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# Intermittent Theta Burst Stimulation (iTBS) for Treatment of Chronic Post-Stroke Aphasia: Results of a Pilot Randomized, Double-Blind, Sham-Controlled Trial

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**Background:** Research indicates intermittent theta burst stimulation (iTBS) is a potential treatment of post-stroke aphasia.

**Material/Methods:** In this double-blind, sham-controlled trial (NCT 01512264) participants were randomized to receive 3 weeks of sham ( $G_0$ ), 1 week of iTBS/2 weeks of sham ( $G_1$ ), 2 weeks of iTBS/1 week of sham ( $G_2$ ), or 3 weeks of iTBS ( $G_3$ ). fMRI localized residual language function in the left hemisphere; iTBS was applied to the maximum fMRI activation in the residual language cortex in the left frontal lobe. fMRI and aphasia testing were conducted pre-treatment, at  $\leq 1$  week after completing treatment, and at 3 months follow-up.

**Results:** 27/36 participants completed the trial. We compared  $G_0$  to each of the individual treatment group and to all iTBS treatment groups combined ( $G_{1-3}$ ). In individual groups, participants gained (of moderate or large effect sizes; some significant at  $P < 0.05$ ) on the Boston Naming Test (BNT), the Semantic Fluency Test (SFT), and the Aphasia Quotient of the Western Aphasia Battery-Revised (WAB-R AQ). In  $G_{1-3}$ , BNT, and SFT improved immediately after treatment, while the WAB-R AQ improved at 3 months. Compared to  $G_0$ , the other groups showed greater fMRI activation in both hemispheres and non-significant increases in language lateralization to the left hemisphere. Changes in IFG connectivity were noted with iTBS, showing differences between time-points, with some of them correlating with the behavioral measures.

**Conclusions:** The results of this pilot trial support the hypothesis that iTBS applied to the ipsilesional hemisphere can improve aphasia and result in cortical plasticity.

**Keywords:** Aphasia • Language • Magnetic Resonance Imaging • Rehabilitation • Transcranial Magnetic Stimulation

Full-text PDF: <https://www.medscimonit.com/abstract/index/idArt/931468>

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## Background

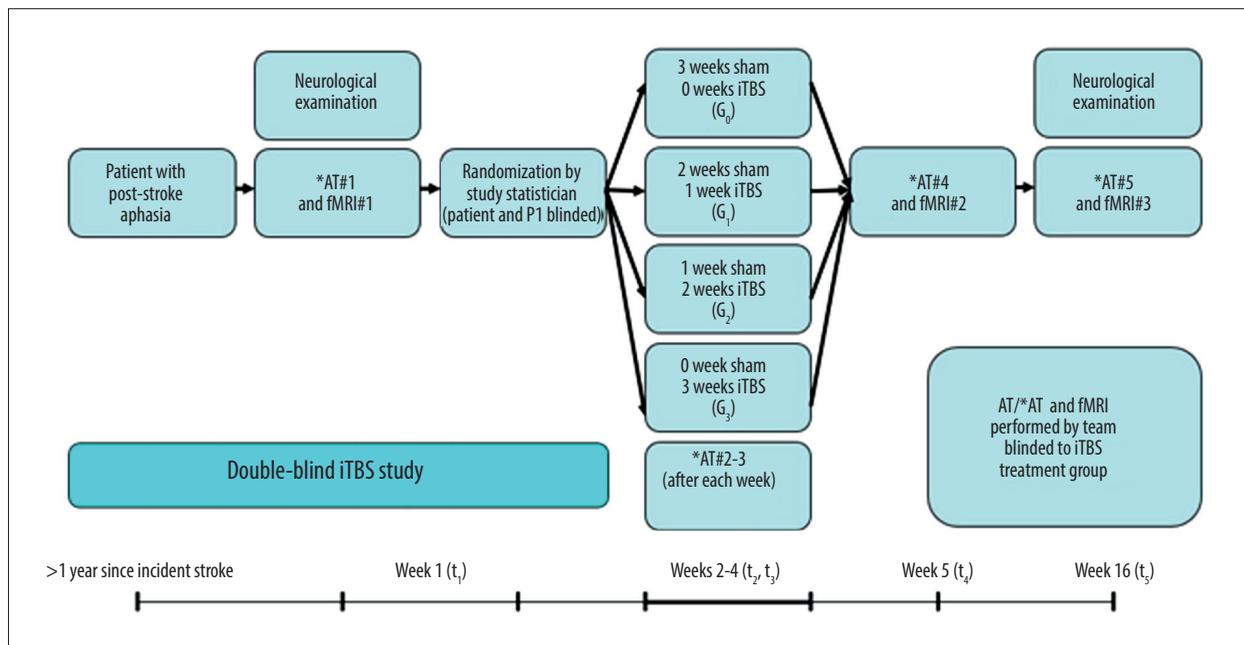
Approximately 30% of the ~800 000 people who suffer from ischemic stroke each year present with aphasia [1]. Stroke with aphasia is more costly than stroke without aphasia, with an additional annual cost in excess of \$2B [2]. The time course and the degree of post-stroke aphasia recovery have been examined in several short- and intermediate-term follow-up studies [3]. Although the subacute post-stroke period is typically associated with spontaneous recovery, minimal or no spontaneous recovery of aphasia is expected beyond the first 6-12 months after presentation [4,5]. Occasionally, aphasia may recover several years after stroke [6,7]. In a recent individual participant data meta-analysis, several factors have been shown to significantly affect outcomes of post-stroke aphasia rehabilitation, including younger age at stroke occurrence and better baseline performance on Boston Naming Test (BNT), Western Aphasia Battery Aphasia Quotient (WAB AQ), and Aachen Aphasia Test (AAT) Spontaneous-Speech Communication subscale [8]. The presence of persistent aphasia is frequently noted as a primary cause of post-stroke social isolation, struggles with mood and depression, and perceived or real cognitive impairments, all of which significantly reduce quality of life and resumption of pre-stroke life activities [9].

Recently, non-invasive brain stimulation has received interest as an intervention for improving chronic post-stroke aphasia and for priming the brain prior to behavioral interventions [10-12]. More specifically, neurostimulation is thought to facilitate neuroplasticity through either changing the perilesional canonical language networks or recruiting compensatory networks [12,13]. In other words, the post-stroke recovery takes advantage of either other networks adapting to perform damaged functions or networks that were dormant prior to stroke activating in the face of increased difficulty in performing a task because of the stroke-related damage [7]. In the realm of neurostimulation, excitatory repetitive transcranial magnetic stimulation (rTMS), which exerts its effects via decreased local GABA-ergic inhibition and increased direct long-term potentiation [12], has been shown to enable language processing in health and disease, including post-stroke aphasia [12]. The use of fMRI to guide selecting the site of stimulation is another advance for administering rTMS. However, another approach is to apply rTMS to the unaffected (right) hemispheric language homologue (see [12] for an extensive review). In a neurostimulation study, stimulating the affected (20Hz) and unaffected (1Hz) hemispheres sequentially followed by rehabilitation for 10 days resulted in an improvement on several measures of aphasia when compared to sham [14]. Similarly, substantial linguistic gains followed applying 1Hz rTMS to the area of maximum fMRI activation in either the left or right hemisphere prior to a 10-day behavioral intervention in participants with fluent and non-fluent aphasia [15].

Despite the therapeutic potential, many participants report adverse effects from rTMS, including headache, muscle twitches, and residual local hypersensitivity [12,13]. A recent development in the realm of neurostimulation is intermittent theta burst stimulation (iTBS) [16-18]. This typically more comfortable and better tolerated form of neurostimulation mimics the electrical firing of the hippocampus that underlies long-term potentiation [17,19]. Based on its mechanism of action, iTBS has high relevance to learning and memory by affecting synaptic plasticity via producing consistent, long-lasting, and powerful effects on behavior and physiology after an application period of 20 to 190 s [19]. However, it is well recognized that excessive or prolonged stimulation can have the reverse effect [17]. Similar to conventional rTMS, iTBS induces changes in the underlying cortex for ~60 min that are associated with benefits such as motor improvement [20,21].

Recently, we have shown that 10 sessions of fMRI-guided iTBS applied to the peristroke region showing maximum language fMRI activation, typically at or near the inferior frontal gyrus (IFG), changed fMRI activation patterns and was associated with both improved communication skills and a trend towards improved aphasia testing (AT); the participants did not report any adverse events [22]. Additionally, fMRI-guided iTBS applied to the peristroke language activation area followed by constraint-induced aphasia therapy (CIAT) resulted in significant fMRI changes and communication gains [18]. iTBS has been also shown to change brain structure and function when applied to the perilesional language area [18,22-24]. Together, these studies support the hypothesis that iTBS applied to the peristroke regions is safe, well-tolerated, not associated with severe adverse events (eg, seizures), and may be associated with linguistic gains through its facilitation of the perilesional canonical language networks; these effects may be independent of or occur in conjunction with cognitive rehabilitation [18,25,26]. Clinical trial data are needed to promote translation of these discoveries into practice.

