Supplementary Materials

Table of Contents

Abbreviations

BNT – Boston Naming Test

CDT – clock-drawing test

COWAT – Controlled Oral Word Association Test

DAN – dorsal attention network

DMN – default mode network

ECN – executive control network

HVLT – Hopkins Verbal Learning Test

l_DLPFC – left dorsolateral prefrontal cortex

l_FI – left frontal insula

l_HIPP – left hippocampus

l_IFG – left inferior frontal gyrus

l_IPS – left intraparietal sulcus

l_mPFC – left medial prefrontal cortex

l_PCC – left posterior cingulate cortex

l_TPole – left temporal pole

LN – language-related network

MN – memory network

MoCA – Montreal Cognitive Assessment

mRS – modified Rankin Score

NIHSS – National Institute of Health Stroke Scale

r_DLPFC – right dorsolateral prefrontal cortex

r_HIPP – right hippocampus

r_FI – right frontal insula

r_IPS – right intraparietal sulcus

RCF – Rey Complex Figure

SCN – structural covariance network

SN – salience network

TMT-A – trail-making test (A)

TMT-B – trail-making test (B).

1. Missing data percentage at subacute and chronic timepoints

Table S1. Domain-specific neuropsychological tests and percentage of missing data at subacute and chronic timepoint.

		Missing Data	
Cognitive Domain	**Neuropsychological Test**	**Subacute**	**Chronic**
Attention	Digit Span Task (WAIS – Third Edition)	3%	3%
	Digit Symbol Substitution Task (WAIS – Third Edition)	3%	3%
	TMT-A	1%	1%
	Simple reaction time task	1%	1%
	Choice reaction time task	1%	1%
	One-back task	1%	3%
Executive Function	RCF-organisation	4%	5%
	TMT-B	3%	3%
	CDT	1%	1%
Language	COWAT-animals	5%	5%
	COWAT-FAS	4%	4%
	BNT	3%	3%
Memory	HVLT-Delay	0%	1%
	HVLT-Retention	0%	1%
	HVLT-Recognition	0%	1%
	RCF-delay	3%	3%
Visuospatial Function	RCF-copy	3%	4%

Abbreviations: BNT – Boston Naming Test; CDT – Clock-Drawing Test; COWAT – Controlled Oral Word Association Test; HVLT – Hopkins Verbal Learning Test; RCF – Rey Complex Figure; TMT-A – Trail-making test (A); TMT-B – Trail-Making Test (B); WAIS – Weschler Adult Intelligence Scale.

2. Seed region and coordinates of structural covariance networks

Table S2. Selected seed region and coordinates of structural covariance networks.

Network	Seed Region	Coordinates			Reference
		X	Y	Z	
Dorsal Attention Network	Left IPS	-31	-55	54	1
	Right IPS	31	-55	54	1
Executive Control Network	Left DLPFC	-44	36	20	2
	Right DLPFC	44	36	20	2
Salience Network	Left FI	-38	26	-10	2
	Right FI	38	26	-10	2
Default mode network	Left PCC	-7	-43	33	3,4
	Left mPFC	-16	48	44	5,6
Language-related Network	Left TPole	-38	10	-28	7
	Left IFG	-50	18	7	7
Memory Network	Left HIPP	-21	-25	-14	8,9
	Right HIPP	24	-19	-21	8,9

Coordinates shown are in Montreal Neurological Institute normalized space. Abbreviations: DLPFC – dorsolateral prefrontal cortex; FI – Frontal Insula; HIPP – hippocampus; IFG – Inferior frontal gyrus; IPS – intraparietal sulcus; MNI – Montreal Neurological Institute; mPFC – medial prefrontal cortex; PCC – posterior cingulate cortex; TPole – temporal pole.

3. ROI seed and lesion overlap

We used FSLMATHS to calculate the percentage of voxels overlapping between the ROI seeds and the lesion maps, both transformed into MNI space. We calculated that the percentage of voxels overlapping between the seeds and infarcts in the group map totalled less than 0.06%. It is therefore unlikely that damage to the seed regions cause by the infarcts is driving the structural covariance networks observed.

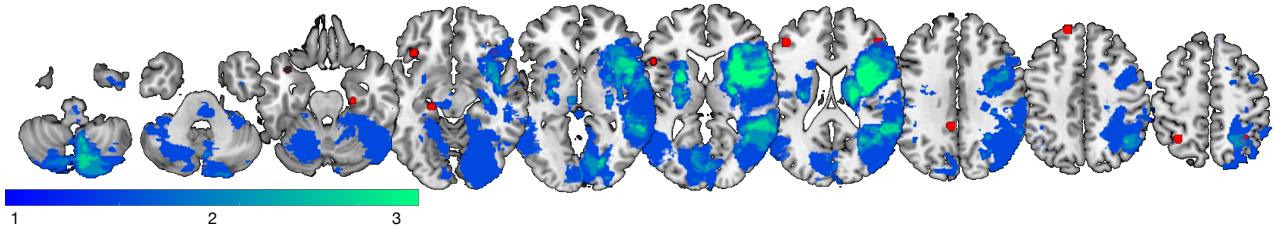


Figure S1. Axial map showing distribution of lesions and overlap with seed ROIs. Seed regions shown in red. Colourbar shows number of patients with overlapping lesions. Neurological orientation (right depicted on the right).

4. Infarct volume control analysis: Behavioural partial least squares analysis without controlling for infarct volume

To examine the influence of infarct volume in confounding the behavioural PLS analysis, we performed two separate analyses without controlling for the log-transformed infarct volume, following the methods outlined for the discovery dataset analysis. The unstandardised residual brain scores (i.e., SCN scores) were input to the following behavioural PLS analyses.

The results of the infarct volume control analysis are illustrated in Figure S2 and S3.

