbins 2003; McAlonan and Brown 2003; Schoenbaum and others 2003).

Reward magnitude also influences response choice (Campbell and Seiden 1974). For example, monkeys show a prepotent disposition to choose a larger quantity of food over a smaller one. The ability to inhibit this bias can be assessed in the reversed reward contingency (RRC) task (Boysen and Berntson 1995; Anderson and others 2000; Kralik and others 2002). In RRC, subjects are presented with 2 amounts of food simultaneously and are allowed to choose between them. Selection of the smaller amount (e.g., 1 half peanut [1P]) yields receipt of the larger amount (e.g., 4 half peanuts [4P]) and vice versa. Recently, Murray and others (2005) showed that macaque monkeys initially selected the larger of the 2 quantities of food as expected but eventually learned to select the smaller quantity of food, as required to receive the larger reward. Accordingly, RRC provides a stringent measure of inhibitory control in the affective domain: monkeys must suppress the prepotent choice of selecting larger food quantities. To test whether PFo subserves this type of inhibitory control, we examined the effects of PFo lesions on rhesus monkeys’ ability to acquire the RRC task.

Materials and Methods
Subjects
Nine male rhesus monkeys (Macaca mulatta) served as subjects in this study. They were housed individually in temperature-controlled rooms (76–80°F) with 12-h light/dark cycles. At the beginning of testing, the monkeys weighed 8–8.7 kg. All monkeys received primate chow (catalog number 5038, PMI Feeds Inc., St Louis, MO) supplemented with fresh fruit and vegetables. Water was available ad libitum. Three monkeys had bilateral ablations of the PFo, and 6 others served as unoperated controls. Data from the 6 controls have been reported previously (Murray and others 2005).

Before entering the present study, all monkeys had received extensive experience with object discrimination problems. In addition, they were evaluated (Izquierdo and others 2004) for the ability to adjust their responses in the face of changes in reward value (i.e., reinforcer devaluation) and reward contingency (i.e., reversal learning). The training histories for all monkeys were virtually identical. All procedures were approved by the National Institute of Mental Health Animal Care and Use Committee.

Surgery
Approximately 33 months prior to the initiation of the present study, 3 monkeys received bilateral ablation of the PFo. A detailed description of the surgical procedures is provided in Izquierdo and others (2004). The monkeys received the lesions in 2 stages balanced for hemisphere (left or right) of first surgery. The boundaries of the intended lesion (see Fig. 1)
were the fundus of the lateral orbital sulcus, laterally, and fundus of the rostral sulcus, medially; the rostral limit of the intended lesion was an imaginary line drawn between the anterior tips of the lateral and medial orbital sulci, and the caudal limit was approximately 7 mm rostral to the junction of the temporal and orbitosinular portions of the piriform cortex. The caudal boundary corresponds approximately to a coronal level of 28.65 mm anterior to the interaural plane in the brain atlas of Paxinos and others (2000). Accordingly, our lesion leaves intact the caudal agranular portions of the orbital surface.

Anesthesia was induced with ketamine hydrochloride (10 mg/kg intramuscularly [i.m.]) and maintained using isoflurane gas (1.0–3.0%, to effect). During surgery, all monkeys received isotonic fluids via an intravenous drip. Heart and respiration rates, body temperature, blood pressure, and expired CO2 were monitored throughout the procedure. After incision and retraction of the skin and galea, rongeurs were used to form a half-moon-shaped craniotomy behind the orbit. The dura mater was then cut along the dorsal edge of the bone opening and reflected ventrally. With the aid of an operating microscope, the boundaries of the lesion were identified. Then, using a combination of suction and electrocautery, PFo was removed by subpial aspiration through a fine-gauge metal sucker, insulated except at the tip. When the lesion was complete, the wound was closed in anatomical layers. Postoperatively, monkeys received dexamethasone sodium phosphate (0.4 mg/kg i.m.; Watson Laboratories, Inc., Corona, CA) to reduce swelling and prevent infection, respectively. Ketoprofen (10–15 mg i.m.; Fort Dodge Animal Health, Fort Dodge, IA) was administered as an analgesic for 3 days and ibuprofen (100 mg) was provided for 5 additional days.

