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Continuous theta-burst stimulation demonstrates language-network-specific causal effects on syntactic processing

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ABSTRACT

Hierarchical syntactic structure processing is proposed to be at the core of the human language faculty. Syntactic processing is supported by the left fronto-temporal language network, including a core area in the inferior frontal gyrus as well as its interaction with the posterior temporal lobe (i.e., "IFG + pTL"). Moreover, during complex syntactic processes, left IFG also interacts with executive control regions, such as the superior parietal lobule (SPL). However, the functional relevance of these network interactions is largely unclear. In particular, it remains to be demonstrated whether the language network plays a specific causal role in comparatively challenging syntactic processes, separable from the interaction between IFG and other general cognitive regions (i.e., "IFG + SPL" in the present study). The present study was designed to address this question. Thirty healthy adult Chinese native speakers underwent four continuous theta-burst stimulation (cTBS) sessions: stimulation over IFG, stimulation over IFG + pTL, stimulation over IFG + SPL, and sham stimulation over IFG + irrelevant region in a pseudo-randomized order. In each session, participants were required to label the syntactic categories of jabberwocky sequences retaining real Chinese function words (e.g., "ム了ウ" is labeled as a verb phrase (VP): "[VP [V]]N]", similar to "ziff-ed a wug", where "ziff" and "wug" are nonsense pseudowords, and the whole phrase is a VP). Contrasted with sham cTBS, change percentage of accuracy rates (Δ ACCR%), reaction times (Δ RT%), and coefficient of variation (Δ CV%) were calculated and compared across conditions. First-order behavioral results showed a significantly higher $\Delta CV\%$ after stimulating IFG + pTL compared to stimulating the IFG + SPL, indicating that syntactic processing became more unstable. Second-order representational similarity analysis (RSA) results revealed that cTBS effects on IFG + pTL selectively depended on the hierarchical embedding depth, a key measure of syntactic hierarchical complexity, whereas the effects on IFG + SPL were sensitive to the dependency length, a crucial index reflecting the working memory load. Collectively, these findings reveal the specific causal relevance of the language areas for hierarchical syntactic processing, separable from other general cognitive (such as working memory) capacities. These results shed light on the uniqueness and the specific causal role of the language network for the human language faculty, further supporting the causally separable view of the functional dissociation between the language network and the domain-general/ multiple-demand network.

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1. Introduction

The human language faculty, a remarkable human capacity, is supported by a left fronto-temporal network (Fedorenko et al., 2024; Friederici, 2017; Hagoort, 2013), which is ubiquitously shared across various language families (Malik-Moraleda et al., 2022). One distinctive feature of human language is the ability to recursively build up syntactic hierarchies for language comprehension. This operation is enabled by the left inferior frontal gyrus (IFG) (esp., pars opercularis, coarsely corresponding to the Brodmann Area (BA) 44) as well as its interaction with the left posterior temporal lobe (pTL), including parts of the superior and middle temporal gyri (i.e., STG and MTG) (e.g., den Ouden et al., 2012; Matchin et al., 2017; Ohta et al., 2013; Pallier et al., 2011; Schell et al., 2017; Zaccarella et al., 2017; Wang et al., 2021; Chen et al., 2023).

In particular, the left IFG was frequently detected for the comparison of complex versus simpler structures (e.g., den Ouden et al., 2012; Friederici et al., 2006; Makuuchi et al., 2009; Pallier et al., 2011; Santi and Grodzinsky, 2010; Wang et al., 2021). For example, a seminal study of Makuuchi et al. (2009) separated the hierarchical embedding depth (i.e., the number of center-embedding relative clauses) from the dependency length (i.e., the linear distance between the dependent syntactic objects) regarding syntactic complexity. In their study, complex German sentences with deeper center-embedded structures showed increased activation in the left IFG (esp., BA 44) when compared with simpler sentences. Similarly, Wang et al. (2021) further demonstrated that complex sentences with more/deeper center-embedded relative clauses showed a significant neural oscillatory effect of IFG (esp., BA 44), representing a successful training-induced change in language processing. A meta-analysis (Zaccarella et al., 2017) synthesizing imaging studies that compared structured sentences with word lists proposed that the left IFG (esp., BA 44) might serve as a syntactic engine for syntactic hierarchy construction. Collectively, these studies consistently showed the critical role of the left IFG in syntactic processing.

However, recent neuroimaging findings suggested that the IFG may not only act as a language-specific area, but rather contribute to language and other cognitive functions in general (Fedorenko and Blank, 2020). Fedorenko and Blank (2020) reviewed previous work on the dissociation of the language-specific network and the multiple-demand network for cognitive control, and argued that the left IFG (the larger Broca's area) was activated for both sentences versus word lists and for harder versus easier non-linguistic cognitive tasks (such as spatial working memory). These findings indicate that Broca's area might be functionally heterogeneous, with separable contributions to the language network and domain-general (multiple-demand) network, respectively. This notion is reminiscent of previous arguments that Broca's area might be a region for verbal working memory (Rogalsky and Hickok, 2011) or, more generally, executive control (Clos et al., 2013; Duncan, 2010).

Nevertheless, the specific role of the left IFG might be defined by its interaction with other regions to compose functionally different brain networks (Friederici and Gierhan, 2013). Using functional localizers during neuroimaging experiments, several studies demonstrated that the fronto-temporal language network is functionally separable from the domain-general multiple-demand network for executive control processes (Hiersche et al., 2024; Fedorenko et al., 2010, 2011; Fedorenko and Thompson-Schill, 2014). Specifically, the language network only responded to the contrast between sentence and nonword list process-ing, separable from working memory, executive control, and other complex non-linguistic tasks, in which the multiple-demand network was engaged (Blank and Fedorenko, 2017; Quillen et al., 2021). While this points towards a high functional-anatomical specificity of brain areas for language processing, several questions remain open.

