Appendices

Appendix e-1. *Additional data on patient E*

Since age 15, patient E presented with progressive motor disturbances, including gait and balance alterations, dysarthria, and hand tremors. Although three years later he began to exhibit cognitive deficiencies, he successfully completed 13 years of education. For a full account of the patient's clinical, neurological, and genetic profile, see [1].

Appendix e-2. *Analysis of demographic and behavioral data*

Demographic and behavioral data of the patient and the controls was compared with Crawford's modified two-tailed *t*-test [2-6]. This test is robust for non-normal distributions, presents low rates of type-I error, and has proved successful in previous single-case studies [7-9], even when the control sample comprises fewer than five subjects [10].

Appendix e-3. *Lexical decision task*

The lexical decision task involved 80 Spanish words (20 action verbs, denoting bodily movements; 20 abstract verbs, denoting cognitive or affective processes; 20 manipulable nouns, denoting graspable objects; and 20 abstract nouns, denoting non-physical notions) and 80 legal pseudowords (created by replacing one or two letters from the real words). All items had between four and seven letters; verbs and nouns were in infinitive and singular form, respectively. As shown by one-way ANOVAs, the four word lists were similar in log frequency [$F(3, 76) = 2.56$, $p = .06$], orthographic length [$F(3, 76) = 2.11$, $p = .10$], mean type bigram frequency [$F(3, 76) = 2$, $p = .12$], and number of orthographic neighbors [$F(3, 76) = 1.5$, $p = .22$] –data extracted from B-Pal [11].

Participants sat facing a laptop and were instructed to press a pre-assigned left or right key on the keyboard to indicate whether each stimulus was a real or an invented word, respectively. The stimuli were pseudorandomly distributed to avoid fatigue-related or strategic effects. Responses were made with the index and middle fingers of the dominant hand. Each trial began with an ocular fixation cross (500 ms), followed by a

blank screen (100 ms) and then by the stimulus (which remained on the screen until a response was made). The task was designed and performed on DMDX software

Note that the lexical decision paradigm is widely used to explore embodied mechanisms –for a review, see [12]. Moreover, since it involves shallow processing (as it allows for task completion without conscious access to meaning), the emergence of semantically-driven effects speaks to their robustness and automaticity.

Appendix e-4.1. ***Structural and functional brain measures***

Structural T1 scans were acquired with the following parameters: matrix size = 256 × 240 × 120, 1 mm isotropic, TR = 7489 ms, TE = 3420 ms, flip angle = 8°. For functional imaging analysis, 33 axial slices (5-mm thick) were acquired parallel to the plane connecting the anterior and posterior commissures and covering the whole brain (TR = 2777 ms, TE = 50 ms, flip angle = 90). The protocol lasted ten minutes and 209 functional brain images were obtained for each subject. During resting-state recordings, all participants were asked not to think about anything in particular during ten minutes and to avoid moving or falling sleep.

Appendix e-4.2. ***Voxel-based morphometry***

Data were preprocessed on the DARTEL Toolbox following validated procedures [13 14] and using the Statistical Parametric Mapping software (SPM12) (http://www.fil.ion.ucl.ac.uk/spm/software/spm12/). Prior to modulation, images were segmented in grey matter, white matter, and cerebrospinal fluid volumes. Next, 12 mm full-width half-maximum kernel images were smoothed as proposed in other reports [14] and normalized to MNI space. Subsequently, the patient's and the controls' smoothed, normalized, and modulated gray matter images were compared via *t*-tests to account for the former's global atrophy pattern, corrected by total intracranial volume. To this end, we used SPM12 software. Statistical significance was set at $p < .001$ uncorrected, extent threshold = 50 voxels [15].

Appendix e-4.3. ***FMRI preprocessing steps***

Images were slice-time corrected, realigned to the middle slice of the volume, normalized, and smoothed using the Data Processing Assistant for Resting-State fMRI

(DPARSF) software [16]. They were also band-pass filtered (0.01-0.08 Hz). Images were smoothed with an eight-mm full-width half-maximum kernel and then normalized to the default EPI template from SPM12. Preprocessing parameters were chosen in accordance with previous studies [17 18]. No participant had movements greater than 3 mm and/or rotations higher than 3º.

Appendix e-4.4. ***Functional connectivity analysis: Seed analysis***

We built one 5-mm spherical ROI from the largest VBM cluster peak (x = -17, y = -60, z = -20) located in the cerebellum. For each participant, we extracted the BOLD signal time-course from the voxels within each seed region. To obtain a functional connectivity map, we then correlated these data to every voxel in the brain (excluding the cerebellum) using Pearson's correlation coefficient. We calculated this with the DPARSF toolbox [16]. Statistical analyses were performed on SPM12 software.

