Right C7 neurotomy at the intervertebral foramen plus intensive speech and language therapy versus intensive speech and language therapy alone for chronic post-stroke aphasia: multicentre, randomised controlled trial - Appendix

Table of contents

1. Participating Clinical Sites

(1). Huashan Hospital, Fudan University
(2). Shanghai Pudong Hospital
(3). Huadong Hospital Affiliated to Fudan University
(4). Shanghai Xuhui Central Hospital

2. Supplement to the Main Text Methodology

2.1 Trial Inclusion and Exclusion Criteria

Inclusion criteria (Meeting all of the following conditions):

1) Aphasia for over 12 months after a single onset of infarction or hemorrhage of the left hemisphere, confirmed by MRI;

2) of 40–65 years, male or female sex, right-handed, native Chinese speakers;

3) Western Aphasia Battery Aphasia Quotient (WAB-AQ) score below 93.8 points;

4) Severity score assessed using the Boston Diagnostic Aphasia Examination (BDAE) test of level 1 and above;

5) Good compliance and ability to cooperate with language rehabilitation training;

6) Ability to understand fully and agree with the doctor's treatment plan and sign the informed consent.

Criteria 3 and 4 will to be confirmed through the diagnostic evaluations of two attending specialists.

Exclusion criteria:

If the subject exhibits any of the following conditions, they will be excluded from participating in the study.

1) Any surgical contraindication, determined by a qualified anesthesiologist or clinician;

2) History of aphasia before the last onset of a stroke;

3) Serious, untreated mental illness;

4) Aphasia due to neurodegenerative diseases or traumatic brain injury;

5) Contraindications for EEG and MRI evaluation;

6) Inability to complete the assessments and rehabilitation required per study design;

7) Severe motor speech disorder and hearing impairment;

8) Having received intensive post-stroke rehabilitation therapy 4 weeks before recruitment.

2.2 Interventions and Standardizations of NC7 Surgery and iSLT

2.2.1 Description of the surgical procedure

During the CC7 operation, we cut the C7 nerve root at the intervertebral foramen to ensure that the C7 nerve on the affected side would provide more nerve fiber length. C7 neurotomy (NC7) is derived from CC7, therefore they are similar in terms of surgical procedures[1,2]. After a satisfactory general anesthesia, the patient is placed in the supine position with pillows under the interscapular region and conventionally sterilized and draped. A 4 cm transverse incision will be made along the height of the two transverse fingers above the right clavicle. We incise the skin, subcutaneous tissue, and the vastus cervicis muscle layer-by-layer, taking care to protect the cutaneous nerve. We retract the sternocleidomastoid muscle medially and separate it toward the deep side to expose the omohyoid muscle. Then we retract the omohyoid muscle upward and laterally to expose the interscalene fat pad. We divide the interscalene fat pad to expose the phrenic nerve on the surface of the anterior scalene muscle and separate the nerve to protect it. We expose and separate nerves roots and trunks of the brachial plexus, and mark them with vessel loops one by one. Then we transect part of the anterior scalene muscle at the medial border of the phrenic nerve to expose the nerve roots of brachial plexus. Identify the nerve roots of the brachial plexus one by one and mark them with vessel loops.

We separate the C7 nerve proximally to the level of the intervertebral foramen, and inject lidocaine under the epineurium of the C7 nerve trunk. Subsequently we transect the C7 nerve at its migration out of the intervertebral foramen and adequately dissociate and dissect the nerve trunk (Supplementary figure 1). Then we fix the epineurium of severed C7 root to the fascia with 4-0 sutures, at the junction of the omohyoid and sternocleidomastoid muscles. Finally, we rinse the wound after sufficient hemostasis with a negative pressure bulb placed for drainage and suture the interscalene fat pad, vastus cervicis muscle, subcutaneous tissue as well as skin layer by layer.

A video showing the surgical procedure (Supplementary Video 1) is available on the website: https://www.mediafire.com/file/1gn5knyut2b1kmj/Procedure_of_NC7.mp4/file.

Reference

1. Xu WD. Surgical Technique of Xu's CC7 Procedure "Contralateral C7 to C7 Cross Nerve Transfer Through a Trans Longus Colli, Prespinal Route for Treating Spastic Arm". Oper Neurosurg (Hagerstown). 2020 Dec 15;20(1):61-68. doi: 10.1093/ons/opaa325. PMID: 33047132.

2. Li T, Feng J, Hu R, et al. Effect and safety of C7 neurotomy at the intervertebral foramen in patients with chronic poststroke aphasia: a multicenter, randomised, controlled study protocol. BMJ Open. 2023 May 2;13(5):e065173. doi: 10.1136/bmjopen-2022-065173. PMID: 37130672; PMCID: PMC10163524.

2.2.2 Description of perioperative care

As previously mentioned, the NC7 procedure is part of the CC7 procedure. Patients who had received NC7 surgery were similar to the changes of the paralyzed hand in the patients after CC7 surgery. Therefore, we follow through CC7 perioperative early nursing program [1,2] to provide care for postoperative patients of NC7.

Preoperative and intraoperative Medication:

1. Antibiotics, including cephalosporins and azithromycin, would be used during or before the surgery to prevent inflammation.
2. Prevention of Bleeding Preoperatively: Medications such as etamsylate injection and aminocaproic acid can be used preoperatively to prevent intraoperative bleeding.

Postoperative Management:

1. Pain Management Routine:

- Pregabalin: 1 capsule tid (three times a day), with an additional 2 capsules at bedtime if necessary. If daytime pain is significant, the dosage can be adjusted to: 2# tid + 2# qn (at night); typically used for 1 month.
- Gabapentin: 1 capsule tid, which can be increased to 2 capsules tid.
- Non-steroidal Anti-inflammatory Drugs (NSAIDs), Flurbiprofen Ester, and other routine medications.
- These medications may be used in combination or alone, typically for a duration of 3 weeks to 1 month.

2. Postoperative Neural Nutrition: Vitamins B1, B6, and mecobalamin tablets (or cobalamin injection) are recommended, 1# tid po (by mouth), for 4 weeks.
3. No cast or cervical gear were used after NC7.

Reference

1. Su F, Xu Y, Wang X, et al. Huashan perioperative nursing program for stroke patients undergoing contralateral seventh cervical nerve transfer. Perioper Med (Lond). 2022 Apr 7;11(1):12. doi: 10.1186/s13741-022-00245-4.

2. Li J, Ying Y, Su F, et al. The Hua-Shan rehabilitation program after contralateral seventh cervical nerve transfer for spastic arm paralysis. Disabil Rehabil. 2022 Feb;44(3):404-411. doi: 10.1080/09638288.2020.1768597.

2.2.3 Description of iSLT protocol

All participants in this study were provided with a three-week intensive speech therapy program as reported in the previous online protocol [1]spanning 15 consecutive treatment days, excluding weekends and public holidays. Throughout this intervention period, each center was staffed with highly experienced speech pathologists licensed by the Health Commission of China. These professionals conducted one-on-one speech therapy sessions with each patient, lasting 90 minutes per day. Typically, the one-on-one therapy sessions were evenly distributed throughout the day, with sessions scheduled in the morning (between 7 a.m. and 12 p.m.) and in the afternoon (between 1 p.m. and 6 p.m.), each lasting 45 minutes. Upon initiating the intensive speech therapy on the first day, each patient was assigned an account for computerized online speech therapy. Therapists trained both the patients and their families to use this account for self-managed language training [2,3,] requiring at least one hour per day and a minimum of five hours per week. In summary, participants thus received a total 37.5 hours of intensive speech therapy [4,5,6,7].

Daily intensive speech therapy application has delivered with the assistant of a computerized language rehabilitation system designed for native Chinese speakers (Tiger Rehabilitation Medical Technology Co., Ltd., Shanghai) [10,11,12]. The use of the computerized language rehabilitation system aims to enhance the transparency, consistency, and comparability of therapy management across different centers. This system includes a series of sections, each featuring four daily-used vocabulary words belonging to the same category (for example, apple, grape, peach, pear—all fruits). Therapy focuses on these words, encompassing both superficial aspects (such as appearance) and deeper concepts (such as function), understanding and judgment of word meanings, repetition of phrases and sentences containing specific vocabulary, naming of words, reading phrases and sentences that include them, transcription and dictation of specific phrases and sentences, matching words with corresponding images and text, syntactic training, language output training (such as storytelling from pictures), and simulation of everyday communication (through role-playing and scenario simulation). Typically, patients undergo three sections of language therapy each day. Self-managed language training is also conducted through a sophisticated online language therapy software (www.66nao.com) [13]. All patients follow the same content for self-managed language training, but the software adjusts the difficulty level based on the accuracy of the patient's responses, ensuring both consistency and individualization of the training. Researchers closely watch whether the speech therapy aligns with the iSLT treatment plan and provide feedback to speech pathologists on their adherence to the guidelines of the treatment manual. The detailed iSLT rehabilitation programs are listed in the Protocol [1].