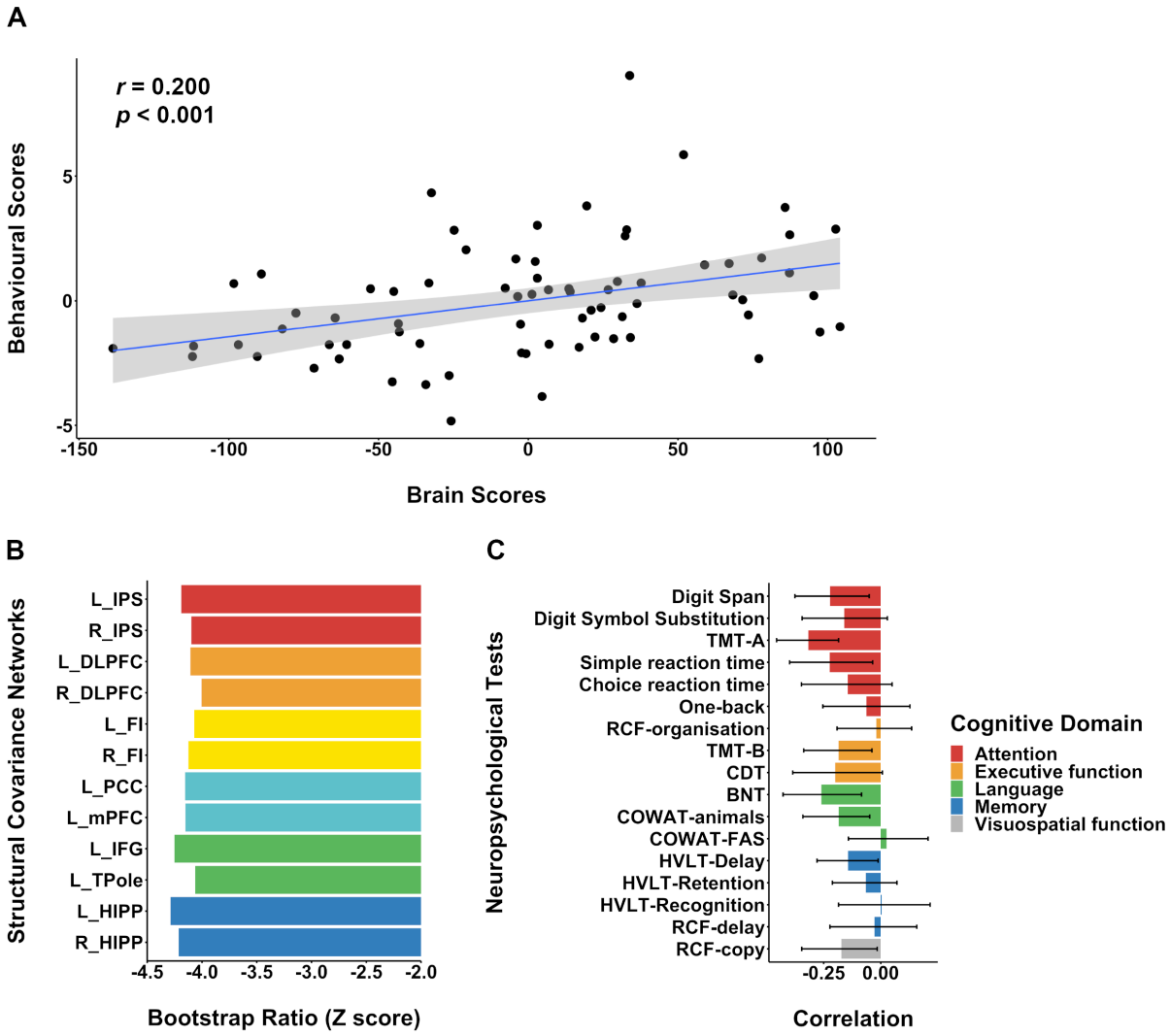


Figure S2. Lower baseline integrity of structural covariance networks was associated with greater impairment in cognitive performance in subacute stroke. These relationships were not affected by infarct volume.

(A) A positive correlation between behavioural and brain scores suggested more damaged SCNs were associated with worse attention, executive function, language, memory, and visuospatial function performance at 3-months post-stroke. (B) The contributions of each SCN

to the covariance between SCNs and neuropsychological tests were revealed by the bootstrap ratio. (C) Extensive negative correlations between SCNs and 17 neuropsychological tests were shown, particularly in the Digit Span Task (r = -0.222, 95% C.I. -0.376 – -0.051), TMT-A (r = -0.317, 95% C.I. -0.456 – -0.185), and simple reaction time task (r = -0.223, 95% C.I. -0.399 – -0.035) within the attention domain, the TMT-B (r = -0.185, 95% C.I. -0.337 – -0.039) and CDT (r = -0.200, 95% C.I. -0.385 – -0.007) within the executive function domain, the BNT (r = -0.260, 95% C.I. -0.427 – -0.084) and COWAT-animals (r = -0.184, 95% C.I. -0.341 – -0.048) within the language domain, the HVLT-Delay (r = -0.143, 95% C.I. -0.279 – -0.012) within the memory domain, and the RCF-copy (r = -0.172, 95% C.I. -0.346 – -0.016) within the visuospatial domain. This infarct volume control analysis also revealed a significant latent variable which could explain 99.84% of the variance of the PLS model.
The error bars indicate 95% confidence interval.

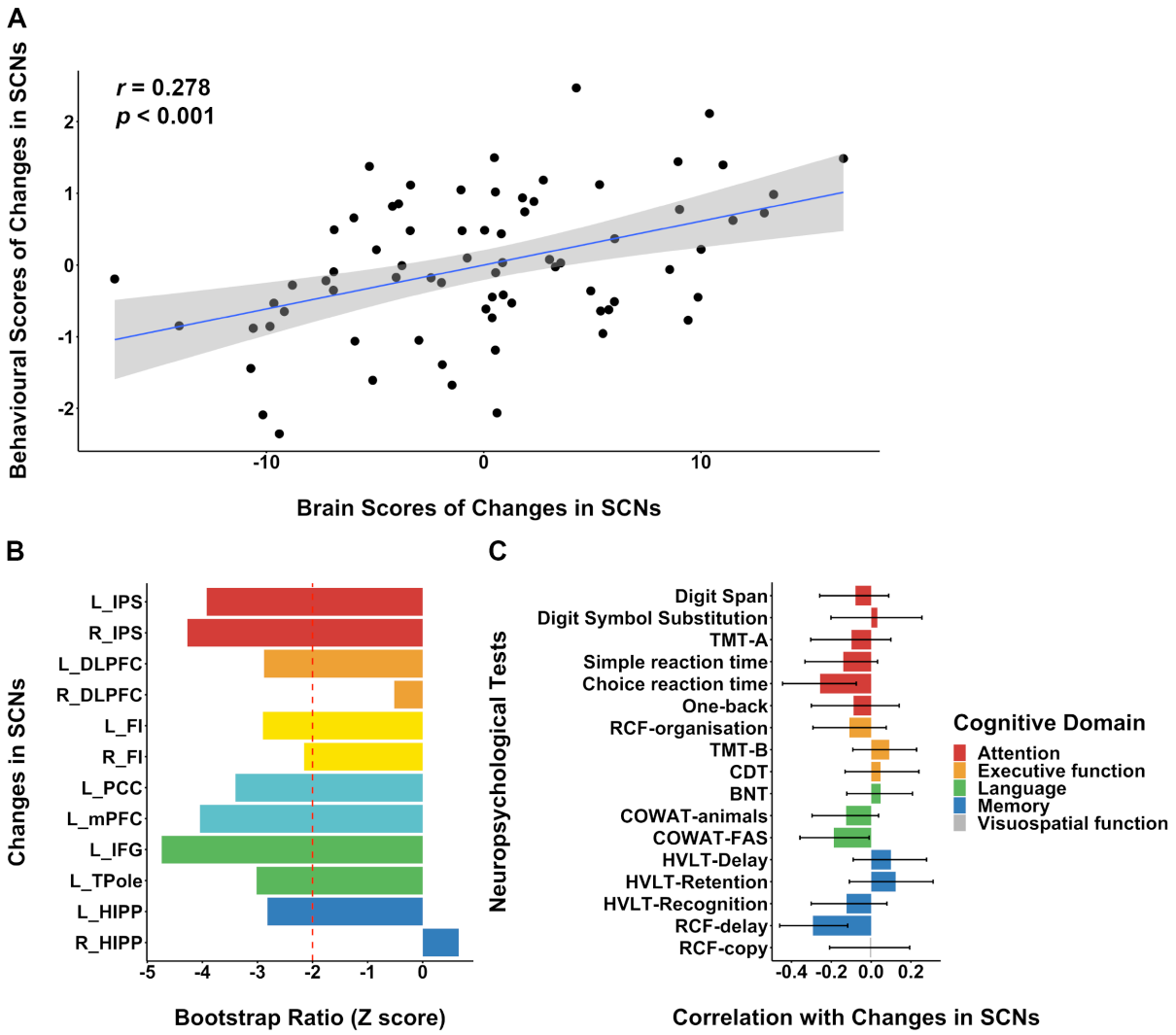


Figure S3. Faster degradation of structural covariance networks was associated with greater longitudinal decline in performance of attention, language and memory from 3-months to 1-year post-stroke (independent of infarct volume).

