Clinical Neurophysiology 128 (2017) 106–114

Contents lists available at ScienceDirect

Clinical Neurophysiology

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

Frequency-independent characteristics of high-frequency oscillations in epileptic and non-epileptic regions

Martin Pail [a,*], Pavel Řehulka [a], Jan Cimbálník [b], Irena Doležalová [a], Jan Chrastina [c], Milan Brázdil [a,d]

[a] Brno Epilepsy Center, Department of Neurology, St. Anne's University Hospital and Faculty of Medicine, Masaryk University, Brno, Czech Republic
[b] International Clinical Research Center, St. Anne's University Hospital, Brno, Czech Republic
[c] Brno Epilepsy Center, Department of Neurosurgery, St. Anne's University Hospital and Faculty of Medicine, Masaryk University, Brno, Czech Republic
[d] Behavioral and Social Neuroscience Research Group, CEITEC – Central European Institute of Technology, Masaryk University, Brno, Czech Republic

ARTICLE INFO

Article history:
Accepted 15 October 2016
Available online 29 October 2016

Keywords:
High frequency oscillations
Ripples
Fast ripples
Temporal lobe epilepsy
Extratemporal lobe epilepsy
Seizure onset zone
Epileptogenic zone
Irritative zone

HIGHLIGHTS

- The highest rate of fast ripples is in the seizure onset zone (SOZ).
- A worse prognosis (no seizure freedom after surgery) is associated with higher amplitudes of fast ripples outside the SOZ.
- In the SOZ, high frequency oscillations are more frequent, shorter, and have higher amplitudes.

ABSTRACT

Objective: The purpose of the presented study is to determine whether there are frequency-independent high-frequency oscillation (HFO) parameters which may differ in epileptic and non-epileptic regions.
Methods: We studied 31 consecutive patients with medically intractable focal (temporal and extratemporal) epilepsies who were examined by either intracerebral or subdural electrodes. Automated detection was used to detect HFO. The characteristics (rate, amplitude, and duration) of HFO were statistically compared within three groups: the seizure onset zone (SOZ), the irritative zone (IZ), and areas outside the IZ and SOZ (nonSOZ/nonIZ).
Results: In all patients, fast ripples (FR) and ripples (R) were significantly more frequent and shorter in the SOZ than in the nonSOZ/nonIZ region. In the group of patients with favorable surgical outcomes, the relative amplitude of FR was higher in the SOZ than in the IZ and nonIZ/nonSOZ regions; in patients with poor outcomes, the results were reversed. The relative amplitude of R was significantly higher in the SOZ, with no difference between patients with poor and favorable surgical outcomes.
Conclusions: FR are more frequent, shorter, and have higher relative amplitudes in the SOZ area than in other regions. The study suggests a worse prognosis in patients with higher amplitudes of FR outside the SOZ.
Significance: Various HFO parameters, especially of FR, differ in epileptic and non-epileptic regions. The amplitude and duration may be as important as the frequency band and rate of HFO in marking the seizure onset region or the epileptogenic area and may provide additional information on epileptogenicity.

1. Introduction

Over the past few years, there has been growing interest in the analysis of interictal high frequency oscillations (HFO), primarily with the goal of understanding their value for identifying the epileptogenic zone and their correlation with epileptogenicity. HFO promise to be more specific than interictal spikes for epileptogenic brain tissue and even more specific than the seizure-onset area (Jacobs et al., 2008).

HFO are short-lasting field potentials, which arise as a result of the synchronization of neuronal populations. HFO have been identified and defined in terms of frequency: ripples (80–250 Hz), fast ripples (250–600 Hz) (reviewed in Bragin et al., 2010; Engel et al.,

* Corresponding author at: Department of Neurology, St. Anne's University Hospital and Faculty of Medicine, Pekařská 53, 656 91 Brno, Czech Republic. Fax: +420 543 182 624.
E-mail address: martin.pail@fnusa.cz (M. Pail).

http://dx.doi.org/10.1016/j.clinph.2016.10.011
1388-2457/

2009), and very high frequency oscillations (1000–2500 Hz) (Usui et al., 2015). HFO have been widely studied in animals and humans, in mesial temporal and neocortical structures, under physiological and pathological conditions, using microelectrodes or commercial macroelectrodes, and during interictal and ictal periods (Bragin et al., 1999a,b; Staba et al., 2002; Worrell et al., 2004, 2008; Urrestarazu et al., 2007; Jacobs et al., 2008, 2009; Bagshaw et al., 2009; Brázdil et al., 2010; Crépon et al., 2010). However, the reliability of HFO as a biomarker of epileptogenicity and the seizure-onset zone remains uncertain (Haegelen et al., 2013; Jobst, 2013; Wang et al., 2013).

Ripples, observed as a physiological finding in the hippocampus and parahippocampal structures, are thought to be functionally involved in memory consolidation (Buzsáki et al., 1992; Axmacher et al., 2010; Lachaux et al., 2012). The spontaneous occurrence of ripples in humans is also believed to be physiological in the primary visual cortex (Nagasawa et al., 2012; Wang et al., 2013) and in the primary motor cortex (Wang et al., 2013). The presumption of the exclusively physiological nature of ripples was, however, impugned by evidence of HFO in ripple ranges recorded in the dentate gyrus after epileptogenic insult in an animal model of kainate-induced status epilepticus (Bragin et al., 1999b, 2004).

Conversely, fast ripples were repeatedly reported as a biomarker of epileptogenesis and epileptogenicity, both in animal models and in human epilepsy (Bragin et al., 1999a,b; Staba et al., 2002). It is important that HFO in the fast ripple range (at about 600 Hz) can also be considered physiological, as they were previously evoked during stimulation of the somatosensory cortex (Curio et al., 1997). Thus the HFO frequency range, in general, is not sufficient to differentiate physiological and pathological HFO (Bragin, 2007). On the other hand, there is evidence of favorable epilepsy surgery outcome after the removal of tissue generating interictal HFO, especially fast ripples (Jacobs et al., 2010; Wu et al., 2010; Akiyama et al., 2011).

Several authors have tried to distinguish between pathological and physiological HFO on the basis of a specific regional distribution in respective mesial temporal structures (Jiruska and Bragin, 2011); some have investigated the difference between task-induced and spontaneous HFO (Nagasawa et al., 2012; Matsumoto et al., 2013; Brázdil et al., 2015); others have studied the association of HFO with epileptiform discharges (Crépon et al., 2010; Urrestarazu et al., 2007; Wang et al., 2013). Interictal HFO (both ripples and fast ripples) rates were proven significantly higher within the seizure onset zone (SOZ) than outside it (Jacobs et al., 2009).

