Supporting Information

Boschin et al. 10.1073/pnas.1419649112

SI Experimental Procedures

Animals. For all animals, water was always available ad libitum in the home enclosure; each monkey's daily food ration was delivered in the test box at the end of each behavioral session and was supplemented with fruit in the home enclosure. All animals were provided with environmental enrichment and, where possible, animals were housed in social groups.

The set of seven animals was divided into two subgroups (four animals with FPC lesions and three intact control animals) and, where possible, analyses were conducted on group differences on sensitive within-subject differences (e.g., preoperative vs. postoperative performance). Many previous studies have verified that these sizes of groups provide sufficient power to show statistically significant performance differences between groups in identical tasks or tasks very similar to those tasks we have used. In the interests of "reduction," we used no more animals than are required to determine if similar behavioral differences can be detected after FPC lesions. Some examples include concurrent objects-in-scenes learning (1), successive single problem learning (2), concurrent discrimination learning for objects (3), and DMS (4).

Surgery. For the FPC lesions, a bone flap was raised over the right and left anterior prefrontal cortex, the dura mater was cut and reflected, and the craniotomy was extended anteriorly by rongeurs to provide access to make the lesion. All cortex anterior to the limit of the lesion on the dorsal, medial, and orbital surfaces was removed by aspiration using a small-gauge metal aspirator. White matter was spared whenever possible, except in the most rostral part of the lesion. When the lesions had been completed, the dura mater was sewn back, the bone flap was replaced, and the wound was closed in layers. The operated animals rested for 2 wk after surgery before beginning postoperative testing. Unoperated control animals rested for the same period between preoperative and postoperative testing.

The operations were performed in sterile conditions with the aid of an operating microscope. The monkeys were first sedated with both ketamine (10 mg/kg) and xylazine (0.25–0.5 mg/kg) given i.m., intubated, and then artificially respirated and anesthetized throughout surgery with isoflurane (1.0–2.0% to effect). In general, steroids (20 mg/kg of methylprednisolone) were given i.m. the night before surgery, and three doses were given 4–6 h apart (i.v. or i.m.) on the day of surgery to protect against intraoperative edema and postoperative inflammation. The monkeys were given atropine (0.05 mg/kg) to reduce secretions, an antibiotic (8.75 mg/kg of amoxicillin) for prophylaxis of infection, an opioid [0.01 mg/kg of buprenorphine given i.v., repeated twice (i.v. or i.m.) at 4- to 6-h intervals on the day of surgery], a nonsteroidal anti-inflammatory (0.2 mg/kg of meloxicam given i.v.) agent for analgesia, and an H2 receptor antagonist (1 mg/kg of ranitidine given i.v.) to protect against gastric ulceration as a side effect of the combination of steroid and nonsteroidal anti-inflammatory treatment. Heart rate, oxygen saturation of hemoglobin, mean arterial blood pressure, end-tidal CO_2, body temperature, and respiration rate were monitored continuously throughout surgery.

This study is, to our knowledge, the first instance of selective FPC lesions in any primate species. In our laboratory, we are very experienced in the different advantages and disadvantages of the aspiration vs. neurotoxic lesion approach, which varies depending on the location of the target structure. The FPC is a particularly demanding target to lesion selectively due to its very anterior location deep below the brow of the macaque, wherein it is not particularly amenable to the neurotoxic injection approach in the absence of proximal stereotaxic or visible landmarks. A new approach was developed to access this region, and the neurosurgeon deemed it more likely, on balance, to achieve a complete lesion by the aspiration method. This choice naturally raised the concern that an aspiration lesion in the frontal pole area might have damaged the underlying white matter bundles/fascicles, particularly in light of a recent study by Rudebeck et al. (5), which concluded that aspiration lesions in orbitofrontal cortex (OFC) could possibly damage the underlying white matter fascicles. Significant practical disadvantages of the neurotoxic lesion method for the OFC have previously been discussed (6). However, unlike under the OFC, there is no such white matter fascicle or bundle that passes below area 10 that distributes fibers to other cortical areas for the obvious anatomical reason that all of the white matter underlying area 10 at the frontal pole cortex comprises afferent and efferent connections between area 10 and other cortical regions; therefore, damage to these connections of area 10 is within the experimental goal of this study to disable the function of the frontal pole cortex. We have carefully examined the literature to see whether there is any evidence of monoaminergic (dopaminergic or noradrenergic fibers) passing under area 10 in macaque monkeys. One study in monkeys (7) reports that a lesion in a frontal area led to changes in monoaminergic innervations of the posterior cortex. However, the lesion in this study was clearly far from the frontal pole cortex (area 10) because figure 1 of ref. 7 shows that the frontal lesion is at the level of where the caudate, putamen, and lateral ventricle can be clearly seen. Therefore, there is no evidence in primate species to show that monoaminergic pathways pass under area 10 at the frontal pole cortex. We conclude that the FPC lesion approach is entirely appropriate because it would not be possible to obtain similarly complete lesions via the neurotoxic approach.

