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Brain Activation during Smooth-Pursuit Eye Movements

Jody Tanabe,* Jason Tregellas,† David Miller,* Randal G. Ross,‡ and Robert Freedman†,‡

Department of Radiology, ‡Department of Psychiatry, and †Department of Pharmacology, University of Colorado Health Sciences Center, Denver, Colorado 80262

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A potential application of studying eye movements with functional MRI (fMRI) is to examine patient populations with known eye movement dysfunction, but the reliability with which normal subjects demonstrate activity in specific brain regions has not been established. To date, fMRI studies of smooth-pursuit eye movements have used relatively small numbers of subjects and have been restricted to fixed-effects analyses. We extend these studies to whole brain imaging at 1.5 T, properly accounting for intersubject variation using random effects analysis. Smooth-pursuit eye movements elicited activation consistently in dorsal cortical eye fields and cerebellum. Subcortical activation was greatly attenuated, but not eliminated, with the random-effects second-level analysis. In addition, session-dependent changes in activation were greater in some regions than others and may indicate areas of brain, such as the supplementary eye fields, that are sensitive to attentional modulation of eye movements.

INTRODUCTION

Smooth-pursuit eye tracking is an oculomotor system that evolved in humans, nonhuman primates, and other carnivores and that enables the continuous maintenance of sharp vision on a moving target, whether the target, animal's head, or both are moving (1, 2). The role of frontal, parietal, and supplementary eye fields in oculomotor control has been studied in great detail using electrophysiological methods in monkeys. At the subcortical level, the lateral geniculate nucleus (LGN), paramedian pons, superior colliculus, and cerebellum are also involved (3). Two types of eye movements are saccades and pursuit, which focus on stationary or moving stimuli, respectively. For pursuit eye tracking, the middle temporal (MT or V5) and middle superior temporal (MST or V5A) cortices are involved in addition to the dorsal cortical eye fields.

In humans, positron emission tomography (PET) studies have shown activation in cortical regions during saccadic (4–6) and smooth-pursuit eye movements (4) that are the same or homologous to those observed in primate brain. More recently, functional MRI (fMRI) methods have been applied to eye movement tasks. Petit *et al.* confirmed activation in frontal, parietal, and supplementary eye fields and occipital temporal cortex during pursuit eye movements (7). Berman *et al.* extended these observations and also demonstrated activation in the cingulate gyrus using a high-field (3 T) MR system (8). There was significant overlap in brain regions serving saccades and pursuit. Nonetheless, because of the high spatial resolution afforded by fMRI methods, both investigators demonstrated subtle differences in localization between saccades and pursuit (9, 10).

Previous studies of fMRI of smooth-pursuit eye movements (SPEM) have been restricted to fixed-effects analyses, however, which fail to account for random variation between subjects. The high sensitivity of the fixed-effects analysis (due to low scan-to-scan compared with high subject-to-subject error variance) limits the inferences that can be made about group behavior (11). One would like to be able to make inferences about SPEM activation in subject groups because SPEM is a trait marker for specific neuropsychiatric diseases. Numerous studies have demonstrated pursuit eye movement abnormalities in schizophrenics and their relatives, for example. SPEM abnormalities have also been found in Alzheimer's disease, bipolar disorders, old age, and posterior cerebral lesions (2), and abnormal saccades have been described in Parkinson's disease (2) and, recently, autism (12). The pursuit response, however, can show considerable intersubject variation and it is likely that the functional brain response is also variable. Therefore, the first aim of this study was to extend previous fMRI studies of SPEM to a larger number of subjects and control for intersubject variance with a random-effects model analysis.

Within the eye movement neural pathways, different activation patterns have been observed depending on whether a stimulus has been predictable or nonpredictable (13) or repeated over multiple sessions (14). These differences are significant because they can point to functional specialization, such as motor, learning, and

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attentional aspects of eye movements. Therefore, the second aim of this study was to evaluate the effect of repeating the smooth-pursuit eye task brain activation. Specifically, we wanted to determine whether neural structures thought to be sensitive to attentional factors modulating eye movements showed a session-dependent change in activation.

METHODS

Subjects. Seventeen subjects participated in this study. Two subjects were excluded because of abnormal smooth-pursuit task performance measured outside the magnet (see Data Analysis). Data are shown for 15 controls (8 female/7 male, ages 23–50 years, mean age 32 years). Subjects provided written, informed consent approved by the human Colorado Multiple Institutional Review Board.

Magnetic resonance parameters. All studies were performed on a 1.5 T magnetic resonance system (Magnetom VISION; Siemens AG, Iselin, NJ) using standard quadrature head coil. First, a high-resolution 3D T1-weighted anatomical scan was acquired (TR/TE/FA 45ms/20ms/45°, matrix 2562, FOV 240 mm^2, 1.6-mm thick partitions). Functional images were acquired using gradient–echo EPI (TR 2500ms/TE 50ms). A total of 80 data sets (volumes) were collected with a 64^2 matrix over a 240 mm^2 FOV. Each data set consisted of 20 slices angled parallel to the planum sphenoidale, 6 mm thick, with a 1-mm gap. For each run, the first four scans were excluded to control for saturation effects. This 10-s equilibration period was followed by alternating 25 s rest/25 s task for four cycles. Twenty volumes were acquired per cycle. Four runs (sessions) were collected from each subject over 20 min, spaced equally apart by 5 min.

