Website publication 11 March 1998

NeuroReport 9, 865–870 (1998)

Thalamic deactivation during early implicit sequence learning: a functional MRI study

Scott L. Rauch,[1,2,3,CA] Paul J. Whalen,[1,2,3] Tim Curran,[4] Sean McInerney,[1] Stephan Heckers[1,3] and Cary R. Savage[1,2,3]

[1]Psychiatric Neuroscience Program and [2]NMR Center, Mass General Hospital East, 9th Floor, Bldg 149, Thirteenth Street, Charlestown, MA 02129; [3]Department of Psychiatry, Havard Medical School, Boston, MA; [4]Department of Psychology, Case Western Reserve University, Cleveland, OH, USA

[1,CA]Corresponding Author and Address

PREVIOUS research has implicated the striatum in implicit sequence learning. However, imaging findings have been inconsistent with regard to activity within the thalamus during performance of such tasks. Contemporary models of cortico-striato-thalamic circuitry suggest opposing influences on thalamic activity; suppression of thalamic activity is mediated by the *indirect* pathway and enhancement is mediated by the *direct* pathway. Using functional magnetic resonance imaging, we studied activity within human thalamus during early and late phases of an implicit sequence learning task known to reliably recruit the striatum. Significant deactivation (decreased signal relative to a baseline condition) was observed within the thalamus during early implicit learning. This finding is consistent with models of cortico-striato-thalamic function and specifically supports a profile of early 'thalamic gating' via the indirect pathway. *NeuroReport* 9: 865–870 © 1998 Rapid Science Ltd.

Key words: Basal ganglia; Memory; Procedural learning; Striatum

Introduction

Over the past decade, it has become widely accepted that the basal ganglia play an important role in cognition and affect as well as sensorimotor functions.[1,2] For example, convergent evidence has accrued to show that cortico-striatal systems support implicit (i.e. non-conscious) forms of learning: studies of patients with known striatal pathology (e.g. Huntington's disease) have demonstrated selective performance deficits on procedural learning tasks,[3,4] and a series of functional imaging studies have shown that healthy subjects reliably recruit the striatum during the performance of implicit sequence learning[5–9] as well as other procedural learning tasks.[10]

Contemporary models of information processing involving the basal ganglia suggest that striatal activity influences thalamic transmission in the service of learning and memory, as well as attention.[11,12] Specifically, it is hypothesized that the *direct* pathway from striatum has a net effect of enhancing thalamic activity (also called amplification), whereas the *indirect* pathway has a net effect of suppressing thalamic activity (also called 'thalamic gating'; see Fig. 1).[1,11–13] These opposing influences are believed to provide a basis for refining corticothalamic output as well as thalamocortical input.[11,12]

In the case of implicit sequence learning, the system portrayed in Fig. 1 mediates the development of a reaction time advantage when subjects are repeatedly exposed to a sequence of visual stimuli that cue a corresponding sequence of motor responses.[6,14] As the sequence is learned, each subsequent element of the sequence can be predicted based on the preceding elements. Thus, the striatum is proposed to operationally predict each subsequent element by enhancing transmission through the correct channel and suppressing transmission through incorrect or extraneous channels within the thalamus. This is consistent with the purported physiology of striatal spiny neurons which exhibit plasticity by developing preferential responses to specific temporospatial cortical input constellations.[11]

Much of what is known regarding the normal structure and function of basal ganglia-thalamocortical system has been gleaned from animal research.[1,2,11,13] Functional imaging techniques provide a means for delineating homologous human physiology *in vivo*. In the current experiments, we employed functional magnetic resonance imaging (fMRI) to measure thalamic activity in normal human subjects performing an implicit sequence learning task[14] that had already been shown to reliably recruit the striatum.[6,9] In this manner, we were

