

Two types of clinical ictal direct current shifts in invasive EEG of intractable focal epilepsy identified by waveform cluster analysis

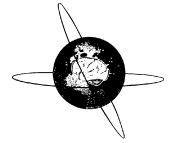
(難治部分てんかん患者の侵襲的脳波を用いたクラスター分析によって同定された発作時直流緩電位変化の2分類)

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Two types of clinical ictal direct current shifts in invasive EEG of intractable focal epilepsy identified by waveform cluster analysis

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HIGHLIGHTS

- Ictal direct current shifts can be observed at a time constant of 2 s.
- Two types of ictal direct current shifts were revealed by the waveform property.
- Each type of direct current shift may be correlated with the corresponding pathology.

ABSTRACT

Objective: To determine clinically ictal direct current (DC) shifts that can be identified by a time constant (TC) of 2 s and to delineate different types of DC shifts by different attenuation patterns between TC of 10 s and 2 s.

Methods: Twenty-one patients who underwent subdural electrode implantation for epilepsy surgery were investigated. For habitual seizures, we compared (1) the peak amplitude and (2) peak latency of the earliest ictal DC shifts between TC of 10 s and 2 s. Cluster and logistic regression analyses were performed based on the attenuation rate of amplitude and peak latency with TC 10 s.

Results: Ictal DC shifts in 120 seizures were analyzed; 89.1% of which were appropriately depicted even by a TC of 2 s. Cluster and logistic regression analyses revealed two types of ictal DC shift. Namely, a rapid development pattern was defined as the ictal DC shifts with a shorter peak latency and they also showed smaller attenuation rate of amplitude (73/120 seizures). Slow development pattern was defined as the ictal DC shifts with crosscurrent of a rapid development pattern, i.e., a longer peak latency and larger attenuation rate of amplitude (47/120 seizures). Focal cortical dysplasia (FCD) 1A tended to show a rapid development pattern (22/29 seizures) and FCD2A tended to show a slow development pattern (13/18 seizures), indicating there might be some correlations between two types of ictal DC shift and certain pathologies.

Abbreviations: AC, alternative current; AQP4, Aquaporin-4; ATP, adenosine triphosphate disodium hydrate; Cx, connexin; DC, direct current; ECoG, electrocorticograms; FCD, focal cortical dysplasia; HFO, high frequency oscillations; HS, hippocampal sclerosis; iEEG, intracranial electroencephalography; IIR, infinite impulse response; LFF, low frequency filter; STFT, short-time Fourier transform; TC, time constant; TLE, temporal lobe epilepsy.

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Conclusions: Ictal DC shifts, especially rapid development pattern, can be recorded and identified by the AC amplifiers of TC of 2 s which is widely used in many institutes compared to that of TC of 10 s. Two types of ictal DC shifts were identified with possibility of corresponding pathology.

Significance: Ictal DC shifts can be distinguished by their attenuation patterns.

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1. Introduction

Currently, an intracranial electroencephalography (iEEG), that is, electrocorticograms (ECoG) is the gold standard for identifying the epileptogenic zone before epilepsy surgery. Owing to recent technological advances, it is now possible to record iEEG (ECoG) from extremely slow to very rapid frequencies (Worrel et al., 2008). The so-called ictal direct current (DC) shifts, usually less than 0.1 Hz, are now reported as a surrogate marker of epileptogenicity (Ikeda et al., 1996, 1999; Modur et al., 2012; Rodin and Modur, 2008; Vanhatalo et al., 2003; Bragin et al., 2005; Bastany et al., 2020; Thompson et al., 2016). In contrast, high-frequency oscillations (HFOs) of more than 80 Hz are considered to be specific markers of the epileptogenic zone (Jacobs et al., 2008, 2009). HFO is thought to reflect the activity of synchronized principal neurons (Jiruska et al., 2017), whereas ictal DC shifts are considered to be astroglial depolarizations (Ikeda et al., 2020). Recent research has confirmed the importance of astroglia, at least partly, in epileptogenesis (Ikeda et al., 2020; Tian et al., 2005; Amzica and Massimini, 2002), and the astroglia are now a candidate target for treatment in patients with focal epilepsy (Walrave et al., 2018).

In the past, we found that ictal DC shifts preceded ictal HFO in patients with intractable focal epilepsy, especially of neocortical origin and to a lesser degree of mesial temporal origin (Imamura et al., 2011, Kanazawa et al., 2015, Nakatani et al., 2019), suggesting active depolarizations of astrocytes preceding ictal firing of neurons. It was later referred to as active DC shift by our institute (Ikeda et al., 2020). Further investigations of active ictal DC shifts, particularly as an active marker of the astroglia, could shed light on the pathophysiology of human focal epilepsy. However, there are two practical limitations in recording and investigating ictal DC shifts. (1) Ictal DC shifts have been reported using a long time constant (TC) of 10 s (low frequency filter [LFF] of 0.016 Hz) (Kanazawa et al., 2015; Wu et al., 2014; Thompson et al., 2016). Many institutes can only record iEEG with the longest possible value of TC of 2 s (LFF, 0.08 Hz), which may hinder the clinical application of ictal DC shifts. (2) Ictal DC shifts have been operationally defined as slow shifts of 3 s or longer in duration by a previous study (Ikeda et al., 1999); however, recent subsequently accumulated data until 2018 revealed (Kajikawa et al., 2018) that time differences between the onset and peak varied from 1 s to 20 s. Recently, two types of ictal DC shifts were proposed (Gnatkvosky et al., 2019), but the systematic logical analysis of waveform features of ictal DC shifts using different display TC and cluster analysis methods and the verification of those characteristics has not been performed. In addition, pathological correlations with certain waveform patterns of ictal DC shifts have been reported very rarely (Gnatkvosky et al., 2019).

