The Contribution of the Medial Prefrontal Cortex, Orbitofrontal Cortex, and Dorsomedial Striatum to Behavioral Flexibility

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ABSTRACT: Behavioral flexibility refers to the ability to shift strategies or response patterns with a change in environmental contingencies. The frontal lobe and basal ganglia are two brain regions implicated in various components for successfully adapting to changed environmental contingencies. This paper discusses a series of experiments that investigate the contributions of the rat prefrontal area, infralimbic area, orbitofrontal cortex, and dorsomedial striatum to behavioral flexibility. Orbitofrontal cortex inactivation did not impair initial learning of discrimination tests, but it impaired reversal learning due to perseverance in the previously learned choice pattern. Inactivation of the prelimbic area did not affect acquisition or reversal learning of different discrimination tests, but it selectively impaired learning when rats had to inhibit one strategy and shift to using a new strategy. However, comparable to orbitofrontal cortex inactivation, strategy-switching deficits following prelimbic inactivation resulted from a perseverance of the previously relevant strategy. Fewer studies have examined the infralimbic region, but there is some evidence suggesting that this region supports reversal learning by maintaining the reliable execution of a new choice pattern. Dorsomedial striatal inactivation impaired both reversal learning and strategy switching. The behavioral flexibility deficits following dorsomedial striatal inactivation resulted from the inability to maintain a new choice pattern once selected. Taken together, the results suggest that orbitofrontal and prelimbic sub-regions differentially contribute to behavioral flexibility, but they are both critical for the initial inhibition of a previously learned strategy, while the dorsomedial striatum plays a broader role in behavioral flexibility and supports a process that allows the reliable execution of a new strategy once selected.

KEYWORDS: orbitofrontal cortex; prelimbic; infralimbic; striatum; learning

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INTRODUCTION

In an ever-changing environment, the ability to adapt new choice patterns is essential for daily living and often survival. Considerable evidence indicates that the frontal cortex supports learning when conditions require inhibition of a previously relevant strategy and acquiring a new strategy.\textsuperscript{1–5} The frontal cortex, however, consists of several subregions, which raises the important issue of whether these separate subareas support distinct behavioral flexibility functions. The frontal cortex is also known to project heavily to the basal ganglia in a highly organized manner in which there are distinct cortico-basal ganglia loops.\textsuperscript{6} This raises a further issue of whether and how distinct cortico-striatal circuits in this highly organized network may differentially support a shift in strategy.

A model put forth by Wise and colleagues to explain the functional organization of the primate frontal cortex proposes that different conditions require different types of cognitive/behavioral processes to facilitate behavioral flexibility, and these processes are mediated by separate primate prefrontal cortex areas.\textsuperscript{7} Specifically, the model proposes that there is a lower-order process for the shifting of specific choices within a dimension. This process allows the approach to and avoidance of a particular stimulus or scene as required in discrimination tasks that involve reversal learning. The model also states that there is a higher-order process when conditions demand learning about stimulus attributes as opposed to a stimulus as a whole. In these cases, learning must go beyond simply attaching a positive or negative valence to stimuli within a particular dimension and instead require attention to components of an object or scene or abstract rules about component objects or scenes. This may involve learning the relationship between different stimulus components—e.g., paired associate learning—or complete response inhibition to stimuli in one dimension while learning what stimulus in a different dimension is correct. For example, one may use street signs to reach a location during the day but use directional information, such as memory for turn sequences and distance, to reach the same location at night. Thus, higher-order processing enables a subject to reconceptualize his or her approach to a task and attend to a new type of information. This type of flexibility is required during extradimensional shifts. Reversal learning is representative of lower-order processing because it involves a change only in exemplar, not in category. The extradimensional shift is representative of higher-order processing because it requires taking a fundamentally new approach to solving a task that entails using a new strategy. Although the terms lower-order and higher-order might suggest a serial hierarchical organization, the model explicitly hypothesizes that different operations are subserved by different prefrontal subregions that function independently of each other.

To explore the model proposed by Wise and colleagues\textsuperscript{7} related to rodent prefrontal cortex functioning, we conducted a series of experiments that
investigated whether temporarily inactivating the prelimbic cortex or the orbitofrontal cortex affected separate processes that enabled behavioral flexibility (see Fig. 1). The experiments were conducted using a variety of discrimination tasks that tested the effects of inactivation on initial learning, reversal learning, and times when conditions required a shift in strategy. Because the orbitofrontal cortex and the prelimbic area both project to the dorsomedial striatum, the effects of dorsomedial striatal inactivation were also on examined on various discrimination tests.