The purpose of the present study is to identify whether there are any other frequency-independent HFO parameters that potentially differ in areas within the SOZ, within the irritative zone (IZ), and in areas outside the IZ/SOZ.

2. Methods

2.1. Subjects

Our sample comprised 31 patients (19 females; 12 males) ranging in age from 13 to 56 years (mean age 33.4 years, SD = 10.5), all with medically intractable focal epilepsies (Table 1). All the subjects fulfilled the diagnostic criteria for either medically intractable temporal lobe epilepsy (TLE) – 22 subjects or extratemporal lobe epilepsy (ETLE) – 9 subjects. The diagnosis was made according the ILAE criteria (Commission on Classification and Terminology of ILAE, 1989). The main demographic and clinical characteristics of the included subjects are shown in Table 1.

2.1.1. Presurgical evaluation

All 31 patients underwent a comprehensive presurgical evaluation, including a detailed history and neurological examination, magnetic resonance imaging (MRI), neuropsychological testing, and scalp and invasive video-EEG monitoring (Table 1). Most of the subjects had not previously undergone intracranial surgery. One subject underwent resection of venous malformation within the left P-O region before SEEG and re-operation; in one subject a resection of the pole of the left temporal lobe had been performed, and in one subject a limited left AMTR was performed before SEEG monitoring. Prior to invasive EEG, two subjects had a vagus nerve stimulation system implanted, with unfavorable seizure frequency outcome. The duration of clinical monitoring and the location and number of implanted electrodes were determined in accordance with clinical considerations.

2.1.2. Surgery and outcome measure

Most of the patients (28) underwent surgical intervention (implantation of VNS was performed in 7 patients and brain resection in 21 patients; details are shown in Table 1). The follow-up interval after epilepsy surgery was at least 12 months. After surgical resection, 8 patients were rated as Engel IA, one patient was Engel IIA, and 11 patients were Engel III or IV; the Engel rating is unknown for one patient (due the loss to follow-up care).

This study was approved by the St. Anne's University Hospital Research Ethics Committee and the Ethics Committee of Masaryk University. All patients signed an informed consent form.

2.2. SEEG

Depth electrodes (mostly SEEG; grids and strips were used in two patients) were implanted to localize the seizure origin prior to surgical treatment. The sites of electrode placements were individualized according to seizure semiology, clinical history, noninvasive EEG investigations, and neuroimaging results. Standard intracerebral 5-, 10-, and 15-contact Micro Deep semi-flexible multicontact platinum electrodes (ALCIS) were used with a diameter of 0.8 mm, a contact length of 2 mm, an inter-contact distance of 1.5 mm, and a contact surface area of 5 mm^2. Their position within the brain was afterwards verified by MRI with electrodes in situ (see Table 1). In two patients, platinum subdural strip and grid electrodes (Radionics) were used. Thirty minutes of artifact-free continuous interictal SEEG data (recorded during wakefulness) was analyzed for each subject. This period is sufficient based on the results of previously published papers (Jacobs et al., 2008; Zelmann et al., 2009; Andrade-Valença et al., 2012). The EEGs were acquired at 25 kHz sampling frequency and subsequently low-pass filtered and downsampled to 5 kHz. High harmonics produced by the system (artificial harmonics) are accounted for during EEG acquisition. A reference average montage was used for the analysis.

2.2.1. Labeling of analyzed contacts

The contacts in each subject were divided into three groups for further HFO analysis. The distribution was done visually (by co-authors M.P. and I.D.) by a standard analysis of ictal and interictal SEEG recordings. The first group was labelled SOZ contacts: the channels that revealed the first ictal activity. The second group was labelled IZ contacts: the channels where interictal epileptiform discharges were detected, but no seizure onset was detected. The remaining non-spiking contacts were labelled nonSOZ/nonIZ. Only contacts localized in gray matter were included in the study.

2.2.2. Automated detection of HFO in resting awake SEEG

The algorithm for HFO detection is explained in more detail in the Supplementum. HFO were detected by a custom Matlab detection algorithm. The raw signal (Supplementary Fig. S1) was divided

Table 1
Demographic and clinical data of the patients.