Visual smooth-pursuit paradigm. The visual target was generated by a Windows-based program written for a PC laptop and back-projected onto a screen. The screen was 15 inches from the subjects' eyes and viewed using a mirror attached to the head coil. The task consisted of a white dot traversing a black background at 16.7°/sec. The movement of the dot subtended a visual angle of 28°. During "rest" subjects were instructed to look straight ahead at the black screen.

Eye-tracking outside magnet. Subjects' eye movements were recorded outside the magnet with a task similar to the one used during the fMRI procedure (15). Subjects were seated 46 cm in front of a screen on which the smooth-pursuit target was displayed against a black background. The dot traversed the background at the same velocity as was used in the fMRI task and included a brief pause at the sides to allow calibration. Eye-tracking data were collected over 1 min, after which time the instrument was recalibrated and the subject rested. The rest and recalibration interval lasted about 3 min. The procedure was then repeated four times. Thus, compared to the fMRI experiment, the interval between "sessions" and the number of sessions was nearly identical. The duration of a session, however, was shorter outside the magnet. Head movement was minimized with a bite bar and head rest. Eye movements were recorded using an infrared photoelectrode limbus eye-tracking device with an accuracy of 0.25° of visual angle and a time constant of 4 ms. The analog output was sampled at 500 Hz using a 12-bit analog-to-digital converter.

Data Analysis

fMRI data analysis. Data were analyzed off-line on a PC using SPM99 (Wellcome Department of Cognitive Neurology, London, UK) and IDL (Interactive Data Language, RSInc., Boulder, CO) software. Spatial preprocessing, model specification and estimation, and statistical inference were carried out with SPM99. An anatomic region-of-interest (ROI) analysis was carried out using IDL. The first four image volumes were excluded for saturation effects. The four sessions were concatenated. Images were motion-corrected, normalized to the Montreal Neurological Institute template and subsequently converted to Talairach space. Data were then smoothed with a 4-mm FWHM Gaussian kernel. After an accounting was made for reslicing for motion correction, normalization to stereotactic space, and the applied smoothing kernel, the effective smoothing was approximately $8 \times 8 \times 8$ mm^3. The model consisted of a simple boxcar convolved with a hemodynamic response function.

Qualitative assessment. The 15 individual activation maps were thresholded at $T = 4.75$ ($P < .05$, corrected for multiple comparisons, using family-wise error) and assessed qualitatively.

Random effects vs fixed effects. In fitting the data to the general linear model, a spatial map of parameter estimates is produced for each subject. By contrasting for the main effect of condition, the parameter estimates provide a measure of the size of the effect of task for a subject. To identify brain regions constituting a significant group response, a second-level random-effects model was implemented by entering the parameter estimate map of interest for each subject into a one-sample *t* test (11). The activation maps produced by fixed effects and random effects were compared qualitatively and quantitatively. Since intersubject variability and loss in degrees of freedom contribute to the lower sensitivity of the random- compared to fixed-effects analyses, these factors were assessed quantitatively.

The intersubject variability was quantified using an ROI analysis. Anatomic ROIs were defined by templates based on Brodmann's areas (16), similar to the method used by Bookheimer *et al.* (17) in their fMRI

analysis. Using an in-house program written in IDL, we overlaid the set of Brodmann templates upon the high-resolution T1 images normalized to Talairach space. Next, the templates were coregistered to the anatomical image using a least-squares method. The total number of activated voxels was determined in each ROI. To compare the variability in the response across subjects and in different brain regions, we normalized the number of activated voxels in each ROI to the mean number of activated voxels across all subjects. Thus, if all subjects showed a similar response within a ROI, the normalized spatial extent should have hovered around 1.

Much of the reduced sensitivity of the random-effects compared to fixed-effects analysis is due to the substantial loss in degrees of freedom based on the number of scans (fixed effects) or the number of subjects (random effects). To compare the sensitivity of the two methods, we quantified the increase in activated voxels as a function of the number of sessions (scans) or subjects.

All statistical parametric maps were set at a threshold of $P < .05$, corrected for multiple comparisons using the false-discovery-rate (FDR) method. The false discovery rate is a correction procedure that applies to any multiple-testing situation. Compared to the Bonferroni correction, which tends to be overly conservative for neuroimaging data, the FDR method is less conservative because it controls the expected proportion of voxels for which the null hypothesis was rejected. In contrast, the Bonferroni correction controls for *all* voxels (18, 19). Theoretical advantages and practicality of FDR methods for analyzing fMRI data have been shown recently by Genovese *et al.* (19).

Effect of repetition. The effect of task repetition on activation was evaluated in two ways: (a) the condition-by-session interaction was modeled explicitly and contrasted for an increase and a decrease and (b) the spatial extent of activation was quantified within the anatomical ROIs and compared across the four sessions.