(A) A positive correlation between behavioural and brain scores suggested faster SCN decline was associated with greater longitudinal decline in performance in neuropsychological tests from 3-months to 1-year post-stroke. (B) The contribution of each SCN to the covariance between SCNs and neuropsychological tests were revealed by the bootstrap ratio. (C) The significant correlation between each neuropsychological test and SCNs was shown in the choice reaction time task (r = -0.256, 95% C.I. -0.445 – -0.074) within the attention domain, the COWAT-FAS (r = -0.188, 95% C.I. -0.357 – -0.010) within the language domain, and the RCF-delay (r = -0.293, 95% C.I. -0.459 – -0.117) within the memory domain. A significant latent variable was found in the infarct volume control analysis and it could explain 63.72% of the variance of the PLS model. The error bars indicate 95% confidence interval.

5. Control for global atrophy: Discovery dataset

To ensure our results were not driven by global atrophy, we replaced total intracranial volume as a confounding variable with global atrophy estimated as 1. grey matter volume normalised by total intracranial volume (GM/TIV) and 2. total brain volume (grey and white matter volume) as a proportion of total intracranial volume (GM+WM/TIV).

5.1 Subacute timepoint

We noted no changes to the overall pattern of significant results when controlling for grey matter/total intracranial volume (Figure S4.1) or grey and white matter/total intracranial volume (Figure S4.2). In both analysis, one significant latent variable accounted for 99.78% and 99.68% of the variance in the behavioural PLS model, respectively. The significant correlation between behavioural and brain scores remained in both control analyses. We are therefore confident the results are not driven by global atrophy as estimated in these two different ways.

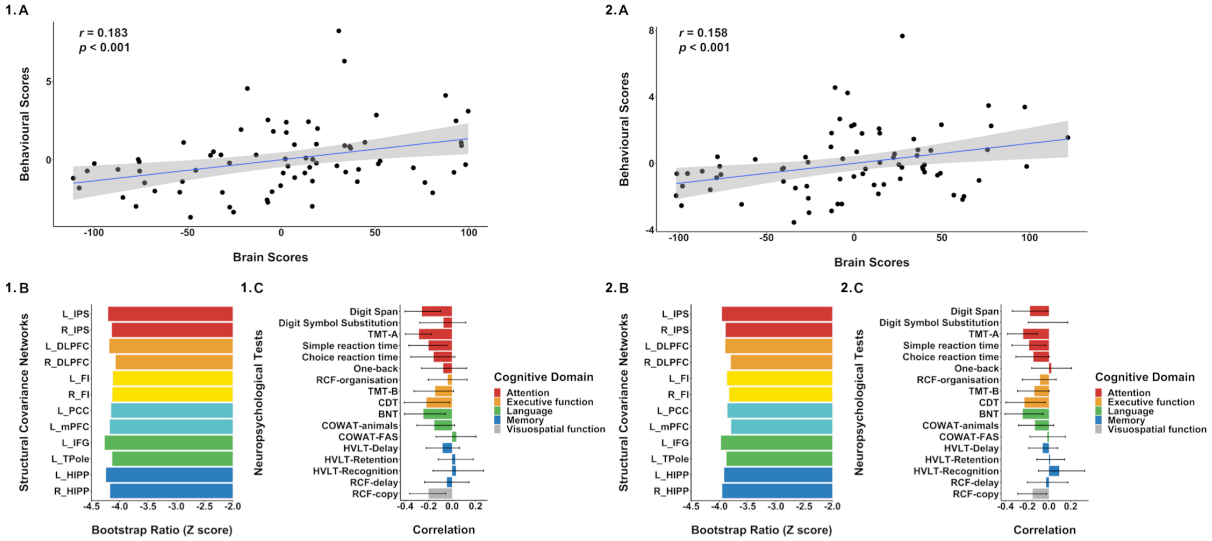


Figure S4. Lower baseline integrity of structural covariance networks was associated with greater impairment in cognitive performance in subacute stroke. These relationships were not driven by global atrophy.

Panel 1 represents results after controlling for grey matter/total intracranial volume and panel 2 represents results after controlling for grey and white matter/total intracranial volume. (1A) A positive correlation between behavioural and brain scores suggested more damaged SCNs were associated with worse attention, executive function, language, and visuospatial function performance at 3-months post-stroke. (1B) The contributions of each SCN to the covariance between SCNs and neuropsychological tests were revealed by the bootstrap ratio, which indicated a fairly equal contribution of SCNs. (1C) Extensive negative correlations between SCNs and 17 neuropsychological tests were shown, particularly in the Digit Span Task (r = -0.255, 95% C.I. -0.400 – -0.096), TMT-A (r = -0.366, 95% C.I. -0.517 – -0.213), and simple reaction time task (r = -0.199, 95% C.I. -0.367 – -0.044) within the attention domain, the CDT (r = -0.318, 95% C.I. -0.407 – -0.021) within the executive function domain, the BNT (r = -0.243, 95% C.I. -0.406 – -0.055) within the language domain, and the RCF-copy (r = -0.201, 95% C.I. -0.362 – -0.054) within the visuospatial domain. (2A) Similar to 1A, a positive correlation was also noted in the analysis controlling for grey and white matter/total intracranial volume. (2B) Similar to 1B, each SCN contributed comparatively. (2C) Similar to 1C, significant correlations between SCNs and neuropsychological tests were in the Digit Span Task (r = -0.175, 95% C.I. -0.338 – -0.008), TMT-A (r = -0.238, 95% C.I. -0.382 – -0.110), and simple reaction time task (r = -0.183, 95% C.I. -0.340 – -0.031) within the attention domain, the CDT (r = -0.226, 95% C.I. -0.398 – -0.035) within the executive function domain,

the BNT (r = -0.242, 95% C.I. -0.404 – -0.054) within the language domain, and the RCF-copy (r = -0.151, 95% C.I. -0.288 – -0.022) within the visuospatial domain.

The error bars indicate 95% confidence interval.