Reference

1. Li T, Feng J, Hu R, et al. Effect and safety of C7 neurotomy at the intervertebral foramen in patients with chronic poststroke aphasia: a multicentre, randomised, controlled study protocol. BMJ Open. 2023;13(5):e065173. Published 2023 May 2. doi:10.1136/bmjopen-2022-065173.

2. Palmer, R.; Dimairo, M.; Cooper, C.; Enderby, P.; Brady, M.; Bowen, A.; Latimer, N.; Julious, S.; Cross, E.; Alshreef, A.; et al. Self-managed, computerised speech and language therapy for patients with chronic aphasia post-stroke compared with usual care or attention control (Big CACTUS): a multicentre, single-blinded, randomised controlled trial. The Lancet. Neurology 2019, 18, 821-833, doi:10.1016/s1474-4422(19)30192-9.

3. Breitenstein, C.; Grewe, T.; Flöel, A.; Ziegler, W.; Springer, L.; Martus, P.; Huber, W.; Willmes, K.; Ringelstein, E.B.; Haeusler, K.G.; et al. Intensive speech and language therapy in patients with chronic

aphasia after stroke: a randomised, open-label, blinded-endpoint, controlled trial in a health-care setting. Lancet (London, England) 2017, 389, 1528-1538, doi:10.1016/s0140-6736(17)30067-3.
4. Stahl, B.; Mohr, B.; Büscher, V.; Dreyer, F.R.; Lucchese, G.; Pulvermüller, F. Efficacy of intensive aphasia therapy in patients with chronic stroke: a randomised controlled trial. Journal of neurology, neurosurgery, and psychiatry 2018, 89, 586-592, doi:10.1136/jnnp-2017-315962.
5. Doppelbauer, L.; Mohr, B.; Dreyer, F.R.; Stahl, B.; Büscher, V.; Pulvermüller, F. Long-Term Stability of Short-Term Intensive Language-Action Therapy in Chronic Aphasia: A 1-2 year Follow-Up Study. Neurorehabilitation and neural repair 2021, 35, 861-870, doi:10.1177/15459683211029235.
6. Monnelly, K.; Marshall, J.; Dipper, L.; Cruice, M. Intensive and comprehensive aphasia therapy-a survey of the definitions, practices and views of speech and language therapists in the United Kingdom. International journal of language & communication disorders 2023, 58, 2077-2102, doi:10.1111/1460-6984.12918.
7. Dosage, Intensity, and Frequency of Language Therapy for Aphasia: A Systematic Review-Based, Individual Participant Data Network Meta-Analysis. Stroke 2022, 53, 956-967, doi:10.1161/strokeaha.121.035216.
8. Zhang Tong, Li Shengli, Bai Yulong, et al. Expert consensus on clinical management of post-stroke aphasia. Chinese Journal of Rehabilitation Theory and Practice, 2022;28(01):15-23. (Chinese)
9. Chen Zhuoming. Current status and prospects of computer-assisted assessment of language disorders. Journal of Chinese Physical Medicine and Rehabilitation, 2005;(02):62-64.(Chinese)
10. Wang Xiaowen, Tang Runkai, Qian Hong, et al. The effect of speech and language training therapy under the ICF framework on naming disorders in stroke patients. Journal of Audiology and Speech Pathology, 2021;29(05):499-503.(Chinese)
11. Ge Zhengqing, Liu Jie, Zhao Yingchu, et al. Precise rehabilitation of aphasia naming disorder under the concept of ICF. China Rehabilitation, 2020;35(02):65-67.(Chinese)
12. Cao Keyong, Wang Ying, Huang Zhaoming, et al. Language cognitive assessment training and Communication Instrument treatment for stroke patients with repetition disorders. Journal of Audiology and Speech Pathology, 2021;29(04):411-414. (Chinese)
13. Tang, Y.; Xing, Y.; Zhu, Z.; He, Y.; Li, F.; Yang, J.; Liu, Q.; Li, F.; Teipel, S.J.; Zhao, G.; et al. The effects of 7-week cognitive training in patients with vascular cognitive impairment, no dementia (the Cog-VACCINE study): A randomized controlled trial. Alzheimer's & dementia: the journal of the Alzheimer's Association 2019, 15, 605-614, doi:10.1016/j.jalz.2019.01.009.

2.3 Methods for Functional Magnetic Resonance Imaging (fMRI) Assessments

2.3.1 Data acquisition

All imaging data were acquired using a GE MR750 3.0T scanner. During each scan, patients were in a supine position and could see the screen through a mirror placed above the head coil. An MRI-compatible microphone was placed near the patient's mouth. The following gradient echo-planar sequence parameters were used for the acquisition of 4D functional images: TR = 2,000 ms, TE = 30.0 ms, FA = 90 degrees, FOV = 240 mm * 240 mm, resulting in a voxel resolution of 3.5 mm * 3.5 mm * 4.2 mm. The images acquired in the first 12 s were discarded because of magnetic field instability. The following 3D-SPGR sequence parameters were used for the acquisition of high-resolution structural images: TR = 8.2 ms, TE = 3.2 ms, FA = 8 degrees, FOV = 256 mm * 256 mm, resulting in a voxel resolution of 1 mm * 1 mm * 1 mm.

2.3.2 Picture naming task

Patients performed a picture naming task during each scan. A total of 180 line-drawings were selected from the Snodgrass and Vanderwart (1980) corpus of pictures[1] and were used as picture stimuli in different scans. In each scan, 90 pictures were distributed into three runs, and were presented in a random order. Baseline scans were acquired before surgery or rehabilitation using 90 pictures. Another two scans were acquired three weeks and six months after surgery in the NC7 plus iSLT group, or after rehabilitation in the iSLT group, respectively, using 45 pictures the same as those used in the baseline scans, and 45 novel pictures.

Each trial began with a 400-ms fixation point at the center of the screen, and then each picture was presented for 6,000 ms. The patients were asked to overtly name the object in the picture during picture presentation. Inter-trial interval was 3,000 to 7,000 ms during which a blank screen was presented. Immediately prior to formal scanning, patients underwent training and practice outside and inside the scanner, so that full comprehension on the task was achieved.

2.3.3 Data analysis

There were 23 patients in NC7 plus iSLT group and 20 patients in iSLT group who successfully completed all three scans at baseline, month1, and month 6. One patient in NC7 plus iSLT group was excluded due to technical error, and another patient in NC7 plus iSLT group was excluded due to poor cooperation (did not response to any stimulus during two of three runs) identified by inspecting the audio files recorded during each scan.

Neuroimaging data were analyzed using FSL (FMRIB Software Library, version 6.00)[2]. Standard preprocessing procedures were run including high-pass filtering (100s cut-off), rigid-body motion correction, slice timing correction, and spatial smoothing with a Gaussian full-width, half-max (FWHM) of 6 mm. Individual functional images were coregistered to the structural image using the FLIRT program in FSL[3], and individual structural images were registered to the standard MNI space[4] using ANTs (Advanced Normalization Tool) package[5] optimized for lesioned brains.

In the first-level analysis, a univariate general linear model (GLM) analysis was performed for each run with overt picture naming modeled by one regressor that covered all picture presentation phases (6000ms in each trial) [6]. Head motion parameters were added as covariates of no interest. Inter-trial intervals served as resting baseline. Contrast images of overt picture naming versus rest were calculated. At the second level, beta estimate maps of the three runs within each patient at each time point were averaged[7]. The averaged beta maps were then coregistered to the MNI space to perform the whole-brain correlation analyses across individuals.

Lesion map of each patient was estimated with LINDA package (Lesion Identification with Neighborhood Data Analysis)[8] in R based on the structural image collected at baseline and manually corrected using ITK-SNAP Software[9]. Individual lesion maps were then transformed to the MNI space and overlapped within NC7 plus iSLT group and iSLT group, respectively. Brain areas with a lesion overlap of over 20% patients were excluded from further analyses.