Subject gender	Age at seizure onset	Age at invasive EEG	Seizure frequency/monthly	Febrile seizures/precipitating events	MRI (signs of)	Brain lobes explored and number of implanted electrodes	Region SOZ (epileptogenic zone)	Intervention/histopathology	Postsurgical outcomes (Engel) (years)
1 (F)	16	20	CPS/4plus	0	Normal	T (3), T′(2)	H bilaterally	VNS	–
2 (F)	19	57	CPS/2plus, sGTCS/6	0	Postischemic lesions within left T-O and H	F′(1), T′(5)	Left H	AMTR/gliosis, hemosiderin within T pole	IA (3.5)
3 (F)	4	34	CPS/50	0	Hypotrophic right H	T′(3), T(3)	Right H	AMTR/hippocampal sclerosis grade III	IA (3.5)
4 (F)	6	19	CPS/4	Perinatal hypoxia	Gliosis and cortical abnormalities left PO	O′(2), T′(2), P′(1), O(2), F(2)	Left TPO region	Cortectomy/negat	IV (3)
5 (F)	2	34	SPS,CPS/4	Perinatal asphyxia	Polymicrogyria left F, postoperative changes of left PO	F′ (8), P′(2), T′(1)	Left P lobe, pericentral area	VNS	–
6 (F)	16	33	CPS/5	Perinatal hypoxia	Bilat. HS	T′(4), T(4)	H bilaterally (mainly right side)	Right AMTR/negat	III (2.5)
7 (M)	0.5	21	CPS/30, MS/300	0	Abnormality of left H	F′(9), P′(1), T′(1), F(2)	Left SMA (myoclonic seizures), left F pole (CPS)	Resection of left F pole/negat	IV (2.5)
8 (F)	9.5	13	CPS/30	Perinatal asphyxia	Malrotation of left H	P′(5), T′(5), F′(1), O(1), P(1)	Undetected	–	–
9 (M)	9	27	CPS/30 plus	Perinatal asphyxia	Bilat. abnormal gyrification and gliotic changes of PO	O (2), T (3), P(3), O′(3), T′(1), P′(3)	O lobe bilaterally (mainly right side)	Partial resection of right O lobe/ ulegyria, FCD IIIA	IIIA (2)
10 (F)	9	30	CPS/3	Perinatal asphyxia	Normal	T (3), T′(8), O′(2)	Left lateral T lobe, GTS	Incomplete resection of left GTS/ negat	IVA (2)
11 (F)	17	26	CPS/12	Meningoencephalitis	Normal	T (1), T′(8)	Lateral mesial T lobe	Left AMTR/FCD IB	IA (2)
12 (F)	28	56	CPS/8	0	Right hippocampal sclerosis	T (2), T′(3)	Right H	Right AMTR/negat	IIIA (2)
13 (M)	1	40	CPS/2 plus	0	Hypotrophic left H	T′(6), P′(2), O′(2)	Left H	Left AMTR/negat	IA (2)
14 (F)	11	27	sGTCS/3	0	Normal	F(4), F′(8)	Left GFS, GFM, GFMed	Partial cortectomy of left F lobe	IIIA (2)
15 (M)	19	26	CPS/4	0	FCD left T and hyperintensity of H	T′(7), F′(1), T (1)	Left anterotemporal, right mesial temporal	VNS	–
16 (F)	10	34	SPS, CPS/10 plus	Perinatal asphyxia	Normal	F′(5), P′(2), F(6), P(2)	Right dorsolateral parasagittal premotoric area	Partial cortectomy of right F lobe, partial callosotomy/FCD 1C	Not available
17 (M)	27	38	CPS/25 plus	0	Normal	T(3), I′(2), T′(6), P′(1), O′(1), F′(1)	Mesial T region bilaterally	VNS	–
18 (F)	6	48	CPS/60 plus	Febrile seizures	Postoperative gliosis right P	P(6), T(3), O(1)	Right TP cortex, right hippocampus	VNS	–
19 (M)	33	41	CPS/30	0	Focal hyperintensity right basal T	T(4), T′(3)	Right H	Right AMTR/FCD IIIb gangliogliom	IA(1.5)
20 (M)	11	25	CPS/12 plus	0	Normal	TPO grids	Right lateral T lobe	Cortical resection of right T a TO region/negat	IIA(2)
21 (F)	17	40	CPS/20	0	Normal	F′(1),O′(2),T′(7),T(2)	Undetected	–	–
22 (F)	17	40	CPS/20	0	Normal	FTPO′ grids	Left laterobasal posterior T lobe	Cortectomy of left laterobasal posterior T lobe/negat	IIIA (1.5)
23 (M)	8	29	CPS/20 plus	0	Normal	T′(7), F′(2), T(2)	Bitemporal	Left AMTR/negat	IVA (1)
24 (F)	2	33	CPS/6 plus	Meningoencephalitis	Postencephalitic changes left T	T(2), T′(6), F′(3)	Left H	Left AMTR/hippocampal sclerosis, postmeningoencephalitic changes	IA (1)
25 (M)	12	40	CPS/40 plus	Trauma	Posttraumatic changes of left T and GFI	F′(6), T′(5)	Left T pole, lateral T, lateral prefrontal area	Resection of temporal pole, lesionectomy F left/posttraumatic changes	IIIA (1)
26 (F)	2	33	sGTCS/10	0	Postoperative changes right PO	T(3), P(2), O(2)	Right GTS, LPI	–	–
27 (F)	2	22	CPS/4	0	Postoperative changes right PO	P(2), F(2), T(3)	Undetected (right posterior quadrant)	VNS	–

Table 1 (*continued*)

Subject gender	Age at seizure onset	Age at invasive EEG	Seizure frequency/monthly	Febrile seizures/precipitating events	MRI (signs of)	Brain lobes explored and number of implanted electrodes	Region SOZ (epileptogenic zone)	Intervention/histopathology	Postsurgical outcomes (Engel) (years)
28 (M)	21	35	CPS/5	Commotio cerebri	Bilat. hypotrophic hippocampi	T(6), T′(2)	H bilaterally (mainly right side)	VNS	–
29 (M)	31	37	CPS/4	Prolonged febrile seizures	Normal	T′(8), F′(2), T(2)	H bilaterally (mainly left side)	Left AMTR/negat	IA (1)
30 (F)	9	27	CPS/5	Meningoencephalitis	Left HS	T′(8), T(2), P′(1), I′(1), F′(1)	Left H	Left AMTR/FCDIIIA	IIIA (1)
31 (M)	2	51	CPS/3 plus	Febrile seizures	Right H atrophy, slight changes of density	T(5), F(3), T′(2)	Right H	Right AMTR/negat	IA (1)

CPS – complex partial seizure; MS – myoclonic seizures; plus – sporadic ictal generalization; SOZ – seizure onset zone; SMA – supplementary motor area; LPI – lobulus parietalis inferior; GTS – gyrus temporalis superior; GFS – gyrus frontalis superior; GFM – gyrus frontalis medius; GFMed – gyrus frontalis medius; H – hippocampus; HS – hippocampal sclerosis; DNET – dysembryoplastic neuroepithelial tumor; FCD – focal cortical dysplasia; AMTR – anteromedial temporal resection; E – extratemporal; T – temporal; R – right; L – left; FCD – focal cortical dysplasia; F – frontal; P – parietal; T – temporal; O – occipital, I – insular; ′ - left.

into sliding statistical windows (10 s) and filtered in a series of overlapping, logarithmically spaced frequency bands. Each band was processed as follows:

The power envelope was computed using the Hilbert transform and normalized by Eq. (1) to stress the signal peaks (Supplementary Fig. S2).

$$x_{normed} = \frac{x - P_{2/3}}{P_{1/3} - P_{2/3}} \quad (1)$$

To overcome the effects of Gibb's phenomenon, a "frequency stability" metric was computed. The band passed filtered signal (narrow band) was transformed to a cosine representation of its phase. The same transformation was applied to a band passed filtered signal with the same high cut-off frequency but a four times lower low cut-off frequency (broad band) (Supplementary Fig. S3a, b). A sliding window with the width of four cycles of the narrow band low cut-off frequency was applied to the narrow band signal and the root mean square (RMS) was calculated, thus generating the "signal". Similarly, the RMS was calculated on the signal created by subtracting the narrow band signal from the broad band signal, generating "noise". The frequency stability was calculated as a "signal-to-noise" ratio. The produced signal was normalized Eq. (1).

The dot product of the amplitude and frequency stability metric was calculated. Negative values were set to 0. Putative HFO detections were obtained by thresholding the normalized Eq. (1) dot product of the power envelopes and frequency stability with 0.1 (Supplementary Fig. S4).