**PRELIMBIC INVOLVEMENT IN BEHAVIORAL FLEXIBILITY**

In initial experiments, the role of the medial prefrontal cortex in switching strategies was examined. In particular, the contribution of the prelimbic
subregion was investigated on tasks that involved learning a discrimination based on one type of strategy and then being required to learn a new strategy in the same context, e.g., to make a choice based on visuospatial information first and then base a choice on odor information. In one set of studies, rats were permanently implanted with guide cannula aimed toward the prelimbic subregion (see Fig. 1A). Subsequently, rats were trained to dig in a sand cup to receive a cereal reinforcement using a procedure similar to that of Bunsey and Eichenbaum. The sands cups contained a spice, such as cinnamon or nutmeg, mixed in with the sand to provide a distinct odor to the sand cups. The two odor cups were randomly switched between two different spatial locations across trials. In this task, rats first learned to base their choice on either odor information or visuospatial information (location of the sand cup). After learning to discriminate based on one of these strategies in one session, rats had to switch their choice pattern based on the other strategy in the following session; e.g., odor information could be relevant on acquisition and spatial information could be relevant on the shift phase. Five minutes prior to each test session, a rat received an intracranial infusion of either saline or the local anesthetic, bupivacaine, into the prelimbic area. The learning criterion for each session was ten consecutive correct trials. The finding revealed that prelimbic inactivation did not impair the initial learning of either odor or spatial discrimination. In contrast, prelimbic inactivation did impair learning when rats had to shift between an odor and spatial discrimination (see Fig. 2A). This was the case whether rats first learned an odor discrimination or first learned a spatial discrimination. The pattern of results suggests that the prelimbic area supports behavioral flexibility when conditions require a shift in strategy.

The contribution of the prelimbic area to extradimensional shifts does not appear to be limited to specific attribute information. For example, the prelimbic area does not facilitate extradimensional shifts that only involve the flexible use of spatial or visual cue information. This is because neurotoxic lesions of the prelimbic and infralimbic areas impair a shift between the use of odor information and the use of texture information. Furthermore, dopamine receptor blockade or \( N \)-methyl-\( D \)-aspartate (NMDA) receptor blockade in the prelimbic area impairs a shift between egocentric response and visual cue strategies. Together, these findings suggest that the prelimbic area enables a shift in strategy across a variety of stimulus dimensions.

Several studies have explored what process or processes are disrupted following prelimbic inactivation that produces a deficit in shifting strategies. One possibility is that the prelimbic region facilitates the ability to initially inhibit a previously relevant strategy and/or to generate a new strategy. In this case, prelimbic inactivation should produce a predominance of errors during the initial trials in the shift phase. These errors are commonly referred to as perseverative errors. Another possibility is that the prelimbic area supports a process that allows an individual to reliably execute or learn a new strategy once the new strategy is selected. This process would prevent or minimize regressions to
FIGURE 2. (A) Mean (± s.e.m.) trials to criterion in odor acquisition and shift to a spatial discrimination following bilateral infusions of saline or bupivacaine into the prelimbic area. Prelimbic inactivation did not affect acquisition, but did impair a shift to a spatial strategy. The treatment received in each phase of testing is in bold. SAL = saline; BUP = bupivacaine. *P < 0.05 versus controls. (B) Mean (± s.e.m.) number of perseverative and regressive errors in reversal learning after bilateral infusions of saline or bupivacaine into the prelimbic area. There was a significant increase in perseverative, but not regressive errors following prelimbic inactivation. SAL = saline; BUP = bupivacaine. *P < 0.05 versus controls.

the previously relevant strategy once the new, presently relevant strategy is selected. In this case, prelimbic inactivation should not produce a significant increase in errors during the initial trials of the shift phase, but rather should lead to a greater number of errors once a rat has selected the new, presently
relevant strategy. We have referred to these errors as regressive errors because a subject has chosen the new correct choice and has been reinforced for it, but regresses to the previous strategy that is no longer reinforced. There have been different operational definitions used to define perseverative and regressive errors but the patterns of results have remained the same whatever definition was used. More specifically, in some studies perseveration was defined as making the previously correct choice three or four times in consecutive blocks of four trials. Once a rat made less than three previously correct choices in a block the errors were no longer counted as perseverative errors, but rather were counted as regressive errors. With this definition, a rat was considered to be perseverating when it was choosing the previously correct choice on the majority of trials. Once it was just as likely to make the new correct choice as the previous correct choice, the errors were considered regressive. We have also defined perseveration as the number of trials a rat continued to make the previously correct choice after making an initial error until it made the new correct choice. Subsequently, every error after making the first correct choice was counted as a regressive error. In multiple experiments in which manipulations of the prelimbic area impaired a shift in strategy, the deficit resulted from an increase in perseverative errors but not regressive errors. This includes the study described above in which prelimbic inactivation impaired a shift from an odor to a spatial strategy (see FIG. 2B). The pattern of results suggests that the prelimbic area enables a shift in strategy by facilitating the initial inhibition of a previously learned strategy and/or the generation of a new strategy.