Firstly, some studies revealed overlap of both networks in the IFG (e. g., MacGregor et al., 2022), consistent with the argument "Broca's area is NOT a natural kind" (Fedorenko and Blank, 2020). This suggests that

merely considering task-related activation of IFG alone during complex language tasks may be insufficient to disentangle the specificity versus overlap of language and other general cognitive abilities. Rather, this finding argues for a network perspective, considering the interaction of distributed language and domain-general cognitive areas (see also Chen et al., 2021).

Secondly, studies on syntactic complexity may be confounded with task difficulty and/or working memory load (Makuuchi et al., 2009; Pallier et al., 2011; Santi and Grodzinsky, 2010; Wang et al., 2021). Consequently, to demonstrate specific functions of the language (esp., syntactic) network relative to domain-general cognitive networks such as the multiple-demand network, direct task comparisons are needed. Accordingly, further work showed that the language network was recruited for complex syntactic processing in the first language (L1) (Chen et al., 2023). Chen et al. (2023) developed a function-word-based jabberwocky sentence processing paradigm in Mandarin Chinese, in which content words were replaced by nonsense pseudowords while real function words and Chinese syntactic structures were preserved. For example, "为了口只生的さ" is a sequence token of the structure type "V 了Num只Adj.的N", in which "为, 口, 出, and さ" are pseudowords, whereas "了, 只, and 的" are function words: "了" is a dynamic auxiliary, following the verbs, functionally similar to the past participle (such as "-ed") in English; "只" is a measure word/quantifier, used between number and noun; "的" is a structural auxiliary, used between a modifier (such as an adjective) and a noun. This would be like the English example, "ziff-ed ruflix-ty lic-es of mout-ing wug-s" in which the Italic suffixes and function words are real words to reveal the syntactic relations. Chinese native speakers had to label the syntactic categories of the jabberwocky sequences based on the function words [i.e., "为了口只 出的さ" would be labeled as a verb phrase (similarly, in English, "ziffed ruflixty lices of mouting wugs" should also be labeled as a verb phrase)]. The comparison of structured sequences versus word list showed that the left IFG, in interaction with the left pTL, was crucially involved in Mandarin Chinese syntactic hierarchy building.

In contrast, the same function-word-based jabberwocky sentence processing paradigm was utilized in a subsequent Chinese L2 (second language) syntactic processing study (Gao, 2022; Gao et al., 2024), and a network including IFG and the left superior parietal lobule (SPL), a core region of the multiple-demand network (Fedorenko et al., 2013), was detected during Chinese L2 complex syntactic structure building. In line with this study, a recent study indicated that L2 syntactic processing might resort to a broader brain network including key regions of the multiple-demand network such as the SPL (Hou et al., 2024). It is noteworthy that the SPL has been assigned a critical role in the multiple-demand network (e.g., Blank and Fedorenko, 2017; Chen et al., 2021; Diachek et al., 2020; Fedorenko, 2014; MacGregor et al., 2022). For instance, Diachek et al. (2020) conducted a large-scale fMRI investigation and showed that the domain-general multiple-demand network, including bilateral fronto-parietal areas, exhibited a stronger response in explicit tasks than in passive reading/listening paradigms, when compared with the language-selective fronto-temporal network. According to a series of studies (Assem et al., 2020; Diachek et al., 2020; Fedorenko et al., 2011; 2013, 2024), the activation of the parietal lobe (including SPL) was coherently stronger for the harder non-language cognitive tasks in various domains, including verbal and spatial working memory. Therefore, the interaction of IFG and SPL ("IFG + SPL") might play a role in general cognitive functions during complex syntactic processing.

However, neuroimaging data can only provide correlational evidence for brain-behavior relationships (Poldrack, 2006; Logothetis, 2008). Consequently, it is largely unknown whether the language network causally contributes to complex syntactic processing in a specific fashion, separable from general executive control regions such as the SPL.

Transcranial magnetic stimulation (TMS), a non-invasive brain stimulation technique, has been utilized to investigate the causal role of the left IFG in language processing (see reviews in Devlin and Watkins, 2007; Hartwigsen, 2015). Several studies showed that perturbation of the IFG affected syntactic processing (Meyer et al., 2018; Uddén et al., 2017; Wu et al., 2024; van der Burght et al., 2023). For examples, Meyer et al. (2018) explored the causal role of the left IFG in chunking words into syntactic phrases. Ambiguous sentences, which could be parsed into either short or long syntactic phrases, served as experimental materials. The results revealed that stimulation of the left IFG impaired the ability to process longer syntactic chunks. Uddén et al. (2017) also reported that the left IFG played a causal role in processing non-adjacent dependencies during artificial grammar discrimination. These TMS findings provide evidence for the causal role of the left IFG in syntactic processing.

Nevertheless, although the efficacy of TMS for the identification of the causal role of the neural substrates for different cognitive operations has been demonstrated by a recent meta-analysis (Qu et al., 2022), the spatial resolution of TMS is limited for specifying the role of left IFG subregions for syntactic versus domain-general processing. TMS studies targeting more than one site within the same experiment to explore the causal relevance of multiple areas or intact network interactions (e.g., via the "multifocal TMS" approach) (Hartwigsen, 2015; Hartwigsen et al., 2010, 2016) are still relatively sparse. Multifocal TMS outperforms single-node TMS by enabling the stimulation of connected brain regions, thereby achieving network-level causal effects. This approach has been shown to mitigate rapid functional compensation within brain networks (Hartwigsen et al., 2010), facilitate the investigation of how one region's function depends on the integrity of other regions within a network (Hartwigsen et al., 2016), and increase behavioral effects (Sinisalo et al., 2024). To our knowledge, no TMS studies have yet targeted multiple areas of the language network to explore the specific causal role of the language network in complex syntactic processing.

