

Modulation of language areas with functional MR image-guided magnetic stimulation

J. Andoh,^a E. Artiges,^a C. Pallier,^b D. Rivière,^c J.F. Mangin,^{a,c} A. Cachia,^a M. Plaze,^a
M.L. Paillère-Martinot,^{a,d} and J.L. Martinot^{a,*}

^aINSERM-CEA Research Unit ERM 02-05, National Institute for Health and Medical Research (INSERM) and Atomic Energy Commission (CEA), IFR49, Frédéric Joliot hospital department, 4 place Gl. Leclerc, 91401 Orsay, Paris Descartes University, France

^bINSERM-CEA U562, Frédéric Joliot hospital department, Orsay, France

^cImage Analysis and Structural Anatomy Group, IFR49, Frédéric Joliot hospital department, Orsay, France

^dAP-HP, Psychiatry department, A. Chenevier and H. Mondor Hospitals, Créteil, France

Received 24 May 2005; revised 11 July 2005; accepted 18 July 2005

Available online 15 September 2005

Repetitive transcranial magnetic stimulation (rTMS) can interfere with linguistic performance when delivered over language areas. At low frequency (1 Hz), rTMS is assumed to decrease cortical excitability; however, the degree of TMS effect on cortical language areas may depend on the localization of the stimulation coil with respect to the inter-individual anatomo-functional variations.

Hence, we aimed at investigating individual brain areas involved in semantic and phonological auditory processes. We hypothesized that active rTMS targeted over Wernicke's area might modify the performance during a language-fragment-detection task. Sentences in native or foreign languages were presented to 12 right-handed male healthy volunteers during functional magnetic resonance imaging (fMRI). 3D-functional maps localized the posterior temporal activation (Wernicke) in each subject and MRI anatomical cortical landmarks were used to define Broca's pars opercularis (F3Op). A frameless stereotaxy system was used to guide the TMS coil position over Wernicke's and F3Op areas in each subject. Active and placebo randomized rTMS sessions were applied at 1 Hz, 110% of motor threshold, during the same language-fragment-detection task. Accuracy and response time (RT) were recorded. RT was significantly decreased by active rTMS compared to placebo over Wernicke's area, and was more decreased for native than for foreign languages. No significant RT change was observed for F3Op area. rTMS conditions did not impair participants' accuracy. Thus, low-frequency rTMS over Wernicke's area can speed-up the response to a task tapping on native language perception in healthy volunteers. This individually-guided stimulation study confirms that facilitatory effects are not confined to high-frequency rTMS. © 2005 Elsevier Inc. All rights reserved.

Keywords: Transcranial magnetic stimulation; Functional magnetic resonance imaging; Neuronavigation; Language; Broca; Wernicke

Introduction

Transcranial magnetic stimulation (TMS) provides a non-invasive method to investigate cognitive functions, such as language processing (Epstein, 1998). By inducing a local current in the cortical tissue, TMS interferes with ongoing brain activity, thus creating a temporary “virtual lesion” (Pascual-Leone et al., 2000). This TMS specificity is used to interfere transiently with brain activity in a specific region to characterize behavioral effects (Matthews et al., 2003).

TMS recently highlighted the functional demand on language-related areas during linguistic tasks, and further questioned the classical model of language localization (Broca, 1861; Wernicke, 1874), which assigns planning and production of speech to the anterior perisylvian area (Broca's area) and both speech recognition and sound representations to the posterior perisylvian area (Wernicke's area).

Broca's area is an anatomically complex region including the pars opercularis (F3Op), the pars triangularis (F3Tr), and the pars orbitalis (F3Or) (Fig. 1A), which shows anatomic variability between subjects (Burton et al., 2001; Tomaiuolo et al., 1999). Investigators have used TMS to clarify the function of these subregions and reported differential effects on syntactic and semantic processing.

High-frequency or single-pulse TMS has implicated F3Or in semantic processing (Devlin et al., 2003; Matthews et al., 2003). In addition, F3Op has been involved in syntactic processing (Sakai et al., 2002), in phonological processing (Devlin et al., 2003; Nixon et al., 2004) and in the motor control of speech (Watkins et al., 2003), and has been suggested to operate as an interface of perception and action (Aziz-Zadeh et al., 2004; Watkins and Paus, 2004). However, other authors have reported no effect of low-frequency or high-frequency repetitive TMS (rTMS) applied over Broca's area (Sparing et al., 2001).

The role of Wernicke's area has also been revisited. Wernicke's area's precise location is still debated, within the posterior

* Corresponding author.

E-mail address: jlmartinot@cea.fr (J.L. Martinot).

Available online on ScienceDirect (www.sciencedirect.com).

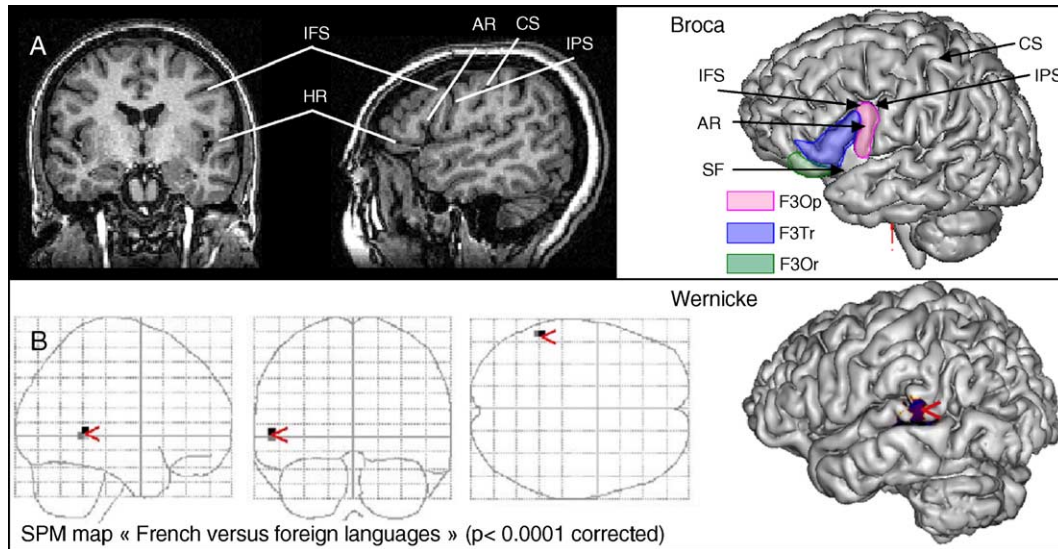


Fig. 1. (A) F3Op definition using anatomical landmarks. Cortical landmarks used to identify the pars opercularis (F3Op) of Broca's area: AR/HR = ascending and horizontal rami of the Sylvian fissure, CS = central sulcus, IFS = inferior frontal sulcus, IPS = inferior branch of the precentral sulcus. Coronal (left) and sagittal views of the left hemisphere in a nonnormalized MRI. Cortex surface rendering (right) of the MRI series with landmarks superimposed to indicate the pars orbitalis (F3Or), the pars triangularis (F3Tr) and the pars opercularis (F3Op) (Duvernoy, 1999). (B) Wernicke's area definition using functional landmarks. (Left) Individual statistical parametrical maps for the fMRI image subtraction "French minus foreign languages" (height threshold: P corrected < 0.0001). The cortex cluster with the highest t value was clearly visible and corresponded to Wernicke's area (peak voxel at $-50, -46, 1; Z > 6.44$) in the Talairach and Tournoux atlas (Talairach and Tournoux, 1988). (Right) Brainvisa software allowed reporting the cluster of maximal activity onto the subject's unnormalized 3D cortex rendering.