The findings described above suggest that the prelimbic area contributes to behavioral flexibility, at least in part, by supporting a shift in strategy. A number of other experiments have also investigated whether the prelimbic area plays a role in other types of behavioral flexibility, such as reversal learning. In one study using the sand-digging test, rats were trained on acquisition and reversal learning of a two-choice odor discrimination. Prelimbic inactivation did not impair acquisition or reversal learning of a two-choice odor discrimination (see FIG. 3A). This lack of effect on reversal learning has also been reported with prelimbic inactivation or lesions on other types of reversal-learning tests, such as spatial, egocentric response, and texture. Thus, based on multiple behavioral-flexibility tests, the findings might suggest that the prelimbic area supports a higher-order process that involves a shift in strategy, but not a lower-order process as assessed by reversal learning.

One issue that arises when examining a brain region on extradimensional shifts versus reversal-learning tests is that extradimensional shifts are often more difficult than reversal-learning tests. Because of the difference in level of difficulty between reversal-learning tests and extradimensional shifts, it is unknown whether a differential effect of prefrontal subregion inactivation in these tasks is related to the behavioral operation necessary to learn the task or due to the level of difficulty. To address this issue, a two-choice odor
FIGURE 3. (A) Mean (± s.e.m.) trials to criterion in two-choice odor acquisition and reversal learning following bilateral infusions of saline or bupivacaine into the prelimbic area. Prelimbic inactivation did not affect acquisition or reversal learning. The treatment received in each phase of testing is in bold. SAL = saline; BUP = bupivacaine. (B) Mean (± s.e.m.) trials to criterion in four-choice odor acquisition and reversal learning following bilateral infusions of saline or bupivacaine into the prelimbic area. Prelimbic inactivation did not affect acquisition or reversal learning. The treatment received in each phase of testing is in bold. SAL = saline; BUP = bupivacaine.

reversal-learning test was changed to a four-choice odor reversal-learning test. A four-choice discrimination test required significantly more trials to acquire and reverse than a two-choice discrimination, but it was comparable to an extradimensional shift. Thus, the same behavioral operation was required in the four-choice reversal-learning test as in the two-choice reversal-learning test, but the four-choice test required a greater number of trials to learn. Even in the four-choice odor discrimination test, prelimbic inactivation did
not impair reversal learning (see Fig. 3B). This suggests that the prelimbic area supports a higher-order process that enables a shift in strategy but is not involved in a lower-order process in which the same general strategy remains the same, but a shift in specific choices is reversed.

A possible alternative interpretation of prelimbic inactivation’s affecting extradimensional shifts, but not reversal-learning tests, is not related to the behavioral operation required but rather is due to different reinforcement contingencies in the shift phase. In particular, in a reversal-learning task the previously reinforced choice is never reinforced during the reversal phase (e.g., the cinnamon odor would be reinforced in 100% of the trials on acquisition and in 0% of the trials on reversal). In an extradimensional shift, the previously reinforced choice or strategy is reinforced in 50% of the trials during the shift phase. For example, in acquisition a rat first learns to choose the cinnamon odor cue and to avoid the nutmeg odor cue, as well as to disregard the spatial location of the cup. During the shift, a rat learns to choose spatial location A and to avoid spatial location B while ignoring odor information. However, the different odor cues and spatial locations are pseudorandomly combined such that for half the trials the previously relevant cinnamon odor cue is in spatial location A and therefore is still reinforced in 50% of the trials. Thus, manipulation of the prelimbic area may lead to a deficit in an extradimensional shift, but not in reversal-learning, because of a difficulty abandoning a previously relevant strategy or choice that is still being partially reinforced.