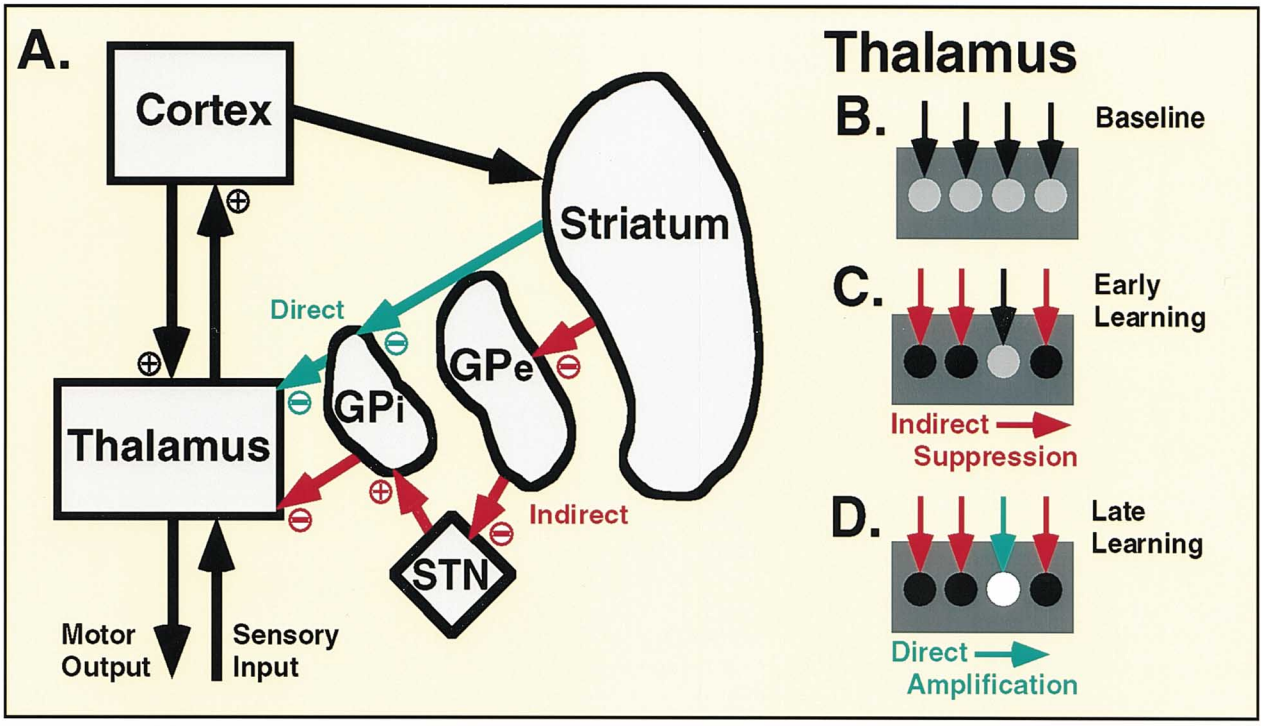

FIG. 1. A schematic diagram illustrating the direct (green arrows) and indirect (red arrows) cortico-striato thalamic pathways (**A**). GPi, globus pallidus interna; GPe, globus pallidus externa; STN, subthalamic nucleus. (–), inhibition; (+), excitation. (**B**–**D**) Heuristic for how these pathways might serve to refine transmission through the thalamus, thereby mediating implicit learning. Four closed circles represent separate transmission channels within the thalamus, corresponding to either the four cue positions (visual input channels) or the four possible finger movement responses (motor output channels). The influence of the direct and indirect pathways on activity within the thalamus is represented on a gray scale, where black = suppressed, white = enhanced, gray = intermediate. During the baseline condition (B) balanced influences of the direct and indirect pathways across all transmission channels produce no bias (gray circles). During early learning (C) predominance of the indirect pathway serves to suppress (or gate) incorrect input or output channels (black circles). During late learning (D) recruitment of the direct pathway serves to enhance input or output via the correct target transmission channel (white circle). This model is consistent with the current findings of decreased thalamic activity during early learning and a trend toward increased thalamic activity during late learning, in the context of striatal recruitment across runs and the development of a significant reaction time advantage.

able to demonstrate relative decreases in thalamic activity, consistent with current models of thalamic gating via basal ganglia-thalamocortical circuitry.[11]