(a) Interaction between task and session. To test the hypothesis that brain regions involved in the attentional modulation of SPEM demonstrate less activation over sessions compared with regions that are not involved in attention, we modeled the interaction between condition (task) and session (run). Interactions were modeled as both a linear decrease and a linear increase over sessions (14). The statistical threshold was set at $P < 0.05$ (corrected). We confined the interaction analysis to those voxels considered active under the main effect of condition (e.g., of the voxels activated by the smooth-pursuit task, which of those voxels best fit the model?). This was achieved by masking the map of interaction effects with the main effects (masking threshold $P < 0.05$, corrected).

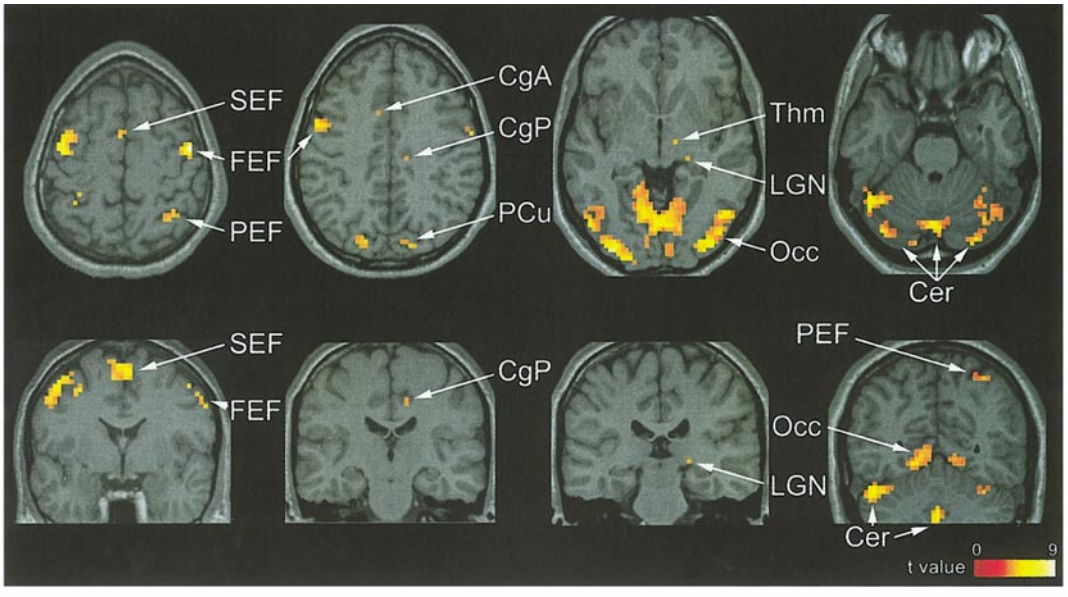
(b) Spatial extent of activation. In addition to a global map of session-dependent changes, we wanted a semiquantitative measure of the changes in activation. For each of the four sessions (acquired at 5-min intervals), group average statistical maps were generated at a significance threshold of $P < 0.05$ (corrected). Using the templates described above (an example of three ROIs of the template is shown in Fig. 3), the percentage of activated voxels within an ROI was automatically calculated for each of the four maps (sessions 1–4). ROIs were selected based on their known role in pursuit eye movements: frontal, parietal, supplementary eye field, cingulate gyrus, and cerebellar hemispheres.

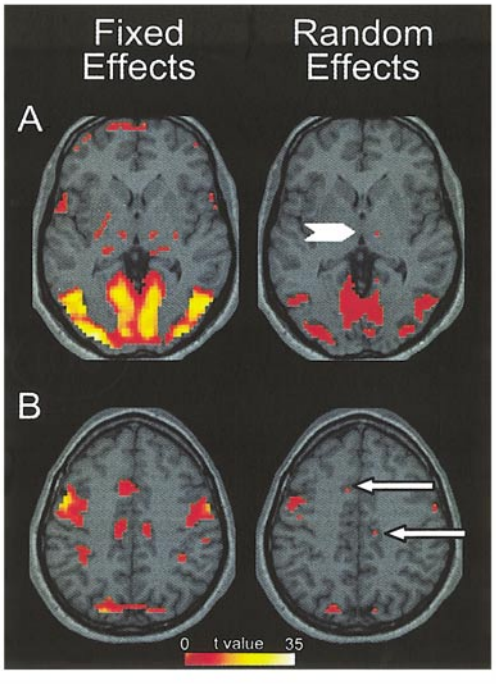
Eye-tracking data. Data were visually inspected for quality. The data were divided into discrete segments, and each segment was defined as saccade, smooth pursuit, or artifact. Pattern-recognition software removed artifacts caused by blinking. Segments not classified as either saccade or artifacts were considered smooth pursuit. All segments of pursuit were included in the gain calculation representing a global measure of task performance (20). Task performance was measured by the mean gain (mean eye velocity divided by target velocity) of all intervals of smooth pursuit. Optimal mean gain is 1.0, with eye velocity equal to target velocity (21). As age has a strong effect on smooth-pursuit performance (15), gain scores were converted to age-adjusted Z scores. Subjects were excluded if the age-adjusted Z score for gain was less than -1.0. Two subjects had abnormal smooth-pursuit gain and were excluded from the study. The remaining 15 subjects had normal smooth-pursuit parameters.