To address these issues, this study reasoned that the left IFG should interact with different regions to compose functionally separable networks during complex syntax processing. Specifically, we expected interactions with the pTL for language-network-specific contributions and with the SPL for domain-general cognitive contributions. Therefore, we investigated the impact of TMS on the language network (represented by IFG + pTL) during complex syntactic processing, while considering a high-level multiple-demand network condition by targeting IFG and SPL (i.e., IFG + SPL), which was assumed to reflect domain-general cognitive capacities. This allowed us to assess whether the language network plays a specific role in syntactic processing performance or indeed merely depends on general cognitive capacities. To clarify, the "specificity" of the causal effects in the language network is twofold:

(1) The causal contribution of the language network could be separable from those of the multiple-demand network for domain general cognitive capacities. For example, TMS might selectively affect syntactic processing when applied over the language network but not the multiple-demand network. The relevance of the language network might be specific to certain syntactic operations, different from those of the multiple-demand network. As implied by previous neuroimaging results (Makuuchi and Friederici, 2013; Makuuchi et al., 2009), the fronto-temporal language-specific network, responsible for constructing hierarchical syntactic structures, interacted closely with the fronto-parietal working memory network involved in dependency length processing as the syntactic complexity of sentences increases. Consequently, we may expect that the language network played a causal role in syntactic hierarchy construction but not in the processing of the dependency length which might be related to the working memory capacity supported by the multiple-demand network.

Therefore, we hypothesized that, due to the complexity of the current syntactic processing task, TMS over IFG alone might not be sufficient to affect task performance, because fast functional compensation from either the pTL in the language network or SPL in the multiple-demand network condition might help to maintain task function at a high level. Moreover, if the language network plays a specific causal role in syntactic processing, separable from domain-general function, then we would expect a strong and selective modulation of task performance for the combined perturbation of "IFG + pTL" but not for "IFG + SPL".

2. Methods

2.1. Participants

Thirty healthy adult Chinese native speakers were recruited for the study [male: 13, female: 17; age: mean = 20.3 years, Standard deviation (SD) = 1.55 years], whose syntactic processing capacity was deemed fully matured (Skeide and Friederici, 2016). All participants had normal or corrected-to-normal vision and were right-handed, with no history of psychiatric or neurological diseases. Each participant came to the laboratory once every week, for a total of four experimental sessions. All participants gave signed informed consent before the experiment and received remuneration for participation. This study was approved by the Tianjin Normal University Ethics Committee.

2.2. Materials

We adopted the well-established Chinese function-word-based jabberwocky sentence reading paradigm from our previous functional magnetic resonance imaging (fMRI) study (Chen et al., 2023), retaining only real Chinese function words in the structures while replacing content words with pseudowords, thereby reducing the semantic interference on syntactic processing. For details of the experimental materials, the reader is referred to our previous study (Chen et al., 2023).

In brief, typical Chinese function words were selected (Tang, 2019), encompassing dynamic and structural auxiliaries, prepositions, and quantifiers (see Fig. 1A). These function words are highly abstract without concrete meanings (Huang and Liao, 2011; Tang, 2019). Pseudo-content words were generated using unfamiliar symbols, including Japanese Katakana and traditional Bopomofo (Zhuyin) used in Taiwan Province, which were like Chinese characters but unpronounceable and semantically meaningless for the participants (see Fig. 1A). These symbols had simple strokes and clear configurations and were assigned to five syntactic categories (including verbs, nouns, adjectives, numerals, and adverbs). Considering that the components of Chinese characters often reveal the syntactic/semantic categories of the corresponding characters (Yeh et al., 2017; Zhang et al., 2019), pseudowords within the same word category were designed to share certain visual similarities to facilitate association of visual information with syntactic categories. All participants confirmed that they memorized word categories based on these visual similarities or cues. Moreover, semantic associations between the pseudowords and orthographically similar real words were assessed via a 5-point Likert rating scale, and results showed that these pseudowords could be hardly related to any concrete semantic concepts (difficulty scores are shown in Fig. 1A; see also Chen et al., 2023).

These function words could be combined with other pseudowords to construct hierarchical syntactic structures, either verb phrases or noun phrases. Please note that the jabberwocky sequences were generated based on typical Chinese syntactic structures, which relied on the function words to reveal syntactic relations, a key feature of Chinese grammar which does not depend on morphosyntactic cues, therefore separating syntactic processing from the morphosyntactic level. Consequently, short grammatical sequences were built, including 16 noun phrases and 16 verb phrases [see Fig. 1B for examples of N(P) (noun phrase) and V(P) (verb phrase)], with each structure comprising one function word and two pseudo-content words. Furthermore, in case



Fig. 1. Experimental materials. A: word categories and the corresponding tokens, including pseudo-content words and real function words. Semantic-related mean scores with their 95 % confidence intervals are provided alongside pseudo-content words. All categories showed difficulty in relating the word tokens to real content words (each category's difficulty was significantly higher than the intermediate level (=3) ($ts(23) \ge 2.94$, ps < 0.01, $ds \ge 0.60$) and showed no statistical difference from the ``very difficult'' level (=4) ($-1.54 \le ts(23) \le 1.92$, $ps \ge 0.14$), according to two separate one-sample *t*-tests, see Chen et al., 2023 for more details). B: experimental sequences, including short and long variations, encompassing grammatical structures (NP and VP) and ungrammatical sequences (illustrated by `` *''). English examples are provided below the grammatical sequences to facilitate comprehension of the language materials. The gray shadow marks the word category violation in the examples and the dashed curve denotes truncation of the violated part while keeping the rest of the elements mergeable. *Abbreviations*: V, verb; N, noun; Adj., adjective; Num., number; Adv., adverb; Dyn-Aux., dynamic auxiliary; Str-Aux.', structural auxiliary (for verb modifiers); Str-Aux., structural auxiliary (for noun modifiers); Prep., preposition; Q, quantifier; NP, noun phrase; VP, verb phrase; *, word category violation.

participants could identify the sequence category solely by focusing on the function word without processing the entire sequence, 16 short ungrammatical sequences were designed, in which the category of a content word violated the syntactic rules (see the example "*" in Fig. 1B and also Supporting Information Section 1).