5.2 Chronic timepoint – controlling for longitudinal atrophy

For the longitudinal analysis we additionally controlled for scan interval (time between subacute and chronic MRI scan) and longitudinal atrophy estimated as the difference between the atrophy measure at the chronic timepoint and the subacute timepoint. As with the subacute timepoint, we did this for GM/TIV and GM+WM/TIV. Here, the first latent variable accounted for 39.43% (GM/TIV) and 54.02% (GM+WM/TIV) of the variance in the PLS model. The main results were largely unchanged including significant correlations of similar magnitude between behavioural change and change in brain scores (Figure S5).

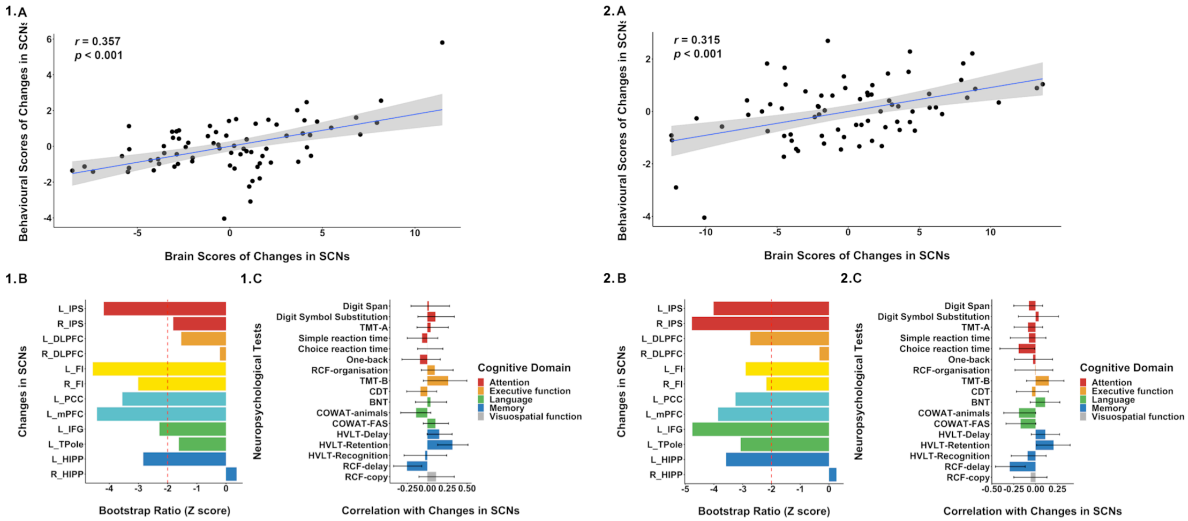


Figure S5. Faster degradation of structural covariance networks was associated with greater longitudinal decline in performance of attention, language and memory from 3-months to 1-year post-stroke (independent of brain atrophy).

Panel 1 represents results after controlling for grey matter/total intracranial volume and panel 2 represents results after controlling for grey and white matter/total intracranial volume. (1A, 2A) A positive correlation between behavioural and brain scores suggested faster SCN decline was associated with greater longitudinal decline in performance in neuropsychological tests from 3-months to 1-year post-stroke. (1B, 2B) The contribution of each SCN to the covariance between SCNs and neuropsychological tests were revealed by the bootstrap ratio. (1C) The significant correlations between each neuropsychological test and SCNs were shown in the HVLT-Retention (r = 0.317, 95% C.I. 0.130 – 0.511) and RCF-delay (r = -0.256, 95% C.I. -0.424 – -0.071) within the memory domain. (2C) The significant correlations between each neuropsychological test and SCNs were noted in the Choice reaction time task (r = -0.194, 95% C.I. -0.407– 0.009) within the attention domain and RCF-delay (r = -0.296, 95% C.I. -0.457 – -0.119) within the memory domain.

6. Validation analysis

To investigate the reproducibility of our findings, we performed validation analyses using an independent sample with 26 participants recruited from the same cohort of the original sample [10]. After identical quality control to the discovery dataset, data from four patients was excluded due to movement or lack of cognitive data. To ensure the two samples were comparable, independent *t*-tests were executed for continuous variables including age, education, infarct volume, Montreal Cognitive Assessment (MoCA), and scan interval. Chi-square tests were used for categorical variables including sex and handedness. Median tests were performed for ordinal variables including National Institute of Health Stroke Scale (NIHSS) and modified Rankin Score (mRS) at both admission and three months. No significant differences between the original and independent samples were noted (Table S3). Identical procedures of SCN analysis and validation analysis of behavioural PLS were followed. A latent variable was found and it explained 87.18% of the variance in the PLS model (Figure S6).

We additionally controlled for GM/TIV or GM+WM/TIV in the validation analysis. Again, we replicated the main pattern of results, finding one significant latent variable accounting for 98.11% and 98.20% of the variance in the PLS model respectively. In both control analyses, all SCNs contributed fairly equally to the latent variable (less than -2 bootstrap ratio across all SCNs). The correlation between behavioural and brain scores remained, again at similar magnitude, $r = 0.330$, $p < 0.001$ (GM/TIV) and $r = 0.314$, $p < 0.001$ (GM+WM/TIV). We are confident that the validation is not driven by these estimated of global atrophy either. The results of the control analyses are illustrated in Figure S7.

Table S3. Participant demographic and behavioural characteristics

Demographics	Discovery sample	Validation sample	*p*-value (2-tailed)
Age (years), mean (SD)	67.41 (12.13)	68.77 (9.65)	0.631
Sex (male/female)	51/22	16/6	0.796
Handedness (left/right)	6/67	1/21	0.563
Education (years), mean (SD)	12.89 (3.75)	12.36 (4.03)	0.572
Infarct volume (mm^3), mean (SD)	5786.62 (9316.65)	12377.65 (18062.69)	0.112
NIHSS on admission, median (25^{th}, 75^{th} percentile)	2 (1,4)	2 (1,3)	0.893
NIHSS at three months, median (25^{th}, 75^{th} percentile)	0 (0,2)	1 (0,1.25)	0.289
mRS on admission, median (25^{th}, 75^{th} percentile)	1 (1,2)	1 (1,2)	0.683
mRS at three months, median (25^{th}, 75^{th} percentile)	1 (1,2)	1 (0.75,2)	0.120
MoCA, mean (SD)	24.26 (3.43)	24.00 (2.62)	0.869
Scan interval (days), mean (SD)	276.92 (26.14)	281.95 (30.34)	0.447

Abbreviations: MoCA – Montreal Cognitive Assessment; mRS – modified Rankin Score; NIHSS – National Institute of Health Stroke Scale; SD – standard deviation.