The aim of the present study was to demonstrate the feasibility of conducting a pilot randomized, double-blind, sham-controlled trial comparing the effects of sole fMRI-guided iTBS to sole sham iTBS on chronic aphasia in patients with a single left (dominant) hemispheric ischemic stroke, and to determine whether any observed initial treatment response would be sustained. We specifically excluded providing cognitive intervention in this trial (ie, using iTBS as a primer for cognitive intervention); a separate open-label study was recently completed by our group to evaluate the potential efficacy of combination of iTBS and constraint-induced aphasia therapy [18,25]. We also aimed to assess the relationship between the duration of iTBS treatment (1 vs 2 vs 3 weeks) and the linguistic and fMRI outcomes. We hypothesized that iTBS alone, when compared to sham alone treatment, would



**Figure 1.** Diagram of the randomized, double-blind, sham-controlled treatment protocol, and associated testing (\*AT – aphasia testing/ AT – reflects AT without obtaining the Aphasia Quotient (AQ) of the Western Aphasia Battery-Revised (WAB-R), iTBS – intermittent theta burst stimulation; fMRI – functional magnetic resonance imaging;  $t_{1-5}$  – study time-points;  $G_{0-3}$  – study groups).

improve linguistic performance in patients with aphasia in a dose-dependent fashion.

## Material and Methods

### Participants

Clinicians identified and referred eligible participants from their neurology and rehabilitation clinics. Eligible participants were at least 1 year after a single left, middle cerebral artery (LMCA) ischemic stroke with documented persistent aphasia and had not received speech-language therapy within the 3 months preceding study enrollment. Medical history was confirmed by records review including admission notes and imaging of the brain (CT or MRI). We used the most recent available brain imaging results to confirm the diagnosis and to exclude participants with more than 1 stroke. Potential participants underwent a screening Token Test (TT) and were qualified for study participation if the results showed at least mild aphasia ( $TT \leq 40$ ) [27]. Patients were excluded if they had a history of a neurodegenerative (eg, dementia), metabolic (eg, encephalopathy), or supervening medical disorder (eg, brain tumor or other cancer), history of severe depression or other mental illness, contraindication to 3T MRI, or positive pregnancy test on the day of MRI scanning in women of childbearing age. The Institutional Review Boards of the participating institutions approved the study, and the trial was registered at clinicaltrials.

gov (NCT 01512264). All participants (or their legal representatives if the participants were judged clinically to have impaired speech comprehension) signed the informed consent prior to initiating any study procedures.

### Study Design

Participants were randomized into 1 of 4 therapy groups (Figure 1): Group 0 ( $G_0$ ) received 3 weeks of sham iTBS, Group 1 ( $G_1$ ) received 2 weeks of sham and 1 week of iTBS, Group 2 ( $G_2$ ) received 1 week of sham and 2 weeks of iTBS, and Group 3 ( $G_3$ ) received 3 weeks of iTBS. Randomization envelopes were prepared prior to initiating the study by the study statistician (CJL); envelopes containing group assignment were opened sequentially. All study staff with the exception of the iTBS treatment staff were blinded to group allocation; however, they were blinded to the results of the pre-enrollment ( $t_1$ ) AT. Blinding was maintained until all participants completed the study. Each participant received fMRI and AT within 1 week prior to initiating the intervention (Figure 1;  $t_1$ ), within 1 week following the intervention ( $t_2$ ), and 3 months later ( $t_3$ ). AT was also performed at the end of each treatment week ( $t_2$  and  $t_3$ ). Since the optimal number of therapy sessions needed for the improved language outcomes is unknown, treatment dosing (1, 2, or 3 weeks) was also implemented in the design of the trial.

## Assessments

Due to the pilot nature of the study, we did not select a specific primary outcome measure. Rather, we used a suite of AT to broadly gauge language comprehension and production, and to explore which measures may be most sensitive to iTBS. The Aphasia Quotient (AQ) of the Western Aphasia Battery-Revised (WAB-R) [28] and fMRI were administered at baseline ( $t_1$ ), after intervention ( $t_2$ ), and at 3 months ( $t_3$ ). The Boston Naming Test (BNT) [29], Semantic Fluency Test (SFT) [30], and Controlled Oral Word Association Test (COWAT) [31] were administered at baseline ( $t_1$ ), after each week of treatment ( $t_{2-4}$ ), and at 3 months ( $t_3$ ). For each group, paired samples  $t$  tests were performed to examine changes between time-points, and effect sizes (Cohen's  $d$ ) were computed for each.

## Transcranial Magnetic Stimulation

iTBS sessions were given for 5 consecutive weekdays over 3 weeks, resulting in 15 treatment sessions. Each session involved either stimulation or sham, depending on group assignment. Prior to the first treatment session [22,23], we established the resting (RMT) and active motor (AMT) thresholds by applying a single-pulse TMS to the right hemisphere motor cortex (MRI-guided localization) using a Magstim Rapid2® figure-of-eight coil (Magstim Co., Wales, UK) with EMG leads placed over the first dorsal interosseous (FDI) muscle of the left hand. The stimulation coil was placed tangentially to the skull with the handle parallel to the sagittal axis and over the primary motor cortex in the right (unaffected) hemisphere at the optimal site for obtaining a motor evoked potential in the FDI muscle. After the RMT and AMT were determined from the right hemisphere motor cortex, participants were given a 10-minute break while the iTBS treatment staff opened the randomization envelope that indicated group assignment. Thereafter, iTBS or sham iTBS was performed over the left hemisphere using either the stimulation or sham Magstim Rapid2® coils, respectively, with intensity set at 80% of AMT obtained from the right hemisphere. Stimulation was targeted towards the residual language-responsive cortex in the left frontal lobe, typically at or near the inferior frontal gyrus (IFG). The precise location was based on fMRI results (see below) with the individual peak activation results entered into the Brainsight neuronavigation system (Rogue Research, Inc., Montreal, Canada). Stimulation parameters were selected based on Huang et al [19] and consisted of 600 iTBS pulses, with 3 pulses at 50 Hz given every 200 milliseconds in 2-second trains at 10-second intervals over a 200-second period [22]. Each session took about 10-15 minutes, with participants monitored for adverse events during and after each session.

## Functional MRI Block-design Tasks

Participants completed 2 runs of a well-established semantic decision/tone decision (SDTD) task that was presented in 30-second blocks with 2 alternating conditions: the control (TD, tone decision) and the active condition (SD, semantic decision) [32-34]. Each run included eleven 30-second blocks, starting with a TD block, for a total of 330 seconds. In the TD condition, subjects heard brief sequences of four to seven 500- and 750-Hz tones every 3.75 seconds and responded with a left-hand button press (ie, index finger) to any sequence containing 2 750-Hz tones. In the SD condition, subjects heard spoken English nouns designating animals every 3.75 seconds and responded with the same left-hand button press to stimuli that met 2 criteria of being native to the United States and commonly used by humans (eg, cow). If the presented items did not fulfill the corresponding criteria, participants pressed the "no" button (ie, middle finger) also with the left hand. While performance was measured by recording responses to the SD and TD conditions, the overarching goal was to engage the brain language area that remained functional after the stroke to provide a target for stimulation [18,22].

## MRI Data Acquisition

Data on the initial 3 participants were acquired on a 3.0 Tesla research-dedicated Philips MRI system using an 8-channel coil. For these subjects, EPI fMRI scans were performed using thirty-two 4 mm thick axial slices covering the entire brain. EPI images were obtained using a T2\*-weighted gradient-echo EPI pulse sequence (TR/TE=2000/38 ms, FOV=24.0×24.0 cm, matrix=64×64, slice thickness=4 mm). In addition, a high-resolution T1-weighted three-dimensional anatomical scan was obtained (TR/TE=8.1/3.7 ms, FOV 25.0×21.1×18.0 cm, matrix 252×211, flip angle 8°, slice thickness=1 mm) for localizing brain regions. On the remaining subjects, we performed imaging on research-dedicated 3.0 Tesla MR Siemens systems, initially using a circular polarized head coil (Allegra) and a 20-channel head coil after scanner upgrade (Prisma). Echo planar imaging (EPI) fMRI scans were performed using thirty 4-mm-thick axial slices covering the entire brain. EPI images were obtained using a T2\*-weighted gradient-echo EPI pulse sequence (TR/TE=2000/38 ms, FOV=24.0×24.0 cm, matrix=64×64, slice thickness=4 mm; after scanner upgrade, TR/TE=2000/35 ms, FOV=24.0×24.0 cm, matrix=64×64, slice thickness=4 mm). In addition, a high-resolution T1-weighted three-dimensional anatomical scan was obtained (TR/TE=2300/2.17 ms, FOV 25.6×25.6×19.2 cm, matrix 256×256, flip angle 9°, slice thickness=1 mm; after scanner upgrade, TR/TE=2300/3.37 ms, FOV 25.6×25.6×19.2 cm, matrix 256×256, flip angle 9°, slice thickness=1 mm) for localization of brain regions. For each fMRI run, 165 whole-brain scans were acquired.