temporal or inferior parietal regions (Burton et al., 2001; McDermott et al., 2003; Peelle et al., 2004; Seghier et al., 2004); it is often defined on a functional basis (Friederici, 2002; Luke et al., 2002) and therefore varies across subjects. Wernicke's area has been variously defined in TMS studies which did not take into account its inter-subject functional variability. For instance, some investigators have approximated Wernicke's area with respect to different electrode locations in the international 10–20 electrode system (Claus et al., 1993; Sparing et al., 2001; Knecht et al., 2002). Low-frequency rTMS (1 Hz) over Wernicke's area suppressed language functions in a word-generation task (Knecht et al., 2002), single-pulse TMS or high-frequency (5 to 20 Hz) rTMS applied to the same area before picture presentation facilitated picture naming (Mottaghy et al., 1999; Topper et al., 1998). Thus, the degree of variability of the TMS effects on language processing may depend on the diversity of language tasks used by the investigators (Binder et al., 1997), or on the stimulation parameters used (Maeda et al., 2000; Sparing et al., 2001), but also on factors such as the localization of the stimulation coil with respect to the inter-individual variations of cortical areas.

While most TMS studies did not accurately localize the stimulated cortical areas, a higher precision should be attainable in repositioning the TMS stimulation coil in order to control for inter-subject morphologic or functional variability. Indeed, the variability of brain folding and functional activations in the left inferior frontal (Broca) and superior temporal (Wernicke) regions has been highlighted in functional magnetic resonance imaging (fMRI) studies during tasks that involve words and sounds as auditory stimuli (Malogiannis et al., 2003; Pallier et al., 2003; Seghier et al., 2004). However, most fMRI reports were based on analyses that blurred the regional inter-individual variations through normalized group-activation maps.

Recently, the analysis of the individual activation maps in a fMRI study contrasting the processing of audio sentences in the native vs. unknown languages (Pallier et al., 2003) has shown the involvement of the posterior superior temporal gyrus (STG) in each subject. Such individual fMRI determination should enable to target a specific location in each subject, and to guide the TMS coil position for stimulation (Bohning et al., 1999; Herwig et al., 2001).

In this study, we used individual mapping and frameless stereotaxy to target and stimulate brain regions involved in semantic and phonological auditory processes while they performed the same language task outside the MR imager. Because high-frequency rTMS may increase the risk of inducing seizures (Anand and Hotson, 2002; Wassermann et al., 1996), we investigated whether low-frequency rTMS has an effect in healthy volunteers. According to literature, we did not expect any modification of task performance for rTMS over the anterior perisylvian target since F3Op has rather been involved in phonological or speech generation processes (e.g., Zatorre et al., 1996; Price, 2000). We hypothesized that low-frequency rTMS over Wernicke's area may modify task performance since this region might implement auditory language identification functions (Pallier et al., 2003; Binder et al., 1997).

Methods

Subjects

Twelve healthy male subjects were included (age range, 20 to 26 years; mean age, 22 years); they were native French speakers, not familiar with Polish or Korean languages, and right-handed according to the Annett's questionnaire (1967) (range 90 to 100%, mean $97.3 \pm 1.0\%$). Their educational level was homogeneous

(9.8 ± 1.5 years of education). Exclusion criteria were alcoholism, drug addiction and history of psychiatric or neurological disease.

All experiments followed the safety guidelines of TMS experiment (Chen et al., 1997; Wassermann et al., 1996) and approval was obtained from the Paris-Pitié-Salpêtrière ethics committee. Each subject gave written informed consent after the nature and possible consequences of the study had been explained.

Task description

The same language task was used in both the fMRI and TMS experiments and was modified from Pallier et al. (2003). Subjects listened to sentences in their native language and in languages unknown to them (Korean, Polish) and were required to perform a fragment-detection task. Following each 3 s sentence and after a 500 ms delay, a 500 ms fragment was played (Fig. 2).

Subjects had to indicate by pressing one of two response buttons whether this fragment had appeared in the sentence or not: the instruction was to respond ‘present’ by pressing the right button with the right thumb, and if not, by pressing the left button with the left thumb. The inter-trial interval was 5000 ms. Languages and side of response were presented in a different randomized order for each subject. A PC running E-Prime software (Psychology Software Tools Inc., Pittsburgh, USA) allowed the listening of auditory stimuli and recording of the responses.

Anatomical MRI acquisition

3D anatomical images were acquired using a 1.5 T General Electric Signa System scanner (General Electric Medical Systems, Milwaukee, WI, USA). MRI scanning parameters included a slice thickness of 1.3 mm, a field of view of 24 cm and an acquisition matrix of $256 \times 256 \times 128$ voxels. A conjugate synthesis in combination with an interleaved acquisition resulted in 124 contiguous double-echo slices whose voxel dimensions were $0.94 \times 0.94 \times 1.3$ mm³. These high-resolution fSPGR T1-weighted images enabled reconstruction of the fine individual cortex folding (Mangin et al., 2004), which was used as anatomical landmarks for the TMS targets.

fMRI design and acquisition

The brain language areas were mapped using auditory events synchronized with functional MR image volume acquisitions. During presentation of auditory stimulations, a gradient-echo echo-planar imaging sequence sensitive to brain oxygen-level dependant (BOLD) contrast was used (18 slices, TR/TE = 2000/60 ms, FOV 24 cm, 64×64 matrix, voxel size $3.75 \times 3.75 \times 6$ mm³).