To increase algorithm specificity, amplitude, and frequency stability, the dot product and duration minimum/maximum thresholds were applied on putative HFO detections. The metric threshold values were obtained from cumulative distribution functions fitted to the HFO metric distributions previously marked by expert reviewers. The parameters of the fitted gamma functions were specific for each band. The parameters (Supplementary Table S1) and examples of true positive detection and false positive detection (Supplementary Fig. S5) are included in the Supplementum.

2.3. Statistical analysis

The rates of HFO per contacts within the three groups (SOZ, IZ, and nonSOZ/nonIZ) were statistically compared (SOZ × IZ; SOZ × nonSOZ/nonIZ; IZ × nonSOZ/nonIZ). This statistical analysis was performed using an independent two sample *t*-test separately for ripple range and fast ripple range. We performed statistical analyses on the whole dataset and separately for patients with favorable postoperative outcomes (Engel I or II – 9 patients) and the other patients with "poor outcomes" (postsurgical outcomes of Engel III or IV and patients with presumed poor outcomes due to more than one detected SOZ according to SEEG). Furthermore, we performed a statistical analysis comparing the duration and relative amplitude of HFO (separately for R and FR ranges) for contacts in the areas, as defined above. This analysis was performed in all patients and subsequently in patients with favorable or poor surgical outcomes. For this analysis, we again used an independent two sample *t*-test. Bonferroni's correction for multiple comparisons was applied where needed.

3. Results

The statistical analysis and complete results of HFO detection (rates, duration, and amplitudes) are shown in Table 2 and Supplementary Table S2, respectively.

3.1. Rates

As expected, the HFO rate per contact in all patients was statistically higher in the SOZ than in the nonSOZ/nonIZ regions (Fig. 1) in the fast ripple frequency range ($p = 0.018$) and ripple range ($p = 0.038$).

Specifically, HFO in the ripple range were identified. The mean number of HFO per contact (per 30 min) was 109.13 within the SOZ (SD = 79.19), 96.03 in the IZ (SD = 82.24) and 68.65 for nonSOZ/nonIZ regions (SD = 49.24). The differences between groups of contacts were significant only between SOZ and nonSOZ/nonIZ regions. In the fast ripple range, the mean HFO count per contact (per 30 min) were 50.02 in the SOZ (SD = 50.75), 31.56 in the IZ (SD = 34.89) and 23.23 in nonSOZ/nonIZ (SD = 21.84). The only significant result was in the comparison of SOZ and nonSOZ/nonIZ regions; see above.

No statistically significant results were seen in comparison of regions separately in groups of patients with favorable or poor outcomes.

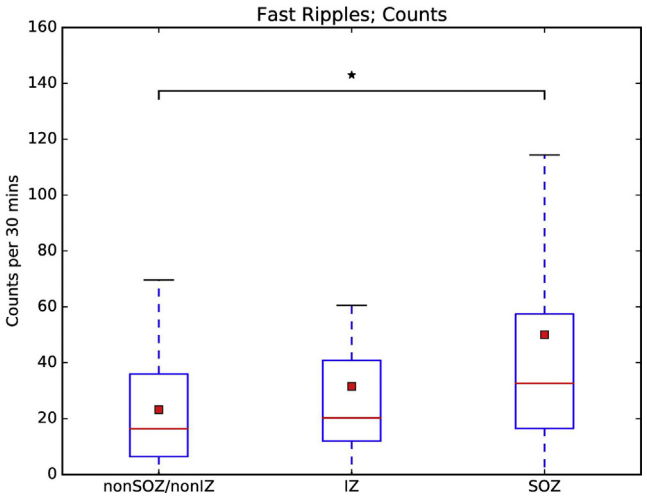

Fig. 1. Counts of fast ripples per 30 min in particular areas (SOZ, IZ, and nonSOZ/nonIZ) in all patients.

3.2. Duration

3.2.1. Ripples

The mean duration of detected HFO was 54.40 ms in the SOZ (SD = 25.58), 56.31 ms in the IZ (SD = 27.52) and 56.12 ms in nonSOZ/nonIZ regions (SD = 29.16). Statistical analysis showed significantly shorter durations of HFO in the SOZ than in either the IZ or nonSOZ/nonIZ regions ($p < 0.001$); the difference between the IZ and nonSOZ/nonIZ regions was not significant ($p = 0.361$).

3.2.2. Fast ripples

Similarly, automated detection in the fast ripple range detected the mean durations of HFO of 27.43 ms in the SOZ (SD = 16.04), 29.13 ms in the IZ (SD = 18.30) and 30.41 ms, in nonSOZ/nonIZ regions (SD = 20.94); the shortest durations of FR were in the

Table 2
The results of statistical analysis (two sample *t*-test, *p* values) comparing the rates, duration, and relative amplitudes of HFO (all over separately for ripples [first column] and fast ripples range [second column]) per contacts in the redefined areas (SOZ, IZ, nonSOZ/nonIZ); also in this analysis particularly in all patients and in patients with favorable or poor outcomes.

Rates

	nonSOZ/nonIZ						IZ					
	All		Favorable outcome		Poor outcome		All		Favorable outcome		Poor outcome	
SOZ	0.038	0.018	0.417	0.143	0.102	0.076	1.078	0.228	0.102	0.092	0.974	0.407
IZ	0.245	0.543	1.243	1.136	0.464	0.491	x	x	x	x	x	x
nonSOZ/nonIZ	x	x	x	x	x	x						

Duration

	nonSOZ/nonIZ						IZ					
	All		Favorable outcome		Poor outcome		All		Favorable outcome		Poor outcome	
SOZ	<<0.001	<<0.001	<<0.001	<<0.001	0.877	<<0.001	<<0.001	<<0.001	<<0.001	<0.001	<<0.001	<<0.001
IZ	0.361	<<0.001	<<0.001	0.460	<<0.001	0.027	x	x	x	x	x	x
nonSOZ/nonIZ	x	x	x	x	x	x						

Amplitude

	nonSOZ/nonIZ						IZ					
	All		Favorable outcome		Poor outcome		All		Favorable outcome		Poor outcome	
SOZ	<<0.001	0.430	<<0.001	<<0.001	<<0.001	<<0.001	<<0.001	0.309	<<0.001	0.014	0.030	<<0.001
IZ	<<0.001	0.013	<<0.001	<<0.001	<<0.001	1.136	x	x	x	x	x	x
nonSOZ/nonIZ	x	x	x	x	x	x						

SOZ, HFO durations were longer in nonSOZ/nonIZ regions than in the IZ ($p < 0.001$) and in nonSOZ/nonIZ regions than in the SOZ ($p < 0.001$). The difference between the IZ and SOZ was also significant ($p < 0.001$).