**Lesion Assessment**

Lesions were assessed using postoperative **T**1-weighted magnetic resonance (MR) imaging scans (1.5-T magnet; fast spoiled gradient; echo time 5.8; repetition time 13.1; flip angle 30; number of excitations 8; 256 square matrix; field of view 100 mm; 1-mm slices) that were performed an average of 11.3 months after surgery. The lesion was mainly as intended and included Walker's areas 11, 13, 14, and the caudal performed an average of 11.3 months after surgery. The lesion was complete, the wound was closed in anatomical layers. Postoperatively, monkeys received dexamethasone sodium phosphate (0.4 mg/kg i.m.) and Cefazolin antibiotic (20 mg/kg i.m.; Watson Laboratories, Inc., Corona, CA) to reduce swelling and prevent infection, respectively. Ketoprofen (10–15 mg i.m.; Fort Dodge Animal Health, Fort Dodge, IA) was administered as an analgesic for 3 days and ibuprofen (100 mg) was provided for 5 additional days.

**Apparatus and Testing Procedure**

All testing was conducted in a Wisconsin General Test Apparatus, which comprised a test compartment lit by two 60-W light bulbs and a monkey compartment that was unlit during testing. An opaque screen separated the compartments during intertrial intervals. The test compartment included a test tray containing 2 food wells (diameter 38 mm; depth 6 mm) located 290 mm apart, center to center, on the midline of the tray. All testing was done in a darkened room.

![Figure 1. Left, ventral view of standard rhesus monkey brain showing location and extent of intended lesion (shaded region). Top right; coronal section 30 mm anterior to the interaural plane showing location and extent of intended PFo lesion (shaded region). Bottom right, representative **T**1-weighted MR image from PFo 1 at matching level. Numerical indicates distance (mm) from the interaural plane (0).](https://academic.oup.com/cercor/article-abstract/17/5/1154/344042)

On each trial, the opaque screen separating the monkey from the test compartment was raised, and the monkey was confronted with a choice: 1P (small reward) or 4P (large reward). The peanuts were placed in the palms of the experimenter’s gloved hands, and the 2 hands presented over the 2 food wells, in full view of the monkey. If the monkey selected the 4P quantity by touching or reaching toward the experimenter’s hand, the experimenter immediately closed both hands, turned her fists upside down, and dropped the 1P reward into the underlying food well. If the monkey selected the 1P quantity, the experimenter followed the same general procedure but dropped the 4P reward. The experimenter then quickly withdrew her hands and allowed the monkey to retrieve the food reward. A correct response was recorded when the animal selected 1P. Once the monkey had retrieved the food from the food well, the opaque door was lowered, thereby terminating the trial. Each monkey received a total of 20 trials, each separated by 20 s, per daily session. The side of presentation of the larger food amount was determined pseudorandomly, balanced within each block of 10 trials. Criterion was set at a mean of 90% correct responses over 5 consecutive sessions (i.e., 10 errors or fewer in 100 trials).

We also tested the possibility that the monkeys were using inadvertent cues provided by the experimenter to perform RRC. After the monkeys had reached criterion, they received additional test sessions administered by a second experimenter using the same procedure as described above. The monkeys were required to reattain criterion. If the monkeys were applying the rule, we expected that they would continue to perform well even when the task was administered by the second experimenter.

**Results**

All monkeys successfully learned the RRC task to a high degree of accuracy, with no significant differences between the operated and control groups in terms of the number of sessions (Mann–Whitney U test, U = 10; P = 0.8), trials (U = 10; P = 0.8), or errors (U = 12; P = 0.4) accrued up to, but not including, the criterion run. In addition, when tested by the second experimenter, 8 of the 9 monkeys quickly reattained criterion (see Table 2). The one monkey that required several sessions to reattain criterion, PFo 3, scored 70% correct in the first session

<table>
<thead>
<tr>
<th>Table 1</th>
<th>Percentage of PFo removed in each hemisphere</th>
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<tbody>
<tr>
<td>Monkey</td>
<td>Left</td>
</tr>
<tr>
<td>PFo 1</td>
<td>92.5</td>
</tr>
<tr>
<td>PFo 2</td>
<td>76.1</td>
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<tr>
<td>PFo 3</td>
<td>85.0</td>
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Note: Left, left hemisphere; Right, right hemisphere.