Appendix e-4.5. ***Gene-atrophy overlap***

To assess a potential link between the patient's atrophy pattern and the *STUB1* gene expression, we calculated their overlap using data from the Allen Human Brain data base [19]. We established the localization of the gene (in MNI coordinates) from an anonymous healthy donor demographically matched with our patient [specimen name: H0351.1012 (31 years-old, male, white); probe name: A_23_P37870]. As in previous reports [20], five-mm radius spherical ROIs were constructed with each coordinate to create the gene expression map. We reported regions in which the overlap covered at least 50 voxels.

Table e-1. ***Behavioral results.***

Lexical decision accuracy					
Category	Patient	Control group*	*p*-value	*t*-value	Effect size (*Z*-score)
Action verbs	2	16.67 (4.41)	.03	-3.08	-3.33
Abstract verbs	8	16.5 (5.04)	.18	-1.56	-1.69
Manual nouns	9	16.33 (4.96)	.23	-1.37	-1.48

Abstract nouns	11	16.67 (4.17)	.26	-1.26	-1.36

* Data expressed as mean (*SD*).

Table e-2. ***Brain sites showing significant atrophy in patient E.***

Brain region	Cluster *k*	Peak *t*	Peak *z*	Peak *p* (uncl)	x	y	z
Cerebellum L*	25553	232.15	6.66	< .001	-16.5	-60	-19.5
		202.15	6.55	< .001	-22.5	-64.5	-16.5
Cerebellum Vermis Crus 2 L		118.33	6.14	< .001	-25.5	-79.5	-39
Cerebellum Vermis Crus 1 L	79	29.90	4.94	< .001	-42	-81	-6
Median cingulate and paracingulate gyri L	67	11.05	3.88	< .001	-6	-30	45
Insula L	905	46.17	5.35	< .001	-34.5	15	3
		34.81	5.08	< .001	-40.5	4.5	1.5
Insula L	823	19.59	4.51	< .001	-28.5	7.5	-18
Insula R	170	24.46	4.74	< .001	31.5	13.5	-13.5
Insula R	351	20.38	4.55	< .001	36	18	9
Insula R	102	8.06	3.49	< .001	43.5	-10.5	1.5
Putamen L	905	28.36	4.89	< .001	-28.5	10.5	6
Putamen R	102	10.41	3.81	< .001	33	-12	10.5
Superior medial orbital frontal gyrus R	118	9.37	3.68	< .001	24	64.5	-12
		9.19	3.66	< .001	18	58.5	-18
Gyrus rectus L	87	11.47	3.92	< .001	-3	48	-27
Inferior frontal gyrus, triangular part L	78	14.81	4.21	< .001	-42	39	12
		14.09	4.16	< .001	-42	33	6
Inferior frontal gyrus, triangular part L	102	13.56	4.11	< .001	-37.5	33	25.5
Inferior frontal gyrus, opercular part R	351	12.36	4.01	< .001	46.5	15	13.5
Inferior frontal gyrus, orbital part R	73	18.45	4.45	< .001	31.5	33	-7.5
Superior dorsolateral frontal gyrus L	73	15.11	4.23	< .001	-15	36	51
Superior dorsolateral frontal gyrus L	208	12.55	4.03	< .001	-12	57	22.5
Superior dorsolateral frontal gyrus R	61	8.09	3.50	< .001	25.5	43.5	43.5
		7.94	3.48	< .001	18	34.5	43.5
Superior dorsolateral frontal gyrus L	208	12.55	4.03	< .001	-12	57	22.5
Superior medial frontal gyrus L		8.57	3.57	< .001	-9	60	31.5
Superior medial frontal gyrus R	394	28.07	4.88	< .001	3	45	40.5
		25.67	4.79	< .001	6	39	46.5
		21.29	4.60	< .001	6	49.5	46.5
Middle frontal gyrus L	53	45.72	5.34	< .001	-28.5	28.5	52.5
Middle frontal gyrus L	64	9.91	3.75	< .001	-27	-9	51