Before scanning, subjects performed a practice run of 10 trials. Stimuli were administered in two 14 min blocks, each comprising 96 trials: 32 sentences in French, 32 sentences in foreign languages and 32 silence periods. The first four trials were excluded from the analyses to allow for stability in magnetization. Two blocks with 416 functional images (=832) were acquired for each subject.

fMRI data analyses

Functional time series data were processed using SPM99 (Wellcome Department of cognitive Neurology, London; <http://fil.ion.ucl.ac.uk/spm>), with MATLAB® 6.1 (<http://www.mathworks.com/products/matlab>). Preprocessing of functional time series included slice timing, spatial realignment (correction of movements). The functional time series data were then normalized using nonlinear spatial normalization to the stereotaxic space devised in the Montreal Neurological Institute (MNI), based on the T1 structural images of each subject to classify the activated areas in a standard stereotaxic space. Finally, each image was smoothed with a 5-mm at full-width half maximum (FWHM) Gaussian filter.

Two separate analyses of these data allowed (1) to detect cerebral activation during right and left thumb movements and (2) to detect the activation during semantic processing (French language stimuli vs. foreign languages stimuli).

The statistical parametrical map (SPM) reflecting the t-contrast (t-map) between right and left thumb movements was obtained and allowed detecting the right thumb movement activation in the motor cortex in a standard stereotaxic space. The activated area in the left hemisphere corresponded to the unnormalized set that was targeted with TMS and used for motor threshold determination (see below).

A linear model was then defined by two categories of events: *French* and *foreign languages*. We chose the *foreign languages* condition as a baseline that was “neutral” for all the subjects, and determined the areas where *French* stimuli elicited stronger activations than *foreign languages* stimuli. Individual activation maps (Fig. 1B), defined as separate activated clusters on each subject’s functional map, were identified according to their x , y , z Talairach’s coordinates and their corresponding anatomical landmarks.

In the posterior temporal region (Wernicke), Talairach’s mean coordinates of the maximum fMRI activation (peak voxel) were computed for the group on the basis of individual response location; also, we assessed inter-individual variability in Talairach’s coordinates, and frequency of activation within the group (Seghier et al., 2004; Xiong et al., 2000).

The coordinates for the peak voxel within the activated regions were obtained using the SPM99 software. We used an algorithm (<http://www.mrc-cby.cam.ac.uk/Imaging/mnispace.html>) for an

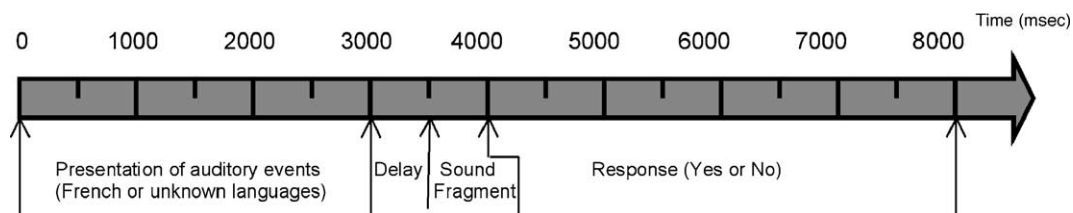


Fig. 2. Schemata of one trial in the fragment-detection language task, during fMRI or rTMS. Participants listened to sentences in their native language and in languages unknown to them (and silence periods for fMRI experiment). The instruction was to press one of the two buttons responses whether a short fragment did belong to a sentence just heard. Vertical lines symbolize the 1 Hz-pulses during the TMS experiment.

approximate conversion to relate these peak coordinates to the Atlas of Talairach and Tournoux (1988).

Determination of the TMS sites from MR images

The t-maps constructed from the fMRI data (defined in Talairach's space) described above (i.e., right thumb's and Wernicke's areas) were loaded into Brainvisa software (<http://brainvisa.info>).

For a voxel-maximum located in the cortex, the software provided automatically the closest barycenter on the head surface (defined in the subject's MRI space) as the optimal coil position for TMS stimulation (Fig. 3C).

The identification of the appropriate sulci and gyri that assists in defining the pars opercularis (F3Op) of Broca's area was made on the basis of each subject's anatomical MRI scan (Devlin et al., 2003; Nixon et al., 2004; Stewart et al., 2001), with reference to an anatomical brain atlas (Duvernoy, 1999). The F3Op was delimited rostrally by the ascending rami of the Sylvian fissure (AR); dorsally, by the inferior frontal sulcus (IFS) and ventrally, by the Sylvian fissure (SF). The identification of the cortical folds (AR, IFS and SF) was obtained from coronal and sagittal MRI slices and 3D rendering of the cortex relief (Fig. 1A).

TMS procedure

The same subjects participated to the TMS experiment that took place a few days after MRI data acquisition. An optical computerized frameless stereotaxy system (Brainsight, Rogue Research, Montreal, Canada; <http://www.rogue-research.com>), plugged to Brainvisa software, was used to track in real-time the positioning of the coil and to guide it to the optimal coil position determined on the MRI for each target site. Subjects were positioned on a platform to hold their head relatively immobile using a chin and a forehead rest. Constant coil positioning was assured by monitoring the coil position display over the subject's head. Because the targets were defined in each subject's MRI space, it was necessary to coregister this space with the actual space where the subject and the coil were. The registration process used four anatomical landmarks (tip of the nose, bridge of the nose, left and right ear tragus) selected on both the subject's MRI and the subject's head. The registration allowed locating the coil position with respect to the subject's head surface and brain.

To obtain 3D anatomical display, the construction of 3-dimensional MRI representations of each volunteer's brain, head and scalp target was carried out using the Brainvisa software. This software included a dedicated TMS guidance procedure that