To solve these issues, we attempted to determine whether ictal DC shifts can be recorded by TC at not only 10 s but also 2 s and performed cluster and logistic regression analyses to confirm the presence of two types of ictal DC shifts by adapting the number of clusters. We also delineated a clear positive association between the two types of ictal DC shifts and pathology. The more logically proven presence of two types of ictal DC shifts could further help

us clarify the active role of astroglia in seizure generation in chronic epilepsy and the development mechanisms of epileptogenesis in patients with intractable focal epilepsy.

2. Patients and methods

2.1. Patients

We retrospectively evaluated 21 of 30 patients with intractable focal epilepsy in whom chronic subdural electrodes were implanted before resection surgery at Kyoto University Hospital between June 2008 and October 2014. The demographic and clinical characteristics of the patients (9 women and 12 men) are summarized in Table 1. Twelve of the 21 patients were previously reported by Kanazawa et al. in 2015, with an entirely different purpose from the present study. And other 9 patients were also enrolled in other study to reveal the correlation between ictal DC shifts, ictal HFO, and other standard EEG and between ictal DC shifts and surgical outcome. Therefore, current study we focused on only waveform property of ictal DC shifts. Since the present study further analyzed the waveform property of the ictal DC shifts, the inclusion criteria of the seizures were the same as those done previously by Kanazawa et al. (2015) as follows: (1) at least two habitual clinical seizures or at least one clinical seizure and subclinical seizure, (2) seizures with reproducible morphology and distribution of ictal DC shifts, (3) and absence of multifocal seizures. The research protocol was approved by the Ethical Committee of Kyoto University Graduate School of Medicine (approval R0603) and performed in accordance with the Code of Ethics of the World Medical Association (Declaration of Helsinki). Written informed consent was not required because of the retrospective observational design.

Subdural grid electrodes were implanted in all patients, with strip electrodes added in 18 and depth electrodes in seven patients. The mean number of implanted electrodes per patient was 84.4 ± 25.7 (range, 48–125). The intracranial electrodes were in place for a mean duration of 12.9 ± 2.4 (range, 6–14) days. The mean patient age at seizure onset was 16.4 ± 10.5 (range, 0.83–43) years and 33.4 ± 11.1 years at the time of epilepsy surgery (range, 17–61).

Pathological diagnosis of the resected brain tissues of patients 1, 2, and 13 was confirmed by one of the authors (AK), while others were performed at our institute. All patients underwent magnetic resonance imaging of the brain, long-term video-scalp EEG monitoring, interictal fluorodeoxyglucose positron emission tomography scanning, and neuropsychological and Wada tests as part of their presurgical evaluation.

2.2. Implantation of invasive electrodes

The number and location of the implanted EEG electrodes were individualized according to the findings of the pre-surgical evaluation. Most of the subdural electrodes and additional depth electrodes were made of platinum. The subdural grid or strip electrodes had a recording surface with a diameter of 2.3 mm

Table 1
Patient demographics and clinical characteristics.

Patient	Age at onset/at surgery Gender	Clinical epilepsy diagnosis	Location and type of electrode	The number of electrodes	Location of the earliest DC shifts (the number of Sz in each region)	pathology	Number of Sz analyzed /Total number of Sz	The number of type#1 and type#2 seizures (typ#1/type#2)
1	16 / 22 / M	L Fr-TLE	L Fr/T (grid, strip)	56 grids, 10 strip	L Fr	FCD IA	2/18	2/0
2	15/32/F	L T-PLE	L Fr/T/P (grid, strip, depth)	80 grids, 10 strip, 6 depth	L P	FCD IA	10/16	10/0
3	19/41/F	L TLE	L Fr/T/P (grid, strip, depth)	92 grid, 10 strip, 16 depth	L T	FCD IA	9/13	4/5
4	15/45/M	L FrLE	L Fr/P (grid, strip)	100 grid, 4 strip	L Fr	FCD IA	3/14	2/1
5	18/29/M	R FrLE	R Fr/P (strip, depth)	74 grid, 26 strip, depth 12	R Fr	FCD IA	5/53	4/1
6	11/17/F	L TLE	L Fr/T/P (strip)	56 grid, 4 strip	L T	FCD IB	3/437	1/2
7	10/24/M	L FrLE	L Fr/T/P, (grid)	86 grid	L Fr	FCD IB	5/29	2/3
8	10 m/27/ M	R T-PLE	R Fr/P/O (grid)	56 grid	R Fr (8), P(1)	FCD IIA	9/28	0/9
9	8/38/F	L TLE	L Fr/T/P (grid, strip)	76 grid, 10 strip	L T	FCD IIA	2/5	0/2
10	16/23/M	R OLE	R O (grid, strip)	40 grid, 12 strip	R O	FCD IIA	3/3	1/2
11	23/30/F	R TLE	R T/P/O (grid, strip, depth)	92 grid, 10 strip, 14 depth	R T	FCD IIA/mild gliosis	4/23	3/1
12	9/27/F	R PLE	R O/P (grid, strip, depth)	52 grid, 20 strip, depth 30	R O	FCD IIB	7/12	3/4
13	40/40/M	L Fr-PLE	L Fr/P (grid)	56 grid	L Fr	Oligodendroglioma	9/46	6/3
14	11/44/M	R FrLE	R Fr/P (grid, strip)	36 grid, 12 strip	R Fr	Mixed oligoastrocytoma/ FCD IA	10/142	9/1
15	28/61/M	L PLE	L T/P (grid, strip)	92 grid, 14 strip	L P	Oligoastrocytoma	3/13	2/1
16	1220/F	R TLE	R Fr/T (grid, strip)	46 grid, 4 strip	R T	Oligodendroglioma/ FCD IB	10/73	0/10
17	24/28/F	R PLE	R Fr/P (grid, strip, depth)	20 grid, 28 strip, 8 depth	R Fr (9), P(1)	Low-grade glioma	10/43	10/0
18	8/34/M	R T-PLE	R Fr/T/P (grid, strip)	60 grid, 22 strip	R T	posttraumatic change(parietal)/scar(temporal)/HS	2/13	2/0
19	12/28/F	L TLE	L Fr/T/P (grid, strip)	96 grid, 10 strip	L T	Non neoplastic brain tissue	5/90	5/0
20	43/52/M	L TLE	L Fr/T/P/O (grid, strip, depth)	92 grid, 18 stirp, 15 depth	L T	Arteriovenous malformation/ gliosis	3/3	3/0
21	6/39/M	R TLE	R Fr/T/P (grid, strip)	76 grid, 14 strip	R Fr (2)/T(4)	HS/FCD IA	6/21	3/3