To investigate this possibility, we examined the effects of prelimbic inactivation on a test that has the same reinforcement contingencies as an extradimensional shift, but involves reversal learning. Specifically, prelimbic inactivation on spatial reversal learning was examined. To inactivate the prelimbic area, the gamma aminobutyric acid (GABA)-A agonist muscimol was infused into the prelimbic area. The same learning criterion of ten consecutive correct trials was used as in past experiments. Acquisition of the spatial discrimination was the same as past studies such that a rat learned to choose the sand cup in spatial location A and to avoid the sand cup in spatial location B. In reversal learning, the previously correct spatial location A was still reinforced in 50% of the trials when chosen, while location B was reinforced in 100% of the trials when chosen. This mimics the reinforcement contingencies in the extradimensional shift in which the stimulus choice that was reinforced on acquisition is still reinforced in 50% of the trials when chosen. However, in this test, the relevant stimulus information remained the same in the switch. Prelimbic inactivation did not impair spatial reversal learning using a 50%/100% reinforcement contingency. These findings suggest that the prelimbic inactivation-induced deficit in an extradimensional shift, but not a reversal-learning task, cannot be due to a difference in reinforcement contingencies, but rather is due to a difference in the strategic requirements of the two tasks. Therefore, the findings suggest that the prelimbic areas support particular types of behavioral flexibility based on the behavioral operation required.
As stated above, a lower-order process allows the approach to and avoidance of a particular stimulus or scene. A lower-order process treats a stimulus or scene as a whole by applying a positive or negative valence. A higher-order process is required when conditions demand learning about stimulus attributes as opposed to a stimulus as a whole. In these cases, learning must go beyond simply attaching a positive or negative valence to stimuli within a particular dimension and instead requires attention to components of an object or scene—or abstract rules about component objects or scenes. Based on this idea of a higher-order process, there should be other conditions besides extradimensional shifts that would require a higher-order process. In one experiment, Dias and Aggleton\textsuperscript{17} studied learning in a spatial-delayed match-to-sample test and nonmatch-to-sample test. Rats have a natural tendency to alternate\textsuperscript{18} and thus can readily acquire a nonmatch-to-sample task that requires a subject to alternate choices between the sample and the test phase. This tendency to alternate has been commonly shown in spontaneous alternation tests in which a rat tends to choose the least-recently visited arm of a maze.\textsuperscript{19} If rats are first trained to acquire a match-to-sample rule and therefore must switch away from their natural bias of using a nonmatch-to-sample strategy, prelimbic lesions impair acquisition.\textsuperscript{17} Furthermore, if rats are first trained on a match-to-sample task and then switched to the nonmatch-to-sample version, prelimbic lesions still impair the shift. Thus, based on a situation where a valence could not simply be applied to either spatial location, but a more abstract rule applied, the prelimbic area appears critical for behavioral flexibility.

Paired-associate learning represents another way to examine whether a prefrontal cortex subregion supports a higher-order process. In paired-associate learning, a subject must learn to make a choice when two specific stimuli are associated together, but avoid choosing either stimulus when associated with other stimuli. Under these conditions a valence cannot be simply attributed to a particular stimulus. To examine whether the prelimbic area is critical for paired-associate learning, rats were trained on an object-spatial location paired-associate test using a go/no-go procedure.\textsuperscript{20} Two objects, black and white blocks, were each placed in two different spatial locations. An object in one spatial location was associated with reinforcement, while in the other spatial location the object was never associated with reinforcement. The two objects had opposite pairings of spatial locations associated with reinforcement or no reinforcement. Therefore, in this task a rat must learn the association between spatial and visual object pairs and flexibly adapt responding. Prelimbic lesions impaired the learning of this paired-associate test. However, prelimbic lesions do not impair learning of either a visual object discrimination or a spatial discrimination.\textsuperscript{10}

In summary, the prelimbic areas support behavioral flexibility in a number of conditions that include extradimensional shifts, switching between match-to-sample and nonmatch-to-sample rules, and paired-associate learning. In contrast, the prelimbic area does not appear critical for behavioral flexibility.
when conditions require the use of the same general strategy, but a shift in specific choices is required in reversal learning. Furthermore, when conditions require a shift in strategies, the prelimbic area may support the initial inhibition of the previously learned strategy and/or the ability to generate a new strategy.