6.1 Validation analysis, independent dataset, subacute timepoint

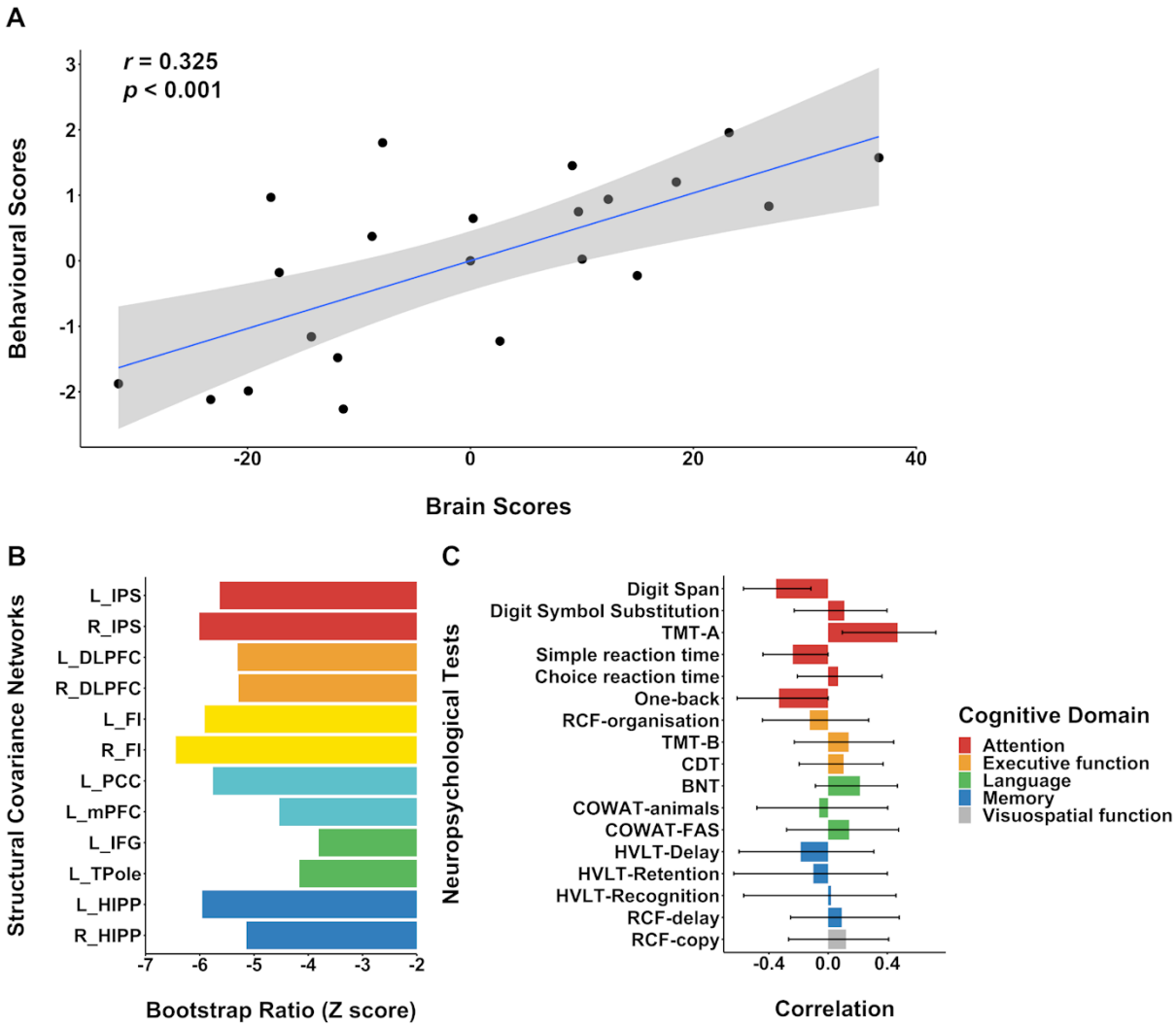


Figure S6. Lower baseline integrity of structural covariance networks was associated with greater impairment in attention in validation dataset

This validation analysis demonstrated a significant latent variable which could explain 87.18% of the variance of the PLS model. (A) A positive correlation between behavioural and brain scores suggested more damaged SCNs were associated with worse attention, executive function, language, and visuospatial function performance at 3-months post-stroke. (B) The contributions of each SCN to the covariance between SCNs and neuropsychological tests were revealed by the bootstrap ratio. (C) Significant correlations between SCNs and 17 neuropsychological TMT-A (r = 0.470, 95% C.I. 0.095 – 0.728) within the attention domain. The error bars indicate 95% confidence interval.

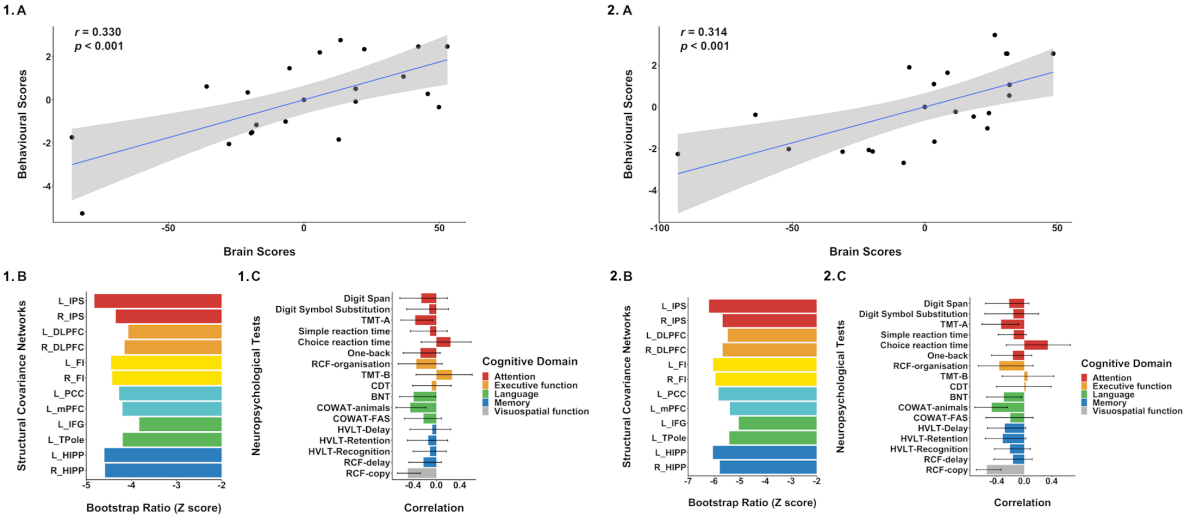


Figure S7. Lower baseline integrity of structural covariance networks was associated with greater impairment in cognitive performance in validation dataset. These relationships were not driven by global atrophy.

Panel 1 represents results after controlling for grey matter/total intracranial volume and panel 2 represent results after controlling for grey and white matter/total intracranial volume. (1A, 2A) A positive correlation between behavioural and brain scores suggested more damaged SCNs were associated with worse attention, executive function, language, and visuospatial function performance at 3-months post-stroke. (1B, 2B) The contributions of each SCN to the covariance between SCNs and neuropsychological tests were revealed by the bootstrap ratio, which indicated a fairly equal contribution of SCNs. (1C) Extensive negative correlations between SCNs and 17 neuropsychological tests were shown in the TMT-A (r = -0.369, 95% C.I. -0.604 – -0.066) within the attention domain, the COWAT-animals (r = -0.449, 95% C.I. -0.693 – -0.181) within the language domain, and the RCF-copy (r = -0.489, 95% C.I. -0.666 – -0.277) within the visuospatial domain. (2C) Similar to 1C, significant correlations between SCNs and neuropsychological tests were in the TMT-A (r = -0.337, 95% C.I. -0.616 – -0.084) within the attention domain, the BNT (r = -0.296, 95% C.I. -0.547 – -0.038) and COWAT-animals (r = -0.474, 95% C.I. -0.422 – -0.242) within the language domain, and the RCF-copy (r = -0.543, 95% C.I. -0.698 – -0.337) within the visuospatial domain.

