


## ORIGINAL RESEARCH

# Effects of seizure burden on structural global brain networks in patients with unilateral hippocampal sclerosis

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## Abstract

**Background and purpose:** Temporal lobe epilepsy secondary to hippocampal sclerosis is related to epileptogenic networks rather than a focal epileptogenic source. Graph-theoretical gray and white matter networks may help to identify alterations within these epileptogenic networks.

**Methods:** Twenty-seven patients with hippocampal sclerosis and 14 controls underwent magnetic resonance imaging, including 3D-T1, fluid-attenuated inversion recovery, and diffusion tensor imaging. Subject-specific structural gray and white matter network properties (normalized path length, clustering, and small-worldness) were reconstructed. Group differences and differences between those with higher and lower seizure burden (<4 vs.  $\geq 4$  average monthly seizures in the last year) in network parameters were evaluated. Additionally, correlations between network properties and disease-related variables were calculated.

**Results:** All patients with hippocampal sclerosis as one group did not have altered gray or white matter network properties (all  $p > .05$ ). Patients with lower seizure burden had significantly lower gray matter small-worldness and normalized clustering compared to controls and those with higher seizure burden (all  $p < .04$ ). A higher number of monthly seizures was significantly associated with increased gray and white matter small-worldness, indicating a more rigid network.

**Conclusion:** Overall, there were no differences in network properties in this group of patients with hippocampal sclerosis. However, patients with lower seizure burden had significantly lower gray matter network indices, indicating a more random organization. The correlation between higher monthly seizures and a more rigid network is driven by those with higher seizure burden, who presented a more rigid network compared to those with a lower seizure burden.

## KEYWORDS

graph theory, gray matter network, hippocampal sclerosis, magnetic resonance imaging, white matter network

## 1 | INTRODUCTION

Hippocampal sclerosis is the most common pathological abnormality associated with mesial temporal lobe epilepsy (Dutra et al., 2015). Although the pathophysiology of hippocampal sclerosis remains unclear, there is evidence that a large network of areas connected to the hippocampus is involved in the generation and maintenance of seizures (Bonilha et al., 2004).

The brain is organized as a complex and flexible network of interconnected neuronal structures, which has topological properties that can be represented as a graph. This topological organization of the brain, as networks, is important for cognitive functions, behavior, and daily functioning (Bullmore & Sporns, 2012; Stam, 2010). Graph theory is a mathematical framework that allows quantitative modeling and analysis of networks, which can be used to quantify structural gray and white matter networks. This technique defines networks as sets of nodes and edges. Nodes can represent brain regions or clusters of voxels, while edges denote connections between these nodes. The strength of an edge interconnecting two nodes can be derived from various structural and functional neuroimaging (Bernhardt et al., 2015; Stam, 2010). For example, structural networks may be derived from covariance patterns of morphological magnetic resonance imaging (MRI) markers, such as cortical thickness or regional gray matter volume, assessed through T1-weighted imaging. White matter networks can be derived from diffusion tensor imaging (DTI) parameters such as voxel-wise fractional anisotropy (Bernhardt et al., 2015).

The most commonly employed graph-theoretical parameters are clustering coefficient and path length (Bernhardt et al., 2015). Clustering coefficient measures the probability that the neighbors of a node will also be connected to each other, indicating the extent to which its neighbors are mutually connected or clustered together. Path length is the average number of edges (connections) that have to be used to get from one node to another; it quantifies the average number of shortest paths between any two nodes in the network (Bernhardt et al., 2015; Stam, 2010). Interestingly, the human brain has a global network topology that is considered small-world. Small-worldness is characterized by high clustering and short path lengths, combining a high efficiency (short path length) and high clustering (Bullmore & Sporns, 2012). A network with higher clustering coefficient and longer path length would be more rigidly ordered, as it is more difficult to get to other nodes outside the cluster. Consequently, the network loses part of its small-worldness and becomes less efficient. On the contrary, a network with a lower clustering coefficient and a shorter path length would become more random, as clusters would be less organized and more difficult to reach. This network would also lose its small-worldness and become less efficient (Stam, 2010). Thus, alterations in this trade-off between clustering and path length will cause networks to become less efficient and lose its small-worldness.

Graph-theoretical measures have been found to capture this complex network structure of both gray and white matter in health and disease (Bullmore & Sporns, 2012; Stam, 2010). In temporal lobe epilepsy, relatively few studies have assessed graph-theoretical properties of

brain networks derived from structural MRI (Bernhardt et al., 2011; Bonilha et al., 2012; Haneef & Chiang, 2014; Liu et al., 2014). In general, these studies have shown that temporal lobe epilepsy presents with alterations in local and global networks, generally characterized by a lower clustering coefficient and increased path length. However, such studies have assessed networks at a group level, which means these studies did not capture intra- and interindividual differences in network properties, and, consequently, missed valuable information. Furthermore, in the absence of subject-specific networks, correlations between network parameters and disease parameters cannot be studied.