**INFRALIMBIC INVOLVEMENT IN BEHAVIORAL FLEXIBILITY**

In several of the experiments described above, inactivation or lesions of the prelimbic area also partly encompassed the infralimbic area, which is found ventral to the prelimbic cortex. Thus, the deficits observed on certain set-shifting tests may have arisen because of damage to the infralimbic area or to a combination of the prelimbic and infralimbic subregions. A few experiments have specifically studied the infralimbic cortex in behavioral flexibility. Taken together, the experiments have not led to a clear understanding of how the infralimbic cortex may contribute to behavioral flexibility. For example, a couple of experiments have found that infralimbic lesions do not impair initial acquisition of a visual cue discrimination, but do impair visual cue reversal learning.\(^{21,22}\) Interestingly, one study found that the reversal-learning deficit was not due to perseveration of the previously relevant choice pattern, but was due to the inability to reliably execute the new choice pattern once selected.\(^{21}\) However, other studies reported that infralimbic lesions do not impair extinction of tone-shock pairings,\(^{23}\) spatial reversal learning,\(^{24}\) or latent inhibition.\(^{25}\) Thus, infralimbic lesions impair learning when conditions require a shift in choice patterns in certain studies but not in others.

Killcross and Coutureau have investigated the effects of infralimbic lesions on the formation of habits with instrumental conditioning.\(^{26}\) The study follows from the idea that in the early stage of instrumental conditioning a response is based on acquiring an action–outcome association, and the goal-directed outcome must be actively maintained for accurate responding.\(^{27,28}\) With more extensive training, a stimulus–response habit is formed in which the goal-directed outcome no longer is actively maintained. This idea of the formation of actions and habits has received empirical support using instrumental devaluation procedures.\(^{29}\) In particular, with limited instrumental training of a bar-press response for a food reward, instrumental performance reduces when the food reward or outcome is devalued either by pairing the food with lithium chloride or by satiating the rat with the food prior to the test session. In contrast, with extensive training, devaluing the outcome does not reduce instrumental performance. Lesions or temporary inactivation of the infralimbic area prevented the insensitivity to the instrumental devaluation observed with extensive training such that infralimbic lesions now showed a decreased instrumental performance.\(^{26}\) These findings suggest that the infralimbic area may be important for the development of a stimulus–response habit or a more routed choice
pattern. These results may be comparable to the reversal-learning deficit observed by Chudasama and Robbins following infralimbic lesions. In this case, a reversal-learning deficit did not cause an increase in perseverative errors, but it did cause an increase in regressive errors. More specifically, if the infralimbic region is critical for the formation and maintenance of a habit or routine choice pattern, then lesions of the infralimbic area will not lead to perseveration of the learned choice pattern when conditions are reversed and a subject must inhibit the expression of the previously relevant choice pattern. That is, a subject will be more likely to initially give up the previously relevant choice pattern, but will have difficulty forming the new relevant choice pattern because the infralimbic region is critical for habit formation. In reversal-learning, this would not lead to an increase in perseveration, but it would lead to an increase in reliably executing the new choice pattern once selected. This is the pattern observed with infralimbic lesions in a visual cue reversal-learning task.

One limitation of the interpretation provided above is that the findings from Killcross and Coutureau suggest that the infralimbic area is critical for the development of a habit formation, and thus in the acquisition phase of these discrimination tests infralimbic lesions should impair this initial learning. However, different studies have reported that infralimbic lesions do not impair acquisition. One possibility is that in these tasks a subject is still operating in an action–outcome mode even at the end of the acquisition phase, and thus the infralimbic area is not engaged. An alternative possibility is that in many of the discrimination tests there is extensive pretraining before acquisition testing begins, and this pretraining influences the initial learning process. Clearly, there is a need for a more systematic examination of experiments of the infralimbic area to better understand how this area may contribute to behavioral flexibility.

**ORBITOFRONTAL CORTEX INVOLVEMENT IN BEHAVIORAL FLEXIBILITY**

The rat orbitofrontal cortex is located predominantly in the lateral portion of the frontal cortex. Comparable to the medial frontal cortex in rats, it contains different subdivisions. The majority of experiments that have investigated the role of the rat orbitofrontal cortex in behavioral flexibility have focused on the lateral orbital, ventral orbital, and/or agranular insular regions. The findings described below come from studies that involve manipulations of the lateral areas of the orbitofrontal cortex.