In the group of patients with favorable outcomes, shorter R and FR durations were seen in the SOZ ($p < 0.001$) than in nonSOZ/nonIZ and IZ regions (see Fig. 2).

In the group of patients with poor outcomes, the longest duration of R was in the IZ ($p < 0.001$); the difference between nonSOZ/nonIZ × SOZ was not significant ($p = 0.877$). In both subgroups of patients, the shortest FR duration was seen in the SOZ ($p < 0.001$).

3.3. Amplitudes

3.3.1. Ripples

Automated detection in the ripple range detected the relative amplitudes of HFO in the SOZ of 77.47 (SD = 164.35), in the IZ of 66.14 (SD = 231.74) and in nonSOZ/nonIZ regions of 56.30 (SD = 151.74). Statistical analysis showed significantly higher amplitudes of HFO in the SOZ than in either the IZ or nonSOZ/nonIZ regions ($p < 0.001$); the difference between the IZ and nonSOZ/nonIZ regions was also significant ($p < 0.001$).

3.3.2. Fast ripples

Automated detection in the fast ripple range detected relative amplitudes of HFO in the SOZ of 76.04 (SD = 164.95), in the IZ of 80.93 (SD = 458.49) and in nonSOZ/nonIZ regions of 73.79 (SD = 241.17). The differences between SOZ and IZ or nonSOZ/nonIZ were not significant.

Nevertheless, in the group of patients with favorable outcomes, the relative amplitude of HFO (both R and FR) was higher in the SOZ than in the IZ and nonIZ/nonSOZ region ($p < 0.001$) (see Figs. 3 and 4). In the group of patients with poor outcomes, the highest amplitude of R was seen in the SOZ ($p < 0.001$) (versus nonSOZ/IZ); $p = 0.030$ (IZ) and the lowest in nonIZ/nonSOZ region ($p < 0.001$). The relative amplitude of FR was lower in the SOZ than in either the IZ or nonSOZ/nonIZ regions ($p < 0.001$) (see Fig. 5).

4. Discussion

Interictal HFO analyses in patients with epilepsy have been reported useful for SOZ identification (Urrestarazu et al., 2007;

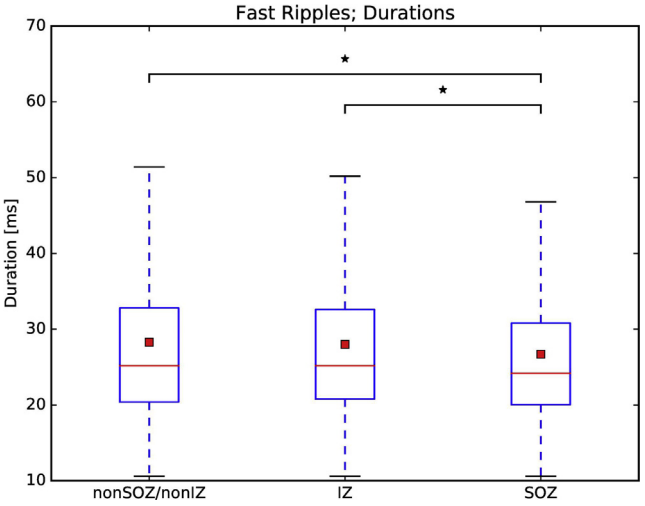

Fig. 2. Durations (ms) of fast ripples in particular areas (SOZ, IZ, and nonSOZ/nonIZ) in patients with favorable outcomes.

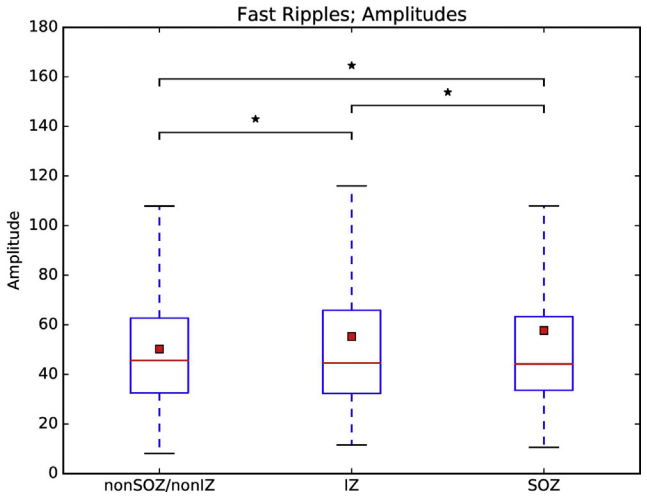

Fig. 3. Relative amplitudes of fast ripples in particular areas (SOZ, IZ, and nonSOZ/nonIZ) in patients with favorable outcomes.

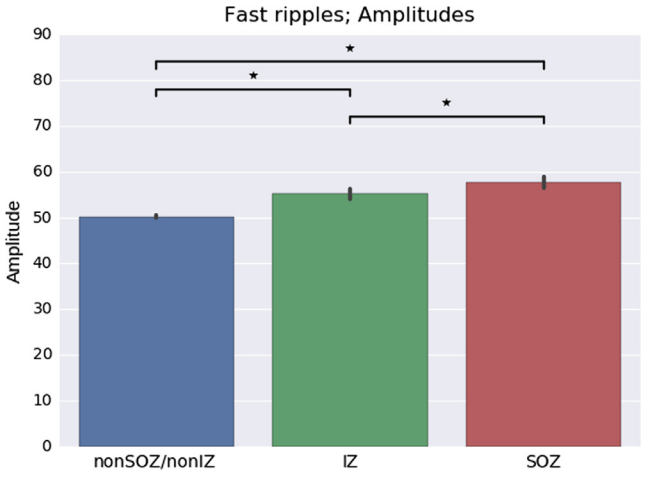

Fig. 4. Relative amplitudes of fast ripples in particular areas (SOZ, IZ, and nonSOZ/nonIZ) in patients with favorable outcomes. Bar graphs represent the population mean. Ticks represent a 95% confidence interval of the mean calculation.