RESULTS

Qualitatively, all subjects demonstrated pursuit-related activation in frontal eye fields (FEF), parietal eye fields (PEF), MT/MST, and cerebellum. Of the 15 subjects, 9 demonstrated activation in the anterior cingulate gyrus, 14 the posterior cingulate gyrus, 12 the supplementary eye fields (SEF), and 7 the thalamus. There was moderate reduction in activation in all regions after controlling for random effects (Fig. 1). The loss of sensitivity incurred after the second-level analysis is shown qualitatively in Fig. 2. Although the BOLD response in subcortical structures significantly decreased, several voxels in the lateral geniculate nucleus of the thalamus and right anterior and left posterior cingulate gyri continued to exceed the significance threshold (Fig. 1). The Talairach coordinates of these regions agree with those reported by others (8, 9) and are shown in Table 1.

While all subjects demonstrated some activity related to SPEM in the specified regions, there was a moderate amount of subject-to-subject variability in the extent of activation, depending on the region. Figure 3 shows an example of the intersubject variability





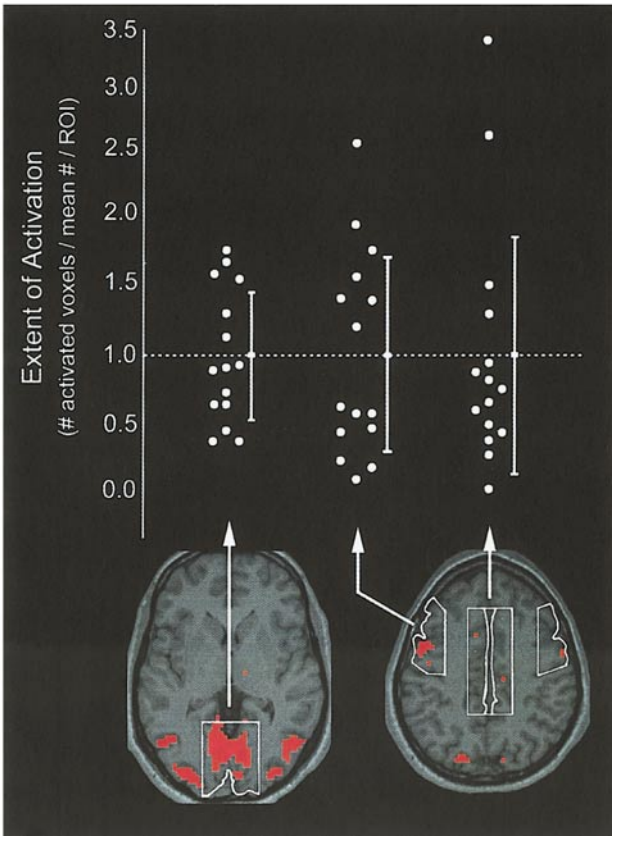
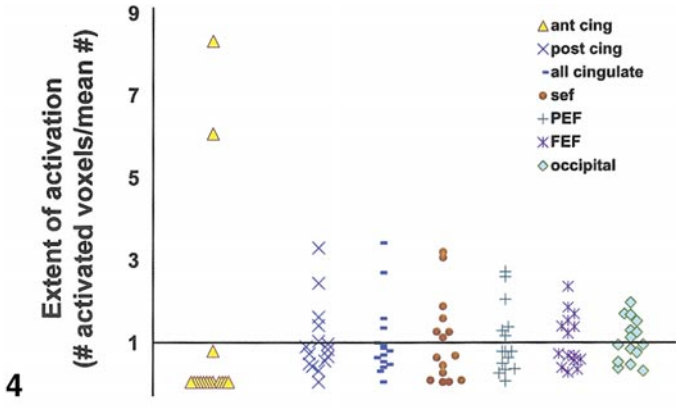


FIG. 1. Random-effects analysis of smooth-pursuit eye tracking shows activation in frontal (FEF), parietal (PEF), supplementary (SEF), precuneus (PCu), primary visual, and lateral temporal occipital (Occ) cortex. Subcortical activation is seen in cingulate gyrus (Cg), left lateral geniculate nucleus (LGN), left medial thalamus (Thm), cerebellar hemispheres, and vermis (Cer). Data are thresholded at $P < .05$ (corrected for multiple comparisons).

FIG. 2. Compared to the fixed-effects analysis, there is a moderate reduction in activation in all regions after the random-effects analysis. Nonetheless, several voxels remain significant in cingulate gyrus (arrows), medial thalamus (arrowhead), and LGN (not shown).

FIG. 3. Example of the Brodmann-based templates (16) used for ROI analyses. Shown are individual subjects' activations for three ROIs (occipital, FEF, and posterior cingulate). To compare variability across ROIs, individual responses were normalized to the mean for that ROI. There was greater variability in the cingulate gyrus than in the visual cortex.