To account for the relationship between brain activities and language functions, we did a whole-brain voxel-wise correlation analysis between patients' beta maps and BNT scores in NC7 plus iSLT group and iSLT group, at baseline, month1, and month 6, respectively. Pearson's r maps were calculated in Matlab 2021a, with a threshold of $p < 0.005$, and underwent cluster-wise correction of $p < 0.05$. Clusters of over 150 voxels were presented in the final results. The original analyses were performed in volumetric space and projected to brain semi-inflated pial surface using FreeSurfer software (version 7.4.1) for visualization[10].

References:

[1] Snodgrass JG, Vanderwart M. A standardized set of 260 pictures: norms for name agreement, image agreement, familiarity, and visual complexity. J Exp Psychol Hum Learn. 1980 Mar;6(2):174-215.

[2] M.W. Woolrich, S. Jbabdi, B. Patenaude, M. Chappell, S. Makni, T. Behrens, C. Beckmann, M. Jenkinson, S.M. Smith. Bayesian analysis of neuroimaging data in FSL. NeuroImage, 45:S173-86, 2009.

[3] Jenkinson, M., Bannister, P., Brady, J. M. and Smith, S. M. Improved Optimisation for the Robust and Accurate Linear Registration and Motion Correction of Brain Images. NeuroImage, 17(2), 825-841, 2002.

[4] VS Fonov, AC Evans, K Botteron, CR Almli, RC McKinstry, DL Collins and BDCG, Unbiased average age-appropriate atlases for pediatric studies, NeuroImage,Volume 54, Issue 1, January 2011, ISSN 1053–8119.

[5] Tustison, N.J., Cook, P.A., Holbrook, A.J. et al. The ANTsX ecosystem for quantitative biological and medical imaging. Sci Rep 11, 9068 (2021).

[6] Woolrich, M. W., Ripley, B. D., Brady, M., & Smith, S. M. (2001). Temporal Autocorrelation in Univariate Linear Modeling of FMRI Data. NeuroImage, 14(6), 1370–1386.

[7] Woolrich, M. W., Behrens, T. E. J., Beckmann, C. F., Jenkinson, M., & Smith, S. M. (2004). Multilevel linear modelling for FMRI group analysis using Bayesian inference. NeuroImage, 21(4), 1732–1747.

[8] Pustina, D., Coslett, H.B., Turkeltaub, P.E., Tustison, N., Schwartz, M.F. and Avants, B. (2016), Automated segmentation of chronic stroke lesions using LINDA: Lesion identification with neighborhood data analysis. Hum. Brain Mapp., 37: 1405-1421.

[9] Paul A. Yushkevich, Joseph Piven, Heather Cody Hazlett, Rachel Gimpel Smith, Sean Ho, James C. Gee, and Guido Gerig. User-guided 3D active contour segmentation of anatomical structures: Significantly improved efficiency and reliability. Neuroimage. 2006 Jul 1; 31(3):1116-28.

[10] Dale, A.M., Fischl, B., Sereno, M.I., 1999. Cortical surface-based analysis. I. Segmentation and surface reconstruction. Neuroimage 9, 179-194.

2.4. Previous Research Trajectory

In 2018, we published our pioneering work in the ***New England Journal of Medicine (NEJM),*** detailing a surgical treatment, **Contralateral C7 nerve transfer (CC7),** for spastic arm paralysis caused by chronic cerebral injury. This innovative procedure has enhanced motor functions in thousands of patients. Subsequently, we conducted comprehensive post-surgical follow-up investigations. Notably, besides motor disability, a significant number of patients treated with CC7 in our department also exhibit aphasia, a finding consistent with the fact that approximately one-third of stroke survivors suffer from this condition. Interestingly, many patients self-reported improvements in language function after undergoing CC7 surgery, before nerve regeneration taking effect. This clinical observation prompted us to explore ***C7 neurotomy at the intervertebral foramen (NC7)*** as a potential approach to enhance language function. To clearly present our research trajectory and the conceptual foundation of NC7, we have outlined a concise step-by-step process and delineated a timeline (available at

https://www.mediafire.com/view/ka0xis5eq31rfmq/Research_Trajectory_Timeline_for_CC7_to_NC7.png/file) for reference.

Step 1. Observational Insights and Patient Self-Reports.

Our attention was drawn to a curious observation that patients with right arm paralysis and aphasia due to the left hemisphere damage were frequently **self-reporting significant language improvements following CC7 surgery especially in naming ability.** This observation dates back to soon after the publication, "Trial of Contralateral Seventh Cervical Nerve Transfer for Spastic Arm Paralysis," on January 4, 2018, in the NEJM journal. This kind of immediate post-surgical enhancements in language function sparked our research interest.

Step 2. Prospective Cohort Study on Language Improvement.

Motivated by patient self-reports of language improvements, we conducted **a prospective cohort study** in 2022, focusing on patients combining spastic arm paralysis with aphasia who underwent CC7 surgery to validate these self-reports. We shared our findings on ***MedRxiv*** by May 24, 2023, demonstrating that CC7 not only enhances motor function but also improves the aphasia quotient in these patients. Following this, this study has been published on ***European Journal of Neuroscience*** on June 3, 2024.

Step 3. Emergence of a New Strategy.

Given the language improvements observed immediately post-CC7 surgery, before the transferred C7 nerve regeneration, we speculated an alternate mechanism may be playing a role. Drawing on insights from our previous studies on neural plasticity, particularly the impact of **C7 neurotomy on inter-hemispheric functional connections** and **dorsal root ganglia (DRG) changes**, as well as altered brain activity in **animal models**, we proposed that these language function improvements might stem from the neurotomy's effect on neural networks. Specifically, our findings on decreased inter-hemispheric connectivity following brachial plexus injury, combined with the anatomical and imaging evidence of DRG alterations post-surgery, support a neural plasticity-driven mechanism. These studies led us to the hypothesis that **right C7 neurotomy at the intervertebral foramen (NC7)** could serve as a novel approach to improve language function in patients with post-stroke aphasia, using changes in brain connectivity and activity patterns influenced by the surgery.

Step 4. NC7 Pilot Study Design and Preliminary Findings.

To verify the hypothesis, we designed **a prospective single-arm pilot study** to determine whether

combining NC7 surgery with three weeks of intensive speech and language therapy (iSLT) can significantly enhance language recovery. We shared our preliminary NC7 results on ***medRxiv*** in March 2023. The single-arm study design, while imperfect, was an essential step in introducing NC7 to the medical community and advancing our understanding and treatment of post-stroke aphasia.

Step 5. Expanding to a Multi-Center Controlled Trial.

With the groundwork laid and the positive outcomes of the pilot trial motivating us to expand our research scope, we then planned and commenced **a multi-center, randomized, assessor-blinded, controlled trial**. This study was designed to provide a robust evaluation of NC7's efficacy. The trial protocol was subsequently published in ***BMJ Open*** on May 2023, signifying our continued dedication to pioneering treatments that offer hope and recovery for those affected by stroke-induced language impairments.

Through these methodical steps, the trajectory of our research from initial CC7 findings to the broader investigation of NC7 and its effects on language recovery post-stroke becomes clearer. Our dedication to evolving our work, continuing to recruit and follow up with participants in ongoing studies, and remain committed to refining our understanding of NC7 and its potential to revolutionize rehabilitation for aphasia patients worldwide.

3. Supplementary Tables

Table S1. Mean score at the indicated follow-up.

Outcome	NC7 plus iSLT group（N=25） Mean (SD)	iSLT group（N=25） Mean (SD)
BNT score †		
day 3	26.48 (12.71)	22.36 (13.01)
month 1	32.36 (15.35)	24.88 (14.23)
month 6	31.44 (16.11)	24.25 (13.95)
WAB-AQ ‡		
day 3	55.26 (14.38)	50.74 (14.14)
month 1	59.52 (14.60)	53.74 (14.13)
month 6	57.52 (14.60)	51.97 (16.02)
Barthel index ‖		
day 3	82.20 (12.92)	84.40 (15.23)
month 1	86.80 (12.98)	83.60 (16.99)
month 6	88.80 (12.10)	86.09 (15.30)
SADQ-H10 ¶		
day 3	4.60 (2.45)	4.12 (2.77)
month 1	3.56 (2.16)	4.20 (2.83)
month 6	2.64 (1.91)	4.13 (2.18)

* Day 3 referred to 3 days post-NC7 in the NC7 plus iSLT group or day 3 from iSLT deferral in the iSLT group; month 1 referred to 1-month from surgery or beginning of iSLT deferral; month 6 referred to 24 weeks after iSLT onset; NC7, C7 neurotomy at the intervertebral foramen; iSLT, intensive speech and language therapy.