F, female; Fr, frontal; FCD, focal cortical dysplasia; HS, hippocampal sclerosis; L, left; M, male; O, occipital; P, parietal; R, right; Sz, seizure; T, temporal. Abbreviations: F, female/frontal; FCD, focal cortical dysplasia; HS, hippocampal sclerosis; L, left; M, male; O, occipital; P, parietal; R, right; T, temporal.

(Ad-Tech, Racine, WI, USA) or 3 mm (Unique Medical Co., Ltd., Tokyo, Japan) and a center-to-center interelectrode distance of 1 cm. In four patients (Patients 4, 11, 15, and 17), grid electrodes with a recording diameter of 1.5 mm and a center-to-center interelectrode distance of 5 mm were also used. All depth electrodes had a diameter of 1.5 mm, a contact length of 1 mm, and a center-to-center interelectrode distance of 5 mm.

2.3. Recording, waveform analysis, and pathological correlation

For data recording, the bandpass filter for iEEG (ECoG) was set from 0.016 (TC of 10 s) to 300 Hz or 600 Hz with a corresponding sampling rate of 1000 Hz or 2000 Hz (EEG1100, AC amplifier with input impedance of 200 MX, Nihon Kohden, Tokyo, Japan). We set a recording reference or system reference applied to the skin during the contralateral mastoid process. With regard to the display reference, we applied an averaged reference, including all intracranial electrodes.

We analyzed ictal DC shifts using a wide-band time frequency analysis program (Nihon Kohden, Tokyo, Japan). For the purpose of this study, we selected one electrode per seizure showing the earliest ictal DC shift. And we analyzed the maximum of 10 seizures in each patient as the previous study sign (Kanazawa et al., 2015). Since DC shifts did not always occur but 70–100% of the

recorded seizures, and thus in some patients, less than 10 seizures were analyzed for DC shifts in those patients.

Finally, we analyzed 119 seizures from the grid or strip electrodes in most patients. And only 1 of the 10 seizures in Patient 17, the earliest DC shifts were chosen from the depth electrode. In this study, we adopted the previous definition of ictal DC shifts as follows (Kanazawa et al., 2015). The subdurally recorded ictal DC shifts were defined as “a sustained negative and/or positive potential longer than 3 s and higher than 200 μ V, preferentially more than 1000 μ V” with a TC of 10 s. The analysis window was set to 120 s, including the EEG segment 60 s before and after conventional ictal EEG onset. Only Patient 14 did not show conventional ictal EEG; we identified ictal onset as the earliest EMG change in accordance with a previous study (Kanazawa et al., 2015). The baseline was set to the first 12 s, that is, 10% of the analysis window.

We introduced the parameters of the individual ictal DC shifts produced by the following two conditions for comparison: (1) original data without applying digital LFF, and (2) data applied by LFF corresponding to TC of 2 s digitally (infinite impulse response [IIR]) on the original data. Because TC is a first-order low-frequency filter, it is suitable to apply LFF. However, as TC is usually set in an amplifier in many manufactures, we verified the DC shift difference by changing the TC. In both conditions, a high-frequency filter (IIR

Table 2

A comparison between ictal DC shifts with the rapid development pattern and those with the slow development pattern among 120 seizures and 21 patients.

	Type#1 Rapid development pattern	Type#2 Slow development pattern
Number of Sz	73/120 (60.8%)	47/120 (39.2%)
Number of patients	14/21 (66.7%)	7/21 (33.3%)
Waveform displayed by TC 10 s		
Peak amplitude (μV)	693.1 ± 297.7	1018.3 ± 484.1
Peak latency (s)	4.5 ± 2.4	15.8 ± 6.6
Waveform displayed by TC 2 s		
Peak amplitude (μV)	484.6 ± 235.8	386.0 ± 221.8
Peak latency (s)	2.8 ± 2.1	10.2 ± 6.1
Amp. attenuation rate from TC 10 s to TC 2 s	Small (36.4%)	Large (65.3%)

DC, direct current; Sz, seizure; TC, time constant.

Butterworth 4th order zero-phase shift applied bidirectionally with a cutoff frequency of 1 Hz) was applied. Then, the DC onset and DC peak were manually decided, and the two following parameters were assessed compared between the above two conditions: amplitude (value of the electrical potential from the onset to peak of DC shifts) and peak latency (duration from the onset to the peak of DC shifts) (Fig. 1). The measurements of these two parameters and the conventional EEG onset were analyzed visually, in the same way as reported in a previous study (Kanazawa et al., 2015).