Materials and Methods

Two separate experiments are presented. This investigation was conducted in accordance with the guidelines of the Subcommittee on Human Studies of the Massachusetts General Hospital; all subjects gave written informed consent. All subjects were right-handed as established by the Edinburgh Handedness Inventory,[15] and were without significant psychiatric, neurological or medical illness as established by a brief screening interview. No subjects were taking psychotropic or cardiovascular medications at the time of study or during the preceding 4 weeks, as established by self report.

fMRI-SRT paradigm: The serial reaction time task (SRT) is well established as a paradigm for the study of implicit sequence learning.[14] A version of the SRT adapted for use with fMRI (fMRI-SRT) has also been described previously:[9] it involves presentation of an asterisk, appearing serially in each of four boxes which are arranged horizontally. The stimuli are projected onto a viewing screen within the magnet bore. Subjects are instructed to respond to each visual cue by pressing, as quickly and accurately as possible, the corresponding one of four keys on a keypad, which is positioned horizontally on the subject's torso, centered beneath the viewing screen. The task is performed bimanually, using the first two fingers of each hand; each finger corresponds to one of the four keys. For each trial, asterisks are programmed to appear in one of the four boxes for a fixed duration (1.0 s), followed by a fixed period with no asterisk projected (0.2 s), followed by reappearance of the asterisk in one of the three other boxes, and so on. Reaction time (RT) and the accuracy of response are measured for each presentation. In the implicit learning conditions, unbeknownst to the subjects, the stimuli follow a 12-item sequence (position: 1-2-1-4-2-3-4-1-3-2-4-3) that repeats six times for a total of 72 trials. The baseline conditions include 24 stimuli for which stimulus locations are pseudo-

randomly determined, with the constraint that no location is immediately repeated. Each subject performs an initial practice run (2 min of pseudo-random stimuli) followed by two experimental runs of ~6 min and 15 s each. Each experimental run consists of contiguous alternating baseline (B) and implicit learning (IL) conditions arranged in the following order: B-IL-B-IL-B-IL-B. Since the subjects are not informed that a sequence is present, and there is no demarcation between B and IL epochs, each run is experienced by the subjects as a seamless entity. The successive runs are separated by rest periods of ~5 min duration.

Immediately following the scanning session, subjects complete a debriefing procedure which is administered in an automated fashion via computer (as previously described).[6] Subjects are informed that a sequence had been present and are asked to attempt to recall the sequence by making a series of 15 key presses. The recall task is scored based on the longest consecutive string of correct responses. Each subject's performance on the recall task is then compared with a chance distribution as an index of significant explicit knowledge.

Imaging protocol: Each subject spent a total of 75 – 90 min within the scanner. Images were obtained with a quadrature head-coil and a 1.5T MR scanner (General Electric, Milwaukee WI) modified for echo-planar imaging (Advanced NMR Systems, Wilmington, MA) according to the following protocol: (1) a sagittal localizer scan was performed to orient 15 contiguous, 8 mm transaxial slices parallel to the intercommissural plane; (2) an automated shimming technique was used to minimize B_0 inhomogeneity;[16] (3) a spoiled gradient recall (SPGR) T1-weighted flow-compensated scan (resolution $1.6 \times 1.6 \times 8$ mm) was obtained for use as an angiogram; (4) a T1-weighted echo-planar spin echo sequence was used to obtain high resolution structural images; (5) an asymmetric spin echo T2*-weighted sequence (TR = 2750 ms; TE = 70 ms; refocusing pulse offset by –25 ms) was used to obtain functional images (i.e. reflecting local blood oxygenation-level dependent [BOLD] signal intensity).[17] Data analysis entailed movement correction, coronal reslicing, Talairach transformation,[18] and construction of statistical non-parametric maps using the Kolmogorov–Smirnov (KS) statistic. For each fMRI run, 136 time points were acquired: 40 corresponded with the baseline condition, 90 corresponded with the IL condition, and six were excluded as transitional points overlapping consecutive epochs. KS maps contrasting the IL *vs* B conditions were generated for each run, concatenated across the entire cohort of subjects.