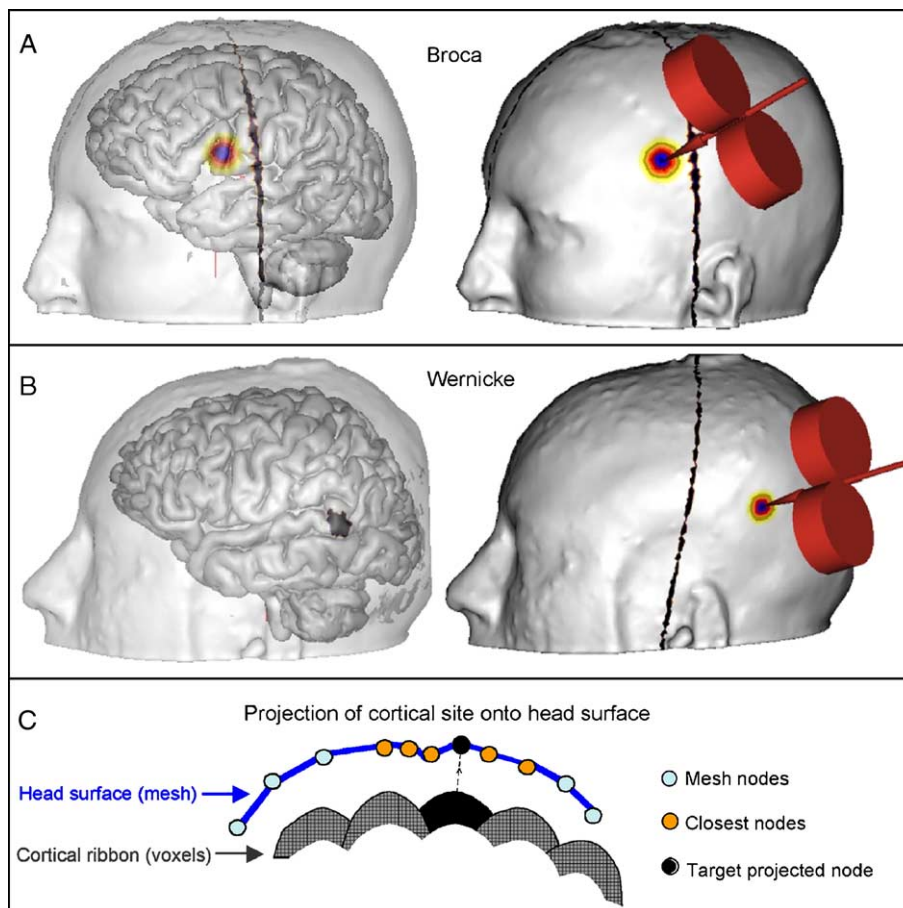


Fig. 3. Definition of the TMS target sites on the head surface, for F3Op (A) and Wernicke's (B) areas. The targets (F3Op and Wernicke's areas) were localized individually and guided the on-line coil positioning; in these figures, the coil is represented with an angulation allowing target visualization. (C) Schematic cortical target projection on a head surface reconstruction. Brainvisa uses a robust approach where the surface target (black dot) is defined as the barycenter of the n ($n = 10$) surface mesh-nodes (orange dots) that are closest to the cortex cluster. Indeed, inaccuracies in head surface reconstruction may induce local geometric artifacts confounding the target projection on the head surface. Hence, the closest scalp point may not be the optimal target position, while the barycenter minimizes the deviation.

enabled to visualize in real-time the coil position according to the functional and anatomical data. Hence, targets were individually monitored and stimulated.

TMS was applied using a MagPro R30 stimulator (Medtronic-Dantec, Skovlunde, Denmark; <http://www.medtronic.com>) with an 8-shaped coil where each wing measured 50 mm in diameter (maximum output 2 T). The coil was fixed in place by a mechanical arm and was adjusted until the central axis of the coil wings intersected perpendicularly the target stimulation site on the MRI display (see Figs. 3A,B). The coil was placed tangential to the head at the point of contact and held such that its handle pointed down and backward at an angle approximately 45° (Hoffman et al., 2003; Maeda et al., 2002; Rizzo et al., 2004).

A placebo coil (Medtronic-Dantec) was also used in order to reproduce the same acoustic stimulation as the active coil, but did not induce the magnetic effects associated with TMS. The subjects were unaware that they would not receive any stimulation.

The level of stimulation used was determined with reference to each subject's resting motor threshold. Surface electrodes were placed on the right abductor pollicis brevis (APB) and a round ground electrode was placed on the wrist and connected to a Dantec CounterPoint electromyograph. The optimal scalp site, i.e., the scalp position where TMS induced motor-evoked potentials (MEPs) of maximal amplitude in the APB hand muscle, was determined from the motor cortex target obtained in the left hemisphere with MR imaging. The resting motor threshold (RMT) was defined for each subject as the minimal intensity of stimulator output capable of inducing MEPs > 50 μ V peak-to-peak amplitude in at least 6 of 10 consecutive trials (Grosbras and Paus, 2002; Mottaghy et al., 1999). Motor thresholds ranged between 40 and 70% of maximum stimulator output (mean $59 \pm 3\%$).

The TMS generator was connected to a PC running the E-Prime software to trigger and to control the timing of the trains of stimulation. The experiment consisted in the presentation via earphones of auditory events synchronized with the TMS trains (Fig. 2). The audio stimuli were identical to those used in the fMRI acquisition; only silence periods were suppressed because of their noninterest.

Before TMS experiment, subjects performed a practice block without any stimulation to achieve stability of responses. Thereafter, subjects received 10 min TMS active stimulation or placebo stimulation (1-Hz for 600 s at 10% above the subject's motor threshold) applied to one of the target sites (Wernicke or F3Op), in a randomized manner.

The order of target sites and stimulation mode (i.e., active or placebo) were randomized across subjects: the coil was placed over each of the two sites in turn, changing site or stimulation mode at the end of each block. Thus, subjects were administered four stimulation blocks of 10 min; each one was followed by a 15-min rest interval to avoid carry-over effects from one block to the next (Knecht et al., 2003; Rossi et al., 2000).

Statistical analysis of behavioral data

Statistical analyses were performed with the use of StatView® 5.0 (SAS Institute, USA). Reaction times (RTs) and error rate (ER) were chosen as dependent variables for analysis of the effects of TMS on task performance (Devlin et al., 2003; Nixon et al., 2004). Outlier response times, i.e., responses higher than 3000 ms, were discarded from analyses. For all subjects, we calculated the percentage of correct response (accuracy) and the mean reaction

time (RT) for each TMS condition. We performed two repeated-measures ANOVAs with three within-subject factors: location (Wernicke, F3Op), language (French, foreign languages) and stimulation mode (active, placebo) to analyze accuracy and RT. In order to investigate the effect of TMS on RT in each location, two other ANOVAs were performed separately for each target site (Wernicke, F3Op). We used unpaired *t* tests for post hoc analysis.

Results

fMRI results

The statistical parametrical map (SPM) reflecting the thumb movement activity allowed detection of a clear activation of the left motor cortex (Brodmann area 4) in each subject.