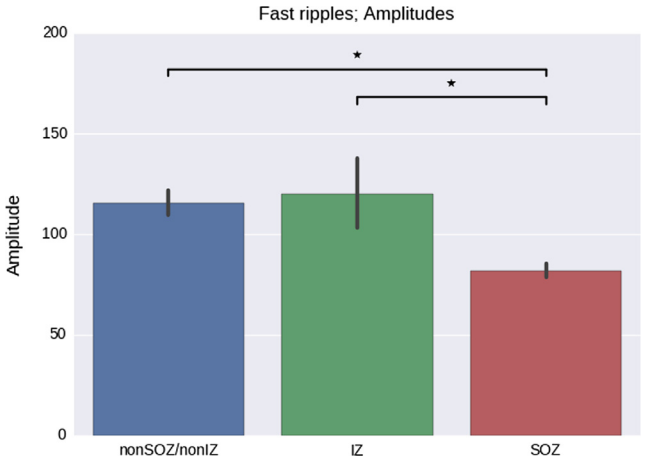

Fig. 5. Relative amplitudes of fast ripples in particular areas (SOZ, IZ, and nonSOZ/nonIZ) in patients with poor outcomes. Bar graphs represent the population mean. Ticks represent a 95% confidence interval of the mean calculation.

Jacobs et al., 2008, 2009). In the present study, patients with both temporal and extratemporal epilepsy were examined to ascertain facts about HFO characteristics in areas within and outside the SOZ and the IZ. For these purposes, we used the concept of an epileptogenic zone (Lüders and Awad, 1992; Rosenow and Lüders, 2001), and used the term "irritative zone" as it has been used elsewhere (Blanco et al., 2011) to investigate HFO characteristics in more detail, to observe the area between the SOZ and non-SOZ, and to eliminate potential overlap of interictal active ("spiking") channels within SOZ and non-SOZ. Only patients who underwent surgical resections and had good postsurgical outcomes reflect the correct determination of the seizure onset zone. For that reason, we divided patients into two subgroups: favorable outcome and others ("poor outcome").

In our study, HFO were detected in all (SOZ, IZ, and nonSOZ/nonIZ) areas, with a higher absolute HFO rate for events in the R range than in the FR range. These findings are not surprising; the occurrence of high frequency activity unassociated with the SOZ is well documented in the hippocampi (Staba et al., 2002; Axmacher et al., 2010) and in the primary motor, somatosensory, and visual cortices (Curio, 2000; Nagasawa et al., 2012; Matsumoto et al., 2013; Wang et al., 2013). The results indicate that the detection algorithm, in addition to detecting pathological HFO, may also detect HFO that might be physiological, might be a fragment or propagation of pathological HFO arising from elsewhere (Crépon et al., 2010), or might be false positive detections. Higher HFO rates in the R range than in the FR range may be explained by an association between HFO and interictal spiking (Urrestarazu et al., 2007; Jacobs et al., 2008; Wang et al., 2013). There is evidence that larger networks are involved in R than in FR generation in SOZ areas (Chrobak and Buzsáki, 1996; Bragin et al., 2002) and that FR generators are spatially localized to a region of less than 1 mm^3 (Bragin et al., 2002). Furthermore there is evidence of HFO under detection, especially in higher frequency bands when using macroelectrodes (as in our cases), i.e. in the FR range, which results in a lower mean frequency of detected HFO (Worrell et al., 2008).

According to previously published data, which is further corroborated by our study, the FR and R rate are significantly higher in the SOZ than in nonSOZ/nonIZ regions (Urrestarazu et al., 2007; Jacobs et al., 2008, 2009; Andrade-Valença et al., 2012). Interestingly, in patients with favorable outcomes, a prominent (though not significant) difference between rates of FR and R in the SOZ and the IZ can be observed, which might represent a more focal generator (epileptogenic tissue) of these pathological HFO; this difference was less seen in other patients. As in a study by Jacobs et al. (2008), the distinction between pathological and normal areas was worse for R than for FR. Nevertheless, the explanation for the high rates of R and FR in nonSOZ/nonIZ regions is unclear. It cannot be definitely demonstrated whether all of these marked events were actually pathological, physiological, or the propagation of pathological HFO arising from elsewhere, as was mentioned above. Spontaneous HFO of a physiological nature are difficult to distinguish from epileptogenic ones, particularly during wakefulness (Jacobs et al., 2008; Curio, 2000) since the frequency and amplitude measures alone cannot be used for this purpose (Engel et al., 2009; Nagasawa et al., 2012; Matsumoto et al., 2013). Finally, the overlap (in both amplitude and duration) between the pathological and physiological ripples is extensive (Alkawadri et al., 2014).

The main reason for focusing on the IZ is evidence of HFO linking with interictal epileptiform discharges in both mesiotemporal and neocortical epilepsy (Urrestarazu et al., 2007; Jacobs et al., 2008; Wang et al., 2013). The vast majority of interictal HFO (up to 73% of R and 92% of FR) is associated with interictal spikes or sharp waves (Urrestarazu et al., 2007; Jacobs et al., 2008). A comparison of HFO associated with interictal epileptiform discharges and unassociated HFO revealed no differences in terms of the duration (Urrestarazu et al., 2007), or the associated HFO had longer durations than the unassociated HFO with spikes (Jacobs et al., 2008). Wang and colleagues (2013) showed HFO detected in the SOZ area were of shorter duration than those not correlated to the SOZ area. As in the study by Wang et al. (2013), our data suggest longer durations of both R and FR in the IZ or nonSOZ/nonIZ than in the SOZ. These findings might be also a consequence of analyzing awake recordings with less expressed interictal discharges, and so more HFO detected this phenomenon and with shorter duration. This finding might be also explained by the work of Nagasawa (2012), who revealed that the duration of spontaneous HFO in the ripple range (namely from the occipital cortex) of a physiological nature were significantly longer than that of epileptogenic ripple HFO. Similar results were presented by Alkawadri et al. (2014). These observations are still consistent with the hypothesis that longer durations of HFO may represent longer excitatory neural processing (Niessing et al., 2005; Nishida et al., 2008; Koch et al., 2009; Manning et al., 2009; Nagasawa et al., 2012). In other studies, however, the duration of pathological (in the SOZ) and physiological (nonSOZ region) FR was not diverse (Nagasawa et al., 2012; Alkawadri et al., 2014), or the longer duration of both R (Brázdil et al., 2015) and FR was revealed within the SOZ (Jacobs et al., 2008; Matsumoto et al., 2013). The discrepancies among studies in the duration of R and FR in various regions might be explained by different proportions of included focal epilepsies (hippocampus vs. neocortex).

