Neurobiology of Disease 124 (2019) 373–378

Contents lists available at ScienceDirect

Neurobiology of Disease

journal homepage: www.elsevier.com/locate/ynbdi

Changes of dimension of EEG/ECoG nonlinear dynamics predict epileptogenesis and therapy outcomes

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ARTICLE INFO

Keywords:
Biomarker
Animal models of epilepsy
Recurrence quantification analysis
Disease modification treatments

ABSTRACT

The lack of early biomarkers of epileptogenesis precludes a sound prediction of epilepsy development after acute brain injuries and of the natural course of the disease thus impairing the development of antiepileptogenic treatments. We investigated whether the dimensional changes of nonlinear dynamics in EEG/ECoG signals, that were recorded in the early aftermath of different epileptogenic injuries, provide a measure to be exploited as a sensitive prognostic and predictive biomarker for epilepsy. Using three different models of epilepsy in two rodent species, we report a common and significant decrease of nonlinear dynamics *dimension* in EEG/ECoG tracings during early epileptogenesis. In particular, the magnitude of this dimensional decrease predicts the severity of ensuing epilepsy, and this measure is modulated by disease-modifying or antiepileptogenic treatments. The broad application of EEG/ECoG monitoring in epilepsy underlines the translational value of these findings for enriching the population of patients at risk for developing epilepsy in clinical investigations.

1. Introduction

Epilepsy affects approximately 65 million of people worldwide of whom 15–60% (resident in western world and developing countries, respectively) develop epilepsy following acute brain injuries such as neurotrauma, stroke, status epilepticus and encephalities (Klein and Tyrlikova, 2017).

There is no possibility as yet to identify people at risk of developing epilepsy following brain injury, or to predict the clinical course of the disease since the available clinical indicators have poor sensitivity (Pitkänen et al., 2016). The lack of biomarkers also affects the preclinical development of antiepileptogenic therapies and impairs their clinical translation due to demanding resources for carrying out sufficiently powered and long-lasting clinical studies (Pitkänen et al., 2016).

Intensive research has been devoted to identify molecular signatures in body fluids or by brain imaging, and EEG/ECoG signal analyses for discovering sensitive and specific biomarkers of epileptogenesis (Pitkänen et al., 2016). Clinical and preclinical investigations routinely rely upon EEG/ECoG monitoring suggesting that a marker of epileptogenesis identified by examining brain electrical activity could be easily combined with other biomarkers and translated to a clinical setting. In this context, we have recently shown by nonlinearity tests that the presence of nonlinear dynamics in the EEG/ECoG signals was increased during the early phase of epileptogenesis in rodents exposed to an acute brain injury (Rizzi et al., 2016a, 2016b). We characterized these nonlinear dynamics as intermittency by observing an increased incidence of laminarity which was abolished by antiepileptogenic treatments (Rizzi et al., 2016b). However, laminarity was not sensitive enough to reliably predict epilepsy development in animal models therefore requiring the search for additional measures.

Notably, the emergence of nonlinear dynamics is usually associated with a reduction of the number of degrees of freedom of the underlying system (Heylighen, 2002; Negru, 2016). Since EEG/ECoG signal *dimension* is a measure of the number of degrees of freedom of the dynamic system (Heylighen, 2002), our findings suggest that signal *dimension* may decrease during epileptogenesis, thus providing a

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https://doi.org/10.1016/j.nbd.2018.12.014
Received 28 September 2018; Received in revised form 23 November 2018; Accepted 21 December 2018
Available online 24 December 2018
0969-9961/

potential biomarker of the diseased brain. In support of this hypothesis, EEG *dimension* is lower in human epileptic foci than in peri-focal areas (Lehnertz and Elger, 1995). This decrease is inversely correlated with the serum levels of carbamazepine, thus supporting that the extent of decrease in the EEG dimension reflects the propensity of the epileptic focus to generate seizures (Lehnertz and Elger, 1997).

Based on this evidence, we studied whether the establishment of ictogenic networks during epileptogenesis is accompanied by a progressive decrease of the EEG/ECoG dimension, and if the magnitude of such decrease predicts the severity of the ensuing epilepsy. To this aim, we measured EEG/ECoG dimension variations during epileptogenesis in three well-established animal models of epilepsy arising following different brain insults and with differing neuropathologic sequelae (Weissberg et al., 2015; Brandt et al., 2016; Pauletti et al., 2017; Walker et al., 2017). We also determined whether the EEG/ECoG dimension is a sensitive measure of the effect of disease-modifying or anti-epileptogenic treatments which were previously tested in each of these models (Weissberg et al., 2015; Brandt et al., 2016; Pauletti et al., 2017; Walker et al., 2017).