† The Boston Naming Test (BNT) scale is a measure of language impairment; scores range from 0 to 60, with higher scores indicating better function.

‡ The Western Aphasia Battery - Aphasia Quotient (WAB-AQ) is a weighted average of all subtest scores relating to spoken language, measuring language ability; scores range from 0 to 100. Negative numbers indicate a decrease and positive numbers an increase in language function from baseline to day 3, month 1 and month 6.

‖ The Barthel Index measures the degree of assistance required by an individual on ten mobility and self-care activity of daily living items; scores range from 0 to 100, with higher scores indicating better functional in daily activities. Negative numbers indicate a decrease and positive numbers an increase functional independence from baseline to day 3, month 1 and month 6.

¶ The Stroke Aphasic Depression Questionnaire Hospital Version (SADQ-H10) is scored by assigning corresponding numeric values to observer selections (range from 0-3), with a higher score indicating more depressive symptoms. Total score of SADQ-H10 is 30. Negative numbers indicate a decrease and positive numbers an increase depression symptom from baseline to day 3, month 1 and month 6.

Table S2. Stability of changes in BNT score at month 1 and month 6 in both groups.

Time point	Month 1 Mean (S.D.)	Month 6 Mean (S.D.)	Mean difference† (95%CI)	Estimated mean difference‡ (95% CI)
NC7 plus iSLT group	11.16 (7.10)	10.24 (8.75)	-0.92 (-3.47 to 1.63)	-0.92 (-3.49 to 1.65)
iSLT group	2.72 (3.40)	2.08 (3.99)	-0.79 (-2.19 to 0.61)	-0.71 (-3.31 to 1.90)

* The mixed model was conducted to analysis the longitudinal BNT and WAB score. The treatment group, interaction of the treatment group and time, the center and the score at baseline were included as fixed factors. The model also included a patient-specific random intercept. Using this model, we estimated marginal means for each time point and treatment group.

† Mean difference measures the absolute difference between the mean value in two time points.

‡ With the mixed model, we estimated marginal means for each time point and the difference values.

Table S3. Changes in BNT and WAB-AQ scale with mixed effect model*.

Outcome	NC7 plus iSLT group（N=25）	iSLT group（N=25）	Difference (95% CI)
Change from baseline (95%CI)			
BNT score			
day 3	5.30 (3.17 to 7.44)	0.18 (-1.96 to 2.31)	5.12 (2.11 to 8.14)
month 1	11.18 (9.05 to 13.32)	2.70 (0.56 to 4.83)	8.48 (5.47 to 11.5)
month 6	10.26 (8.13 to 12.40)	1.99 (-0.17 to 4.16)	8.27 (5.23 to 11.31)
WAB-AQ total			
day 3	6.85 (5.13 to 8.56)	1.01 (-0.70 to 2.73)	5.83 (3.41 to 8.26)
month 1	11.11 (9.40 to 12.83)	4.01 (2.30 to 5.73)	7.10 (4.67 to 9.52)
month 6	9.11 (7.39 to 10.82)	2.86 (1.10 to 4.61)	6.25 (3.80 to 8.70)

* The mixed model was conducted to analysis the longitudinal BNT and WAB score. The treatment group, interaction of the treatment group and time, the center and the score at baseline were included as fixed factors. The model also included a patient-specific random intercept. Using this model, we estimated marginal means for each time point and treatment group.

Table S4. Sensitivity analysis of primary outcome.

Outcome	NC7 plus iSLT group (N=25)	iSLT group (N=25)	Adjusted Mean Difference* (95% CI)
Sensitivity Analysis 1			
BNT imputed by LOCF method			
Change in total BNT score from baseline to day 3	5·3 (3·17 to 7·43)	0·18 (-1·95 to 2·31)	5·12 (2·11 to 8·14)
Change in total BNT score from baseline to month 1	11·18 (9·05 to 13·31)	2·7 (0·57 to 4·83)	8·48 (5·47 to 11·5)
Change in total BNT score from baseline to month 6	10·26 (8·13 to 12·39)	1·94 (-0·19 to 4·07)	8·32 (5·31 to 11·34)
Sensitivity Analysis 2			
BNT imputed by mean imputation			
Change in total BNT score from baseline to day 3	5·3 (3·17 to 7·43)	0·18 (-1·95 to 2·31)	5·12 (2·11 to 8·13)
Change in total BNT score from baseline to month 1	11·18 (9·05 to 13·31)	2·70 (0·57 to 4·83)	8·48 (5·47 to 11·49)
Change in total BNT score from baseline to month 6	10·26 (8·13 to 12·39)	2·07 (-0·06 to 4·2)	8·19 (5·18 to 11·2)
Sensitivity Analysis 3			
BNT imputed by regression imputation			
Change in total BNT score from baseline to day 3	5·30 (3·17 to 7·43)	0·18 (-1·95 to 2·31)	5·12 (2·11 to 8·13)
Change in total BNT score from baseline to month 1	11·18 (9·05 to 13·31)	2·70 (0·57 to 4·83)	8·48 (5·47 to 11·49)
Change in total BNT score from baseline to month 6	10·26 (8·13 to 12·39)	2·06 (-0·07 to 4·19)	8·20 (5·19 to 11·21)

Table S5. Investigator-assessed results of the BNT score and WAB-AQ score.

	NC7 plus iSLT group (N=25)	iSLT group (N=25)	Adjusted Mean Difference* (95% CI)
BNT			
Change in total BNT score from baseline to day 3	5.20 (3.77)	-0.08 (2.00)	5.26 (3.5 to 7.03)
Change in total BNT score from baseline to month 1	11.12 (7.01)	2.52 (3.47)	8.65 (5.45 to 11.85)
Change in total BNT score from baseline to month 6	10.12 (9.18)	1.96 (3.94)	8.24 (3.98 to 12.51)
WAB-AQ			
Change in total WAB-AQ score from baseline to day 3	6.77 (3.16)	0.75 (3.35)	5.90 (4.09 to 7.72)
Change in total WAB-AQ score from baseline to month 1	10.97 (5.25)	3.83 (3.77)	7.01 (4.41 to 9.61)
Change in total WAB-AQ score from baseline to month 6	9.25 (5.13)	3.08 (4.56)	6.21 (3.28 to 9.15)

*Adjusted mean changes from baseline to day 3, month 1 and month 6 based on analysis of covariance (ANCOVA) were presented, with the model adjusted for baseline values and study center as covariates.

Table S6. Brain areas with activation correlated with the BNT score in the NC7 plus iSLT group.

Timepoint	Region	MNI Coordinate			Number of Voxels	Peak r
		X	Y	Z		
Month 1	Right IFG (pars opercularis)	40	12	16	217	0.68
	Right SMG	68	-24	38	151	0.72
Month 6	Right SMG	56	-32	50	189	0.74
	Left ITG	-54	-52	-8	182	0.75

Pearson's correlation between the session-averaged parameter estimation maps and BNT scores was performed across subjects, with a threshold of $P< 0.005$, and underwent cluster-wise correction of $P < 0.05$. Clusters of over 150 voxels were presented above. No clusters larger than 150 voxels survived at baseline. IFG: inferior frontal gyrus. SMG: supramarginal gyrus. ITG: inferior temporal gyrus.

Table S7. Brain areas with activation correlated with the BNT score in iSLT group.

Timepoint	Region	MNI Coordinate			Voxels in Cluster	Peak r
		X	Y	Z		
Month 1	Left Precentral gyrus	-50	8	36	258	0.78
	Left Fusiform gyrus	-44	-54	-22	233	0.71
	Right Lingual gyrus	22	-80	-8	373	0.73
Month 6	Left MFG	-44	24	46	197	0.78
	Left Precentral gyrus	-46	12	32	193	0.76

Pearson's correlation between the session-averaged parameter estimation maps and BNT scores was performed across subjects, with a threshold of $P< 0.005$, and underwent cluster-wise correction of $P < 0.05$. Clusters of over 150 voxels were presented above. No clusters larger than 150 voxels survived at baseline. MFG: middle frontal gyrus.