## MRI Data Preprocessing and Statistical Analysis

fMRI data preprocessing and modeling were completed using MATLAB toolbox SPM12 (<http://www.fil.ion.ucl.ac.uk/spm/software/spm12/>) [18,35-37]. The processing followed standard steps that included discarding the first 30 seconds of the control block, followed by co-registering and aligning all scans using the *coregister* and *realign* functions in SPM. Unified segmentation computed on the anatomical scan was used to normalize functional scans [38]. Functional scans were then spatially smoothed with an 8 mm full-width half-maximum kernel, and general linear modeling (GLM) was performed using the fMRI time series from the block-design task as boxcar regressor convolved with the canonical hemodynamic response function (HRF). In addition, we utilized the 24-parameter model [39] to regress out head motion effects from the realigned data (ie, 6 head motion parameters, 6 head motion parameters 1 time-point before, and the 12 corresponding squared items) plus 2 regressors accounting for the number of runs. Group random effects were computed using one-sample *t* tests. Data were compared between time-points using paired *t* tests. To avoid confounds from participants' individual brain lesions, combined lesion-frequency maps were used as a mask to exclude lesioned voxels from group statistical analyses. Finally, all imaging data analyses were co-varied for the type of scanner used in this study.

## Lateralization Index (LI)

It is well recognized that chronic stroke directly affects language lateralization [40-42]. The LI is commonly used in functional neuroimaging studies to describe the hemispheric or regional distribution of activations in response to specific tasks (eg, language or memory). It ranges from -1 (pure right-hemispheric) to 1 (pure left-hemispheric) [42,43]. For each map, a threshold was computed using a bootstrap algorithm with values above an internal threshold added together to generate a global value for each hemisphere within a region of interest (mask) [44]. Then, the LI was calculated using the LI-toolbox on contrast maps obtained by combining HRF and derivatives contrasts [45]. Three different atlas-based masks were used for calculating LI, including frontal, cerebellum, and whole-brain (frontal+temporal+parietal) masks [45]. For all masks, voxels within a 20-mm area around the midline (10 mm left and 10 mm right) in an axial plane, and the parts of the ROIs that were affected by the stroke were excluded from analyses [40]. For each group, paired samples *t* tests were performed to examine changes between time-points.

## Lesion-Frequency Maps

A previously developed MATLAB plugin (R2017b, MathWorks) was used to compute the stroke-induced lesion area for each

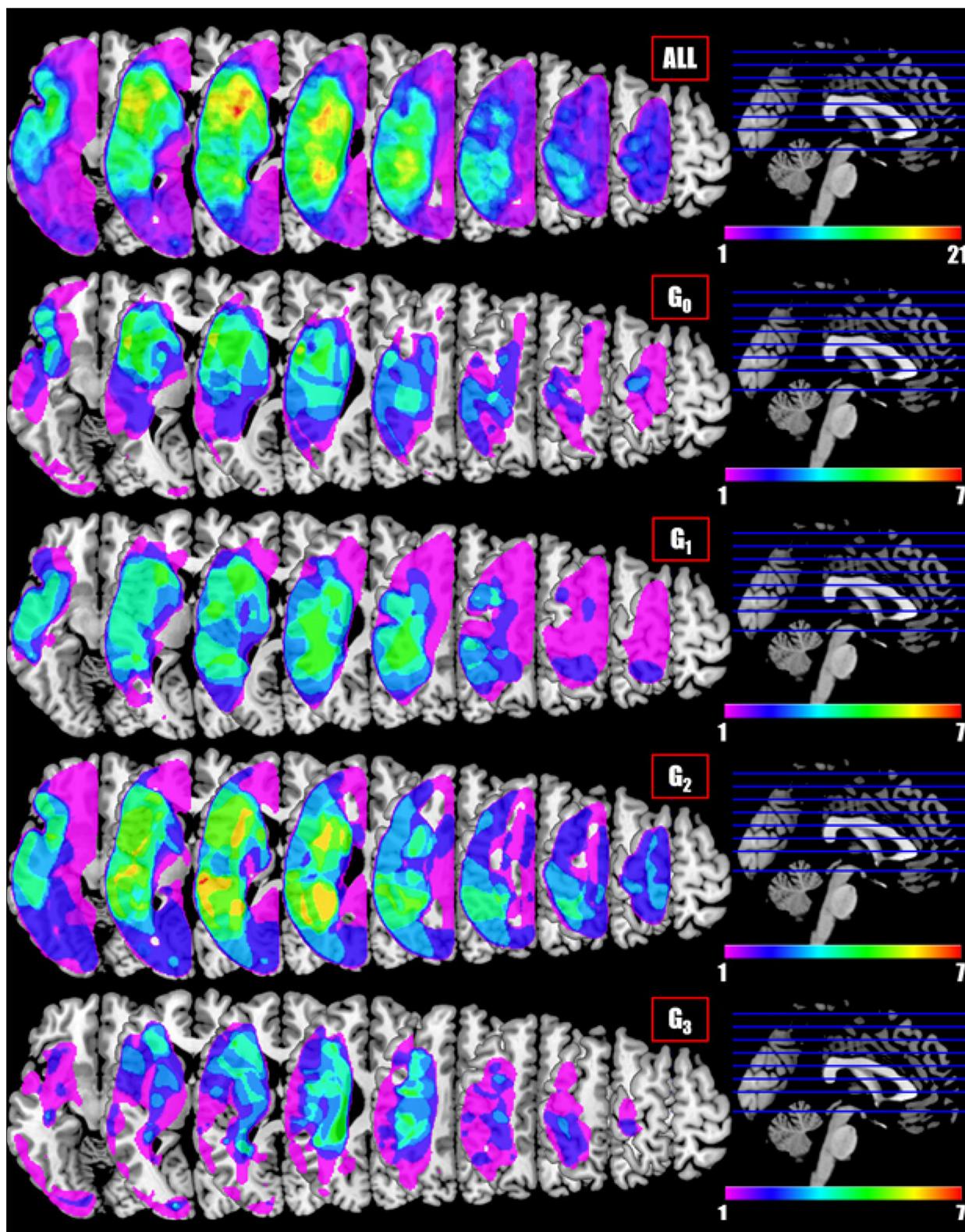
stroke patient [46]. Probabilistic tissue segmentation and image algebra with naïve Bayes classification were used to create feature maps encoding information about missing and abnormal tissue. All maps were binarized then summed into 1 image (separately for each group) in which the value of each voxel represents the frequency of lesion at this particular cortical location (Figure 2).

## Connectivity analysis

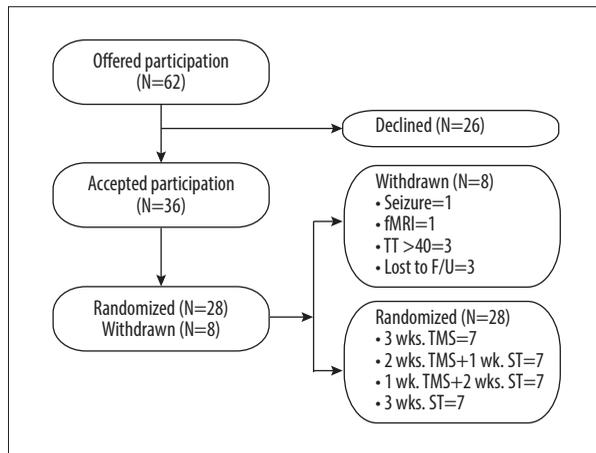
Generalized psychophysiological interaction (gPPI) is an analysis for fMRI data that is conducted between regions of interest, is context-dependent, and is performed to assess dynamic changes over time. Exploratory gPPI analysis was conducted using the toolbox in SPM [47] to model context-specific changes (ie, changes in AT measures) in the relationship between activity in 1 seed brain region and activity in the other brain regions by including a term specifying an interaction effect between the seed region time series and the task time series in each first-level GLM [48]. The gPPI effects are interpreted as changes in interregional connectivity that are driven by psychological states related to factors such as the AT measure [47,49]. This makes gPPI an appropriate tool for testing the hypothesis that longitudinal changes occur in functional connectivity of cognitive probes such as language tasks in response to iTBS. For each participant, the first principal component of the time series from each scan was extracted from the right and left anatomically defined IFG (WFU\_pickatlas toolbox: [https://www.nitrc.org/projects/wfu\\_pickatlas/](https://www.nitrc.org/projects/wfu_pickatlas/)) and entered as a seed time series for the gPPI analysis. Cerebrospinal fluid (CSF) and white-matter signals were included as nuisance variables in the gPPI model in order to reduce the influence of non-neural signals on estimates of task-dependent connectivity [50]. Next, for each participant, gPPI estimates quantifying the level of condition-dependent connectivity from right and left IFG to the rest of the cortex during each session were extracted from the gPPI model into connectivity maps. These connectivity maps were used in paired *t* test analyses to compare connectivity between time-points within stroke groups.

## Relationship Between gPPI Connectivity, LI, and Behavioral Scores

To investigate the relationship between the changes in connectivity and behavior over time, regression analyses were performed in SPM. For each participant and each seed region, a difference in connectivity between pairs of time-points was computed and regressed with a difference of each behavioral score between the same pair of time-points. Results were corrected for multiple comparison (FWE), with significance set at  $P < 0.05$ . Additionally, correlations between changes in LI between time-points and changes in behavioral scores were computed.