We further designed long complex sequences to delve deeper into the syntactic processing complexity. Long grammatical sequences were built including 24 noun phrases and 24 verb phrases, each comprising three function words and four pseudo-content words (see Fig. 1B for examples and Supporting Information Section 1). Additionally, 24 long ungrammatical sequences were developed where the word category violations were placed at the second or third pseudoword position to mitigate edge-effect (e.g., Endress et al. 2010; Sonnweber et al. 2015). Across all experimental materials, the frequency of different pseudo-content-word tokens within the same word category, the ratio of grammatical to ungrammatical trials, and the distribution of trials with violations at different positions were balanced carefully to prevent judgment biases from participants. Please note that since both short and long sequences contained real function words for hierarchical syntactic structure build-up, we were not interested in assessing the sequence length influence on the TMS effects by comparing short and long sequence processing performance.

2.3. Procedures

2.3.1. Main procedures for each session

The experimental procedures followed Chen et al. (2023) and are depicted in Fig. 2B. During preparation, the participants were provided with the pseudo-content-word list (Fig. 1A) three days before the experiment and had to memorize the category of each pseudoword. After memorizing, they underwent a time-limited pseudoword category test administered via the online test website (Sojump, https://www.wjx.

cn). Only participants who scored 90 (out of 100) were permitted to proceed to the formal experiment, ensuring that they had effectively acquired the word category knowledge of the pseudowords in advance.

Before the actual experiment, participants underwent a behavioral adaptation phase consisting of two sessions. Firstly, a vocabulary category test, identical to the earlier online test, was conducted. Only participants whose accuracy rates reached 90 % in the two successive blocks, with 20 trials per block, were able to proceed. We deliberately decided for this high level of accuracy to maintain consistency with our previous fMRI study (Chen et al., 2023). We reasoned that this would ensure successful performance in the subsequent syntactic structure processing tasks, as supported by pilot testing in the fMRI study (Chen et al., 2023). After that, in the practising phase, participants had to complete the category identification task for short 3-word sequences and long 7-word sequences, respectively. The task required them to combine the words to identify the category of the sequence (NP, VP, and violation of word category). After identification, feedback indicating correctness or incorrectness was provided. The short sequence comprised 32 trials, while the long sequence consisted of 24 trials. The formal experiment commenced only if both test accuracy rates reached 90 %, ensuring that participants had proficiently grasped the experimental rules. Given that the syntactic structures used in the experiment were familiar to the participants, and their accuracy rates during the practice phase reached relatively high levels, age differences between participants might be negligible for syntactic processing.

After practising, the actual experiment began, comprising a brain stimulation phase and two testing phases. The TMS phase of the experiment, the independent within-group variable, contained continuous theta-burst stimulation (cTBS) to IFG, IFG + pTL, IFG + SPL, and sham IFG + task-irrelevant brain region. Each participant underwent four stimulation sessions, with a minimum interval of seven days between sessions to avoid potential carry-over effects as shown in Fig. 2A.



Fig. 2. Experimental procedure. A: Brain stimulation sessions, the within-group independent variable, containing continuous theta-burst stimulation (cTBS) to IFG, IFG + pTL, IFG + SPL, and sham IFG + unrelated brain region. Brain stimulation sessions were counterbalanced across participants. The experiment was conducted on four separate days, and the between-session interval was longer than seven days. B: Main procedures. Participants first completed the pseudoword category familiarization. Prior to the actual experiment, participants were required to pass the word category identification test and the sequence category identification task, including short 3-word sequences and long 7-word sequences, with each task achieving 90 % accuracy rate. During the actual experiment, after brain stimulation, participants completed category identification tests for both short sequences and long sequences, following the same procedure as in the practice phase but without feedback. C: Trial examples. Trial examples for each test along with their timing parameters are shown. *Abbreviations*: IFG, inferior frontal gyrus; pTL, posterior temporal lobe; SPL, superior parietal lobule.

The Latin Square randomization method was used to balance the order of the four TMS sessions across participants, effectively controlling for potential order effects. Detailed stimulation settings are provided in Section 2.3.2 below. During the testing phase, participants performed category identification tasks for both short sequences and long sequences, following the same procedure as in the practice session but without receiving feedback. The short sequence test consisted of one run with 48 trials, while the long sequence test included two runs, each with 36 trials. Participants were allowed a 30 s rest between runs. The detailed presentation times for each trial are shown in Fig. 2C. The adaptation phase lasted approximately 1 h, while the testing phase took about 20 min. It is worth noting that, in each session, experiment materials were different in their tokens (i.e., different pseudowords could compose various sequence tokens) and in the orders of appearance, but the structure types remained consistent across all stimulation sessions. On one hand, different sequence tokens appearing in various orders could prevent participants from training-effects based on stimulus similarities. On the other hand, keeping the structure types similar made the comparisons among the four stimulation conditions fair, excluding confounding effects of differences in structure types. Consequently, we chose this option. Structure similarity was also critical for performing the subsequent representational similarity analysis, as each structure type should have the same number of stimulation conditions.

2.3.2. TMS protocol

To ensure precise coil placement during neuronavigation, highresolution T1-weighted images were acquired prior to the experiment from all participants using a 3T MRI Scanner (Siemens Prisma) to facilitate subsequent coil placement with neuronavigation. Individual anatomical data were obtained for co-registration with the following imaging parameters: repeated time (TR) = 2530 ms; echo time (TE) = 2.98 ms; flip angle = 7°; field-of-view (FOV) = $256 \times 256 \text{ mm}^2$; matrix size = 256×256 mm²; in-plane resolution within slices = 1.0×1.0 mm; slice thickness = 1.00 mm; number of slices = 192.

Each participant's high-resolution T1-weighted MRI scan was coregistered with their head in real time using a frameless stereotaxic navigation system (Localite GmbH, Bonn, Germany). The system was recalibrated at the beginning of each TMS session to ensure consistent coil placement across sessions. Specifically, anatomical landmarks, such as the anterior and posterior commissures and the falx cerebri, were utilized to guide the co-registration process. Reflective markers attached to a headband worn by participants enabled real-time tracking and dynamic adjustments of coil positioning over the predefined target region. These procedures were identical across all four TMS sessions to minimize variability and enhance the reliability of the stimulation process.