Studies investigating the contribution of the orbitofrontal cortex to behavioral flexibility have predominantly used reversal-learning tests. Studies involving lesions centered in the orbitofrontal cortex have commonly found that orbitofrontal damage does not impair acquisition of different discrimination
tests, but does impair reversal-learning.\textsuperscript{21,24,30–33} This occurred in a number of different types of reversal-learning tests that involved the flexible use of odor, visual cue, tactile, or spatial information. In a comparable manner, we found that infusion of muscimol into the lateral orbital or ventral orbital frontal cortex did not impair acquisition of a two-choice odor discrimination, but did impair reversal-learning (see Fig. 4A).\textsuperscript{34} The reversal-learning impairment observed with orbitofrontal cortex inactivation was comparable whether muscimol injections were centered in the lateral or the ventral regions of the orbitofrontal cortex. Similar to that observed with prelimbic inactivation in extradimensional shifts, orbitofrontal cortex inactivation impaired reversal learning by selectively increasing perseverative errors (see Fig. 4B). This selective increase in perseverative errors has also been observed in other studies in which neurotoxic lesions of the orbitofrontal cortex impaired reversal learning.\textsuperscript{21,24} These findings suggest that the orbitofrontal cortex supports learning when conditions require a switch in choice patterns as required in reversal learning and facilitates the ability to initially inhibit a previously relevant choice pattern and/or to generate a new choice pattern.

Orbitofrontal cortex lesions or inactivation producing perseveration during reversal learning has been reported for two-choice reversal-learning tests.\textsuperscript{21,24,30–34} We explored whether a similar deficit would occur with orbitofrontal cortex inactivation during a four-choice odor discrimination. One advantage of using a four-choice discrimination is that two of the stimuli are never reinforced during acquisition or reversal learning. Therefore, this can provide information about whether the orbitofrontal cortex is important in reducing interference to irrelevant stimuli. In this study, irrelevant errors during the reversal-learning phase were counted as the number of trials in which a rat chose the never-reinforced stimuli after an initial choice. Comparable to the two-choice discrimination, orbitofrontal cortex inactivation did not impair initial learning, but did impair reversal learning (see Fig. 5A). However, the error pattern that emerged was unique. Specifically, orbitofrontal cortex inactivation increased perseverative, regressive, and irrelevant errors (see Fig. 5B). This pattern of results suggests that under these conditions the orbitofrontal cortex is not only critical for initial inhibition of a previously relevant choice pattern, but also critical for maintaining a choice pattern once selected, as well as reducing interference to irrelevant stimuli. With the increased number of stimuli to discriminate among, orbitofrontal cortex inactivation may lead to this broader range of errors in reversal learning because the orbitofrontal cortex is critical for a lower-order process that applies a valence to particular stimuli. Therefore, increasing the number of stimuli that must be inhibited from choosing requires an increased elimination process that manifests itself following orbitofrontal cortex inactivation as an increase in perseverative, regressive, and irrelevant errors.

There are findings from devaluation paradigms that also suggest that the orbitofrontal cortex is critical for flexible responding when a valence can
FIGURE 4. (A) Mean (± s.e.m.) trials to criterion in two-choice odor acquisition and reversal learning following bilateral infusions of saline or muscimol into the orbitofrontal cortex. Orbitofrontal cortex inactivation did not affect acquisition but impaired reversal learning. The treatment received in each phase of testing is in bold. SAL = saline; MUS = muscimol. *P < 0.05 versus controls. (B) Mean (± s.e.m.) number of perseverative and regressive errors in two-choice odor reversal learning after bilateral infusions of saline or muscimol into the orbitofrontal cortex. There was a significant increase in perseverative, but not regressive errors following orbitofrontal cortex inactivation. SAL = saline; MUS = muscimol. *P < 0.05 versus controls.

be applied to a stimulus. More specifically, orbitofrontal cortex lesions fail to decrease responding to a conditioned stimulus following devaluation of a reinforcer.35,36 In devaluation paradigms a subject must first learn to respond to a cue that is associated with presentation of a food reinforcer. After the
FIGURE 5. (A) Mean (± s.e.m.) trials to criterion in four-choice odor acquisition and reversal learning following bilateral infusions of saline or muscimol into the orbitofrontal cortex. Orbitofrontal cortex inactivation did not affect acquisition but impaired reversal learning. The treatment received in each phase of testing is in bold. SAL = saline; MUS = muscimol. *P < 0.05 versus controls. (B) Mean (± s.e.m.) number of perseverative, regressive, and irrelevant errors in four-choice odor reversal learning after bilateral infusions of saline or muscimol into the orbitofrontal cortex. There was a significant increase in perseverative, regressive, and irrelevant errors following orbitofrontal cortex inactivation. SAL = saline; PVSR = perseverative; MUS = muscimol. *P < 0.05 versus controls.

reinforcer is devalued by pairing the food with illness, this new association leads a normal rat to decrease responding to the previously associated cue, but orbitofrontal cortex lesions prevent rats from adjusting their responding to the cue.