**Figure 2.** Lesion maps for the 27 included stroke patients. Each voxel value is the number of participants whose stroke lesion extends to that particular voxel (all pictures in neurological convention – left in the figure corresponds to left in the brain). Top lesion map depicts all participants together, then divided by groups.



**Figure 3.** Consort Statement (fMRI – functional MRI; TT – token test; F/U – follow-up; TMS – transcranial magnetic stimulation; ST – sham treatment).

**Table 1.** Demographic data of stroke participants included in the analyses (N=27).

Subject	Token	Handedness	Gender	Age at scan	TSS(Y)	fMRI sessions	rTMS weeks
PART001	28	Right	F	79	3.4	3	1
PART002	33	Right	M	64.6	14	3	0
PART003	12	Right	F	57.8	13	3	2
PART006	6	Right	M	49.6	2.9	3	3
PART008	9	Right	M	57	2.1	3	2
PART009	21	Right	M	50.7	1.1	3	3
PART010	34	Right	M	43.1	1.3	3	1
PART011	6	Right	F	74	1.65	2	3
PART012	21	Right	F	23.8	2.3	2	0
PART013	10	Right	F	66.6	2.2	3	1
PART014	39	Left	F	61.8	4.4	3	3
PART015	4	Right	M	30	0.9	2	2
PART019	34	Right	F	43.6	2.2	3	0
PART020	33	Right	M	62.1	2.7	3	3
PART021	9	Right	M	46.4	1.7	2	1
PART022	23	Right	M	53.3	1.2	3	2
PART023	9	Right	M	54.6	3.7	3	0
PART024	41	Left	M	44.1	3.3	3	0
PART026	28	Right	M	61.1	9.6	3	3
PART027	24	Right	M	67.4	12.7	2	2
PART028	7	Right	F	78.4	1.9	2	1
PART030	31	Right	M	84.7	1.3	2	1
PART032	39	Right	F	57.2	1.1	3	3
PART033	27	Right	M	54	1	2	2
PART034	12	Right	M	47.3	0.9	3	1
PART035	4	Right	M	46.2	1.2	3	2
PART036	32	Right	M	63	2.2	3	0

TT – Token Test, TSS – Time since stroke. Based on the randomization procedure, the subjects received a variable number of active and sham TMS treatments. The number of active treatment weeks is included in the “rTMS weeks” column.

Results

From 62 potential participants with chronic aphasia resulting from LMCA stroke referred to the study, we recruited 36 (Figure 3 – Consort Statement). Out of the 36 participants, 5 were found not to be eligible based on screening tests, and 3 completed less than 2 assessments and were withdrawn (no fMRI or iTBS were administered to these participants). Of the 28 who were randomized, 1 participant received pre-intervention testing (t<sub>1</sub>) and the intervention, but was unable to complete any of the post-intervention measures and was not included in the final analyses. This was related to the participant travelling to the study site from a great distance and needing to

return home on short notice. Thus, 19/28 participants included in final analysis completed all 3 sessions and 8/28 completed at least the pre- and immediate post-treatments sessions (t<sub>1</sub> and t<sub>4</sub>). The 27 participants included in the analyses are described in Table 1 and Figure 3. Subjects' handedness was determined using the Edinburgh Handedness Inventory [51]. Of the 27 included participants, 25 were right-handed prior to the stroke (handedness index >91) while the other 2 had atypical handedness. All patients had a single LMCA distribution stroke confirmed by review of the MRI or MRI report prior to enrollment.

Table 2A. Behavioral results. Numbers represent the difference of the mean between time-points (TP) for each group using paired samples t tests.

TP		Group 0			Group 1			Group 2			Group 3			Group 123		
		t	p	d	t	p	d	t	p	d	t	p	d	t	p	d
1->2	BNT	-2.53	0.053	-1.03	1.76	0.129	0.66	-2.53	0.052	-1.03	-2.49	<b>0.047*</b>	-0.94	-1.83	0.082	-0.41
	SFT	-2.24	0.076	-0.91	-0.60	0.569	-0.23	-0.33	0.754	-0.14	-0.99	0.362	-0.37	-1.22	0.236	-0.27
	COWAT	-2.42	0.060	-0.99	0.00	1.000	0.00	-0.25	0.816	-0.10	-0.55	0.604	-0.21	-0.57	0.575	-0.13
2->3	BNT	-1.50	0.194	-0.61	-4.80	<b>0.003**</b>	-1.82	0.09	0.933	0.04	-0.95	0.377	-0.36	-2.23	<b>0.039*</b>	-0.51
	SFT	0.43	0.684	0.18	-0.90	0.403	-0.34	-0.26	0.805	-0.11	-0.11	0.916	-0.04	-0.65	0.523	-0.15
	COWAT	-0.43	0.688	-0.17	0.28	0.788	0.11	0.88	0.421	0.36	-1.10	0.314	-0.42	-0.39	0.700	-0.09
3->4	BNT	-1.55	0.182	-0.63	0.14	0.892	0.05	-2.92	0.054	-1.31	-2.49	<b>0.047*</b>	-0.94	-2.36	<b>0.029*</b>	-0.54
	SFT	-0.36	0.735	-0.15	0.76	0.476	0.29	0.17	0.868	0.07	0.08	0.936	0.03	0.43	0.674	0.10
	COWAT	2.91	<b>0.03*</b>	1.19	-0.79	0.457	-0.30	-1.57	0.178	-0.64	-0.59	0.579	-0.22	-1.39	0.181	-0.31
4->5	BNT	2.76	<b>0.050*</b>	1.23	3.58	<b>0.015*</b>	1.46	3.65	<b>0.021*</b>	1.63	2.75	<b>0.040*</b>	1.12	5.38	<b>0.000***</b>	1.30
	SFT	0.24	0.821	0.11	-2.00	0.102	-0.82	0.30	0.778	0.13	-0.07	0.950	-0.03	-0.27	0.792	-0.07
	COWAT	-2.15	0.098	-0.96	1.08	0.328	0.44	0.79	0.472	0.36	1.58	0.176	0.64	1.92	0.073	0.47
1->4	BNT	-4.00	<b>0.010**</b>	-1.63	-1.43	0.203	-0.54	-3.98	<b>0.010*</b>	-1.62	-4.19	<b>0.005**</b>	-1.58	-5.03	<b>0.000***</b>	-1.12
	SFT	-1.71	0.147	-0.70	-0.97	0.368	-0.37	-0.75	0.490	-0.30	-1.82	0.118	-0.69	-2.13	<b>0.046*</b>	-0.48
	COWAT	-1.05	0.341	-0.43	-0.18	0.864	-0.07	-0.74	0.493	-0.30	-1.88	0.109	-0.71	-1.85	0.081	-0.41
1->5	BNT	-3.83	<b>0.018*</b>	-1.71	0.00	1.000	0.00	-0.52	0.628	-0.23	-2.99	<b>0.030*</b>	-1.22	-2.05	0.056	-0.50
	SFT	-0.71	0.518	-0.32	-1.58	0.175	-0.65	0.41	0.704	0.18	-0.90	0.410	-0.37	-0.99	0.338	-0.24
	COWAT	-1.84	0.140	-0.82	0.00	1.000	0.00	0.69	0.530	0.31	0.13	0.899	0.05	0.67	0.513	0.16
TP		t	p	d	t	p	d	t	p	d	t	p	d	t	p	d
4->5	WAB-R AQ	-1.03	0.413	-0.59	-1.84	0.163	-0.92	-0.38	0.730	-0.19	-2.17	0.096	-0.97	-1.98	0.071	-0.55
1->4	WAB-R AQ	0.31	0.776	0.16	-1.94	0.148	-0.97	-0.27	0.800	-0.12	-0.69	0.520	-0.28	-1.71	0.110	-0.44
1->5	WAB-R AQ	0.44	0.689	0.22	-2.30	0.105	-1.15	-2.07	0.107	-0.93	-1.34	0.239	-0.55	-3.19	<b>0.007*</b>	-0.82

Significance is as follow: (\*) P<0.05, (\*\*) P<0.01, (\*\*\*) P<0.001. WAB-R AQ was collected at time-points 1, 4, and 5 only. For each significant finding, t-value, P-value and effect size (Cohen's d) are provided.

**Table 2B.** Behavioral results. For each group and time-points, the number of missing data-points are provided.

TP		Group 0	Group 1	Group 2	Group 3
1	BNT	0	0	1	0
	SFT	0	0	1	0
	COWAT	0	0	1	0
	WAB	0	2	1	0
2	BNT	0	0	1	0
	SFT	0	0	1	0
	COWAT	0	0	1	0
3	BNT	0	0	2	0
	SFT	0	0	1	0
	COWAT	0	0	1	0
4	BNT	0	0	1	0
	SFT	0	0	1	0
	COWAT	0	0	1	0
	WAB	1	2	2	1
5	BNT	1	1	2	1
	SFT	1	1	2	1
	COWAT	1	1	2	1
	WAB	1	2	2	1

**Table 2C.** Behavioral results. Mean and standard deviation of behavioral scores for each group and each time-point. Values were computed using every available score for each time-point.