The error bars indicate 95% confidence interval.

6.2 Validation analysis, split half analysis, longitudinal changes

Due to a lack of data of sufficient quality at the chronic timepoint (total n = 14), we were unable to do a validation on an independent dataset for the chronic timepoint. We ran a split half analysis, randomly selecting patients to groups (n = 36 and 37, respectively) and repeating this five times to generate ten samples following the procedure and corrections outlined for the longitudinal discovery dataset. Figure S8 shows the results of one iteration of the split half analysis. The first latent variable accounted for 45.40% of the variance in the PLS model in sample 1 and 60.10% of the variance in sample 2. The pattern of results were similar across validation samples, and a significant correlation between behavioural and brain score change was maintained at $r = 0.438$, $p < 0.001$ in sample 1 and $r = 0.465$ $p < 0.001$ in sample 2.

Table S4. Split half validation analysis, longitudinal changes.

Iteration	Latent variable variance explained		p-value	
	Sample 1	**Sample 2**	**Sample 1**	**Sample 2**
1	45.40%	60.10%	< 0.001	< 0.001
2	50.90%	52.98%	< 0.001	< 0.001
3	53.38%	53.96%	< 0.001	< 0.001
4	43.36%	56.55%	< 0.001	< 0.001
5	59.70%	41.15%	< 0.001	< 0.001

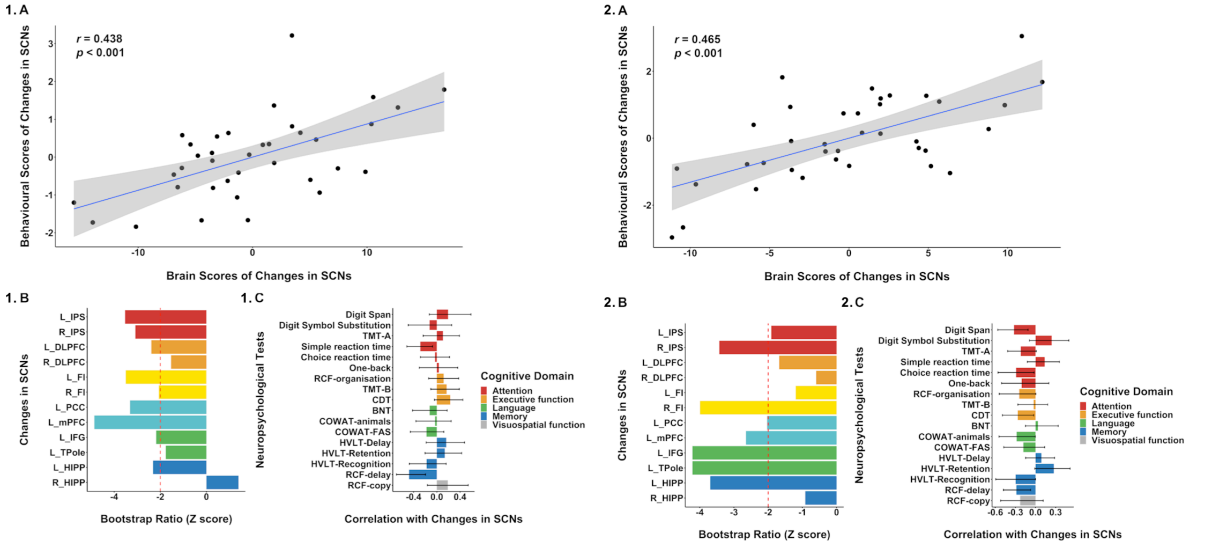


Figure S8. Faster degradation of structural covariance networks was associated with greater longitudinal cognitive decline from 3-months to 1-year post-stroke in split half validation dataset.

Panel 1 and 2 represent behavioural PLS results from two non-overlapping samples of one representative split-half run. The remaining 4 runs demonstrated similar results. (1A, 2A) A positive correlation between behavioural and brain scores suggested faster SCN decline was associated with greater longitudinal decline in performance in neuropsychological tests from 3-months to 1-year post-stroke. (1B, 2B) The contribution of each SCN to the covariance between SCNs and neuropsychological tests were revealed by the bootstrap ratio. (1C) The significant correlations between each neuropsychological test and SCNs were shown in the Simple reaction time task (r = -0.284, 95% C.I. 0.516 – 0.074) within the attention domain and RCF-delay (r = -0.479, 95% C.I. -0.694 – -0.196) within the memory domain. (2C) The significant correlations between each neuropsychological test and SCNs were noted in the Digit Span (r = -0.322, 95% C.I. -0.562– 0.112) and Choice reaction time task (r = -0.289, 95% C.I. -0.559– 0.022) within the attention domain, CDT (r = -0.265, 95% C.I. -0.493– 0.027) within the executive function domain, COWAT-animals (r = -0.282, 95% C.I. -0.544– 0.002) within the language domain, and RCF-delay (r = -0.284, 95% C.I. -0.477 – -0.073) within the memory domain.