2. Materials and methods

2.1. Experimental models of epileptogenesis

Our retrospective investigations were based on the analysis of either EEG or ECoG recordings previously acquired from rats exposed to status epilepticus (SE) evoked by electrical stimulation of the basolateral amygdala (SE-BLA; Brandt et al., 2016) or the ventral hippocampus (SE-HIPPO; Pauletti et al., 2017; Walker et al., 2017), or mice exposed to subchronic intracerebroventricular (ICV) infusion of albumin (ICV-ALB; Weissberg et al., 2015). These former studies either refined existing epilepsy models (Brandt et al., 2016) or established new epilepsy models (Weissberg et al., 2015) or tested new antiepileptogenic treatments (Pauletti et al., 2017; Walker et al., 2017; Weissberg et al., 2015). Supplementary Table 1 reports the animal models and describes the treatments given during epileptogenesis and the related therapeutic outcomes.

2.2. EEG/ECoG signal acquisition process

The original EEG/ECoG recordings were acquired using different filtering settings and sampling rates, as described: SE-BLA model, low-pass filter 50 Hz, sampling rate 200 Hz; SE-HIPPO model, low-pass filter 70 Hz, notch filter 50 Hz (power grid frequency), sampling rate 400 Hz; ALB-ICV model, low-pass filter 100 Hz, notch filter 50 Hz (power grid frequency), sampling rate 500–1000 Hz (re-sampled at 400 Hz for analysis). We restricted the frequency range of signals by a low-pass 50 Hz filter so that all time-series analyzed presented the same frequency range, which was also compatible with the different sampling rates. Importantly, although filtering may alter or exclude potential nonlinear signalling embedded in the original time-series, our settings always included frequency bands (< 50 Hz) which were shown useful to investigate on dynamics of the epileptic brain, including nonlinear phenomena (Chauvière et al., 2009; Seo et al., 2013; Rizzi et al., 2016a).

In rats exposed to electrically-induced SE, the EEG signals used for nonlinear analysis originated from the stimulating electrode while in ICV-ALB exposed animals the recording electrode was placed over the somatosensory cortex. In sham animals (not exposed to electrical stimulation or injected ICV with vehicle), the EEG/ECoG signals originated from electrodes located in brain areas matched with those exposed to the epileptogenic injury. Supplementary Table 2 reports the number of animals in each experimental group and the total EEG/ECoG epochs analyzed.

2.3. Animal exclusion criteria

We used EEG/ECoG tracings from animals without acute symptomatic seizures during the entire observation period corresponding to the epileptogenesis phase prodromal to spontaneous seizures onset. I.e., no seizures were observed in the ICV-ALB for 144 h from the start of albumin perfusion, or for 120 h in the SE-HIPPO model or for 120 h in the SE-BLA model except for one SE-BLA rat which was therefore excluded from the analysis.

2.4. Estimate of EEG/ECoG signals dimension by the recurrence quantification analysis

To estimate the EEG/ECoG signals dimension, we exploited the Recurrence Quantification Analysis (RQA) (Webber and Zbilut, 1994; Marwan et al., 2007), a powerful and sensitive mathematical tool that was developed for detecting and measuring several aspects of nonlinear dynamics in short, noisy and nonstationary time-series, as are those of biological origins, without any assumption on the nature of those dynamics. RQA has been successfully used for investigating epileptic phenomena (Ouyang et al., 2008; Ngamga et al., 2016; Rizzi et al., 2016a, 2016b). The type of dimension we have measured is technically defined *embedding dimension* (**Supplementary Material**). Thus, we will use the term *estimated embedding dimension (Est-ED)*, or for simplicity *dimension,* unless differently specified. Our goal was to estimate an embedding dimension that can be considered sufficiently close to the actual *minimal* embedding dimension necessary for the unfolding of trajectories of the main nonlinear dynamics characterizing EEG/ECoG signals. Basically, the analytical protocol was implemented by taking into consideration the general principle according to which no significant changes are expected to occur for the values of recurrence variables across embedding dimensions, until an insufficient embedding is applied (Zbilut et al., 2002a,b; Webber and Zbilut, 2005). We evaluated the embedding dimension-dependent variations of the fundamental variables *determinism* and *laminarity* which strictly depend on the percent of recurrences and are directly related to the main recurrence patterns induced by nonlinear dynamics (see Supplementary Material for details). Importantly, since the embedding dimension is an input parameter of the RQA, each EEG/ECoG epoch was analyzed using the progressively decreasing embedding dimensions 25, 21, 17, 13 and 9, a set of dimensions appropriately selected as detailed in Supplementary Material. Moreover, to test the robustness of variations of RQA variables across the selected embedding dimensions, five confidence intervals were calculated by nonparametric bootstrapping technique and compared for each dataset.