Posterior sylvian clusters of activation resulting from the *French minus foreign languages* contrast were detected in each subject (see Table 1). Furthermore, in order to assess inter-individual variability in the whole sample, mean fMRI activations were mapped into Talairach's space. Significant activation was detected in the left superior temporal gyrus in 6 subjects (mean \pm SD peak voxel *x, y, z* Talairach's coordinates in mm: $-49 \pm 7, -44 \pm 7, -14 \pm 4$; $Z > 6.2$; extent: 976 ± 1696 voxels), and in the left middle temporal gyrus in 9 subjects (mean peak voxel at $-53 \pm 4, -41 \pm 9, -0.7 \pm 4$; $Z > 5.85$; extent 3216 ± 4829 voxels). Group peak voxel Talairach's coordinates of Wernicke's area were *x, y, z*: $51 \pm 5, -45 \pm 8, 9 \pm 8$ ($Z > 5.85$; extent 3701 ± 4679 voxels); maximum inter-subject variations reached 17, 24 and 18 mm, respectively. In addition, the anterior cingulate was activated in nine subjects (mean peak voxel at $-2 \pm 4, 3 \pm 6, 48 \pm 6$; $Z > 6.18$; extent: 2682 ± 3546 voxels). Activation was also observed in the left inferior frontal gyrus (IFG; Broca's area) in eight subjects, including the pars opercularis and/or the pars triangularis. Five subjects activated the F3Op (mean peak voxel at $-50 \pm 3, 12 \pm 3, 17 \pm 8$; $Z > 6.1$; extent: 1930 ± 4065 voxels) and three subjects activated the PTr (mean peak voxel at $-44 \pm 4, 20 \pm 8, 18 \pm 9$; $Z > 6.4$; extent 512 ± 1615 voxels). The right middle temporal gyrus was activated in four subjects only (mean peak voxel at $48 \pm 5, -21 \pm 17, -6 \pm 9$; $Z > 5.9$; extent: 640 ± 1224 voxels).

TMS effects on the targeted language areas

One subject could not tolerate active stimulation, which triggered a panic attack, and was excluded from the TMS data analysis. Two subjects reported discomfort caused by active rTMS over F3Op (e.g. blinking of the eyes, squeaking of the teeth); however, their mean RT for this site [(1080 and 1322 ms) for French language and (1203 and 1834 ms) for foreign languages] was within the RT range of the subjects' sample [(794 to 1449 ms) for French language and (1098 to 1904 ms) for foreign languages].

Regarding general effects, a significant effect of language was detected for RT ($F_{(1,10)} = 120.35, P < 0.0001$) and for accuracy ($F_{(1,10)} = 49.19, P < 0.0001$). There was a significant main effect of stimulation mode on RTs ($F_{(1,10)} = 10.85, P < 0.008$); RT of active stimulation was faster (1229 ± 46 ms) relative to placebo stimulation (1339 ± 46 ms). Also, there was an interaction between stimulation mode and language ($F_{(1,10)} = 7.61, P < 0.02$). Moreover, this analysis showed a trend towards significance for the interaction between location, stimulation mode and language ($F_{(1,10)} = 4.23, P = 0.06$).

Table 1

Individual characteristics of posterior language area detected for French vs. foreign languages stimuli (height threshold $P < 0.0001$ corrected for multiple comparisons)

Subjects	Talairach's coordinates (mm)			Region	BA	Voxel level		Cluster level
	x	y	z			t value	SPM (Z)	Extent (mm ³)
S1	-46	-42	18	L. STG	22	13.75	$Z > 8$	16,256
S2	-50	-42	-2	L. MTG	21	11.33	$Z > 8$	7424
S3	-50	-46	18	L. STG	22	9.09	$Z > 8$	2432
S4	-60	-34	-6	L. MTG	22	6.00	5.85	64
S5	-46	-54	18	L. STG	21	10.40	$Z > 8$	1920
S6	-50	-58	4	L. MTG	21	7.57	7.29	2688
S7	-60	-38	12	L. STG	22	9.09	$Z > 8$	1280
S8	-50	-46	1	L. MTG	21	6.63	6.44	320
S9	-43	-49	11	L. STG	22	8.41	$Z > 8$	5632
S10	-53	-38	4	L. MTG	22	6.71	6.51	384
S11	-57	-57	18	L. MTG	22	7.56	7.28	5952
S12	-50	-38	12	L. STG	22	6.38	6.20	64

BA: Brodmann's area.

L.STG = posterior part of the left superior temporal gyrus.

L.MTG = posterior part of the left middle temporal gyrus.

Regarding the effects on reaction time of rTMS in Wernicke's and Broca's areas, ANOVA conducted for each site showed that relative to placebo stimulation, active rTMS over Wernicke's area induced a significant decrease in RTs ($F_{(1,10)} = 19.04$, $P = 0.001$) with a mean decrease of 199 ± 39 ms for French language and 110 ± 40 ms for foreign languages (Fig. 4). Moreover, a significant interaction of stimulation mode \times language ($F_{(1,10)} = 7.88$, $P = 0.01$) indicated that active stimulation on Wernicke's area reduced RT for French language more than RT for foreign languages.

Over the F3Op area, the stimulation mode effect was not significant ($F_{(1,10)} = 1.04$, $P = 0.33$, ns) with a mean decrease of 55 ± 38 ms, and there was no interaction between stimulation mode and language ($F_{(1,10)} = 0.007$, $P = 0.94$, ns) with a mean decrease of 54 ± 55 ms for French language and 56 ± 57 ms for foreign languages.

Error rate was not significantly modified by TMS, either in the stimulation mode ($F_{(1,10)} = 0.35$, $P = 0.56$, ns) or in the stimulation location ($F_{(1,10)} = 0.03$, $P = 0.87$, ns) comparisons.

Finally, post hoc analyzes showed that baselines (i.e., placebo stimulations over the two sites) did not differ (paired t test: $t > 1.005$, $P = 0.33$).

Discussion

Low-frequency rTMS applied over Wernicke's area resulted in a decreased reaction-time response to a language-fragment-detection task. This facilitatory effect appeared to be specific for stimulation of Wernicke's area, as we did not observe any change after stimulation of F3Op area. These findings substantiate the view that posterior temporal activations detected in each subject with fMRI reflect the contribution of posterior temporal areas to the present language task. They also illustrate the feasibility of individual fMRI determination of cortical language-related targets for neuronavigated rTMS.

rTMS on Wernicke's area had an effect on the latencies of the language-fragment-detection task, which resulted in a shortening of response time without affecting the accuracy of the response. This significant decrease in RT observed for TMS over Wernicke's area was approximately three times larger than the decrease reported by other studies, which also stimulated Wernicke's area without individual anatomical localization (Mottaghy et al., 1999; Topper et al., 1998). Wernicke's area stimulation was associated with a response time significantly shorter for French than for foreign