Although there is convincing evidence that the rat orbitofrontal cortex is involved in learning when contingencies are reversed, few studies have
examined the rat orbitofrontal cortex in other tests that require behavioral flexibility. One investigation studied whether orbitofrontal cortex lesions affect an extradimensional shift involving the use of odor and tactile information.\textsuperscript{32} The study found that orbitofrontal cortex lesions do not impair performance in an extradimensional shift, but the same lesions do impair reversal learning. Taken together with the findings from prelimbic inactivation, these findings suggest that the prelimbic and orbitofrontal cortex support different behavioral operations to enable behavioral flexibility. This double dissociation observed between the orbitofrontal cortex and the prelimbic cortex on reversal-learning tests and extradimensional shifts cannot be explained by differences in the level of difficulty because orbitofrontal cortex inactivation also leads to impairments on a four-choice reversal-learning test, but prelimbic inactivation does not impair four-choice reversal learning. Instead, the pattern of results suggests that these separate prefrontal cortex regions differentially contribute to behavioral flexibility.

**DORSOMEDIAL STRIATAL INVOLVEMENT IN BEHAVIORAL FLEXIBILITY**

There is evidence that the orbitofrontal cortex and the prelimbic cortex differentially contribute to behavioral flexibility related to the behavioral operation required to flexibly adapt. Both the orbitofrontal cortex and the prelimbic area project to the dorsomedial striatum.\textsuperscript{8} Because the dorsomedial striatum receives input from both of these prefrontal cortex areas, this brain area may also contribute to behavioral flexibility. Using a similar approach to investigate the contributions of the prefrontal cortex subregions to behavioral flexibility, a series of experiments have examined the effects of dorsomedial striatal inactivation on acquisition, reversal learning, and extradimensional shifts of different discrimination tests.

In the initial experiment, the effects of tetracaine injections into the dorsomedial striatum were investigated on the acquisition of either a visual cue or an egocentric response discrimination, then a shift to the other strategy.\textsuperscript{37} In the egocentric response discrimination, a rat was required to make the same turn relative to its body (left or right) in order to receive a cereal reinforcement. In an egocentric response discrimination, the idea is that a rat learns to use proprioceptive and vestibular information to guide its response. Dorsomedial striatal inactivation did not impair acquisition of either a visual cue or an egocentric response strategy. Dorsomedial striatal inactivation did impair a shift between a visual cue and an egocentric response strategy. These results are comparable to those observed with manipulations of the prelimbic area. However, an examination of the error pattern revealed that dorsomedial striatal inactivation did not increase perseveration, but rather led to an increase in regressive errors. This was the opposite pattern to that observed with prelimbic
inactivation, suggesting that these two areas both enable a shift in strategy, but they support different processes to facilitate an adaptation in strategy.

The dorsomedial striatum also plays a role in reversal learning. While inactivation of dorsomedial striatum did not impair acquisition of different discrimination tests, inactivation did impair reversal learning.\textsuperscript{38,39} This was the case for spatial reversal learning as well as egocentric response reversal learning (see Fig. 6A). These findings are comparable to previous studies demonstrating that dorsomedial striatal lesions impair reversal learning.\textsuperscript{40–42} In our experiments, an examination of the error pattern revealed that dorsomedial striatal inactivation also led to a selective increase in regressive errors during reversal learning comparable to that observed during the extradimensional shift (see Fig. 6B). Again, this is the opposite pattern to that observed in two-choice reversal-learning tests following orbitofrontal cortex inactivation.

The findings from studying the effects of dorsomedial striatal inactivation suggest that this striatal region plays a broader role in behavioral flexibility then either the prelimbic area or the orbitofrontal cortex alone. This may not be surprising because both regions project to the dorsomedial striatum. Furthermore, the dorsomedial striatum appears critical for the maintenance or reliable execution of a strategy once selected, but it is not critical for the initial inhibition of the previously relevant strategy or the generation of a new strategy. These results suggest that the dorsomedial striatum may dynamically interact with multiple prefrontal cortex subregions to facilitate behavioral flexibility in a distinct but complementary manner. More specifically, prefrontal cortex subregions may be critical for the generation of a new strategy. This allows the initial inhibition of the previously relevant strategy. However, once a new strategy is generated, it must be executed into the appropriate response pattern. The striatum, in coordination with different prefrontal cortex areas, may facilitate the execution of the appropriate response pattern for a particular strategy that is generated. Thus, the striatum in linking a particular response pattern with a specific strategy allows the reliable execution of a strategy once generated, as well as continual inhibition of previously relevant strategies.