Importantly, the quality of nonlinear signals we measured is not influenced by the impedance of the surface vs depth electrodes since we previously performed nonlinearity tests in the ICV-ALB and SE-HIPPO models. These tests showed that the different electrode impedances do not impair the detection of nonlinearities in EEG/ECoG signals, thus allowing reliable measurements of nonlinear variables (Rizzi et al., 2016a, 2016b).

2.5. Statistical analysis

Time-dependent changes of Est-EDs were analyzed by two-way ANOVA. Cumulative changes of Est-EDs were analyzed by nonparametric Mann-Whitney or Kruskal-Wallis tests, as appropriate, with correction for ties among values. Correlation analysis was performed by nonparametric Spearman's test. Statistics and graphical representation of results were performed using GraphPad Prism (version 7.03 for Windows, www.graphpad.com) except for calculations of confidence intervals by nonparametric bootstrapping (see Supplementary Material) and Youden index which were performed using software packages developed for the statistical computing environment R (Davison and Hinkley, 1997; Robin et al., 2011; RStudio Team, 2015; R Core Team,

2016; Canty and Ripley, 2017).

2.6. Computational resources and applications

High-Throughput-Computing (Barbera et al., 2011) and Cloud-Computing technologies were adopted for shortening calculation times. Specifically, computational and storage resources were provided by the European Grid Infrastructure (EGI), the Europe's leading e-infrastructure co-funded by the European Union in the context of the 6th and 7th Framework Program.

The RQA was implemented by the applications *RQE* and *RQH* developed by Webber (freely available for Windows operating systems at http://homepages.luc.edu/~cwebber), after adapting and recompiling the source codes for Linux operating systems. Other applications were *notch*, *mutual, minima* and *extrema*, all included in the TISEAN software package (Hegger et al., 1999) (freely available at http://www.mpipks-dresden.mpg.de/~tisean).

2.7. Data availability

All datasets of RQA variables and related CIs used to estimate EEG/ECoG signals dimension are available from corresponding author upon request. Original EEG/ECoG recordings are available upon request at the respective laboratories.

3. Results

3.1. Changes in estimated embedding dimension in EEG/ECoG signals during epileptogenesis

Our first approach was to investigate whether the estimated embedding dimension (Est-ED) undergoes changes during epileptogenesis in animals developing epilepsy after different brain insults.

In SE-BLA rats which developed epilepsy (Epi, $n = 8$; Fig. 1A,B), Est-ED was significantly reduced during the first 24 h post-insult reaching a nadir at 48 h, then recovering to sham values (in rats not exposed to SE and without seizures, $n = 10$) by 96 h. Similarly, ICV-ALB mice which developed epilepsy in the absence of acute SE (Epi, $n = 7$; Fig. 1D,E) showed a significant reduction in Est-ED compared to vehicle-exposed mice (without seizures; n = 10). This decrease was longer lasting compared to SE-BLA since Est-ED did not recover to vehicle values during 144 h of recording (Fig. 1D).

In SE-BLA and ICV-ALB models, one group of animals did not develop epilepsy in spite all animals were exposed to the same injury, thus providing a powerful tool for biomarker validation. In particular, the SE-BLA model was refined by calibrating SE duration in order to evoke epilepsy in around 50% of rats (Brandt et al., 2016). Differently, in the ICV-ALB model, the cohort of mice which did not develop epilepsy was treated with SJN2511, a specific ALK5/TGF-β inhibitor, during albumin infusion ($n = 6$; Weissberg et al., 2015). In both models, the Est-ED reduction in animals not developing epilepsy (No Epi) - either because of the natural course of the disease (SE-BLA) or because of drug treatment (ICV-ALB) - was transient since it occurred only during the first 24 h post-insult, then returning to sham values within 48 h (Fig. 1A,D). This evidence shows that the decrease of temporal dynamics of Est-ED differentiate animals developing or not developing epilepsy starting from 48 h post-insult. ROC analysis of values measured from 48 h to 72 h post-injury showed an AUC = 0.8863 ($p < .01$) in SE-BLA rats (Fig. 1C) and an AUC = 0.905 ($p < .01$) in ICV-ALB mice (Fig. 1F). We tested also specificity and sensitivity of the Est-ED measure for discriminating between animals developing or not developing epilepsy using the cumulative Est-ED values measured in the SE-BLA and ALB-ICV models. ROC analysis showed an excellent discriminating power of this measure (AUC = 0.8869; $p < .01$), providing a cut-off threshold = 21 (Youden index).