The target location of the IFG (MNI: x = -54, y = 20, z = 26) was defined by averaging the peak MNI coordinates of IFG in Chinese native speakers (Chen et al., 2023) and Chinese L2 learners (Gao, 2022) when processing the jabberwocky sequences. Moreover, the Chinese native speakers recruited the left pTL (MNI: x = -58, y = -42, z = 20) (Chen et al., 2023), while the left SPL (MNI: x = -16, y = -78, z = 50) was activated at the whole-brain level for the Chinese L2 learners, indicating a higher syntactic task demand for less proficient participants (Gao, 2022). It is noteworthy that in our previous study, only the left IFG was activated for the comparison of structure versus word list for Chinese native speakers (Chen et al., 2023). In contrast, a Chinese L2 experiment using the same materials revealed that only the left SPL survived at the whole-brain level analysis (Gao, 2022; Gao et al., 2024). Therefore, based on the same jabberwocky sequence processing paradigm, the present study adopted the SPL coordinates identified from the L2 study to compare the causal effects of TMS over the language network critical for native/L1 syntactic processing and the multiple-demand network which was identified as crucial for L2 syntactic processing. Consequently, both pTL and SPL were defined as two stimulation sites in the present study, rendering IFG + pTL as representative language network condition and IFG + SPL as the multiple-demand network condition. Since the network stimulation contained both the frontal (IFG) and posterior regions (pTL and SPL), a posterior task-irrelevant region, the mid-point between pTL and SPL (MNI: x = -37, y = -60 z = 35), was deliberately defined as control brain region.

Stimulation was delivered using a MagPro X100 TMS stimulator (MagVenture) equipped with a standard 70 mm figure-of-eight coil (MagVenture MCF-B65). The stimulation protocol followed procedures described in our previous research (Wu et al., 2024, 2025) and conformed to established guidelines (Rossini et al., 2015). To determine the resting motor threshold (RMT), single pulses were delivered at the beginning of the first TMS session to the left primary motor cortex (M1) targeting the hand area. Motor-evoked potentials (MEPs) were recorded from the right first dorsal interosseous muscle using electromyography (Boux and Pulvermüller, 2023; Obeso et al., 2013). Electrodes were configured in a belly-tendon montage, with the ground electrode attached to the left wrist. Participants sat comfortably and were instructed to keep their hands relaxed during the procedure. The TMS coil was positioned tangentially to the scalp at a 45° angle relative to the midline of the central sulcus, with the handle pointing laterally and posteriorly (Wu et al., 2025). RMT was defined as the minimum stimulation intensity required to generate MEPs with a peak-to-peak amplitude of at least 50 µV in five or more out of ten consecutive trials (Rossini et al., 2015; Steel et al., 2016). Participants' RMT values ranged from 43 % to 75 % of the maximum stimulator output, with a mean threshold of 58.467 % (SD = 8.437 %). Stimulation intensity was set to 80 % of each participant's RMT, consistent with prior studies employing theta-burst stimulation protocols (Jung and Lambon Ralph, 2021; Steel et al., 2016; Wu et al., 2025).

Subsequently, cTBS was applied. The cTBS protocol was configured to deliver triplets of stimulation pulses at 50 Hz, repeated every 200 ms (equivalent to 5 Hz), in a continuous 40-second train, resulting in a total of 600 pulses (Huang et al., 2005; Steel et al., 2016). To validate the reliability of the experimental results, sham stimulation was also conducted. Sham stimulation with the coil flipped was achieved by delivering stimulation to IFG along with a task-irrelevant brain region (i.e., the unrelated region in this study).

2.4. Data analyses

2.4.1. First-order behavioral analyses

The accuracy rate (ACCR) and reaction times (RT) of participants were collected during the actual experimental phase. Please note that RT-related indices were selectively analyzed for correct responses, and trials with RT shorter than 200 ms were excluded for each participant (see also Chen et al., 2019). To assess processing stability, we calculated the coefficient of variation (CV) based on RT (Segalowitz and Segalowitz, 1993): $CV = SD_{RT} / mean RT$. This measure has been suggested as a reliable and robust indicator of automatization in language learning and processing (e.g., Jeon and Friederici, 2013; Lim and Godfroid, 2015; Segalowitz and Segalowitz, 1993; Segalowitz and Hulstijn, 2005; Wu et al., 2024). In this study, we considered both ACCR and RT as indices of processing quality, and CV as an indicator of the response/processing state.

Behavioral change percentage (` Δ %'') was computed for ACCR, RT, and CV under each condition, calculated by "(effective - sham cTBS) / sham cTBS". Therefore, the Δ % for each participant's behavioral indices in both the short sequence test and the long sequence test were computed. We did not differentiate between short and long sequences. Both short and long sequences were jabberwocky structures composed by real Chinese function words and pseudowords, and all sequences were included in the first-level ANOVA tests to guarantee sufficient statistical power. Since we only had 48 trials for short sequences, analyzing them separately might have resulted in a lack of power, and the comparison between short and long sequences might have been unbalanced because of the difference in the number of trials. Therefore, the averages across both tests were obtained to derive the Δ ACCR%, Δ RT%, and Δ CV% for each participant under different brain stimulation conditions. If necessary, outliers of the behavioral change rates for each index were interpolated by "Q1 – 1.5 IQR" or "Q3 + 1.5 IQR" respectively (Q: quantile; IQR: interquartile range).

The $\Delta\%$ for each index were compared to 0 using one-sample T-tests to assess whether cTBS could induce significant changes under specific conditions. Subsequently, one-way repeated measures ANOVAs were conducted to analyze the differences in $\Delta\%$ among the three cTBS stimulation conditions (IFG, IFG + pTL, & IFG + SPL) for each behavioral index, and *p*-values of post-hoc tests were Bonferroni-corrected. Specifically, for each index (e.g., Δ CV%), a one-way repeated measures ANOVA was performed to compare the three real stimulation conditions IFG, IFG + pTL, and IFG + SPL. Conditional on significant effects, the following post-hoc paired-samples *t*-tests were performed: IFG vs. IFG + pTL, IFG vs. IFG + SPL, and IFG + pTL vs. IFG + SPL. The alpha-level was divided by three (because the *t*-tests were performed three times), resulting in a corrected threshold of *p* < 0.017 to reduce the Type I error. Data analyses were performed in JASP 0.17.1.0 (https://jasp-stats.org/).