6.3 Validation analysis, split half analysis, longitudinal changes – global atrophy controlled

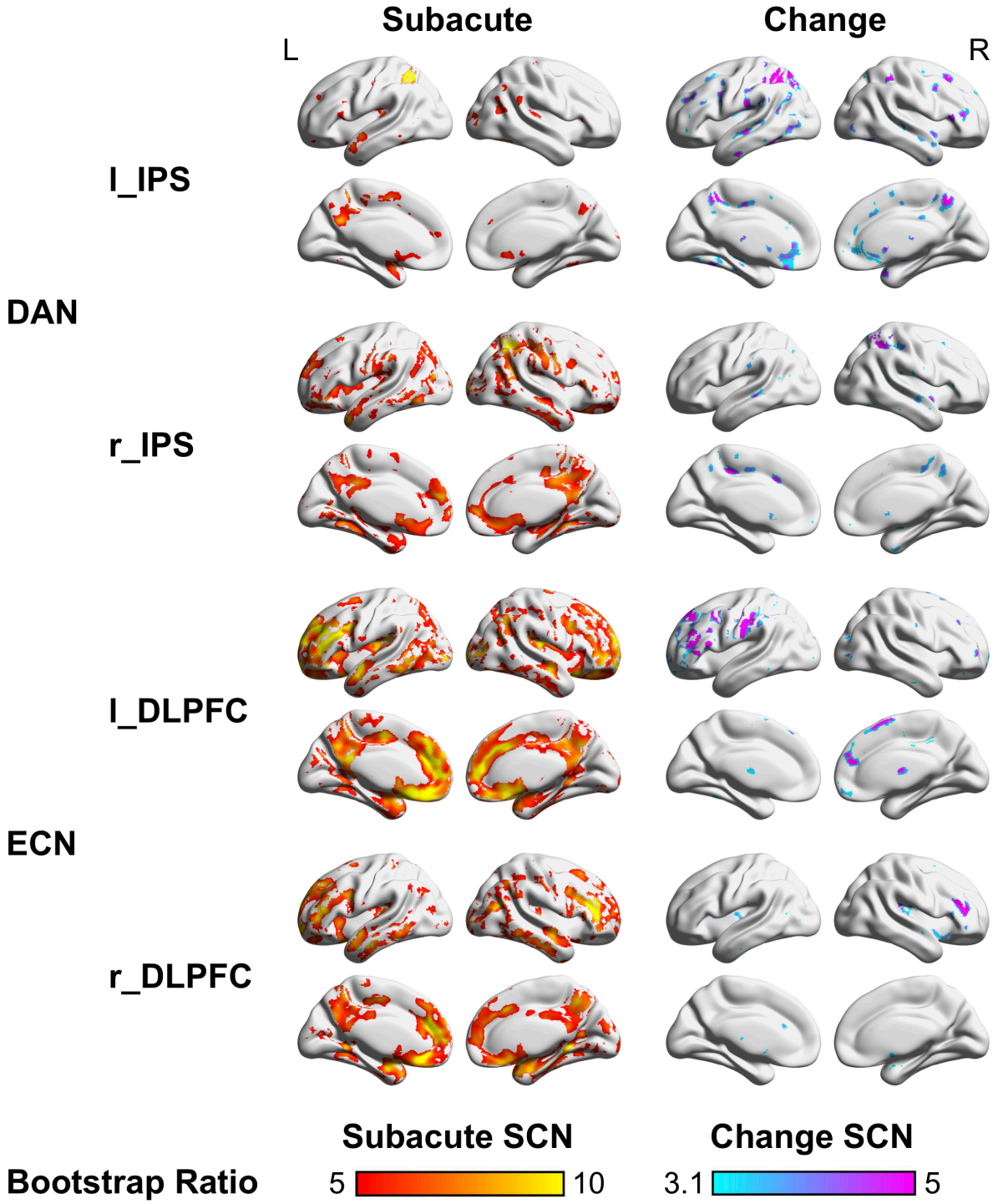
We ran the split half analysis additionally controlling for global atrophy as estimated by GM+WM/TIV. We once again found broadly similar results to the longitudinal discovery dataset and present the results of the 10 split half samples in Table S5.

Table S5. Split half validation analysis, longitudinal changes – global atrophy controlled.

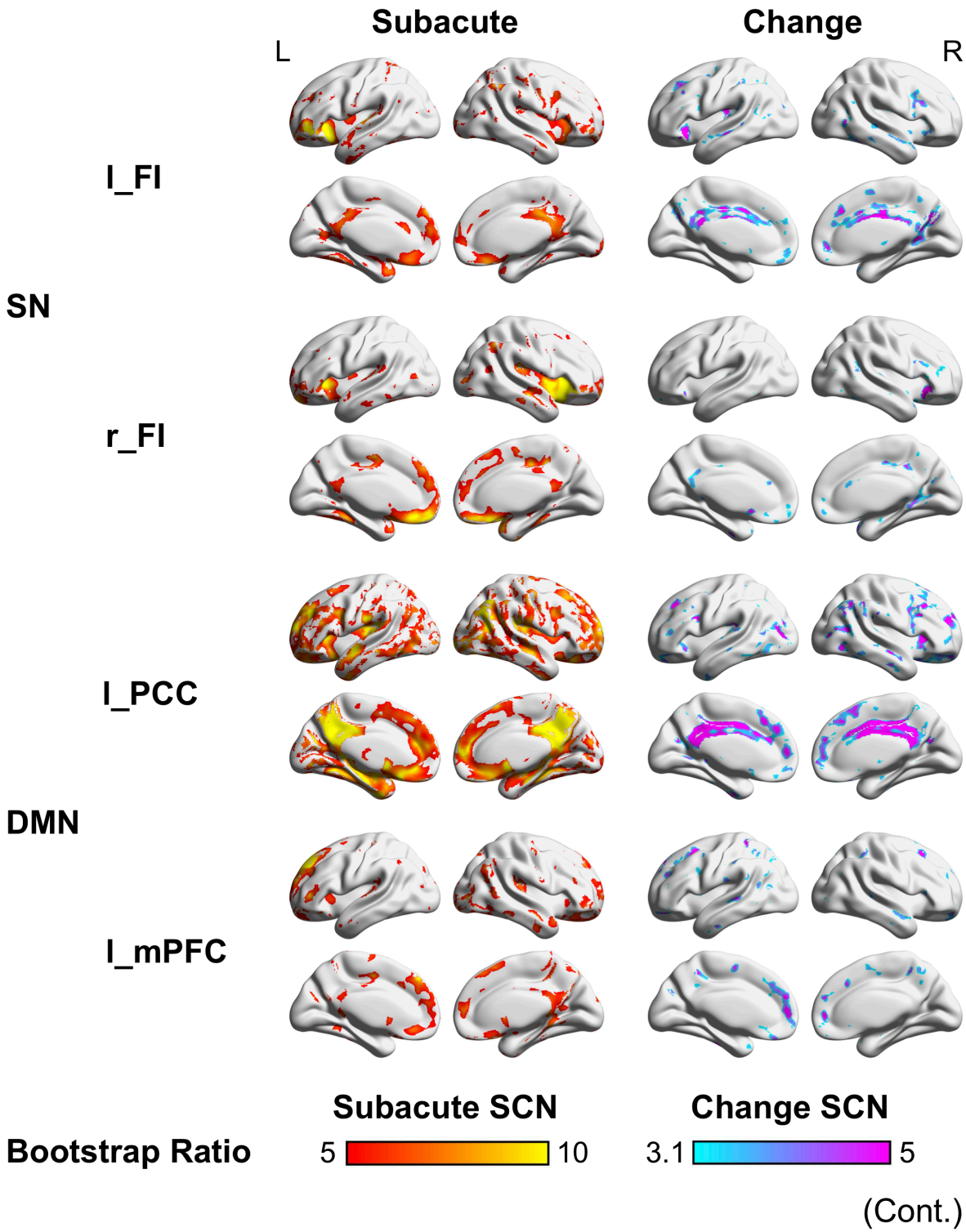
Iteration	Latent variable variance explained		p-value	
	Sample 1	**Sample 2**	**Sample 1**	**Sample 2**
1	50.66%	54.80%	< 0.001	< 0.001
2	48.33%	58.05%	< 0.001	< 0.001
3	53.81%	55.18%	< 0.001	< 0.001
4	42.18%	52.56%	< 0.001	= 0.001
5	56.07%	41.87%	< 0.001	< 0.001

7. Surface-rendered structural covariance networks in subacute and chronic patients

To enable visualisation of the entire networks at the subacute and longitudinal timepoints we surface rendered all the SCNs.