TP	Group 0		Group 1		Group 2		Group 3		Group 123		
	M	SD	M	SD	M	SD	M	SD	M	SD	
1	BNT	45	10.5	25.2	24.6	21	19.3	33	19.3	27.1	20.6
	SFT	27.8	7.73	9.17	9.83	9.4	8.32	17.2	14	12.1	11.1
	COWAT	9.4	6.35	4.67	4.32	6	6.89	9.83	8.77	6.88	6.86
	WAB	87.6	2.9	26.7	5.3	35.6	27.5	70.5	34.2	51.2	33.4
2	BNT	48	10.1	22.8	25.7	23.5	20.8	37	21.7	28.3	22.6
	SFT	33	8.86	9	9.44	9.8	8.96	19.2	13	12.8	11.1
	COWAT	11.2	6.5	5.17	5.64	6.2	6.1	9.83	8.95	7.12	6.98
3	BNT	50.2	8.53	27	24.4	23.8	19.4	38.7	21.3	30.6	21.6
	SFT	30.8	3.11	9.83	10.6	11	12	19.3	13.2	13.5	12.1
	COWAT	11	7	5	5.9	5.2	6.38	11.8	9.6	7.47	7.8
4	BNT	51.6	8.62	28	25.7	30	19.8	42.2	20.1	33.8	21.9
	SFT	31.4	8.79	9.17	9.47	9.2	9.09	18.8	14.7	12.6	11.8
	COWAT	10	6.2	5.33	5.79	5.8	5.93	12.2	11.1	7.88	8.26
	WAB	85.2	6.72	28.7	1.48	36.9	25.3	71.3	34.9	52.4	33
5	BNT	47.6	9.56	25.2	24.7	23.3	17.1	38.5	20.4	29.7	21.2
	SFT	30.4	5.32	9.83	9.39	9	9.03	19	12.7	12.8	11
	COWAT	12.6	5.5	4.67	5.47	4	4.3	9.67	8.38	6.24	6.54
	WAB	82.6	2.76	29.1	2.9	37.6	28.4	72.1	35.4	53.1	33.9

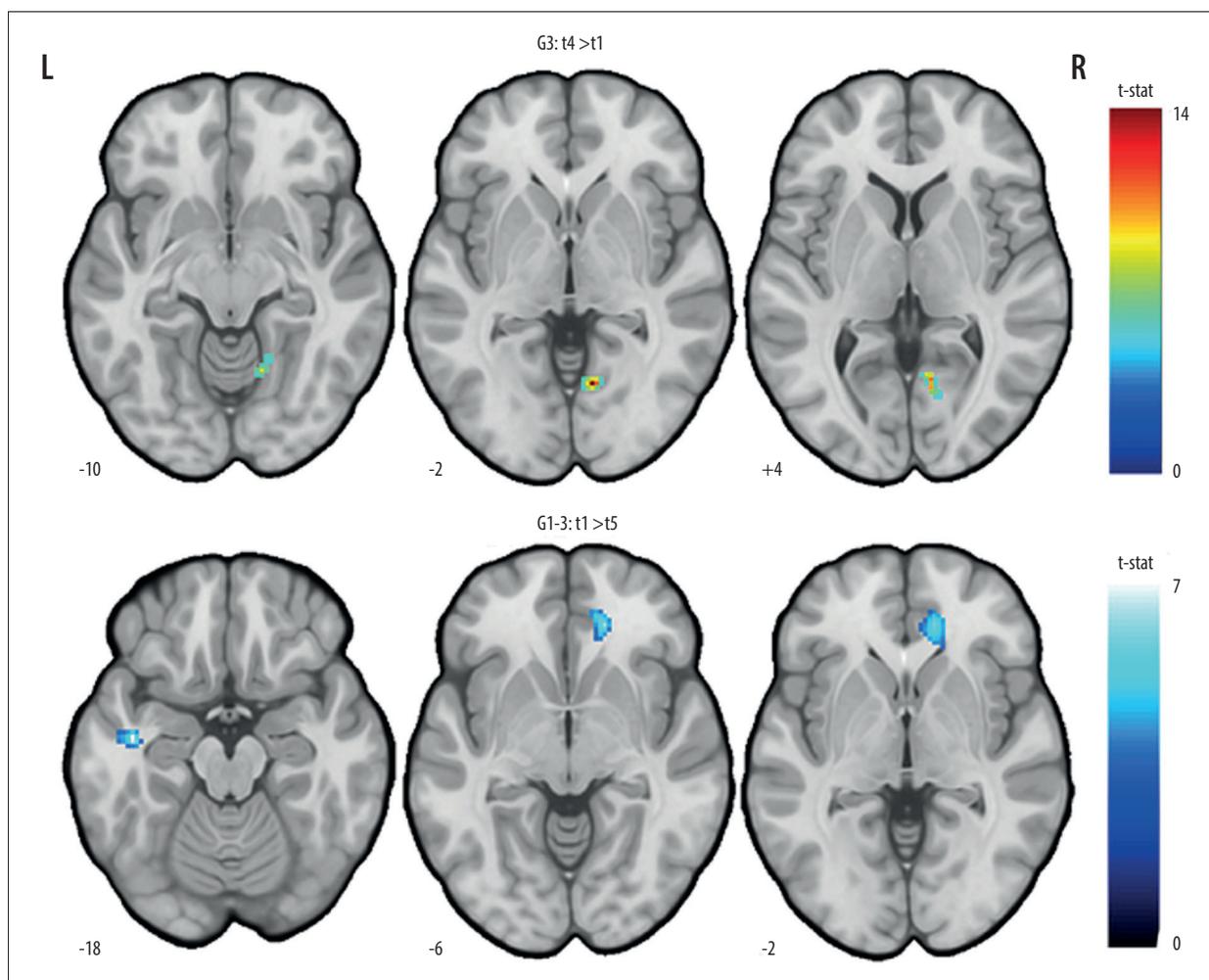
**Behavioral Results**

Behavioral results, including effect sizes (Cohen’s *d*), are presented in detail in **Table 2A** (due to the debilitating nature of their condition, some participants were not able to complete some of the behavioral tests; most of the effect sizes were medium or large. Missing scores are reported in **Table 2B** and mean and standard deviation scores are reported in **Table 2C**. One-way ANOVAs showed no significant differences between groups in any of the behavioral measures at  $t_1$  except for SFT ( $F=4.13, P=0.017$ ). Post hoc tests showed that  $G_1$  and  $G_2$  had lower SFT scores compared to  $G_0$  ( $P=0.038$  and  $P=0.02$ , respectively) but there was no significant difference between  $G_0$  and  $G_3$  or  $G_{1-3}$ . Due to the small number of subjects, we analyzed the data in groups, and we also combined groups 1-3 into 1 treatment group ( $G_{1-3}$ ). Paired samples *t* tests were used to compare scores between time-points ( $t_1, t_4$  and  $t_5$ ) for  $G_0$  and  $G_{1-3}$ . Results are reported in **Table 2A**. For BNT, several of the

groups, including  $G_0$  and  $G_{1-3}$ , significantly improved between baseline ( $t_1$ ) and  $t_4$  or  $t_5$ . The SFT scores increased between  $t_1$  and  $t_4$  for the combined group ( $P=0.046$ ). The WAB-R AQ significantly improved between  $t_1$  and  $t_5$  for the combined group ( $P=0.007$ ). The COWAT did not significantly change except at 1 point for the  $G_0$  group ( $t_4-t_3; P=0.03$ ). Due to the pilot nature of the study, corrections for multiple comparisons were not performed.