Middle frontal gyrus L	76	8.72	3.59	< .001	-24	12	55.5
Middle frontal gyrus L	65	7.43	3.39	< .001	-43.5	10.5	37.5
Middle frontal gyrus R	346	16.92	4.36	< .001	34.5	-3	55.5
		12.72	4.04	< .001	37.5	-1.5	63
Middle frontal gyrus R	60	11.24	3.90	< .001	28.5	16.5	60
		7.32	3.37	< .001	28.5	21	52.5
Precentral gyrus L	65	8.70	3.59	< .001	-42	4.5	31.5
		8.21	3.52	< .001	-34.5	6	37.5
Precentral gyrus L	73	8.26	3.52	< .001	-42	7.5	48
Precentral gyrus R	346	21.84	4.63	< .001	46.5	1.5	51
Precentral gyrus R	76	7.43	3.39	< .001	-30	10.5	61.5
Lingual gyrus R	53	18.90	4.47	< .001	19.5	-37.5	-1.5
Middle temporal gyrus R	143	19.36	4.50	< .001	48	-63	1.5
Inferior temporal gyrus R		11.69	3.94	< .001	48	-60	-6
Middle temporal gyrus R		8.97	3.63	< .001	42	-67.5	7.5
Middle temporal gyrus R	91	11.67	3.94	< .001	-61.5	-61.5	-1.5
Superior temporal gyrus R	349	22.44	4.65	< .001	55.5	-36	15
		13.82	4.13	< .001	66	-30	16.5
Middle temporal gyrus R		11.06	3.88	< .001	52.5	-42	6
Superior temporal gyrus L	823	30.81	4.97	< .001	-37.5	22.5	-25.5
		24.22	4.73	< .001	-31.5	4.5	-33
Superior temporal gyrus L	72	17.90	4.42	< .001	-43.5	-12	-12
Superior temporal gyrus R	132	10.69	3.84	< .001	52.5	-25.5	16.5
		9.09	3.64	< .001	49.5	-28.5	7.5
		8.84	3.61	< .001	48	-16.5	-3
Inferior parietal L	171	16.83	4.35	< .001	31.5	-43.5	51
Inferior parietal R		14.44	4.18	< .001	37.5	-39	46.5
Postcentral gyrus R	76	10.80	3.85	< .001	-30	-31.5	52.5
Postcentral gyrus L		8.34	3.54	< .001	-28.5	-39	61.5
Precuneus L	63	13.09	4.07	< .001	0	-57	49.5
Precuneus R		7.93	3.47	< .001	1.5	-64.5	55.5
Cuneus cortex L	68	28.28	4.88	< .001	-1.5	-79.5	34.5
		8.41	3.55	< .001	-10.5	-85.5	27
Superior occipital gyrus R	198	23.85	4.72	< .001	19.5	-91.5	25.5
Cuneus cortex R		21.65	4.62	< .001	13.5	-90	19.5
Middle occipital gyrus R		13.46	4.10	< .001	25.5	-90	16.5
Middle occipital gyrus L	95	10.81	3.85	< .001	-46.5	-70.5	10.5
Inferior occipital gyrus R	75	47.20	5.37	< .001	34.5	-85.5	-6
Inferior occipital gyrus R	59	12.38	4.01	< .001	39	-70.5	-12

Reported areas are based on Automated Anatomical Labeling (AAL) [21]. R = right; L = left. Asterisk (*) indicates the peak of atrophy used to establish the seed used in seed analysis.

Table e-3. ***Seed analysis results*****.**

Brain region	Cluster *k*	Peak *t*	Peak *z*	Peak *p* (uncl)	x	y	z
Inferior frontal gyrus, opercular part R	211	17.28	4.42	< .001	48	16	28
Inferior frontal gyrus, orbital part R	76	10.94	3.90	< .001	36	30	-8
		8.84	3.64	< .001	30	30	-16
		7.14	3.37	< .001	22	24	-14
Gyrus rectus L	94	12.84	4.09	< .001	-6	48	-20
Gyrus rectus R	103	11.01	3.91	< .001	4	36	-26
		8.93	3.65	< .001	-2	32	-30
Superior frontal gyrus, dorsolateral R	59	14.08	4.19	< .001	22	8	48
Middle frontal gyrus, orbital part	56	7.33	3.40	< .001	28	48	-14
Middle frontal gyrus, orbital part R		6.31	3.20	< .001	22	54	-18
Middle frontal gyrus L	62	12.74	4.08	< .001	-24	-8	48
Supramarginal gyrus L	72	18.89	4.52	< .001	-60	-44	26
Precuneus R	24985	65.34	5.71	< .001	12	-80	26
Middle occipital gyrus L		61.81	5.66	< .001	-34	-82	10
Fusiform gyrus R		58.55	5.61	< .001	38	-58	-20
Inferior parietal, excluding supramarginal and angular gyri L	181	9.65	3.75	< .001	-40	-46	40

Reported areas are based on Automated Anatomical Labeling (AAL) [21].