Histology. At the conclusion of the experiments, the FPC-lesioned animals were deeply anesthetized and then perfused through the heart with saline, followed by formol-saline solution. Their brains were blocked in the stereotaxic plane, removed from the skull, put in sucrose-formalin solution until the block sank, and subsequently cut in 50-μm horizontal sections on a freezing microtome. Every fifth or tenth section was retained and stained with cresyl violet.

Apparatus. The subject sat, unrestrained, in a wheeled transport cage fixed in position in front of a touch-sensitive screen on which the stimuli were displayed. An IR camera monitored the general status of the monkey, especially pellet uptake and consumption. A computer, with a millisecond accuracy timer-card to record reaction times, controlled the experiment and conducted data acquisition. The apparatus and hardware were as similar as possible between laboratories; the automated task control software was always identical.

Behavioral Tasks.

Experiment 1 (concurrent objects-in-scenes learning). The scene-generating algorithm was based on a random number generator; each scene was unique in that the scenes varied in several randomly selected attributes, including the background color of the screen; the location of ellipses on the screen; the color, size, and orientation of ellipse segments; and the large typographic character

that was a component part of the background in every scene and was clearly distinct in size from the foreground (i.e., S^+, S^-) objects, as well as the color of the typographic character. All colors used were assigned with the constraint that the foreground objects should be visible (i.e., there was larger than a certain separation in color space between the colors of a foreground object and the color of any element of its local background). Because these scenes were randomly generated, an infinite number of unique scenes could be presented and 20 new ones were used each day in the full task. Before working on the full task and gathering experimental data for analysis, animals were trained to perform the task in stages.

First, each monkey learned (with reinforcement) to touch single foreground objects against a black background. Then, additional scene elements were introduced until the monkey reliably identified and accurately touched a single foreground presented within a full new scene. Second, problems were then introduced in which there were two foreground objects (one correct and one incorrect, as described above); the number of scenes given in each session was gradually increased at a rate that was based on each monkey's performance. Initially, with few scenes (e.g., three unique scenes per day), there were many repetitions of each scene per session. As the number of unique scenes per day increased, the number of repetitions decreased until within-session learning in the final version of the task involved 20 new scenes in each session that were repeated eight times each (always in the same order).

When the monkey completed the final trial of a session, the lunchbox opened automatically; the monkey received the large food reward and was allowed ~10 min to eat some of the food and take the remainder into its cheek pouches before being returned to the home enclosure. If the final trial was incorrect, a correction trial was given so that the monkey only ever received the large food reward after a correct response. The dependent measure was the number of errors (initial touches of the incorrect foreground object) in each repetition block of the 20 scenes. Training continued until performance was stable at 20 scenes per day, after which we tested the animals on 15 more consecutive sessions, which constituted the preoperative data. After approximately 2 wk, animals were tested again (during which time those animals assigned to FPC lesion group received their lesion and then rested ~2 wk, whereas those animals assigned to CON group rested for an equivalent time) for 15 more consecutive sessions, which constituted the postoperative data. Preoperative and postoperative percent error data were arcsin-transformed to approximate normality before analyses better.

Experiment 2 (successive single problem learning). The stimuli used in the successive single problem learning task were individual clipart images obtained from commercially available internet sources. Each clipart image was 128 × 128 pixels in size and comprised a unique foreground colored image imposed on a dark gray-colored, screen-congruent background. A very large pool of several thousand unique images was used for this task, and stimuli were used from the pool in random order without replacement, ensuring that the stimuli used in this task were trial-unique. A touch-sensitive area matching the size of each image was used to ensure the detection of stimulus-directed responses on the touch-screen.