**Task fMRI Results**

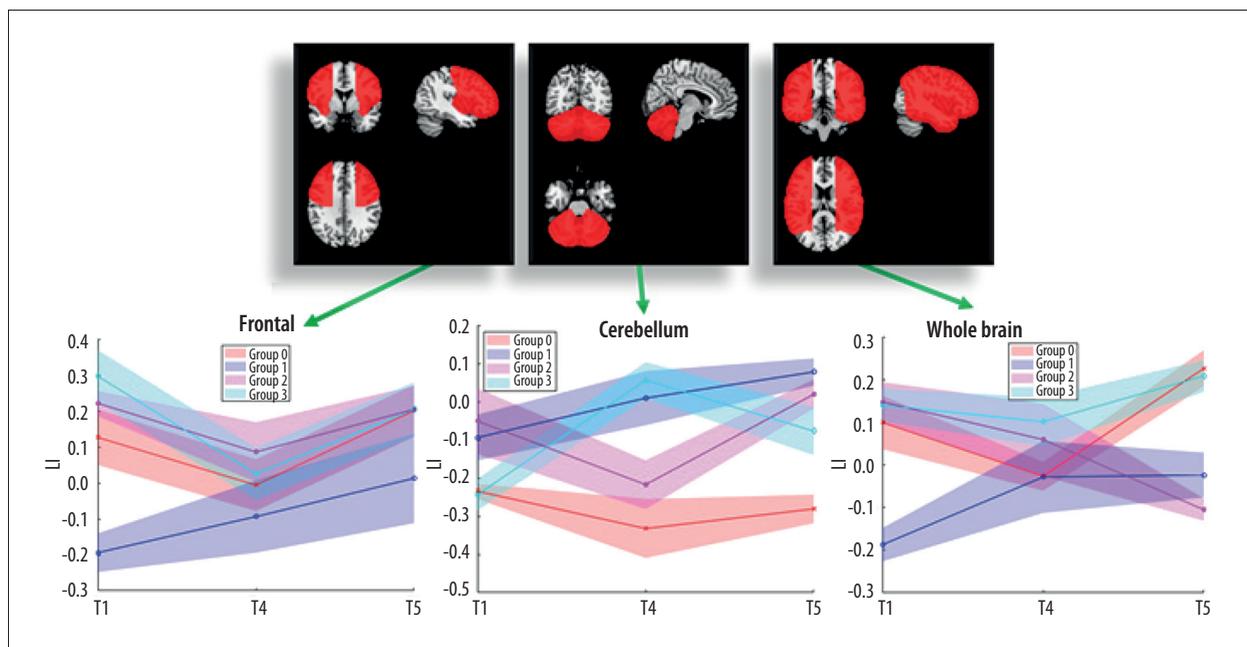
Second level paired *t* tests were used to assess fMRI (SDTD task) changes between time-points within groups on a voxel-wise basis. A cluster-wise FDR algorithm was used to correct for multiple comparisons in SPM (cluster  $P<0.05$ ). Results are depicted in **Figure 4** and **Table 3**. The BOLD signal significantly increased in the right lingual gyrus in  $G_3$  between  $t_1$  and  $t_4$ .  $G_{1-3}$  analysis showed a significant decrease between  $t_1$



**Figure 4.** fMRI semantic decision/tone decision (SDTD) task result (general linear model). Paired *t* tests were computed between time-points for each iTBS group (contrast SD >TD). For every contrast, corrected data are provided (Cluster-wise FDR,  $P<0.05$ ) with peak coordinates in Table 3. All pictures are in neurological convention (left in the figure corresponds to left in the brain).

**Table 3.** Main peak coordinates of paired *t* tests on general linear model analysis results included in Figure 4. Significant differences in cortical activity were found for Group 3 ( $G_3$ ), between pre-treatment and post-treatment ( $t_4 > t_1$ ), and for the combined group ( $G_{1-3}$ ) between pre-treatment and 3-month follow-up ( $t_1 > t_3$ ). Table shows all local maxima separated by more than 1 mm.

	Region Label	t-value	MNI coordinates		
			x	y	z
G3	Right Lingual gyrus	14.286	10	-64	-2
		10.971	10	-62	4
		9.701	14	-58	-8
		6.093	18	-52	-10
	Temporal_Mid_L	4.853	-50	-10	-20
	Temporal_Mid_L	4.629	-56	-14	-24
G1-3	Temporal_Mid_L	4.562	-54	-12	-22
	Frontal_Med_Orb_R	6.259	12	40	-2
	Frontal_Med_Orb_R	6.131	14	38	-4
	Frontal_Med_Orb_R	6.089	12	44	-4



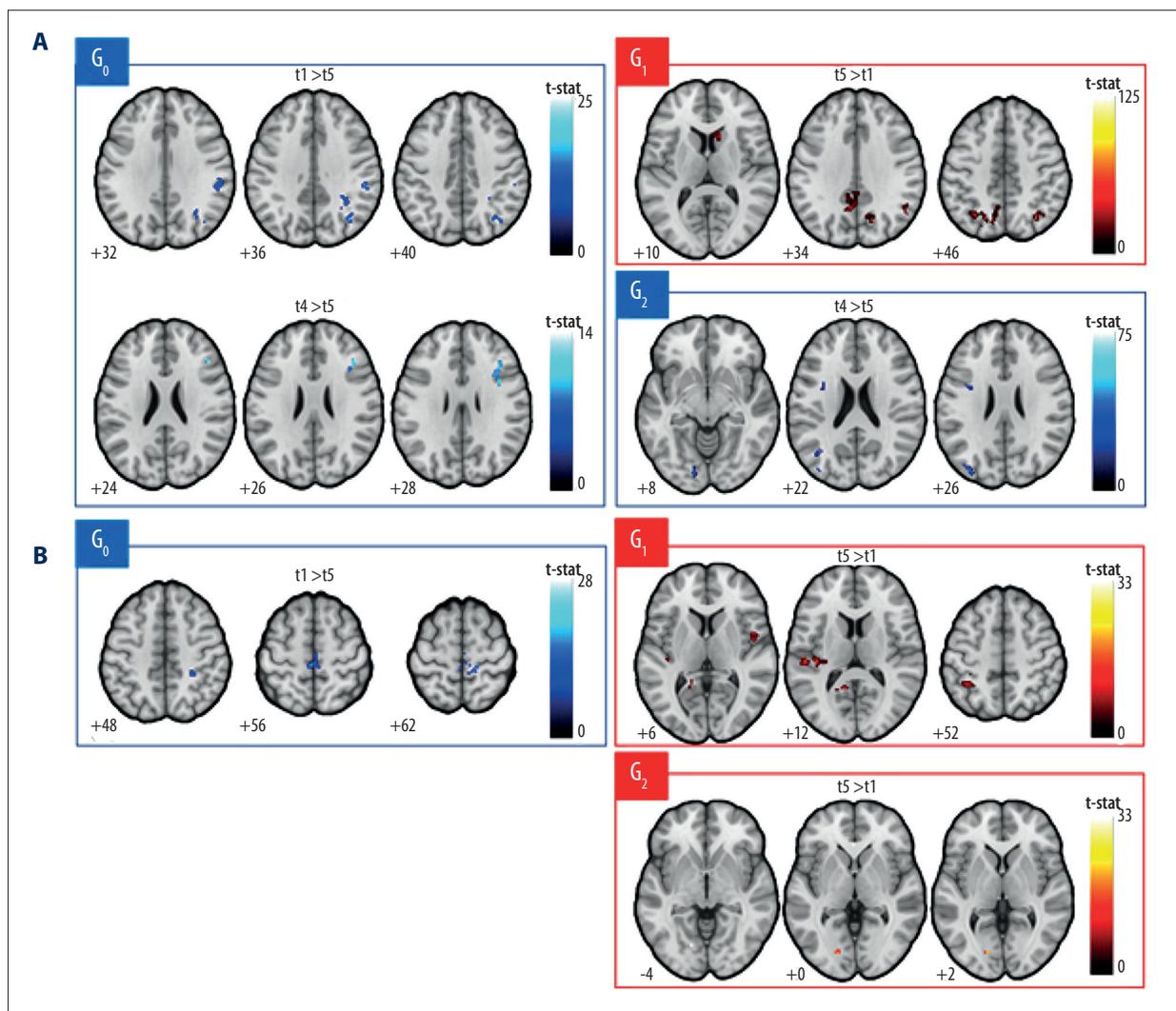
**Figure 5.** Results of Laterality Index (LI) analyses. Results are depicted for frontal mask (left), cerebellum mask (center), and whole-brain mask (right). Paired *t* tests revealed a significant increase between  $t_1$  and  $t_4$  for  $G_3$  (3 weeks of iTBS,  $P=0.02$ ) with cerebellum mask, and a significant increase between  $t_4$  and  $t_5$  for  $G_0$  ( $P=0.04$ ) with whole-brain mask.

and  $t_5$  in left middle temporal gyrus and in right medial fronto-orbital gyrus.

### Laterality Index Results

LIs were computed for each participant, each fMRI, each session, and each region of interest (Figure 5). The LIs for the  $G_{1-3}$  did not differ from  $G_0$  at any time-point for any mask. Paired *t* tests revealed a significant increase between  $t_1$  and  $t_4$  for  $G_3$

(3 weeks of iTBS,  $P=0.02$ ), with the cerebellum mask indicating stronger lateralization to the right cerebellar hemisphere between these 2 time-points. Also noted was a significant increase between  $t_4$  and  $t_5$  for  $G_0$  ( $P=0.04$ ) with the whole-brain mask indicating stronger lateralization to the left cerebral hemisphere between these 2 time-points.



**Figure 6.** gPPI connectivity results. For each participant, the first principal component of the BOLD time series from each scan was extracted from the left (A) and right (B) inferior frontal gyrus (IFG) and entered as a seed time series for the gPPI analysis. Location of the BOLD signal changes is provided in Table 4. Red and Blue frames refer to increases and decreases over time, respectively.

**gPPI Connectivity Results**

The  $G_0$  exhibited decreases in functional connectivity for the left IFG as seed region: between  $t_1$  and  $t_5$ , functional connectivity decreased in the right supramarginal, middle occipital, and angular gyri (Figure 6A). For the left IFG seed, between  $t_4$  and  $t_5$ , functional connectivity decreased also in the right inferior frontal region (Figure 6A). With seed in the right IFG, between  $t_1$  and  $t_5$  functional connectivity decreased in the right pre- and post-central gyri and in left and right paracentral lobules (Figure 6B).