The figure plotting the individual seizures that displayed TC of 10 s was then constructed with the attenuation rate on the vertical axis and the peak latency on the horizontal axis. The correlation between the attenuation amplitude rate and peak latency with a TC of 10 s was examined using Spearman's correlation coefficient method (Fig. 2A).

Based on the correlation between amplitude attenuation rate and the degree of peak latency values at a TC of 10 s in 120 seizures, as the next step, we attempted to classify the ictal DC shifts by cluster analysis in order to identify which types and how many subtypes of DC shifts at TC of 10 s were more attenuated at TC of 2 s. We assumed that the number of clusters could not be so high because of its biological and physiological validity. Therefore, the number of clusters was not initially given; thus, hierarchical clustering was used. The details of the method are as follows:.

Let B and C be two clusters including one plot cluster and A be the union of B and C, and the distances of B and C were defined using the following formula:

$$d(B, C) = \sum_{i=1}^{N_A} (A_i - \mu_A)^2 - \sum_{i=1}^{N_B} (B_i - \mu_B)^2 - \sum_{i=1}^{N_C} (C_i - \mu_C)^2$$

where μ_x is the average of each cluster x, and N_x is the number of elements in each cluster x. We could find a pair of two clusters B

and C, minimizing D (B, C). The new cluster is then identified as A (union of B and C).

The cluster became hierarchically larger step-by-step until finally, a dendrogram showing the smallest clustering on the bottom and the largest clustering on the top was produced. Subsequently, the Calinski-Harabasz method was applied to determine the optimal number of clusters. This method calculates the Calinski-Harabasz index, which is the ratio of the between-cluster variance to the total within-cluster variance, as follows:

$$\text{Calinski-Harabasz index}_k = \frac{SS_B}{SS_w} \times \frac{(N-k)}{(k-1)}$$

where SS_B is the between-cluster variance, SS_w is the total within-cluster variance, N is the total number of seizures, and k is the number of clusters. The clustering number showing the largest Calinski-Harabasz index is suitable for optimal clustering.

In this study, the Calinski-Harabasz index showed the highest values for 2, 9, and 10 clustering. Therefore, we adopted two clusters for the classification of ictal DC shifts because of the assumption mentioned above.

We then compared each parameter between detected clusters. Furthermore, to obtain a separation line on the peak latency-attenuation plot, we applied the following logistic regression analysis:.

Letting x be the peak latency and y be the attenuation of a seizure, the point (x,y) was labeled according to the two classes identified by the cluster analysis. Label 1 corresponded to a slow development pattern and label 0 to a rapid development pattern.

To apply logistic regression, we first defined the following function:.

$$f(x, y) = 1 / (1 + e^{(ax+by+c)})$$

This is a sigmoidal function, and the label function $l(x, y)$ indicates the label (x,y).

Second, using the least squares method, we found the values of a, b, and c, which minimized the absolute values of the sum over all the pairs of (x,y) of the square as follows:

$$(f(x, y) - l(x, y))^2$$

Finally, the separation line was obtained as:

$$ax + by + c = 0$$

because the value of $f(x, y)$ equals 1/2 on the separation line, where the line shows the midpoint of the two classes of point (x,y) as either $l(x, y) = 1$ or $l(x, y) = 0$.

Given that these analyses were based on bivariate logistic regression, we also applied a univariate logistic regression to determine the threshold of peak latency for the clinical situation. There-

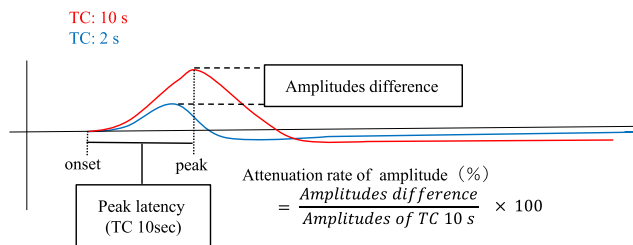


Fig. 1. Measuring parameters of ictal direct current (DC) shifts. It shows the schema of ictal DC shifts with a time constant (TC) of 10 s (red) and that with TC of 2 s (blue). In each TC, the peak latency and amplitude, from onset to peak, were investigated. The difference between the amplitude by TC of 10 s and TC of 2 s was divided by the amplitude by TC of 10 s, namely, the amplitude attenuation rate was calculated.