Experiment 3 (concurrent discrimination learning for objects). The stimuli for the concurrent discrimination learning task were the same kinds of stimuli described in the successive single problem learning task but without any overlap in the identity of stimuli used between these two tasks.

As described in *Experimental Procedures*, making an incorrect choice made the subject wait longer than usual for the opportunity to obtain the next reward pellet and, consequently, the lunchbox. Therefore, there was an incentive to make correct choices and also an incentive not merely to choose randomly at each trial, because this type of behavior would increase the time before the lunchbox opened. After all 10 pairs in the set had been presented and the response to the last S^+ had been made, the lunchbox immediately opened and the food within became available. The subjects were left for 10 min to eat out of the lunchbox.

Experiment 4 (DMS) and experiment 5 (DNMS) task acquisition, performance tests, and subsequent alternations. The visual stimuli used in this task were also clipart images similar to, but not overlapping with, those clipart images used for the successive single problem learning task or the concurrent object discrimination learning task described earlier. A very large pool of several thousand unique images was used, stimuli were used from the pool in random order without replacement within each daily test session until all were used up, and the set was then reused (hence, it would be several weeks before an animal saw any given image again, making stimuli close to trial-unique).

When we repeated the testing, reversing the rule (until the animals had experienced the following sequence of rule reversals: DNMS, DMS, DNMS, DMS), we found that one FPC-lesioned animal completed only two of the four reversals but performed these reversals as efficiently as the other animals; the mean errors to criterion show no performance difference between groups (Fig. 4*C*). Thus, the FPC group had no difficulty compared with the CON group in postoperative learning and switching between new abstract rules.

Experiment 6 [learning new abstract rule ("smaller than")] and experiment 7 [combining two rules ("smaller than" and "same as". The animals were trained on a series of seven different training stages. During stage 1, a randomly selected piece of clipart, the sample item, was presented in the center of the screen. Once the subject touched the sample on the screen, a delay of 2 s began. After the delay, the sample was removed and two test items appeared on the screen: one "match" item identical to the sample item and one novel "nonmatch" item (both were of the same size as the sample in this and previous experiments, which we will refer to as "large"). These items were presented randomly in two of the four corners of the screen. The subject was rewarded with a pellet for selecting the match item, therefore being reinforced for applying the standard DMS ("same as") rule. Once a correct response was performed, 8 s elapsed before the following trial. If an incorrect response was performed, no reward was provided and a 16-s ITI with a blank screen was introduced before the following trial. Stage 2 was like stage 1, but once the sample response was executed, the sample item remained on the screen when the test items appeared, making the task a simultaneous, as opposed to DMS, paradigm. Stage 3 also followed the same procedure as the previous stage, with the addition of an extra nonmatch item to one of the other possible corner locations, making the task three-choice as opposed to two-choice. Then, in stage 4, an additional nonmatch item was added, bringing the total number of possible choices up to four. In stage 5, the four possible choices included two match items in two different sizes (one large, and hence the same size as the sample item, and one smaller than the sample item), as well as two nonmatch items that were both large, and hence the same size as the sample item. The subject was still rewarded for selecting the same size, match item. Stages 1–5 were each carried out by the subject over several consecutive days until an accuracy criterion of 80% or more was achieved, which triggered progression to the next stage on the next day.

Stage 6 was the crucial stage for analysis because we switched the rule in this stage and the subject was now required to learn to apply the smaller than rule for the first time. The four possible choices still included two match items, one of the same size and one smaller than the sample item, and two nonmatch items, both of the same size as the sample item. The subject was rewarded for picking the smallest item of the four. During this stage, the subject carried out 100 trials a day over 3 consecutive days.

Stage 7 involved the introduction of correction trials. In this stage, when the subject made more than five errors in a row, the computer did not present the big-match item on the next trial so as to encourage the acquisition of the new smaller than rule. This stage was carried out over several consecutive days until an accuracy criterion of 90% or more was reached. There was no difference between groups in their ability to attain the stage 7 criterion (the CON group made a mean of 34 errors to criterion and the FPC group also made a mean of 34 errors to criterion). The subjects were then moved on to the final stage, which combined the two aforementioned rules.