2.4.2. Second-order representational similarity analysis

Given that long sequences contained various types of structures (i.e., 6 structures for NP and VP respectively) at the same length, while short sequences were composed of only three words and lacked sufficient feature variability, the relationships between syntactic complexity of long sequences and cTBS modulation effects for both "IFG + pTL" and "IFG + SPL" could be further scrutinized via representational similarity analysis (RSA). RSA has prevailed into the investigation of higher-order neural representations as well as the comparison of distinctive cognitive computation models on the basis of the second-order isomorphism between the materials and responses (Kriegeskorte et al., 2008).

For each type of long sequences, the mean embedding depth and the mean dependency length were calculated by an in-house developed program called "L2C-Rater" [https://github.com/iris2hu/L2C-rater; see Wang and Hu (2021) for details]. The embedding depth feature was assumed to reflect the syntactic complexity of hierarchical processes, while the dependency length feature was deemed to stand for the syntactic complexity of multi-level associations, more related to the working memory capacity necessary for complex structure processing (Gibson, 1998; Makuuchi et al., 2009). Thus, two kinds of material feature representational dissimilarity matrices (RDMs) were built-an embedding depth RDM and a dependency length RDM-with the dissimilarity between each pair of structures calculated by the Euclidean distance. Correspondingly, for each behavioral index (e.g., RT) of each network, each structure had all the participants' after-cTBS behavioral data, and therefore, by calculating the behavioral data Euclidean distance between each pair of structures, a behavioral performance RDM could be established (see Fig. 4). Subsequently, a Spearman correlation test was performed for each pair of behavioral performance RDM and material feature RDM with 3000-times permutation. For each behavioral index (e.g., ACCR) within each condition (e.g., IFG + pTL), *p*-values were Bonferroni-corrected to test against the null hypothesis that, for instance, after-cTBS, ACCR of IFG + pTL had no significant correlation with either embedding depth or dependency distance. This allowed us to explore the sensitivity of "IFG + pTL" and "IFG + SPL" to different dimensions of syntactic complexities, respectively.

3. Results

3.1. First-order behavioral analysis results

The following results were obtained for the dependent measures in the sham condition: ACCR (M = 0.809, SD = 0.092), RT (M = 913.825

ms, SD = 173.833 ms), and CV (M = 0.321, SD = 0.070). These results show that participants were able to perform the task. The sham condition served as baseline for calculating the change rate for the critical comparisons among effective stimulation conditions. Accordingly, the summary of descriptive behavioral data is presented in Table 1. As shown in Fig. 3, the results indicate that only the Δ CV% under the "IFG + pTL" condition significantly exceeded 0 [M = 0.064, t(29) = 2.333, p = 0.027, Cohen's d = 0.426].

The one-way repeated measures ANOVAs results revealed a significant main effect of stimulation only for the Δ CV% [*F*(2, 58) = 4.668, *p* = 0.013, $\eta_p^2 = 0.139$], and post-hoc paired-samples *t*-tests further demonstrated the Δ CV% was significantly higher under the "IFG + pTL" condition than under the "IFG + SPL" condition [*t*(29) = 2.992, *p*_{bonf} = 0.012, Cohen's *d* = 0.617]. No significant differences were found for both Δ ACCR% and Δ RT% among the different stimulation conditions (*F*s \leq 0.822, *p*s \geq 0.444).

Taking together, the results of the first-order analyses implied that the language network (IFG + pTL) might show inhibitory TMS effects, and their casual role should be separable from the null effects of the multiple-demand network (IFG + SPL).

3.2. Second-order representational similarity analysis results

Although the multiple-demand network showed null effects for the first order as reported in Section 3.1, it is still beneficial to take this network into consideration at the second-order RSA: On the one hand, to probe the specificity of the language network for certain syntactic operations, one should take the multiple-demand network into account; on the other hand, it is still not clear whether IFG + SPL play an indirect role in complex syntactic processing at the second order.

RSA results (Fig. 4) showed that the embedding depth was positively correlated with the after-cTBS RT of "IFG + pTL" (rho = 0.25, $p_{\text{bonf}} = 0.048$). Moreover, the dependency length was positively correlated with the after-cTBS CV of "IFG + SPL" (rho = 0.34, $p_{\text{bonf}} < 0.001$).

4. Discussion

The present TMS study is the first to explore whether core areas of the language network (i.e., IFG + pTL) play a specific causal role in complex syntactic processing, and whether such a contribution of the language network is separable from the domain-general areas for other cognitive abilities such as working memory. We found that inhibiting the language network reduced the processing automaticity, leading to a more unstable state, as revealed by the larger change rate of the coefficient of variation (i.e., Δ CV%) after stimulation of the language network. Notably, this change was larger than the impact of TMS on the multiple-demand network "IFG + SPL". In contrast, $\Delta ACCR\%$ and ΔRT % did not show significant changes under the different stimulation conditions. Given that the entire sequence was presented on a single screen, the task per se reduced working memory load. More critically, we assumed that for native speakers, syntactic processing primarily relied on the language network, with less involvement of the multiple-demand network, unless the syntactic structures were extremely difficult to process as shown in first language (e.g., Makuuchi et al., 2009; Wang

Table 1

Summary of the behavioral data.