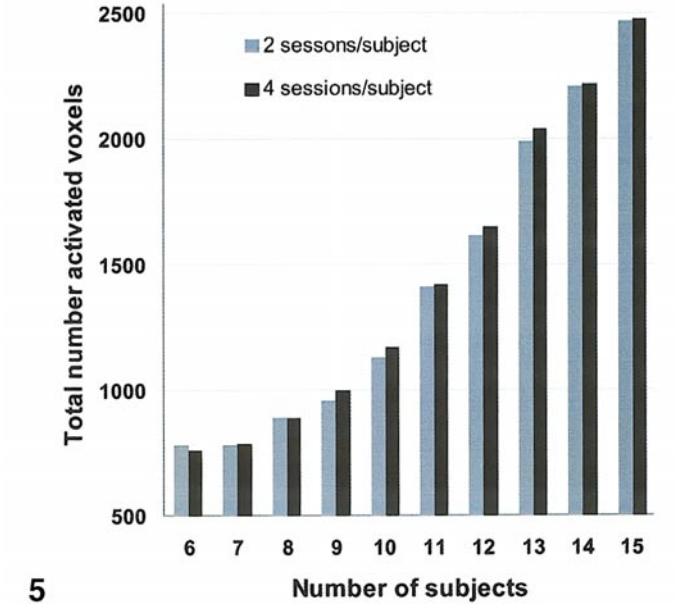


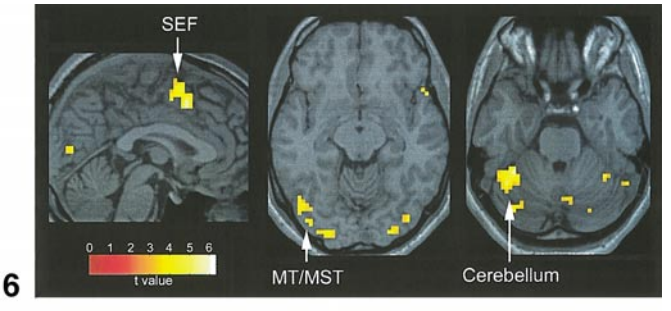
FIG. 4. Intersubject variability is shown for several brain regions. Only four subjects demonstrated significant activation in the anterior cingulate gyrus. All subjects demonstrated significant activity in the posterior cingulate gyrus. The occipital lobe showed the least variability.

FIG. 5. For random-effects analysis, whole-brain-activated voxels increased significantly by increasing the number of subjects, whereas doubling the number of scans from two to four sessions (160 to 320 scans) had little effect. This difference can be attributed to the lower degrees of freedom for the fixed compared to random effects.

in the activation for three ROIs. The cingulate region had the most variability, while the occipital region had the least. Scatter plots of intersubject variability are shown for additional ROIs in Fig. 4. The ROIs containing occipital cortex and frontal eye fields had the least subject-to-subject variation. The greatest intersubject variability was observed in the anterior cingulate gyrus, if analyzed as an isolated structure, whereas the variability was lower if anterior and posterior cingulate gyri were analyzed together.

Figure 5 demonstrates that increasing the number of subjects dramatically increased the sensitivity for detecting activity related to SPEM. In contrast, increasing the number of scans per subject from two to four sessions (160 to 320 scans) had little effect.

Effect of session. There was a general decrease in activation throughout cortex and cerebellum with repeated sessions. Figure 6 shows the interaction between condition and session, contrasting a *decrease* in activation. Not all brain regions decreased uniformly. Relatively greater reductions were observed in supplementary eye field, right parietal eye field, lateral occipital temporal cortex, and cerebellar hemisphere (Fig. 6). On the other hand, there was no significant interaction between condition and session, contrasting an *increase* in activation. In addition to modeling the interaction of task *over* sessions, we also modeled a parametric linear decrease in activation *within* a session. We found no significant reduction in activation within the short, 3-min session. To quantify the session-dependent decrease, we used the ROI approach, comparing the extent of activation for five ROIs. To allow comparisons across regions, activated voxesl were normalized to the ROI. Figure 7 shows that the session-



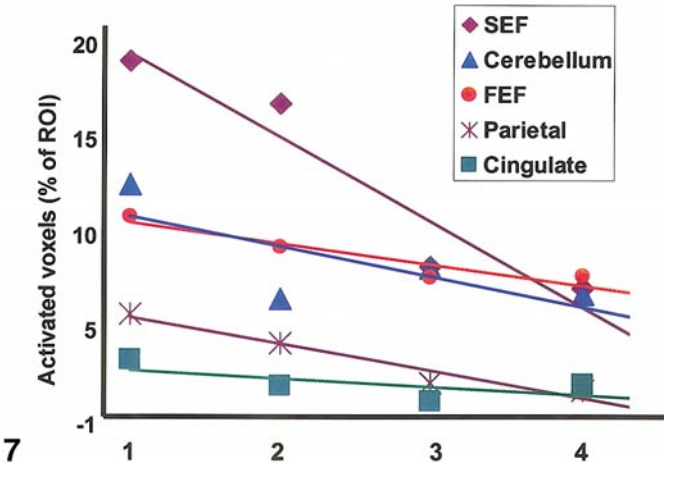


FIG. 6. Regions of the brain where there was a significant interaction between condition and session, contrasted for a decrease in activation. The SEF, lateral temporal occipital, and right cerebellum showed the greatest session-dependent reduction. Data are thresholded at $P < 0.05$ (corrected for multiple comparisons).