**SUMMARY AND CONCLUSIONS**

The findings from a series of experiments indicate that the prelimbic cortex and the orbitofrontal cortex differentially contribute to behavioral flexibility. The prelimbic area supports behavioral flexibility when conditions require a shift in strategies or rules. This is observed in conditions in which an individual must inhibit responding based on one stimulus dimension and instead respond based on a different dimension. Under these conditions, the prelimbic area facilitates the ability to initially shift away from a previously relevant strategy, but once a new strategy is selected the prelimbic area is not critical for the reliable execution of that strategy. The prelimbic area is also involved in adaptive
FIGURE 6. (A) Mean (± s.e.m.) trials to criterion in two-choice egocentric response acquisition and reversal learning following bilateral infusions of saline or bupivacaine into the dorsomedial striatum. Dorsomedial striatal inactivation did not affect acquisition but impaired reversal learning. The treatment received in each phase of testing is in bold. SAL = saline; BUP = bupivacaine. *P < 0.05 versus controls. (B) Mean (± s.e.m.) number of perseverative and regressive errors in two-choice egocentric response reversal learning after bilateral infusions of saline or bupivacaine into the dorsomedial striatum. There was a significant increase in regressive, but not perseverative, errors following dorsomedial striatal inactivation. SAL = saline; BUP = bupivacaine. *P < 0.05 versus controls.

responses when more abstract rules must be used in a context, e.g., switching between match-to-sample and nonmatch-to-sample rule. Furthermore, the pre-limbic area supports learning when different stimuli must be integrated and the relationship between different stimuli learned as in paired-associate tasks. In contrast, the pre-limbic area does not appear critical for flexibly adapting when
conditions require a reversal in choice patterns or exemplars. Taken together, the pattern of findings suggests that the prelimbic area supports a higher-order process for flexibly adapting to a change in environmental conditions.

Several studies indicate that the orbitofrontal cortex supports learning when the general strategy remains the same but a shift in specific choices is required. This is commonly observed in reversal-learning tasks. Comparable to the prelimbic area, the orbitofrontal cortex supports a process that enables the ability to initially shift away from a previously relevant choice. However, as the level of difficulty is increased with more stimuli to discriminate among, the orbitofrontal cortex may support multiple processes for facilitating a switch in choice patterns. One study examined the effects of rat orbitofrontal cortex lesions on an extradimensional shift and revealed that there was no deficit. These results suggest that the orbitofrontal cortex and the prelimbic cortex support different behavioral operations to enable behavioral flexibility. However, future studies are clearly needed to better understand how the orbitofrontal cortex contributes to behavioral flexibility. One issue is that within the orbitofrontal cortex there are different subregions. It is unknown whether the separate orbitofrontal subregions differentially contribute to behavioral flexibility. Related to this point, it should be noted that at the rostral pole in the ventromedial frontal cortex the rat contains a medial orbital subregion that makes up part of the larger orbitofrontal cortex. There is a paucity of experiments that have investigated the medial orbital region.

There is some evidence that the infralimbic subregion contributes to behavioral flexibility. A role for the infralimbic region in behavioral flexibility has come from studies that have demonstrated that infralimbic lesions impair reversal learning but do not impair the initial acquisition of a discrimination. One study further demonstrated that the reversal-learning impairment was not due to perseverance of the previously relevant strategy, but once a new choice pattern was selected infralimbic lesions prevented the maintenance of that new strategy. This contrasts with the prelimbic and orbitofrontal regions, which are important for the initial inhibition of a previously relevant strategy. Although there is some evidence that the infralimbic region supports behavioral flexibility, not all studies have demonstrated this. Furthermore, the range of tests to examine infralimbic involvement in behavioral flexibility has been limited. A more comprehensive examination of possible infralimbic contributions to behavioral flexibility is needed. Future studies should utilize a wider range of behavioral paradigms.

The dorsomedial striatum, which receives input from both the orbitofrontal cortex and the prelimbic area, plays a broader role in behavioral flexibility in that this region supports both reversal-learning and extradimensional shifts. In contrast to prefrontal cortex subregions, the dorsomedial striatum does not facilitate the initial shift away from a previous strategy, but it is critical for the maintenance of a new strategy once selected. These results indicate that the dorsomedial striatum, in conjunction with prefrontal cortex subregions,
can enable behavioral flexibility by supporting distinct but complementary functions. Thus, these areas together are part of a larger neural system that allows one to adapt successfully in an ever-changing environment.

REFERENCES