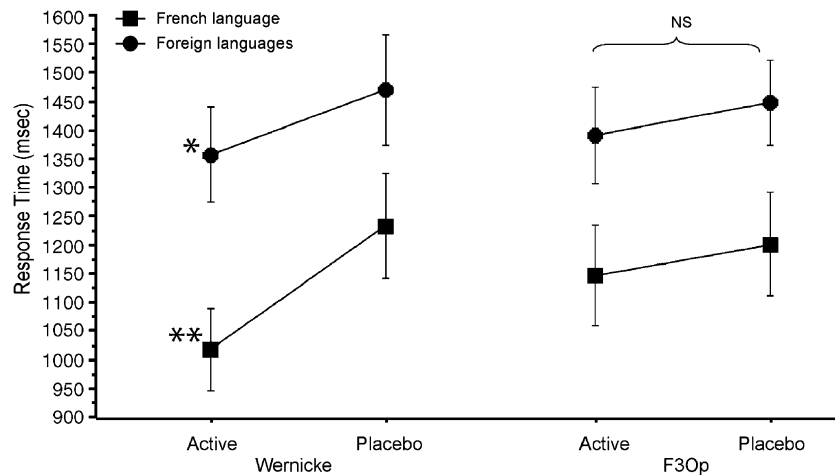


Fig. 4. Mean response time pooled for 11 individuals. Active coil compared to placebo coil: $*F_{(1,10)} = F = 6.39$ ($P = 0.03$), for foreign languages and $**F_{(1,10)} = 30.281$ ($P = 0.0003$), for French language. NS = others conditions showed no significant effect. Error bars indicate standard error.

languages, consistently with the involvement of this region in semantic processes (Malogiannis et al., 2003). This behavioral gain of performance for the native language was not detected for F3Op stimulation.

Absence of TMS effect on Broca's area has previously been reported (Mottaghy et al., 1999; Sparing et al., 2001) and might be related to the paradigm used. Indeed, functional neuroimaging studies have rather attributed the cortical activations of F3Op to phonological processing or to motor program for speech generation (e.g. Zatorre et al., 1996; Price, 2000) than to language identification as in the task used here.

We found that low-frequency rTMS over Wernicke's area speeded reaction times and consequently can lead to a behavioral gain for the language-fragment-detection task performance. The factors underlying a facilitatory effect of rTMS are poorly known. A facilitatory effect has previously been reported with high-frequency rTMS over language areas, rather than with low-frequency rTMS. The facilitatory effect with high-frequency is not always observed, as it can vary with the characteristics of the stimulation parameters, such as duration and intensity (Chen et al., 1997; Pascual-Leone et al., 1998); it can even be reversed to a slowing effect. Indeed, two recent studies have combined fMRI and TMS to investigate various durations of stimulation and have reported either acceleration or slowing of RT during a language task (Devlin et al., 2003; Nixon et al., 2004).

Low-frequency rTMS has been demonstrated to decrease motor cortex excitability, providing evidence that low-frequency pulses applied to the motor cortex can have an inhibitory effect in this area (Chen et al., 1997; Wassermann, 1998). The neurophysiologic mechanisms responsible for the changes in cortical excitability in other cerebral regions are still undetermined.

Thus, we can only speculate that a putative mechanism accounting for enhanced performance when disrupting Wernicke's area could be the change in excitability of a site functionally interconnected with Wernicke's area. In line with this hypothesis, rTMS would have modified the trade-off between semantic and phonologic processes. Low rTMS over Wernicke's would inhibit semantic processes, indirectly accelerating the information transfer to other sites implementing the phonological processes engaged during the sound-fragment-detection-task. Alternatively, our results might be in agreement with a recent low-frequency rTMS study, showing improved motor performance by suppressing intracortical inhibition (Kobayashi et al., 2004). However, the present study was not designed to disentangle whether the decreased RT results from an excitatory process, or from a removal of ongoing inhibition.

Although it may engage comprehension processes, the language-fragment-detection task used here probably did not constraint the subjects' linguistic activity exclusively to semantic processes. The individual activation maps derived from the contrast of *French minus foreign* trials likely reflect areas mainly engaged in semantic processes, although not exclusively. Indeed, each trial included during 3 s either a maternal language sentence automatically engaging semantic processes or a foreign sentence incomprehensible to the subjects. The remaining time (4 s) of each trial was allocated to the fragment-detection task.

We observed that the group activation maps resulting from the image subtraction *French minus foreign languages* involved the middle and superior temporal gyrus, the left inferior frontal gyrus (IFG) and the right superior temporal region. These regions have been involved in both phonologic and semantic processes. They have been reported in a word comprehension task (Malogiannis et

al., 2003), in a phonologic and semantic task (Binder et al., 1997) and in an auditory language comprehension task (Friederici, 2002). However, we are not aware of any fMRI paradigm that allows for detecting activations related to semantic processes only, with a signal to noise ratio sufficient for individual localization at 1.5 T. In the present study, we rather used the language-fragment-detection task to determine a TMS targeting procedure based on individual functional image analysis.

The present results indeed highlight the reliability of the language-fragment-detection task for individual localization of language-related targets with event-related fMRI. The analysis of individual activation maps provided evidence that the posterior perisylvian region (Brodmann's areas 21, 22) was involved in language processing with a probability of detection up to 100%. The activation of these areas during language comprehension tasks has been reported by other authors with a lower frequency of activation (Seghier et al., 2004; Xiong et al., 2000). The location of the targeted posterior temporal region varied notably between subjects (inter-subject maximal difference for x , y , z Talairach's axes: 17, 24, 18 mm), and its variance was equivalent to that observed in other language studies (Burton et al., 2001; Seghier et al., 2004; Xiong et al., 2000).

At variance with the posterior regions where activation was detected in each subject, within the inferior frontal gyrus (IFG) only eight subjects activated F3Op or F3Tr. This lower detection rate in the IFG may be accounted for by the co-occurrence of phonologic and semantic stimuli. Indeed, our fMRI task used natural sentences in order to isolate brain areas involved in semantic processing, by subtracting the activation induced by sentences in foreign languages from the activation induced by sentences in French. However, one consequence of this logic subtraction is that some areas, particularly involved in phonologic processing may be subtracted away because of their equal activation by known and unknown languages (Pallier et al., 2003). This difficulty in detecting activations in anterior language areas further motivates the targeting of this region using criteria based on individual morphology. Indeed, F3Op (and F3Tr) boundaries were readily visible on each subject's T1 MR image and more specifically on their 3D cortex rendering.

Limitations due to inter-subject differences that can affect TMS effects may preclude the generalization of the results.

Indeed, the triple interaction between the conditions only yielded a trend towards significance, thus the possibility of a nonspecific effect cannot be excluded. The small size of the present sample might account for this statistical trend, although the number of subjects was analogous to that of other studies (e.g. Devlin et al., 2003; Mottaghy et al., 1999; Sparing et al., 2001). While we cannot dismiss the possibility of a slight nonspecific effect of active TMS leading to a facilitation of task performance, the stimulation mode effect and the interaction between stimulation mode and language were significant in Wernicke's area site only, and were far from significance in F3Op. Also, we were able to preclude the possibility that active rTMS might function as a warning stimulus leading to an increase of attention and thereby resulting in a shorter reaction time, as the noise produced by the placebo stimulator had no impact on response time. Finally, active rTMS effect was not due to a nonspecific local sensation induced by the TMS pulses because no such effect was reported during the stimulation of Wernicke's area.