FIG. 7. The plot demonstrates reductions in the spatial extent of activation across session in all brain regions. However, SEF showed the most decrement over time. Data are normalized to enable comparisons across regions.

TABLE 1

Smooth-Pursuit Eye Movement Activation (Random-Effects Analysis)

		Coordinates				
		x	*y*	*z*	*t* value[a]	*P* value[b]
Cortical area						
FEF	(R)	42	−6	48	7.52	0.005
	(L)	−48	−6	54	8.88	0.005
PEF	(R)	30	−39	48	5.52	0.007
	(L)	−33	−57	57	4.47	0.014
SEF	(R)	6	3	60	5.89	0.006
	(L)	−3	0	60	4.99	0.010
Cingulate, anterior	(R)	9	12	42	3.37	0.046
	(L)	—	—	—	—	—
Cingulate, posterior	(R)	—	—	—	—	—
	(L)	−12	−21	42	3.47	0.041
Striate	(R)	6	−72	15	7.90	0.005
	(L)	6	−69	9	7.63	0.005
MT/MST	(R)	42	−87	−9	7.64	0.005
	(L)	−30	−90	−15	7.61	0.005
Subcortical area						
Thalamus, medial	(R)	—	—	—	—	—
	(L)	12	−15	−3	4.10	0.020
Thalamus, LGN	(R)	—	—	—	—	—
	(L)	21	−27	−3	3.81	0.027
Cerebellar hemisphere	(R)	42	−45	−36	3.47	0.041
	(L)	−36	−36	−39	4.86	0.010
Vermis		0	−60	−45	6.99	0.005

Note. x, y, and *z* (mm) refer to local maxima of BOLD response at stereotactic coordinates based on the dimensions of the atlas of Talairach and Tourneaux.

[a] *t* value indicates the significance of the activation change.

[b] *P* value corrected using false discovery rate.

dependent decrease in the spatial response to SPEM varied across regions. There was a relatively greater decrease in SEF compared to other regions. Because of the large number of regions evaluated, we did not test for statistical significance.

DISCUSSION

The main finding of this paper is that BOLD activity related to smooth-pursuit eye movements was consistent across subjects to the extent that all cortical and some subcortical regions constituted a significant response among normals (11). The anatomical pathways associated with eye movements in humans previously studied using PET imaging have demonstrated activation in frontal, parietal, and supplementary eye fields. Most of the previous work has focused on saccades (4, 14), but a few studies have observed activation in similar dorsal cortical regions during SPEM (8, 9). Using fMRI techniques, Petit *et al.* observed activation related to SPEM in the MT/MST areas as well as the dorsal cortical regions, but their study was limited to five subjects and the eye-tracking performance was not verified (7, 9). Berman *et al.* reported activation in the anterior cingulate gyrus in 7 of 11 subjects and posterior cingulate gyrus in most of the subjects performing SPEM and imaged at 3 T (8). These fMRI studies of eye movements have been restricted to fixed-effects analyses, which are highly sensitive due to large degrees of freedom and low scan-to-scan variation. Statistical methods that account for intersubejct variability, however, have not received attention and are particularly important for assessing activity within small structures such as the cingulate gyrus and thalamus, which elicit modest activation compared to surface cortex. SPEM is known to be abnormal in specific neuropsychiatric conditions such as schizophrenia, Parkinson's disease, genetic disorders, and hemispheric lesions (2). Defining the functional anatomy of smooth pursuit could elucidate the pathophysiology of these diseases, but the reliability with which a region responds must be established before inferences regarding these patient populations are made. SPEM deteriorate with old age (15), supporting a further need to establish intersubject reliability, even among controls. Friston *et al.* (22) emphasized the need to select the appropriate analysis depending on the level of inference desired. Since the goal would be to extend one's inferences about fMRI of SPEM to the population, a random-effects analysis is critical.

The current study shows that activations in FEF, PEF, areas MT/MST, posterior cingulate gyrus, and

cerebellum are highly consistent across subjects. In contrast, activations in SEF, anterior cingulate gyrus, and thalamus (including LGN) are more variable and nearly eliminated following the second-level analysis (Figs. 1 and 2). The loss in sensitivity after the second level of analysis stems from two separate but related factors: intersubject variance and the loss of degrees of freedom. For a given region, if all subjects demonstrated a similar response, the region would survive the second level analysis. If, on the other hand, only one or two subjects demonstrated a strong response, activation in this might exceed significance threshold for a fixed-, but not random-, effects analysis. Our results show that in the visual cortex normal subjects demonstrated a very similar response to SPEM, whereas there was a good deal of variability in other brain regions, especially the anterior cingulate gyrus (Fig. 4). Larger cohorts are required for random-compared to fixed-effects analyses because of a precipitous decline in degrees of freedom, as shown by the current study (Fig. 5). fMRI experiments, while easier to implement than PET studies, are still time consuming and expensive. Given limited resources, these results strongly support the philosophy that the payoff in statistical power is to reduce the number of scans per subject and to increase the number of subjects.