We also analyzed Est-ED in SE-HIPPO rats developing epilepsy associated with relatively high seizures number (*severe epilepsy*: 20.2 ± 5.0 seizures/2 weeks, $n = 18$ vehicle injected rats) or with lower seizure number (*mild epilepsy*: 9.3 ± 4.1 seizures/2 weeks, n = 18 drug treated rats) ($n = 9$ rats/treatment or vehicle group). The animals with less seizures (*mild epilepsy*) were treated during epileptogenesis with antiinflammatory (Fig. 2A,B; Walker et al., 2017) or antioxidant (Fig. 2C,D; Pauletti et al., 2017) drugs (Suppl. Table 1). In all epileptic rats, Est-ED was significantly reduced below sham values (in rats not exposed to SE) within 48 h post-SE and for additional 72 h ($n = 25$ rats, Fig. 2A,C). Notably, the animals with severe epilepsy (vehicle-treated) displayed a greater Est-ED reduction than rats with milder epilepsy (drug-treated) when compared to shams (Fig. 2B,D). We also ranked the severity of epilepsy in each experimental group by reckoning the progression index, i.e., the ratio between the median number of seizures at 5 months vs. 2.5 months post-SE (2 weeks 24/7 EEG recording at each time point) (see **Supplementary Material** for details). A significant inverse correlation was found between the median cumulative Est-ED measure and the severity of epilepsy in the animals (Table 1).

We also analyzed if a reduction in Est-ED was a sensitive measure of epilepsy severity due to the natural course of the disease. We analyzed therefore Est-ED in all vehicle-injected rats (n = 18; 9 rats in each vehicle injected group) by grouping animals according to the number of seizures they displayed in the chronic disease phase (**Supplementary Table 3**). The Est-ED was progressively reduced in rats with increasing number of spontaneous seizures (Fig. 2E). This set of data provides the proof-of-concept evidence that the magnitude of decrease in Est-ED is a measure of disease severity also in the absence of treatments (Fig. 2E).

4. Discussion

Our data provide novel evidence that the EEG/ECoG dimension is reduced during the early phase of epileptogenesis compared to healthy animals. Notably, the temporal dynamics of this decrease predict which animals will develop epilepsy. In fact, if Est-ED is only transiently reduced after the acute brain injury (resolving by 48 h) then rats or mice do not develop spontaneous seizures, as shown in the SE-BLA and ICV-ALB models. On the contrary, a longer lasting reduction (72–144 h) is a common feature of animals developing epilepsy independently on their inciting event.

Notably, *the extent of the dimensional decrease* also predicts the severity of ensuing epilepsy based on the number of spontaneous seizures in the chronic disease phase. This was demonstrated by two different approaches.

First, Est-ED prospectively differentiates animals developing severe epilepsy vs. animals developing a milder form of epilepsy due to a treatment. In fact, the epileptogenesis models we have examined responded to disease-modifying treatments with variable outcomes ranging from prevention of epilepsy (ICV-ALB model; Weissberg et al., 2015) to an overall 50% decrease in spontaneous seizures frequency and prevention in seizure progression (SE-HIPPO model; Pauletti et al., 2017; Walker et al., 2017). Thus, our findings show that the decrease in Est-ED during epileptogenesis is a sensitive measure to disease-modifying drugs.

Second, the magnitude of the Est-ED reduction was larger in the vehicle-injected rats with higher number of spontaneous seizures, thus providing a measure of disease severity independently on treatments.