Experiment 1 entailed the re-analysis of our previously published fMRI-SRT study,[9] in order to generate testable hypotheses regarding the temporal pattern of thalamic activity associated with implicit sequence learning, in the context of documented striatal recruitment. The study sample comprised 10 males (20–35 years of age). Separately for run 1 (early learning) and run 2 (late learning), the volume of the thalamus was systematically inspected for foci of activation or deactivation, with respect to the IL *vs* baseline contrast. A liberal statistical threshold was employed ($p < 0.005$ uncorrected), since this was a hypothesis generating exercise.

Experiment 2 utilized the fMRI-MultiSRT,[19] a statistically more powerful version of the above paradigm, to test the hypotheses generated by experiment 1. The fMRI-MultiSRT entailed four runs, rather than two: runs 1 and 2 involved learning one sequence and runs 3 and 4 involved learning an entirely different sequence. In this way, greater statistical power could be garnered by effectively producing replicates of the early and late learning conditions. Thus, runs 1 and 3 were averaged to reflect early learning; runs 2 and 4 were averaged to reflect late learning. The two sequences employed were sequence A (from the fMRI-SRT) and sequence B (positions = 3-4-2-1-2-4-1-4-3-2-3-1); the order of the sequences used was counterbalanced across subjects (A-A-B-B or B-B-A-A). The sample comprised 10 males (19–36 years of age). Based on the findings from experiment 1, we sought to test the hypothesis that early implicit sequence learning was characterized by thalamic deactivation (decreased BOLD signal for the IL *vs* B contrast). The volume of the thalamus was systematically inspected for loci of deactivation (as well as activation) during early learning (as well as late learning). A stringent statistical threshold was employed ($p < 5.0 \times 10^{-4}$, uncorrected; corresponding to $p < 0.05$ with Bonferroni correction for multiple comparisons, based on the number of voxels within the thalamic search volume).

Results

Experiment 1: Error rates were < 10% for every subject. The group exhibited a significant RT advantage for the IL *vs* B condition (mean ± s.d. median RT = 403.9 ± 50.9 ms *vs* 437.9 ± 64.1 ms; $t = 5.89$, $df = 9$; $p < 0.0002$). The results of the debriefing task, which assessed subjects' ability to explicitly recall the sequence,[6] demonstrate that they did not perform significantly better than chance; both across the entire cohort (mean maximum consecutive correct responses 3.10 ± 1.19; chance performance

3.71 ± 1.26) as well as in each individual case (all ⩽ 4; individual scores of > 6 are suggestive of significant explicit knowledge).

As reported previously,[9] foci of significant striatal activation associated with the concatenated group IL *vs* B contrast were found within right caudate and an inferior region of the right putamen (uncorrected $p = 3.2 \times 10^{-4}$). Regarding the current analysis, for the IL *vs* B contrast, early learning (run 1) was associated with decreased BOLD signal within the thalamus bilaterally, whereas late learning (run 2) was associated with increased BOLD signal within the left thalamus (Table 1; Fig. 2).

Experiment 2: The results of the debriefing task indicated that one subject exhibited significant explicit knowledge of the sequence (explicit recall score = 7). Therefore, all subsequent analyses for experiment 2 were performed using the nine subjects who did not demonstrate explicit knowledge. Error rates were < 10% for each of the subjects. The group again exhibited a significant RT advantage for the IL *vs* B condition (t = 8.4, df = 8, $p = 0.0001$) indicating that implicit learning had occurred.

The finding of significant right striatal activation was replicated in the concatenated group IL *vs* B contrast. Regarding the current analysis, for the IL *vs* B contrast, early learning (runs 1 and 3) was associated with significant BOLD signal decreases within the thalamus bilaterally; late learning (runs 2 and 4) was associated with no significant change in BOLD signal (although the greatest trend present for this contrast was in the direction of BOLD signal increases ($p = 2.0 \times 10^{-3}$; Table 1, Figure 2).

Table 1. Thalamic foci of fMRI BOLD signal change for the implicit learning *vs* baseline contrast.