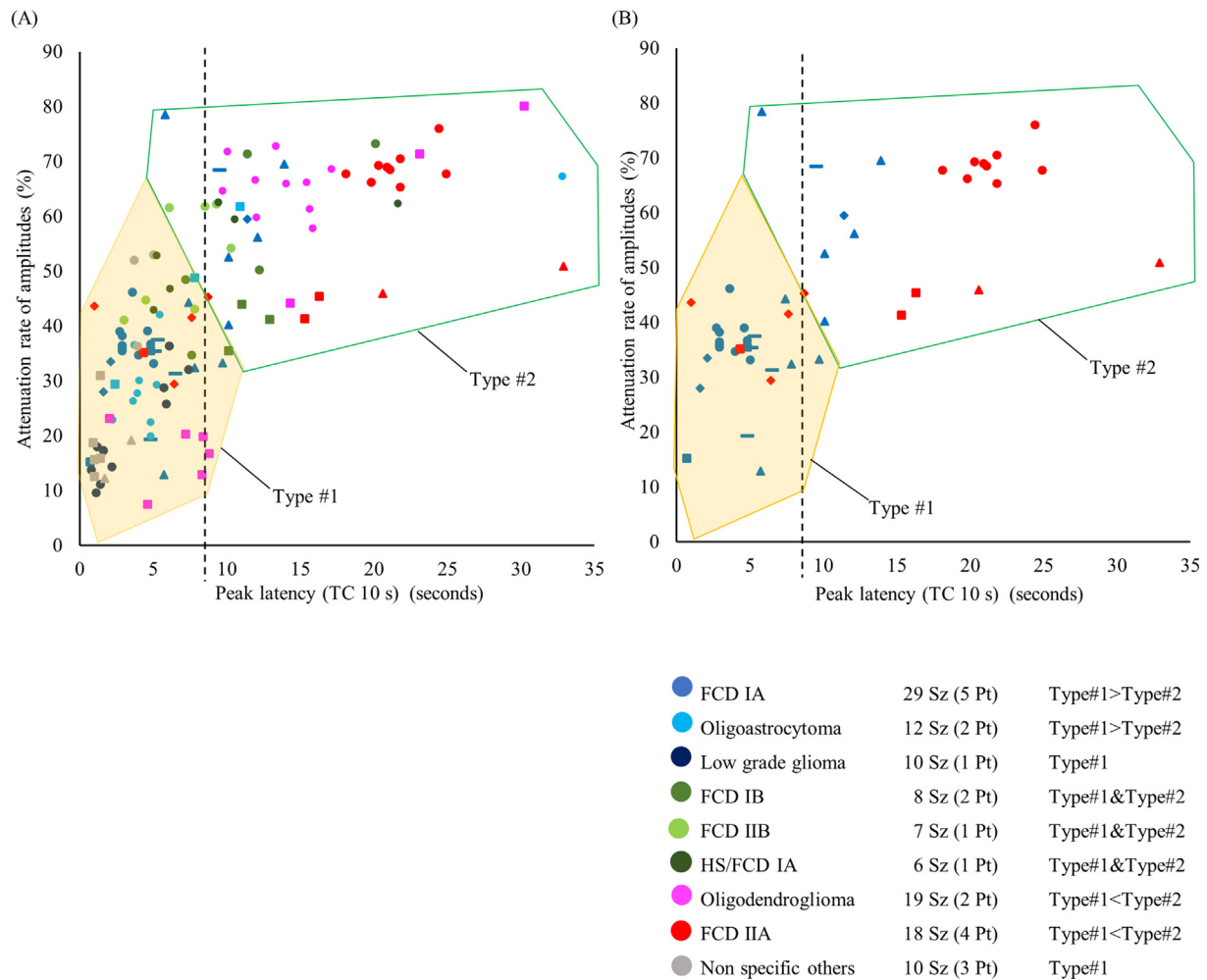


Fig. 2. Correlation between amplitude attenuation rate and peak latency. It was made with a TC of 10 s, as determined by cluster and logistic regression analyses. The plot shows the parameters of the ictal direct current shifts for each seizure. The vertical axis shows the amplitude attenuation rate, and the horizontal axis shows the peak latency of the ictal direct current shifts described with a TC of 10 s. (A) Specific colors were assigned to each pathology. If several patients had the same pathology, each patient was distinguished by different symbols (i.e., circle, square, triangle, rhombic, and stick). The results of the cluster analysis are represented as enclosed yellow (shaded) or green line circling areas. Type #1 were circled by yellow line, and type #2 were circled by green line. The dotted line indicates the results of logistic regression analysis with univariate analysis in black. The peak latency threshold was 8.9 s. (B) Figure focused on focal cortical dysplasia (FCD) IA and FCD IIA from Fig. 2A. Plots color, symbols, yellow and green line circling, and the dotted line demonstrate same as Fig. 2A. HS; hippocampal sclerosis.

fore, the following function was also applied to the univariate logistic regression:

$$f(x) = 1 / (1 + e^{(ax+b)})$$

Additionally, we performed cluster and logistic regression analyses with bivariate peak latency using a TC of 2 s and an amplitude attenuation rate. Univariate logistic regression of peak latency with TC of 2 s was also performed to confirm whether or not we could apply the threshold at TC of 2 s.

With regard to the pathological association, we assessed the correlation between two clusters of ictal DC shifts and pathological findings, based on the hypothesis that the degree of astroglial dysfunction, especially the impairment of extracellular potassium homeostasis, may differ according to the pathology, and it could modify the generator mechanism of ictal DC shifts in onset time, degree of steepness after onset, peak amplitude, peak latency,

etc. The relationship between the two types of ictal DC shift and pathology, if any, was investigated using Fisher's exact test.

3. Results

3.1. Overall wave form characteristics by means of different display TC (10 s vs. 2 s)

We analyzed 120 seizures in 21 patients. As documented in Table 1, DC shifts did not always appear in all seizures in each patient. And we analyzed max 10 seizures per patient. The mean number of seizures per patient (with standard deviation) was 5.7 ± 3.1 . The mean amplitude of the ictal DC shifts was $820.5 \pm 411.9 \mu\text{V}$ by TC of 10 s and $446.0 \pm 234.5 \mu\text{V}$ by TC of 2 s. A total of 89.1% (107/120) of the seizures showed ictal DC shifts greater than $200 \mu\text{V}$ with TC of 2 s. The mean peak latency values are 8.9 ± 7.1 s

and 5.7 ± 5.5 s, by TC of 10 s and of 2 s, respectively. The mean amplitude attenuation rate for the ictal DC shifts was $42.9\% \pm 19.0\%$ in all seizures.

3.2. Two types of ictal DC shifts by means of wave form parameters (TCs 10 s vs. 2 s) (Figs. 2 and 3, Table 2)

Fig. 2A shows the relationship between the amplitude attenuation rate and peak latency of ictal DC shifts by TC for 10 s in all seizures. The correlation between the two factors was significant ($r_s = 0.76$, $p < 0.0001$). Namely, the more the peak amplitude is attenuated by changing TC from 10 s to 2 s, the longer the peak latency in the TC of 10 s was coupled.