Since our findings are based on EEG/ECoG monitoring routinely used in the clinic, they provide a rapidly translatable measure for clinical validation. Moreover, our measure may be exploited to improve investigations on basic mechanisms of epileptogenesis, for developing antiepileptogenic therapies and discover biomarkers. Indeed, the possibility to predict early post-injury which animals will develop epilepsy, and the associated disease's course, allows to enrich the experimental groups and also may reduce both unnecessary treatments and the follow-up time for determining the success of a therapy. Additionally,

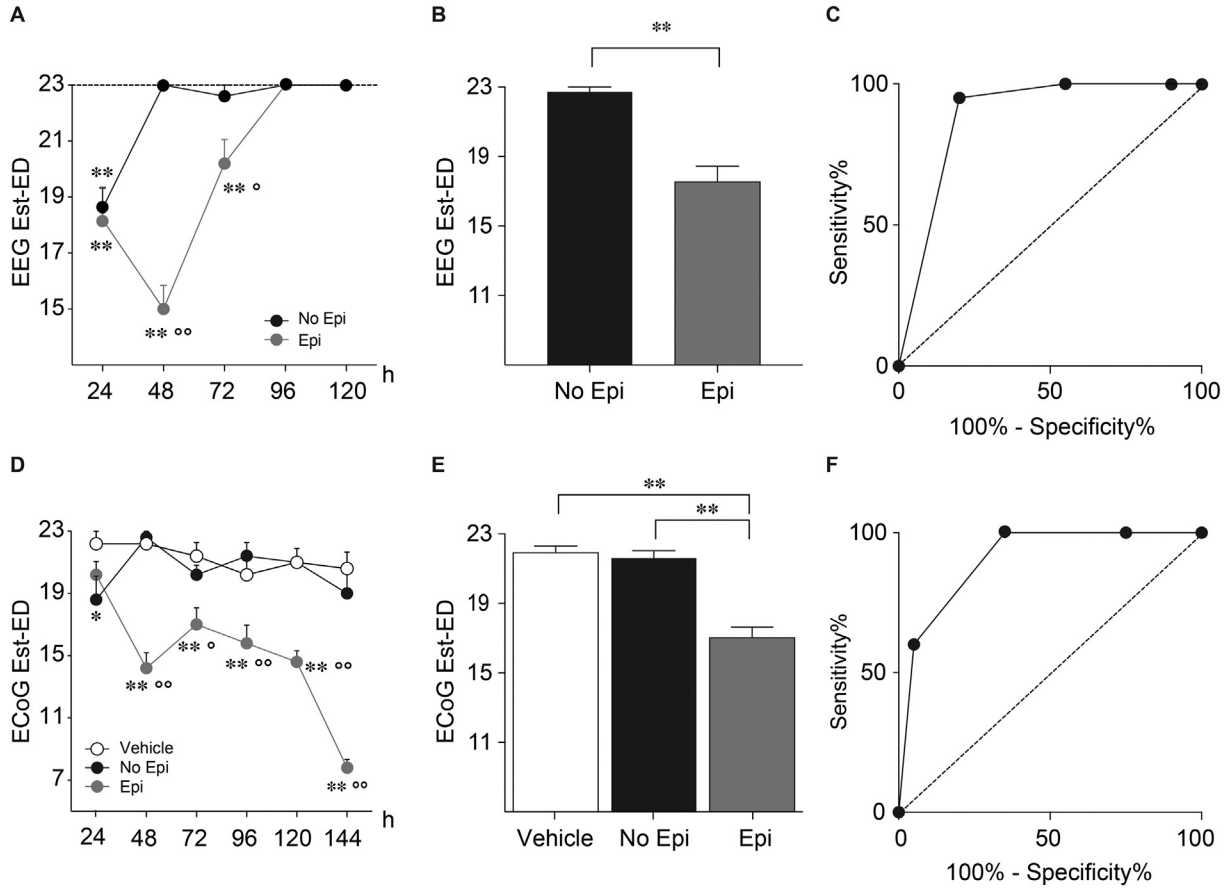


Fig. 1. *Time-dependent changes in Est-ED during epileptogenesis.*

Panels A and D (mean ± SEM; $n = 10$ replicates of Est-ED measure/time interval) depict the dynamics of dimensional changes in rats exposed to status epilepticus induced by electrical stimulation of the basolateral amygdala (A) or in mice infused icv with serum albumin and receiving either SJN2511 (no Epi) or saline (Epi) (D). Brain activity was measured by depth electrode EEG recordings in the stimulated amygdala (A,B) or by ECoG recordings with screw cortical electrodes (D,E). *No Epi*, animals not developing epilepsy; *Epi*, animals developing epilepsy; *Vehicle*, animals treated icv with albumin solution vehicle.