In stage 8, one item was identical in both size and identity to the sample item ("big match"), one item shared the same identity as the sample item but was smaller in size ("small match"), one item was unrelated in identity to the sample item but of the same size ("big nonmatch"), and one item was unrelated in identity to the sample but related in identity to the big nonmatch item and was smaller in size ("small nonmatch"). Therefore, the subject needed to integrate the two rules, "same as" and "smaller than", to select the correct item because simply selecting the smaller item would cause the animal to perform at chance (two of four items were small). A schematic of a typical trial is provided in Fig. 5*A*. Successful performance on the task was defined as an accuracy criterion of 80% or more correct trials over the course of one experimental session. As with all other tasks, the automated lunchbox opened after completion of the daily session.

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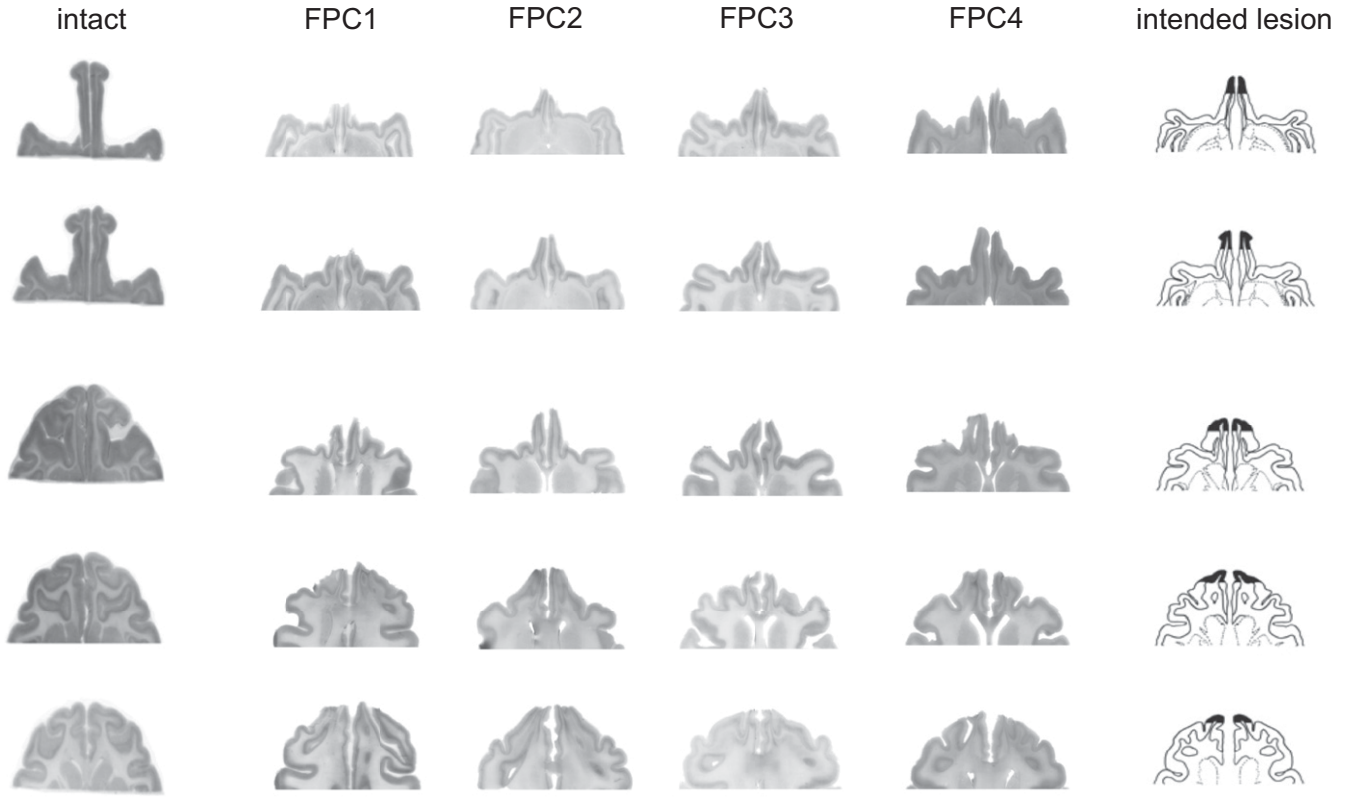