There have been noteworthy results regarding relative HFO amplitude in epileptic and non-epileptic regions in particular subgroups of patients. Interestingly, in the group of patients with favorable outcomes, the relative HFO amplitude (especially FR) was higher in the SOZ than in other regions; in other patients with "poor outcomes", the results of FR analysis were reversed. These results for both R and FR might contribute to neurosurgical resection planning, showing possible worse prognosis in patients with higher amplitudes especially of FR outside the SOZ. This indicates that it is possible that the true SOZ was not adequately detected. Another reason may be a more dispersed or multifocal epileptogenic zone/SOZ, a pattern which is too diffuse to permit a successful resective strategy, and so usually results in VNS implantation. Yet another reason for poor outcome may be that the result of the epileptogenic zone may include the actual epileptogenic zone (generating seizures before surgery) as well as a potential epileptogenic zone which is an area of the cortex that may generate seizures after the presurgical SOZ has been resected (Rosenow and Lüders, 2001). Seizures originating from areas not covered by electrodes but propagating to the actual electrode positions might lead to misinterpretation (Zijlmans et al., 2012).

HFO are more stable and more expressed, and the likelihood for artifact contamination is lower, during sleep. However, a review of recently presented data indicates that the effect of sleep on HFO expression differs among regions (Dümpelmann et al., 2015) and so this phenomenon might influence the observation. Some patients also experience postoperative nausea and general discomfort so they rarely reach deep stages of sleep. The differences in HFO rates between the SOZ and other remote areas were disclosed in wakefulness periods as significant (Bagshaw et al., 2009). Based on this data we decided to analyze awake recordings.

Our findings emphasize the importance of the careful interpretation of HFO, especially in cases with extensive spatial sampling or when there is an overlap between the epileptic and physiologic areas (Alkawadri et al., 2014). Based on our data, it is useful to include both ripples and fast ripples in the evaluation of the potential epileptogenic region (Zijlmans et al., 2012). Currently, there are no established criteria for distinguishing physiological from pathological HFO and SOZ areas (Engel et al., 2009; Jacobs et al.,

2016). Nevertheless, it seems that FR generated in the SOZ are more frequent, shorter, and have higher relative amplitudes than in other regions. There is, however, no clear cut-off value for these characteristics which can separate the SOZ, the IZ, and other regions.

5. Conclusion

HFO parameters (rate, amplitude, and duration) differ in epileptic and non-epileptic regions. We suggest that amplitude and duration may be as important as frequency band and rate of HFO in marking the seizure onset region or epileptogenic area and may provide additional information on epileptogenicity. To conclude, FR are more frequent, shorter, and have higher relative amplitudes in the SOZ area than in other regions. The study suggests a possible worse prognosis in patients with higher amplitudes of FR outside the SOZ.

Acknowledgments

The study was supported by the "CEITEC – Central European Institute of Technology" project (CZ.1.05/1.1.00/02.0068) from the European Regional Development Fund and by MŠMT ČR Research Program no. MSM0021622404. The technical part of the study was supported by the GACR project P103/11/0933 and the Application Laboratories of Advanced Microtechnologies and Nanotechnologies (CZ.1.05/2.1.00/01.0017), co-funded by the Operational Programme "Research and Development for Innovations", the European Regional Development Fund, and the state budget.

Supported by European Regional Development Fund - Project FNUSA-ICRC (No. CZ.1.05/1.1.00/02.0123) and Ministry of Education, Youth and Sports of the Czech Republic project no. LH15047 (KONTAKT II).

Conflict of interest: None of the authors has any conflict of interest to disclose.

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.clinph.2016.10.011.

References

Akiyama T, McCoy B, Go CY, Ochi A, Elliott IM, Akiyama M, et al. Focal resection of fast ripples on extraoperative intracranial EEG improves seizure outcome in pediatric epilepsy. Epilepsia 2011;52:1802–11.

Alkawadri R, Gaspard N, Goncharova II, Spencer DD, Gerrard JL, Zaveri H, et al. The spatial and signal characteristics of physiologic high frequency oscillations. Epilepsia 2014;55:1986–95.

Andrade-Valença L, Mari F, Jacobs J, Zijlmans M, Olivier A, Gotman J, et al. Interictal high frequency oscillations (HFOs) in patients with focal epilepsy and normal MRI. Clin Neurophysiol 2012;123:100–5.

Axmacher N, Cohen MX, Fell J, Haupt S, Dümpelmann M, Elger CE, et al. Intracranial EEG correlates of expectancy and memory formation in the human hippocampus and nucleus accumbens. Neuron 2010;65:541–9.

Bagshaw AP, Jacobs J, LeVan P, Dubeau F, Gotman J. Effect of sleep stage on interictal high-frequency oscillations recorded from depth macroelectrodes in patients with focal epilepsy. Epilepsia 2009;50:617–28.

Blanco JA, Stead M, Krieger A, Stacey W, Maus D, Marsh E, et al. Data mining neocortical high-frequency oscillations in epilepsy and controls. Brain 2011;134:2948–59.

Bragin A, Wilson CL, Engel Jr J. Voltage depth profiles of high-frequency oscillations after kainic acid-induced status epilepticus. Epilepsia 2007;48(Suppl 5):35–40. Erratum in: Epilepsia 2007; 48:2379.

Bragin A, Engel Jr J, Wilson CL, Fried I, Buzsáki G. High-frequency oscillations in human brain. Hippocampus 1999;9:137–42.

Bragin A, Engel Jr J, Wilson CL, Fried I, Mathern GW. Hippocampal and entorhinal cortex high-frequency oscillations (100–500 Hz) in human epileptic brain and in kainic acid-treated rats with chronic seizures. Epilepsia 1999;40:127–37.

Bragin A, Mody I, Wilson CL, Engel Jr J. Local generation of fast ripples in epileptic brain. J Neurosci 2002;22:2012–21.

Bragin A, Wilson CL, Almajano J, Mody I, Engel Jr J. High-frequency oscillations after status epilepticus: epileptogenesis and seizure genesis. Epilepsia 2004;45:1017–23.

Bragin A, Engel Jr J, Staba RJ. High-frequency oscillations in epileptic brain. Curr Opin Neurol 2010;23:151–6.

Brázdil M, Halámek J, Jurák P, Daniel P, Kuba R, Chrastina J, et al. Interictal high-frequency oscillations indicate seizure onset zone in patients with focal cortical dysplasia. Epilepsy Res 2010;90:28–32.

Brázdil M, Cimbálník J, Roman R, Shaw DJ, Stead MM, Daniel P, et al. Impact of cognitive stimulation on ripples within human epileptic and non-epileptic hippocampus. BMC Neurosci 2015;16:47.

Buzsáki G, Horváth Z, Urioste R, Hetke J, Wise K. High frequency network oscillation in the hippocampus. Science 1992;256(5059):1025–7.