Panels B, E: Cumulative Est-ED values (mean ± SEM; $n = 20$ replicates of Est-ED measure/group) measured 48 h to 72 h post-insult.

In A: ** $p < .01$ vs. Sham (rats not exposed to status epilepticus; median value = 23; *dotted line*) by Kruskal-Wallis test; °$p < .05$, °°$p < .01$ vs. No Epi by two-way ANOVA. In D: *p < .05, ** p < .01 vs. Sham °p < .05, °°p < .01 vs. No Epi by two-way ANOVA. In B: ** p < .01 vs. No Epi by one-tailed Mann-Whitney test. In E: ** p < .01 vs. No Epi and Sham by Kruskal-Wallis test.

Panels C,F: ROC curves for values measured from 48 h to 72 h post-injury; AUC = 0.8863 (C), AUC = 0.905 (F), $p < .01$.

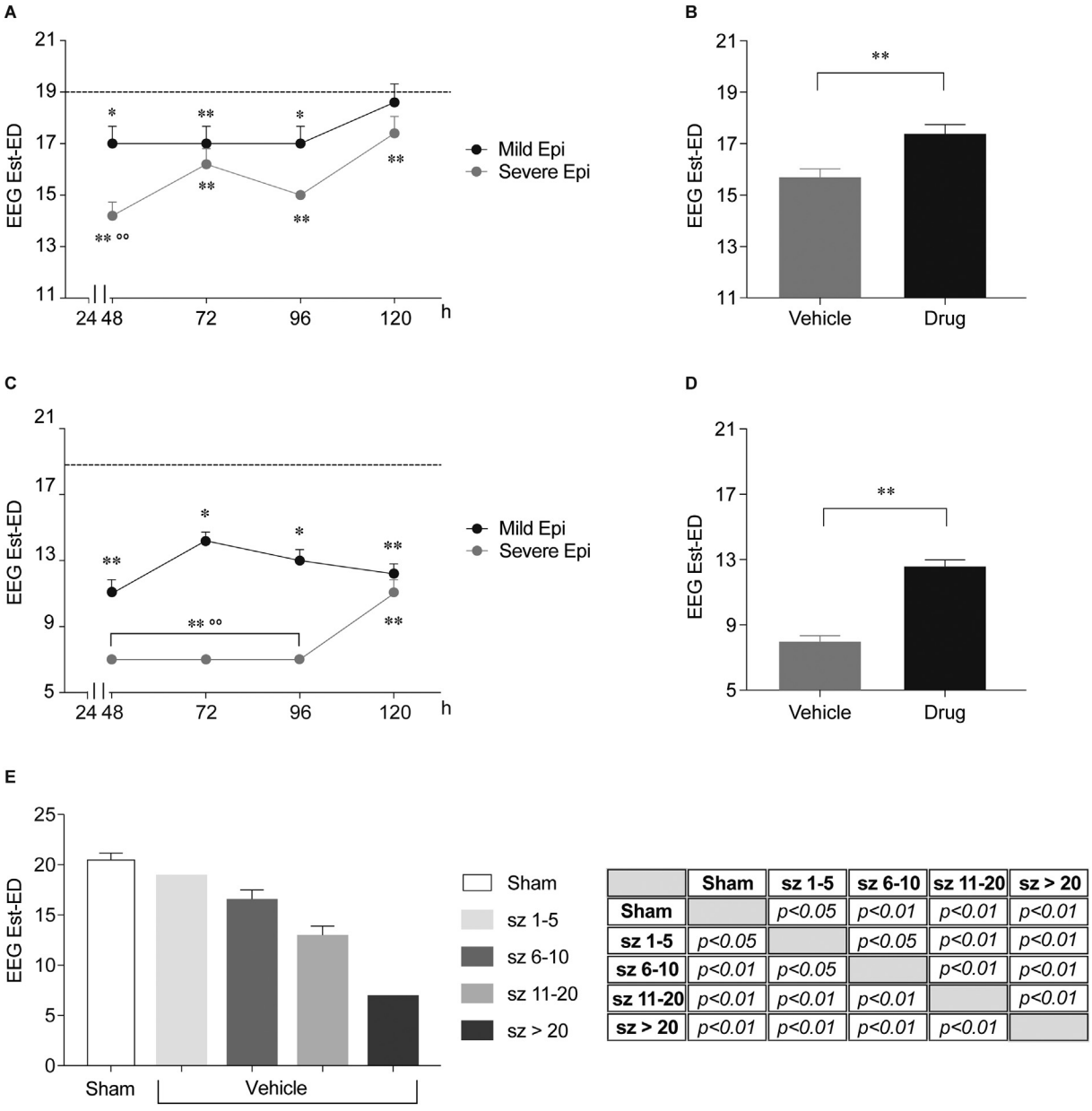
the correlation between signal dimension decrease and the severity of epilepsy may be exploited to predict the epileptogenicity of a given brain injury.