Cluster analysis by means of peak latency with TC of 10 s and the amplitude attenuation rate in combination could classify all seizures appropriately into two types: type #1, which tended to have a smaller amplitude attenuation rate and a shorter peak latency (named as rapid development pattern, Fig. 3A), and type #2, which showed the opposite tendency (slow development pattern; Fig. 3B). In total, 60.8% of the seizures (73/120) were type #1 and 39.2% (47/120) were type #2.

The parameters for the two patterns by display TC of 10 s and 2 s are listed in Table 2. Type #1 showed a smaller amplitude and a shorter peak latency in TC of 10 s compared to type #2, whereas the amplitude of type #1 was larger than that of type #2 in TC of 2 s. The amplitude attenuation rate was smaller in type #1. Logistic regression analysis showed that the best threshold was 8.9 s. This threshold could correctly identify type #1 (rapid development pattern) in 97.3% of seizures (71/73) and type #2 (slow development pattern) in 93.6% (44/47).

3.3. Correlation between subtypes of ictal DC shifts and pathology (Fig. 2)

While in all patients except for Patients 1 and 2 with focal cortical dysplasia (FCD) the respective brain tissue was diagnosed using the Palmini definition in our institute, the Blümcke definition

was used in Patients 1 and 2 by AK. To unify the definition, we applied the Palmini definition to the latter two patients so that there was no contradiction.

Finally, the pathological classification was FCD in 13 patients, glioma in five, and “non-specific others” (i.e., non-specific changes as a scar or gliosis comorbid with arteriovenous malformation) in three. Two out of five patients with glioma were comorbid with FCD (Patient 14, Patient 16). Only one patient with hippocampal sclerosis (HS) with FCD was also included (Patient 21).

Fig. 2B and C show a color-coded plot of the seizures for each type of pathology. Individual patients were assigned a specific color according to the type of pathology.

Type #1 (rapid development pattern) was mainly observed in FCD IA (22/29 seizures, 75.8%; 4/5 patients, 80%), followed by “non-specific others” (10/10 seizures, 100%; 3/3 patients, 100%), oligoastrocytoma (11/13 seizures, 84.6%; 2/2 patients, 100%), and low-grade glioma (10/10 seizures 100%; 1/1 patient, 100%). Type #2 (slow development pattern) was seen in FCD IIA (13/18 seizures, 72.2%; 3/4 patients, 75%), followed by oligodendroglioma (13/19 seizures, 68%; 2/2 patients, 100%).

Some pathologies showed both types of DC shifts as follows. 1) In FCD IB (two patients) and FCD IIB (one patient), 5/8 seizures (62.5%) and 4/7 seizures (57.1%) showed type #2, and in FCD IIB 3/5 seizures and 3/7 seizures were type #1. 2) HS comorbid with FCD IA (1 patient) also showed both types of ictal DC shifts (out of six seizures, three showed type #1 and three showed type #2).

In summary, there is a minor overlap of type #1 and #2 DC shifts in the same patients (Patients 6, 7, 12, 21) and in the same pathology (FCD IB, FCD IIB, HS/FCD IA); however, we were able to successfully extract the major features of two types of DC shifts corresponding to FCD IA (five patients and two with comorbidity) and FCD IIA (four patients and one with comorbidity) since both pathologies were dominant in number compared to others. We found a significant correlation between FCD IA and type #1 (rapid development pattern) ($68.9\% = 33/44$ seizures) and FCD IIA for type #2 (slow development pattern) ($72.2\% = 23/28$ seizures) ($p < 0.01$).