Therefore, we aimed to assess both gray and white matter global structural networks in epileptic patients with confirmed hippocampal sclerosis, compared to healthy controls, on a subject-specific level. Thus, including important intra- and interindividual information and allowing for the calculation of correlations between network properties and clinical variables. Assessment of structural networks through graph theory may help identify subtle alterations within the hippocampal epileptic structural network. We hypothesized that patients with hippocampal sclerosis would present reduced small-world topology, correlated with the severity of the disease.

## 2 | MATERIALS AND METHODS

### 2.1 | Subjects

This study was approved by the institutional review board of the University Hospital, and all subjects and/or their legal guardians gave written informed consent. Between February 2014 and September 2017, 27 participants with hippocampal sclerosis were included from the epilepsy clinics of the University Hospital. All patients met the criteria for hippocampal sclerosis, including clinical symptoms, age of onset, and conventional MRI and electroencephalography (EEG) characteristics. All patients had unilateral crisis onset documented by video-EEG, which corresponded to the side of hippocampal sclerosis seen on MRI. No patient had a history of status epilepticus and all patients and/or their legal guardians denied having a history of generalized tonic-clonic seizures within 3 years prior to MRI examination. Exclusion criteria included a diagnosis of bilateral hippocampal sclerosis, video-EEG demonstrating bilateral seizure activity, discordant EEG-MRI findings with respect to laterality, dual pathology (assessed on MRI) or other neurological disorders.

A healthy control group without a clinical indication for MRI examination, was also included. Exclusion criteria for controls included any history of epilepsy, neurological disorders, or other major medical illnesses. All controls had normal MRI scans or only nonspecific hyperintensities in deep frontal or parietal white matter on fluid-attenuated inversion recovery images. All participants (both epilepsy

patients and controls) were right-handed and underwent the same MRI protocol.

## 2.2 | MRI acquisition

A 1.5-T scanner (Magnetom Avanto, Siemens, Erlangen, Germany) was used. For this study, a T1-3D Magnetization Prepared Rapid Gradient Echo (MPRAGE) (TR, 2730 ms; TE, 3.26 ms; inversion time, 1000 ms; FOV, 256 mm; matrix,  $192 \times 256$ ; flip angle,  $7^\circ$ ; voxel size,  $1.3 \times 1.0 \times 1.3$  mm), and an axial diffusion tensor single-shot echo-planar imaging (EPI) with bipolar diffusion gradients applied along 30 noncolinear directions ( $b_0 = 0$  and  $b_1 = 900$  s/mm<sup>2</sup>); TR, 10,100 ms; TE, 94 ms; FOV, 256 mm; matrix,  $122 \times 120$ ; 65 slices with 2.1-mm thickness without gap) were used. All MRI datasets were reviewed by an experienced neuroradiologist and were of good quality and no participant had unexpected clinical findings, except for isolated punctiform white matter hyperintensities.

## 2.3 | Gray matter network reconstruction

Using Statistical Parametric Mapping version 12 (SPM12) the origin of the scans was set to the anterior commissure, after which they were segmented into cerebrospinal fluid, gray and white matter. Gray matter segmentations were resliced into  $2 \text{ mm}^3$  isotropic voxels. The labels of the automated anatomical labeling (AAL) (Tzourio-Mazoyer et al., 2002) atlas were then nonlinearly warped to native T1-MPRAGE space.

Next, using the native space gray matter segmentations, networks were extracted using a software developed in Matlab v7.12.0.635 ([https://github.com/bettytijms/Single\\_Subject\\_Grey\\_Matter\\_Networks](https://github.com/bettytijms/Single_Subject_Grey_Matter_Networks); for technical details please see Tijms et al., 2012). In short, the nodes of the network were defined as small regions of interest of  $3 \times 3 \times 3$  voxels, which were connected by edges when they showed structural similarity as quantified by Pearson's correlations. Given the large number of possible correlations that comes with this approach, it is necessary to apply a threshold to each individual's correlation matrix so that the number of spurious (i.e., false-positive) correlations is kept at the statistical level of 5% (Noble, 2009). Briefly, an empirical null model was derived for each participant, which tested structural similarity between regions from which all spatial structure was removed, while keeping intact first order moments. Then, through permutation, the subject-specific threshold for significance was set such that the area under the curve of the empirical null model was 5%, which corresponds to the expected number of spurious (i.e., potential false-positive) correlations that may occur for that threshold. Therefore, each network was binarized using this threshold correcting for multiple testing. Although continuous weighted networks would contain the most information, we binarized the networks to compute basic network topology measures, in order to compare these findings from previous studies.