Table S8. Perioperative situation of the NC7 plus iSLT group

	Mean (n=25)	**Center 1 (n=10)**	**Center 2 (n=5)**	**Center 3 (n=5)**	**Center 4 (n=5)**
Duration of surgery, mins (mean±SD)	71.74±12.99	72.6±11.44	66±11.94	87.33±12.1	66.4±12.50
Duration of anesthesia, mins (mean±SD)	112.16±28.95	131.1±31.02	88±22.25	119.2±9.23	91.4±7.5
Postoperative drainage volume, ml (mean±SD)					
Day 1 po-op	30.48±16.12	27.6±14.39	42±21.68	26±19.49	29.2±5.4
Day 2 po-op	14.84±11.6	19.7±9.20	15±16.96	12±13.04	7.8±5.36
Day 3 po-op	5.16±14.37	1.7±3.95	17±30.33	5±7.07	0.4±0.89
Patients using analgesics for neuropathic pain post-operatively, n	15	7	3	3	2
Post-operative analgesics duration, days (mean±SD)	31.96±16.04	24.8±16.53	43.2±14.29	30.4±7.44	36.6±18.81

4. Supplementary Figures

Figure S1. Schematic diagram of NC7 surgery

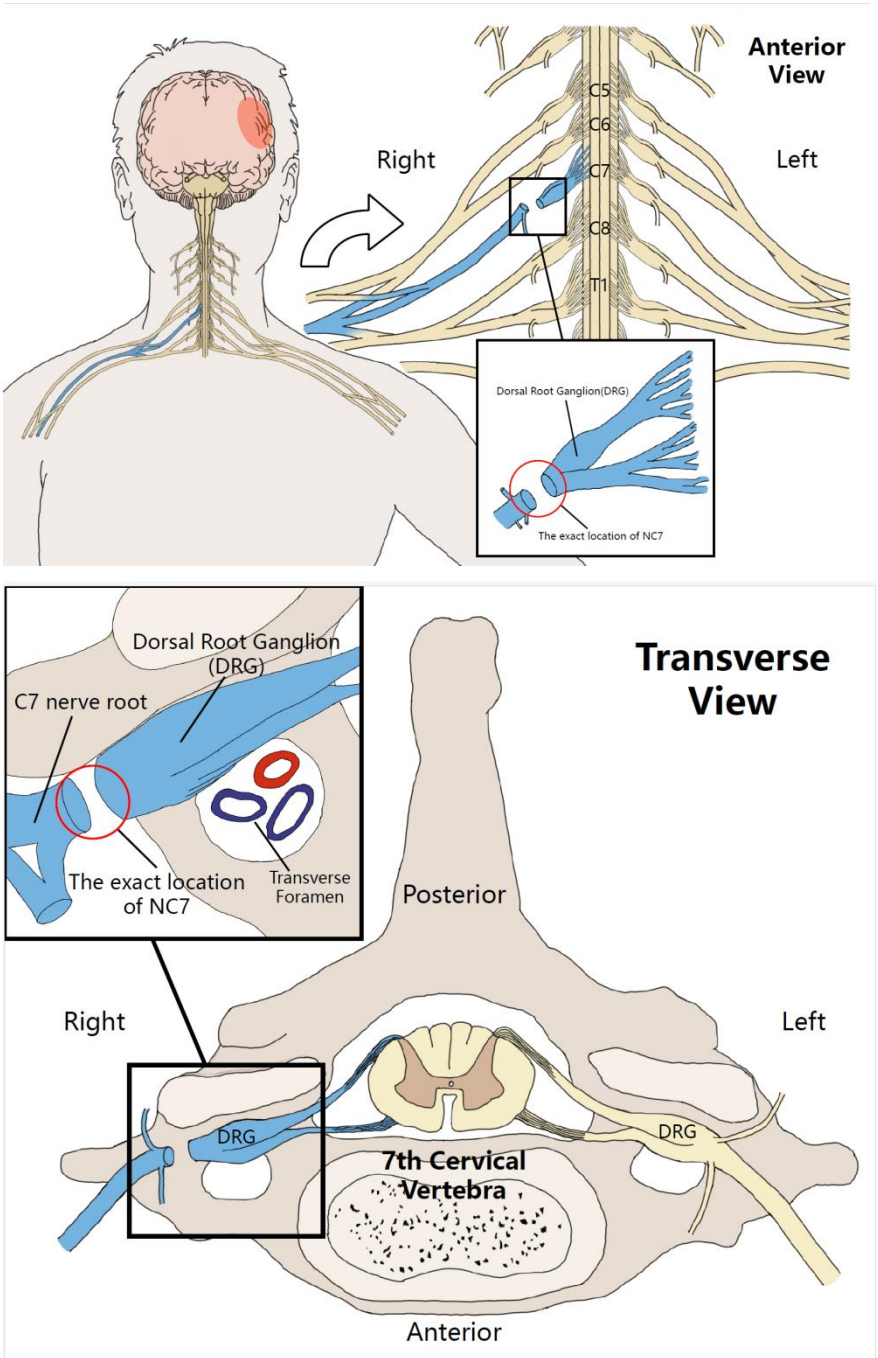
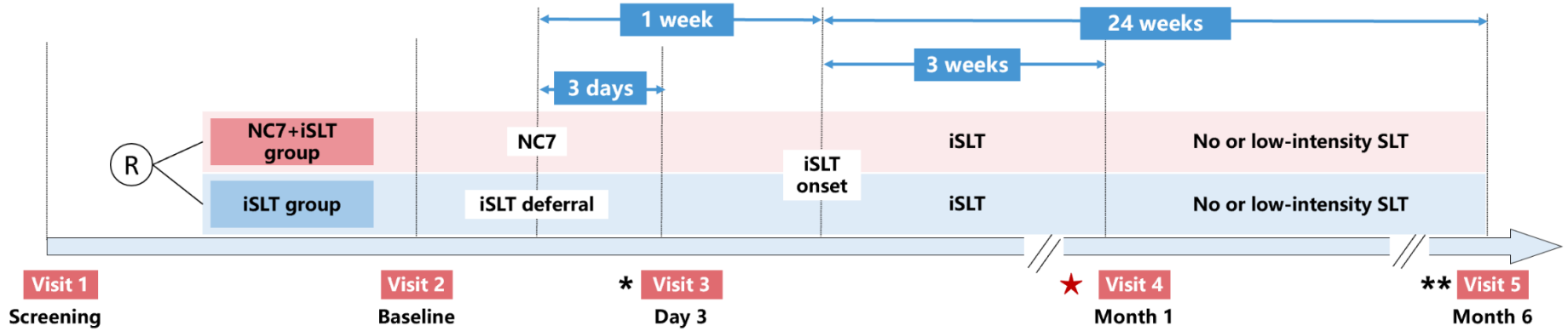


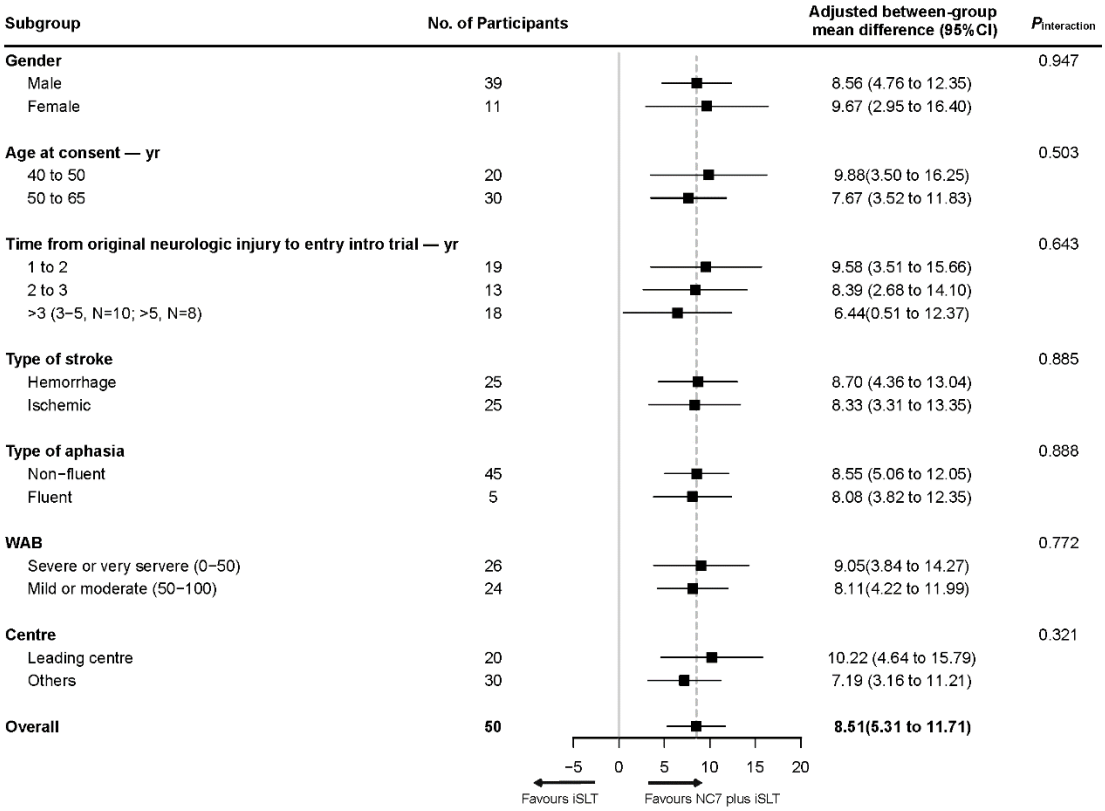
Diagram of the previously described C7 neurotomy (NC7) including anterior view and transverse view. We magnify the exact location of NC7 to show anatomical relationships. We transect the C7 nerve at its migration out of the intervertebral foramen. This is a redrawn figure based on the following source: Zheng MX, Hua XY, Feng JT, et al. Trial of Contralateral Seventh Cervical Nerve Transfer for Spastic Arm Paralysis. N Engl J Med. 2018 Jan 4;378(1):22-34. doi: 10.1056/NEJMoa1615208.)