The second finding was that a decrease in activation occurred for some brain regions more than others with the repetition of SPEM. Factors that could explain this temporal decrease are greater noise due to motion, alterations in pursuit performance, or true physiological changes related to attention, learning, or adaptation. A motion artifact is unlikely to contribute greatly, as it would not explain the different degrees of reduced activation among brain regions of similar volume. Task performance is also unlikely to play a significant role, as smooth-pursuit performance is stable both across and within runs (23). Previous PET and fMRI studies reporting similar session-dependent changes in activation in specific brain structures (13, 14, 24–26) have attributed such temporal differences to physiological adaptation or cognitive processes (e.g., learning) or states (e.g., attention). We found session-dependent decreases in activation in the right cerebellum, right temporal occipital cortex, and SEF (Fig. 6). These results are consistent with the reduction in cerebellar activity associated with repeated motor activity (24) and self-paced saccades (14). If attentional set (readiness) plays a role in the session effect we observed, then the data support the hypothesis that cerebellar hemispheres are involved in attentional control, whereas the vermis is associated with ocular movement per se (27, 28). The region showing greatest attenuation in terms of both condition by session interaction and spatial extent of activation (Figs. 6 and 7) was the SEF. This is in contrast to work by Schmid *et al.* who observed session-dependent *increases* in SEF/supplementary motor area (SMA) activation using smooth pursuit. A major difference between the current study and that of Schmid *et al.* was their use of a task that specifically addressed the learning of eye movements (by contrasting predictable with nonpredictable conditions). This is significantly different from our task, which was highly predictable and involved no planning or learning. Our results also differ from those of Dejardin *et al.*, who found session-dependent increases in SEF activation with saccades (14). This is not too surprising, given the different physiology of saccades compared to pursuit. Specifically, spatial working memory is thought to be an important component of saccades and may be related to the role of SEF in oculomotor learning (3). In the context of top-down attentional modulation of processing visual information, we hypothesize that the reduced activation in SEF, lateral occipital temporal cortex, and cerebellum reflected a decreased need to attend to the smoothly moving dot. Another potential explanation for the session-dependent decreased activation is that it resulted not from task-related activity, but rather from an increase in activation over the rest periods. Since we did not track eye movements in the scanner and since rest periods did not involve fixation, this remains a possibility. To test for this, we explicitly contrasted for a session-dependent *increase* in activation during rest periods and found no effect (data not shown). Establishing attention-related effects for distinct brain structures involved in SPEM clearly has implications for future paradigm design, either as a variable of interest or one for which there needs to be control.

In the following sections we discuss the results with regard to specific structures and their functions.

Frontal and Supplementary Eye Fields

The function of FEF in humans is incompletely understood. Lesions to the FEF impair performance on saccadic and pursuit tasks (1, 29), but these studies are difficult to interpret because lesions are rarely discrete. Behavioral and motor effects are confounded by lesion extension to other areas such as prefrontal cortex (29, 30). FEF activation has been observed in both saccadic and smooth-pursuit eye tracking in humans, detected with PET cerebral blood flow (4) and fMRI (7, 8, 31) methods. Lesion and activation studies, together suggest that FEF is involved in task execution, while prefrontal association cortex and dorsolateral prefrontal cortex contribute to initiation and monitoring eye movements (30, 32). The activation we observed in the medial dorsal frontal cortex is consistent with previous studies localizing human SEF (33, 34). Differentiating SEF from SMA was beyond the resolution of this study and the two are considered together for this paper. The SEF is thought to be important in the intent to perform rather than the execution of eye movement (33, 35).

Furthermore, there is flexibility in that the effect of electrical stimulation of SEF depends on what a monkey has been trained to do previously, suggesting it plays a role in adaptation and learning (3). Thus, the relative stability of FEF compared to the significant decrease in SEF activation across sessions is consistent with these functions. As noted in the previous section, the session-dependent *decrease* we observed and the *increase* observed by Schmid *et al.* could be explained by the fact that our task was largely a simple sensorimotor one, while Schmid specifically evaluated the learning aspects of SPEM. These results support a putative role of SEF for planning and attending to eye movements.

Cingulate Gyrus

The cingulate cortex is also heavily implicated in attention-related functions. Mesulam has described it as the "paralimbic belt, providing a cytoarchitectonic transition zone between limbic areas and frontoparietal cortex" (36). Within the cingulate cortex there appears to be specialization of the anterior compared to posterior cingulate gyrus. Regarding eye movements, the posterior cingulate gyrus is involved in integrating sensorimotor signals to maintain ongoing eye movement (37). In keeping with this hypothesis, Berman *et al.* (8) reported robust posterior cingulate gyrus activation in 11 of 11 subjects during pursuit eye movements. We also observed consistent (14 of 15 subjects) activation and the fixed-effects analysis yielded strong bilateral activation in the posterior cingulate gyrus. The substantial decrease in activation following a second-level analysis, however, suggests that at 1.5 T, the fMRI-related signal change observed in posterior cingulate gyrus is weak, albeit consistent. The anterior cingulate gyrus has been implicated in complex as opposed to simple sensorimotor tasks. Similar to the Berman study, we found that activation in the anterior cingulate gyrus was smaller than that in the posterior cingulate gyrus and showed the greatest intersubject variability.