## 2.4 | White matter network reconstruction

Preprocessing in ExploreDTI version 4.8.6 included signal drift correction using  $b = 900$  s/mm<sup>2</sup> (Vos et al., 2017), Gibbs ringing artifact correction (Perrone et al., 2015), closing of Venetian Blinds, if present. Both DTI and T1 images were then cropped to remove nonbrain data, after which DTI images were corrected for subject motion and eddy currents, and the  $b$ -matrix was rotated (Leemans & Jones, 2009). Lastly, to correct for EPI deformations, the corrected DTI images were nonlinearly warped to the participant's T1-scan (Kennis et al., 2016). Tractography was performed using constrained spherical deconvolution (Jeurissen et al., 2011).

Also in ExploreDTI, each participant's fiber tract reconstruction was segmented into the AAL atlas, which was first nonlinearly warped to subject-space using FMRIB's Software Library version 6.0. Connectivity matrices based on fractional anisotropy of the connections were used in this study. These have shown high reproducibility and low within-subject variability (Welton et al., 2015). To exclude partial volume effect of gray matter and cerebrospinal fluid, the fractional anisotropy (FA) threshold was set at 0.2 because gray matter tends to have an FA value lower than 0.2.

## 2.5 | Network measures

All graph theory properties were calculated using scripts from the Brain Connectivity Toolbox ([brain-connectivity-toolbox.net](http://brain-connectivity-toolbox.net)), which were modified for large networks. The networks are template free, so the atlas is used only to compare the measures. More technical details of all graph properties, their references and their interpretation can be found in Rubinov and Sporns (2010).

For each node of both gray and white matter networks, we calculated degree (number of connections of a node), connectivity density, path length, and clustering coefficient. Then, normalized whole-brain path length ( $\lambda$ -lambda) and clustering coefficient ( $\gamma$ -gamma) were calculated by averaging the raw path length and clustering coefficient across the nodes for each graph and then dividing these whole-brain properties by those averaged from 25 randomized reference graphs with an identical size and degree distribution (Maslov & Sneppen, 2002). By dividing  $\gamma/\lambda$ , small-worldness was calculated. A network has small-world properties when the clustering coefficient is higher than one ( $\gamma > 1$ ) and the path length is similar to that of the averaged random reference graph of identical size and degree ( $\lambda \approx 1$ ).

## 2.6 | Statistical analysis

Subject characteristics were analyzed using Student's  $t$ -test, for variables with a normal distribution (age, years since diagnosis, age at epilepsy onset, and affected hippocampal volume); and chi-square test, for categorical variables (sex distribution). Normality was tested using the Kolmogorov-Smirnov test. No variables had a nonparametric

**TABLE 1** Sociodemographic and clinical data of participants with hippocampal sclerosis and healthy controls

	Hippocampal sclerosis	Controls	p-Value
Age (years)	36.0 (SD: 11.09; range: 13–55; median: 38; IQR: 17.5)	35.07 (SD: 11.95; range: 18–55; median: 38; IQR: 20.75)	.81
Sex	19 men/8 women	10 men/4 women	.94
Affected hippocampus normalized volume	4.82 (SD: 0.88; range: 2.54–6.38; median: 4.92; IQR: 1.08)		
Years since diagnosis	20.96 (SD: 11.23; range: 3–43; median: 20; IQR: 16)		
Age at epilepsy onset (years)	15.07 (SD: 13.73; range: 1–45; median: 11; IQR: 17.5)		
Average monthly number of seizures in the last year	3.37 (SD: 4.46; range: 0–20; median: 2; IQR: 4.5)		

Abbreviations: IQR, interquartile range; SD, standard deviation.

distribution. Considering all patients as one group, gray and white matter normalized path length and clustering and small-worldness were compared to control subjects, using analysis of variance (ANOVA). Spearman's correlations were calculated between global network properties and years since diagnosis, age at epilepsy onset, average monthly number of seizures in the last year, and affected hippocampus volume, in the patients group.

Additionally, patients were classified according to lower or higher seizure burden. This was determined on the basis of the average monthly seizures in the past year. Those with less than four average monthly seizures were determined to have lower seizure burden and those with four or more average monthly seizures in the past year were characterized as having higher seizure burden. Graph-theoretical measures between these two groups and controls were also compared using an ANOVA. All analyses were performed with IBM-SPSS version 20 (Chicago, IL, USA). A  $p < .05$  was considered statistically significant.