In contrast to connectivity decreases in  $G_0$ ,  $G_1$  significantly increased in functional connectivity between  $t_1$  and  $t_5$  both with left and right IFG as seed region in multiple cortical areas

(Figure 6, Table 4).  $G_2$  showed decreased functional connectivity with left IFG as seed region between  $t_4$  and  $t_5$  in left middle occipital gyrus, left middle temporal gyrus, left lingual gyrus, left calcarine, and left frontal inferior operculum (Figure 6A, Table 4), and increased functional connectivity with right IFG as seed region between  $t_1$  and  $t_5$  in left lingual gyrus (Figure 6B, Table 4). Functional connectivity did not significantly change for  $G_3$  or for  $G_{1-3}$ .

**Relationship Between gPPI Connectivity and Behavioral Scores**

For the left IFG as seed, significant results were found for  $G_1$ ,  $G_2$ , and  $G_{1-3}$ , with a difference between  $t_4$  and  $t_1$  (Figure 7A, Table 5). For  $G_1$ , a difference in connectivity in the left middle

**Table 4.** Main peak MNI coordinates (x, y, z) for significant results of gPPI connectivity analysis. For each participant, the first principal component of the BOLD time series from each scan was extracted from the right and left inferior frontal gyrus (IFG) and entered as a seed time series for the gPPI analysis.

Left IFG					Right IFG								
Group/time	Region label	t-value	MNI coordinates			Group/time	Region label	t-value	MNI coordinates				
			x	y	z				x	y	z		
G0/t1>t5	SupraMarginal_R	49.1	52	-32	36	G0/t1>t5	Postcentral_R	28.7	14	-34	62		
	Occipital_Mid_R	26.5	38	-70	38		Paracentral_Lobule_L	21.7	-4	-30	58		
	Angular_R	17.3	38	-66	40		Paracentral_Lobule_R	20.0	10	-30	52		
G0/t2>t5	Frontal_Inf_Tri_R	14.2	44	28	24	G0/t2>t5	Precentral_R	17.7	16	-30	66		
	Frontal_Inf_Oper_R	11.8	38	8	28		Precentral_L	89.7	-32	-4	42		
G1/t5>t1	Parietal_Sup_L	125.3	-28	-54	64	G1/t5>t1	Angular_L	73.6	-42	-58	30		
	Precuneus_L	64.4	-8	-58	38		Parietal_Inf_L	66.1	-32	-46	54		
	Paracentral_Lobule_R	111.2	10	-38	58		Postcentral_L	36.7	-34	-38	50		
	Angular_R	103.5	34	-62	42		Lingual_R	51.0	14	-30	-12		
	Cingulate_Post_L	20.9	-6	-38	16		Vermis_3	25.5	4	-36	-8		
	Parietal_Sup_R	52.0	22	-58	58		Temporal_Sup_L	49.4	-54	-20	12		
	Cuneus_R	50.5	16	-68	34		Heschl_L	47.5	-40	-24	12		
	Precuneus_R	24.0	12	-60	30		Calcarine_L	38.8	-16	-50	8		
	Caudate_R	34.8	12	18	10		Precuneus_L	13.0	-8	-54	10		
	Temporal_Sup_R	23.2	48	-56	22		Cingulate_Post_L	10.9	-14	-44	8		
	Temporal_Mid_R	12.2	48	-62	18		Rolandic_Oper_R	27.4	48	-2	6		
	G2/t4>t5	Occipital_Mid_L	75.8	-38	-64		22	G2/t5>t1	Lingual_L	33.7	-16	-72	-4
		Temporal_Mid_L	23.4	-40	-66		20						
		Lingual_L	32.8	-14	-86		-8						
		Calcarine_L	13.9	-12	-90		-6						
Frontal_Inf_Oper_L		29.2	-40	4	26								

temporal gyrus and right precuneus was negatively correlated with SFT score. For  $G_2$ , a difference in connectivity in left occipital inferior gyrus was negatively correlated with COWAT score. For  $G_{1-3}$ , a difference in connectivity in the left superior frontal gyrus was negatively correlated with BNT score.

For the right IFG as seed, significant results were found between  $t_4$  and  $t_1$  for  $G_1$  and between  $t_5$  and  $t_4$  for  $G_3$ . For  $G_1$  (Figure 7B), the differences in connectivity in left planum temporale end left precuneus were positively correlated with COWAT scores, respectively. For  $G_3$ , a difference in connectivity in right anterior insula was positively correlated with BNT score.

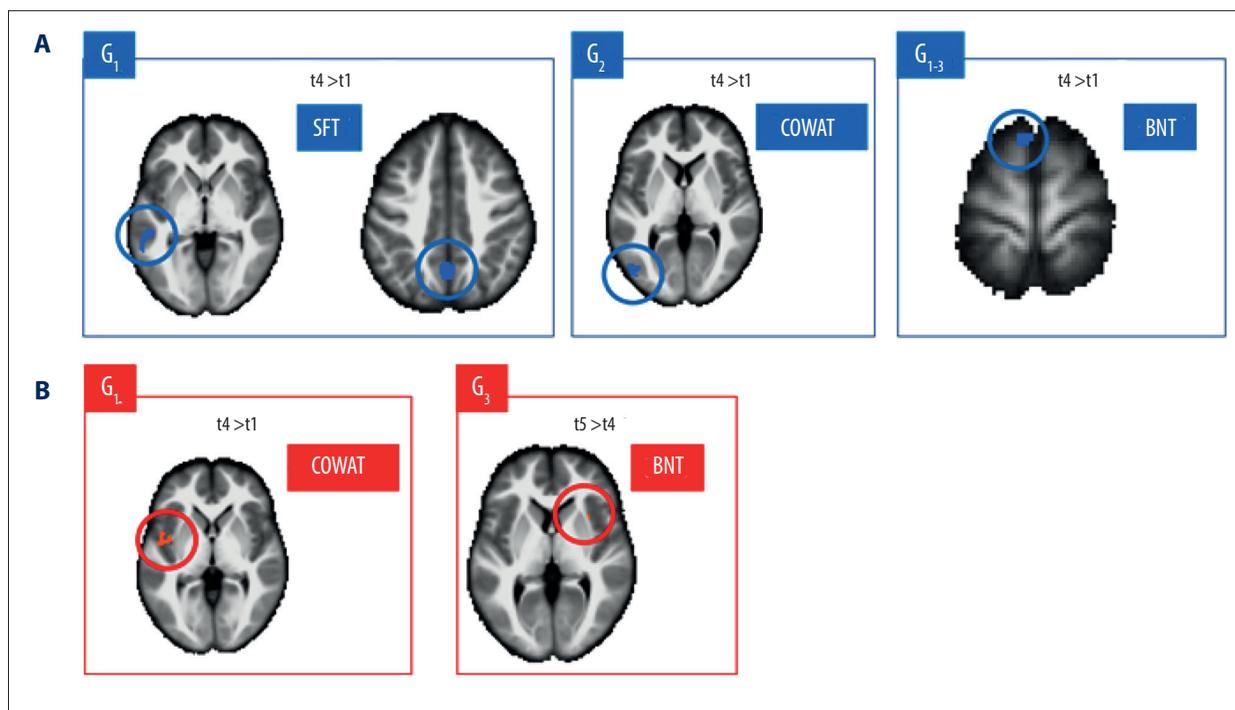
### Relationship Between Laterality Index and Behavioral Scores

No significant correlation was found between changes in LI in cerebellum and behavioral scores. Significant correlations were

found for  $G_0$  and  $G_2$ : for  $G_0$ , changes over time in LI with frontal mask were significantly correlated with BNT ( $t_1-t_5$ ,  $r=-0.95$ ,  $P=0.009$ ). For  $G_2$ , changes over time in LI with frontal mask were significantly correlated with SFT ( $t_1-t_4$ ,  $r=0.92$ ,  $P=0.008$ ), and changes over time with whole-brain mask were significantly correlated with BNT ( $t_4-t_5$ ,  $r=-0.95$ ,  $P=0.027$ ).

### Discussion

This was the first randomized, double-blind, sham-controlled trial of fMRI-guided iTBS for the treatment of post-stroke aphasia following LMCA stroke. Some of the language measures improved with iTBS, with these improvements corresponding to changes in fMRI language activation patterns, including decreases in right-hemispheric activation and increases in left frontal language lateralization in close proximity to the stimulation site and opposite changes in cerebellar fMRI signal



**Figure 7.** Results of regression analysis between change over time in gPPI connectivity coefficients and change over time in behavioral measures. Results for changes in the left IFG seed are shown in (A) and for the right IFG seed are shown in (B). Location of the BOLD signal changes is provided in Table 5. Red and Blue frames refer to positive and negative regression coefficients, respectively.

**Table 5.** Main peak MNI coordinates (x, y, z) for significant results of regression analyses between changes over time in gPPI connectivity coefficients and changes over time in behavioral measures. Results are depicted in Figure 7.