<table>
<thead>
<tr>
<th>Table 2</th>
<th>Number of sessions, trials, and errors to attain criterion (Experimenter 1) and reattain criterion (Experimenter 2) on the RRC task</th>
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<tbody>
<tr>
<td>Monkey</td>
<td>Experimenter 1</td>
</tr>
<tr>
<td></td>
<td>Sessions</td>
</tr>
<tr>
<td>PFo 1</td>
<td>62</td>
</tr>
<tr>
<td>PFo 2</td>
<td>37</td>
</tr>
<tr>
<td>PFo 3</td>
<td>25</td>
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<tr>
<td>Mean (±SEM)</td>
<td>41 (13)</td>
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<tr>
<td>Control 1</td>
<td>39</td>
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<tr>
<td>Control 2</td>
<td>53</td>
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<tr>
<td>Control 3</td>
<td>25</td>
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<tr>
<td>Control 4</td>
<td>133</td>
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<tr>
<td>Control 5</td>
<td>62</td>
</tr>
<tr>
<td>Control 6</td>
<td>13</td>
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<tr>
<td>Mean (±SEM)</td>
<td>54 (18)</td>
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Note: SEM, standard error of mean.

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with Experimenter 2 followed by 85% in the second, thereby showing good transfer. As was the case for initial acquisition, the groups did not differ in the number of sessions (Mann–Whitney U test, \( U = 7; P = 0.6 \)), trials (\( U = 7; P = 0.6 \)), or errors required to reattain criterion (\( U = 6.5; P = 0.5 \)). Thus, the monkeys were unlikely to be performing the RRC task on the basis of inadvertent cues provided by the experimenter.

The learning curves are illustrated in Figure 2. Initially, all animals chose the 4P reward, which counted as an error. For the first session, the PFo group made 16.6 ± 1.8 errors in 20 trials (mean ± standard error of mean), and the controls made 17.7 ± 0.6 errors, scores that did not significantly differ (Mann–Whitney U test, \( U = 6.5; P = 0.5 \)). Thus, both groups exhibited a prepotent response, as evidenced by their scoring significantly below chance levels during the first session (PFo group: \( t_{2} = 9.44, P = 0.01 \); controls: \( t_{5} = 28.74, P = 0.001 \)).

To examine whether monkeys with PFo lesions might have exhibited perseveration, we operationally defined 2 error types: perseverative (errors in sessions with >14 errors in 20 trials) and nonperseverative (13 errors or fewer in 20 trials), corresponding to below chance (binomial test, \( P = 0.037 \)) versus chance and above levels of performance, respectively. Figure 3 illustrates the number of errors of each type scored by each group. A 2-way analysis of variance with factors group (control, PFo) and error type (perseverative, nonperseverative) revealed neither a main effect of group (\( F_{1,7} = 0.62, P = 0.4 \)) nor a group by error type interaction (\( F_{1,7} = 0.07, P = 0.8 \)). Thus, by this measure, monkeys with PFo lesions made no more perseverative errors than did controls.

**Discussion**

In the RRC task, monkeys must detect the relationship between their response and the magnitude of reward outcome, and they must inhibit the prepotent tendency to reach for the large reward. Our results showed that monkeys with PFo lesions and their controls initially selected the large (4P) quantity of reward, thereby exhibiting a strong prepotent response (Fig. 2). With extended training, all monkeys learned to select the small (1P) quantity in order to receive the large reward. Like the unoperated controls, the PFo group overcame their prepotent tendency and solved the RRC problem. There was no evidence that the 2 groups of monkeys acquired the RRC task at a different rate or exhibited any differences in response perseveration. Although monkeys with PFo lesions were impaired in acquisition of the RRC, these same monkeys were impaired on a test of instrumental extinction that was conducted after completion of the present study (Izquierdo and Murray 2005). Consequently, neither extensive postoperative training nor recovery of function after the PFo lesion could account for the present negative result.

To our knowledge, this is the first demonstration that PFo lesions in rhesus monkeys leave intact the ability to suppress a response in the face of changed reward contingencies. Our findings with the RRC task thus contrast with evidence derived from other tests of affective inhibitory control, such as ODRL and extinction.