### 3 | RESULTS

#### 3.1 | Subject and network characteristics

The group of patients with hippocampal sclerosis included 27 participants (13 participants with right-sided and 14 participants with left-sided hippocampal sclerosis; 8 women and 19 men; mean age  $36.00 \pm 11.90$  years, range: 13–55 years; median: 38; interquartile range [IQR]: 17.5). At the time of MRI examination, all patients were at least 1-week seizure-free. All 27 hippocampal sclerosis participants were taking antiepileptic medications, and remained on their treatment throughout the study. The control group included 4 women and 10 men (mean age:  $35.07 \pm 11.95$  years; range: 18–55 years; median: 38; IQR: 20.75). As can be found in Table 1, there were no differences in age or sex between the groups ( $p > .05$ ).

#### 3.2 | Whole-brain gray and white matter network parameters between hippocampal sclerosis patients and controls

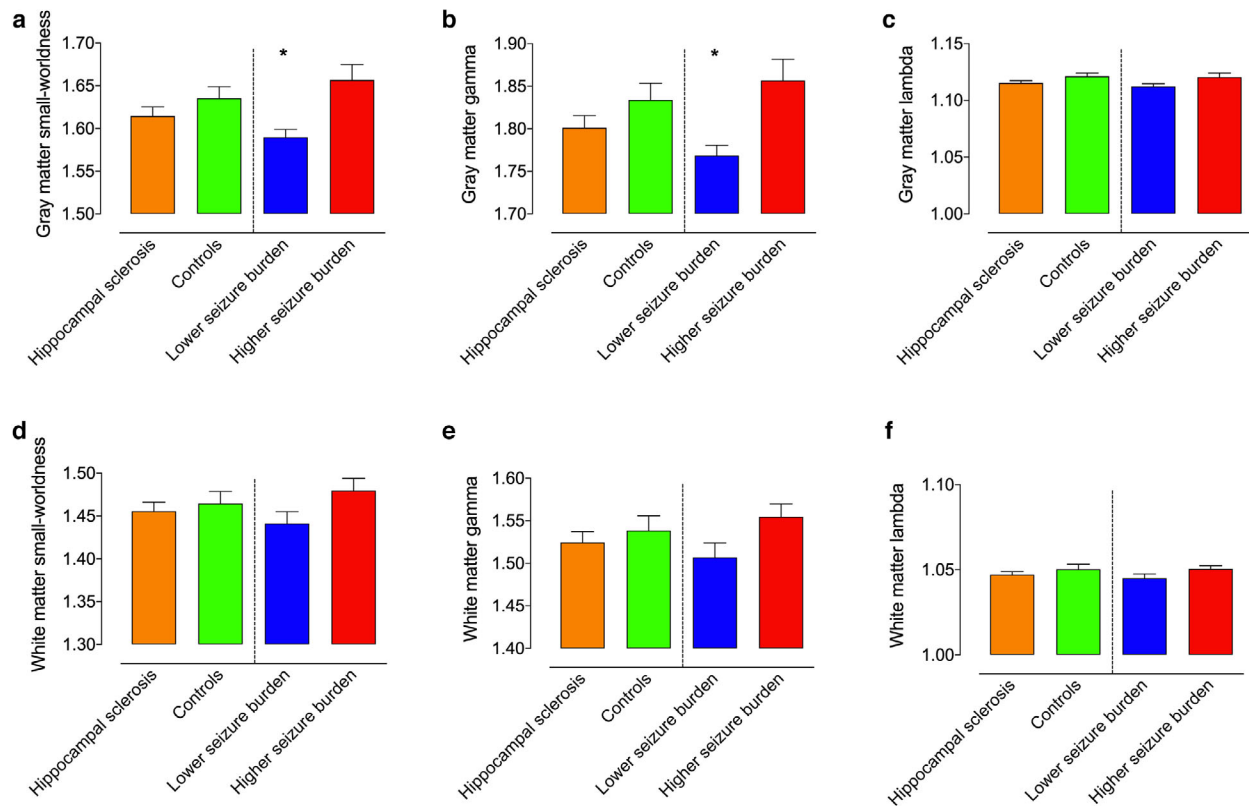
In both groups, gray and white matter networks had a small-world architecture ( $\gamma/\lambda \geq 1$ ), albeit means for all 27 patients were lower than for controls for small-worldness (more random network) and for  $\gamma$  and  $\lambda$  (less efficiently connected network; Figure 1 and Table 2). These effects were more pronounced for the gray matter network, reaching moderate effect sizes (Cohen's  $\delta$  for the gray matter gamma:  $-0.45$ ; Cohen's  $\delta$  for the gray matter lambda:  $-0.56$ ; Cohen's  $\delta$  for the gray matter small-worldness:  $-0.38$ ). However, none of these differences reached statistical significance ( $p > .05$ ).