Thalamic foci of fMRI signal change	*p* value	Coordinates* x	y	z
Experiment 1 (fMRI-SRT)				
Early learning (run 1)				
Right thalamus (decrease)	3.4×10^{-4}	21	–30	6
Left thalamus (decrease)	1.0×10^{-3}	–6	–21	6
Late learning (run 2)				
Right thalamus (increase)	2.2×10^{-4}	–15	–30	3
Experiment 2 (fMRI-MultiSRT)				
Early learning (runs 1 and 3)				
Right thalamus (decrease)	2.4×10^{-5}	18	–27	6
Left thalamus (decrease)	3.7×10^{-5}	–15	–21	9
Late learning (runs 2 and 4)				
None				

*Coordinates are presented according to the convention of Talairach and Tournoux[18], in ml; the origin is the anterior commissure at the midsagittal plane, with x > 0 corresponding to right of midsagittal, y = 0 corresponding to anterior, and z > 0 corresponding to superior.

Discussion

The current findings demonstrate reliable bilateral BOLD signal decreases in human thalamus during the initial phase of an implicit sequence learning paradigm which is known to recruit striatum. Thalamic deactivation, in the face of striatal activation, is in keeping with contemporary models of basal ganglia-thalamocortical information processing.[11] One interpretation is that these findings represent an illustration of thalamic gating via the indirect striato-thalamic pathway during early implicit sequence learning in humans.

The observed thalamic deactivation was restricted to an early phase of implicit sequence learning: during late learning, there was no evidence of thalamic deactivation, but rather significant BOLD signal increases (experiment 1) or trends toward increases (experiment 2). This is consistent with results from previous studies which have shown significant thalamic activation when initial epochs of implicit sequence learning were excluded from the analysis,[6] monotonic increases in thalamic activity over successive trials of various implicit learning tasks,[10,20] and no significant thalamic activation when data were integrated over early and later stages of implicit learning together.[7–9] This suggests that, in the context of the SRT, early learning is mediated predominantly by thalamic suppression (presumably via the indirect pathway), whereas late learning may involve enhancement or amplification of thalamic activity (presumably via the direct pathway, see Fig. 1).

Other possible interpretations must also be entertained. It is possible that the decreased BOLD signal in the thalamus reflects decreased synaptic activity involving pallido-thalamic inputs that are primarily inhibitory. In other words, apparent thalamic deactivation (as measured by fMRl) could actually represent disinhibition, rather than decreased activity of the neurons intrinsic to the thalamus. It is also possible that the observed decreased BOLD signal within the thalamus reflects changes that are not attributable to influences from the cortico-pallido-thalamic pathway, but rather direct projections from other cortical territories (such as visual cortex). This seems unlikely, however, because no cortical regions exhibited a parallel pattern of activity (i.e, deactivation during early learning). Nonetheless, although the initial interpretation of these findings, focusing on the role of basal ganglia-thalamocortical systems and emphasizing the concept of thalamic gating, is appealing heuristically, additional studies will be necessary to confirm it.

The designations of 'early' and 'late' learning reflect the time scale of a paradigm that was completed during a single visit to the imaging suite.