**Comparison with Previous Studies of Behavioral Inhibition**

As indicated earlier, several studies have shown that PFo lesions in monkeys yield an impairment on ODRL (e.g., Butter 1969; Iversen and Mishkin 1970; Jones and Mishkin 1972; Dias and others 1996). In addition, the same monkeys with PFo lesions studied here were also found to be significantly impaired on...
ODRL (Izquierdo and others 2004). Consequently, we can be confident that the same PFO removal that was effective in disrupting ODRL had no effect on the RRC task. Although both ODRL and RRC require monkeys to suppress a response to a stimulus that has been associated with greater reward in the recent past, the tasks differ in at least 2 ways.

Perhaps the most important difference is that in RRC, unlike in ODRL, food serves as both the discriminative stimulus and the reward outcome. Because the RRC task relies on the monkeys’ inherent, prepotent tendency to reach for a larger amount of food, it arguably tests inhibitory control in a more ethologically meaningful way than does ODRL, which involves objects associated arbitrarily with some value determined by the experimenter. Perhaps the previous findings of deficits on ODRL, compared with our negative results with RRC, indicate that PFO plays a more important role for learning the changing associations between nonfood objects and their current biological value (ODRL) than for comparable decisions based on food quantities (RRC) (see also Baxter and Murray 2002).

Another difference between RRC and ODRL is the rate of learning. Whereas a single reversal in ODRL is typically acquired in a few sessions, mastering the RRC task required roughly 50 sessions. Although there is no principled account of why PFO might be essential only in those situations in which responses are rapidly reversed, the difference between ODRL and RRC might be explained by a phenomenon related to speed of learning. Murray and Gaffan (2006) have previously argued that learning set is an important consideration in discrimination learning. Furthermore, learning set may be important in mediating repeated reversals of visual discriminations. Given that Browning and others (2006) have found that lesions of frontal cortex disrupt learning set, a possible explanation of the pattern of effects of PFO lesions is that learning set is important for ODRL but not for the learning of the single RRC problem administered here.

In another test of behavioral inhibition, object retrieval, marmosets with PFO lesions show perseverative reaching to food that is located directly in front of them but blocked by a solid, transparent barrier. These monkeys have a prepotent tendency to reach directly to the food but can inhibit this behavior if previously overtrained to reach around an opaque barrier (Wallis and others 2001; see also Diamond 1990). Setting aside possible species differences, in marmosets, PFO is rendered unnecessary by this intermediate training step. Could intermediate training account for our negative result? This seems unlikely because our monkeys with PFO lesions learned the RRC task without remedial training (e.g., correction trials) or symbolic mediation (e.g., use of Arabic numerals as discriminanda), methods which have been used by others to facilitate performance on this task (Boyesen and Berntson 1995; Silberberg and Fujita 1996; Kralik 2005) and which serve as intermediate training steps. A more likely account is that, although both object retrieval and RRC require suppressing reaches to visible food, in RRC, monkeys reach instead to a smaller quantity of visible food, but in object retrieval, they substitute a different reach trajectory to reach the same amount of food.

In neuroimaging and clinical studies of decision making, magnitude of reward outcome is an important factor. Although PFO is strongly activated in decision-making paradigms in which outcomes must be predicted on the basis of changing reward and penalty contingencies, such as ODRL tasks (Kringelbach and Rolls 2003; O’Doherty and others 2003), PFO is not activated when there is a high degree of conflict between predicted reward magnitude and outcome probability (Rogers and others 1999). In RRC, the food items serve as both discriminanda and reward. As a result, the visual information at the time of choice contradicts the expected outcome, at least during learning: selection of the “large reward” results in a small outcome. Thus, in the context of the finding of Rogers and others (1999), the absence of an essential contribution of PFO to the RRC task is not as surprising as it might first appear. In addition, in many settings, patients with PFO damage are not disinhibited in decision-making tasks per se (Bechara and others 1996; Rogers and others 1999; Manes and others 2002). These findings contradict the idea that PFO damage invariably results in impulsive decision making and thus accords with the present results.