#### 3.3 | Correlation between the network properties and clinical data

Having a higher average number of monthly seizures in the last year was associated with higher small-world topology (i.e., more rigid network) of the gray ( $r_s = .407$ ;  $p = .035$ ) and white matter structural network ( $r_s = .385$ ;  $p = .047$ ), driven by higher white matter clustering ( $r_s = .477$ ;  $p = .012$ ) and path length ( $r_s = .479$ ;  $p = .011$ ) (Figure 2 and Table 3).

There were no significant correlations between normalized volume of the affected hippocampus and the average number of monthly seizures in the last year ( $r_s = -.016$ ;  $p = .935$ ), years since diagnosis ( $r_s = .074$ ;  $p = .710$ ), and age at disease onset ( $r_s = .060$ ;  $p = .765$ ).

Also, there were no significant correlations between the average number of monthly seizures in the last year and current age ( $r_s = .014$ ;  $p = .944$ ), years since diagnosis ( $r_s = .156$ ;  $p = .435$ ), and age at first seizure ( $r_s = -.060$ ;  $p = .765$ ).



**FIGURE 1** Bar graphs demonstrating the gray matter small-worldness (a), gray matter gamma (b), gray matter lambda (c), white matter small-worldness (d), white matter gamma (e), and white matter lambda (f) of hippocampal sclerosis patients, controls, hippocampal sclerosis patients with a lower seizure burden and patients with a higher seizure burden.

Note: (\*) means significantly lower values in patients with a lower seizure burden compared to controls, and patients with a higher seizure burden.

**TABLE 2** Mean values of whole-brain network measures of hippocampal sclerosis patients versus controls

	Gray matter				White matter			
	Hippocampal sclerosis	Controls	Cohen's $\delta$	$p$ -Value	Hippocampal sclerosis	Controls	Cohen's $\delta$	$p$ -Value
Size	6663 $\pm$ 547	6715 $\pm$ 891	-0.08	0.817	. <sup>a</sup>	. <sup>a</sup>	. <sup>a</sup>	. <sup>a</sup>
Degree	1202 $\pm$ 136	1225 $\pm$ 184	-0.16	0.644	20.24 $\pm$ 1.13	19.98 $\pm$ 1.46	0.21	.532
Connectivity density	18.02 $\pm$ 1.21	18.22 $\pm$ 0.85	-0.19	0.590	0.233 $\pm$ 0.013	0.230 $\pm$ 0.017	0.21	.532
Gamma	1.801 $\pm$ 0.074	1.833 $\pm$ 0.073	-0.45	0.189	1.524 $\pm$ 0.066	1.538 $\pm$ 0.064	-0.21	.522
Lambda	1.115 $\pm$ 0.011	1.121 $\pm$ 0.011	-0.56	0.118	1.047 $\pm$ 0.009	1.050 $\pm$ 0.011	-0.32	.340
Small-worldness	1.615 $\pm$ 0.056	1.635 $\pm$ 0.051	-0.38	0.255	1.456 $\pm$ 0.055	1.465 $\pm$ 0.053	-0.17	.618

Note: Data are presented as means with standard deviation. An effect size of 0.2 is considered small, an effect size of 0.5 is considered medium, and an effect size of 0.8 is considered high.

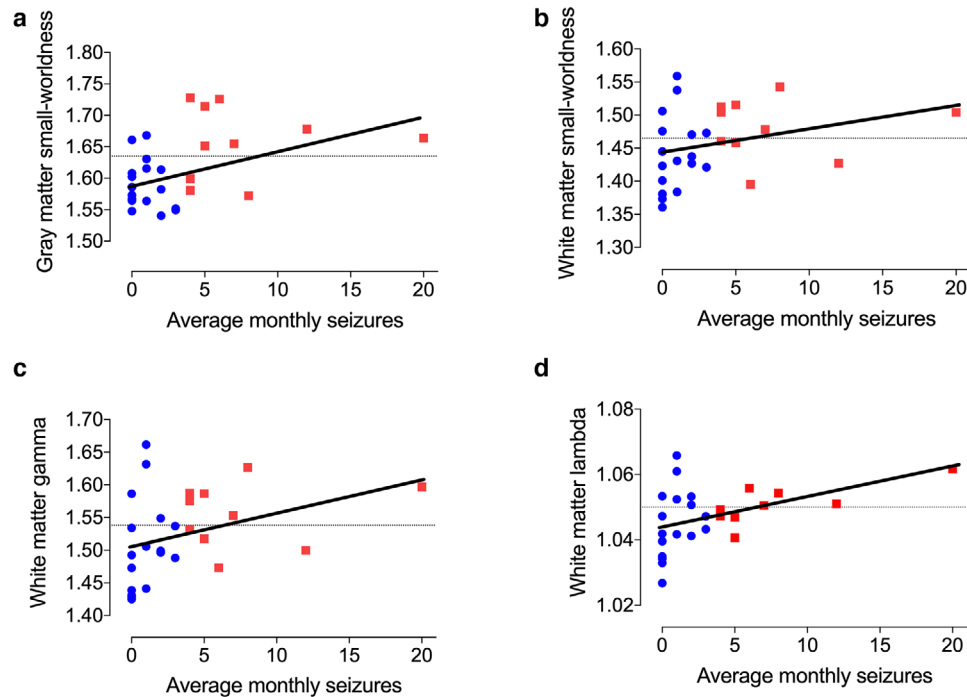
<sup>a</sup>As there were no disconnected nodes, the graph size was 89 (90 AAL regions minus Heschl) in all participants.