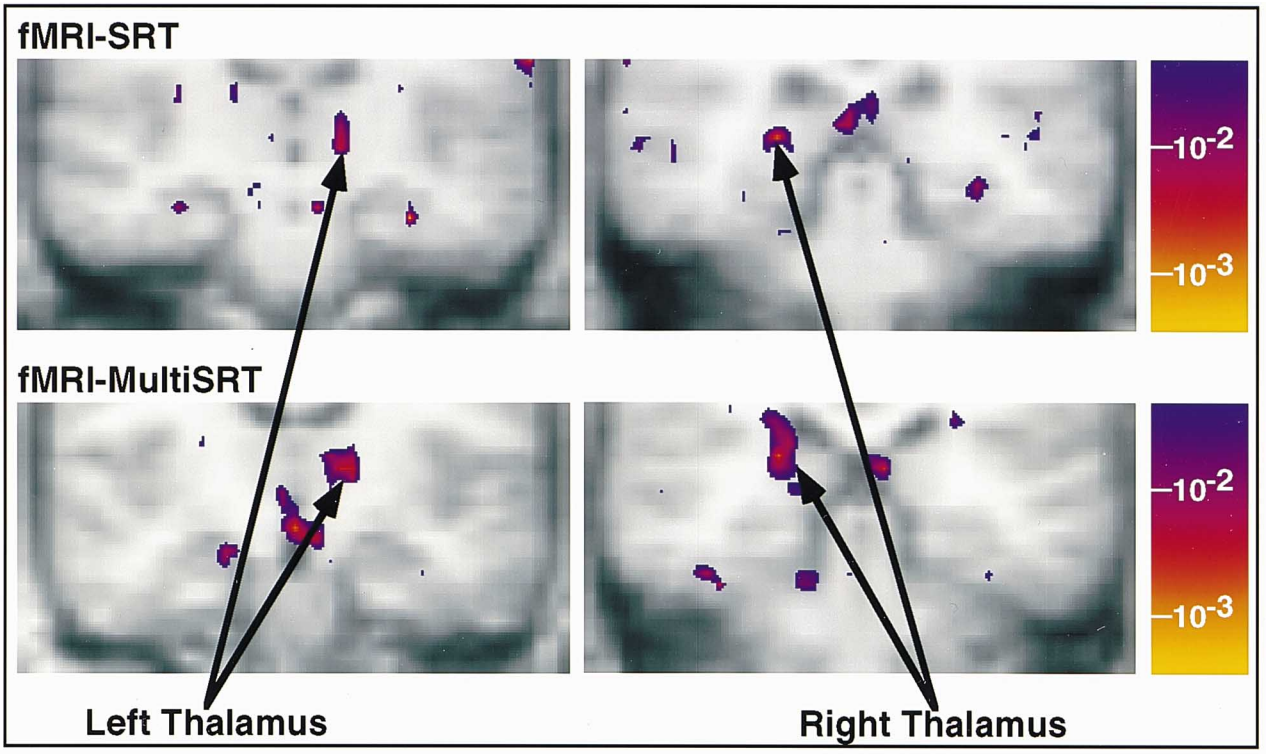

FIG. 2. KS-maps illustrating significant bilateral thalamic deactivation during early learning runs, based on the concatenated IL *vs* B contrast for the entire cohort. The upper panel depicts the results from experiment 1; the lower panel depicts the results from experiment 2. The KS statistical color maps are superimposed over T1-weighted high resolution coronal images that have been averaged for all subjects in each group. Functional and structural data have been Talairach transformed.[18]

It is important to appreciate that other investigators have sought to characterize functional adaptions that occur during sequence learning trials spread over multiple scanning sessions across days to weeks.[21]

Limitations of the current study include the temporal and spatial resolution limits of the techniques employed. The subtle nature of the behavioural effect (implicit learning), together with the contingencies of assessing and preventing explicit contamination, presented a substantial challenge in terms of generating sufficient statistical power. In the current study, these factors necessitated averaging over time and across subjects.

Although our spatial resolution was ample for localizing activation foci within the thalamus, resolution of the specific thalamic nuclei cannot be claimed with confidence. Still, it is noteworthy that the peak loci of thalamic deactivation map to the right pulvinar, left mediodorsal nucleus, and left ventral tier nuclei (based on the Talairach atlas).[18] Foremost, this speaks to the reproducibility of the pulvinar finding across two independent data sets. Moreover, it suggests that the thalamic nuclei involved include those associated with frontal (mediodorsal nucleus), motor (ventral tier nuclei), and parieto-occipital (pulvinar) cortical domains. This is consistent with the nature of the SRT, which entails the learning and integration of sequential visuospatial cues together with corresponding sequential motor responses.

The motivation for this line of inquiry included our interest in developing functional imaging tools for studying neuropsychiatric disorders. Current neurobiological models of several diseases, including major depression, obsessive compulsive disorder and Tourette syndrome, posit dysfunctional cortico-striato-thalamic systems.[22,23] A recent PET-SRT study showed that patients with obsessive compulsive disorder failed to normally recruit striatum, and exhibited aberrant activation within medial temporal regions instead.[24] The fMRI-SRT and fMRI-MultiSRT paradigms should enable replication and extension of those results in various patient groups. Moreover, the current findings with regard to thalamic deactivation suggest a candidate probe for testing hypotheses pertaining to abnormalities involving striato-pallido-thalamic transmission, and specifically dysfunctional thalamic gating.