(Cont.)


Subacute
Change
L
R
l_FI
SN
r_FI
l_PCC
DMN
l_mPFC
Subacute SCN
Change SCN
Bootstrap Ratio
5
10
3.1
5

(Cont.)

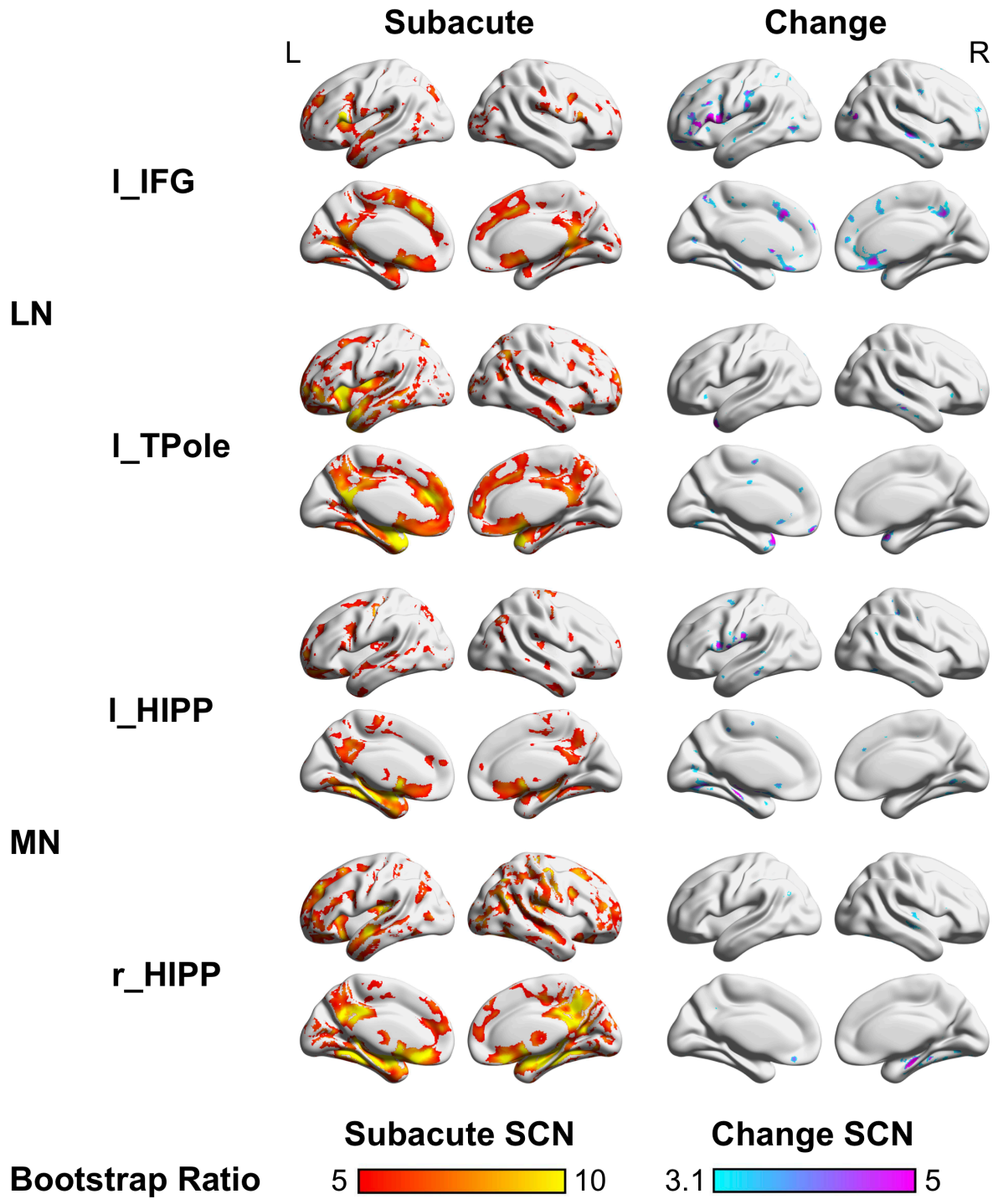


Figure S9. Surface-rendered structural covariance networks in subacute and chronic stroke patients.

The derived structural covariance networks using twelve seeds from six canonical brain networks at the subacute timepoint were depicted in the middle column in hot colour and those for change from subacute to chronic stroke were depicted in the right column in cold colour.

REFERENCES

1 Fox, M. D., Corbetta, M., Snyder, A. Z., Vincent, J. L. & Raichle, M. E. Spontaneous neuronal activity distinguishes human dorsal and ventral attention systems. *Proceedings of the National Academy of Sciences* **103**, 10046, doi:10.1073/pnas.0604187103 (2006).

2 Seeley, W. W. *et al.* Dissociable intrinsic connectivity networks for salience processing and executive control. *The Journal of Neuroscience* **27**, 2349, doi:10.1523/JNEUROSCI.5587-06.2007 (2007).

3 Sridharan, D., Levitin, D. J. & Menon, V. A critical role for the right fronto-insular cortex in switching between central-executive and default-mode networks. *Proceedings of the National Academy of Sciences* **105**, 12569, doi:10.1073/pnas.0800005105 (2008).

4 Chong, J. S. X. *et al.* Influence of cerebrovascular disease on brain networks in prodromal and clinical Alzheimer's disease. *Brain* **140**, 3012-3022, doi:10.1093/brain/awx224 (2017).

5 Greicius, M. D., Krasnow, B., Reiss, A. L. & Menon, V. Functional connectivity in the resting brain: A network analysis of the default mode hypothesis. *Proceedings of the National Academy of Sciences* **100**, 253, doi:10.1073/pnas.0135058100 (2003).

6 Vipin, A. *et al.* Cerebrovascular disease influences functional and structural network connectivity in patients with amnestic mild cognitive impairment and Alzheimer's disease. *Alzheimer's Research & Therapy* **10**, 82, doi:10.1186/s13195-018-0413-8 (2018).

7 Zielinski, B. A., Gennatas, E. D., Zhou, J. & Seeley, W. W. Network-level structural covariance in the developing brain. *Proceedings of the National Academy of Sciences* **107**, 18191, doi:10.1073/pnas.1003109107 (2010).

8 Vincent, J. L., Kahn, I., Snyder, A. Z., Raichle, M. E. & Buckner, R. L. Evidence for a frontoparietal control system revealed by intrinsic functional connectivity. *J Neurophysiol* **100**, 3328-3342, doi:10.1152/jn.90355.2008 (2008).

9 Koechlin, E., Basso, G., Pietrini, P., Panzer, S. & Grafman, J. The role of the anterior prefrontal cortex in human cognition. *Nature* **399**, 148-151, doi:10.1038/20178 (1999).

10 Brodtmann, A. *et al.* Charting cognitive and volumetric trajectories after stroke: Protocol for the cognition and neocortical volume after stroke (CANVAS) study. *International Journal of Stroke* **9**, 824-828, doi:10.1111/ijs.12301 (2014).