Figure S2. Trial design diagram



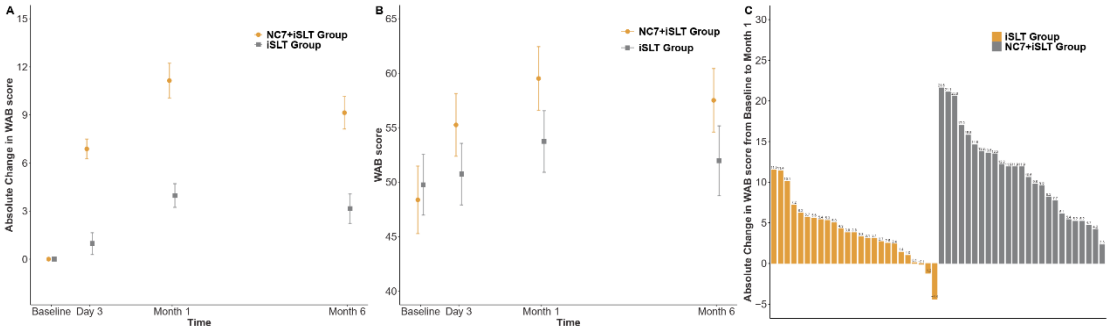
Trial design in detail and patient flow chart. NC7, C7 neurotomy at the intervertebral foramen; iSLT, intensive speech and language therapy; R: Randomization. ★(in red), primary endpoint; *, short-term efficacy assessments; **, long-term efficacy assessments. This is a redrawn figure based on the following source: Li T, Feng J, Hu R, et al. Effect and safety of C7 neurotomy at the intervertebral foramen in patients with chronic poststroke aphasia: a multicenter, randomised, controlled study protocol. BMJ Open. 2023;13(5):e065173. Published 2023 May 2. doi:10.1136/bmjopen-2022-065173.

Figure S3. Prespecified and post-hoc subgroup analysis for the change in BNT score.



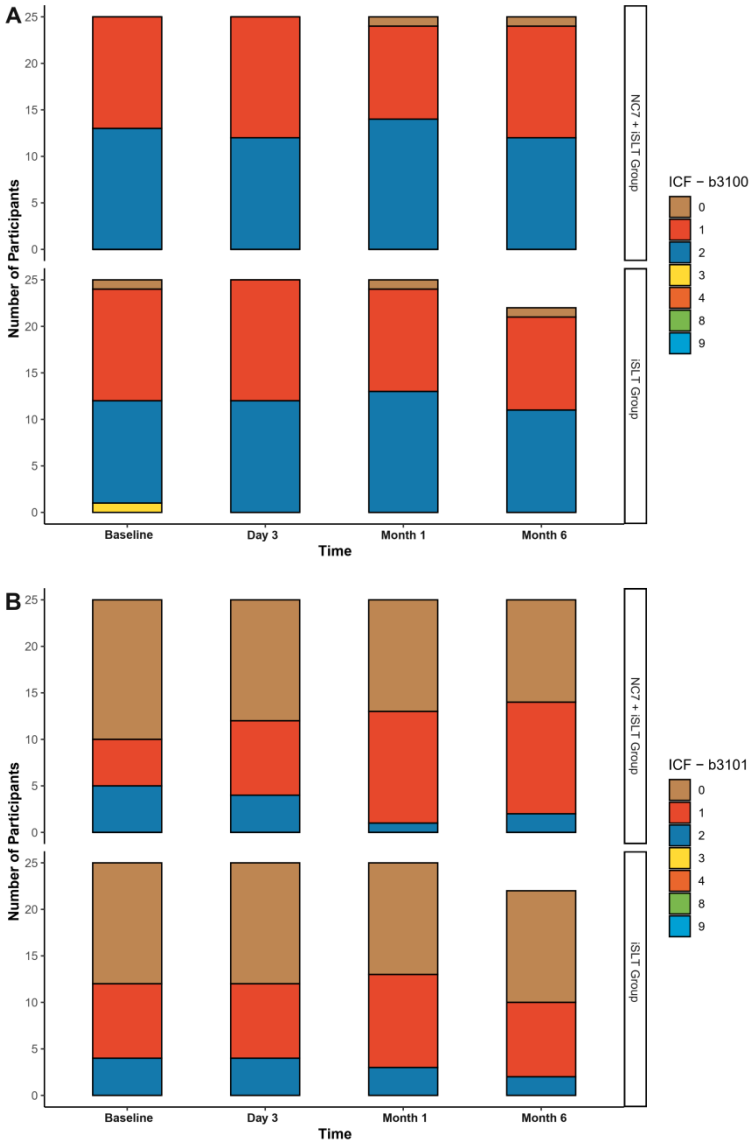
Adjusted between-group mean difference (95%CI) represents the between-group effect of the primary outcome in each subgroup. P-value for interaction shows the results of subgroup effect analysis. WAB denotes Western Aphasia Battery score. Leading center is Huashan hospital, and others include 3 sub-centers.

Figure S4. Changes of WAB-AQ score



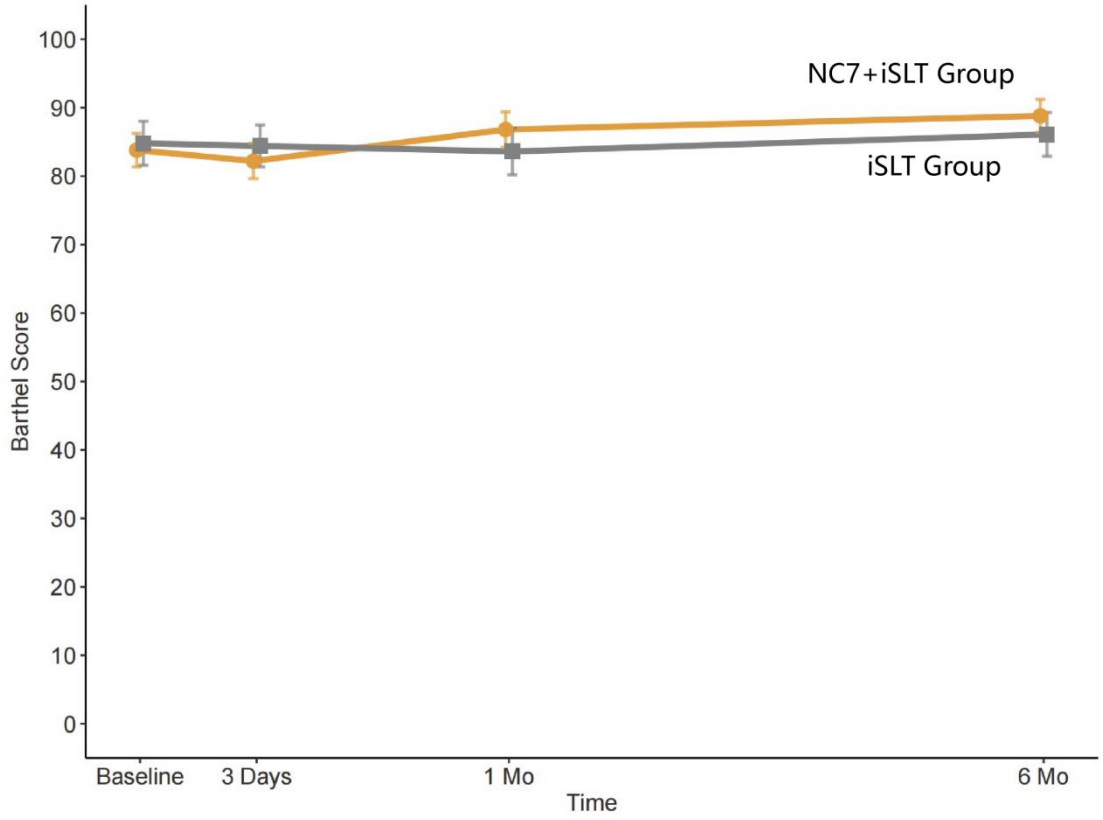
Panels A shows the absolute change from baseline in WAB score for patients who were assigned to NC7 plus iSLT group or iSLT group, and I bars indicate standard errors. Panel B shows the mean WAB scores in the NC7 plus iSLT group or iSLT group, and I bars indicate standard errors. Panel C shows observed data of the absolute change in WAB score from baseline to month 1 after intervention from participants who underwent randomization.