Uncertainty about the precise coil position at which the TMS effect will be maximal is inherent to the TMS method. While we

controlled for the location of target areas, we did not precisely control for the variable depth of the targeted regions (McConnell et al., 2001; Nixon et al., 2004). Nevertheless, we could check in real-time with the Brainvisa software that both F3Op and Wernicke's targets were 2 cm or less below the coil.

Conclusion

This study investigated language processing using a method combining individual fMRI and rTMS. The results suggest that low-frequency rTMS over Wernicke's area speeds-up response to a task tapping on native language perception in healthy volunteers. This finding may be relevant to consider for research on putative usefulness of rTMS in speech disorders, as it confirms that facilitatory effects are not confined to high-frequency stimulations.

Acknowledgments

J. Andoh is supported by the French Foundation for Medical Research (FRM). R. Comeau is acknowledged for his advice regarding the implementation of the neuronavigation system. The authors thank Dr. T. Paus (University of Nottingham, UK) for thoughtful comments on the manuscript. The authors thank the healthy subjects who participated in the study.

References

- Anand, S., Hotson, J., 2002. Transcranial magnetic stimulation: neurophysiological applications and safety. *Brain Cogn.* 50, 366–386.
- Aziz-Zadeh, L., Iacoboni, M., Zaidel, E., Wilson, S., Mazziotta, J., 2004. Left hemisphere motor facilitation in response to manual action sounds. *Eur. J. Neurosci.* 19, 2609–2612.
- Binder, J.R., Frost, J.A., Hammeke, T.A., Cox, R.W., Rao, S.M., Prieto, T., 1997. Human brain language areas identified by functional magnetic resonance imaging. *J. Neurosci.* 17, 353–362.
- Bohning, D.E., Shastri, A., McConnell, K.A., Nahas, Z., Lorberbaum, J.P., Roberts, D.R., Teneback, C., Vincent, D.J., George, M.S., 1999. A combined TMS/fMRI study of intensity-dependent TMS over motor cortex. *Biol. Psychiatry* 45, 385–394.
- Broca, P.M., 1861. Remarques sur le siège de la faculté du langage articulé, suivies d'une observation d'aphémie (perte de la parole). *Bull. Mem. Soc. Anat. Paris* 36, 330–357.
- Burton, M.W., Noll, D.C., Small, S.L., 2001. The anatomy of auditory word processing: individual variability. *Brain Lang.* 77, 119–131.
- Chen, R., Gerloff, C., Classen, J., Wassermann, E.M., Hallett, M., Cohen, L.G., 1997. Safety of different inter-train intervals for repetitive transcranial magnetic stimulation and recommendations for safe ranges of stimulation parameters. *Electroencephalogr. Clin. Neurophysiol.* 105, 415–421.
- Claus, D., Weis, M., Treig, T., Lang, C., Eichhorn, K.F., Sembach, O., 1993. Influence of repetitive magnetic stimuli on verbal comprehension. *J. Neurol.* 240, 149–150.
- Devlin, J.T., Matthews, P.M., Rushworth, M.F., 2003. Semantic processing in the left inferior prefrontal cortex: a combined functional magnetic resonance imaging and transcranial magnetic stimulation study. *J. Cogn. Neurosci.* 15, 71–84.
- Duvernoy, H.M., 1999. *The Human Brain Surface, Three-Dimensional Sectional Anatomy with MRI, and Blood Supply*. Springer-Verlag, New York.
- Epstein, C.M., 1998. Transcranial magnetic stimulation: language function. *J. Clin. Neurophysiol.* 15, 325–332.
- Friederici, A.D., 2002. Towards a neural basis of auditory sentence processing. *Trends Cogn. Sci.* 6, 78–84.
- Grosbras, M.H., Paus, T., 2002. Transcranial magnetic stimulation of the human frontal eye field: effects on visual perception and attention. *J. Cogn. Neurosci.* 14, 1109–1120.
- Herwig, U., Schonfeldt-Lecuona, C., Wunderlich, A.P., von Tiesenhäuser, C., Thielscher, A., Walter, H., Spitzer, M., 2001. The navigation of transcranial magnetic stimulation. *Psychiatry Res.* 108, 123–131.
- Hoffman, R.E., Hawkins, K.A., Gueorguieva, R., Gueorguieva, R., Boutros, N.N., Rachid, F., Carroll, K., Krystal, J.H., 2003. Transcranial magnetic stimulation of left temporoparietal cortex and medication-resistant auditory hallucinations. *Arch. Gen. Psychiatry* 60, 49–56.
- Knecht, S., Floel, A., Dräger, B., Breitenstein, C., Sommer, J., Henningsen, H., Ringelstein, E.B., Pascual-Leone, A., 2002. Degree of language lateralization determines susceptibility to unilateral brain lesions. *Nat. Neurosci.* 5, 695–699.
- Knecht, S., Ellger, T., Breitenstein, C., Bernd Ringelstein, E., Henningsen, H., 2003. Changing cortical excitability with low-frequency transcranial magnetic stimulation can induce sustained disruption of tactile perception. *Biol. Psychiatry* 53, 175–179.
- Kobayashi, M., Hutchinson, S., Theoret, H., Schlaug, G., Pascual-Leone, A., 2004. Repetitive TMS of the motor cortex improves ipsilateral sequential simple finger movements. *Neurology* 62, 91–98.
- Luke, K.K., Liu, H.L., Wai, Y.Y., Wan, Y.L., Tan, L.H., 2002. Functional anatomy of syntactic and semantic processing in language comprehension. *Hum. Brain Mapp.* 16, 133–145.
- Maeda, F., Keenan, J.P., Tormos, J.M., Topka, H., Pascual-Leone, A., 2000. Interindividual variability of the modulatory effects of repetitive transcranial magnetic stimulation on cortical excitability. *Exp. Brain Res.* 133, 425–430.
- Maeda, F., Gangitano, M., Thall, M., Pascual-Leone, A., 2002. Inter- and intra-individual variability of paired-pulse curves with transcranial magnetic stimulation (TMS). *Clin. Neurophysiol.* 113, 376–382.
- Malogiannis, I.A., Valaki, C., Smyrnis, N., Papathanasiou, M., Evdokimidis, I., Baras, P., Mantas, A., Kelekis, D., Christodoulou, G.N., 2003. Functional magnetic resonance imaging (fMRI) during a language comprehension task. *J. Neurolinguist.* 16, 407–416.
- Mangin, J.F., Rivière, D., Cachia, A., Duchesnay, E., Cointepas, Y., Papadopoulos-Orfanos, D., Scifo, P., Ochiai, T., Brunelle, F., Regis, J., 2004. A framework to study the cortical folding patterns. *Neuroimage* 23, S129–S138.
- Matthews, P.M., Adcock, J., Chen, Y., Fu, S., Devlin, J.T., Rushworth, M.F., Smith, S., Beckmann, C., Iversen, S., 2003. Towards understanding language organization in the brain using fMRI. *Hum. Brain Mapp.* 18, 239–247.
- McConnell, K.A., Nahas, Z., Shastri, A., Lorberbaum, J.P., Kozel, F.A., Bohning, D.E., George, M.S., 2001. The transcranial magnetic stimulation motor threshold depends on the distance from coil to underlying cortex: a replication in healthy adults comparing two methods of assessing the distance to cortex. *Biol. Psychiatry* 49, 454–459.
- McDermott, K.B., Petersen, S.E., Watson, J.M., Ojemann, J.G., 2003. A procedure for identifying regions preferentially activated by attention to semantic and phonological relations using functional magnetic resonance imaging. *Neuropsychologia* 41, 293–303.
- Mottaghy, F.M., Hungs, M., Brugmann, M., Sparing, R., Boroojerdi, B., Foltys, H., Huber, W., Topper, R., 1999. Facilitation of picture naming after repetitive transcranial magnetic stimulation. *Neurology* 53, 1806–1812.
- Nixon, P., Lazarova, J., Hodinott-Hill, I., Gough, P., Passingham, R., 2004. The inferior frontal gyrus and phonological processing: an investigation using rTMS. *J. Cogn. Neurosci.* 16, 289–300.
- Pallier, C., Dehaene, S., Poline, J.B., LeBihan, D., Argenti, A.M., Dupoux, E., Mehler, J., 2003. Brain imaging of language plasticity