Our findings have mechanistic implications since the reduction in EEG/ECoG signal dimension likely results from underlying pathogenic events affecting neuronal networks. In support, the duration of this effect is influenced by the nature of the epileptogenic insult as well as by therapeutic interventions that improve the disease outcomes. In this context, the first 48 h post-injury are pivotal for distinguishing which animals will develop epilepsy supporting the view that anti-epileptogenic interventions should be applied early post-insult to increase the chances of therapeutic success (Pitkänen et al., 2016).

A prerequisite for the clinical validation of the EEG/ECoG signal dimension reduction as an early biomarker of epileptogenesis is that this phenomenon indeed occurs in humans exposed to different brain insults. In support, our results show that the reduction of signal dimension is a common feature of three different models of epilepsy in two species where animals are exposed to diverse brain injuries. In particular, signal dimension was decreased after both convulsive (SE-HIPPO, SE-BLA) and non-convulsive stimuli (ICV-ALB), independently on the presence or absence of brain lesions, and preceded both progressive (SE-HIPPO) and non-progressive (SE-BLA, ALB-ICV) forms of epilepsy. These findings suggest that the dimensional decrease is a common characteristic of brain activity associated with the establishment of an epileptogenic network. Therefore, it is likely that similar changes occur also in humans exposed to differing epileptogenic injuries. Notably, a dimension decrease of EEG signals recorded from depth electrodes was previously reported in epileptic *foci* of epilepsy patients showing that this phenomenon is not restricted to experimental models (Lehnertz and Elger, 1995, 1997).

We measured a decrease in signal dimension not only using depth electrodes but also analyzing ECoG tracings, therefore extending the applicability of this analysis to epidural clinical monitoring. Moreover, our measurements are based on a single recording electrode. It is likely that if multiple recording sites are analyzed, the changes in signal dimension may be detected with higher sensitivity in individual patients. Potential difficulties in clinical translation should be taken into account, such as the unknown location of the epileptogenic area or its distance from the scalp recording electrodes which may impair the detection of the Est-ED measure with sufficient sensitivity and reliability.

In summary, signal dimension measurements following exposure to epileptogenic insults provide a potential biophysical marker of epileptogenesis. If this measure is validated in prospective clinical studies, it will be of significant value to enrich the patient population for testing novel therapeutics and to predict the response to therapies improving



	Sham	sz 1-5	sz 6-10	sz 11-20	sz > 20
Sham		*p<0.05*	*p<0.01*	*p<0.01*	*p<0.01*
sz 1-5	*p<0.05*		*p<0.05*	*p<0.01*	*p<0.01*
sz 6-10	*p<0.01*	*p<0.05*		*p<0.01*	*p<0.01*
sz 11-20	*p<0.01*	*p<0.01*	*p<0.01*		*p<0.01*
sz > 20	*p<0.01*	*p<0.01*	*p<0.01*	*p<0.01*	

Fig. 2. *Modulation of Est-ED by treatments.*
Panels A,C: dynamics of EEG dimensional changes in the hippocampus of rats exposed to status epilepticus induced by electrical stimulation of the ventral hippocampus. Rats with mild epilepsy (Mild Epi) were treated 1 h after status epilepticus onset (A) for 1 week with antiinflammatory drugs (Anakinra + BoxA; n = 10 Est-ED replicates/time) or (C) for 2 weeks with antioxidant drugs (N-Acetylcysteine + Sulforaphane; n = 10 Est-ED replicates/time). Vehicle-injected SE rats developed more spontaneous seizures than treated animals (Severe Epi; see text for details). *Panels B,D*: *cumulative Est-ED* values measured 48–120 h post-SE (*n* = 40 Est-ED replicates/ group).
Panel E reports the *cumulative Est-ED* values measured 48–120 h post-SE (n = 10 Est-ED replicates/ group) in all vehicle-injected rats with increasing number of spontaneous seizures in the chronic epilepsy phase. Details on the number of animals and seizures in each group are reported in Suppl. Tables 2 and 3.
Data are mean ± SEM. Panels *A,B*: *p < .05, **p < .01 vs. Sham (rats not exposed to status epilepticus; median value = 19; *dotted line*) by Kruskal-Wallis test; °°p < .01 vs. Mild Epilepsy by two-way ANOVA. EEG dimensional changes at 24 h post-SE were not available. *Panels B,D*:
** p < .01 by one-tailed Mann-Whitney test. *Panel E*: the table reports statistical level of significance by one-tailed Mann-Whitney test.

disease prognosis.