### 3.4 | Influence of seizures burden in the whole-brain structural networks parameters

Although in all patients together small-world values were nonsignificantly lower than those of controls (Figure 1), there was a significant correlation between higher small-worldness and more average monthly seizures in the past year (Figure 2). Upon closer inspection of the data presented in Figure 2, it is clear that patients with four or more

average monthly seizures seem to have higher small-worldness values. To further investigate the potential effect of monthly seizures we created two groups of patients: one with less than four and the other with four or more average monthly seizures in the past year, and compared network parameters between these groups.

Table 4 shows the distribution of the average monthly seizures in the past year. Seventeen patients had an average of less than four seizures and were characterized as having lower seizure burden. Ten patients



**FIGURE 2** Scatter plots of the significant associations between the average number of monthly seizures in the last year and graph theory properties, such as (a) gray matter small-worldness ( $r_s = .407$ ;  $p: .035$ ); (b) white matter small-worldness ( $r_s = .385$ ;  $p: .047$ ); (c) white matter gamma ( $r_s = .477$ ;  $p: .012$ ); and (d) white matter lambda ( $r_s = .479$ ;  $p: .011$ ).

Note: Blue circles represent patients with less than four seizures per month in the last year, red squares represent patients with four or more seizures per month in the last year. The dotted line represents the mean value of the control group for that property.

**TABLE 3** Correlations between global gray and white matter network properties and affected hippocampal volume, and clinical data of hippocampal sclerosis patients

	Years since diagnosis		Epilepsy onset age		Average monthly seizures in the last year	
	Gray matter	White matter	Gray matter	White matter	Gray matter	White matter
Gamma	0.101 (.615)	0.207 (.299)	-0.215 (.281)	-0.121 (.548)	0.378 (.052)	<u>0.477 (.012)</u>
Lambda	-0.169 (.399)	0.133 (.508)	-0.132 (.511)	-0.125 (.535)	0.101 (.616)	<u>0.479 (.011)</u>
Small-worldness	0.175 (.383)	0.201 (.315)	-0.222 (.266)	-0.126 (.532)	<u>0.407 (.035)</u>	<u>0.385 (.047)</u>
Affected hippocampus normalized volume	0.074 (.710)	-	0.060 (.765)	-	-0.016 (.935)	-

Note: Correlations are given as Spearman's rho with  $p$ -values between parentheses. The underlined values show statistically significant differences, considering a  $p < .05$ .

had an average of four or more seizures per month and were considered to have a higher seizure burden. As can be found in Table 4, these two groups were matched for age, sex, years of disease duration, and age at epilepsy onset.

As can be found in Figure 1 and Table 5, hippocampal sclerosis patients with a lower seizure burden had smaller gray matter small-worldness and gray matter gamma compared to the patients with a higher seizure burden and controls ( $p < .05$ ). Although those with a higher seizure burden had, on average, higher mean values than the controls, these differences did not reach statistical significance ( $p > .05$ ). Additional adjustment for age did not alter statistical significance of these findings.

## 4 | DISCUSSION

This study aimed at identifying global structural network properties in patients with hippocampal sclerosis compared with controls. In all patients, both gray and white matter network properties were statistically similar to those of controls, despite reaching moderate effect sizes for gray matter parameters. Interestingly, patients who had lower seizure burden, measured as less than four seizures per month on average over the last year, showed significantly decreased gray matter small-worldness and clustering. Thus, the network of these patients was more random and less efficient in nature. Although patients with higher seizure burden had higher average values than controls, these



**TABLE 4** Sociodemographic data of patients with higher burden seizures compared to patients with lower burden seizures