Conditions	$\Delta ACCR\%$		ΔRT%		$\Delta CV\%$	
	Mean	SD	Mean	SD	Mean	SD
IFG	0.029	0.118	0.004	0.120	0.004	0.144
IFG + pTL	0.056	0.154	0.000	0.127	0.064	0.150
IFG + SPL	0.061	0.182	0.004	0.166	-0.025	0.136

Abbreviations: ACCR, accuracy rate; RT, reaction time; CV, coefficient of variation; SD, standard deviation; IFG, inferior frontal gyrus; pTL, posterior temporal lobe; SPL, superior parietal lobule. et al., 2021) and second language processing (e.g., Hou et al., 2024). Moreover, based on the RSA results, syntactic complexity differently interacted with TMS in the language network compared to the multiple-demand network: The observed TMS effect on the language network was selectively correlated with the embedding depth (i.e., larger embedding depth was related to longer response times), separable from the TMS effects on the multiple-demand network "IFG + SPL," which was modulated by the dependency distance / length.

Single-node TMS over the IFG did not yield significant changes of the behavioral indices. This was expected because our complex syntactic processing task should require intact interactions between IFG and pTL as suggested by Chen et al. (2023). In particular, our task required participants to explicitly identify the syntactic categories of the jabberwocky sequences with the semantics being largely deprived, which should require communication between IFG and pTL to merge the constituents hierarchically and label the whole sequences. This hypothesis was also motivated by previous work emphasizing the relevance of the pathway connecting IFG (esp., BA 44) and pTL (esp., pSTG) for complex sentence or syntactic processing (den Ouden et al., 2012; Matchin et al., 2017; Ohta et al., 2013; Pallier et al., 2011; Schell et al., 2017; Zaccarella et al., 2017; Wang et al., 2021; Chen et al., 2023). Accordingly, a temporal "virtual lesion" of IFG by means of TMS might trigger rapid functional compensation from either pTL or other general cognitive regions (i.e., SPL in this study) (See Maran et al., 2022 for a similar discussion). Crucially, the IFG + pTL effect was also significant against zero (i.e., against baseline), which further evidences that a combined inhibition effect of the language network should be larger than the effect of the single IFG perturbation. Alternatively, given the high syntactic complexity of the task, IFG should interact with SPL to support general task processing with comparatively high demands. Accordingly, Duncan (2010) found that the fronto-parietal network plays an important role in complex cognitive control tasks such as solving matrix problems with various dimensional changes. These authors argued that the function of the IFG might depend on its interaction with distinct posterior regions, depending on the specific task at hand. Hence, our present results support the notion that stimulating IFG alone might be prone to yield null results because of its dynamic functional interaction with distinct posterior regions: Fast compensation in the language network (IFG + pTL) and / or the multiple-demand network (IFG + SPL) might prevent performance deterioration when applying a single perturbation to the IFG.

An intriguing question is if single note perturbation of the pTL would affect language processing. A recent TMS study by Schroën et al. (2023) reported an early effect of left pTL stimulation (targeting the superior temporal gyrus and adjacent superior temporal sulcus) during sentence comprehension. Specifically, TMS over left pTL applied at verb onset during sentences with varying cloze probability reduced the N400 amplitude of the final sentence noun measured with simultaneous EEG. This effect was suggested to reflect modulation of phonological and lexical-semantic processes. Moreover, a study by Kuhnke et al. (2017) demonstrated that TMS over the left IFG but not pTL (planum temporale) affected reordering during complex auditory sentence processing in German. Our visually presented materials did not have concrete lexical-semantic meanings or phonological features. Therefore, stimulation on pTL alone might not be sufficient to reveal a syntactic effect in this study. Moreover, as argued in the introduction, the language network and the multiple-demand network overlap in frontal regions (especially in the left IFG) but not in the pTL. Consequently, we did not consider to select single site stimulation of the pTL in this study. Nevertheless, we do think that it would be worthwhile to probe the causal role of the pTL in syntactic processing in a follow-up study.

Stimulation of the language network, as represented by "IFG + pTL" in the current study, was demonstrated to cause complex syntactic processing performance change that were not observed after targeting IFG and an executive control region. Specifically, analyses of the processing stability at the first-order revealed a significant inhibitory TMS



Fig. 3. Behavioral results. Differences between behavioral indices (Δ ACCR%, Δ RT%, and Δ CV%) among three brain stimulation conditions (IFG, IFG + pTL, & IFG + SPL). *Abbreviations*: ACCR, accuracy rate; RT, reaction time; CV, coefficient of variation; IFG, inferior frontal gyrus; pTL, posterior temporal lobe; SPL, superior parietal lobule. Bold and *, significant differences between conditions.



Fig. 4. RSA results. V: verb phrases/structures; N: noun phrases/structures. Correlation between the embedding depth representational dissimilarity matrices (RDM) and the dependency distance RDM and each behavioral performance RDM for each condition ("IFG + pTL" (upper panel) & "IFG + SPL" (lower panel)) via Spearman correlations with 3000-times permutation tests, and with Bonferroni-corrected p-values. Black arrows indicated significant correlation results after correction. As shown in the figure, for the language network (LN), the increase (\uparrow) of hierarchical embedding depth (ED) was positively correlated (\sim) with larger RT difference, noted as "LN: ED $\uparrow \sim RT\uparrow$ "; for the multiple-demand network (MDN), the increase of the dependency distance (DD) was positively correlated with larger CV difference, that is, "MDN: DD $\uparrow \sim CV\uparrow$ ".

effect on the language network, and this effect was significantly stronger than stimulation of the multiple-demand network (i.e., "IFG + SPL"). Previous work (Jeon and Friederici, 2013) proposed that language processing is modulated by the degree of processing automaticity (i.e., the extent of how automatic language processing is) as syntactic processing in the first language (L1) activated the posterior part of IFG (i.e., BA 44), and its signal change was negatively related to the coefficient of variation (CV), the index also adopted in the present study. Moreover, these authors identified the arcuate fasciculus, connecting BA 44 and the pTL, as the core structure underlying complex syntactic processing, providing further support for the role of automaticity. In line with these findings, a recent TMS study on natural sentence processing in Mandarin Chinese also showed significantly lower processing stability after cTBS-induced inhibition of left IFG, as indexed by the increased coefficient of variation as well (Wu et al., 2024). Although the present syntactic task was more difficult due to the explicit syntactic category identification with little semantic facilitation, which is distinct from the previous TMS work on natural syntactic processing (Wu et al., 2024), L1 participants of the present study shifted to recruit the language network for syntactic operations and similarly showed reduced processing

stability after stimulation.