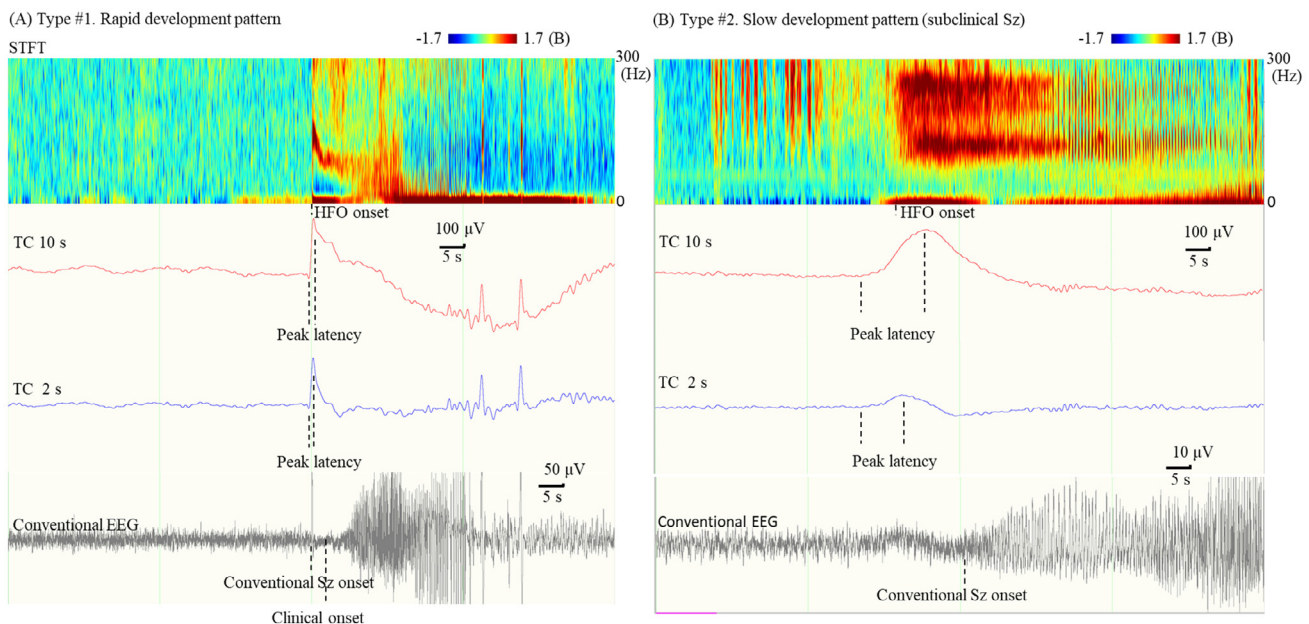


Fig. 3. Representative waveforms of two types of ictal direct current (DC) shift. (A). Example of type #1 (rapid development pattern) ictal DC shifts of Patient 17 (clinical seizure) is shown. (B) Representative example of type #2 (slow development pattern). The figures were obtained from Patient 16 (subclinical seizure). Both (A) and (B) show the ictal DC shifts with a TC of 10 s (red) and 2 s (blue), and conventional seizure patterns which were observed by bandpass filter of 1.6–120 Hz (black). To describe the time differences between ictal DC shifts and ictal HFO, the figure also shows short-time Fourier transform (STFT) (top), ranging between 0 and 600 Hz of frequency. The STFT was analyzed with a window frequency of 10 Hz. High frequency filter = 1 Hz, HFO; high-frequency oscillations, STFT; short-time Fourier transform. Sz; seizure.

4. Discussion

We found the three important findings in this study from intractable epilepsy patients before surgery as follows: 1) ictal DC shifts can be divided into two types: type #1 rapid development pattern of short duration with less amplitude attenuation, and type #2 slow development pattern of long duration with more amplitude attenuation, 2) and thus ictal DC shifts can be recorded at least partly even by an alternate current-coupled amplifier with TC of 2 s, where the type #1 pattern could be easily identified, and 3) the two types of ictal DC shifts may be correlated with an underlying pathology mainly for FCD IA vs. IIA, respectively, although this result was still rather limited because of a small sample size.

Current study suggests that AC amplifier of TC of 2 s, which is widely used in many institutes rather than that of TC of 10 s, is at least partly sufficient to record and identify ictal DC shifts. This finding could facilitate clinical application of ictal DC shifts for epilepsy surgery.

It also clearly demonstrated that two types of ictal DC shifts were defined. Namely, type #1 (rapid development pattern) could be well detected by TC of 2 s because it is lesser attenuated by TC of 2 s, and it may tend to be pathologically FCD IA. Therefore, it may also lead to pathological correlation with two types of ictal DC shifts, and to different epileptogenicity with regard to the contribution of glia.

In current study, patients were partly recruited from [Kanazawa et al., 2015](#). And the relationship of DC shifts and HFOs or conventional iEEG (ECoG) were documented in [Kanazawa et al., 2015](#). The results were described in abstract as follows; the onset of ictal DC shifts preceded that of ictal HFOs with statistical significance. And we accumulated other patients from other research that also mentioned same results as Kanazawa et al. ([Nakatani et al., 2019](#)). Therefore, we focused on only ictal DC shifts of different type.

4.1. Presence of two types of ictal DC shifts

We were able to classify ictal DC shifts into two types based on waveform analysis and cluster analysis. Type #1 showed a shorter peak latency and smaller amplitude attenuation rate that appeared once we reduced the display TC from 10 s to 2 s. Type #2 showed a longer peak latency and larger amplitude attenuation rate that appeared once we reduced the display TC from 10 s to 2 s. On EEG, the TC operates as an LFF to remove slower artifacts, and thus, a smaller TC tends to attenuate lower frequency waveforms. In our study, this characteristic successfully allowed us to discriminate between types #1 and #2 from the viewpoint of waveform easily.

TC is the first-order forward low-frequency filter; thus, not only the spectral characteristics but also the slope and morphology of the ascending part of the ictal DC shift could affect the attenuation pattern. Two types of ictal DC shifts can be distinguished based on these phenomena, which would reflect a different generator mechanism discussed later.

And it is the same signal that is just being filtered in different ways, therefore a shift is expected. It is very important as the attenuation factor is being used to define the two types of DC shifts and understanding the signal properties might reflect underlying mechanisms.

A recently published study proposed two possible types of classification of ictal DC shifts ([Gnatkovsky et al., 2019](#)). Their findings are similar to those of the present study, but our study provided two important, differently new points as follows. (1) The two types consist of the amplitude attenuation rate and peak latency, as explained above, by extracting the features of the waveform based on the function of different TC values. Furthermore, we also performed cluster analysis and logistic regression analysis, and a opti-

mal peak latency cut off value (8.9 s) was obtained to classify the ictal DC shifts quantitatively for clinical utility. And the number of two clusters was also adapted, whereas in the previous study, these objective processes were not introduced. We also set the peak latency threshold for ictal DC shifts with TC of 10 s as 8.9 s for classification purposes based on logistic regression analysis. This threshold can be applied to clinical situations. (2) In a previous study, they analyzed data using stereo-EEG electrodes, whereas we adopted subdural or strip grid electrodes (except for one seizure).

4.2. Ictal DC shifts with TC of 2 s

In previous research on the clinical situation of epilepsy surgery since the 1990 s, ictal DC shifts were usually recorded by TC of 10 s (LFF of 0.016 Hz), and only two studies adopted an LFF of 0.1 Hz with depth electrodes in patients with temporal lobe epilepsy patients ([Mader et al., 2005](#); [Rampp and Stefan, 2012](#)), suggesting that ictal DC shifts are clinically useful, even if they employed an LFF of 0.1 Hz.