Fig. S1. Related to *Experimental Procedures*. Intended extent of the FPC lesion and histological verification. The intended lesion extent is shown on the right in a series of drawings of horizontal sections. The posterior extent of the lesions was a vertical line 2 mm posterior to the rostral tip of the principal sulcus, and all gray matter tissue anterior to this line on the dorsal, medial, and orbital surfaces was removed. The histological sections on the left (one column per FPC-lesioned animal and one intact animal) show the actual extent of the lesions, as evident from the horizontal stained sections (compare with the drawings of horizontal sections).

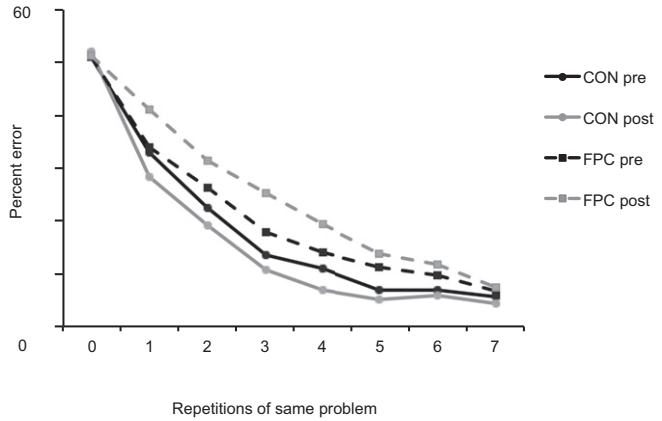

Fig. S2. Related to *Experimental Procedures*. Comparison of preoperative (pre) and postoperative (post) learning rates for concurrent objects-in-scenes learning. Mean preoperative and postoperative error rates for the CON group and FPC-lesioned group, averaged across all within-session repetitions of concurrently presented objects-in-scenes problems, are shown. The repetition corresponding to zero is the first presentation of a problem to which the animals necessarily have to guess (hence, averaging around 50%).

Table S1. Summary of behavioral effects of FPC lesions across seven different behavioral tasks in terms of their resulting in impairment or unimpairment of behavioral performance; in each case, the nature of the cognitive process probed by the task and the interpretation of each result in light of our model are discussed in summary form

Task	Lesion-induced behavioral change	Intact cognitive processes of note	Why the result is consistent with the hypothesis that the FPC is necessary for exploring the relative value of unfamiliar alternatives
Tasks affected by the FPC lesion			
Objects-in-scenes learning	One-trial learning impaired	Slow gradual reinforcement-based learning	Deficit selective to the most rapid phase of learning; this stage is also the stage in which animals stand to benefit most from assigning relative value on the basis of feedback to both chosen and unchosen stimuli
Successive single problem learning	One-trial learning impaired	Slow gradual reinforcement-based learning; prospective encoding of next choice across delay	Deficit selective to the most rapid phase of learning; this stage is also the stage in which animals stand to benefit most from assigning relative value on the basis of feedback to both chosen and unchosen stimuli
Learning new abstract rule	FPC-lesioned animals fail to select the S^+ ("smaller than") item often in first 3 d		Slowed learning of the "smaller than" rule in the FPC group attributed to less exploration of the relative value of the newly emerging alternative rule
Tasks unaffected by the FPC lesion			
Concurrent object discrimination learning	None	Slow gradual reinforcement-based learning	No impairment expected because this task is slowly acquired, without a rapid learning stage during which animals with an intact FPC would benefit by being able to assign relative value on the basis of feedback to both chosen and unchosen stimuli
DMS	None	Working memory for stimulus	No impairment expected because there are no alternatives to value in the sample phase
DNMS	None	Working memory for stimulus	No impairment expected because there are no alternatives to value in the sample phase
Applying two rules at the same time	None	Applying two abstract rules at the same time; combining the result of multiple cognitive operations	FPC does not influence choosing between abstract rules once they are highly familiar, even in combination