Conclusion

The current project utilized fMRI to study *in vivo* human thalamic activity during an implicit sequence learning task. The findings demonstrate thalamic

deactivation during an early phase of implicit learning. The results are interpreted as illustrating thalamic gating mediated by the indirect striato-pallido-thalamic pathway. Future research will be necessary to replicate, clarify, and expand upon these observations. In particular, application of this paradigm to the study of basal ganglia disorders may prove useful.

References

1. Alexander GE, Crutcher MD and Delong MR. *Prog Brain Res* **85**, 119–146 (1990).
2. Wise SP, Murray EA, Gerfen CR. *Crit Rev Neurobiol* **10**, 317–356 (1996).
3. Knopman D and Nissen MJ. *Neuropsychologia* **29**, 245–254 (1991).
4. Willingham DB and Koroshetz WJ. *Psychobiology* **21**, 173–182 (1993).
5. Grafton ST, Hazeltine E, Ivry R. *J Cog Neurosci* **7**, 497–510 (1995).
6. Rauch SL, Savage CR, Brown HD *et al. Human Brain Mapp* **3**, 271–286 (1995).
7. Doyon J, Owen AM, Petrides M *et al. Eur J Neurosci* **8**, 637–648 (1996).
8. Berns GS, Cohen JD, Mintun MA. *Science* **276**, 1272–1275 (1997).
9. Rauch SL, Whalen PJ, Savage CR *et al. Human Brain Mapp* **5**, 124–132 (1997).
10. Grafton ST, Mazziotta JC, Presty S *et al. J Neurosci* **12**, 2542–2548 (1992).
11. Houk JC. Davis JL and Beiser DG, eds. *Models of Information Processing in the Basal Ganglia*. Cambridge: MIT Press, 1995.
12. Jackson S and Houghton G. Sensorimotor selection and the basal ganglia: a neural network model. In: Houk JC, Davis JL and Beiser DG, eds. *Models of Information Processing in the Basal Ganglia*. Cambridge: MIT Press, 1995: 337–367.
13. Graybiel AM. *Curr Opin Neurobiol* **5**, 733–741 (1995).
14. Nissen MJ and Bullemer P. *Cogn Psychol* **19**, 1–32 (1987).
15. Oldfield RC. *Neuropsychologia* **9**, 97–113 (1971).
16. Reese TG, Davis TL and Weisskoff RM. *J Magn Reson Imaging* **5**, 739–745 (1995).
17. Kwong KK, Belliveau JW, Chesler DA *et al. Proc Natl Acad Sci USA* **89**, 5675–5679 (1992).
18. Talairach J and Tournoux P. *Co-Planar Stereotaxic Atlas of the Human Brain*. New York: Thieme, 1988.
19. Rauch SL, Whalen PJ, Savage CR *et al. Neuroimage* **5**, S600 (1997).
20. Hazeltine E, Grafton ST and Ivry R. *Brain* **120**, 123–140 (1997).
21. Karni A, Meyer G, Jezzard P *et al. Nature* **377**, 155–158 (1995).
22. Swerdlow NR and Koob GF. *Behav Brain Sci* **10**, 197–245 (1987).
23. Miguel EC, Rauch SL, Leckman JF, eds. *Neuropsychiatry of the Basal Ganglia. Psychiatric Clinics of North America*. Philadelphia: W.B. Saunders **20**, 691–991 (1997).
24. Rauch SL, Savage CR, Alpert NM *et al. J Neuropsychiatry* **9**, 568–593.

ACKNOWLEDGEMENTS: Support was provided by the Tourette Syndrome Association, Inc., NIMH (grants MH01215 & MH01230). and the David Judah Research Fund. P.W. was supported as a Fellow in the Clinical Research Training Program of Harvard Medical School. We acknowledge Halle Brown, Terry Campbell, Mary Foley, Michael Jenike, Linda Leahy, Michael Lee, Bruce Rosen and Mike Vevea for their assistance.

Received 3 December 1997; accepted 18 January 1998