In a complementary approach, RSA analyses at the second order were used here for the first time in combination with TMS data to provide more elaborate information on how the language network might specifically respond to the syntactic features of jabberwocky sequences. These analyses showed that the TMS effect on the language network was selectively related to the embedding depth (as revealed by the increases in response times), indicating that sequences having deeper embedded constituents might be processed significantly slower after perturbation on the language network. This further demonstrates that the interaction between IFG and pTL is critical for hierarchical syntactic structure construction as suggested by previous work (den Ouden et al., 2012; Matchin et al., 2017; Ohta et al., 2013; Pallier et al., 2011; Schell et al., 2017; Zaccarella et al., 2017; Wang et al., 2021; Chen et al., 2023). However, TMS perturbation of the multiple-demand network "IFG + SPL" showed more domain-general effects: Behavioral changes were modulated by the dependency distance, and longer dependency length lowered the processing stability after stimulating "IFG + SPL". The SPL might be a key region for verbal working memory (Fedorenko et al., 2013; Jonides et al., 1998; Emch et al., 2019), and interact with the IFG during harder verbal and spatial working memory tasks, as reflected in increased multiple-demand network activity with increasing task complexity (Fedorenko et al., 2011; Fedorenko and Blank, 2020). Accordingly, Koenigs et al. (2009) found that patients with SPL lesions had deficits in manipulating information in working memory. Necka et al. (2021) further reported that working memory training increased the correlation between behavioral accuracy and SPL activation. These findings suggest that the SPL might has a critical role in working memory. Similarly, a fronto-parietal pathway was identified for working memory processes during syntactic operations (Makuuchi and Friederici, 2013), with the corresponding parietal region showing increased responses to the dependency length (Makuuchi et al., 2009). Nevertheless, the previous studies (Makuuchi et al., 2009; Makuuchi and Friederici, 2013) focused solely on complex sentences with center-embedded relative clauses, leaving it unclear whether the effect of dependency length might generalize to other syntactic structures. In the current study, we designed various syntactic structures to cover a range of hierarchical embedding depths and dependency lengths. Our results provide further evidence for a causal role of both networks suggesting that such a division of labor may be reflected at the second-order/isomorphic level as indicated by our RSA results. Consequently, RSA is an efficient approach to map the relationships between stimulus features and task performance. Hence, we speculate that the observed causal contribution of "IFG + SPL" during complex syntactic processes in our study might be related to the participants' working memory capacity. Interestingly, the left SPL did not show significant activation at the whole-brain level when native Chinese speakers processed jabberwocky sequences in the previous study (Chen et al., 2023). Yet, the present TMS effects after combined stimulation of IFG + SPL showed a prominent second-order correlation with the dependency distance of the complex syntactic structures, thus specifying the way how SPL was involved in complex syntactic processes.

5. Limitations

Since each participant underwent four cTBS sessions (one sham and three real stimulation sessions) with an inter-session interval of at least a week, it was challenging to recruit a large sample of participants willing to commit to a "long-term" experiment. Future studies should include larger sample sizes to enhance statistical robustness. Additionally, individual responses to TMS may vary due to differences in cognitive abilities, brain anatomy, and other factors, which are inherent in all TMS studies. Although we tried to standardize stimulation parameters and controlled for individual differences in the present study, these factors may still have influenced our results which should therefore be interpreted with caution. Furthermore, it should be cautious that multifocal TMS also presents challenges, including complex timing control, optimized stimulation sequences, unintended neural interactions, and nonlinear dose-response relationships (Hartwigsen, 2015; Bergmann and Hartwigsen, 2021), emphasizing the need for rigorous design and interpretation.

6. Conclusion

Utilizing a multi-focal TMS approach with continuous theta-burst stimulation, this study revealed that the language network plays a causal role in complex syntactic processing: Our study identified a high degree of specificity of the language network since the observed TMS effects could be distinguished from the impact of perturbing IFG and SPLat both the first and the second order. Collectively, the current findings deepen our understanding of the neural mechanisms of the human language faculty and further indicate the efficacy of TMS in research of causality of brain networks in the field of neurolinguistics.

Moreover, this study also illuminates that multi-focal TMS approaches might be suitable to identify the neural correlates of various syntactic operations in future studies, for example, the differences in the causal role of the language network for processing anaphora, topicalization, and wh-movement which generate predictable nonadjacent dependencies but may differ in their underlying neurocognitive mechanisms (see also Santi and Grodzinsky, 2012). Multi-focal TMS might also be used to address bilingual/cross-linguistic syntactic processing differences. For instance, inhibition of the IFG + SPL might result in significantly worse L2 syntactic processing performance, contrary to the results of L1 syntactic processing as identified by this study. Furthermore, we also suggest that multi-focal TMS could be applied to other non-linguistic complex cognitive tasks. In particular, the causal role of the multiple-demand network could be specified with regard to its contribution to various tasks in working memory, music, arithmetic, and action domains.

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CRediT authorship contribution statement

Chenyang Gao: Writing – original draft, Investigation, Formal analysis, Data curation, Conceptualization. **Junjie Wu:** Writing – original draft, Investigation, Formal analysis, Data curation, Conceptualization. **Yao Cheng:** Writing – review & editing, Investigation, Formal analysis. **Yuming Ke:** Writing – review & editing, Investigation, Formal analysis. **Xingfang Qu:** Writing – review & editing. **Mingchuan Yang:** Writing – review & editing, Funding acquisition. **Luyao Chen:** Writing – review & editing, Supervision, Funding acquisition, Conceptualization.

Declaration of competing interest

The authors declare that they have no conflict of interest.

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Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.neuroimage.2025.121014.

Data availability

Anonymized data will be made available upon reasonable requests and collaborative agreement addressed to the coauthors. Researchers wishing to obtain the data must contact the Max Planck Partner Group, School of International Chinese Language Education, Beijing Normal University, to sign a formal data sharing agreement.

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