Area MT and MST

The middle temporal visual area is analogous to the superior temporal sulcus in the monkey (2) and receives projections from the striate cortex. Discrete lesions in MT disrupt smooth-pursuit initiation without necessarily impairing saccades. Thus, MT is involved in the processing of visual motion. Adjacent to MT is the medial superior temporal visual area, which receives major projections from area MT (38). Neurons in MST differ from MT by also receiving input relating to eye and head movement (39). Robust activation was observed in primary visual cortex and lateral occipital temporal cortex (V5 and V5a). Similar to Petit, we found consistent MT/MST activation across all subjects (7). There was a strong session-dependent reduction in right lateral occipital temporal activation. The interpretation is that, like SEF, region MT/MST is relatively sensitive to attentional state. In support of this hypothesis, Barton *et al.* found more activation in the lateral occipital temporal cortex during a pursuit compared with a moving grating task. The authors attributed this relative increase in signal, despite a decrease in retinal image motion, to extraretinal factors such as attention (40).

Lateral Geniculate Nucleus and Thalamus

In the fixed-effects analysis, moderate activation in both medial thalami and lateral geniculate nuclei was observed, but only two voxels in the left LGN remained significant after the second-level analysis. Chen *et al.* reported consistent activation of LGN at a nominal spatial resolution similar to that used in the current studies, but at 4 T (41). Buchel *et al.* acquired images at 27 mm^3 at 2 T and reported that LGN activation was preserved after spatial normalization and group averaging (42). The current study differs from the previous studies in two important aspects: data were acquired at lower field strength and the visual stimulus was subtler. Berman *et al.* reported thalamic activity in two of five subjects (at 3 T), but the location was not described (8). The volume of human LGN ranges between 70 and 150 mm^3 (43). Given a nominal spatial resolution of 84 mm^3, the LGN would correspond to one or two voxels. Based on a twofold variability in LGN volume in normal subjects (43), it is not surprising that LGN activation was dramatically reduced after second-level analysis.

Cerebellum

The current study provides the first evidence using fMRI that the cerebellum plays a significant role in SPEM. Petit *et al.* acquired whole brain fMRI data, but did not report cerebellar activity (7). Dieterech *et al.* found cerebellar activation using optokinetic stimulation and self-guided saccades (27), but not pursuit. Precise identification of pursuit-specific cerebellar structures is complicated by massive cerebral cortical input via pontine pathways. Specific neurons that discharge for pursuit are found in the flocculus and vermis, but other cerebellar structures likely contribute as well (1, 2). While large regions of activity were found in both cerebellar hemispheres, the maxima localized to the flocculi. We also observed strong response in the vermis. Interestingly, there was a significant session-dependent reduction in the hemisphere activation (especially the right) compared with the vermis. This is consistent with the idea that activation in the vermis is more stable since it is associated with oculomotor activity, whereas activation in the cerebellar hemisphere

is more sensitive to repetition because it is involved in attention, as suggested in other work (27).

A possible limitation of this study was that the relatively wide visual angle incurred eye motion and not just retinal motion. This may have resulted in greater activation of FEF and SEF due to involvement of the efferent motor system, but was chosen because we wanted to use task parameters that matched our eye-tracking paradigm outside the magnet (15). We did not quantify eye movements during fMRI scanning, only outside of the magnet, which is a limitation for the condition by session-interaction analysis. The templates used for ROI analyses were somewhat arbitrary. The differentiation of small structures, such as of the anterior from the posterior cingulate gyrus, for example, may not have been as accurate as a manually traced ROI. Since most of the cortical regions of interest were large relative to the template ROIs, this was not a problem. Advantages of the Brodmann-based templates were automated fitting to anatomic images (e.g., there was no subjective input), the use of the template by previously published studies (16, 17), and wide availability. Another limitation common to all *in vivo* imaging studies is that activity does not mean the area is necessary for control of the task, but merely that it is involved. Only lesion studies can determine areas that are critical.

CONCLUSION

In addition to providing information about task-related activity in individuals, many fMRI studies strive to make generalizable comments about a population. In the latter case, the analyses should extend beyond a fixed (first)-level to include a random (second)-level statistical analyses. This paper extends previous work on fMRI of SPEM by controlling for random effects. While it is possible to detect subcortical activations related to SPEM at 1.5 T, we remain cautious regarding second-level inferences, depending on the size, spatial resolution, and intersubject variability for that region. We show empirically that to improve the sensitivity of the random-effects analysis, studies should aim to maximize the number of subjects. Finally, a session-dependent decrease in activity related to SPEM was different among brain regions and may indicate areas that are sensitive to the attentional modulation of eye movements. This reduction in activity may be an important experimental variable which is often overlooked.

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