	Higher seizure burden hippocampal sclerosis	Lower seizure burden hippocampal sclerosis	p-value
Age (years)	32.70 (SD: 11.71; range: 13–48; median: 33; IQR: 9.5)	37.94 (SD: 11.92; range: 16–55; median: 41; IQR: 17)	.27
Sex	8 men / 2 women	11 men / 6 women	.66
Affected hippocampus normalized volume	4.76 (SD: 0.99; range: 2.54–5.82; median: 4.89; IQR: 1.21)	4.86 (SD: 0.83; range: 3.58–6.38; median: 4.92; IQR: 0.96)	.97
Years since diagnosis	20.70 (SD: 8.23; range: 8–34; median: 20.5; IQR: 9.75)	21.12 (SD: 12.91; range: 3–43; median: 19; IQR: 18.25)	.92
Age at epilepsy onset (years)	12.00 (SD: 11.92; range: 1–40; median: 8; IQR: 12.5)	16.88 (SD: 14.73; range: 1–45; median: 16; IQR: 20.5)	.38
Average monthly number of seizures in the last year	4 seizures: 3 patients 5 seizures: 2 patients 6 seizures: 1 patient 7 seizures: 1 patient 8 seizures: 1 patient 12 seizures: 1 patient 20 seizures: 1 patient	0 seizures: 8 patients 1 seizure: 4 patients 2 seizures: 3 patients 3 seizures: 2 patients	
Carbamazepine use			
Monotherapy (%)	0 (0)	3 (18)	
Combination therapy (%)	6 (60)	8 (47)	
Oxcarbamazepine use			
Monotherapy (%)	0 (0)	1 (6)	
Combination therapy (%)	0 (0)	2 (12)	

**TABLE 5** Mean values of whole-brain network measures of hippocampal sclerosis patients with a higher burden of seizures per month versus hippocampal sclerosis patients with a lower burden of seizures per month and controls

	Gray matter				White matter			
	Higher burden hippocampal sclerosis	Lower burden hippocampal sclerosis	Controls	p-Value	Higher burden hippocampal sclerosis	Lower burden hippocampal sclerosis	Controls	p-Value
Gamma	1.856 ± 0.078	1.768 ± 0.050	1.833 ± 0.073	.003*	1.554 ± 0.048	1.506 ± 0.070	1.538 ± 0.064	.149
Lambda	1.120 ± 0.011	1.112 ± 0.009	1.121 ± 0.011	.051	1.051 ± 0.005	1.045 ± 0.010	1.050 ± 0.011	.238
Small-worldness	1.656 ± 0.054	1.589 ± 0.038	1.635 ± 0.051	.003 <sup>#</sup>	1.480 ± 0.045	1.441 ± 0.056	1.465 ± 0.053	.180

Note: Data are presented as means with standard deviation.

\*Bonferroni: Gray matter gamma—Higher burden hippocampal sclerosis versus controls ( $p = .409$ ); higher burden hippocampal sclerosis versus lower burden hippocampal sclerosis ( $p = .002$ ); lower burden hippocampal sclerosis versus controls ( $p = .009$ ).

<sup>#</sup>Bonferroni: Gray matter small-worldness—Higher burden hippocampal sclerosis versus controls ( $p = .867$ ); higher burden hippocampal sclerosis versus lower burden hippocampal sclerosis ( $p = .004$ ); lower burden hippocampal sclerosis versus controls ( $p = .037$ ).

did not reach statistical significance. However, they did influence the positive correlation between higher monthly seizure number in the past year and higher gray and white matter network properties, indicating a relationship with a more rigid structural network.

The small-world characteristics of brain networks enable the specialization and integration of information transfer at relatively low wiring costs (Haneef & Chiang, 2014; Stam, 2010). In the present study, all patients and controls had a small-world topology, whereas network parameters were not different when comparing all patients as one

group to controls. Our overall results are somewhat different from the results of other studies. One study, using cortical thickness to create structural networks, found increased clustering and path length (i.e., a more rigid network) with a more central role for the paralimbic structures within the network of patients with refractory temporal lobe epilepsy compared to controls (Bernhardt et al., 2011). Other studies, using DTI for white matter network reconstruction, found that patients with mesial temporal lobe epilepsy exhibited increased global network clustering coefficient compared with controls. Regionally, the hip-

pocampus became less well connected (reduced clustering), whereas limbic structures, the insula and thalamus, and other extratemporal regions were found to have increased clustering and less efficiency, indicating more rigidity of the network (Bonilha et al., 2012; Liu et al., 2014).