Although our definition of ictal DC shifts was slightly different from that used previously, the findings of the present study could also support the clinical value of ictal DC shifts even by TC of 2 s (LFF, 0.08 Hz). Furthermore, unlike the earlier studies, we included neocortical epilepsy, and almost all of the iEEG (ECoG) data in our study were obtained from the subdural grid and strip electrodes. This suggests that ictal DC shifts could be recorded with a short TC of 2 s not only from depth but also from subdural electrodes, regardless of the electrodes' implanted locations. We also recently reported patients showing ictal DC shifts even on scalp EEG by TC of 2 s ([Murai et al., 2020](#)). This needs to be validated by more data on ictal DC shifts by scalp EEG.

4.3. Correlation between pathology and development pattern of ictal DC shifts

The present study showed a correlation between FCD IA and type #1 and FCD IIA and type #2 ictal DC shifts.

From a previous report, it is well-known that glial depolarization is triggered by an increase in extracellular potassium concentration $[K^+]_o$ passively ([Speckmann et al., 2011](#)), and that astrocytes also play a role in potassium buffering to maintain extracellular K^+ homeostasis, which could be related to the active depolarization of astrocytes per se. There is increasing research interest in the role of inwardly rectifying potassium (Kir) 4.1 channels on astrocytes to control spatial K^+ buffering. These channels are now considered to be involved in the development of epilepsy or epileptogenicity ([Kinboshi et al., 2019](#)) and their loss of function could cause elevation of $[K^+]_o$, leading to glial depolarization and triggering seizures, at least in chronic epilepsy ([Kanazawa et al., 2015](#)). Aquaporin-4 (AQP4) water channels, which are distributed in the end feet of astrocytes, work in concert with Kir 4.1 channels to buffer K^+ ([Nagelhus et al., 2004](#)). Gap junctions between astrocytes that form a functional syncytium, known as connexin 43 and 30 (Cx43 and Cx30, respectively), are thought to assist in spatial K^+ buffering by promoting the formation of large networks of astrocytes (functional syncytium) that redistribute K^+ within the cytosol ([Giaume et al., 2010](#)).

Thus, there might be qualitative and/or quantitative differences in astroglia among various pathologies that could modify the development pattern of ictal DC shifts into several types in the present study. Several studies have demonstrated astroglial dysfunction in the context of FCD, including loss of Kir 4.1 channels, redistribution of AQP4 channels, and abnormalities in Cx43 and Cx30 ([Sukigara et al., 2014](#); [Kielbinski et al., 2016](#)). Furthermore, our group recently found that (1) loss of Kir 4.1 channels happened in the area generating ictal DC shifts and (2) it was more evident in

FCD I than in FCD II (Kobayashi et al., 2019), as appeared in the abstract. Indeed, in this study type #1 ictal DC shifts tended to be shown in FCD I (3/5 patients) whereas type #2 in FCD II (2/2 patients), consistent with the current study.

This again suggests that the mechanisms underlying the development of chronic epilepsy and the generator mechanisms of ictal DC shifts may be different between the two pathologies (FCD IA and FCD IIA). However, within the same patient, both types #1 and #2 of ictal DC shifts were observed, possibly because homeostasis of variables such as pH, adenosine triphosphate disodium hydrate, and blood flow may fluctuate overtime once seizures begin to occur repeatedly. This could then affect the mechanism generating dichotic ictal DC shifts (Pinto et al., 2017). And seizure intensity might also affect the waveform of ictal DC shifts.

Finally, we could pay attention to the following findings: type #1 DC shifts (rapid development pattern) occurred exclusively in FCD I and that Kir4.1 channels might be downregulated mainly in FCD I (Kobayashi et al., 2019). Inversely, type #2 DC shifts (slow development pattern) occurred in FCD II, and Kir4.1 channels were not down-regulated although the number of patients were still limited. Therefore, our assumption is that decreased Kir4.1 activity for K homeostasis could be responsible for the very initial part of rapid development period of ictal DC shifts, and the following later period may represent the mixture of much lesser part of type #1 DC shifts and passive depolarization of astroglia produced by massive neuronal ictal activity, resulting in subsequent accumulation of extracellular K (Zuckermann and Glaser, 1970). Since this is still speculative, we need to warrant this hypothesis in the future.

4.4. Limitations

This study has several limitations. First, the sample size of patients was small, especially to search for a significant correlation between the two types of ictal DC shift and pathology we found. Second, our subtype of ictal DC shifts was analyzed only with a TC of 10 s. Therefore, another approach by means of TC of 2 s may be warranted in the future by increasing the sample size of patients.

Nevertheless, our research data clearly demonstrate that ictal DC shifts can be recorded by a TC of 2 s, which is widely used throughout the world. This strategy could help us detect the epileptogenic zone and identify patients with favorable surgical outcomes. Furthermore, the accumulation of these data on ictal DC shifts will delineate the pathophysiology of human epilepsy.

5. Conclusions

Our present findings suggest that invasive ictal DC shifts before epilepsy surgery can be recorded with a TC of 2 s and can be clearly classified into two types according to waveform property and its cluster analysis. Two types of ictal DC shifts and pathological correlation were at least partly suggested, and it is still rather limited because of the small sample size. Therefore, further analysis of ictal DC shifts from both basic and clinical perspectives is warranted to reveal the pathomechanisms of chronic epilepsy especially from view point of the glia.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Author contributions

SK, MM, KK, and AI contributed to the conception and design of the study; SK, MM, KK, MD, YY, TK1, KY, TK2, TK3, AK, TN, IT, and AI contributed to acquisition and analysis of the data; and SK, MM, KK, TH, RM, TH, RM, SM, RT, and AI contributed to drafting the text and preparing the figures.

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