Figure S5. ICF Score at each follow-up



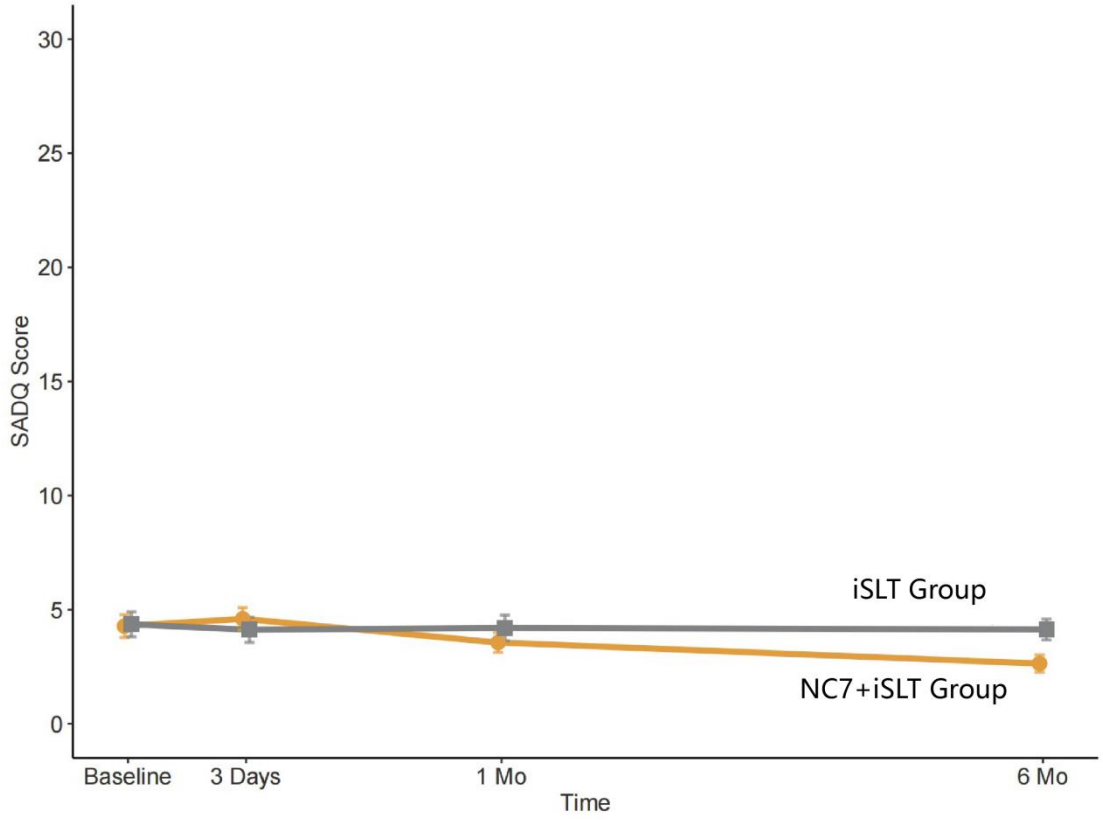
Panel A and B show the distribution of the ICF-b3100 and b3101 class at baseline and each follow-up period. The ICF scale ranges from 0 to 9.

Figure S6. Mean Barthel Index Score at each follow-up



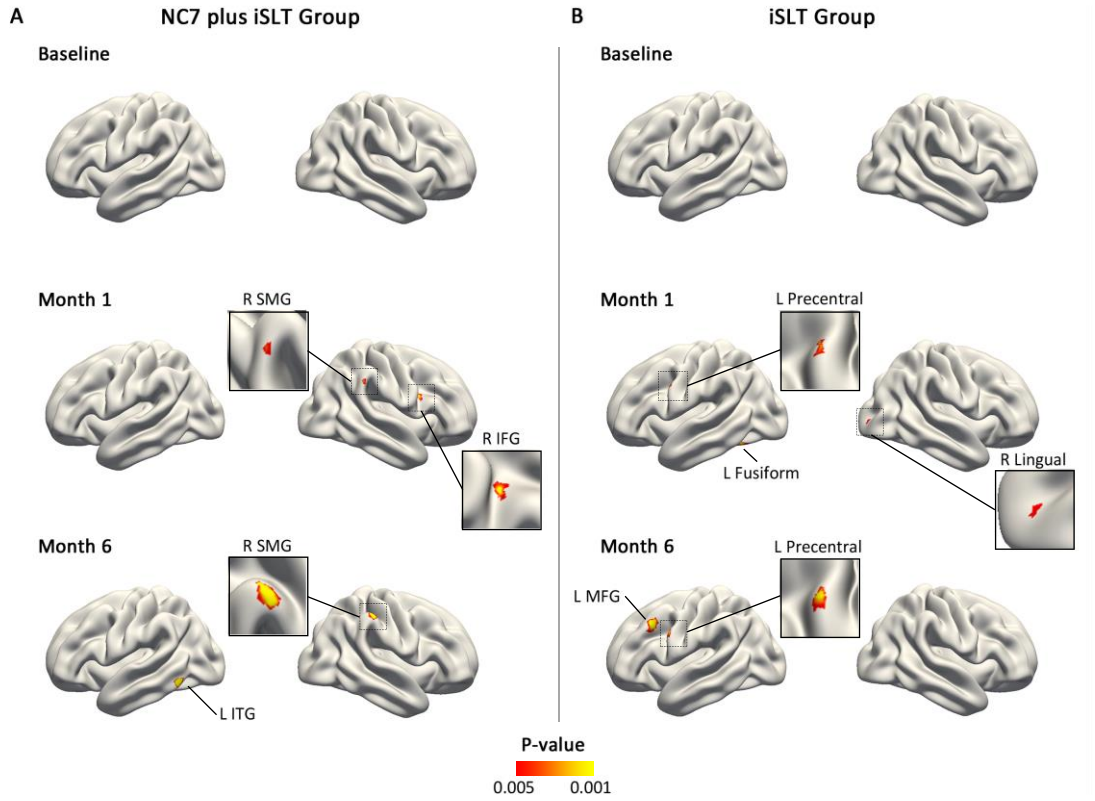
The mean Barthel Index Score in the NC7 plus iSLT group or iSLT, and I bars indicate standard errors.