Upon further inspection, this absence of statistically significant differences in network properties within all patients as one group was due to a clear distinction in these network parameters between patients with lower (less than four) and higher (four or more) seizure burden. This cutoff was guided by our data, as there are no clinical guidelines determining what constitutes as lower and higher seizure burden. Our patients with lower seizure burden (17 in total) showed significantly decreased gray matter small-worldness and clustering, whereas these measures were nonsignificantly increased in those with higher seizure burden compared to controls. For white matter small-worldness and clustering a similar trend was observed. This distinction explains why, when pooling all patients, there were no statistically significant alterations between patients and controls. Figure 2 shows that for gray matter small-worldness the effect of the number of seizures is very clear. Only two patients with a lower seizure burden (11.8%), relative to seven of those with a higher seizure burden (70%) had values above that of the mean of controls (Figure 2 dotted line). For the white matter network, the results are more mixed, which may explain the lack of statistically significant results here. That only the gray matter network was significantly altered may be related to the fact that seizures are generated in the gray matter. Further studies in larger samples with more accurate seizure frequency determination are needed to assess the influence of seizure frequency on networks.

Small-worldness of both gray and white matter networks captured clinically relevant information, as it was correlated with the number of seizures. Here, a higher number of seizures was related to a less flexible and efficient, and a more regular network topology (i.e., higher small-worldness values). However, it is clear that this correlation is driven by the 10 patients with a higher seizure burden, and that we lack statistical power to determine the correlations in both groups separately. As described above, both a more random and a more rigid network hamper efficient information exchange within the network and could be pathologically important with different effects for patients. Future studies will need to determine the exact effects of a more random and more rigid network in patients with hippocampal sclerosis, for example, in terms of treatment response, surgical outcome, and cognitive functioning. One study, interestingly, has shown that a more rigid gray matter network topology was related to poorer surgery outcome, highlighting the clinical importance of small-worldness (Bernhardt et al., 2015).

Previous studies have shown that in patients who are not seizure-free after temporal lobe surgery their presurgery global networks were not different from those of patients who obtained seizure freedom after surgery (Bonilha et al., 2013). However, the local presurgery network of patients who were not seizure-free after temporal lobe surgery had a higher degree of connectivity between the entorhinal cortex and superior temporal gyrus, between contralateral temporal pole and contralateral supramarginal gyrus, and between ipsilateral parahippocam-

pal gyrus and ipsilateral superior parietal gyrus (Bonilha et al., 2013). This corroborates the hypothesis that the effects of mesial temporal lobe epilepsy may be more significant in local networks, which not necessarily transfer to global alterations. In addition, Lee and Park (2020) found that patients with drug-resistant temporal lobe epilepsy due to hippocampal sclerosis have more severe intrinsic hippocampal network disruptions than patients with drug-controlled epilepsy due to hippocampal sclerosis.

The use of carbamazepine or oxcarbazepine in temporal lobe epilepsy has been associated with a lower functional betweenness centrality, compared with those patients not using one of these medications (Haneef et al., 2015). In our sample, some patients were using carbamazepine or oxcarbazepine, as monotherapy or in combination with other antiepileptic drugs (Table 4). Given the low number of patients using these medications, and the combination therapy that most patients were on, it was not possible in this study to determine any medication effect on network parameters. Additionally, in patients with chronic epilepsy, a negative correlation has been found between clustering coefficient/local efficiency of white matter structural networks and cognitive function (Vaessen et al., 2012). Furthermore, epileptic activity can reduce long-range connectivity and enhance local connectivity in the postseizure functional connectivity compared to the preseizure functional connectivity (Liang et al., 2020). As we included, in the current study, only patients using antiepileptic drugs, and some were well controlled for several months, this may also explain the absence of statistically significant differences in the global brain structural network properties between patients with hippocampal sclerosis and healthy controls.

Limitations include the relatively small sample size. Although we defined strict inclusion criteria to include homogenous groups, the results may be an underestimation. However, despite the limited sample size, we were able to demonstrate a strong effect of seizure burden, especially on gray matter network topology. Differences may exist between left- and right-sided temporal lobe epilepsy regarding network measures. In this study, we did not observe any such effect, possibly due to either the small sample size and subsequent lack of statistical power or due to the influence of seizure burden. Also, we did not perform multiple comparisons correction in our correlation analysis. Although this increases the risk of false-positives, we feel this approach is justified given the limited knowledge of disease-related correlates of structural network in mesial temporal lobe epilepsy and the small sample size. Because resting-state functional MRI was not acquired and we could not assess the effects of hippocampal sclerosis on the functional network.

## 5 | CONCLUSION

Patients with lower seizure burden had significantly lower gray matter network indices, indicating a more random organization. The correlation between higher monthly seizures and a more rigid network is driven by those with higher seizure burden, who presented with a more rigid network compared to those with a lower seizure burden. Future



studies, in larger samples, should determine the extent and importance of small-worldness and other global and local network properties in hippocampal sclerosis patients.

## CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

## PEER REVIEW

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