As for rats, it has been observed that PFO lesions cause an inhibitory control deficit, but not a general one. Specifically, rats with PFO lesions do not show an inability to withhold prepotent “go” responses in go–no go discrimination tasks but are impaired instead at responding to changes in the relationship between odor or visual stimuli and predicted outcomes (Schoenbaum and others 2002, 2003; Chudasama and Robbins 2003; see also McAlonan and Brown 2003). These data, too, are consistent with the present findings.

Perseveration

Given the widely held view that perseveration follows damage to PFO, it may seem surprising that there was no evidence for perseverative responding by monkeys with PFO lesions on the RRC task. As already indicated, the same monkeys with PFO lesions tested in the present study were found to have a deficit on the ODRL task (Izquierdo and others 2004). In that study, too, there was no evidence for a greater number of perseverative errors by monkeys with PFO lesions relative to controls. Although this seems to conflict with the report of Jones and Mishkin (1972), who found an increase in perseverative errors after PFO lesions, we note that their PFO removals included not only the orbital surface medial to the lateral orbital sulcus, as here, but also the cortex on the ventral convexity. In agreement with past findings (Iversen and Mishkin 1970), lesions limited to the orbital surface do not lead to an increase in perseverative errors on ODRL. Moreover, perseverative responses after PFO damage are not found in other settings. For example, Bussey and others (2001) found no perseveration in monkeys acquiring associations between stimuli and actions after removal of PFO and the adjacent cortex on the ventral convexity.

Possible Neural Substrates of RRC Task Performance

The finding that monkeys with PFO lesions successfully change their behavioral response according to RRCs implicates other brain regions as neural substrates for this task.

One candidate region for mediating RRC is the medial PF (PFm; areas 24, 32, and 25), which has been implicated in linking actions with reward. For example, lesions within PFm disrupt preoperatively acquired reward conditional response associations (Hadland and others 2003). In addition, the activity of neurons in anterior cingulate cortex (area 24) reflect changes in reward magnitude for various responses (Shima and Tanji 1998; Matsumoto and Tanaka 2004). Furthermore, human neuroimaging studies report increased activation in the PFm when reward levels vary and responses have to be changed (Bush and others 2002; O’Doherty and others 2003; Elliott and
others 2004; Rogers and others 2004). Perhaps in RRC, as in the reward conditional response selection task used by Hadland and others (2003), the small and large quantities of food serve as conditional cues to guide response selection. If so, RRC may depend on the integrity of PfO. Finally, anterior cingulate cortex may be involved in response selection during attentional conflicts (Pardo and others 1990; Botvinik and others 2004), which may also involve aspects of inhibitory control.

Ventrolateral PfO (Pfvl) could also contribute to the RRC task. Pfvl, along with PfO, is a key area in the arbitrary association of objects with actions and with other objects (Eacott and Gaffan 1992; Bussey and others 2001, 2002). Pfvl is also involved in learning associations in which the presence or absence of food signals the appropriate motor response (Parker and Gaffan 1998). A role for Pfvl is consistent with the idea that arbitrary associations between the stimuli and actions may underlie performance on the RRC task (Murray and others 2005). In addition, Diamond (1990) reported that rhesus monkeys with dorsolateral Pf lesions were unable to suppress inappropriate barrier reaches to a transparent box on the object retrieval task, suggesting that the dorsolateral Pf may also play a role in acquiring the RRC task.

Finally, the caudal, largely agranular orbital cortex might also underlie performance of the RRC task. These areas are likely homologues of what is called PfO in rats and other mammals (Preuss 1995) but were spared by our lesions. Little is known about the function of these areas, but data from monkeys show them to be extensively interconnected with both gustatory and olfactory cortices, as well as with PfO and its visual inputs (Carmichael and Price 1996), which could be important in RRC.

Conclusion

An inability to adjust to changes in reward contingency is associated with damage to PfO in monkeys (Jones and Mishkin 1972; Dias and others 1996; Izquierdo and others 2004) and humans (Bechara and others 1994; Rahman and others 1999; Blair 2001; Fellows and Farah 2003). Yet, our data show that in rhesus monkeys, selecting among conflicting reward options and overcoming prepotent responses does not require an intact PfO. We conclude that the concept of inhibitory control used to characterize PfO function is underspecified and that the neural mechanisms that guide response selection require further investigation.

Notes

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