- in adopted adults: can a second language replace the first? *Cereb. Cortex* 13, 155–161.
- Pascual-Leone, A., Tormos, J.M., Keenan, J., Tarazona, F., Canete, C., Catala, M.D., 1998. Study and modulation of human cortical excitability with transcranial magnetic stimulation. *J. Clin. Neurophysiol.* 15, 333–343.
- Pascual-Leone, A., Walsh, V., Rothwell, J., 2000. Transcranial magnetic stimulation in cognitive neuroscience—virtual lesion, chronometry, and functional connectivity. *Curr. Opin. Neurobiol.* 10, 232–237.
- Peelle, J.E., McMillan, C., Moore, P., Grossman, M., Wingfield, A., 2004. Dissociable patterns of brain activity during comprehension of rapid and syntactically complex speech: evidence from fMRI. *Brain Lang.* 91, 315–325.
- Price, C.J., 2000. The anatomy of language: contributions from functional neuroimaging. *J. Anat.* 197, 335–359.
- Rizzo, V., Siebner, H.R., Modugno, N., Pesenti, A., Munchau, A., Gerschlagel, W., Webb, R.M., Rothwell, J.C., 2004. Shaping the excitability of human motor cortex with premotor rTMS. *J. Physiol.* 554, 483–495.
- Rossi, S., Pasqualetti, P., Rossini, P.M., Feige, B., Olivelli, M., Glocker, F.X., Battistini, N., Lucking, C.H., Kristeva-Feige, R., 2000. Effects of repetitive transcranial magnetic stimulation on movement-related cortical activity in humans. *Cereb. Cortex* 10, 802–808.
- Sakai, K.L., Noguchi, Y., Takeuchi, T., Watanabe, E., 2002. Selective priming of syntactic processing by event-related transcranial magnetic stimulation of Broca's area. *Neuron* 35, 1177–1182.
- Seghier, M.L., Lazeyras, F., Pegna, A.J., Annoni, J.M., Zimine, I., Mayer, E., Michel, C.M., Khateb, A., 2004. Variability of fMRI activation during a phonological and semantic language task in healthy subjects. *Hum. Brain Mapp.* 23, 140–155.
- Sparing, R., Mottaghy, F.M., Hungs, M., Brugmann, M., Foltys, H., Huber, W., Topper, R., 2001. Repetitive transcranial magnetic stimulation effects on language function depend on the stimulation parameters. *J. Clin. Neurophysiol.* 18, 326–330.
- Stewart, L., Ellison, A., Walsh, V., Cowey, A., 2001. The role of transcranial magnetic stimulation (TMS) in studies of vision, attention and cognition. *Acta Psychol. (Amst)* 107, 275–291.
- Talairach, J., Tournoux, P., 1988. *Co-Planar Stereotaxic Atlas of the Human Brain*. Thieme Medical Publisher, New York.
- Tomaiuolo, F., MacDonald, J.D., Caramanos, Z., Posner, G., Chiavaras, M., Evans, A.C., Petrides, M., 1999. Morphology, morphometry and probability mapping of the pars opercularis of the inferior frontal gyrus: an in vivo MRI analysis. *Eur. J. Neurosci.* 11, 3033–3046.
- Topper, R., Mottaghy, F.M., Brugmann, M., Noth, J., Huber, W., 1998. Facilitation of picture naming by focal transcranial magnetic stimulation of Wernicke's area. *Exp. Brain Res.* 121, 371–378.
- Wassermann, E.M., 1998. Risk and safety of repetitive transcranial magnetic stimulation: report and suggested guidelines from the International Workshop on the Safety of Repetitive Transcranial Magnetic Stimulation, June 5–7, 1996. *Electroencephalogr. Clin. Neurophysiol.* 108, 1–16.
- Wassermann, E.M., Grafman, J., Berry, C., Hollnagel, C., Wild, K., Clark, K., Hallett, M., 1996. Use and safety of a new repetitive transcranial magnetic stimulator. *Electroencephalogr. Clin. Neurophysiol.* 101, 412–417.
- Watkins, K., Paus, T., 2004. Modulation of motor excitability during speech perception: the role of Broca's area. *J. Cogn. Neurosci.* 16, 978–987.
- Watkins, K.E., Strafella, A.P., Paus, T., 2003. Seeing and hearing speech excites the motor system involved in speech production. *Neuropsychologia* 41, 989–994.
- Wernicke, C., 1874. *Der Aphasische Symptomenkomplex*. Cohn and Weigert, Breslau.
- Xiong, J., Rao, S., Jerabek, P., Zamarripa, F., Woldorff, M., Lancaster, J., Fox, P.T., 2000. Intersubject variability in cortical activations during a complex language task. *Neuroimage* 12, 326–339.
- Zatorre, R.J., Meyer, E., Gjedde, A., Evans, A.C., 1996. PET studies of phonetic processing of speech: review, replication, and reanalysis. *Cereb. Cortex* 6, 21–30.