Left IFG				Right IFG							
Group/time/score	Region label	t-value	MNI coordinates			Group/time/score	Region label	t-value	MNI coordinates		
			x	y	z				x	y	z
G1	Temporal_Mid_L	36.17	-54	-34	-4	G1	Planum	44.07	-50	-4	0
T4 > T1	Precuneus_R	35.8	2	-60	48	T4 > T1	Polare_L				
SFT_NEG						COWAT_POS					
G2	Occipital_Inf_L	34.18	-48	-74	4	G3	Insula_Ant_R	58.97	32	16	4
T4 > T1						T5 > T4					
COWAT_NEG						BNT_POS					
G123	Frontal_Sup_L	7.11	-6	8	76						
T4 > T1											
BNT_NEG											

lateralization. Additional analyses showed variable changes in connectivity between the left and right IFG (anatomical seed near or around the area of stimulation or its right homologue) and other left and right hemispheric brain regions in response to iTBS but not in response to sham; these connectivity changes correlated with some of the behavioral measures. These findings demonstrate dynamic language recovery following stroke and generally support our initial hypotheses regarding linguistic improvements and neuroplasticity changes in response to iTBS.

### Improvements in Language Skills in Response to iTBS

Our observed improvements in AT in response to iTBS were variable when comparing groups  $G_1$ - $G_3$ , with the most consistent improvements noted for the BNT (Table 2A), where the statistically significant changes were of medium or large sizes. In the combined  $G_{1-3}$  group, the BNT responses significantly improved after the intervention ( $t_4$ ), which was sustained at 3-month follow-up ( $t_5$ ). This is consistent with the results of a combined CIAT and iTBS intervention we recently reported [18,25] and implies that while iTBS can improve language in patients with chronic stroke-induced aphasia, additional treatments some weeks to months after initial therapy to maintain the gains may be needed. The notion of booster treatment is similar to the idea of a transfer package, which consists of a set of techniques designed to maintain and possibly improve gains from the initial treatment. A transfer package is commonly used in rehabilitation studies to reinforce maintenance and possible further improvement of the initial post-intervention function increases [52,53].

However, it is important to note for future studies that while the collected measures improved immediately after treatment ( $t_4$ ), there was some regression at follow-up ( $t_5$ ). We further note that the BNT remained improved for the 3 months after therapy ( $P=0.056$ ), with the WAB-R score also significantly improved at the  $t_5$  time-point ( $P=0.007$ ), indicating that some improvements may be sustained over time and may parallel the changes observed in the fMRI measures. It is also important to acknowledge that  $G_0$  showed some gains as well, indicating that some of the changes observed in BNT could be due to test-retest learning effects or spontaneous recovery. While some of the tests we used may be subject to practice effects, this was minimized by rotating different versions of each test. In addition, recent research has shown relative stability of the WAB-R when repeated over short periods [54]. Finally, our AT results are consistent with the results of approximately 20 uncontrolled studies that have assessed short- and intermediate-term effects of TMS on language improvement in post-stroke aphasia patients [10].

Only 4 of the previous neurostimulation studies used an approach similar to ours (ie, applying rTMS to the lesioned rather than the intact hemisphere) [14,15,18,55]. In the first study, trends for language improvement were observed in 8 patients who received the same treatment protocol as in the present study [22]. In the second study, low-frequency (1 Hz) rTMS was applied to either the inferior frontal or superior temporal gyri (based on the type of aphasia and the region most-activated with fMRI) followed by 60 minutes of intensive speech therapy [15]. Participants in the study significantly improved in several aphasia measures. The third study's participants were randomized to rTMS and language training vs sham and language training [14]. Combined low-frequency rTMS over the non-dominant hemisphere's IFG (inhibition) and higher frequency rTMS over the dominant hemisphere's IFG (excitation) in 30 patients with post-stroke non-fluent aphasia, followed by speech/language training, resulted in improved language after active vs sham treatment in the language section of the Hemispheric Stroke Scale and other measures of post-intervention outcomes (eg, NIHSS). Finally, in our previous study that combined iTBS immediately followed by CIAT, the WAB-R AQ improved after 10 treatment sessions [18]. While these studies have used different TMS stimulation parameters, guidance methods, targets, and additional interventions, their results converge with the current pilot RCT results, suggesting that neurostimulation can improve short- and intermediate-term language outcomes in patients with chronic post-stroke aphasia.

### Effects of TMS on Language Lateralization and Connectivity in Post-stroke Aphasia

In our prior research on iTBS, we reported changes in fMRI language lateralization and activation in the dominant and non-dominant hemispheres after stimulation of the left-hemisphere targets, indicating both local and global effects of the stimulation [55]. We have subsequently documented additional effects of iTBS on brain anatomy and structural connectivity in patients with post-stroke aphasia [23-25]. It is important to note that while the majority of the TMS intervention studies in aphasia used structural MRI to localize the stimulation area, only a few studies have used other neuroimaging methods for this purpose [15,18,22,56,57]. In our studies, we have used the SDTD fMRI task to visualize the target for stimulation on pre-intervention imaging. Others have used either  $O^{15}$ PET and a verb generation task [57], or fMRI or functional near-infrared spectroscopy (fNIRS) with a word repetition task [15,56].

Functional neuroimaging has proven effective for identifying the active language area in the peristroke area and in the contralateral unaffected hemisphere [58]. Beyond targeting therapy, conducting follow-up neuroimaging allows exploration of the cortical changes in response to an intervention [59]. Our study

was specifically designed to test and is the first to report the effect of neurostimulation as the sole rehabilitative modality; other studies have used neurostimulation to prime the brain prior to a behavioral intervention. Here, although iTBS was the sole intervention, we showed cortical plasticity with non-significant shifts of language lateralization to the dominant (affected) hemisphere and concurrent shifts in lateralization to the right cerebellar hemisphere. Additionally, we showed significant correlations between changes in connectivity over time and changes in behavioral performance using gPPI for all active treatment groups, while these effects were not observed in the sham group ( $G_0$ ). Moreover, the vast majority of the significant results were found between  $t_1$  and  $t_4$ , which are the time-points around the stimulation period. This suggests that TMS can have a positive impact on cerebral networks linked to behavioral performance and that the observed effect can partially dissipate with time after completion of the treatment, again suggesting the need for additional (booster) therapy to possibly maintain the gains from the initial intervention. Significant correlations were also found between changes in LI over time in frontal and whole-brain mask and behavioral scores. These results possibly suggest that laterality of cortical activity plays a role in the observed behavioral performance, which is a phenomenon previously observed in neuroimaging studies [7,60]. These findings underscore the importance of the canonical language regions for post-stroke language recovery [37,58,61]. They further underscore the importance of brain plasticity in post-stroke recovery, which is a concept that has been at the core of post-stroke rehabilitation [7,62]. Whether the shifts in activation patterns and eventual recovery rely on the peristroke areas, other areas of the ipsilateral hemisphere, or the contralateral hemisphere may depend on multiple factors including handedness [4], preservation of the white-matter tracts (bottlenecks) [63,64], age at the time of the stroke [42], lesion extent [65], intensity and duration of the intervention [3], or other currently unknown factors such as genetics [66].

This study's limitations need to be considered for future neurostimulation and post-stroke aphasia rehabilitation studies. First, recruitment and retention in randomized trials are challenging, and although we identified the planned number of participants, a number of screening failures and dropouts decreased our sample size, which limited our ability to detect differences between groups and time-points and prevented a meaningful comparison of treatment duration effects. Blinding is important and was maintained through separating the treatment team

from the testing/imaging team, and via the use of a sham coil. However, we did not ask the participants if they could determine whether they received experimental treatment vs sham, and it is possible that covert awareness of the form of treatment could have inadvertently unblinded them. Further, a "transfer package" is being used in many rehabilitation studies to reinforce maintenance and possible further improvement of the initial treatment effects [52]. Such a package, whether behavioral or TMS-based, needs to be developed and validated for aphasia rehabilitation to sustain treatment gains. We also recommend measuring discourse productivity as a functional outcome in patients who undergo post-stroke aphasia rehabilitation [67]. Participants who received sham treatment were noted to have improvements in BNT. This finding questions the validity of using BNT in rehabilitation studies and underscores the importance of validating measures for longitudinal use prior to selecting them as the primary outcome measures. Of note is that there were baseline differences between groups, including degree of aphasia and level of performance on AT. These differences may disappear when larger samples are included or may need to be controlled for in subsequent studies; level of education will need to be either controlled for or included as a co-variate in final analyses. Further, the AT results analyses were not adjusted for multiplicity and we cannot rule out the possibility of Type I error. However, the consistency of direction, magnitude, and timing of effects and their biological plausibility support those exploratory findings. Finally, in larger studies, stratification by potential confounders such as education, age, size and location of the lesion, aphasia type (eg, fluent vs non-fluent), and aphasia severity may increase the ability to observe between-group effects.

## Conclusions

The results of this randomized, double-blind, sham-controlled pilot study support the hypothesis that neurostimulation, as the sole therapeutic approach, can improve post-stroke aphasia and induce short- and intermediate-term cortical plasticity in human brain networks involved in language function. Understanding the strengths and potential limitations of the current study will inform the design of future trials.

## Conflict of Interest

None.

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