Chrobak JJ, Buzsáki G. High-frequency oscillations in the output networks of the hippocampal–entorhinal axis of the freely behaving rat. J Neurosci 1996;16:3056–66.

Crépon B, Navarro V, Hasboun D, Clemenceau S, Martinerie J, Baulac M, et al. Mapping interictal oscillations greater than 200 Hz recorded with intracranial macroelectrodes in human epilepsy. Brain 2010;133:33–45.

Curio G. Linking 600 Hz "spikelike" EEG/MEG wavelets ("ς-bursts") to cellular substrates: concepts and caveats. J Clin Neurophysiol 2000;17:377–96.

Curio G, Mackert BM, Burghoff M, Neumann J, Nolte G, Scherg M, et al. Somatotopic source arrangement of 600 Hz oscillatory magnetic fields at the human primary somatosensory hand cortex. Neurosci Lett 1997;234:131–4.

Dümpelmann M, Jacobs J, Schulze-Bonhage A. Temporal and spatial characteristics of high frequency oscillations as a new biomarker in epilepsy. Epilepsia 2015;56:197–206.

Engel Jr J, Bragin A, Staba R, Mody I. High-frequency oscillations: what is normal and what is not? Epilepsia 2009;50:598–604.

Haegelen C, Perucca P, Châtillon C-E, Andrade-Valença L, Zelmann R, Jacobs J, et al. High-frequency oscillations, extent of surgical resection, and surgical outcome in drug-resistant focal epilepsy. Epilepsia 2013;54:848–57.

Jacobs J, LeVan P, Chander R, Hall J, Dubeau F, Gotman J. Interictal high-frequency oscillations (80–500 Hz) are an indicator of seizure onset areas independent of spikes in the human epileptic brain. Epilepsia 2008;49:1893–907.

Jacobs J, LeVan P, Châtillon C-E, Olivier A, Dubeau F, Gotman J. High frequency oscillations in intracranial EEGs mark epileptogenicity rather than lesion type. Brain 2009;132:1022–37.

Jacobs J, Zijlmans M, Zelmann R, Châtillon C-E, Hall J, Olivier A, et al. High-frequency electroencephalographic oscillations correlate with outcome of epilepsy surgery. Ann Neurol 2010;67:209–20.

Jacobs J, Vogt C, LeVan P, Zelmann R, Gotman J, Kobayashi K. The identification of distinct high-frequency oscillations during spikes delineates the seizure onset zone better than high-frequency spectral power changes. Clin Neurophysiol 2016;127(1):129–42.

Jiruska P, Bragin A. High-frequency activity in experimental and clinical epileptic foci. Epilepsy Res 2011;97:300–7.

Jobst BC. Are HFOs still UFOs? The known and unknown about high frequency oscillations in epilepsy surgery. Epilepsy Curr. 2013;13:273–5.

Koch SP, Werner P, Steinbrink J, Fries P, Obrig H. Stimulus-induced and state-dependent sustained gamma activity is tightly coupled to the hemodynamic response in humans. J Neurosci 2009;29:13962–70.

Lachaux JP, Axmacher N, Mormann F, Halgren E, Crone NE. High-frequency neural activity and human cognition: past, present and possible future of intracranial EEG research. Prog Neurobiol 2012;98:279–301.

Lüders HO, Awad I. Conceptual considerations. In: Lüders HO, editor. Epilepsy surgery. New York: Raven Press; 1992. p. 51–62.

Manning JR, Jacobs J, Fried I, Kahana MJ. Broadband shifts in local field potential power spectra are correlated with single-neuron spiking in humans. J Neurosci 2009;29:13613–20.

Matsumoto A, Brinkmann BH, Stead SM, Matsumoto J, Kucewicz MT, Marsh WR, et al. Pathological and physiological high-frequency oscillations in focal human epilepsy. J Neurophysiol 2013;110:1958–64.

Nagasawa T, Juhász C, Rothermel R, Hoechstetter K, Sood S, Asano E. Spontaneous and visually driven high-frequency oscillations in the occipital cortex: intracranial recording in epileptic patients. Hum Brain Mapp 2012;33:569–83.

Niessing J, Ebisch B, Schmidt KE, Niessing M, Singer W, Galuske RA. Hemodynamic signals correlate tightly with synchronized gamma oscillations. Science 2005;309(5736):948–51.

Nishida M, Juhász C, Sood S, Chugani HT, Asano E. Cortical glucose metabolism positively correlates with gamma-oscillations in nonlesional focal epilepsy. Neuroimage 2008;42:1275–84.

Rosenow F, Lüders H. Presurgical evaluation of epilepsy. Brain 2001;124:1683–700.

Staba RJ, Wilson CL, Bragin A, Fried I, Engel Jr J. Quantitative analysis of high-frequency oscillations (80–500 Hz) recorded in human epileptic hippocampus and entorhinal cortex. J Neurophysiol 2002;88:1743–52.

Urrestarazu E, Chander R, Dubeau F, Gotman J. Interictal high-frequency oscillations (100–500 Hz) in the intracerebral EEG of epileptic patients. Brain 2007;130:2354–66.

Usui N, Terada K, Baba K, Matsuda K, Usui K, Tottori T, et al. Significance of very-high-frequency oscillations (over 1000 Hz) in epilepsy. Ann Neurol 2015;78:295–302.

Wang S, Wang IZ, Bulacio JC, Mosher JC, Gonzalez-Martinez J, Alexopoulos AV, et al. Ripple classification helps to localize the seizure-onset zone in neocortical epilepsy. Epilepsia 2013;54:370–6.

Worrell GA, Parish L, Cranstoun SD, Jonas R, Baltuch G, Litt B. High-frequency oscillations and seizure generation in neocortical epilepsy. Brain 2004;127:1496–550.

Worrell GA, Gardner AB, Stead SM, Hu S, Goerss S, Cascino GJ, et al. High-frequency oscillations in human temporal lobe: simultaneous microwire and clinical macroelectrode recordings. Brain 2008;131:928–37.

Wu JY, Sankar R, Lerner JT, Matsumoto JH, Vinters HV, Mathern GW. Removing interictal fast ripples on electrocorticography linked with seizure freedom in children. Neurology 2010;75:1686–94.

Zelmann R, Zijlmans M, Jacobs J, Châtillon C-E, Gotman J. Improving the identification of high frequency oscillations. Clin Neurophysiol 2009;120:1457–64.

Zijlmans M, Jiruska P, Zelmann R, Leijten FS, Jefferys JG, Gotman J. High-frequency oscillations as a new biomarker in epilepsy. Ann Neurol 2012;71:169–78.