As far as concern other candidate markers of epileptogenesis based on EEG/ECoG analysis, we recently reported a reduced incidence of slower frequencies (*theta* band) and their inverse relationship with the incidence of higher frequencies (*gamma* band) during the initial 24–72 h of epileptogenesis (Milikovsky et al., 2017). Changes in these frequency bands may be a consequence of the progressive decrease of signal dimension during this time-window, as suggested by recurrence plot analysis (Webber and Zbilut, 2005; Marwan et al., 2007). High-frequency oscillations (80–600 Hz) (which we did not test due to sampling rate limitations) and spiking activity are additional EEG features of epileptogenesis under consideration as candidate biomarkers (White et al., 2010; Engel and da Silva, 2012). Although we do not know whether there is a relationship with the dimensional changes we report here, these EEG signals overall reflect neuronal network alterations, therefore their combination may significantly increase the predictive value of each individual measure alone.

Table 1
Correlation analysis between the EEG Est-ED and the rank of severity of epilepsy.

Experimental treatment	Progression Index	Rank of epilepsy severity	EEG Est-ED (median)	Spearman's correlation coefficient (EEG Est-ED vs. Rank)
Vehicle NAS	8.50	4	7	$r_s = -0.975$
NAS	3.00	3	11	$p = .017$
Vehicle AB	2.67	2	15	(one-tailed)
AB	2.33	1	19	
SHAM	*nd*	0	19	

Rank values are expressed using an ordinal arbitrary scale (see Supplementary Material for details). For sham group, rank = 0. The higher the rank, the more severe the epilepsy. NAS, N-Acetylcysteine + Sulforaphane (antioxidant drugs); AB, Anakinra + BoxA (antiinflammatory drugs); *nd*, not determined.

Acknowledgements

We are grateful to Prof. Charles Webber (Loyola University Chicago, Health Sciences Division, Chicago, USA) who generously provided the source codes of RQA applications. We wish to thank also Dr. Wiebke Theilmann (Department of Pharmacology, Toxicology and Pharmacy, University of Veterinary Medicine, Hannover, Germany) for her contribution to this study.

This work used the EGI Applications On Demand Service, which is co-funded by the EGI-Engage project (Horizon 2020) under Grant number 654142. We are grateful to Dr. Giuseppe La Rocca and Dr. Gergely Sipos (Customer and Technical Outreach Managers - The EGI Foundation, Science Park 140 - 1098 XG Amsterdam, The Netherlands) for their crucial technical support related to computational resources. Specifically, this work benefited from services and resources provided by the following Virtual Organizations: *fedcloud.egi.eu*, supported by the national resource providers of the EGI Federation; *biomed*, with the dedicated support of resource centers BEINJING-LCG2, IN2P3-IRES, OBSPM, INFN-FERRARA, GARR-01-DIR, INFN-CATANIA, INFN-ROMA3, INFN-BARI, CREATIS-INSA-LYON, NCG-INGRID-PT, INFN-PISA, CESNET-MetaCloud and CLOUFIN, resource centers in UK hosted by GridPP collaboration, and the additional support of the resource centers listed here: http://operations-portal.egi.eu/vapor/resources/GL2Browser?VOfilter=biomed.

Funding

This work was supported by European Union's Seventh Framework Programme (FP7/2007-2013) under grant agreement n. 602102 (EPITARGET to A.V., A.F., W.L.) and by ARCEM association (M.R.).

Declarations of interest

None.

Supplementary Material provides details on the implementation of RQA, including the choice of parameters and the procedure for estimating EEG/ECoG signals dimension variations during epileptogenesis. *Supplementary Material* include Suppl. Table 1, Suppl. Table 2, Suppl. Table 3, Suppl. Table 4 and Suppl. Fig.1.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.nbd.2018.12.014.

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