Table e-4. ***Overlap between patient's E atrophy and sites of expression of the*** **STUB1** ***gene*****.**

Regions	Number of voxels
Cerebellum (Hemispheric lobule IV/V) L	124
Cerebellum (Hemispheric lobule IV/V) R	99
Cerebellum (Hemispheric lobule VI) L	103
Cerebellum (Hemispheric lobule VI) R	108
Cerebellum (Hemispheric lobule VIII) R	115
Cerebellum Crus 1 L	85

Cerebellum Crus 2 L	118
Fusiform gyrus L	77
Fusiform gyrus R	104
Superior temporal gyrus R	66

Reported areas are based on Automated Anatomical Labeling (AAL) [21]. R = right; L = left.

References

1 Cordoba M., et al., Ataxia plus myoclonus in a 23-year-old patient due to STUB1 mutations. *Neurology* 2014;83(3):287–8.

2 Crawford JR & Garthwaite, PH, Single-case research in neuropsychology: a comparison of five forms of t-test for comparing a case to controls. *Cortex* 2012;48(8):1009–16.

3 Crawford JR & Garthwaite, PH, Investigation of the single case in neuropsychology: confidence limits on the abnormality of test scores and test score differences. *Neuropsychologia* 2002;40(8):1196–1208.

4 Crawford JR, Garthwaite, PH, & Howell, DC, On comparing a single case with a control sample: an alternative perspective. *Neuropsychologia* 2009;47(13):2690–5.

5 Crawford JR & Howell, DC, Regression equations in clinical neuropsychology: an evaluation of statistical methods for comparing predicted and obtained scores. *J Clin Exp Neuropsychol* 1998;20(5):755–62.

6 Crawford JR, Garthwaite, PH, & Ryan, K, Comparing a single case to a control sample: testing for neuropsychological deficits and dissociations in the presence of covariates. *Cortex* 2011;47(10):1166–78.

7 Couto B, et al., Insular networks for emotional processing and social cognition: comparison of two case reports with either cortical or subcortical involvement. *Cortex* 2013;49(5):1420–34.

8 Straube T, et al., No impairment of recognition and experience of disgust in a patient with a right-hemispheric lesion of the insula and basal ganglia. *Neuropsychologia* 2010;48(6):1735–41.

9 Couto B, et al., The man who feels two hearts: the different pathways of interoception. *Soc Cogn Affect Neurosci* 2014;9(9):1253–60.

10 Crawford JR, Comparing an individual's test score against norms derived from small samples. *Clin Neuropsych* 1998;12(4):482–6.

11 Davis CJ & Perea, M, BuscaPalabras: a program for deriving orthographic and phonological neighborhood statistics and other psycholinguistic indices in Spanish. *Behav Res Methods* 2005;37(4):665–71.

12 García AM & Ibáñez A, A touch with words: Dynamic synergies between manual actions and language. *Neuro Bio Rev* 2016;68:59–95.

13 Ashburner J & Friston, KJ, Voxel-based morphometry--the methods. *Neuroimage* 2000;11(6 Pt 1):805–21.

14 Couto B, et al., Structural neuroimaging of social cognition in progressive non-fluent aphasia and behavioral variant of frontotemporal dementia. *Front Hum Neurosci* 2013;7:467.

15 Irish M, et al., Common and unique gray matter correlates of episodic memory dysfunction in frontotemporal dementia and Alzheimer's disease. *Hum Brain Mapp* 2014;35(4):1422–35.

16 Chao-Gan Y & Yu-Feng, Z, DPARSF: A MATLAB Toolbox for "Pipeline" Data Analysis of Resting-State fMRI. *Front Syst Neurosci* 2010;4:13.

17 Sedeno L, et al., Brain network organization and social executive performance in frontotemporal dementia. *J Int Neuropsychol Soc* 2016;22(2):250–62.

18 Sedeno L, et al., How do you feel when you can't feel your body? Interoception, functional connectivity and emotional processing in depersonalization-derealization disorder. *PLoS One* 2014;9(6):e98769.

19 Hawrylycz MJ, et al., An anatomically comprehensive atlas of the adult human brain transcriptome. *Nature* 2012;489(7416): 391–9.

20 Baez S, et al., Tracking the cognitive, social, and neuroanatomical profile in early neurodegeneration: Type III Cockayne syndrome. *Front Aging Neurosci* 2013;5:80.

21 Tzourio-Mazoyer N, et al., Automated anatomical labeling of activations in SPM using a macroscopic anatomical parcellation of the MNI MRI single-subject brain. *Neuroimage* 2002;15(1):273–89.