Figure S7. Mean Stroke Aphasic Depression Questionnaire Score at each follow-up



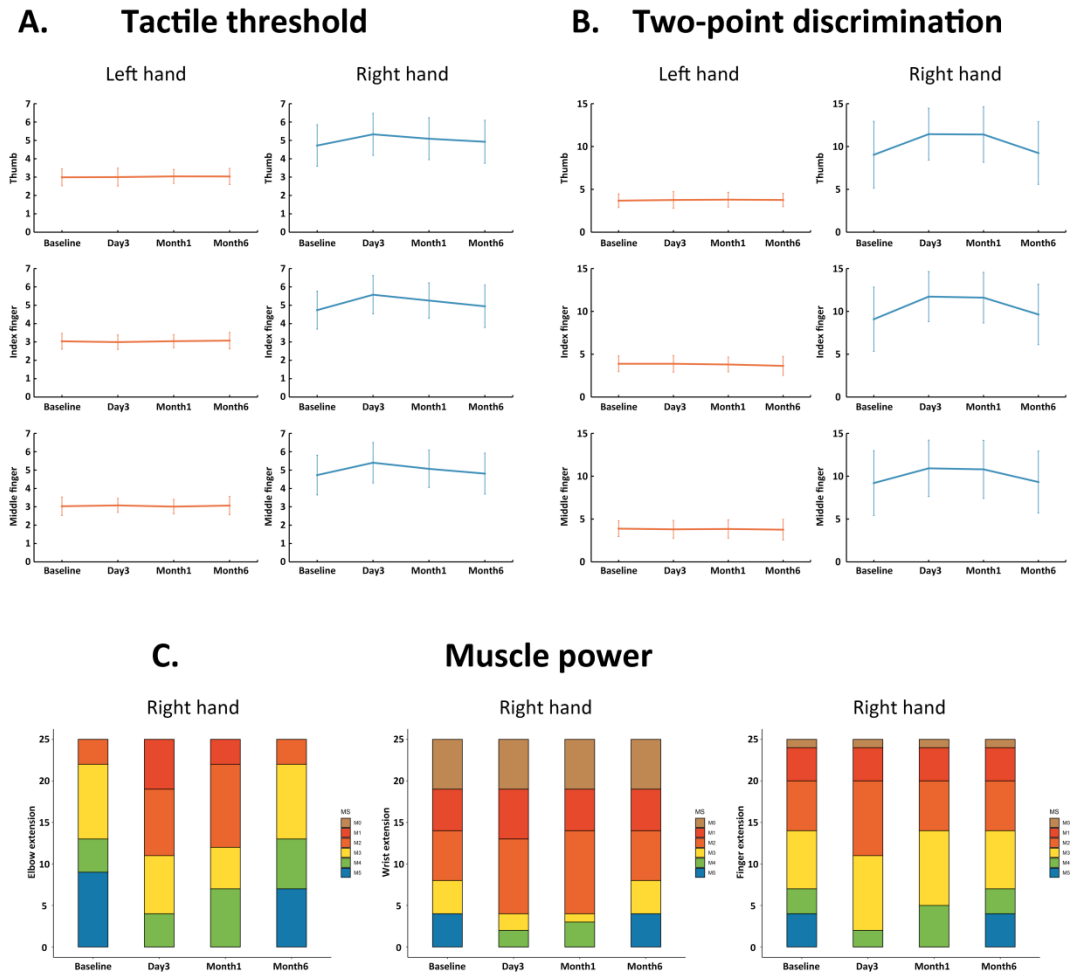
The mean Stroke Aphasic Depression Questionnaire Score in the NC7 plus iSLT group or iSLT, and I bars indicate standard errors.

Figure S8. Correlation map between BNT score and brain activation during overt picture naming in NC7 plus iSLT Group (A) and iSLT Group (B)



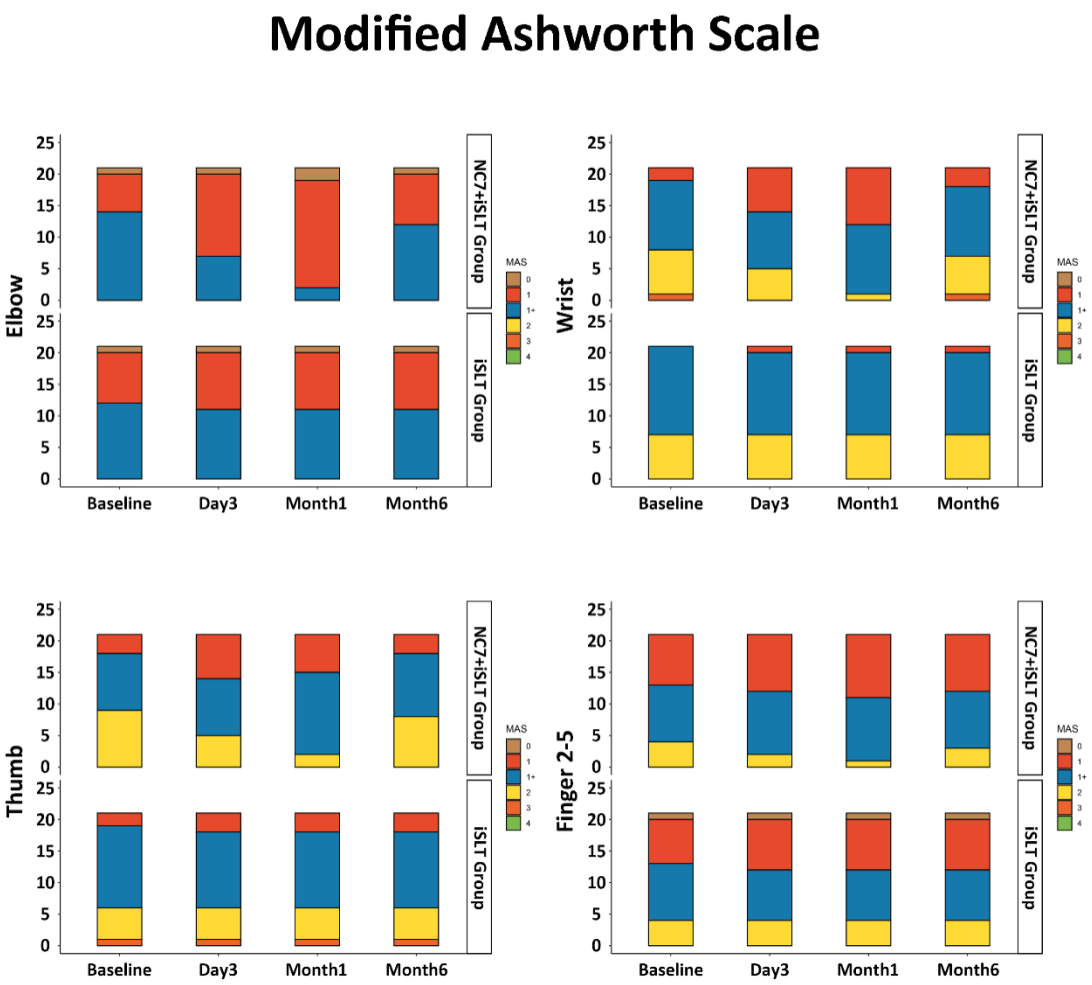
A: In NC7 plus iSLT group, BNT score correlated with brain activation in right supramarginal gyrus (SMG) and left inferior temporal gyrus (ITG) at month 1, and with right SMG and right inferior frontal gyrus (IFG) at month 6. B: In iSLT group, BNT score correlated with brain activations in left precentral gyrus, left fusiform gyrus, and right lingual gyrus at month 1, and with left precentral gyrus, left middle frontal gyrus (MFG) at month 6. No significant cluster was observed at baseline in either group. In both groups, all images were thresholded at $P < 0.005$, and cluster-wise corrected at $P < 0.05$. Inset images are magnified and rotated to better visualize sulcal clusters. L: left. R: right. Precentral: precentral gyrus. Fusiform: fusiform gyrus. Lingual: lingual gyrus. MFG: middle frontal gyrus.

Figure S9. Sensorimotor functions of bilateral hands at each follow-up.



Panel A and B show the tactile threshold and two-point discrimination (2-PD) results in the NC7 plus iSLT group (mean ± SD). Panel C shows the frequency of each muscle power grade in elbow, wrist and finger extension. The vertical coordinate is the number of participants, 25 in total, and the horizontal coordinate is the time point of visit.

Figure S10. Modified Ashworth Scale Score at each follow-up.



In the NC7 plus iSLT group, four patients without limb spasm were excluded from the statistical analysis of the Modified Ashworth Scale. This figure displays the frequency of muscle spasticity levels in the elbow, wrist, thumb, and fingers for patients in the NC7 plus iSLT group. The vertical axis represents the number of participants in the NC7 plus iSLT group (total n = 21), while the horizontal axis indicates the time points of the visits (Baseline, Day 3, Month 1, and Month 6). The different colors within each bar correspond to the Modified Ashworth Scale (MAS) grades.

5. Supplementary Videos

Video S1. Surgical procedure of NC7

This video showing the description of NC7 procedure.

Video S2 to Video S4.

Videos of 9 patients from the NC7 plus iSLT group to show their changes in language function across 6 months follow-up.

Video S5.

Videos of 3 patients from the iSLT alone group to show their changes in language function across 6 months follow-up.